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Editor

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AQUACULTURE RESEARCH PROGRESS

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Expert Commentary A

**BEHAVIOURAL INDICATORS OF “WILDNESS” AS
USEFUL TOOLS FOR RESTOCKING: THE EUROPEAN
SEA BASS (*DICENTRARCHUS LABRAX* L.) JUVENILE AS
A STUDY MODEL**

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ABSTRACT

The present contribution aims to resume the main results obtained within a three years research project, analysing and discussing their potential use for the development of behavioural indicators of “wildness” of the cultured fish. The species we used as a study model was the European sea bass (*Dicentrarchus labrax* L.), which is cultured and seeded in the Northern Adriatic lagoons to sustain a traditional, specialised form of Italian aquaculture, named “vallicoltura”. We focused on the antipredator responses of the juvenile phase of this species (about 2 cm of total length) towards different stimuli associated both to a fish predator and to a potential bird predator (overhead stimuli).

We developed a series of experimental apparatuses and protocols to quantitatively assess some components of the antipredator behaviour, which were compared between hatchery-reared and wild fish juveniles. Two components resulted to differ significantly between the wild and the hatchery groups, being good candidates for the development of indices of “wildness”. These two components are the shoaling index, that is the level of cohesiveness of the group as a response to a fish predator, and the freezing behaviour, i.e. time spent motionless resting on the bottom as a response to overhead fright stimuli. For both measures it would be necessary to develop a reference system and to further improve and possibly simplify the experimental protocols for a wider and routine use of the behavioural test in the aquaculture practise related to restocking.

In response to the worldwide decline of fish populations, stock enhancement has been attempted as a way to rejuvenate populations, by rearing a large number of fish under artificial conditions and subsequently releasing them into the natural environment (Kellison et al. 2000).

During the recent years, a growing body of evidence has suggested that the success of stock enhancement programmes is highly related to the behavioural profile of the juvenile fish produced in the hatchery, as the artificial environment of the hatchery may generate a number of behavioural deficits potentially detrimental for the survival of the released fish in the wild (Einung and Fleming, 2001; Brown and Day, 2002; Huntingford, 2004). These behavioural deficits have been documented through a relatively large but constantly increasing number of studies, conducted to compare the behavioural performance between hatchery-reared fish and their wild counterparts. The main and more significant differences in behavioural traits between hatchery and wild fish have been shown especially at the level of feeding, aggressive, and antipredator behaviour (Einung and Fleming, 2001, Huntingford 2004). A large part of these differences seem to have an environmental origin, since hatchery fish experience a psychosensory-deprived environment with respect to the wild counterparts (Olla et al., 1998), and it has been shown that in many cases a relatively short exposure to a relevant and proper experience may adjust the behavioural performance reducing the gap between the wild and the hatchery fish (Huntingford, 2004). On the basis of this body of knowledge, "life skills training" and procedures designed to expose the fish to appropriate stimuli and to increase the complexity of the hatchery environment, have been recently proposed (Brown and Day, 2002). Although these procedures aim to increase both the well-being of captive animals and the related success of the released fish in the wild, it has been noted that they are costly and difficult, in certain cases, to be applied (Huntingford, 2004). On the other hand, if the aim of a release program is to increase the viability of a natural population, the detailed knowledge of the behavioural profile of the reared fish and the eventual differences with respect to the wild counterparts, is an indispensable requisite before fish releasing.

In this framework, it is therefore necessary to develop methods for a rapid and effective evaluation of the behavioural "quality" of the juvenile fish destined to release, as a part of a routine practice of any hatchery/restocking plan.

The behavioural quality of the hatchery fish should be assessed in terms of an array of behavioural indicators of "wildness", enabling to provide a measure of the distance between the hatchery and the wild fish at the different life stages of the fish before releasing. These indicators could constitute the basis to evaluate whether and how to proceed with programs of environmental enrichment, including the life skills training, and to eventually assess, in a second time, the efficacy of these programmes.

