

Study of the development of the caudal endoskeleton of the turbot *Scophthalmus maximus* L., 1758 (Pleuronectiformes: Scophthalmidae).

Bruno Chanet¹ and France Wagemans²

¹Institut Universitaire Européen de la Mer, pl. Nicolas Copernic, Technopôle, Brest-Iroise, F-29280 Plouzané, France

²Laboratoire de Morphologie fonctionnelle et évolutive, Université de Liège, Institut de Chimie (B6), Sart-Tilman, B-4000 Liège (Belgium)

ABSTRACT. The development of the caudal endoskeleton of the turbot (*Scophthalmus maximus* L.) is described and compared to the setting-up of the caudal endoskeleton in other flatfishes. In the turbot, caudal bony elements develop very early, before the eye migration. The same pattern occurs in the flatfishes of the bothoid group, while in the Soleidae the supporting caudal fin elements develop once the eye has migrated. This discrepancy is discussed; it points out that more work on the ontogeny of flatfishes is needed.

KEY WORDS: Pleuronectiformes, turbot, caudal endoskeleton, development.

INTRODUCTION

Several authors have shown the importance of studying the caudal endoskeleton in order to define major events in the phylogeny of the pleuronectiform fishes (HENSLEY & AHLSTROM, 1984; CHAPLEAU, 1993; COOPER & CHAPLEAU, 1998). In this supporting caudal fin structure, different features can be identified as unique, or apomorphic, for several flatfish groups. However, the sister group of the Pleuronectiformes within the Percomorpha being still uncertain (JOHNSON, 1993; CHAPLEAU, 1993), it is not always easy to decide the character states through out-group comparison and some problems of homology remain. Thus, HENSLEY (1997) insisted that phylogenetic works associated with the study of the ontogeny were very badly needed. In that perspective, we present here data concerning the development of the caudal endoskele-

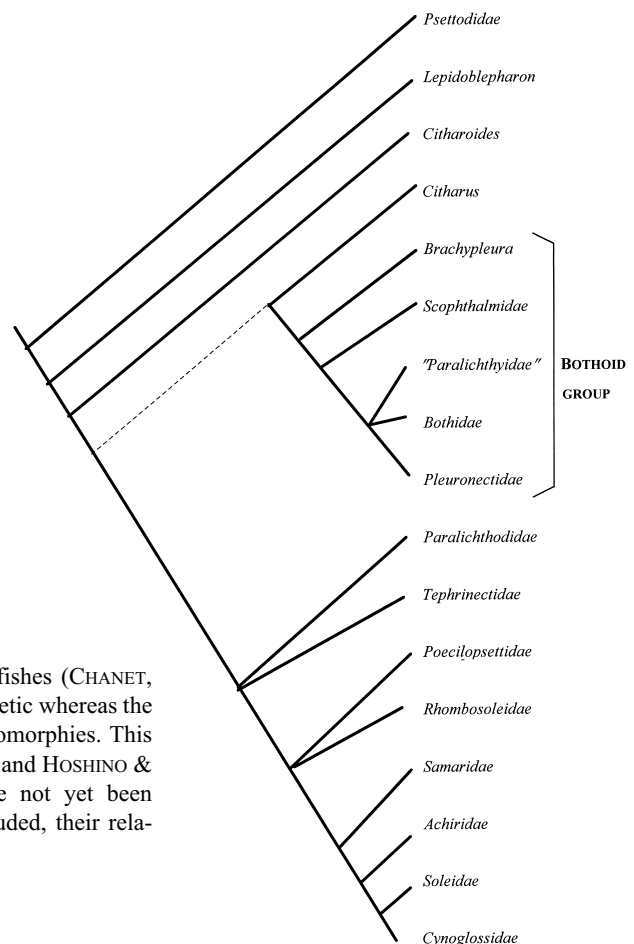


Fig. 1. – Tree of relationships between the major groups of flatfishes (CHANET, 1999). The inverted commas (") indicate that the group is paraphyletic whereas the dotted line shows a clade that is not well corroborated by synapomorphies. This tree is reconstructed from the data of COOPER & CHAPLEAU (1998) and HOSHINO & AMAOKA (1998). Because Pleuronectiformes relationships have not yet been analysed with both the Paralichthodidae and Tephrinectidae included, their relationships with other taxa are indicated by a polytomy.

