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Anatomical Studies on the Eye Muscle Nerves and Ciliary Ganglion of the Leaping Grey Mullet, *Liza saliens* (Risso, 1810)

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Abstract: The aim of this study is to analyze the ocular muscle nerves and the ciliary ganglion of the bony fish *Liza saliens*. This leaping grey mullet fish was collected from Kafr El-Sheikh and has been undergone to a serious of permanent histological preparation to be willing for examination. The ocular muscle nerves comprise the nervi oculomotorius, trochlearis and abducens. The oculomotor nerve arises from the pars peduncularis mesencephalon of the brain to leave the cranial cavity through its own foramen. It innervates four eye muscles; rectus superior, rectus inferior, rectus medialis and the obliquus inferior muscles. It carries pure somatic motor fibers and visceromotor (parasympathetic) ones. The ciliary ganglion is small and has no radix ciliaris brevis. It has only the radix ciliaris longa that originates from the ramus opthalmicus profundus. Only one ciliary nerve arises from the ciliary ganglion that terminates in the muscle of the iris. The trochlear nerve passes extracranially through its trochlear foramen and carries pure somatic motor fibers to the obliquus superior muscle. The nervus abducens arises from the medulla oblongata by a single root. It leaves the cranial cavity through its own foramen to enter the posterior eye muscle canal (myodome). It carries pure somatic motor fibers to the rectus lateralis muscle.

Key words: Abducens · Ciliary Ganglion · *Liza saliens* · Ocular Muscle Nerves · Oculomotor · Trochlear

INTRODUCTION

The cranial nerves are an important collection of nerves, where, they connect the brain with all the important centers of perception of the outer surface of the head, as well as the inner surface of the buccopharyngeal and other visceral regions. They travel directly to the brain rather than through the spinal cord. The cranial nerves have several functions vital for day-to-day life.

Of the first investigations done on the cranial nerves of osteichthyes were those of Stannius [1] and Goronowitsch [2] on *Acipenser ruthenus*; these classical studies are still useful to investigators. Dakrory [3] on *Ctenopharyngodon idellus*, Dakrory [4] studied the ciliary ganglion and its anatomical relations in some bony fishes, Hussein [5] on *Mugilcephalus*, Taha [6] on *Hypophthalmichthys molitrix*, Matter [7] on *Gambusia affinis affinis* give details account on the morphological anatomy of the cranial nerves.

It is quite evident from the above historical reviews that there are numerous works on the cranial nerves of fishes, but few studies has been carried out concerning the cranial nerves of species belonging to mugillid fishes which is an interesting group among teleosts. Thus it was recommended that a detailed microscopic investigation on the eye muscle nerves in *Liza saliens* belonging to family Mugillidae will be very fruitful.

This investigation will highlight the nerve's origin, route, intracranial pathway, location of exit from the cranium, extracranial course and to its innervation. In addition, the current study will give a concise and comprehensive explanation of the important characters of the nerve including its components, correlated ganglion

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(Ganglia), relevant branches. It is worth mentioning that, they may add some knowledge on this important subject, their distribution is associated with the habits and habitats and behavior pattern of teleosts (Feeding, defense, spawning, schooling orientation, migration, etc.) in addition, they show an evolutionary and phylogeny trend among this group of fishes.

MATERIALS AND METHODS

Liza (Leaping grey mullet) *saliens* (Family: Mugilidae) is a pelagic species found in various habitats, from shallow brackish and marine waters, to lagoons, estuaries, rivers and freshwater lakes [8]. It spawns at sea. In general, *L. saliens* juveniles with a total length shorter than 35 mm feed mainly on animal prey [9] while longer and older juveniles and adults feed mainly on detritus [10].

The fully formed larvae were collected from fish farm in Kafr El-Sheikh Governorate, Egypt. Fish was anesthetized using tricaine methane sulfonate. The heads of the fully formed larvae were separated and fixed in aqueous Bouin's solution for 24 hours. This was followed by several washing with 70% ethyl alcohol for 3-4 days to get rid of excess fixative and its yellow colour from the specimens. Decalcification to head to ensure the success of the following steps. This was carried out by placing the heads in EDTA solution for about 40 days with changing the solution every 4 days. Thereafter, the embryonic heads were washed several times with 70% ethyl alcohol. The heads were treated with ascending series of ethyl alcohol and then cleared with xylene. Thereafter, the specimens were embedded in a paraffin wax. This is followed by sectioning the heads transversely at 10 micron in thickness by Reichert microtome. The serial sections were mounted and stained by Mallory's triple stain [11]. This stain was found effective where cartilage appears blue, nerves and glands in various shades of violet and muscles in red Experimental protocols and procedures used in this study were approved by the Cairo University, Faculty of Science and Institutional Animal Care and Use Committee (IACUC) (Egypt) (CUFS/S/33/15). All the experimental procedures were carried out in accordance with international guidelines for care and use of laboratory animals.

