

4-2009

Functional Morphology of the Superficial Branchial Musculature of the Dogfish Shark, *Squalus acanthias*

Roy Andermann

Follow this and additional works at: https://digitalcommons.lsu.edu/honors_etd



Part of the [Biology Commons](#)

Functional Morphology of the Superficial Branchial Musculature
of the Dogfish Shark, *Squalus acanthias*

by

Roy J Andermann Jr.

Undergraduate Honors Thesis

Directed by

Dr. Dominique G. Homberger
Professor
Department of Biological Sciences

Submitted to the LSU Honor's College in partial fulfillment of the
Upper Division Honor's Program

April 2009

Louisiana State University and Agricultural & Mechanical College
Baton Rouge, Louisiana

Abstract

Ventilation in sharks and rays, as in piscine vertebrates in general, is a complex mechanical process with one full ventilatory cycle involving a two-step pumping mechanism based on the action of an array of constrictor and adductor muscles of the branchial arches.

Preliminary observations indicated, however, that the standard descriptions of these muscles in sharks were inaccurate and did not lend themselves to a functional analysis of the branchial apparatus. Hence, a new functional-morphological analysis of the superficial branchiomic musculature in the dogfish shark (*Squalus acanthias*) was undertaken within the context of the entire skeleto-muscular system as part of a larger project re-evaluating the evolutionary morphology of sharks and vertebrates. Micro-examination of the origin, insertion, and three-dimensional configuration of the cucullaris muscle and superficial branchial constrictor muscles yielded new morphological details. The cucullaris muscle can be separated into two functionally and evolutionarily distinct moieties, and the dorsal and ventral superficial branchial constrictor muscles can also be subdivided into two portions with separate functions. Based on these new morphological data, the superficial branchial constrictor muscles and one portion of the cucullaris muscle can be identified as muscles that are likely to have been present already in the earliest fishlike ancestors of sharks, whereas the other portion of the cucullaris muscles evolved secondarily from the dorsal superficial branchial constrictor muscles through a process called “scissoring”. The superficial branchiomic muscles, furthermore, are likely to be involved in the undulatory locomotion of the shark, during which not only the trunk, but also the branchial region bends sideways.

Table of Contents

1. Introduction	5
2. Materials and Methods.....	6
2.1 Materials.....	6
2.2 Methods	7
3. Results	9
3.1 The visceral skeleton	9
3.2 The superficial branchial constrictor muscle complex	10
3.2.1 The dorsal superficial branchial constrictor muscles.....	10
3.2.2 The trematic branchial constrictor muscles.....	12
3.2.3 The ventral superficial branchial constrictor muscles	12
3.3 The cucullaris muscle.....	13
3.4 The epibranchial musculature	14
3.5 Movement of the branchial region during undulation.....	15
4. Discussion.....	15
4.1 The significance of morphology for macrotaxonomy and macroevolution	15
4.2 Marion's (1905) description of dorsal superficial branchial constrictor tendons	16
4.3 Biomechanical analysis	17
4.3.1 Biomechanical analysis of the dorsal superficial branchial constrictor muscles	17
4.3.2 Biomechanical analysis of the trematic branchial constrictor muscles	18
4.3.3 Biomechanical analysis of the ventral superficial branchial constrictor muscles.....	18
4.3.4 Biomechanical analysis of the cucullaris muscle	20
4.3.4.1 Insulation of the lateral line	20
4.3.4.2 Function of the scapulocoracoid portion	20
4.3.4.3 Function of the pharyngoepibranchial portion	21
4.3.4.4 Physiologic implications of the alterations of cucullaris muscle fasciculi near the dorsal superficial branchial constrictor muscle tendons.....	21
4.3.5 Biomechanical analysis of the epibranchial musculature	22
4.4 General functional and evolutionary implications	22
4.4.1 The configuration of crossing muscles in sharks.....	22
4.4.1.1 Hypothesis 1: Reduction of the superficial dermal bony plates.....	23

4.4.1.2 Hypothesis 2: Gradual differentiation from a single muscular body; fusion of levator muscles into the cucullaris of derived species.....	26
4.5 Functional significance of the epibranchial musculature	29
4.6 The concatenation of the organ systems in the selachian head	30
4.7 Concluding remarks	30
5. Acknowledgements	31
6. Literature Cited	32
7. Appendices	35
7.1 Appendix I: Tables.....	35
7.2 Appendix II: Figures	40

1. Introduction

Fishes extract oxygen from and release carbon dioxide into the surrounding water by ventilating their gills through a concerted action of the branchiomic musculature. The functional morphology of the gill apparatus of sharks and rays (Chondrichthyes: Subclass Elasmobranchii, Bond 1996) is better known than that of bony fishes (Sarcopterygii and Actinopterygii, Bond 1996) and chimaeras (Chondrichthyes: Subclass Holocephali, Bond 1996) in part because each gill pouch opens on the surface of the body and is, therefore, directly accessible for inspection and experimentation. In bony fishes and chimaeras, in contrast, the individual gill pouches are protected and hidden by a single operculum, which is ossified in bony fishes and fleshy in chimaeras. The operculum in these two taxa was independently and secondarily evolved (Lund & Grogan 1997; Mallatt 1997b). Thus, the configuration of the branchiomic musculature differs in sarcopterygians and actinopterygians, elasmobranchs, and holocephalans, even though it performs the same vital role for gill ventilation.

The general morphology of the musculature of the branchial region of sharks has been described by numerous authors for at least the past century (e.g., Marion 1905; Marinelli & Strenger 1959; Hughes 1960; Hughes & Ballintijn 1965; Metcalfe & Butler 1986; Ferry-Graham 1999; Gudo & Homberger 2002; Summers & Ferry-Graham 2003; Wilga *et al.* 2007). It can be separated into three main muscle groups: (1) the epibranchial muscles are located above the branchial basket and are continuous with the epaxial musculature of the body. They tend to be addressed in studies of feeding behavior and jaw mechanics (Wilga & Motta 1998; Wilga *et al.* 2007) rather than in studies of gill ventilation. (2) The branchiomic muscles are associated with the branchial arches of the branchial basket. They are most frequently studied in analyses

of gill ventilation as they compress the branchial arches (Hughes 1960; Hughes & Ballintijn 1965; Ferry-Graham 1999; Summers & Ferry-Graham 2003). (3) The hypobranchial muscles are located underneath the branchial basket. According to Hughes & Ballintijn (1965), they are usually not active during ventilation and function primarily during feeding. A closer re-examination of the origin and insertion sites of these muscles groups, however, indicated that they are more complex than previously thought. This necessitated a morphological and functional re-analysis.

