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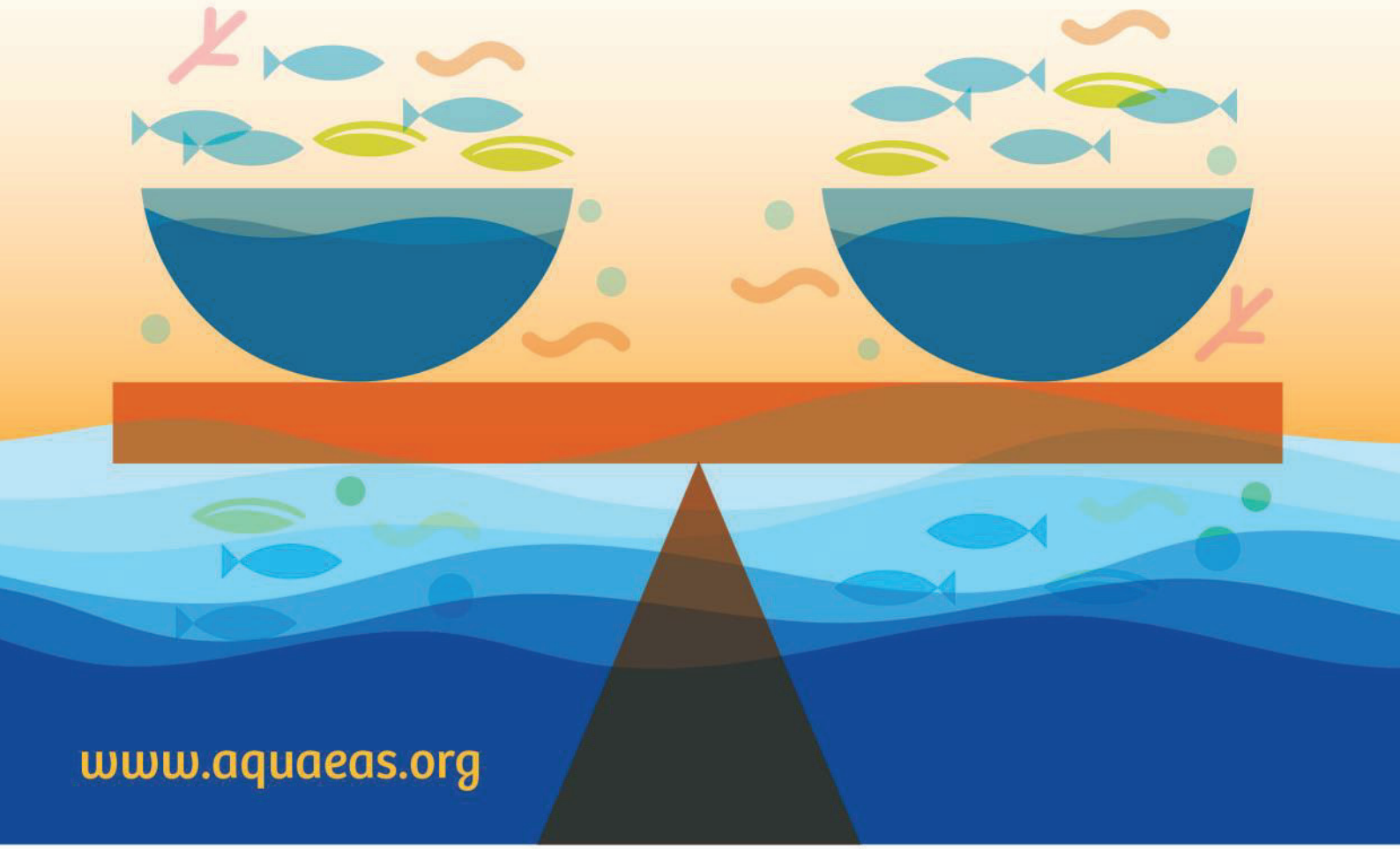

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ABSTRACTS

A RAPID MICRORADIOGRAPHIC METHOD TO DETECT SKELETAL ANOMALIES IN EARLY JUVENILE FISH

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Introduction
Skeletal anomalies affect fish growth, welfare and health, increase mortality and decrease product marketability (Boglione et al., 2013). Skeletal anomalies of early juveniles can be identified through an *in toto* double staining procedure, whose execution takes several days thus delaying diagnoses. Recently, a rapid microradiographic method to identify skeletal anomalies in early sea bream *Sparus aurata* juveniles (total length ≥ 25 mm) was proposed (Pousis et al., 2022). The aim of the present study was to assess the use of microradiography to identify skeletal anomalies in gilthead sea bream of younger age than previously reported and to extend the use of this method to the European sea bass *Dicentrarchus labrax*, the common carp *Cyprinus carpio* and the silver carp *Hypophthalmichthys molitrix*.