ton in the turbot. The turbot (*Scophthalmus maximus* L. 1758) is a well-known flatfish species belonging to the family Scophthalmidae, within the bothoid group in the pleuronectiform order (Fig. 1). We have already noted anomalies in the development of the caudal endoskeleton of the turbot and proposed a mechanism to explain it (CHANET & WAGEMANS, 1997), but here we intend to: i) present the ontogenetic data, ii) compare our observations to previous descriptions of the development of the caudal endoskeleton in other flatfish species.

MATERIAL AND METHODS

Scophthalmus maximus fry were raised in the aquaculture-station of France Turbot-NATA (Noirmoutiers, France) at 15 °C. Samples of 30 fry were sampled on days 0 to 61 post-hatching. The fry were fixed in a CaCO₃ buffered 10% formalin solution and were cleared with trypsin. Some of them were stained with alcian blue to reveal the cartilage and others with alizarine, to stain the calcified bones, according to TAYLOR & VAN DYKE's method (1985). It was possible to stain the oldest stages simultaneously with alizarine and alcian blue. Finally, the fry were stored in glycerin. A 6 month-old specimen was cleared with trypsin, stained with alizarine and stored in glycerin according to TAYLOR and VAN DYKE's method. The specimens -270 larval and juvenile turbot- have been studied with a binocular Wild M10 Leica dissecting microscope at 8x magnification, and a drawing tube. The length from the tip of the snout to the posterior margin of hypural elements (standard length - SL) was measured for each specimen.

RESULTS

The first discernible caudal fin elements appear at day 13: the notochord is already slightly flexed and its ventral edge shows a slender cartilaginous ribbon with a ventrally protrusive expansion (Fig. 2A). At day 14, this ventral ribbon is dissociated into three ventral elements (Fig. 2B). The anterior-most one – the future parhypural – is never in contact with the notochord, the following one – the future hypural 1 and 2 plate – is a broad plate, the posterior one – the future hypural 3 and 4 plate – is smaller and has an irregular dorsal edge. These latter two elements are in contact dorsally with the notochord and ventrally support the first caudal fin rays. Anteriorly, the vertebral cartilaginous structures develop with neural spines dorsally and haemal spines ventrally. At day 22 (Fig. 3A), the notochord is strongly curved with an almost vertically directed distal part. Ventrally, the parhypural and the two hypural plates are larger and their median parts begin to ossify, while their proximal and distal parts remain cartilaginous. A little hypural 5 is visible, two cartilaginous epurals are present dorsally. At day 26 (Fig. 3B), as the hypural elements are well developed, the notochord regresses, hypural 5 is close to its distal tip. The posterior-most epural is smaller and

more ventrally placed than its serial homologue. The haemal and neural spines are more ossified. At day 29 (Fig. 4A), the vertebral centra are fully ossified and fused with their haemal and neural spines. In some specimens (55%), the second preural centrum (PU2) shows two neural and haemal spines (Fig. 4A-B). We showed (CHANET & WAGEMANS, 1997) that this anomaly was the result of a

