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Enigmatic coloration pattern in greater weever *Trachinus draco* Linnaeus, 1758 and its biological significance

by

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Résumé. – Coloration énigmatique chez la grande vive *Trachinus draco* Linnaeus, 1758 et sa signification biologique.

Un total de 58 spécimens de *Trachinus draco* Linnaeus, 1758 ont été capturés dans les eaux de Galice (nord-ouest de l'Espagne) et dans le golfe de Cadix (sud de l'Espagne) entre juin et novembre 2020. Vingt-huit d'entre eux, tous des mâles, présentaient une tâche sombre distincte située au-dessus de la nageoire pectorale et sous la ligne latérale des deux côtés du corps. Les données bibliographiques et photographiques ont montré le même schéma de coloration dans le temps et dans toute leur aire de répartition. Une coloration nuptiale temporaire chez les mâles est proposée pour la première fois comme cause de cette coloration.

Key words. – *Trachinus draco* – Morphology – Nuptial coloration – Reproduction.

The Trachinidae family comprises nine marine species distributed worldwide. On the Atlantic European coasts, only four species belonging to two genera are reported: the lesser weever *Echiichthys vipera* Cuvier, 1829, the greater weever *Trachinus draco* Linnaeus, 1758, the spotted weever *Trachinus araneus* Cuvier, 1829 and the starry weever *Trachinus radiatus* Cuvier, 1829. *Trachinus draco* lives near the eastern Atlantic coastline, from Norway to Mauritania, including the Madeira and Canary Islands, and the Mediterranean, Aegean, and Black Seas (Buz and Başusta, 2015). This species inhabits muddy bottoms, often burrowing into the substrate, from the coastline to about 200 m depth, most common between 20 and 50 m, but migrating into deeper waters (up to 100 m) during the winter (Smith, 2016). The normal coloration pattern of this species is dorsally green with brown or green scales arranged in oblique rows forming numerous dark lines directed downwards and backwards; lateral and ventral surfaces light yellow (Smith, 2016). Colour patterns in animal species are involved in a wide range of functions such as camouflage, successful foraging, thermoregulation, photo protection, mate selection, social signalling and predator avoidance (Cal *et al*., 2018). Fish coloration is produced through specialized cells called chromatophores and iridophores, and is one of the external morphological features traditionally used in the fish species identification. In addition, fish coloration involves many colour variations, called sexual dichromatism and temporary colour changes in the reproductive phase, colour polymorphism, morphological colour changes and abnormal body coloration or malpigmentation.

The present study aims to correlate the enigmatic coloration pattern of *T. draco* with its biological significance.

MATERIAL AND METHODS

The material examined includes a total of 58 specimens, 28 from Galician waters (Northwest Spain) between 221 and 330 mm TL caught in June and August 2020 at a depth of 30 and 36 m, and from Gulf of Cádiz (South Spain) between 176 and 325 mm TL caught in August and November 2020 between 35 and 92 m depth. In the laboratory, total length (TL) to the nearest mm, total weight and gonad weight were recorded and the sex of each individual was identified by inspection of the gonads. An extensive bibliographic and image search of the species was also made. Subsequently, seven Galician specimens caught in June (Fig. 1) were frozen and deposited in the fish collection of the Museo Luis Iglesias de Ciencias Naturais in Santiago de Compostela with the reference numbers MHNUSC 25178-1 to MHNUSC 25178-7. A literature review was also carried out, including available photographs. From the seven voucher specimens, complementary molecular and reproductive analyses were performed. DNA analysis of muscle tissue samples, amplifying part of the sequence of the mitochondrial gene *cytochrome c oxidase* subunit I (COI) was performed as previously described in Bañón *et al*. (2019). DNA sequence data, specimen photographs and other metadata have been deposited in the Barcode of Life Database (BOLD Systems; www.boldsystems.org) as part of the project entitled "Marine Fishes from Galicia" (code FIGAL) with Process IDs FIGAL036-20 to FIGAL042-20. Nucle-

