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ARAŞTIRMA MAKALESİ **RESEARCH PAPER**

Effect of Salinity on Yolk Utilization and Growth of Brook Trout Alevins (*Salvelinus fontinalis*)

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Abstract:
The production of freshwater fish in brackish water is well recognized as an urgent need to provide animal protein in areas where the availability of fresh water is limited. This study compared the yolk utilization and growth rate of brook trout alevins (*Salvelinus fontinalis*) reared in brackish water (4.0‰ and 8.0‰) and fresh water (0‰). The alevins were stocked in brackish and fresh waters during their yolk absorption period and intermittently sampled over 35 days. The yolk utilization efficiency was assessed by yolk conversion efficiency (*YCE*) and yolk consumption rate (*YCR*) while growth rates (mg day^{-1} and mm day^{-1}) were determined by comparing slope values estimated by the linear regression function. These parameters appeared to increase with increasing salinity, but differences between different treatments were not significant except for *YCE*. The highest mean value of *YCE* was estimated for 8.0‰ salinity treatment which was significantly different from the other two salinity treatments. These findings should assist the process of understanding the potential for rearing brook trout in brackish water and to optimize their growth rate.

Keywords: Aquaculture, brackish water, food production, yolk utilization, salinity, Salmonidae.

Tuzluluğun Kaynak Alabalığı (*Salvelinus fontinalis*) Alevinin Büyümesine ve Besin Kesesi Tüketimine Olan Etkisi

Öz: Tatlı su balıklarının acı sularda üretilmesi, tatlı suyun kullanılabilirliğinin sınırlı olduğu alanlarda hayvansal protein temin etme yöntemi olarak kabul edilmektedir. Bu çalışmada acı su (‰4 ve ‰8) ve tatlı suda (‰0) yetiştirilen kaynak alabalıklarının (*Salvelinus fontinalis*) besin kesesi tüketimi ve büyüme oranları karşılaştırılmıştır. Alevinler besin kesesi tüketim aşamasında tatlı su ve acı suda stoklanmış ve 35 gün boyunca balıklar örneklenmiştir. Besin kesesi tüketim verimliliği besin kesesi dönüşüm etkinliği (*YCE*) ve besin kesesi tüketim oranı (*YCR*) hesaplanırken büyüme oranı ise (mg day^{-1} ve mm day^{-1}) lineer regresyon modeli ile oluşturulan eğri değerleri ile belirlenmiştir. Bu parametreler tuzluluğun artmasına paralel olarak kısmen yükselmiş fakat *YCE* değeri hariç farklı tuzluluk değerindeki denemeler arasında istatistiksel bir farklılık tespit edilememiştir. En yüksek ortalama *YCE* değeri ‰8'lik tuzlulukta yetiştirilen grupta belirlenmiş ve bu değerin istatistiksel olarak diğer tuzluluk grubundakilerden farklı olduğu tespit edilmiştir. Bu bulgular, kaynak alabalıklarının acı sularda yetiştirilme potansiyelinin değerlendirilmesine ve büyüme oranlarının optimize edilmesine katkı sağlayacaktır.

Anahtar sözcükler: Akuakültür, acı su, gıda üretimi, besin kesesi tüketimi, tuzluluk, Salmonidae.

INTRODUCTION

The construction of fish farms in areas where fresh water resources are limited has been a challenge for many years. Yet, the production of freshwater fish in brackish water is well recognized as an urgent need to provide animal protein in areas where the availability of fresh water is confined due to factors such as urbanization, industrialization and agricultural activities (Loya et al., 1969; Fridman et al., 2012). Freshwater fish has a blood osmolality (solute concentration) higher than surrounding water and hence, spend a considerable amount of energy upon ion-regulation to prevent diffusive loss of ions (Brix, 2008). At the early stages of development, larval fish generally lack gill filaments, and has no osmoregulatory system as adult fish (Opstad, 2003). Ionic regulations are therefore accomplished within integument cells through Na^+/K^+ ion-transport pumps until gills and intestinal osmoregulatory systems are fully developed (Rombough, 2007).

