



# Effects of light spectrum and tank background colour on Atlantic cod (*Gadus morhua*) and turbot (*Scophthalmus maximus*) larvae performances



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## ARTICLE INFO

### Article history:

Received 9 December 2013

Received in revised form 25 June 2015

Accepted 26 June 2015

Available online 4 July 2015

### Keywords:

Marine teleosts

Larviculture

Light wavelength

Tank colour

Growth

Survival

## ABSTRACT

Light is a key environmental cue involved in the entrainment and regulation of fish development and physiology. Species specific spectral differences in sensitivity are believed to be an adaptation to the species' ecological niche and therefore fish larvae are predisposed to perform better under specific light environments. This study investigated the effects of light spectrum including blue (455 nm), green (530 nm), red (640 nm) and white on larvae performances of two temperate marine species, Atlantic cod (*Gadus morhua*) and turbot (*Scophthalmus maximus*). In both species, larvae exposed to shorter wavelengths (blue and green spectrums) showed significantly enhanced growth in comparison to larvae exposed to longer wavelengths (red). However, green spectrum appeared to reduce survival rates for both species. Larvae performances in the colour background experiment differed between species with cod larvae survival enhanced when exposed to a blue background but growth performances reduced. No significant impacts of background colour were seen in turbot. The results of the present study highlight the importance of considering light environment in marine larvae rearing protocols to enhance larval performance and survival.

**Statement of Relevance:** Improving hatchery performances of marine fish larvae.

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## 1. Introduction

Protocols for the optimal rearing of marine fish larvae have seen significant improvements in the last decade through the definition of the most appropriate rearing environmental and nutritional conditions in culture (including systems, temperature, live feeds) (Hamre et al., 2013; Planas and Cunha, 1999). However, the impact of light has received comparatively little attention (Pittman et al., 2013; Villamizar et al., 2011). Fish feeding and foraging activity involves different sensory mechanisms (e.g. vision, mechanoreception, chemoreception, electroreception) however, none of those systems are fully functional at the early stages of larval development. Eye development is one of the first changes in larvae morphology with the appearance of a duplex retina (rods and cones), that enlarges as the fish grow (Evans and Browman, 2004). Retina development starts by the appearance of cone photoreceptors (Blaxter and Staines, 1970), responsible for colour detection, while rods (intensity reception) develop later during metamorphosis. The basic structure of photoreceptors (retinal and non-retinal) is well conserved across vertebrates, although specific morphological and functional features vary widely across species and within developmental stages

(Blaxter, 1969; Britt et al., 2001). Fish living in different photo-environments thus appear to have adapted their maximum photopigment sensitivity according to the species ecological niche (Kusmic and Gualtieri, 2000). In addition water selectively absorbs light wavelengths with blue wavelengths (~450 nm) being the most efficient at penetrating the sea water column (>100 m depth depending on water turbidity) (Smith, 1974). This explains the increased sensitivity to short wavelengths (e.g. blue/green) displayed by the few marine fish species studied to date (Villamizar et al., 2011).

Photoreception plays a crucial role throughout the whole teleosts' life cycle regulating (directly or indirectly) physiological and behavioural (feeding activity, swimming, schooling, migration and reproduction) responses (Boeuf and Le Bail, 1999; Migaud et al., 2010). In an aquaculture context, incident light, in terms of intensity and spectral content, has been shown to influence larval development, growth and survival in a number of marine teleosts, however response appears to be species specific (Villamizar et al., 2011). Tank colour, which is less well studied, has also been shown to impact on growth and/or development in a limited number of species including haddock (*Melanogrammus aeglefinus*) (Downing and Litvak, 2000), spotted sea bass (*Paralabrax maculatofasciatus*) (Pena et al., 2005), striped trumpeter (*Latris lineta*) (Cobcroft and Battaglione, 2009) and orange spotted grouper (*Epinephelus suillus*)

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(Duray et al., 1996). The drivers behind these physiological effects remain to be defined however visual acuity and thus perception of prey may be one of the main factors (Utne-Palm, 2002).

