Growth Performance, Fillet Quality, and Reproductive Maturity of Rainbow Trout (Oncorhynchus mykiss) Cultured to 5 Kilograms within Freshwater Recirculating Systems

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Abstract
Rainbow trout are commonly cultured within aquaculture systems to one pound or less and marketed as pan-sized fillets. Production of larger rainbow trout provides a distinguishable product. Research that describes the growth performance and fillet quality of large rainbow trout is limited, particularly for trout cultured in recirculating aquaculture systems. A study was conducted evaluating the growth performance and fillet quality attributes of all-female rainbow trout reared using freshwater recirculating systems operated at a mean water temperature of 13°C, under constant lighting, and with around-the-clock feeding. Rainbow trout grew to 4.8 kg in 22 months post-hatch. Growth rates declined with the onset of reproductive maturity. Rainbow trout weighed 5.2 kg at 26 months. The mean ratio of feed provided to biomass gain was 1.36:1 from first feeding to 22 months but increased substantially from 23-25 months. As rainbow trout approached reproductive maturity, 10 fish were collected at specified intervals for assessment of fillet quality attributes. Fillet yield peaked at 20-22 months when trout were 3.8-4.8 kg. Cook yield, cooked fillet firmness, and crude fat decreased; while fillet moisture and raw fillet firmness increased from 24-26 months. Changes in fillet quality coincided with reduced growth rates, decreased feed efficiency, and increasing gonadosomatic index. Two principal components were identified that explained more than 73% of the variation in growth and fillet attribute responses: principal component 1, the growth variable (length, weight, fillet thickness, belly flap thickness, and cook yield) and principal component 2, the quality variable (fillet moisture, fillet fat, and cooked fillet firmness). This research provides rainbow trout growth performance and fillet quality results that can be referenced for the development of recirculating system production plans and for selection of harvest endpoints that balance the requirements of fish farmers and the food industry sector.

Keywords: Rainbow trout; Fillet quality; Recirculating aquaculture systems; Reproductive maturity

Introduction
Recent development and advancements in water recirculating systems for the culture of rainbow trout (Oncorhynchus mykiss) and other salmonids [1-3] provide opportunity to expand current production levels. Commercial production of large 2-5 kg rainbow trout provides a niche product that is distinguished from the pan-sized rainbow trout (≤ 0.5 kg whole body weight) commonly marketed in many countries, including the United States [4]. Recently, there has been growing interest in culturing rainbow trout (a.k.a. steelhead) to harvest sizes >2 kg [4]; however, research that clearly defines growth performance and quality of larger trout is limited, particularly for trout cultured in recirculating systems. Several studies have evaluated fillet quality attributes for large, sexually maturing rainbow trout [4-10]. Most of these studies were focused on physiology during reproduction, were conducted using flow through systems, and did not thoroughly describe rainbow trout growth and fillet quality with age/time. The maximum rainbow trout weights reported in previous aquaculture studies were 3.6 and 3.1 kg [5,6], respectively. Research investigating the culture of large trout (>2 kg) in recirculating aquaculture systems (RAS) is particularly limited. Successful rainbow trout culture to approximately 1.3 kg was reported in a Danish model RAS [11]; and previous on-site studies demonstrated that rainbow trout can be cultured within RAS to approximately 1.3-1.4 kg in one year from hatch [2,12,13]. However, these studies did not attempt to culture trout to larger sizes and evaluation of fillet quality was limited.

One concern when rearing trout to larger sizes is the negative effects of sexual maturation on fillet yield and quality [4,14]. During sexual maturation, female rainbow trout develop ovaries that may account for >20% of the total body weight prior to ovulation [15]. As sexual maturation occurs, female rainbow trout metabolism is reorchestrated to meet nutrient demands of egg growth that, in turn, influences mobilization of energy and other stored nutrients [16]. A primary concern in regard to fillet quality is the mobilization of lipid from energy stores [6,9,10,14,17,18]; particularly intramuscular lipid, which in addition to visceral fat, can be catabolized to support egg development. Lipid mobilization, as well as protein, from muscle profoundly affects fillet quality and reduces fillet yield, as measured by percent separable muscle [4-9,19,20]. These changes in fillet composition and yield often lead to diminished quality of the final product and reduced revenue for the fish farmer.

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Received April 17, 2014; Accepted June 11, 2014; Published June 28, 2014

Citation: Davidson JW, Kenney PB, Manor M, Good CM, Weber GM, et al. (2014) Growth Performance, Fillet Quality, and Reproductive Maturity of Rainbow Trout (Oncorhynchus mykiss) Cultured to 5 Kilograms within Freshwater Recirculating Systems. J Aquac Res Development 5: 238 doi:10.4172/2155-9546.1000238

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Defining optimal rainbow trout fillet quality (and for that matter seafood quality) is a complex task, because quality encompasses a wide range of traits and variables, and the manufacture and sale of product involves many levels (farmers, processors, wholesalers, distributors, retailers, caterers, and consumers) with varying perceptions of quality [21]. For example, taste, odor, nutritional value (fatty acid content), firmness, and food safety are high ranking quality attributes to consumers, chefs, and caterers; while sex, maturity, fillet size, fat content, and product consistency have greater value to wholesalers, distributors, and retailers [19,21]. The form of the final product (i.e. filleted, smoked, canned, etc.) can also influence expected quality. For example, high quality smoked products require the raw material to be higher in fat than fillets used for other, non-smoked purposes [19,22]. At the farm level, multiple variables and their interactions contribute to these quality attributes, including: genetics [23], fish age/growth rate [24], diet [19], feeding regimen [25], photoperiod-which influences sexual maturation [26], and water quality [27]. Hence, the farmer is responsible for production of fish that meet a wide range of quality demands, while also optimizing criteria that are essential to profitability such as rapid growth rates, efficient feed conversion, and maximum fillet yield.