The serial sections were drawn by the aid of a projector microscope. From these graphs a lateral view of an accurate graphical reconstruction for the nervi oculomotorius, trochlearis and abducens and also the ciliary ganglion and the eye was made. In order to avoid confusion of lines, few rami were slightly displaced from their normal position. Also, parts of certain sections were photomicrographed using Zeiss photomicroscope supplied by Canon digital camera to demonstrate the relation of the nerves with other cranial structures.

RESULTS

Nervus Oculomotorius: In *liza saliens*, the nervus oculomotorius arises from the ventrolateral side of the pars peduncularis mesencephalon of the brain by one root (Figs. 1 & 2, RO.III) after the root fibers leave their cells they congregate into a rootlet. The two rootlets, right and left decussate completely inside the brain. The oculomotor nerve runs rostrally (Ventrolaterally) within the cranial cavity extending dorsomedial to the anterior cardinal vein, ventromedial to both the trochlear nerve and profundus ganglion of the nervus trigeminus and ventrolateral to the mid brain. Thereafter, it continues in a forward direction to leave the cranial cavity through its own foramen, the foramen oculomotorius (Fig. 3, F.OC). This foramen lies in the lateral part of the pleurosphenoid bone, ventromedial to the orbital cartilage, dorsal to the rectus lateralis muscle and ventrolateral to the brain. During its exit from its own foramen, the oculomotor nerve divides into two rami: a dorsolateral ramus superior and a ventromedial ramus inferior (Fig. 3)

Ramus Superior: After its separation from the nervus oculomotorius the ramus superior passes in the forward direction and extending dorsolateral to the Ramus inferior, medial to eyeball and dorsolateral to the rectus superior muscle (Fig. 4, R.SP) after a considerable anterior distance, it penetrates the rectus superior muscle from its dorsal side, where it terminates between its fibers.

Ramus Inferior: After its separation from the nervus oculomotorius, the ramus inferior runs anteriorly in a ventromedial direction passing ventromedial to the ramus superior, dorsolateral to the rectus superior muscle and dorsomedial to the ophthalmic vein (Fig. 4, R.IF) here, it gives off a ventromedial branch (Fig. 4, N.OIF) which extends anterior in a ventromedial direction passing ventromedial to the ramus inferior and the ophthalmic vein, lateral to the rectus superior muscle and medial to the rectus lateralis muscle. Shortly forwards the ventromedial branch extend medially passing dorsal to the adductor arcularis palatines muscle, lateral to the rectus medialis muscle, ventrolateral to the origin of the rectus inferior muscle and ventral to blood vessel and the rectus superior muscle. After a short distance medially, the ventromedial branch extend dorsal to the adductor arcularis palatines muscle, dorsolateral to the ramus palatines, ventromedial to the rectus inferior muscle and ventrolateral to the rectus medialis muscle. This branch continues its medial course along the ventral side of the rectus medialis muscle to become ventromedial to the latter muscle. The two branches of both sides approach each other passing medial to the rectus medialis muscle and dorsal to the basisphenoid bone and ventral to the optic chiasma then it becomes medial to the rectus medialis muscle and the optic nerve. Thereafter, the ventromedial branch extend medial to the eyeball and the rectus medialis muscle and lateral to the interorbital septum finally, this branch passes dorsomedial, dorsal then dorsolateral side of the obliquus inferior muscle where it enters this muscle and terminates between its fibers. The main ramus inferior passes anteriorly and approaches to the ciliary ganglion and fused with it (Fig. 4). Slightly anteriorly it gives off a ventromedial nerve. This nerve directly enters the rectus inferior muscle from its ventrolateral side to innervate its fibers.

Anterior to the latter nerve the ramus inferior runs medially passing dorsal to the rectus inferior muscle and dorsolateral to the rectus medialis muscle and ends between the fibers of the latter muscle.

