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Growth and gonadal development in diploid and triploid turbot (*Scophthalmus maximus*)

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Abstract

This study determined the effect of triploidy on the survival, growth and gonadal development of turbot from 6 to 48 months of age. From 6 to 24 months of age (first sexual maturity), survival was similar in both ploidies ($P>0.05$). From 24 to 48 months of age, after the first sexual maturity, survival was 91.9% in diploids and 100% in triploids, which did not exhibit the post-spawning-associated mortality. Growth was similar for both ploidies during the first year of life. After that, triploids grew significantly ($P<0.05$) more that diploids, with more marked differences after each spawning season. From 24 to 48 months, the average weight difference between both ploidies was $11.4 \pm 1.9\%$, ranging from 4.3 to 23.0%. At 47 months of age, the biomass of triploids was 10.3% greater in total weight and 14.3% greater in eviscerated weight. Gonads of triploid males were similar to that of diploids, whereas in triploid females, they were significantly smaller and rudimentary. A histological analysis carried out at 47 months of age showed complete sterility of triploids in both sexes. Sex ratio was 1 male (M):0.6 female (F), for diploids, significantly ($P<0.05$) different from 1:1, and 1 M:3.3 F for triploids, significantly ($P<0.05$) different from 1:1 and from the diploids. Since females grow more than males, culture of triploids benefited from the high female ratio, which helped to reduce size dispersion. In addition, their sterility allowed better performance by avoiding the reduction in growth that takes place during the spawning periods. Together, these observations indicate that triploidy induction can be an interesting option for turbot aquaculture, especially for the production of large-size fish of more than 2 years of age.

Keywords: Triploidy; Growth; Gonadal Development; Turbot (*Scophthalmus maximus*); Sex ratio

1. Introduction

Turbot (*Scophthalmus maximus* L.) is a marine teleost which has an important commercial value. It is essential for European aquaculture industry, especially in Galicia (NW of Spain), where it generates

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85.0% of the Spanish turbot production and 45.4% of the world turbot production (Ojeda, 2003). The growing phase starts using animals weighing 5–10 g which are 3–4 months old. Turbot is a fast growing species which reaches 1.5 kg at an age of 25–29 months. Although it can be marketed as soon as it reaches 0.5 kg, most fish are marketed in the range 1.5–4 kg, depending on the end user (restaurant or domestic consumption). Based on size, fish will be eaten as a whole (~ 0.5 kg) or after being filleted.

Under culture conditions, the first sexual maturity takes place at an age of about 24 months, during the growing period. As it is the case in many fish, the first maturity does not negatively affect growth, since by that time, the gonads are still not too big. Nevertheless, animals which are cultured until they reach 4 kg undergo a second and a third sexual maturity, with more marked effects on growth than in the first sexual maturity, due to their bigger gonads. Sexual maturation disturbs behavior and slows down growth, as fish reject food and are more sensitive to changes in water temperature and low oxygen levels, which translates into mortalities during and after these periods. Therefore, induced sterility could be an effective method to solve or alleviate these problems in the culture of big-sized turbot.

Triploidy induction is an effective way to achieve sterility in fish (Swarup, 1959; Chourrout, 1987; Carrasco et al., 1999; Felip et al., 2001a; Zanuy et al., 2001). Triploidy alters chromosome pairing during meiosis and thus gonadal development is impaired, especially in females, where meiosis occurs after a relatively short period of growth by mitosis. In males, gonadal development is similar to that of diploids, and some species are even capable of producing sperm, although it is very diluted and aneuploid (Benfey, 1999). Sterility confers a potential additional advantage, as the energy invested in reproduction can be diverted to somatic growth (Utter et al., 1983; Ihssen et al., 1990; Benfey, 1999). Triploidy, however, does not always result in larger body size (Felip et al., 2001b).

Triploidy induction by chromosome set manipulation has been successfully carried out in several marine species with commercial interest, such as plaice, *Pleuronectes platessa* (Purdom, 1972), Atlantic salmon, *Salmo salar* (Johnstone, 1985), gilthead sea bream, *Sparus aurata* (Garrido-Ramos et al., 1996), halibut, *Hipoglossus hipoglossus* (Holmefjord and Refstie, 1997), sea bass, *Dicentrarchus labrax* (Felip et al., 1997), and turbot, *Scophthalmus maximus* (Piferrer et al., 2000, 2003). However, many studies have focused on determining the combination of treatment variables to induce triploidy, with less attention being paid on the actual performance of triploids under culture conditions. In those studies in which this has been done, results have been equivocal. Thus, in adult *Platichthys flesus*, triploids grew more than diploids (Lincoln, 1981), in *Paralichthys olivaceus* and *Pagrus major*, triploids grew less (Arai, 2001), and in *Dicentrarchus labrax*, triploids grew equally in the case of juveniles and less in the case of adults (Felip et al., 1999, 2001b). Based on the above, the aim of this study was to determine the survival, growth and gonadal development of triploid turbot during 4 consecutive years.

2. Material and methods

2.1. Animals used

The fish used in this experiment were reared at the Instituto Español de Oceanografía in Vigo (Spain). Eggs, all them from the same batch and from one female, and a pool of sperm from two males were obtained from fish of the same broodstock, in June 1999. Triploidy was induced by applying a cold-shock shortly after fertilization according to Piferrer et al. (2000, 2003). The ploidy level was verified in 6-month-old juveniles by determining the size of the erythrocyte major axis (20–40 erythrocytes per fish, $n=240$ diploids and $n=280$ triploids) in a blood sample, stained with Hemacolor (E. Merck. Darmastadt., Germany) as previously described (Piferrer et al., 2003). Mean erythrocyte major axis length ranged between 10.0 ± 0.21 μm and 11.4 ± 0.21 μm in control diploids and between 13.0 ± 0.16 μm and 15.0 ± 0.26 μm in cold-shocked fish ($P<0.001$). Thus, the fish used in the "triploid" groups were in fact 100% triploids.

Two hundred 6-month-old control diploids and two hundred verified triploids of the same age were placed in four 3800-l tanks. Each tank contained 100 fish, two of them with diploids and two with triploids, all with the same initial mean weight and similar weight