Research that combines the evaluation of rainbow trout growth performance and coinciding fillet quality attributes would provide information for fish farmers and food industry sectors that could be useful for determination of best harvest endpoints. This information would be particularly valuable for trout farmers using or planning to use innovative fish production technologies that recirculate water and optimize environmental variables. Therefore, the primary objective of this study was to characterize rainbow trout growth, processing, and fillet quality attributes as rainbow trout approached sexual maturity, while being cultured to 5 kg in freshwater recirculating systems with relatively constant water temperature averaging 13°C, constant lighting, and around-the-clock feeding. The effects of the culture conditions on the reproductive and growth axes will be described elsewhere.

Materials and Methods

Fish and fish culture systems

All-female diploid, Kamloops strain rainbow trout were purchased as eyed eggs from Troutlodge, Inc. (Sumner, WA, USA). Eyed eggs were incubated and hatched within Heath-Tecnca 8-stack incubator trays (Marisource, Tacoma, WA, USA) in a RAS maintained at a constant water temperature of 10°C. Designation of Day 1 of the growth cycle corresponded with 50% egg hatch. After hatch, water temperature in the incubation system was adjusted to approximately 13°C to acclimate the fry to culture tanks. When rainbow trout had absorbed the incubation system was adjusted to approximately 13°C to acclimate water temperature of 10°C. Designation of Day 1 of the growth cycle (Marisource, Tacoma, WA, USA) in a RAS maintained at a constant incubated and hatched within Heath-Tecnca 8-stack incubator trays as eyed eggs from Troutlodge, Inc. (Sumner, WA, USA). Eyed eggs were approached 80 kg/m², fish were randomly divided and removed from the study to reduce density to 40-50 kg/m². Due to limited space and the need to maintain numbers, rainbow trout density exceeded 80 kg/m² for two brief periods during the fingerling phase. Throughout the study, random population sampling was conducted through crowding and dip-netting.

Photoperiod

A constant 24 h photoperiod (overhead lights on) was provided throughout the study. From 0-12 mo external natural light was limited and the primary photoperiod was provided by overhead metal halide and fluorescent lights. When fish were relocated to the 11 m³ partial reuse system (approximately 12 months), constant 24 h overhead fluorescent lighting was provided. However, the greenhouse-style building in which this system was housed was made of a canvas material that filtered natural light, allowing a diurnal fluctuation in light intensity at the surface of the tank: from ~ 40-200 lux at night, depending on location, to ~ 2000 lux midday.

Feeding

Timer-controlled mechanical feeders were used to provide uniformly distributed feed events that delivered equal feed portions around-the-clock through all phases of culture. From first feeding (18 days post-hatch) to 0.13 kg, Sterner Fish Tech AS, Model 907 feeders (Ski, Norway) were used to feed once every 30 min; from 0.13-1.4 kg, Arvotec, Model T-drum 2000CE feeders (Huuotokoski, Finland) were used to feed once every 2 h; and from 1.4-5 kg, an Aquatic Eco-Systems, Auger Feeder Model FS4022 (Apopka, FL, USA) was used to feed once every 6 h. Daily feeding rates and feed size were determined using standard trout feeding charts as well as observations of feeding activity and wasted feed. Fish were fed to satiation as much as possible. Although we attempted to avoid overfeeding, it did occur on occasion, especially as the fish approached 5 kg. From first feeding to 0.13 kg, trout were fed Silver Cup Feed (Nelson & Sons Inc., Murray, UT, USA) and from 0.13 kg to the end of the study, trout were fed standard trout diets produced by Zeigler Brothers Inc. (Gardners, PA, USA). A slow sinking, extruded commercial trout growers diet, (42% protein: 16% fat) was fed for the majority of the study, with the exception of a 45:20 diet fed from 6 to 8 mo post-hatch. Daily feed amounts were recorded.

Water quality analysis

Dissolved oxygen and temperature were measured 5 times weekly using a handheld dissolved oxygen meter while trout were in the fry system and 11 m³ growout system (Table 1). When trout were

| Parameter                  | Method                                      | Units   |
|----------------------------|---------------------------------------------|---------|
| Dissolved oxygen           | Hach Model HQ10 LDO probe; Hach SC100 Universal Controller with LDO probe | mg/L    |
| Temperature                | Hach Model HQ10 LDO probe; Hach SC100 Universal Controller with LDO probe | °C      |
| Dissolved carbon dioxide   | Standard Methods 2320                       | mg/L    |
| Alkalinity                 | Standard Methods 2302                       | mg/L    |
| Total suspended solids     | Standard Methods 2560                       | mg/L    |
| Total ammonia nitrogen     | Standard Methods 4500-NH₃                   | mg/L    |
| Nitrite-nitrogen           | Standard Methods 4500-NO₂                   | mg/L    |
| Nitrate-nitrogen           | Standard Methods 4500-NO₃                   | mg/L    |
| dBO₅                       | Standard Methods 5210 5-day BOD             | mg/L    |

Table 1: Standard methods [28,29] or a handheld probe were used to measure water quality.
cultured in the six replicated RAS, SC100 Universal Controllers with Luminescent Dissolved Oxygen (LDO) probes (Hach Company, Loveland, CO, USA) displayed oxygen and temperature continuously and these values were recorded daily each morning. Water samples were collected once weekly and analyzed for dissolved carbon dioxide (CO₂), alkalinity, total ammonia nitrogen (TAN), nitrite nitrogen (NO₂⁻), nitrate nitrogen (NO₃⁻), total suspended solids (TSS), and 5-day carbonate biochemical oxygen demand (cBOD₅) according to methods described in Table 1 [28,29].