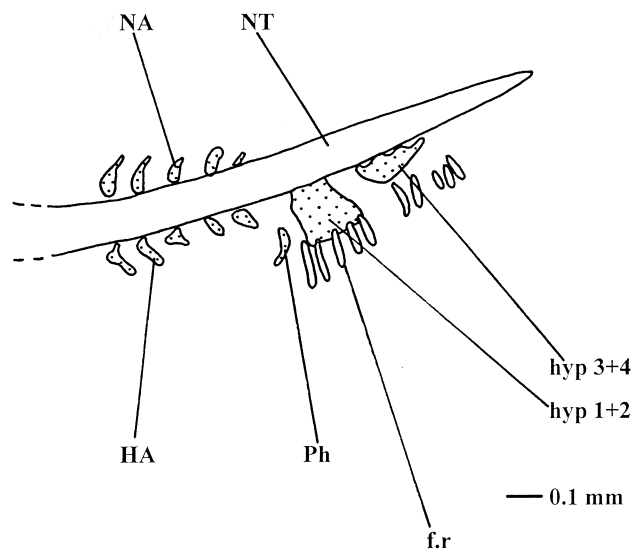
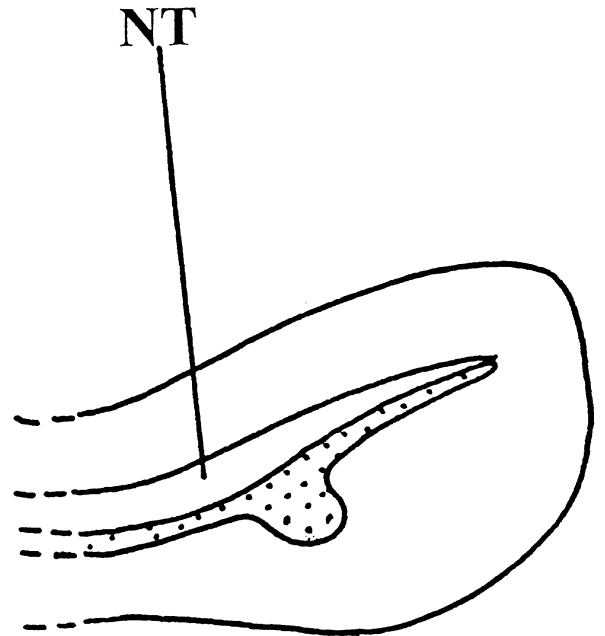


Fig. 2. – A. Caudal endoskeleton of a young turbot at day 13 (LS= 5 mm). B. Caudal endoskeleton of a young turbot at day 14 (LS= 6.5 mm). The stippled areas are cartilaginous regions. f.r: fin ray, HA: haemapophysis, hyp: hypural, NA: neurapophysis, NT notochord, Ph: parhypural.

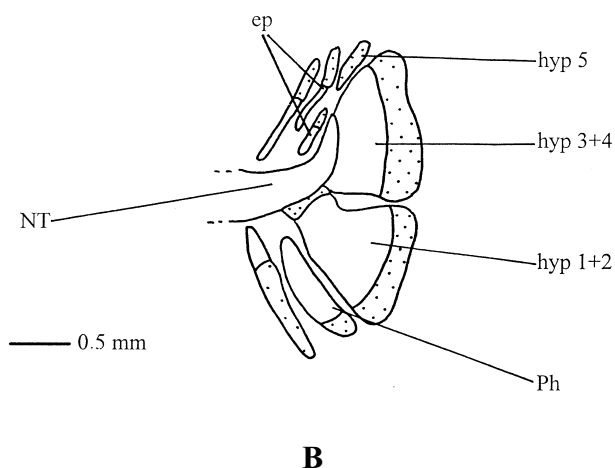
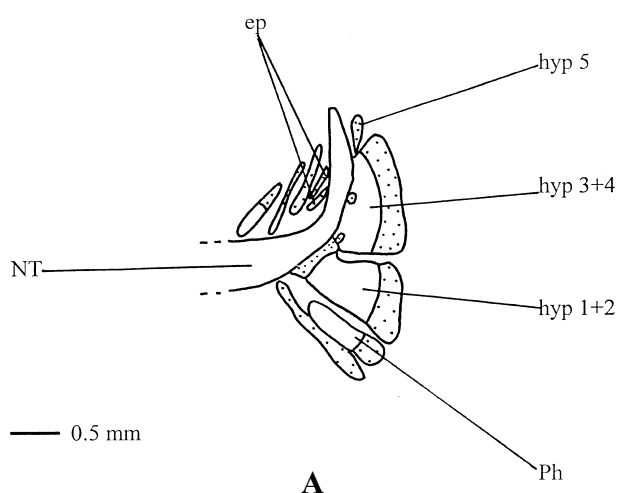


Fig. 3. – A. Caudal endoskeleton of a young turbot at day 22 (LS= 9 mm). B. Caudal endoskeleton of a young turbot at day 26 (LS= 15 mm). The stippled areas are cartilaginous regions. ep: epural.