A common model organism to study elasmobranch ventilation, and the organism used in the present study, is the Spiny Dogfish, *Squalus acanthias* (Squaliformes). *Squalus acanthias* has retained several traits that are ancestral among extant sharks. They possess a fusiform body, a spine in front of the dorsal fins, and an unspecialized spiracle that is neither enlarged for the intake of respiratory water, as in rays, nor reduced, as in many pelagic sharks. According to Wilga & Motta (1998), *Squalus acanthias* has also retained the basic kinematic feeding sequence observed in lower aquatic vertebrates. Nevertheless, the Spiny Dogfish is a model for the ancestral elasmobranch condition only to a limited degree, as Mesozoic and extant sharks differ significantly from Paleozoic species (Grogan & Lund 2009).

2. Materials and Methods

2.1 Materials

Four specimens (three female and one male) of *Squalus acanthias* were obtained from NASCO Biological Supply Company (Fort Atkinson, Wisconsin) and were stored in a 1% 2-phenoxyethanol solution. The three female specimens together represent a series of post-

embryonic shark development: A juvenile (# RJA001) with a total length of 611 mm; a small adult (# RJA002) with a total length of 749 mm; and a large pregnant adult (# RJA003) with a total length of 1132 mm. Observations of the male shark (# RJA004) with a snout to caudal peduncle length of 525 mm (estimated total length of 664 mm, because the tail was lost during the embalming process) showed that the branchial and branchiomic structures apparently are not sexually dimorphic.

2.2 Methods

The specimens were dissected with a pair of manually sharpened precision Watchmaker's forceps (Carolina Biological Supply Company, Burlington, North Carolina) under a stereomicroscope (Wild M3 TYP 355110 with a discussion tube; Leica, Heerbrugg, Switzerland) with a fiber optic ring light (Volpi, Schlieren, Switzerland) fitted with a light-polarizing filter and attached to a Volpi NCL 250 cold light box.

Macroscopic digital photographs of the specimens were obtained with a SPOT Insight Color Closed-Circuit TV Camera and SPOT Advanced Imaging Software (Diagnostic Instruments, Sterling Heights, Michigan). Three separate lenses were used with the camera to provide appropriate fields of view from the least to the greatest magnification: (1) a 6.5 mm Computar TV lens 1:1.8 (object to lens distance 250-775 mm, field of view 378mm x 630mm to 816mm x 1360mm); (2) a 12.5 mm Goldinar TV lens 1:1.3 (object to lens distance 190-1000 mm, field of view 138mm x 230mm to 522mm x 870 mm); and (3) a 25 mm National Electronic CCTV lens F/1.4 (object to lens distance 375 to 1000 mm, field of view 126mm x 210mm to 309mm x 515 mm). The camera was mounted on a Bencher copy stand with an Illuma System Light Control

(Bencher Inc., Antioch, Illinois). The copy stand included 4 Sun-Lite UL E196460 Portable Luminaires, which each mounted a GE Reveal 120 watt/120 volt incandescent bulb with a neodymium coating, which filters out yellow light. When taking pictures, the fluorescent ceiling lights were turned off to avoid glare and flickering. To further reduce glare, the positions of the light bulbs were adjusted and diffusers (i.e., plexiglass covered with visquine) were placed in front of the light bulbs to provide even illumination.

Microscopic digital photographs were obtained with a SPOT Insight Firewire Camera mounted on a Leica MZ6 microscope. The images were processed with ImagePro version 4.5 Software (Media Cybernetics, Bethesda, Maryland). The microscope and mounted camera, as well as the specimens, were placed on a Micro-g anti-vibration table (Model 63-551; Technical Manufacturing Corporation, Peabody, Massachusetts) to prevent vibrations and to ensure a sharp image.

Each digital photograph was retaken with a metric scale at an equal distance to the lens as the point of focus in the original photograph. This metric scale was used to create a scalebar in Adobe Photoshop CS2 to be incorporated into the final image. Each image was processed with Adobe Photoshop CS2 to maximize the light levels and contrast, and to add descriptive labels without modifying the image contents itself.

To determine whether the branchial region participates in locomotory undulation, a video (kindly provided by Anabela Maia and Dr. Cheryl Wilga, University of Rhode Island, Kingston) of one undulation of a specimen of *Squalus acanthias* (total length of 660 mm) swimming at 0.75 body lengths per second was analyzed. Sequential still pictures were captured using Adobe

Photoshop CS2, and the displacement of the rostrum from the midline was determined by measuring the angle of deviation from a virtual line perpendicular and rostral from the scapulocoracoid cartilage.

3. Results

3.1 The visceral skeleton

The splanchnocranium, or viscerocranium, of sharks is composed of seven visceral arches (Figure 1), except the hexanchiform Sixgill Sharks (*Hexanchus spp.*, *Chlamydoselachus anguineus*) and Sevengill Sharks (*Heptranchias perlo*, *Notorhynchus cepedianus*), which have eight and nine visceral arches, respectively. The first visceral arch (i.e., the jaws or mandibular arch) comprises the dorsal palatoquadrate cartilage and the ventral mandibular cartilage. The second visceral arch (i.e., hyoid arch) comprises three elements, namely the dorsal hyomandibular cartilage, the ceratohyal cartilage, and the ventral basihyal cartilage. The third to seventh visceral arches are the branchial arches associated with the gills (branchial arches 1-5). Each is comprised of a series of articulated cartilages (from dorsal to ventral): the pharyngobranchial cartilages, the epibranchial cartilages, the ceratobranchial cartilages, the hypobranchial cartilages, and the basibranchial cartilages. The pharyngobranchial and epibranchial cartilages of the fifth branchial arch are fused together to form the pharyngoepibranchial cartilage, which is further fused to the pharyngobranchial cartilage of the fourth branchial arch. The pharyngobranchial cartilages are articulated with the vertebral column.