Ciliary Ganglion: In the current study, there is a well-developed ciliary ganglion. This ganglion is located in the postorbital region it measures about 120 microns in length and have the same types of cells. The ciliary ganglion is found in a postion surrounded by the ramus inferior of the nervus oculomotorius dorsally, the rectus lateralis muscle laterally, the rectus superior muscle medially and the ophthalmic vein dorsolaterally (Fig. 4, G.CL). The ciliary ganglion is fused with the ramus inferior of the nervus oculomotorius. During its fusion, the ciliary ganglion receives a branch which is the radix ciliares longa which enters the ganglion from its lateral side (Fig. 4, RCL). The radix ciliaris longa originates from the ramus opthalmicus profundus of the nervus trigeminus outside the cranial cavity. Directly after its exist from the cranial cavity, the ramus opthalmicus profundus of the nervus trigeminus fuses completely with the head sympathetic chain. Shortly forwards, the ramus opthalmicus profundus gives off the truncus ciliaris and the radix ciliaris longa .the latter runs anteromedially, ventral to the ophthalmic vein, dorsomedial to the rectus lateralis muscle to enter the ciliary ganglion from its lateral side. The ciliary ganglion gives off from its anterior end, one ciliary nerve (Fig. 5, N.CL). This ciliary nerve runs forwards in a lateral direction accompanying the ophthalmic vein lateral to the ramus inferior and medial to the eyeball. After a long course the ciliary nerve become in a position lateral to the ophthalmic vein, medial to the eyeball, dorsolateral to the rectus inferior and rectus medialis muscles and ventromedial to the rectus superior muscle. It shifts laterally passing between the two arms of the oblique u shaped ophthalmic vein (Fig. 6, N.CL) lateral to the ophthalmic vein, the ciliary nerve runs anteroventrally on the ventral surface of the ophthalmic vein and medial to the eyeball, dorsal to the rectus inferior muscle. After a short distance, it enters the eyeball with the ophthalmic vein through a foramen in the sclera for the passage of the optic nerve and the ophthalmic vein. Within the eyeball, the ciliary nerve passes to the muscle of the iris, where it terminates.

Nervus Trochlearis: In the studied *liza saliens*, the nervus trochlearis originates from the lateral side of the posterior part of the midbrain by a small single root (Fig. 7, RO.IV). The roots of the two opposite nervus decussate completely within the brain forming trochlear decussation and leave the brain, the right nerve from the left side and the left nerve from the right one. After its origin from the brain the trochlear nerve extends ventrolateral in the anterior direction passing lateral to the brain and dorsomedial to the nervus trigeminus and anterodorsal lateral line nerve and dorsomedial to the geniculate ganglion of the nervus facialis. After that, the nervus trochlearis continues passing medial to the anterior part of the profundus ganglion, lateral to the brain, dorsolateral to the root of the nervus oculomotorius. More forwards, the nervus trochlearis continues passing medial to both the orbital cartilage and the ramus opthalmicus superficialis trigeminus, dorsal to the laterosphenoid bone. After along anterior course, the nervus trochlearis leaves the cranial cavity through its own foramen (Fig. 8, F.T). This foramen is found in the laterosphenoid bone. Extracranially, the nervus trochlearis continues forwards passing ventral to the cranial wall, dorsomedial to the rectus superior muscle. Thereafter, it becomes ventromedial to the orbital cartilage and medial to the rectus superior muscle and dorsomedial to the eyeball and ventrolateral to the brain. More anteriorly, it continues passing lateral and ventrolateral to the orbital