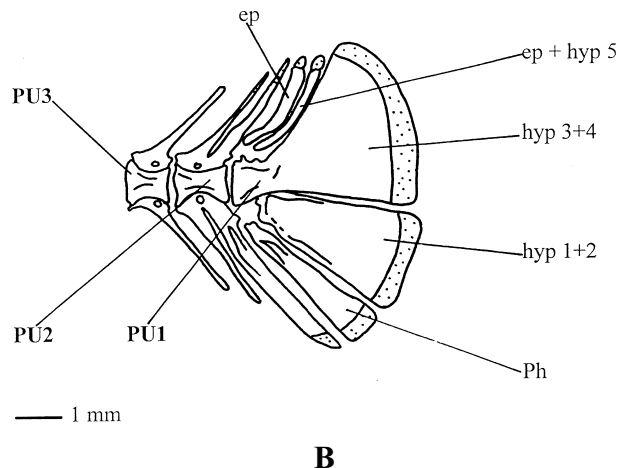
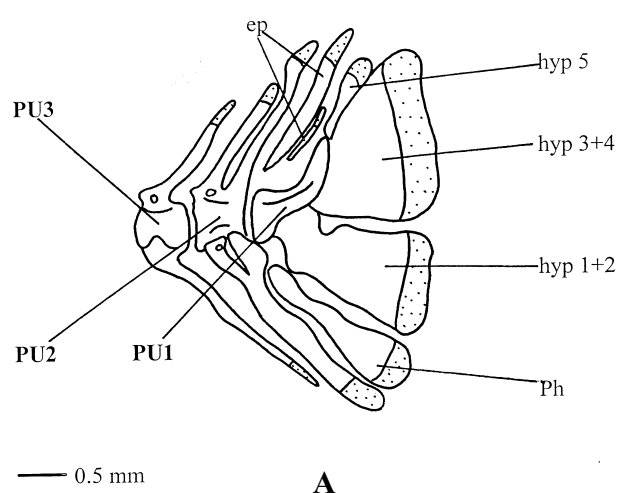


Fig. 4. – A. Caudal endoskeleton of a young turbot at day 29 (LS= 16 mm). B. Caudal endoskeleton of a young turbot at day 61 (LS= 28 mm). PU; preural centrum.

fusion between the third preural centrum and the second preural centrum. At this stage, the hypural plates are large and in contact with the ventral edge of the first preural centrum (PU1), the latter being dorsally curved. At this stage, the posterior epural is still independent, slender and in contact with a large anterior epural. However, at day 61 (Fig. 4B), the posterior epural is fused with the hypural 5 (CHANET & WAGEMANS, 1997). No uroneural is discernible.

DISCUSSION

Different information can be deduced from this description: first, on the development of the endoskeleton among the flatfishes, second, on the setting up of this structure compared to the metamorphosis.

One of the first points to note is that the set-up of the different elements of the caudal skeleton in the turbot is not different to what has been described in three other

bothoid species: one pleuronectid, *Pleuronectes platessa* L., 1758 (BARRINGTON, 1937; COLE & JOHNSTONE, 1902), and two bothids: *Trichopsetta ventralis* (Goode & Bean, 1885) (FUTCH, 1977) and *Engyophrys senta* Ginsburg, 1933 (HENSLEY, 1977). The same bony elements develop in the same pattern and in the same order. Thus, it corroborates previous observations. Moreover, the works of several authors (HENSLEY & AHLSTROM, 1984; CHAPLEAU, 1993; COOPER & CHAPLEAU, 1998) showed that the type of caudal endoskeleton present in the turbot (with hypurals 3 and 4 fused together and to PU1 centrum and hypurals 1 and 2 fused in a plate possessing a ball and socket articulation with the ventral edge of PU1 centrum) is unique among the percomorphs and is characteristic of the flatfishes belonging to the Scopthalmidae, Bothidae, Paralichthyidae, Pleuronectidae and *Brachypleura novaezeelandiae* Günther, 1862 previously referred to “Citharidae” a family recognised now as paraphyletic (CHAPLEAU, 1993). On the basis of this peculiar caudal