**Fish sampling and performance assessment**

During the first five months of culture, after first feeding, representative bulk weight samples were collected monthly. Thereafter, 60-100 fish were sampled monthly from the population to collect individual length and weight data. Fish were returned to culture tanks after each sampling event. Fulton’s condition factor (CF), thermal growth coefficient (TGC), and feed efficiency ratio (FER) were calculated using the following equations. Mean weight was expressed in grams, length in millimeters, and temperature as °C.

\[ CF=100,000 \times \frac{\text{Weight}}{\text{Length}^3} \]
\[ TGC=(\text{End Weight}^{1/3}-\text{Start Weight}^{1/3})/((\text{Days Between} \times \text{Mean Temp.}) \times 1000) \]
\[ FER=\text{Cumulative Feed Delivered to Fish/Fish Biomass Gain} \]

**Fish processing**

Fish were sampled bimonthly to evaluate processing and fillet attributes, from 14-24 mo, and then monthly from 24-26 mo. During each sampling event, at least 10 fish were randomly netted from the tank and humanely euthanized immediately after removal according to approved procedures. The following data were collected on individual fish: total body length; as well as total body, head-on-gutted (HOG), skin-on fillet, skinless fillet, total visceral, and total gonadal weights. These weights were used to calculate HOG, skin-on fillet, and skinless fillet yields (product mass/total body mass *100), as well as gonadosomatic index (GSI; gonad mass/total body weight *100) and viscerosomatic index (VSI; visceral mass/total body mass *100). Fillet thickness and belly-flap thickness for each fish were measured using calipers at three standardized locations along the length of the ventral midline; these locations were just cranial to the pectoral fin, just caudal to the pectoral girdle from each boneless, skinless fillet. The section was thermally processed in a microprocessor-controlled commercial grade blender (Model 51BL31; Waring Commercial; Scarsdale, NY, USA) equipped with a 5-blade kramer shear attachment at a cross speed of 127 mm/min. Values were expressed as peak force generated per gram of sample.

**Cook yield and fillet texture analysis**

A 3 by 8 cm section of the dorsal musculature was removed 2 cm caudal to the pectoral girdle from each boneless, skinless fillet. The section was thermally processed in a microprocessor-controlled smoke oven (model CVU-490, Enviro-Pak, Clackamas, OR, USA) to an internal temperature of 65.5°C. Following cooking, fillet portions were removed from the oven and cooled to room temperature. Cooked portions were weighed to calculate cook yield (cooked weight as a percent of raw weight). The precut, cooked portion was placed, with the skin-side up, in a Kramer Shear Cell. Shear force was measured using a Texture Analyzer (model TA-Hdi, Texture Technologies Corp., Loveland, CO, USA) equipped with a 5-blade kramer shear attachment at a cross speed of 127 mm/min. Values were expressed as peak force generated per gram of sample.

**Statistical analysis**

This experiment was conducted as a completely randomized design with age as an independent variable. One-way ANOVA was used to evaluate the effect of age on growth and fillet attributes using the GLM procedure of SAS system for Windows, version 9.1 [31]. Significance was defined at P<0.05. Principal component analysis (PCA) was used to create a few key variables that characterized as fully as possible the variation in the data set while reducing its dimensionality. The varimax rotation on the first two principal components was computed by FACTOR procedure of SAS system for Windows, version 9.1 [31]. Significance of principal component loadings was defined at 0.50. Linear and nonlinear models were fitted to the rotated standardized principal components (PCs) to characterize their relationships with the variable age. The nonlinear least squares method was used to fit a four-parameter logistic (4PL) model to the first principal component (PC1) scores as a function of age [32]. A segmented regression model was fitted to the second principal component (PC2) scores [33,34]. Regression analysis by PROC REG procedure and Pearson’s correlation analysis by PROC CORR procedure were used to assess the relationships of select fillet attributes, as well as GSI, with fish age [31].

**Results and Discussion**

**Fish growth**

Rainbow trout grew to a mean size of 1.4 kg by 12 mo, to 4.8 kg by 22 mo, and to 5.2 kg and 62.9 ± 0.6 cm in 26 mo (Figure 1 and Table 2). Cumulative growth from 0-26 mo followed a sigmoidal curve typical for trout as they advanced from the larval and juvenile stages to maturing adult [35]. Condition factor increased steadily from 0-12 mo and thereafter remained relatively constant at approximately 2.1 g/cm³ (Table 2). Thermal growth coefficient averaged 2.13 from first feeding to 12 mo (1.4 kg) as fish grew to 1.4 kg, 1.49 as fish grew from 1.4-4.8 kg (13-22 mo), and 0.30 as fish grew to 5.2 kg (23-26 mo) (Table 2). Growth rates declined with onset of sexual maturation, as nutrient...
utilization was refocused from somatic growth toward gonad and gamete development [9,35,36].

To the authors’ knowledge, rainbow trout growth rates (i.e., cumulative weight with age/production time) of this magnitude have not been previously reported for trout cultured to mean sizes > 2 kg [4-6,37]. The study conditions were unique in that trout of this size were cultured in freshwater recirculating systems that provided maximum control of environmental conditions (Table 3). The following culture parameters likely contributed to the rapid growth rates: constant 24 h overhead lighting, around-the-clock feeding to satiation, and near-optimal water temperatures averaging 13°C.