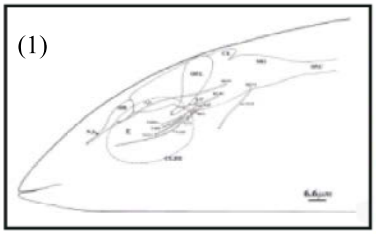
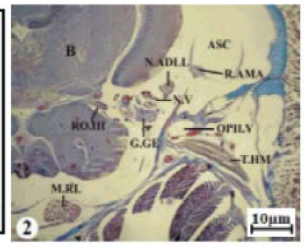
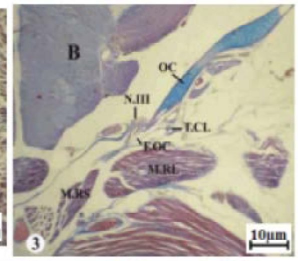
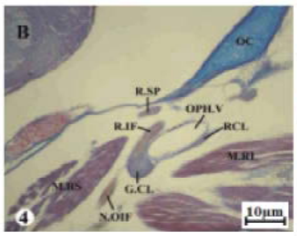
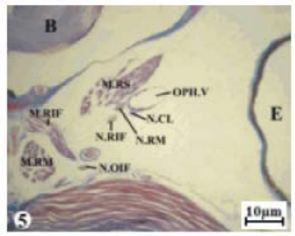
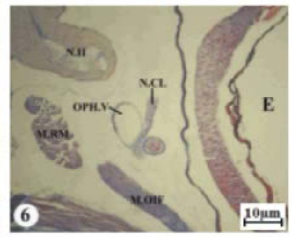
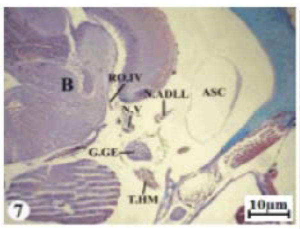
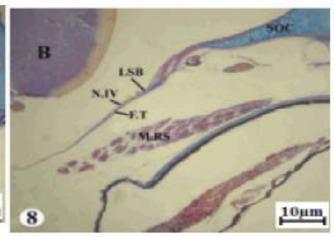
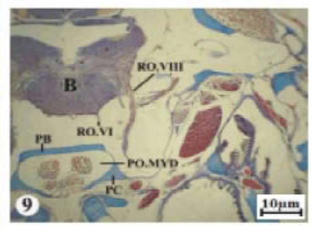
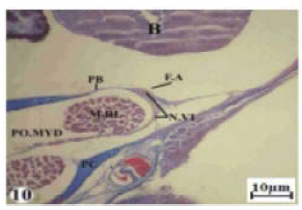

Fig. 1: Graphic reconstruction of the brain, the nervi oculomotorius, trochlearis and abducens and the eye of *Liza saliens* in a lateral view

Fig. 2: A photomicrograph of a part of transverse section passing through the mesencephalon of *Liza saliens* illustrating the root of the nervus oculomotorius

Fig. 3: A photomicrograph of a part of transverse section passing through the postorbital region of *Liza saliens* illustrating the exit of the nervus oculomotorius through the foramen oculomotorius

Fig. 4: A photomicrograph of a part of transverse section passing through the postorbital region of *Liza saliens* showing the two rami superior and inferior of nervus oculomotorius, coalescence between the ramus superior and the ciliary ganglion and radix ciliaris longa

Fig. 5: A photomicrograph of a part of transver section passing through the postorbital region of *Liza saliens* illustrating the ciliary ganglion giving off one ciliary nerve

Fig. 6: A photomicrograph of a part of transverse section passing through the orbital region of *Liza saliens* showing the ciliary nerve between two arms of the oblique U shaped ophthalmic vein

Fig. 7: A photomicrograph of part of transverse section passing through the anterior otic region of *Liza saliens* illustrating the root of the nervus trochlearis

Fig. 8: A photomicrograph of part of transverse section passing through the orbital region of *Liza saliens* illustrating the trochlear foramen and the trochlear nerve

Fig. 9: A photomicrograph of part of transverse section passing through the otic region of *Liza saliens* illustrating the root of the nervus abducens

Fig. 10: A photomicrograph of part of transverse section passing through the postorbital region of *Liza saliens* illustrating the abducens foramen and the abducens nerve entering the posterior myodome

List of Abbreviations:

ASC: Anterior semicircular canal; B: Brain; BSB: Basisphenoid bone; E: Eye; F.A: Foramen of abducens nerve; F.OC: Foramen of oculomotor nerve; F.T: Foramen of trochlear nerve; G.CL: Ciliary ganglion; G.GE: Geniculate ganglion; LSB: Laterosphenoid bone; M.OI: Obliquus inferior muscle; M.OS: Obliquus superior muscle; M.RI: Rectus inferior muscle; M.RL: Rectus lateralis muscle; M.RM: Rectus medialis muscle; M.RS: Rectus superior muscle; OB: Olfactory bulb; N.ADLL: Anterodorsal lateral line nerve; N.CL: Ciliary nerve; N.II: Nervus opticus; N.III: Nervus oculomotorius; N.IV: Nervus trochlearis; N.OIM: Nerve to the obliquus inferior muscle; N.RI: Nerve to the rectus inferior muscle; N.RM: Nerve to the rectus medialis muscle; N.V: Nervus trigeminus; N.VI: Nervus abducens; OC: Orbital cartilage; OPH.V: Opthalmic vein; PB: Prootic bridge; PC: Prootic cartilage; PO.MYD: Posterior myodome; PSB: Pleurosphenoid bone; R.AMA: Ramus ampullaris anterior; R.IF: Ramus inferior; R.SP: Ramus superior; RCL: Radix ciliaris longa; RO.III: Root of nervus oculomotorius; RO.VI: Root of nervus abducens; RO.VIII: Root of nervus octavus; SC: Sclera; SOC: Supra orbital cartilage; T.HM: Truncus hyomandibularis