The anadromous populations of wild brook trout (*Salvelinus fontinalis*) generally migrate from fresh water to coastal seas and stay there for 2–4 months, providing that there is access to the sea i.e. northeastern North America populations (McCormick et al., 1984). However, most of the *S. fontinalis* populations are restricted to fresh water and do not migrate between fresh and sea waters (cited in McCormick et al., 1984). According to McCormick et al. (1984), 4–5 times high growth rate of *S. fontinalis* was observed in sea ranching than freshwater populations.

In this study, *S. fontinalis* alevins were reared in varied treatments of fresh and brackish waters during their yolk absorption period to assess their comparative ability to rear in the brackish water. The findings of this study will help optimize the growth of *S. fontinalis* at their earlier life stages.

MATERIAL and METHOD

The desired salinity for experimental treatments was obtained by diluting the same fresh water used for the controls with different proportions of sea water (Yıldırım et al. 2008). Using this approach, we used three different salinity treatments (0.0‰, 4.0‰ and 8.0‰), each with three replicates. Each treatment had 480 newly emerged *S. fontinalis* alevins (distributed equally into three replicates) from the Surmene Faculty of Marine Sciences, Trabzon. Alevins for each treatment were separately stocked into a ~16-liter fiberglass aquarium. Each aquarium was constantly supplied with recirculating water and oxygen at saturation was supplied by continuous air bubbling.

Up to 12 alevins were taken at every 7-day intervals from each treatment for the determination of growth and yolk utilization efficiency. The total length of sampled alevins was measured to the nearest 0.1 mm. They were then preserved in 10% formalin for three weeks. The preserved alevins were then dissected to separate the yolk-sac from its body and both were separately dried in the oven for each individual sample (Ecocell Drying Oven, MMM Medcenter, Germany) at 60° C for 48 h. The dried alevin body and yolk-sac were finally weighted individually (to nearest 0.01 mg).

The survival rate, growth and yolk utilization efficiency of *S. fontinalis* alevins were determined using the following equations:

i. Survival rate (%):

$$Survival\ rate\ (\%) = N_f / N_i \times 100$$

where N_f = number of alevins at the end, N_i= number of alevins initial stocked.

ii. Growth rate: The relationships between alevins growth and time was best described by the linear regression function (r^2>0.95). The slope values estimated with the linear regression function were used to determine growth rates (weight, mg day^{-1} and length, mm day^{-1}):

$$y = a - \rho t$$

where y is alevins wet weight, dry weights or length at time t, a is the intercept and ρ is the slope. The ρ can be ρ_w, ρ_d and ρ_l for alevins wet and dry weights, and length respectively.

iii. Yolk conversion efficiency:

$$YCE = (W_f - W_o) / (Y_o - Y_f)$$

iv. Yolk consumption rate (mg/day):

$$YCR = (Y_o - Y_f) / t$$

where W_0: initial alevin dry weight, W_f: final alevin dry weight, Y_0: initial yolk-sac dry weight, Y_f: final yolk-sac dry weight, t is a number of days. *YCE* and *YCR* were calculated using dry weight instead of total wet weight since alevins during their yolk-sac absorption phase absorb yolk-sac and synthesize water in their tissue. Thus, it is suggested to use dry weight during the calculation of *YCE* and *YCR* (Hansen, 1985).

The values obtained with these equations were compared using One-way ANOVA to test for significant differences.

RESULTS and DISCUSSION

The water contents of alevins body did not change greatly throughout the study and were found to be significantly similar (P >0.05). Percentage survival rate decreased with increasing salinity (Table 1). The mean (± SE) dry and wet body weights, and length of alevins at time t are shown in Figure 1 along with curve lines provided by the linear regression function. The growth rate of *S. fontinalis* alevins increased with increasing salinity and the highest value of regression slope was obtained in 8.0‰ treatment revealed a higher growth rate. The yolk utilization in *S. fontinalis* alevins slightly increased with increasing salinity but differences between different treatments were not significant except for *YCE* (Table 2). The highest mean value

of *YCE* was estimated for the 8.0‰ salinity group which was significantly different from the other treatments (0% and 4% salinities).

Table 1. Total survival rate (%) and mean (± SE) water content (%) of brook trout alevins (*Salvelinus fontinalis*) in fresh and brackish waters for 35 days.