Survival
Cumulative rainbow trout survival was 92.9% following hatch and yolk sac absorption. After 6 months of culture in the single pass fry culture system, cumulative survival was 87.1%. From 6 to 12 months of the life cycle, while the rainbow trout were reared in six replicated RAS, mortality was minimal. Cumulative survival after 12 months of culture was 86.3%. Rainbow trout mortality from fry stage to 1-1.5 kg was slightly greater than the average mortality rate (4%) for other trout cohorts cultured on-site within the same systems [2]. Slightly increased mortality occurred from 12-22 months while a small portion of the population (330 fish) was slightly greater than the average mortality rate (4%) for other trout cohorts cultured on-site within the same systems [2]. Slightly increased mortality occurred from 12-22 months while a small portion of the population (330 fish) was held in the partial reuse system for growout to 5 kg. From months 12 to 20, 8.4% of the population was lost to mortality. Much of this mortality was related to repeated monthly handling of the same fish for length and weight assessment, as one or two mortalities typically were removed from the tank on days subsequent to each sampling event. Based on these results, frequent handling of large rainbow trout (1-4 kg) is not recommended as one or two mortalities typically were removed from the tank on days subsequent to each sampling event. Based on these results, frequent handling of large rainbow trout (1-4 kg) is not recommended in a commercial setting and production plans should be designed to minimize handling and movement of large trout. From months 20 to 26, an additional 15% of the population was lost to mortality. Some of these fish succumbed to handling stress, while others became egg laden causing them to lose equilibrium in the water column.

Feed efficiency
Feed efficiency ratio (cumulative feed delivered to the fish/biomass gain) ranged from 1.0-2.0 for the majority of the growth cycle and averaged 1.36 from first feeding to 22 mo; however, FER increased from

| System (fish size)         | Temperature ºC | Dissolved O2 mg/L | Dissolved CO2 mg/L | Alkalinity mg/L as CaCO3 | TAN mg/L | NO3-N mg/L | NO2-N mg/L | TSS mg/L | cBOD5 mg/L |
|----------------------------|----------------|-------------------|--------------------|--------------------------|----------|------------|------------|----------|------------|
| Single-pass (0.5-130 g)    | 12.9 ± 0.1     | 9.5 ± 0.2         | 18.4 ± 0.3         | 266 ± 3                  | 0.31 ± 0.01 | 3 ± 0       | 0.01 ± 0.00 | 4.5 ± 0.3 | 1 ± 0      |
| Fully-recycle H            | 13.2 ± 0.0     | 10.0 ± 0.0        | 9.3 ± 0.1          | 228 ± 1                  | 0.29 ± 0.00 | 12 ± 0      | 0.04 ± 0.01 | 2.7 ± 0.1 | 6 ± 1      |
| Fully-recycle L* (130-1400 g) | 13.2 ± 0.1   | 10.0 ± 0.1        | 9.0 ± 0.4          | 214 ± 1                  | 0.48 ± 0.05 | 70 ± 4      | 0.10 ± 0.00 | 6.4 ± 1.2 | 2 ± 0      |
| Partial-reuse (1400-5000 g) | 12.9 ± 0.0    | 11.0 ± 0.1        | 13.6 ± 0.3         | 263 ± 1                  | 0.24 ± 0.01 | 2 ± 0       | 0.01 ± 0.00 | 1.1 ± 0.1 | 0 ± 0      |

*Rainbow trout were raised in both the high (H) and low (L) water flushing treatments.

Table 3: Water quality (mean ± standard error) in the culture tanks during each phase of the 26-month growth period.
2.23 to >20.0 from 23-25 mo (Figure 2 and Table 2). Feed efficiency has been reported to decrease with rainbow trout age [24,38]; but has not been thoroughly evaluated for maturing rainbow trout. During the present study, increased FER corresponded with a rapid decline in TGC and an increase in egg development, as evidenced by increasing GSI. The dramatic increase in FER can be explained in part by the reallocation of energy resources from growth to reproduction; however, it was likely that the primary driver for the increase in FER at 25 mo to >20.0 was a substantial decline in feed consumption and subsequently significant amounts of wasted feed. Between months 25 and 26, FER declined from >20.0 to 0.71 (Figure 2). The substantial drop in FER was due to recognition by the fish culturist that maturing trout were not accepting feed. Feed ration was reduced by more than 50% from months 25-26, in order to rebalance FER (Figure 2). Although, the FER from months 23-25 was far from optimal, these results provided critical information for fish farmers with prospects of raising large rainbow trout. Increased FER or simply a reduction in feeding would negatively impact a producer’s profit margin because feed represents a large fraction of the total cost of commercial aquaculture; however, harvesting rainbow trout prior to 23 mo could possibly evade these problems.

**Gonadosomatic index**

GSI assessment indicated that rapid egg growth occurred primarily from 20-26 mo (Figure 3), coincident with and likely affecting changes in many of the aforementioned trout performance traits (growth, TGC, FER) and subsequent fillet attributes. GSI was positively correlated with age (r=0.73). In addition, the decline in VSI (excluding gonad weight from the viscera weight) occurred in unison with increasing GSI (Figure 3) from 20-24 mo, consistent with lipid mobilization from the viscera to support egg production. Although VSI stabilized after 24 mo, GSI continued to increase suggesting that nutrients were derived from other sources, feed or possibly muscle lipid and protein. The GSI for fish in this study was lower than typically observed. Female rainbow trout can develop ovaries that account for over 20% of total body weight prior to ovulation [15], but fish in this study reached a maximum GSI of only 7.0% at 26 mo. A lower GSI could indicate that a lower fraction of whole body nutrient and energy resources were expended for gonadal growth than ordinarily occurs in maturing female rainbow trout. In addition, larger rainbow trout size, greater than that of previously reported comparisons to gonad mass, could also have been related to reduced GSI.