cartilage, ventral to the supraorbital cartilage and dorsomedial to the eyeball and ventrolateral to the brain. At the anterior half of the orbital region, nervus trochlearis continues forwards passing lateral to the orbital cartilage and dorsomedial to the obliqus superior muscle. Finally, it enters the latter muscle from its dorsal side, where it terminates between its fibers. The nervus trochlearis carries somatic motor fibers.

Nervus abducens: The nervus abducens originates from the ventral side of the medulla oblongata by means of a single root (Fig. 9, RO.VI), where its origin lies ventromedial to the origin of the anterior branch of the octaval nerve. Directly after its origin, it runs intracranially in a ventral direction passing ventral to the brain and ventromedial to the nervus facialis and octaval ganglion and dorsal to the cartilaginous prootic bridge that form the roof of the posterior myodome. Thereafter, it continues anteriorly extending ventromedial to the geniculate ganglion of the nervus facialis and dorsal to the prootic bridge. At the end of its intracranial course, the nervus abducens leaves the cranial cavity and enters the posterior myodome through abducens foramen (Fig. 10, F.A). This foramen lies between the prootic bridge and the prootic cartilage. Within the myodome, it moves forwards passing dorsolateral to the rectus lateralis (Externus) muscle. After a distance in this position, the nervus abducens enters and ends between the fibers of the latter muscle. The nervus abducens carries somatic motor fibers.

DISCUSSION

In the current study, complete decussation was observed in the nervus oculomotorius within the brain near its origin. This finding was observed by Hussein [5] in *Mugilcephalus*, Taha [6] in *Hypophthalmichthys molitrix* and Matter [7] in *Gambusia affinis affinis*. The nervus oculomotorius brings its exit from the cranial cavity through its own foramen, the foramen oculomotorius. Within bony fishes, the nervus oculomotorius leave the cranial cavity through its own foramen as in *Tilapia zillii* [13] *Hypophthalmichthys molitrix* [6] and *Gambusia affinis affinis* [7].

In the present work, the nervus oculomotorius is divided into its two rami superior and inferior during its exit from its specific foramen. This condition agreed with Ray [14] in *Lampanyctus leucopsarus*. The division of the nervus oculomotrius out the cranial cavity was reported by Taha [6], Matter [7] and by Dakrory *et al.* [12] in *Hypophthalmichthys molitrix*, *Gambosia affinis affinis* and *Liza ramada* respectively. In the teleost *Alticus kirkii magnosi* [15] the nervus oculomotorius is divided within the cranial cavity into a posterior branch to the rectus superior muscle and an anterior branch to the other three muscles.

In this study, the ciliary ganglion has only one type of neurons. A similar case was found in *Mugil cephalus* [4]. On the other hand, such ganglion consists of large and small neurons in the batoid *Rhinobatus halavi* and the cyprinid *Ctenopharyngodon idellus* [3] and in *Gambusia affinis affinis* [4] *Tilapia zillii* [13] *Hypophthalmichthys molitrix* [6] and *Gambusia affinis affinis* [7].

In the present study, the sensory root, the radix ciliaris longa, originates from the ramus opthalmicus profundus of the nervus trigeminus. A similar finding was reported in *Mugil cephalus* [4, 5] and in *Gambusia affinis affinis* [7]. On the other hand, it was reported in both the batoid *Rhinobatus halavi*and the cyprinid *Ctenopharyngodon idellus* [3] that the radix ciliaris longa originates from the Gasserian ganglion of the nervus trigeminus.