Material and Methods
Gilthead sea bream (N = 92; total length, TL, 18-31 mm), European sea bass (N = 145; TL 16-21 mm), common carp (N= 51; TL 18-31mm) and silver carp (N= 87; TL 17-23 mm) were sampled in fish farms operating in Italy and in Albania, anaesthetized, fixed in 10% buffered formalin and then stored in 50% ethanol. The fish were then micro-radiographed in latero-lateral projection. Since the high-resolution film is sensitive to water, a transparent polyethylene film was placed between the photographic film and the specimens. X-ray exposure was set up at 12 kV and 18 mA. The specimens <20 mm TL were subjected to 6500 X-ray shots, while the specimens ≥20 mm TL underwent 7500 shots. Films were then developed with Kodak HC-110, fixed in Kodak UNIFIX, and dried at room temperature. After microradiography, all the samples were stained with a routine *in toto* double staining procedure for bone and cartilage (Boglione et al., 2013).

Results and Discussion
In gilthead sea bream, the majority of the analyzed individuals (82.6%) showed at least one skeletal anomaly and most of them showed more than one anomaly (average: 3.3 ± 2.4). A total of 248 anomalies were recorded of which 21.4% were several anomalies such as haemal lordosis, vertebral body shape anomalies, vertebral body fusion and cephalic deformities. In European sea bass specimens, a total of 533 anomalies were recorded of which 10.3% were several anomalies such as cephalic deformities, kyphosis, lordosis and vertebral body fusion. The frequency of individuals with at least one (severe or slight) anomaly was found to be 93.1% while the average of anomalies per individual were 3.9 ± 3.1. In silver and common carp, a total of 1119 and 131 anomalies were respectively recorded. In silver carp 93.1% of the analyzed specimens showed almost one anomaly (average: 13.8 ± 15.4) while in common carp the frequency of deformed specimens was 78.4% (average: 3.5 ± 3.0). The most common severe anomalies in both species were vertebral body shape anomalies and vertebral body fusion, while no cephalic deformities were found. The frequency of specimens with almost one severe anomaly was 57.5% and 41.2%, respectively. In all the analyzed specimens the most common slight anomalies were deformed neural and haemal arches. All the skeletal anomalies observed after double staining method were also identified in the microradiographic plates (Fig. 1).

The present study suggests microradiography as a useful tool for the rapid identification of skeletal anomalies in early juveniles of different species, provided that X-ray exposure conditions appropriate to the size of the examined samples are set up.

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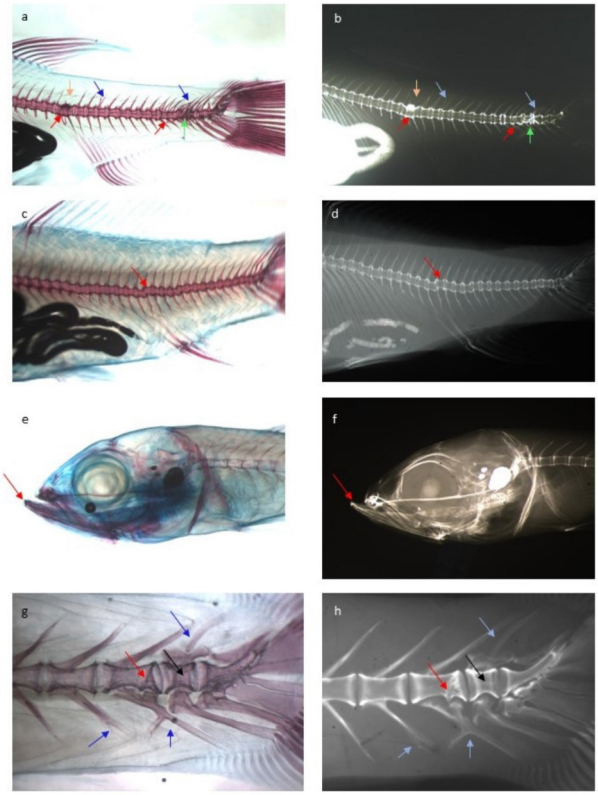

Fig. 1. Skeletal anomalies observed in double stained samples (left) and in microradiographic plates (right). a) and b), silver carp. Orange arrow: absence of neural arches of haemal vertebra; red arrow: fused haemal vertebra; blue arrows: deformed neural arches; green arrow: deformed bodies of haemal and caudal vertebrae. c) and d), common carp. Red arrow: kyphosis and deformed vertebrae between the 21st and the 24th vertebrae. e) and f), sea bass. Red arrow: anomalous dentary. g) and h), gilthead sea bream. Red arrow: partial fusion of two caudal vertebrae; black arrow: deformed body of a caudal vertebra; blue arrows anomalous neural and haemal arches.

References

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