**Fillet yield**

Head-on-gutted yield ranged from 86.2-87.6% from 14 to 24 mo, but declined to 82.8% by 26 mo (Figure 4). Skin-on fillet (butterfly) yield increased from 52.9% at 14 mo to a maximum of 56.8% at 20 mo and then declined to 52.5% by 26 mo (Figure 4). Skinless fillet yield followed a similar trend, beginning at 52.0% at 20 mo, and declining to 48.3% by 26 mo (Figure 4). Separable muscle (skinless fillet) continued to increase through 25 mo; however, when expressed as a percent of whole fish weight, separable muscle peaked at the 20 to 22 mo interval (Figure 5). Percent separable muscle declined from 51.9 ± 0.5% at 22 mo to 48.3 ± 1.2% at 26 mo (Figure 5). Critical changes in final product yield were also related to the timing of egg growth and reproductive development. Previous studies have also reported a maximum fillet yield at roughly 20 mo of age and a corresponding increase in GSI thereafter [6,9].

Small differences in fillet yield can have a substantial economic impact on production and processing economics [5,39]; therefore, prediction and selection of a harvest endpoint to obtain peak fillet yield is important. Based on yield data alone, an optimal harvest endpoint of 20-22 mo (mean weight=3.8-4.8 kg) could be targeted under similar conditions and depending on marketing objectives for the final product.

**Fillet belly flap thickness**

Fillet thickness increased from approximately 2.7 ± 0.1 cm at 14 months.
respectively (Figure 7). The timing of egg growth and development was at 24 mo, but then decreased to 13.5% and 11.7% at 25 and 26 mo, respectively (Figure 7). The timing of changes in fillet lipid content could influence the selection of harvest endpoint because intramuscular lipid represents an important quality parameter that significantly impacts the palatability, flavor, and nutritional value (fatty acid content) of the product [41-43].

Aside from reproductive maturation, many factors can influence the fillet lipid content of cultured salmonoids, including lipid content of the feed [41,44,45], age/size of the fish [46,47]; and genetic heritability [16,48]. Crude fat levels in rainbow trout fillets are typically lower than the peak levels reported here. Pan-sized rainbow trout raised at Clear Springs Foods Inc. (Buhl, ID, USA), the largest rainbow trout producer in the U.S., have an average fillet fat content of 12-13% (personal communication Randy MacMillan, Clear Springs Foods Inc.). Another source [46] reported fillet fat content for pan-sized rainbow trout (0.3 kg) to range from 4-7%, and noted a correlation with fish size. In addition, muscle lipid levels of approximately 10% have been reported for 3.1 kg rainbow trout [6]. The present study demonstrated a relationship of increasing fillet fat content with trout size/age, at least prior to changes related to reproductive development. For example, fillet lipid was 11.9% when compositional analyses were conducted at 14 mo (mean weight=1.8 kg), and increased to 18.1% by 24 mo (mean weight=5 kg). It is noteworthy that these large rainbow trout contained fillet fat levels that were similar to farm-raised Atlantic salmon. For example, an average fillet lipid level of 15% was reported for 4 kg salmon harvested in Norway [49]. Large rainbow trout fillets with lipid and fatty acid composition comparable to Atlantic salmon could be perceived as a unique product with appealing marketing traits, depending on the effect of fat content on other quality attributes such as texture or fillet firmness.

Fillet moisture declined from 67.8 to 63.2% from 14-24 mo, but increased to 66.3% over the final two months of the study (Figure 7). Overall, a negative correlation (P<0.02) was observed between age and fillet moisture (r=-0.27). Likewise, cooked fillet texture declined from 460 to 290 g/g from 14 to 24 mo, but increased to 577 g/g over the final two months of the study, indicating an increase in fillet firmness (Figure 7). Increased cooked fillet moisture and firmness coincided with decreasing lipid from 18.1 to 11.7%.

mo to 4.4 ± 0.2 cm at 26 mo (Figure 6). Fillet thickness was positively correlated with age (r=0.91). Specific data for optimal fillet thickness as it relates to endpoint cooking rate and temperature is lacking for large rainbow trout. Endpoint cooking temperature has been shown to impact quality characteristics such as cook yield and moisture for rainbow trout [40]. The fillet thickness data obtained during this study could provide a reference for trout producers and food industry stakeholders in selecting a harvest endpoint that meets the qualifications of the desired end-product.

Belly flap thickness increased in unison with fillet thickness from 1.4 ± 0.1 cm at 14 mo to 2.1 ± 0.1 cm at 26 mo (Figure 6). Thus, belly flap thickness was also positively correlated with age (r=0.76). An increase in abdominal wall (belly flap) thickness of rainbow trout was also positively correlated with age and gonadal development during a previous study [6]. The belly flap of fish is generally trimmed from the fillet because of the adiposity of the tissue. Therefore, increased belly flap thickness generally correlates with increased carcass loss and a decrease in product yield.