Days post-hatch	Survival rate (%)			Yolk-sac			Alevins		
	Fresh water	Brackish water		Fresh water	Brackish water		Fresh water	Brackish water	
	0.0‰	4.0‰	8.0‰	0.0‰	4.0‰	8.0‰	0.0‰	4.0‰	8.0‰
0	100	100	84.0	36.7 ± 0.1	36.7 ± 0.7	36.7 ± 0.4	81.7 ± 0.3	81.7 ± 0.7	81.7 ± 0.6
7	98.6	100	84.3	44.0 ± 0.6	37.0 ± 0.7	38.0 ± 0.7	83.2 ± 0.7	83.2 ± 0.5	81.9 ± 0.3
14	98.0	96.2	91.5	42.4 ± 0.9	45.3 ± 0.6	44.0 ± 0.5	82.8 ± 0.9	83.7 ± 0.3	82.6 ± 0.1
21	96.9	96.9	93.8	42.2 ± 0.8	32.6 ± 1.0	40.9 ± 0.6	81.3 ± 0.6	83.4 ± 0.7	82.8 ± 0.4
28	97.9	95.7	96.7	57.1 ± 1.2	48.3 ± 0.9	46.0 ± 0.8	84.3 ± 04	84.1 ± 0.1	82.4 ± 0.7
35	100	100	84.0	49.6 ± 1.0	47.8 ± 0.8	56.1 ± 1.5	85.1 ± 1.0	85.1 ± 0.9	85.5 ± 0.5