In the present investigation, there is no radix ciliaris brevis and the motorcomponent of the nervus oculomotorius is transmitted directly to the ganglion through the coalences between them. A similar finding was reported among bony fishes by Dakrory [4] in the cyprinid *Ctenopharyngodon idellus* and in both *Mugil cephalus* and *Gambusia affinis affinis*, by Ali and Dakrory [15] in *Alticus kirkii magnosi*, Taha [6] in *Hypophthalmichthys molitrix* and by Matter [7] in *Gambusia affinis affinis*. On the other hand, a well-developed motor root, the radix ciliaris brevis, was observed in *Tilapia zillii* [13].

In the current study, the *Liza saliens* showed that in addition to the ciliary nerve, a truncus ciliaris profundus enters the eyeball through a foramen excavated in the dorsal side of the sclera just ventral to the obliquus superior muscle. This finding shows that the eye is accommodated by both the ciliary nerve and the truncus ciliaris. Therefore, this reflects the fact that the eye is well developed in the studied fish and this fish depends on vision during its feeding. So, it may be considered as a diurnal animal. A similar observation was found also by Taha [6] in *Hypophthalmichthys molitrix* and Matter [7] in *Gambusia affinis affinis*. In *Polypterus senegalus* [16] described two ciliary nerves.

In this study, the nervus trochlearis originates from the cranial cavity through a special foramen, the trochlear foramen that located in the laterosphenoid bone. A similar finding was found in some fishes such as *Trichiurus lepturus* [17] in the cyprinid fish *Ctenopharyngodon idellus* [3] *Alticus kirkii magnosi* [15], *Hypophthalmichthys molitrix* [6] and *gambusia affinis affinis* [7]. In *Clarias batrachus*, there is a common foramen for the exit of the nervi oculomotorius, trochlearis, abducens and the trigemino-facial complex [18]. In the Goldfish, *Carassius auratus*, the nervus trochlearis leaves the brain case with the ramus ophthalmicus superficialis through an opening on the optic tectum [19]. Nakae and Sasaki [20] reported that the trochlear nerve in *Molamola* emerges from the cranium through the anterior part of the suture between the pterosphenoid and basisphenoid.

In the current study, a trochlear decussation of the two sides of trochlear nerves inside the brain is recorded. This data was in agreement with that observed in *Polypterus senegalus* [16] and in *Mugil cephalus* [5]. However, no trochlear decussation as reported by Ali [13] in *Tilapia zillii*, Ali and Dakrory, [15] in *Alticus kirkii magnosi* and Matter [7] in *Gambusia affinis affinis.*

In the present study, the nervus abducens originated from the medulla oblongata by a single root as reported in *Ctenopharyngodon idellus, Tilapia zilli, Mugil cephalus, Hypophthalmichthys molitrix, Liza ramada* [3, 5, 6, 12, 13] respectively.

The present study proved that the nervus abducens emerges from the cranial cavity through a foramen excavated between the prootic bridge and the prootic cartilage. This is the same as recored in *Hypophthalmichthys molitrix* [6]. While the nervus abducens leaves the cranial cavity through a foramen on the prootic bridge as reported in *Alticus kirkii magnosi* [15]. However, among bony fishes, the exit of the nervus abducens from the cranium was observed through a special foramen as in *Trichiurus lepturus*, *Ctenopharyngodon idellus* [3, 17]. On the other hand, Piotrowski and Northcutt [16] and El-Toubi and Abdel-Aziz [21] dealing with *Polypterus senegalus*, observed that from the foramen trigeminal, the nervus abducens emerges from the cranial cavity with the nervus trigeminus. In *Clarias batrachus*, the nervus abducens issues from the cerebral cavity together with the trigeminofacial complex, through the foramen prooticum [18]. In addition, in *Amphipnous cuchia*, Saxena [22] reported that the nervus abducens runs out of the cranial cavity with the nervus opticus, through a foramen located in the lateral ethmoid bone.

In the present work, the nervus abducens, as in all vertebrates, innervates the rectus lateralis (Externus) muscle. This state was reported in some fishes [3, 6, 7, 13, 15, 20]. In *Tridentiger trigonocephalus*, the rectus externus muscle composed of two different fibers. The latters are innervated by two distinct nerve bundles [23]. However, in *Latimeria chalumnae* and in many tetrapoda, the nervus abducens innervates the rectus externus and the retractor oculi muscles [24].

This current study reported that, the rectus lateralis muscle is located within the posterior myodome (The eye muscle chamber). Others recorded the presence of this myodome in *Ctenopharyngodon idellus* [3] and *Tilapia zillii* [13].

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