**Fillet quality**

Fillet crude fat content increased from 11.9% at 14 mo to 18.1% at 24 mo, but then decreased to 13.5% and 11.7% at 25 and 26 mo, respectively (Figure 7). The timing of egg growth and development was a critical determinant of changes in fillet composition. In the context of an energetically demanding physiological process, such as egg growth and development, changes in adipose tissue are a reflection of the mobilization of stored energy (lipid) to support the development of eggs [6,9,14,17,18]. The timing of changes in fillet lipid content could influence the selection of harvest endpoint because intramuscular lipid represents an important quality parameter that significantly impacts the palatability, flavor, and nutritional value (fatty acid content) of the product [41-43].
Changes in muscle composition during growth and development did not affect cook yield. Cook yield peaked at 22 mo at 89.3 ± 0.7% and then declined slightly thereafter. Cook yield was positively correlated with age (r=0.57). Typically, greater cook yield indicates increased water and fat retention. It is expected that increased fillet thickness (Figure 6) would slow cooking rate and contribute to greater cook yield. Additionally, increased fat content may have reduced cook yield and firmness after 22 months, as the rainbow trout matured. It appears that raw fillets, when cooked, were not able to retain the increasing intramuscular fat (15 to 18%), as fish aged from 18 to 24 months. Fillet fat content was negatively correlated with cook yield (r=−0.35, P=0.0005).

In addition to the aforementioned quality traits, an important parameter to the processing industry and consumers for salmon and possibly for large rainbow trout fillets is bright red/orange fillet color [4,50]. During the present study, rainbow trout were not fed a diet containing carotenoids and therefore had pale, whitish fillet color. Other studies found that reproductive maturity resulted in a significantly lower red fillet color score in large rainbow trout [4]; thus color is another important quality consideration in determining an optimal harvest endpoint. Further investigation into the relationship of maturation, age, and fillet color is needed for rainbow trout cultured to 4-5 kg.

Principal components analysis

In the previous sections, a description of multiple growth and fillet quality responses to age were provided. While this approach was useful in understanding changes in a specific response variable with age, it did not provide a simplified model to analyze and predict the timing of best harvest endpoint for large rainbow trout while considering many variables. Principal components analyses reduced the dimensionality of the data and therefore facilitated identification of best harvest endpoints in terms of overall growth performance and fillet quality.

Two principal components were retained. Principal component 1 grouped length, weight, fillet thickness, belly flap thickness, and cook yield; while principal component 2 grouped moisture content, fat content, and cooked fillet firmness. Examination of these variables indicated that PC1 measured size and "Growth" of the fish, with larger fish receiving larger scores; whereas PC2 described the "Quality" of the fillet.

Interestingly, a scatter plot of PC1 scores versus age suggested a sigmoidal curve (Figure 8) with growth clearly exhibiting a positive association with age, thus corroborating the interpretation of PC1 as measuring fish growth. Based on the 4PL model, the estimated regression function is displayed in Figure 8 along with approximate 95% confidence limits on the conditional expected PC1 score. Figure 8 indicates that harvesting fish at 20-22 mo would coincide with the peak of the linear stage of growth, around the point of inflection on the sigmoidal curve [51].

Fillets that were firmer, with less fat, and higher in moisture received higher PC2 scores. The scatter plot of PC2 scores versus age fitted with a segmented regression model is shown in Figure 9 along with the 95% Working Hotelling confidence band for the regression function. A lower PC2 score should not necessarily be interpreted as a lower quality fillet. More research is needed to determine if the fillet fat composition measured in 3.8-4.8 kg rainbow trout during the present study is acceptable to consumers; and if necessary, methods to achieve optimal fat composition and texture, such as optimization of feeding regime or diet composition, should be evaluated. In the meantime, the plot of PC2 scores with age provides a general basis for expected changes in fillet quality for maturing rainbow trout (Figure 9).

Conclusions

This research provides rainbow trout growth performance and
fillet quality results that can be referenced for the development of recirculating system production plans and for selection of harvest endpoints that balance the requirements of fish farmers and food industry sectors. The results indicated that a commercially available strain of all-female rainbow trout can be cultured in freshwater recirculating systems at approximately 13°C, under constant lighting, and with around-the-clock feeding to a mean size of 5 kg in 24 mo. Principle components analysis indicated that a peak in growth performance variables, as a whole, occurred between 20-22 months. If growth performance was the only consideration for determination of best harvest endpoint, then it is reasonable to estimate that harvesting large rainbow trout at 20-22 mo of age or 3.8-4.8 kg in size might be optimal. However, selection of harvest endpoint may vary depending upon the end-product quality desired by value-chain sectors or relative to the form or preparation of the final product. More research is needed to interpret quality preferences for large rainbow trout fillets, particularly with increased fat content (up to 18%), as was measured needed to interpret quality preferences for large rainbow trout fillets, particularly with increased fat content (up to 18%), as was measured during this study. Overall, the corresponding growth and fillet quality data provided by this research serve as an initial reference for industry, when culturing large rainbow trout under similar conditions.

Acknowledgements

Special thanks to Susan Slider for technical assistance, as well as data collection and organization. Thanks also to the Freshwater Institute water chemistry staff. This research was supported by the USDA Agricultural Research Service under Agreement No. 59-1930-5-010 and 59-1930-0-046. All experimental protocols and methods were in compliance with the Animal Welfare Act (SCFR) requirements and were approved by the Freshwater Institute’s Institutional Animal Care and Use Committee. Use of trade names does not imply endorsement by the U.S. Government.