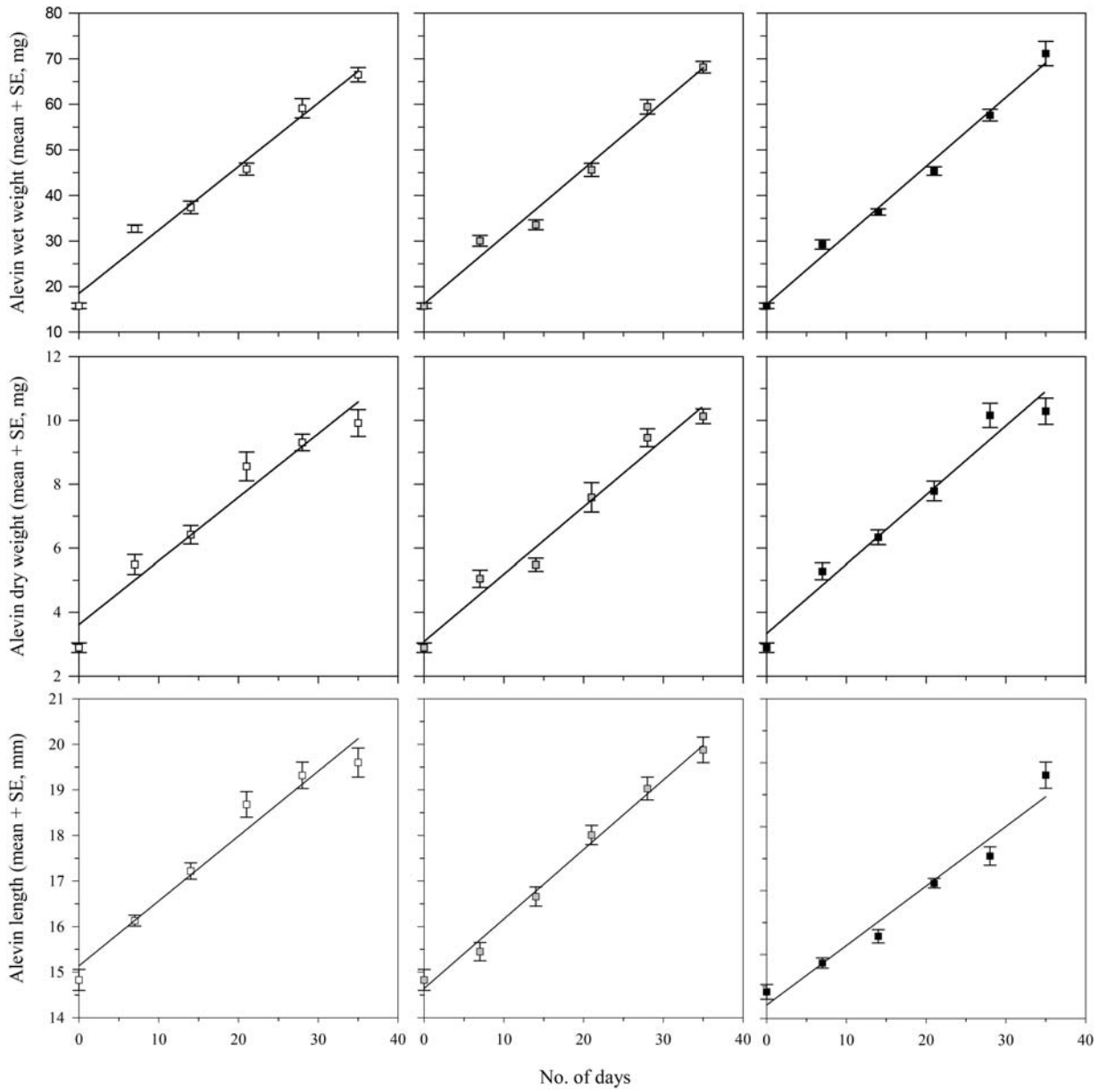

Fig. 1. Growth of brook trout alevins (*Salvelinus fontinalis*) reared in fresh water (□) and brackish water: 4.0‰ (▒) and 8.0‰ (■) salinities.

The present study supports the advantage of rearing *S. fontinalis* alevins in brackish water. The linear relationships between *S. fontinalis* alevins and time steps when subsamples were removed for analysis showed that the slope increased with increasing salinity and the highest growth of *S. fontinalis* alevins were observed in 8.0‰ salinity. These findings are in line with earlier investigations (Watanabe et al., 1989; Yıldırım et al. 2008; Fridman et al., 2012). A higher growth performance of killifish fry *Aphanius chantrei* was obtained in 4‰ salinity than in fresh water (Yıldırım et al. 2008). However, their growth tends to decrease with a further increment in water salinity (e.g., 8‰ and 14‰).

The acclimatization of *S. fontinalis* alevins to 4‰ salinity occurs more efficiently than in 8‰. The 8‰ salinity treatment resulted in higher mortality during the first two weeks (Table 1). This might suggest that the high osmoregulatory challenge faced by *S. fontinalis* alevins in 8‰ treatment during the first two weeks resulted in high mortality. Also, the overall mortality of *S. fontinalis* alevins increased with increasing salinity. Consequently, the growth tends to be higher if the alevin survives the high salinity conditions. According to Fridman et al. (2012) mortality rate of Nile tilapia *Oreochromis niloticus* alevins tends to increase with increasing salinity from 0‰ to 25‰.

In conclusion, based on *YCE* and *YCR*, higher growth rates of *S. fontinalis* alevins were observed in 4.0‰ and 8.0‰ salinities than fresh water. The results of this study can facilitate hatchery management of *S. fontinalis* to help determine the optimal way to culture larger resultant alevins during and just after the yolk utilization phase.

Table 2. Yolk utilization and growth rates of brook trout (*Salvelinus fontinalis*) alevins reared in fresh and brackish water for 35 days. Values represent mean ± SE.

PARAMETER	Fresh water	Brackish water		One-Way ANOVA	
	0.0‰ salinity	4.0‰ salinity	8.0‰ salinity	$F_{2,12}$	P
pCE	0.525 ± 0.016^{a}	0.552 ± 0.014^{b}	0.670 ± 0.029^{ab}	13.8	<0.05
YCR (mg day^{-1})	0.414 ± 0.008	0.422 ± 0.008	0.431 ± 0.009	1.04	0.384
ρ_w (mg day^{-1})	0.199 ± 0.023	0.210 ± 0.016	0.217 ± 0.019	0.216	0.809
ρ_d (mg day^{-1})	1.393 ± 0.101	1.478 ± 0.093	1.514 ± 0.074	0.476	0.632
ρ_l (mm day^{-1})	0.142 ± 0.015	0.152 ± 0.007	0.186 ± 0.018	2.66	0.11

YCE; yolk conversion efficiency, *YCR*; yolk consumption rate, ρ_w, ρ_d & $\rho_l$are rate parameters (slope) estimated by linear regression; see Figure 1 for intercept values.

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