3.2 The superficial branchial constrictor muscle complex

The superficial branchial constrictor muscles comprise the contiguous dorsal superficial branchial constrictor muscles, trematic branchial constrictor muscles, and ventral superficial branchial constrictor muscles. These three main muscle groups are associated in series with each gill slit and gill pouch. Each of the four successive muscles is separated by tendinous myosepta (Figures 2, 3, 4). Though five gill slits are present, only four superficial branchial constrictor series are present because the first (rostralmost) gill slit is associated with the hyoid arch, and it is compressed by the hyoid constrictor muscles (dorsal hyoid constrictor muscle, hyoid trematic constrictor muscle, and ventral hyoid constrictor muscle) instead of superficial branchial constrictor muscles.

At first glance, the superficial branchial constrictor muscles seem to arise from the surface of adjacent medio-dorsal and medio-ventral muscle groups, namely the dorsal superficial branchial constrictor muscles from the cucullaris muscle and the ventral superficial branchial constrictor muscles from the coracoarcual muscle (Homberger & Walker 2004: 134). In reality, however, this is not the case (see 3.2.1, 3.2.3) as can be shown in this study.

3.2.1 The dorsal superficial branchial constrictor muscles

The dorsal superficial branchial constrictor muscles comprise four muscle segments, which are separated from each other by myosepta that are supported by a dorsal extrabranchial cartilage (Marion 1905; Mallatt 1997a; Homberger & Walker 2004: 134). The first dorsal superficial branchial constrictor muscle follows the caudal border of the dorsal hyoid constrictor muscle, and the fourth dorsal superficial branchial constrictor muscle lies rostral to the scapulocoracoid

cartilage (Figure 2). Each dorsal superficial branchial constrictor muscle can be subdivided into two distinct portions (Figure 3): (1) the vertebroseptal portion is situated adjacent and ventral to the cucullaris muscle, and (2) the interseptal portion situated between the vertebroseptal portion dorsally and the trematic branchial constrictor muscle ventrally.

The muscle fiber bundles of the vertebroseptal portion of the first, second, and third dorsal superficial branchial constrictor muscle attach to the dorsal end of the rostral myoseptum and converge caudo-dorsally into a constrictor tendon, which passes through the obliquely oriented cucullaris muscle and the epibranchial musculature to attach to the vertebral column (Figures 5, 6, 7, 8, 9). The muscle fiber bundles of the cucullaris muscle directly above and underneath the constrictor tendon become tendinous, presumably through myotendinous junctions with the rostral and caudal parts of these muscle fiber bundles (Figure 7). These individual cucullaris tendon fiber bundles interweave with individual tendon fibers of the constrictor tendons at their crossing point (Figure 7). The first, second and third constrictor tendons continue medially towards the vertebral column by piercing first through the fascia enveloping the epibranchial musculature and then traversing the epibranchial musculature by piercing through its individual internal tendons (Figures 8, 9). For some of its length, the third constrictor tendon runs medially towards the vertebral column between the anterior cardinal sinus and the overlying cucullaris muscle and epibranchial musculature (Figure 10). The fourth dorsal superficial branchial constrictor muscle is juxtaposed to the scapulocoracoid cartilage and the analogous vertebroseptal portion attaches to the caudally adjacent scapulocoracoid cartilage.

The constrictor tendons can be discerned from the surrounding loose connective tissue due to the densely packed and unidirectional nature of the striated collagen fibrils which are iridescent under polarized light (Figures 6, 9). The tendinous fibers of the constrictor tendon fan out as they approach the vertebral column and thereby increase their attachment areas to such an extent that the attachment sites of the constrictor tendons are contiguous (Figure 9).

The muscle fiber bundles of the first, second and third interseptal portions attach to the rostral myoseptum between the attachment of the vertebroseptal portion and the ventral end of the myoseptum above the gill slit and run caudodorsally to attach to the caudal myoseptum. The fourth interseptal portion attaches caudodorsally to the scapulocoracoid cartilage. The ventralmost muscle fiber bundles next to the gill openings are contiguous to the muscle fiber bundles of the trematic branchial constrictor muscle (see Section 3.2.2).

3.2.2 The trematic branchial constrictor muscles

The muscle fiber bundles of the trematic branchial constrictor muscles are contiguous dorsally and ventrally with the muscle fiber bundles of the dorsal and ventral interseptal portions of the superficial branchial constrictor muscles. The muscle fiber bundles attach along the caudal myoseptum dorsally and ventrally of the gill opening (Figure 2) so that they run dorsoventrally over the surface of the gill flap.

3.2.3 The ventral superficial branchial constrictor muscles

There are four ventral superficial branchial constrictor muscles, which are separated from each other by tendinous myosepta, each of which is supported by a ventral extrabranial cartilage.

The first ventral superficial branchial constrictor muscle follows caudally the ventral hyoid constrictor muscle, and the fourth ventral superficial branchial constrictor muscle lies rostral to the scapulocoracoid cartilage (Figure 4). In this respect, they show a configuration corresponding to that of the dorsal superficial constrictor muscles, with which they form a functional unit for each gill pouch. The orientation of the fasciculi of the ventral constrictor muscles is dorso-rostral to ventro-caudal.

The ventral superficial branchial constrictor muscles can be divided into an interseptal portion and a ventroseptal portion. The fasciculi of the first, second, and third interseptal portion attach to the rostral myoseptum, travel ventro-caudally and attach to the caudal myoseptum. The fourth interseptal portion attaches to the rostral myoseptum, also travels ventro-caudally, and attaches to the scapulocoracoid cartilage. The fasciculi of the ventroseptal portion vary irregularly in their origin. Some fasciculi send tendons superficial to the coracoarcual muscle, some send tendons rostral to the large coracoarcual muscle, some send tendons deep to the coracoarcual muscle, some send tendons into the epimysium of the coracoarcual muscle, and, finally, some send tendons into and constitute part of the pseudomyosepta of the coracoarcual muscle. The midventral raphe serves as the origin for the tendons that are sent superficial or rostral to the coracoarcual muscle, and also the tendons that are continuous with the pseudomyosepta of the coracoarcual muscle (Figure 11).