References

1. Summerfelt ST, Davidson JW, Waldrop TB, Tsukuda SM, Bebak-Williams J (2004) A Partial-Reuse System For Coldwater Aquaculture. Aquacult Eng 31: 157-181.
2. Summerfelt S, Davidson J, Good C, Waldrop T (2010) Rainbow Trout Attain Good Growth, Health in Tank-Based Recirculating Systems. Global Aquaculture Advocate, March/April 2010.
3. Dalsgaard J, Lund I, Thorarinsdottir R, Drengstig A, Arvonen K, et al. (2013) Farming Different Species in RAS in NORDIC Countries: Current Status and Future Perspectives. Aquacult Eng 53: 2-13.
4. Paaver T, Gross R, Ilves P (2004) Growth Rate, Maturation Level and Flesh Quality of Three Strains of Large Rainbow Trout (Oncorhynchus mykiss) Reared in Estonia. Aquacult Int 12: 33-45.
5. Bugjen J, Lefevre F, Cardinal M, Uyanik A, Davenel A, et al. (2010) Flesh Quality in Large Rainbow Trout With High or Low Fillet Yield. J Muscle Foods 21: 702-721.
6. Aussanasuwannakul A, Kenney PB, Weber GM, Yao J, Slider SD, et al. (2011) Effect of Sexual Maturation on Growth, Fillet Composition, and Texture of Female Rainbow Trout (Oncorhynchus mykiss) on a High Nutritional Plane. Aquaculture 317: 79-88.
7. Aussanasuwannakul A, Weber GM, Salem M, Yao J, Slider SD, et al. (2012) Effect of Sexual Maturation on Thermal Stability, Viscoelastic Properties, and Texture of Female Rainbow Trout, Oncorhynchus mykiss, Fillets. J Food Sci 71: 577-583.
8. Cleveland BM, Kenney PB, Manor ML, Weber GM (2012) Effects of Feeding Ration and Sexual Maturation on Carcass and Fillet Characteristics and Indices of Protein Degradation in Rainbow Trout (Oncorhynchus mykiss). Aquaculture 338-341: 228-236.
9. Manor ML, Weber GM, Salem M, Yao J, Aussanasuwannakul A, et al. (2012) Effect of Sexual Maturation and Triploidy on Chemical Composition and Fatty Acid Content of Energy Stores in Female Rainbow Trout, Oncorhynchus mykiss. Aquaculture 364-365: 312-321.
10. Manor ML, Weber GM, Cleveland BM, Kenney PB (2014) Effects of Feeding Level and Sexual Maturation on Fatty Acid Composition of Energy Stores in Diploid and Triploid Rainbow Trout, Oncorhynchus mykiss. Aquaculture 418-419: 17-25.
11. Roque d’Orbcastel E, Ruyet JP, Le Bayon N, Blancheton, JP (2009) Comparative Growth and Welfare in Rainbow Trout Reared in Recirculating and Flow Through Systems. Aquacult Eng 40: 79-86.
12. Davidson J, Good C, Welsh C, Brazl B, Snevik K, et al. (2009) Heavy Metal and Waste Metabolite Accumulation and Their Potential Effect on Rainbow Trout Performance in a Replicated Water Reuse System Operated at Low or High System Flushing Rates. Aquacult Eng 41: 136-145.
13. Davidson J, Good C, Welsh C, Summerfelt S (2011) The Effects of Ozone and Water Exchange Rates on Water Quality and Rainbow Trout Oncorhynchus mykiss Performance in Replicated Water Recirculating Systems. Aquacult Eng 44: 80-96.
14. Shearer KD (1994) Factors Affecting the Proximate Composition of Cultured Fishes with Emphasis on Salmonids. Aquaculture 119: 63-88.
15. Tyler CR, Sumpter JP, Withamms PR (1990) The Dynamics of Oocyte Growth During Vitellogenesis in the Rainbow Trout (Oncorhynchus mykiss). Biol Reprod 43: 202-209.
16. Kause A, Ritola O, Paananen T, Mantsaari E, Eskelinen U (2002) Coupling Body Weight and Its Composition: A Qualitative Genetic Analysis in Rainbow Trout. Aquacult Nutr 21: 65-78.
17. Nassour I, Leger CL (1989) Deposition and Mobilization of Body Fat During Sexual Maturation in Female Trout (Salmo gairdneri Richardson). Aquat Living Resour 2: 153-159.
18. Jonsson N, Jonsson B, Hansen LP (1997) Changes in Proximate Composition and Estimates of Energetic Costs During Upstream Migration and Spawning in Atlantic Salmon Salmo salar. J Anim Ecol 66: 425-436.
19. Rasmussen RS (2001) Quality of Farmed Salmonids with Emphasis on Proximate Composition, Yield and Sensory Characteristics. Aquac Res 32: 767-786.
20. Salem M, Kenney PB, Rexroad III CE, J Yao (2006) Molecular Characterization of Muscle Atrophy and Proteolysis Associated with Spawning in Rainbow Trout. Comp Biochem Physiol Part D 1: 227-237.
21. Setala J, Saami K, Honkanen A (2000) The Quality Perceptions of Rainbow Trout Defined by Different Fish Market Sectors. In: Paper Presented at the Tenth Biennial Conference of The International Institute of Fisheries Economics And Trade (IIFET), Corvallis, Oregon.
22. Einen O, Skrede G (1998) Quality Characteristics in Raw and Smoked Fillets of Atlantic Salmon, Salmo salar, Fed High Energy Diets. Aquacult Nutr 4: 99-108.
23. Morkramer S, Horsbig-Schwark G, Langholz HI (1985) Comparison of Different European Rainbow Trout Populations Under Intensive Production Conditions. Aquaculture 44: 303-320.
24. Azevedo PA, Leeson S, Cho CY, Bureau DP (2004) Growth and Feed Utilization of Large Size Rainbow Trout (Oncorhynchus Mykiss) and Atlantic Salmon (Salmo salar). Aquac Nutr 10: 401-411.
