Measuring effects of different temperatures on swimming activity and social behavior in groups of Mediterranean marine fish with the EthoVision Color-Pro video tracking system

M.Y. Ylieff [1,2,3], C. Sanchez-Colero[1], P. Poncin[1], J. Voss[2] and J.C. Ruwet[1]

1Department of Ethology and Animal Psychology, Institute of Zoology "Ed. van Beneden", University of Liège, Liège, Belgium_[2]Aquarium "M. Dubuisson", Institute of Zoology "Ed. van Beneden", University of Liège,Liège, Belgium_[3]Scientific Research Worker - F.N.R.S. (Belgian National Funds of Scientific Research), Liège, Belgium

In nature, fish are continuously submitted to various external environmental factors (biotic, abiotic) affecting their behavioral responses. We have already conducted studies to determine the effect of environmental variables on sexual behavior in fish. However, other behaviors involved in reproduction, like swimming activity, are also influenced by the environment. Temperature is almost certainly one of the most important environmental effectors influencing swimming activity. In laboratory (aquariums) we used EthoVision Color-Pro 1.96 (Noldus Information Technology), a computerized video tracking system based on digital imaging techniques, to quantify and take into account variables that cannot be measured accurately by usual methods of direct observation.

In a first approach conducted in three experimental aquariums (L100 x W50 x D48 cm) equipped with 2x30W neon tubes, reduced in an arena of L68 x W30 x D48 cm with blue plastic plates), three different temperatures (16 °C, 21 °C, 26 °C) were tested in the Mediterranean damselfish *Chromis chromis* (length: 9 cm). To track several individually identified fishes, we used color markers (fluorescent color plastic pearls of 1 centimeter) attached under the dorsal fin of the fish. In theory, the color tracking system can detect up to eight colors, but in practice we only found two colors (fluorescent green and pink) detectable simultaneously in a water environment. Placing the camera in front of the arena, we measured in two individual damselfish (2/4 fishes tracked at each trial) the distance moved (DM), the velocity (V), the social interactions reflected by the distance between the two animals (DO) and the time spent in the defined zones (IZ) of the aquarium (sandy bottom, middle water, near surface), according to water temperature and

fish density at 21 °C (1, 2 and 4 fishes). It is worth noting that we have tested the potential impact of the pearl on the swimming activity with trials using one fish with and without color mark (monochrome tracking). Each session (track) lasted 1 hour with an image sample rate of 5 samples/s (18000 samples/h).

The pearl has no significant effect (n=6) on swimming behavior (DM, V) and time spent in the defined zones (IZ). Density does not significantly affect the DM, V, IZ, except for the density 4, where the fish swims faster near the surface (n=12, p=0.01). However, densities 2 and 4 influence the DO (larger for density 2) in the arena (n=12, p=0.05) and when a fish is alone, it stays longer on the bottom (n=6, p=0.04). The three temperatures tested do not influence the DM, V, IZ, except for 21 °C, where fish swims faster near the surface (n=12, p=0.01), probably an adapted behavior to escape from birds in nature. An interesting result concerns the DO on the bottom that is inversely proportional to the increase of temperature and highly correlated with this factor (n=12, r=-0.984). This measure could reflect the increase of social interactions observed with reproductive behaviors in nature, which appear with the elevation of water temperature in June.

Our study illustrates the new possibilities of digital imaging techniques for the understanding of the relations between fish and their biotic and abiotic environment.

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