3.3 The cucullaris muscle

The cucullaris muscle is visible superficially between the dorsal superficial branchial constrictor muscle group and the epibranchial musculature, but in reality, it overlaps the ventral portion of

the epibranchial musculature and it is partly covered by the dorsal superficial branchial constrictor muscles (Figure 12). Its superficially visible shape, therefore, differs from its actual shape (Figures 12, 13). The cucullaris muscle appears to be a single sheet-like muscular body at its origin along a horizontal line underneath the lateral line organ, where its muscle fiber bundles become tendinous, presumably through myotendinous junctions, and attach to the internal side of the fascia that envelops the epibranchial musculature and is anchored to the internal surface to the superficial fascia (Figure 14). From their origin, the muscle fiber bundles run caudo-ventrally toward their insertions to two separate skeletal elements, so that two portions can be distinguished for the cucullaris muscle: (1) The scapulocoracoid portion, which originates along the rostral edge of the scapulocoracoid cartilage (Figure 13); and (2) the pharyngoepibranchial portion, which originates from the fused pharyngoepibranchial cartilage of the fifth branchial arch, which is further fused to the pharyngobranchial cartilage of the fourth branchial arch (Figures 1, 12).

The sheet-like cucullaris muscle is pierced by three constrictor tendons (see Section 3.2.1).

Where the constrictor tendons pass, the cucullaris muscle fiber bundles immediately above and underneath the constrictor tendon are tendinous (for details, see Section 3.2.1; Figures 7, 10).

3.4 The epibranchial musculature

The epibranchial musculature is an extension of the epaxial musculature rostral to the scapulocoracoid cartilage that inserts on the occipital region of the endocranium (Figures 1, 2, 3). The muscle fiber bundles are very short and thick, and numerous cone-shaped myosepta subdivide the muscle into myomeres that exhibit a zig-zag-shape on the surface (Figure 3).

Three dimensionally, the myosepta form a complex cone-in-cone shape, and the apices of the zigzags seen on the surface are exaggerated medially. Most muscle fiber bundles run nearly parallel to the vertebral column, and they originate from a rostral myoseptum and insert on the caudal myoseptum (Figures 2, 3).

3.5 Movement of the branchial region during undulation

Based on the analysis of the video of an individual *Squalus acanthias* undulating at a steady cruising speed, it appears that the head is involved in the undulation during locomotion. The rostrum deviates from the midline by an angle of 4.2 degrees just as a new undulation begins in the cranial trunk muscles (Figure 15). The head is bent laterally just before cranial epaxial muscle contraction on the ipsilateral side of the body.

4. Discussion

4.1 The significance of morphology for macrotaxonomy and macroevolution

Because the muscles in sharks generally are not spindle-shaped, but rather sheet-like, they are not always easy to delimit and identify. Probably as a result, earlier descriptions of the structure and function of muscles in sharks are inconsistent and occasionally contradictory. The function of the skeletomuscular system is derived from its configuration, so uncertainties in the morphology prevent a realistic biomechanical analysis. Furthermore, because most inferences in macrotaxonomy and macroevolution are based on morphological data, flawed functional-morphological data may call into question our current understanding of vertebrate evolutionary history. As a classic example of comparative anatomy, the cucullaris muscle can be tracked

from its genesis in basal gnathostomes (jawed vertebrates), often using sharks as a model organism, through transmutations in intermediate organisms, such as amphibians, to its expression as the trapezius muscle in mammals. Therefore, if the basic description of the cucullaris muscle of sharks is incorrect, the interpretations of its evolution through the rest of the vertebrates are incorrect, too.

A compilation of earlier descriptions of the superficial branchiomic musculature is provided in Table 1, which documents that earlier morphological descriptions and functional interpretations vary often significantly and are, therefore, unreliable. Many of the secondary sources appear to have uncritically accepted earlier descriptions and interpretations. Our re-analysis of the superficial branchiomic musculature was, therefore, needed to open new avenues for re-evaluating not only the anatomy and classification of sharks, but also the evolutionary history of vertebrates.

4.2 Marion's (1905) description of the dorsal superficial branchial constrictor tendons

The description by Marion (1905: 897) of the constrictor tendons (Figures 16, 17) has been completely overlooked in the past century, even though it has substantial functional and evolutionary significance:

“Towards the dorsal margin some of the [muscle] fibers converge to a tendon at the posterior edge of the muscle, and these tendons, after piercing the trapezius [i.e., cucullaris muscle]... have their points of origin among the fibers of the dorsal

longitudinal [epibranchial] muscles, the last being also attached to the shoulder girdle...”

Most studies of shark muscles do not mention these tendons but instead place the origin of the dorsal superficial branchial constrictor muscles on the surface of the cucullaris muscle or on the myosepta that separate the dorsal superficial branchial constrictor muscles (Ashley & Chiasson 1988; Rosenzweig 1988; Chiasson & Radke 1993; Kent & Carr 2001; Homberger & Walker 2004: 134). Marion’s (1905) paper was descriptive and comparative (with *Raia Erinacea*) and did not include a functional analysis. It also failed to describe the attachment of the constrictor tendons beyond the epibranchial musculature to the vertebral column. The functional implications of these constrictor tendons will be discussed in Section 4.3.1 and the evolutionary implications in sections 4.4.1.1 and 4.4.1.2

4.3 Biomechanical Analysis

4.3.1 Biomechanical analysis of the dorsal superficial branchial constrictor muscles

The newly discovered fact that the dorsal superficial branchial constrictor muscles are anchored to the vertebral column has distinct functional implications. The superficial branchial constrictor muscle complex (consisting of the combined dorsal and ventral superficial branchial constrictor muscles and the trematic branchial constrictor muscles) arcs from the vertebral column over the branchial apparatus to the midventral raphe. Contraction of the vertebroseptal portion of the dorsal superficial branchial constrictor muscles pulls not only on the constrictor tendons longitudinally, but also straightens them. The straightening of the curved third constrictor tendon, which passes directly above the anterior cardinal sinus, will

press on the sinus. Thus, localized compression of the anterior cardinal sinus must occur periodically in synchrony with the expiration phase of the ventilation process, as both sinus compression and gill compression result from contractions of the superficial branchial constrictor complex. This cyclic compression of the anterior cardinal sinus may assist in returning blood from the head to the heart. Because sharks possess a low-pressure circulatory system, they may benefit from this additional pressure pump, which would work in an analogous manner as the caudal pump in the tail of fishes, which forces blood from the tail towards the kidneys and heart (Satchell 1971; Homberger & Walker 2004: 295). This coordinated function of the superficial dorsal branchial constrictor musculature adds another layer of complexity to the already highly intricate concatenation of ventilation and circulation in the head of sharks.