25. Rasmussen RS, Ostenfeld TH (2000) Effect of Growth Rate on Quality Traits and Feed Utilization of Rainbow Trout (Oncorhynchus mykiss) and Brook Trout (Salvelinus fontinalis). Aquaculture 184: 327-337.
26. Nordgarden U, Omsrud R, Hansen T, Hemre GI (2003) Seasonal Changes in Selected Muscle Quality Parameters in Atlantic Salmon Reared Under Natural and Continuous Light. Aquacult Nutr 9: 161-168.
27. Guttman L, Van Rijn J (2008) Identification Of Conditions Underlying Production of Geosmin and 2-Methylisoborneol in a Recirculating System. Aquaculture 279: 85-91.
28. American Public Health Association (APHA) (2005) Standard Methods for the Examination of Water and Wastewater, (21stedn). Washington, DC.
29. Hach Company (2003) DR/4000 Spectrophotometer Procedures Manual, (11thedn). USA.
30. Association of Official Analytical Chemists (1990) Official Methods of Analysis, (15thedn). Association of Official Analytical Chemists, Washington, DC.
31. SAS Institute Inc. (2004) SAS Users Guide: Basics Version 6.06. Cary, N.C.: Statistical Analysis System Institute Inc.
32. Pinheiro JC, Bates DM (2000) Mixed-Effects Models in S and S-PLUS. Springer-Verlag, New York, NY.
33. Muggeo V (2000) Estimating Regression Models with Unknown Break-Points. Statist Med 22: 3055-3071.
34. Muggeo V (2008) Segmented: An R Package to Fit Regression Models with Broken-Line Relationships. R News 8/1:20-25.
35. Westers H (2001) Production. In: Fish Hatchery Management (2nd edn).
36. Piper RG, Mcelwain IB, Orme LE, Mccraren JP, Fowler LG, et al. (1982) Fish Hatchery Management. United States Department of the Interior Fish and Wildlife Service, Washington DC.
37. Memis D, Gun H (2004) Effects of Different Diets on the Growth Performance, Gonad Development, and Body Composition at First Sexual Maturity of Rainbow Trout. Turk J Vet Anim Sci 28: 315-322.
38. Storebakken T, Austreng E (1987) Ration Levels for Salmonids: II. Growth, Feed Intake, Protein Digestibility, Body Composition, and Feed Conversion in Rainbow Trout Weighing 0.5-1.0 Kg. Aquaculture 60: 207-221.
39. Rora AMB, Morkore T, Einen O (2001) Primary Processing (Evisceration and Filleting). In: Kestin, SC, Warriss, PD (Eds.) Farmed Fish Quality, Blackwell Science, Oxford 249-260.
40. Johansson L, Berglund L, Rudéus H (1992) Cooking Losses of Rainbow Trout –Effects of Final Internal Temperature, Fish Weight, Heating Method and Oven Temperature. In: Eating Quality of Rainbow Trout, Øphel Thesis, Uppsala University, Uppsala, Sweden.
41. Johansson L, Kiessling A, Kiessling KH, Berglund L (2000) Effects of Altered Ration Levels on Sensory Characteristics, Lipid Content, and Fatty Acid Composition of Rainbow Trout (Oncorhynchus mykiss). Food Qual Prefer 11: 247-254.
42. Sargent JR, Bell JG, Mcghee F, Mcvoy J, Webster, JL (2001) The Nutritional Value of Fish. In: Kestin, SC, Warriss, PD (Eds.) Farmed Fish Quality, Blackwell Science, Oxford 3-11.
43. Vemuri M, Kelley DS (2008) The Effects of Dietary Fatty Acids on Lipid Metabolism. In: Chow, CK (Ed.) Fatty Acids in Foods and Their Health Implications (3rd edn), CRC Press Taylor & Francis Group, Florida 591-630.
44. Jobling M, Koskela J, Savolainen R (1998) Influence of Dietary Fat Level and Increased Adiposity on Growth and Fat Deposition in Rainbow Trout, Oncorhynchus mykiss (Walbaum). Aquacult Res 29: 601-607.
45. Chaiyapechara S, Liu KKM, Barrows FT, Hardy RW, Dong FM (2003) Proximate Composition, Lipid Oxidation, and Sensory Characteristics of Fillets From Rainbow Trout Oncorhynchus mykiss Fed Diets Containing 10% to 30% Lipid. J World Aquacult Soc 34: 266-288.
46. Aisted N, Due T, Hjermløy N (1995) Practical Experience with High Energy Diets, FCR, Growth, and Quality. J Appl Ichthyol 11: 329-335.
47. Kiessling A, Pickova J, Johansson L, Aasgard T, Storebakken T, et al. (2001) Changes in Fatty Acid Composition in Muscle and Adipose Tissue of Farmed Rainbow Trout (Oncorhynchus mykiss) in Relation to Ration and Age. Food Chem 73: 271-284.
48. Quillet E, Guillou WL, Aubin J, Fauconneau B (2005) Two-Way Selection for Muscle Lipid Content in Pan-Size Rainbow Trout (Oncorhynchus mykiss). Aquaculture 245: 49-61.
49. Fjellanger K, Obach A, Rosenlund G (2001) Proximate Analysis of Fish with Special Emphasis on Fat. In: Kestin, SC, Warriss, PD (Eds.) Farmed Fish Quality, Blackwell Science, Oxford 307-317.
50. Aftnes F, Guttormsen G, Steine G, Kolstad K (2006) Consumers' Willingness to Pay for The Color of Salmon: A Choice Experiment with Real Economic Incentives. Am J Agr Econ 88: 1050-1061.
51. Gerrard D, Grant A (2006) “Chapter 7: Adipose Tissue Growth and Development.” Principles of Animal Growth and Development. Kendakk/Hunt Publishing Company 119-136.
52. Ritz C, Streibig JC (2008) Nonlinear Regression with R. Springer-Verlag, New York, NY.