endoskeleton, these flatfishes were regrouped in a bothoid group (Fig.1) (HENSLEY & AHLSTROM, 1984; COOPER & CHAPLEAU, 1998). The present description provides data that corroborate the homologies between the different caudal structures, and thus provides elements to confirm the monophyly of the bothoid group.

In the turbot, the eye migration – one the main features of flatfish metamorphosis – occurs between the 23rd and the 60th day of development (WAGEMANS et al., 1998). Then, the caudal endoskeleton is fully developed before the metamorphosis in this species. If we compare this result to what has been described in other bothoids, we can notice that, in each case, the caudal endoskeleton is formed before the eye begins to migrate. FUTCH (1977) noted that, in *Trichopsetta ventralis* (Bothidae), the hypurals were fused when the larva measured 6 mm (SL), whereas the ocular migration occurred when the larva was 28.5-35.7 mm long (SL). In *Engyophrys senta* (Bothidae), the migration begins on 18.9 mm long (SL) larvae and the hypurals are already fused at 4.6 mm (SL) (HENSLEY, 1977). In *Pleuronectes platessa* (Pleuronectidae), the hypurals are already well fused on 15-17 mm long (TL: Total Length) larvae, while the beginning of the migration occurs when the larva is 15 mm long (TL) (COLE & JOHNSTONE, 1902). SCHNAKENBECK (1928) described the morphology of some bothoid larvae, but his data are hardly useful. Nevertheless, in *Glyptocephalus cynoglossus* L. 1758 (Pleuronectidae) caudal elements seem to take place when the larva is 10 mm long and the metamorphosis occurs at 20-26 mm length (SCHNAKENBECK, 1928). In the scophthalmid *Phrynorhombus norvegicus* (GÜNTHER) 1862, the caudal endoskeleton is already well developed when the larva is 7.5 mm long whereas the eye migration takes place between 6 and 10 mm in total length (SCHNAKENBECK, 1928). On the other hand, in Soleidae, the hypurals fuse with the PU1 centrum after metamorphosis. In the sole (*Solea solea* L., 1758), the fusion occurring between hypurals and PU1 appears when the larva is between 18 and 470 mm long (TL) (HENSLEY & AHLSTROM, 1984), while the metamorphosis occurs on 8 mm long (TL) larvae – 18 days after hatching- (WAGEMANS & VANDEWALLE, 1999). In *Dicologlossa cuneata* (Moreau, 1881), the caudal complex develops at 11-22 mm on larvae that have already the two eyes on the right side (LAGARDÈRE & ABOUSSOUAN, 1981).

Pending new evidence, we can only say that the caudal endoskeleton of Pleuronectiformes develops before or after eye migration according to species. Is the relative shift in developmental timing a synapomorphy of the bothoids? Does it mean that heterochronic events occur during the development of some flatfishes? Such hypotheses are yet premature and must be confirmed through the study of many flatfish species -bothoids and non-bothoids. More work on other flatfish species is needed to confirm these ideas.

ACKNOWLEDGEMENTS

We would like to thank here: Dr D. Leclercq (France Turbot, Noirmoutiers, France), Dr E. Lebègue (NATA, Noirmoutiers, France) for the rearing and selecting the studied specimens. We are indebted to Dr P. Janvier (CNRS, URA 12, Paris, France) and Dr. G. Nelson (University of Melbourne, Parkville, Australia) for their advice and acute review of a first version of this article. Dr. F.W is a scientific collaborator researcher at the Fonds pour la Recherche dans l'Industrie et l'Agriculture (FNRS).

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Received: August 4, 2000

Accepted: October 25, 2000