The aim of this study was to test the effects of light spectrum and tank background colour on larvae development and survival of two commercially important fish species inhabiting different ecological niches at the adult stage (pelagic Atlantic cod, *Gadus morhua*, vs. benthic turbot, *Scophthalmus maximus*).

## 2. Materials and methods

### 2.1. Broodstock and egg production

All the experiments were carried out at the facilities of Viking Fish Farms Ltd., Ardtoe Marine Laboratory, Acharacle, Scotland (N 56°46' W 05°53') and conducted in accordance with the Animals Scientific Procedures Act of 1986, UK following independent ethical review. Hatchery reared Atlantic cod broodstock were kept in an indoor circular tank with a flow through system at ambient temperature (7–8 °C spawning season), salinity (~34.5 ppt) and photoperiod (except for experiment 2a where out of season eggs were used). Atlantic cod broodstock spawned naturally in the tanks and eggs were collected daily at 15:00 h through a 100 µm mesh egg collector.

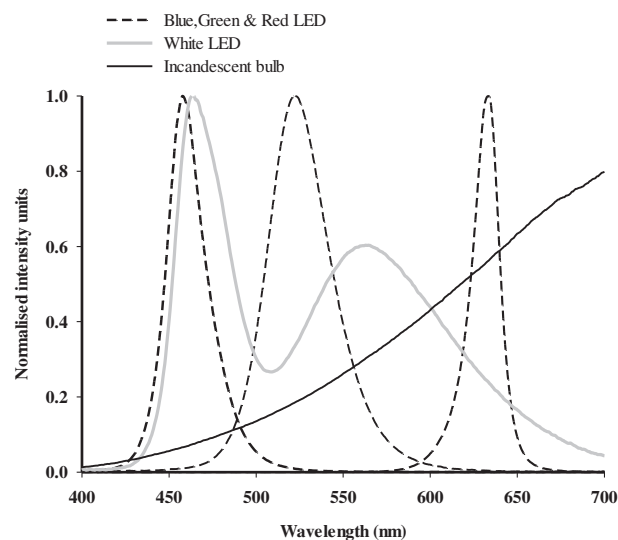
Turbot broodstock were kept in an outdoor tank under natural ambient light conditions, temperature (12–14 °C during spawning season) and salinity (~34.5 ppt). As turbot do not spawn spontaneously in captivity, fish were stripped every third day by gentle abdominal massage according to hatchery procedure. Eggs from several females (3–5) were stripped, pooled and fertilised with milt from at least two males.

Eggs used in the present experiment for both cod and turbot were first checked for quality (floating fraction, development through blastomere scoring, fertilisation rate and total egg production). Cod egg floating fractions were disinfected for 45 s (Kickstart, RS Hygiene, UK, 1:250) and then rinsed in filtered sea water for 30 s, weight measured and transferred to 80 L conical black incubators. Eggs were first incubated in darkness at ambient temperature (7–8 °C and 12–14 °C for cod and turbot eggs, respectively) at a density of 6 g of eggs L<sup>-1</sup> and provided with continuous water flow at 400 mL min<sup>-1</sup>. Cod eggs were transferred to the experimental system at 75–80 °C days (prior to hatching) while turbot eggs were left to hatch in the incubator and transferred at 100 °C days. Numbers were assessed volumetrically from the floating fraction in the incubators prior to transfer.

### 2.2. Experimental setup and treatments

Two sets of experiments were performed in both species: effects of narrow bandwidth (spectrum) light on larvae performances (Exp. 1a and b for cod and turbot, respectively) and effects of tank background colour on larvae performances (Exp. 2a and b for cod and turbot, respectively). All trials were performed in the same experimental setup that consisted of 12 flow through tanks (triplicated design with four treatments). Continuous illumination (24L:0D) was used as it is the industry standard based on results obtained by Puvanendran and Brown (2002) for cod. Temperature was kept at 9.0 ± 1.0 °C for cod trials and 18.5 ± 0.9 °C for turbot trials. Seawater was passed through mechanical filtration systems (sand filters and cartridges; 5 µm particles size) and UV sterilised. All experimental tanks received gentle aeration via a glass tube (0.5 mm) and water was exchanged at 0.2 L min<sup>-1</sup> with temperature and dissolved oxygen monitored daily.