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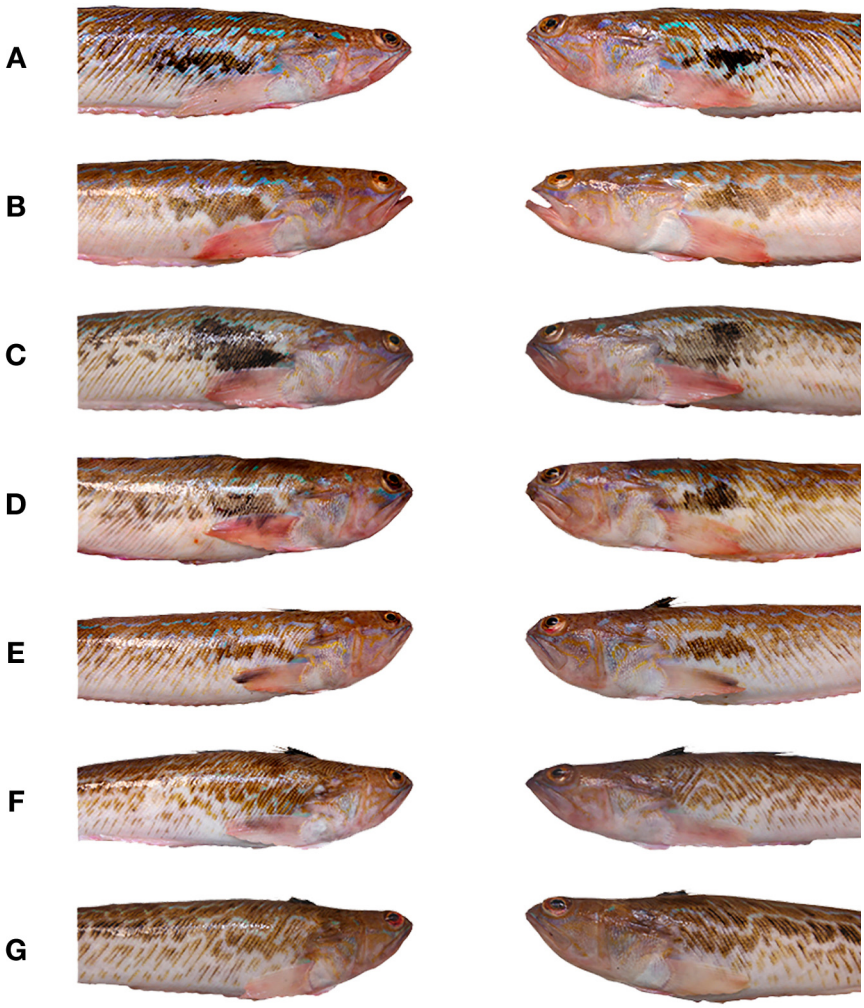

Figure 1. – Right and left side of *Trachinus draco* individuals showing dark patch (A-E) or patch absence (F, G). **A**: Male 297 mm TL; **B**: Male 264 mm TL; **C**: Male 305 mm TL; **D**: Male 268 mm TL; **E**: Male 306 mm TL; **F**: Female 323 mm TL; **G**: Female 317 mm TL.

otide sequences are also available in the GenBank database under accession numbers MT998972-MT998978.

For reproductive analysis, gonads were removed and immediately fixed in 4% formaldehyde buffered with $Na_2HPO_4 \cdot 2H_2O$ (0.046 M) and $NaH_2PO_4 \cdot H_2O$ (0.046 M) (0.029 M). A subsample from the central section of both lobes of the gonad of each individual was extracted, dehydrated and embedded in paraffin, sectioned at 3 µm and stained with haematoxylin-eosin for its analysis under microscope. Histological slides were used to determine microscopic maturity based on the most advanced oocyte developmental stage/spermatogenic stage present, signals of previous spawning, or other structural details (Brown-Peterson *et al*., 2011).

RESULTS

The specimens examined were 28 males between 176 and 320 mm TL and 30 females of 220 and 330 mm TL. All male specimens showed a patent grey to black dark patch located above the pectoral fin and below the lateral line in both sides of the body, which is absent in the females (Fig. 1). Judging by the literature reviewed, this character seems to show a spatial-temporal continuity in the species. Bleeker (1861) reported many individuals of *T. draco* caught in the Mediterranean and bought at the Marseilles fish market, which showed a large blackish spot behind the head and below the lateral line. De Buen (1935) also reported this in two specimens photographed in Palma de Mallorca. A revision of the available images on the web also showed this coloration pattern in specimens at the Olhao (Portugal) fish market in 2009 (http://www.ictioterm.es), in the Canary Islands (https://reeflifesurvey.com) or in the Madeira Islands (https://www.fishbase.de/search.php). However, despite this background, this unusual coloration is unreported, as a character, in the ichthyological literature. A possible reference was recently found in this regard. Heneish and Rizkalla (2021) mentioned the presence of a large black spot above the pectoral fin in the coloration of the species, although this spot is absent in its representative image, showing that is not a common character of the species.

Histological analysis of the voucher specimens showed that all specimens were in the spawning capable phase (SC). Five specimens were males with presence of spermatozoa in the lumen of lobules and/or sperm ducts (Fig. 2A), and two specimens were females in active spawning subphase, due to the presence of germinal vesicle migration oocytes in the lumen (Fig. 2B), a signal of imminent spawning. The presence of individuals in SC phase indicates that June is included in the spawning period of the species in the sampling area.