4.3.2 Biomechanical analysis of the trematic constrictor muscles

The trematic branchial constrictor muscles are located on the caudal edge of each gill flap (Homberger & Walker 2004: 134). They cover the fleshy gill slits and have little association with the cartilages of the branchial arches. Contraction of these muscles closes the gill slits by flattening the gill flap against the underlying tissue of the next caudal gill septum.

4.3.3 Biomechanical analysis of the ventral superficial branchial constrictor muscles

An evolutionary change resulting in new structures can occur only if the mechanical integrity and full functionality of the organism as a whole remains unimpaired and, therefore, may involve transitional expressions of a structure (Gudo & Homberger 2002). The highly irregular and variable medio-ventral attachments of the muscle fiber bundles of the ventral superficial

branchial constrictor muscles may be a manifestation of a structure in gradual transition.

Muscle fiber bundles of the ventroseptal portion of this muscle attach either on the epimysium of the coracoarcual muscle or on the midventral raphe. The tendons that attach to the raphe can travel to the raphe in one of three ways: medio-ventro-rostral to the coracoarcual muscle, over the outer surface of the coracoarcual muscle, or even through the coracoarcual muscle via pseudomyosepta (Figure 11). The strengthening of one of these attachments may on occasion be more effective than the other types and, thus, be selectively favored.

The tendons that travel through the pseudomyosepta can be loosely analogized to the condition seen in the vertebroseptal dorsal superficial branchial constrictor muscles (the constrictor tendons), as the pseudomyosepta are a “straight shot” through a muscle to the collagenous midventral raphe. Though the midventral raphe is not histologically skeletal, it can be grouped with the axial skeleton, as it serves as an anchor point for several muscle groups, such as the intermandibular and interhyoid muscles (Homberger & Walker 2004: 130-132). The coracoarcual muscles are formed embryonically from an anterior migration of somites from the postotic and anterior trunk regions (Liem *et al.* 2001). Thus these muscles are not truly segmented and therefore do not have actual myosepta, as is seen in the axial musculature, which arise from a series of segmented somites. Hence, the term “pseudomyosepta” is more appropriate.

For the reasons provided, giving one simple name to the medial-most ventral superficial constrictor muscle group using attachment sites as criteria for naming is quite difficult, as the fibers may originate from one of five sites. An almost web-like array of fibers converge at the

tip of the muscle fibers of these ventralmost ventral superficial branchial constrictor muscles. A suitable name for this group, while trying to maintain the current naming scheme, is ventroseptal fibers. This term refers to the nature of all origination sites as ventral (or medial) to the collagenous intersepta and the insertion on the rostral interseptum.

4.3.4 Biomechanical analysis of the cucullaris muscle

4.3.4.1 Insulation of the lateral line

The cucullaris muscle is located ventral to the epibranchial musculature, which is active during feeding and undulatory locomotion, and dorsal to the dorsal superficial branchial constrictor muscles, which are active during ventilation. Thus, it is constrained by two cyclically active muscle groups. During feeding, the cucullaris muscle may aid in expanding the orobranchial cavity by elevating the pharyngoepibranchial cartilages of the fourth and fifth branchial arches with the pharyngoepibranchial portion of the muscle. During ventilation, however, expansion of the orobranchial cavity is mediated by the elastic recoil of the branchial arches (Hughes & Ballintijn 1965). The lateral line within the superficial fascia external to the attachment of the cucullaris muscle might be located along a line that is often electrically and mechanical inert and, thereby, insulates the neuromast cells from electrical and mechanical stimuli that are generated by the body.

4.3.4.2 Function of the scapulocoracoid portion

The scapulocoracoid cartilage is a large cartilaginous element that would require a large force to move. In addition, the scapulocoracoid cartilage is held in place by very thick perichondrial

fibers and is bordered on all sides by masses of muscle. Furthermore, the muscle fiber bundles of the cucullaris muscle are long, thin, and lack myosepta, rendering it a relatively weak muscle that can, however, shorten considerably. Thus, it is unlikely that the scapulocoracoid bar can be displaced by muscle force. It is more likely that the contraction of the cucullaris muscle fiber bundles that attach to the scapulocoracoid cartilage would cause the muscle to bulge, straighten, and compress the wall of the anterior cardinal sinus (Homberger & Walker 2004: 134).

4.3.4.3 Function of the pharyngoepibranchial portion

The group of cucullaris muscle fiber bundles that inserts on the pharyngobranchial cartilage and pharyngoepibranchial cartilage of the fourth and fifth branchial arches, respectively, are oriented nearly vertically and can elevate the two caudal branchial arches. The mass of these two branchial arch cartilages much less than that of the scapulocoracoid cartilage, and their dorsal displacement by the pharyngoepibranchial portion of the cucullaris muscle contribute to the expansion the branchial basket.

4.3.4.4 Physiologic implications of the alterations of cucullaris muscle fasciculi near the dorsal superficial branchial constrictor muscle tendons

When the constrictor tendons are moved as a consequence of the contractions of the dorsal superficial branchial constrictor muscles, they compress the surrounding cucullaris fiber bundles, thereby restricting the blood supply to them. The replacement of the muscle fibers by tendon fibers mitigates this problem, because the collagen fibers are not only stronger than muscle fibers, but are also less vascularized and innervated and are nearly incompressible.

4.3.5 Biomechanical analysis of the epibranchial musculature

The epibranchial musculature is the continuation of the epaxial musculature rostral to the scapulocoracoid cartilage (Rosenzweig 1988; Homberger & Walker 2004: 130). According to Wilga *et al.* (2007), the epibranchial musculature raises the chondrocranium during feeding if the head needs to manipulate prey into the proper orientation for deglutition. However, given its morphological and functional continuity with the trunk epaxial muscles, which are responsible for the undulatory locomotion, its innervation by the dorsal rami of occipital and anterior spinal nerves (the anterior portion of the same nervous series that innervates the epaxial musculature) (Liem *et al.* 2001), and the results of video analysis of an undulating shark (section 3.5), the epibranchial musculature has also been determined to serve a role in locomotion.