Our contribution in this field is based on our participation to a project promoted and funded by the "Italian Ministry of Agriculture and Forestry", aimed at assessing the quality of the juvenile hatchery fish of some brackish species, typical of the Italian coastal and lagoon waters. These species are cultured to sustain traditional forms of lagoon aquaculture, such as the Adriatic vallicultura (Ardizzone et al., 1988). The program was therefore an integral part of the strategies aimed at improving stock enhancement, according to the guide lines for a responsible fisheries (FAO, 1995). On the basis of some results obtained within this research project, we aim, in the present context, to propose some suggestions for a methodological approach to the assessment of the behavioural quality of the hatchery-reared fish. We focused

on the juvenile stage of the European sea bass (*Dicentrarchus labrax*), which is routinely seeded in the Adriatic lagoon to sustain “vallicoltura”, using both captured wild fish and fish obtained from controlled spawning under hatchery conditions. We compared some behavioural aspects related to the antipredator behaviour between wild and hatchery juveniles of about 2 cm in Total length. We decided to analyse the antipredator behaviour for two main reasons: this is a fundamental component for the individual fitness of the released fish in the wild, and it has been stated, on the basis of previous studies, that the levels of antipredator behaviour generally differ between hatchery and wild fish (Einum and Fleming, 2001). Although details and results of our investigations are partly found in Malavasi et al. (2004), Torricelli et al. (2005) and Georgalas et al. (2007), we aim, in the present context, to point out some aspects of this investigation, especially those related to the methodological approach. These methodological considerations could be potentially useful for a generalised and quantitative evaluation of the behavioural quality of the hatchery fish, using standardised and relatively simple protocols. We found, for example, that a simple behavioural measure, the shoaling index, differed significantly between the groups of hatchery-reared and that of wild fish juveniles, as a response towards a live fish predator behind a transparent partition (Malavasi et al. 2004). Thus, the level of cohesion of the shoal seems to be a reliable measure of the intensity of antipredator behaviour towards piscivorous predation. We measured this behavioural variable, by calculating the dispersion index (ratio between the variance and the mean) of the distribution of the juvenile fish across the rear tank wall, with the aid of a grid, before and after the introduction of the predatory stimuli, that is the fish predator (a common eel in this case). The behaviour of the juvenile shoals was video-recorded and the shoaling index could be assessed through the analysis of subsequent temporal frames. It was enough to analyse a period of 20 seconds both before and after the introduction of the predator to detect an increase of the index from 3 to 5 in the hatchery group and from 3 to 8 in the wild group. The level of shoal cohesion of the wild fish was therefore about 40% higher than in the hatchery fish. This simple behavioural variable could therefore constitute a behavioural index of wildness useful to assess the gap in the intensity of antipredator behaviour between hatchery and wild fish within the juvenile phase. During the same study, we designed other experimental apparatus to test for the differences between the two groups at the level of the response towards overhead fright stimuli, such as a shadow rapidly created on the water surface and a mechanical bill splashing into the water. In this case, we measured the distance from the bottom and the time spent motionless on the bottom, the so called freezing behaviour, as behavioural variables useful to assess the intensity of the antipredator reaction. Even in this case, we video-recorded the behaviour of the juvenile shoals both before and after the exposure to the stimulus. Video-recording is fundamental to analyse in details the behavioural sequences and to assess quantitatively the intensity of the behaviour at the different temporal frames preceding and following the stimulus, by exporting the recorded sequences into a proper software. In this case, the freezing behaviour resulted to differ between hatchery and wild fish, being therefore this measure an other good candidate as behavioural index of wildness (Torricelli et al. 2005). By extending the use of these behavioural variables, it could be possible to build up a sort of reference system, where the optimal value is that reached by the wild counterpart of a given hatchery species at a given life stage, to which the value reached by the hatchery fish could be compared. This reference system could be useful to assess the effects of experience and training on the intensity of the antipredator behaviour, in order to eventually evaluate a “life skills” training program.

Behavioural variables, such as shoaling index and freezing, can be assessed in any fish species, being very basic and “universal” components of the behavioural repertoire of Teleost fishes. If protocols were standardised, these behavioural variables could be used within a wide comparative approach.

Thus, such kind of laboratory test, aimed at assessing behavioural indices of wildness, could be useful for a general use in the hatchery practice related to restocking for 4 main reasons: 1) The experimental apparatuses are simple and easy to set up in the hatchery 2) The experiment can be performed quickly, as it is enough to transfer the experimental fish in the experimental tank just 24 hours before the experiment 3) The intensity of the behavioural response can be assessed quantitatively within a short term scale 4) The use of standardised procedure and the assessment of universal and relatively simple behavioural variables could be used with a comparative approach, taking also into account a large spectrum of factors, such as: hatchery conditions, generations of captivity, quality of parents, species, populations, experience and life stage. This would allow to obtain a reference system and a related scale of “wildness”, together with a scale of the gaps between wildness and domestication.

Although we are aware that these behavioural test do not solve the complicated problem of the real efficacy of the restocking programmes and do not make the application of “life skills training” easier and more realistic, we think that they could help at least to generate some results of general interest on the patterns of domestication occurring in the hatchery fish, suggesting more proper solutions to improve the rearing and releasing strategies.

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