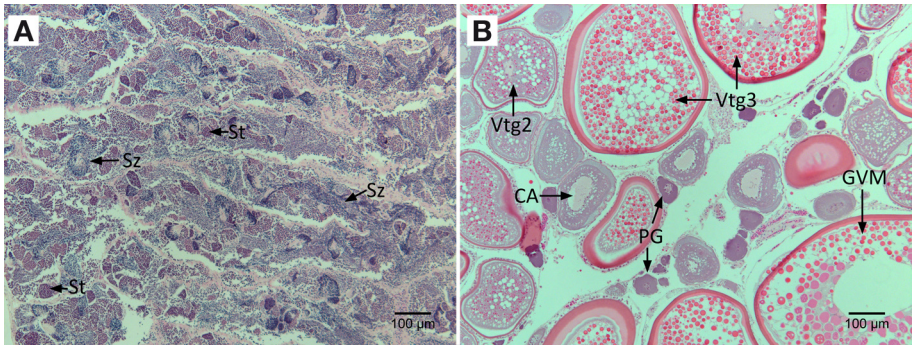

Figure 2. – Gonad transverse sections of *Trachinus draco*. **A**: Detail of functional testicular tissue of male 268 mm TL (St = spermatid; Sz = spermatozoa); **B**: Detail of functional ovarian tissue female 317 mm TL, actively spawning female (PG = primary growth oocyte; CA = cortical alveoli; Vtg2 = secondary vitellogenic oocyte; Vtg3 = tertiary vitellogenic oocyte; GVM = germinal vesicle migration). Based on the classification of Brown-Peterson *et al*., 2011).

DISCUSSION

At first glance, the presence of a dark patch is reminiscent of a case of abnormal melanic colouration. The occurrence of melanism in nature is relatively rare in fish, characterized by more or less dark spots randomly distributed on the head and/or body and fins. However, in the case of *T. draco* specimens, they are uniform or slightly fragmented spots, which are not unusual and follow a pattern, all located above pectoral fin and on both sides of body. These characteristics seem to rule out melanosis as the possible cause of its appearance. A case of sexual dichromatism could be another possible explanation, but these phenomena have not been demonstrated in any Trachinidae species to date (Smith, 2016).

The presence of this dark spot throughout time and range of *T. draco* indicates that this feature is a morphological trait of the species, and considering the results of this study, a case of male nuptial colouration is the most likely explanation. During the breeding season, the male would undergo a colour change in the anterior part of the body, which would assume an elongated dark patch, disappearing once mating is complete. In fact, specimens caught in November have less apparent spots than specimens caught in June and July.

Kodric-Brown (1998) defined the expression of nuptial chromatic traits in fish as temporary colour changes associated with reproduction. Nuptial coloration is reported in many marine fish species, such as the two spotted gobies *Gobiusculus flavescens*, the three-spine sticklebacks *Gasterosteus aculeatus* or blenniiform fishes, mainly members of the Blenniidae, Tripterygiidae and Chaenopsidae families. Temporary dark colorations are not unusual in the breeding season. Black heads and bodies that contrast with the background are reported for nuptial males of several blenniiform fishes and males of painted greenling *Oxylebius pictus* become dark during courtship.

Regarding the reproductive season, it is well known that the sexual development of *T. draco* occurs in the period between April and May and that the spawning period happens between June and August (Ak and Genç, 2013). The June and July samples fall within this period, but the November samples do not, which probably indicates that the black spots remain for some time after the reproductive phase. Differences can be observed in patch size and colour intensity, ranging from grey to black. Changes in intensity and/or area of black, brown, or grey melanic colours are commonly regulated through the melanocyte-stimulating hormone and may indicate static properties such as species identity, sex, or developmental stage, but also the current quality and motivational state of an individual (Price *et al*., 2008).

The presence of June in the spawning period is in accordance with other studies of *T. draco* along its geographical distribution such as in the Mediterranean, Aegean Sea, Kattegat or the Black Sea. Furthermore, the simultaneous presence of oocytes in all stages of development in the female ovaries shows an asynchronous development of secondary growth follicles (Wallace and Selman, 1981). Species with asynchronous oocyte development are assumed to be batch-spawner species, with several spawning events during the same reproductive cycle, a strategy generally associated with indeterminate fecundity (Brown-Peterson *et al*., 2011). Even this batch-spawner strategy has already been mentioned for *E. vipera* (Brook, 1884), another Trachinidae species, this is the first histological evidence described in *T. draco*. Due to the low commercial value of this species, the seasonality of coloration or the lack of research on the descriptive characters, often considered of less scientific interest, may be the most likely reasons why this phenomenon has not been documented so far.

CONCLUSION

It is necessary to develop several lines of research in the future, such as the delimitation of the period in which this colouration occurs, the size of the affected population and the role of the patches during courtship and mating of the species.

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