Gudo and Homberger (2002) hypothesized that the shoulder girdle serves as a mechanical separator between the movements of the epibranchial musculature of the head and the epaxial musculature of the trunk. This does not seem to be the case, however, as undulatory movements of the axial musculature are not restricted to post-scapulocoracoid myomeres.

4.4 General functional and evolutionary implications

4.4.1 The configuration of crossing muscles in sharks

Muscles in general (as known from at least in mammals) are surrounded individually by epimysia and are organized in layers or side by side. Muscles that interdigitate, like the cucullaris muscle and the vertebroseptal portion of the dorsal superficial branchial constrictor

muscles, are rare. There are two hypotheses that could possibly explain the genesis of this configuration. First, the constrictor tendons in sharks may be the remnants of the muscles attaching on the internal surface of bony dermal plates in an ancestral taxon. Second, the pharyngoepibranchial portion of the cucullaris muscle and the dorsal superficial branchial constrictor muscles may have differentiated from a single ancestral muscle mass of a pre-gnathostome.

4.4.1.1 Hypothesis 1: Reduction of the superficial dermal bony plates

Because the skeleton of chondrichthyans is cartilaginous, well-preserved fossils of primitive chondrichthyans are rare (Lund & Grogan 1997). The earliest known chondrichthyan fossils are known almost exclusively from isolated scales, usually of the placoid type, or from teeth and toothplates from the Devonian period (Long 1995; Paxton & Eschmeyer 1998). The identity of the ancestor of sharks has long been debated, but one theory states that they may have evolved from a primitive group of jawed fishes, the placoderms (Moy-Thomas 1939; Long 1995; Paxton & Eschmeyer 1998; Goujet 2001). Janvier (1998) and Johanson (2003) have recognized that the most primitive taxa among the placoderms are sharklike and lack many features that are characteristic of derived placoderms, such as a well-developed mandibulohyoid ligament and operculum, and a clavobranchialis musculature. Primitive placoderms, like chondrichthyans, possessed a coracobranchialis system (Moy-Thomas 1939; Johanson 2003). Denison (1978) proposed that the primitive placoderm in the order Stensioellida may have had no opercular bone at all. In addition, primitive placoderms possessed separate coracomandibular and coracohyoid muscles (Johanson 2003). In a case of Red Queen Dynamics

(i.e., a case of the evolution of a trait in one species in response to an evolving trait in another species, or co-evolution), the Placodermi show a trend toward increased motility and a reduction of the dermal armor near the end of their existence at the end of the Devonian (Helfman *et al.* 1997) when slow moving, armored ostracoderm fishes, such as pteraspidomorphs and cephalaspidomorphs, were becoming extinct, probably because prey species became more agile.

Instead of scales, placoderms possessed overlapping or beveled armored exoskeletal plates, which formed a head shield and a trunk shield that were connected by a joint at the nuchal gap (Miles 1971; Denison 1978; Carroll 1988). The grooves and fossae on the internal side of these shields allow a reconstruction of the morphology of the internal soft structures (Denison 1978). The neck joint at the nuchal gap allowed the head shield to be raised (Benton 1997; Kardong 2009). The penetration of the constrictor tendons through the cucullaris and epibranchial muscles could have arisen as a consequence of the loss of the head shield and its replacement by a simple superficial fascia of connective tissue. In placoderms, the constrictor muscles of the branchial region may have originated from the cucullaris fossae on the caudo-lateral part of the internal surface of the head shield and on the outer surface of the underlying endocranium (Denison 1978). Johanson (2003) noted that the cucullaris muscle inserted on this cucullaris fossa.

Romer and Parsons (1986) and Kardong (2009) stated that muscles have a propensity to merge, become reduced, split into separate bellies or muscles, or change their point of attachment over time. In a developing embryo, muscle anlagen, or primordia, are able to migrate (Kardong

2009). Hence, over time, the gradual reduction of the head shield in placoderms could have been accompanied by a weakening of the osseotendinous connection in the cucullaris fossa, thereby driving the tendons that connected the dorsal superficial branchial constrictor muscle to the inner surface of the headshield to migrate to a more stable anchor. Therefore, the muscle fiber bundles of the dorsal superficial branchial constrictor muscles may have interdigitated with the muscle fibers of the cucullaris muscle as the origin of the dorsal superficial branchial constrictor muscle gradually moved its attachment past the cucullaris muscle from the cucullaris fossa in placoderms to the vertebral column in sharks, which do not have a head shield and, thus, no cucullaris fossa. This scenario, however, is unlikely to be corroborated by the fossil record, because primitive sharks contained no bony plates or bony elements.

Scenarios describing the migration of muscle attachments as a consequence of the loss of the bony cosmoid shields of ancestral placoderms have not yet been described in the literature, but the process of shield reduction has been studied. These bony shields are composed of three histologically distinct layers. The deepest layer consists of cellular or laminar bone, the next layer consists of spongy bone, and the superficial layer is denticulated. These bony denticles are capped by enamel or enameloid, as in modern selachians. The placoid scales of modern chondrichthyans may, hence, be remnants of the bony plate denticles that encased placoderms (Kluge *et al.* 1977; Denison 1978; Liem *et al.* 2001).

New fossil evidence may provide additional information about morphological transitions between ancestral placoderms and extant sharks. For example, newly described iniopterygian

fishes (i.e., a primitive chondrichthyan) possess an intracranial joint above the orbits, which is reminiscent of the intracranial joint present in *Latimeria*, a sarcopterygian bony fish that is considered a “living fossil” (Grogan & Lund 2009). These fishes also possessed two sets of cartilages, the neomorphic and suprascapular linking cartilages, which articulated the scapulocoracoid cartilage with the neurocranium. Because the ancestor of sharks is not known and described, it is possible that a primitive chondrichthyan with additional cranial skeletal elements, such as the new inioptrygian species, may represent a fish that is intermediate between plated taxa (such as the placoderms) and derived taxa that lack dermal bones. The discovery of cartilages that connect the pectoral girdle to the cranium supports Gudo & Homberger’s (2002) hypothesis that the scapulocoracoid cartilage represents an integral part of the head. As the cartilages suggest, paired fins probably evolved in coordination with jaws, rather than just alongside them in the evolutionary history of fishes.

