Development of a microsatellite based multiplex PCR standardized panel for Meagre (*Argyrosomus regius*)

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Summary

In this study, a microsatellite based multiplex PCR panel for meagre (*Argyrosomus regius*) was developed as a useful and single tool in parental assignment and population studies. From the microsatellites published in the genetic map of different species, 21 specific and interspecific microsatellites from different aquaculture species of meagre (*Argyrosomus regius*), meagre (*A. japonicus*), red drum (*Sciaenops ocellatus*) and yellow meagre (*Acoupa weakfish*) were selected. Each microsatellite marker was assessed individually, with four errors being considered: a) inadequate amplification: peak height <300 RFU (relative fluorescent units); b) null allele: preferential amplification of the short allele; c) unclear band pattern: bands that make it difficult to identify between homozygous and heterozygous for adjacent alleles; d) intermediate alleles: loci of di-nucleotides, differ from each other by 1 bp. Finally, a SuperMultiplex for *Argyrosomus regius* (SMAr) was designed with only the best eight microsatellite markers, considering their genetic variability, allelic range and genotype reliability. The panel assessment was performed using a batch of broodstock from one company and a sample of 616 offspring. It was possible to assign 95% of the offspring to a single pair of parents using the exclusion method. Genetic diversity, analyzed by heterozygosity and polymorphism degree of each microsatellite. In our population, for the selected microsatellite markers the mean values and the range were: for number of alleles 7.9 (from 5 to 13), for observed heterozygosity 0.69 (from 0.35 to 0.97), for expected heterozygosity 0.61 (from 0.32 to 0.81) and for polymorphism information content 0.57 (from 0.35 to 0.80). The microsatellite-based multiplex has proven to be an easy procedure, and a robust and low-cost tool for parental assignment to support companies breeding programs and to exchange information between research groups.

Keywords: Meagre (Argyrosomus regius); Microsatellites; Parental assignment; PCR; Population.