Light spectrum experiments (Exp. 1a and 1b) were performed in 150 L black, flat bottom circular tanks running at a 100 L capacity. Light was provided by white (wavelength peaks: 460 and 560 nm), green (530 nm), blue (455 nm) and red (640 nm) dimmable Light-Emitting Diode (LED) lamps (Intravision Aqua AS, Norway) suspended 40 cm above the water surface connected to individual dimmers. Spectral profiles are shown in Fig. 1. Light intensity in the experimental tanks



**Fig. 1.** Normalised spectral profiles from light sources used the experiments. LED peak wavelengths: blue (455 nm), green (530 nm), red (640 nm) and white (460 and 560 nm). LED light was provided in light spectrum experiments 1a (cod) and 2a (turbot). Incandescent tungsten bulb (60 W) was used in tank background experiments 1b (cod) and 2b (turbot). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

was balanced to  $1.39 \pm 0.01 \text{ W m}^{-2}$  at the surface and  $0.37 \pm 0.01 \text{ W m}^{-2}$  at the bottom for all light treatments (equivalent to  $7.4$  and  $1.97 \mu\text{mol m}^{-2} \text{ s}^{-1}$ , respectively).

Tank background colour experiments (Exp. 2a and 2b) were performed in 100 L polyethylene coloured (white, blue, red and black) flat bottom circular tanks (Mailbox mouldings, UK) filled to 75 L. Light was delivered by 60 W dimmable incandescent tungsten bulbs suspended 40 cm above the water surface to provide a comparison with commercial production of turbot and cod on site. Light intensity in the experimental tanks was balanced to  $1.25 \pm 0.04 \text{ W m}^{-2}$  at the surface, and  $0.18 \pm 0.01 \text{ W m}^{-2}$  at the bottom (equivalent to  $6.66$  and  $0.96 \mu\text{mol m}^{-2} \text{ s}^{-1}$ , respectively).

Light irradiance ( $\text{W m}^{-2}$ ) for all treatments was measured with a calibrated single channel light sensor (Skye Instruments Ltd., Powoys, UK). Spectral composition was measured using a portable spectroradiometer with an umbilical fibre optic based sensor head (Stellarnet Inc, USA). Light intensities tested in cod larvae were determined by the performance of the lighting systems used characterised by spectrum specific LED outputs and previous published literature on the effects of light intensity on gadoid larvae growth and survival (Downing and Litvak, 2000, Puvanendran and Brown, 2002). No such data is available for turbot larvae and therefore intensities tested were similar to the cod experiments.

### 2.3. Larval rearing

At 100 °C days Atlantic cod egg hatching rates were determined (Exp. 1a: 96.6% and 2a: 96.0%) and light treatments as well gentle aeration and water inflow started ( $0.1 \text{ L min}^{-1}$ ). Larvae ( $50 \text{ larvae L}^{-1}$  for Exp. 1a and  $75 \text{ larvae L}^{-1}$  for Exp. 2a) were reared in “green water” using 50:50 mixture of *Nannochloropsis atomus* and *Isochrysis galbana* ( $20 \times 10^6$  and  $13 \times 10^6 \text{ cells L}^{-1}$ , respectively). Microalgae were added every day. On the 3rd day post hatch (DPH), rotifers enriched with *Pavlova lutheri* ( $5 \text{ individuals mL}^{-1}$ ) were introduced and water inflow increased ( $0.2 \text{ L min}^{-1}$ ). At 30 DPH (240 °C days) *Artemia salina* nauplii (enriched with AlgaMac-2000, Aquafauna Biomarine Inc., Hawthorne, California, USA) was introduced ( $0.3 \text{ individuals mL}^{-1}$ ). At 36 DPH rotifers and microalgae were completely substituted by  $0.5 \text{ Artemia naupii mL}^{-1}$  (supplied twice a day from 42 DPH to 53 DPH).

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