4.4.1.2 Hypothesis 2: Gradual differentiation from a single muscular body; fusion of levator muscles into the cucullaris muscle of derived species

The cucullaris muscle is located dorsal to the gill arches and may represent the fused levator muscles of the branchial arches (Webster & Webster 1974; Bohensky 1981; Ashley & Chiasson 1988; Rosenzweig 1988; Wingerd 1988; Fishbeck & Sebastiani 2001). This hypothesis, however, is vague in terms of the origin and structure of the levator muscles and with respect to a model organism that may have possessed them. Levator muscles of the branchial arches are functional only if an organism possesses jaws, and Kuratani *et al.* (2002) pointed out that the lamprey possesses no cucullaris muscle or muscles derived from it. If the lamprey effectively

reflects the basic jawless condition of vertebrates, then the individual levator muscles of the branchial arches must have arisen in a basal gnathostome in conjunction with the evolution of jaws and paired pectoral fins. However, the fossil record of basal gnathostomes is exceptionally poor, and no extant model organisms of basal gnathostomes exist, so that this hypothesis remains to be tested.

Intramuscular differentiation may also account for the condition of crossing muscle fiber bundles from two different muscles as seen in the cucullaris and dorsal superficial branchial constrictor muscles of the Dogfish Shark. A developmental-evolutionary mechanism to account for such crossing muscles was proposed by Homberger & de Silva (2003) for the smooth erector and depressor feather muscles in the integument of birds. In this case it was postulated that some muscle fiber bundles of the original erector feather muscles gradually shifted one of their attachments upwards on the feather follicle to while shifting the opposite attachment down on the feather follicle. As a result, the muscle fiber bundles of the erector and depressor feather muscles cross another. The pattern of interdigitation of these two muscle types is highly irregular, which provides supportive evidence for a secondary process (Homberger & de Silva, unpublished observations). This muscle differentiation mechanism has the advantage that it retains the full muscle functionality and whole-body organism integrity throughout the differentiation process.

It is conceivable that a similar differentiation process took place for the differentiation of some of the superficial branchiomic muscles in the shark. It can be hypothesized that originally, in an ancestral placoderm, the dorsal superficial branchial constrictor muscles resembled the ones

in the extant Dogfish Shark with their attachment to the vertebral column. At the same time, the cucullaris muscle inserted only on the scapulocoracoid cartilage, like the scapulocoracoid portion of the extant Dogfish Shark, and originated from the cucullaris fossa on the internal surface of the head shield. It would, thus, pass dorsal to the constrictor tendons with no interweaving of the fiber bundles of the two muscles. Eventually, possibly with the evolution of an articulated branchial basket, some muscle fiber bundles of the dorsal superficial branchial constrictor muscle complex would move one attachment to the underlying pharyngobranchial cartilages of the branchial arches and the other attachment would move to overlying the cucullaris fossa on the internal surface of the head shield rostral to the attachment of the cucullaris muscle. As a result, the muscle fiber bundles of the new pharyngobranchio-cucullaris muscles would cross the muscle fiber bundles of the original dorsal superficial branchial constrictor muscles. In a subsequent step, in the course of the reduction of the head shield, the attachments of the cucullaris and new pharyngobranchio-cucullaris muscle fibers would have originated from the internal surface of the superficial fascia. In a final step, the insertions of the new pharyngobranchio-cucullaris muscles on the first, second and third pharyngobranchial cartilages would have shifted caudally and concentrated on the fused pharyngoepibranchial cartilages of the fourth and fifth branchial arch, thus becoming the pharyngoepibranchial portion of the cucullaris muscle as it is known in the extant Dogfish shark. In the course of this caudal shift of the insertions, the entire cucullaris muscle would assume its oblique muscle fiber direction.

4.5 Functional significance of the epibranchial musculature

The contribution of the epibranchial musculature to locomotory undulation has implications for the feeding and locomotion of sharks. Alternating unilateral contractions of the epibranchial musculature bends the branchial region, but bilateral contractions raises the chondrocranium. On the inside bend of the branchial region, the superficial branchial constrictor muscles will contract to shorten the distances between the gill arches, whereas on the outside bend, the superficial branchial constrictor muscles will be stretched.

Gudo & Homberger's (2002) hypothesis that the scapulocoracoid cartilage acts as a mechanical separator between the head, trunk, and fin musculature is no longer valid. The scapulocoracoid cartilage does effectively separate the fin muscles from the head and branchial region, but the epaxial musculature travels dorsal to and between the suprascapular processes of the scapulocoracoid cartilage and continues without interruption into the head as the epibranchial musculature. Any sharp border between the epaxial and epibranchial musculatures is topographical, though nevertheless artificial and not functional-morphological.

Cyclic ipsilateral contraction of epibranchial myomeres raises many questions regarding the synchrony of ventilation and circulation in the head. The effect of the epibranchial musculature locomotory movements on these systems and possible rhythmic concatenation of epibranchial muscle contraction with ventilation and heartbeat need to be studied *in vivo*.

4.6 The concatenation of organ systems in the selachian head

Pisciform vertebrates have evolved specializations to overcome the inevitable spatial problems that result from a rostrally tapered head, which reduces drag and turbulence during swimming. As described by Gudo & Homberger (2002), the head of fishes houses the centers of the alimentary, circulatory, nervous, and respiratory systems. Terrestrial vertebrates overcame the spatial constraints through a high degree of cephalization and the evolution of a neck, and by relocating the circulatory and respiratory centers in the thorax (Homberger, unpublished observations). In fishlike organisms, however, there is a constant environmental selective regime favoring a fusiform, streamlined body with a tapered head. The mechanical regulation of branchial blood flow during ventilation is the primary mechanism synchronizing the complex processes that involve multiple organ systems in the head of fishes, as Metcalfe & Butler (1984) suggested that the branchiomic vasculature of dogfishes is devoid of any direct motor innervation.

4.7 Concluding remarks

The complexity of the concatenation of the ventilation and circulation systems has been studied by several authors (Lutz & Wyman 1932; Satchell 1962; Hughes & Ballintijn 1965; Metcalfe & Butler 1984; Taylor 1985), but our study suggests that the coordination of these systems is even more multifaceted and intricate than previously thought because locomotion also influences these systems. The epibranchial musculature, which is often omitted from studies of the head musculature, should be included not only in future studies of feeding, but also in those of locomotion.

Further re-evaluating morphological studies should include detailed descriptions of the fibrous arrangement of the remaining branchial muscles and of the muscles associated with the first two visceral arches, as only the superficial muscles of the third through seventh visceral arches have been described here. Also, because the epibranchial musculature contracts rhythmically during undulation, its effect on ventilation and circulation should be addressed. Because ventilation and circulation are both also cyclic processes (heartbeat and ventilation occur in a 1:1 ratio in an undisturbed shark according to Taylor 1985), then perhaps locomotion in a cruising shark is also timed so that the ventilatory muscle action is not inhibited.

5. Acknowledgements

I thank Dr. Dominique G. Homberger for directing and funding the project and for her support, encouragement, and mentoring. I would also like to thank Anabela Maia and Dr. Cheryl Wilga from the University of Rhode Island for providing the video of an undulating shark. Brooke Hopkins, Michelle Osborn, and Brad Wood helped in taking photographs of the specimens and were available for discussion, advice and mentoring. Sigrid Hamilton assisted in scanning the photographs. I would also like to thank Dr. D. Allen Rutherford and Dr. Kurt R. Svoboda for serving on the committee for this Honors Thesis.

6. Literature cited

- Ashley, L.M. & R.B. Chiasson. 1988. Laboratory Anatomy of the Shark, 5th ed. Wm. C. Brown Company Publishers, Dubuque, Iowa.
- Benton, M.J. 1997. Vertebrate Paleontology, 2nd ed. Cambridge University Press, Cambridge, UK. P. 54
- Bohensky, F. 1981. Photo Manual and Dissection Guide of the Shark. Avery Publishing Group Inc., Wayne, New Jersey.
- Bond, C.E. 1996. Biology of Fishes, 2nd ed. Brooks/Cole Thompson Learning, Inc., Florence, Kentucky
- Carroll, R. L. 1988. Vertebrate Paleontology and Evolution. W.H. Freeman and Company, New York, New York. Pp. 46-51, 58-60.
- Chiasson, R.B. & W.J. Radke. 1993. Laboratory Anatomy of the Vertebrates. Wm. C. Brown Publishers, Dubuque, Iowa.
- Denison, R. 1978. Handbook of Paleoichthyology, vol. 2. Gustav Fischer Verlag, Stuttgart, Germany. Pp. 1-15.
- Ferry-Graham, L.A. 1999. Mechanics of ventilation in swellsharks, *Cephaloscyllium ventriosum* (Scyliorhinidae). Journal of Experimental Biology, 202: 1501-1510.
- Fishbeck, D.W. & A. Sebastiani. 2001. Comparative Anatomy: Manual of Vertebrate Dissection. Morton Publishing Company, Englewood, Colorado.
- Gilbert, S.G. 1973. Pictorial Anatomy of the Dogfish. University of Washington Press, Seattle, Washington.
- Grogan, E.D., & R. Lund. (2009). Two new iniopterygians (Chondrichthyes) from the Mississippian (Serpukhovian) Bear Gulch Limestone of Montana with evidence of a new form of neurocranium. Acta Zoologica 90: 134-151
- Goujet, D. 2001. Placoderms and basal gnathostome apomorphies. Pp. 209-222 in Major Events in Early Vertebrate Evolution (P. E. Ahlberg, ed.). Taylor & Francis Routledge, London, New York.
- Gudo, M., & D.G. Homberger. 2002. The functional morphology of the pectoral fin girdle of the Spiny Dogfish (*Squalus acanthias*): Implications for the evolutionary history of the pectoral girdle of vertebrates. Senckenbergiana lethaea, 82 (1): 241-252.
- Helfman, G.S., B.B. Collette, & D.E. Facey. 1997. The Diversity of Fishes. Blackwell Science, Inc., Malden, Massachusetts. P. 157
- Homberger, D.G. & K.N. de Silva. 2003. The role of mechanical forces on the patterning of the avian feather-bearing skin: a biomechanical analysis of the integumentary musculature in birds. Journal of Experimental Zoology 298B: 123-139.
- Homberger, D.G., & W.F. Walker. 2004. Vertebrate Dissection, 9th ed. Brooks/Cole Thompson Learning, Inc., Belmont, California. Pp. 44-47, 133-134, 264-265.

- Hughes, G.M. 1960. The mechanism of gill ventilation in the dogfish and skate. *Journal of Experimental Biology* 37: 11-27.
- Hughes, G.M., & C.M. Ballintijn. 1965. The muscular basis of the respiratory pumps in the dogfish (*Scyliorhinus canicula*). *Journal of Experimental Biology* 43: 363-383.
- Janvier, P. 1998. *Early Vertebrates*. Oxford University Press Inc., New York, New York. Pp. 150-155
- Johanson, Z. 2003. Placoderm branchial and hypobranchial muscles and origins in jawed vertebrates. *Journal of Vertebrate Paleontology* 23 (4): 735-749.
- Kardong, K. 2009. *Vertebrates, Comparative Anatomy, Function, Evolution*, 3rd ed. McGraw-Hill Companies, Inc., New York, New York. Pp. 93, 384, 397-398
- Kardong, K., & E. Zalisko. 2006. *Comparative Vertebrate Anatomy: A Laboratory Dissection Guide*, 4th ed. McGraw-Hill Companies, Inc., New York, New York.
- Kent, G.C., & R.K. Carr. 2001. *Comparative Anatomy of the Vertebrates*, 9th ed. McGraw-Hill Companies, Inc., New York, New York. P. 259
- Kluge, A., B. Frye, K. Johansen, K. Liem, C. Noback, I. Olsen, & A. Waterman. 1977. *Chordate Structure and Function*, 2nd ed. Macmillan Publishing Co., Inc. New York, New York.
- Kuratani, S., S. Kuraku and, & Y. Murakami. 2002. Lamprey as an evo-devo model: lessons from comparative embryology and molecular phylogenetics. *Genesis* 34: 175-183
- Liem, K.F., W.E. Bemis, W.F. Walker, Jr., & L. Grande. 2001. *Functional Anatomy of the Vertebrates, an Evolutionary Perspective*, 3rd ed. Harcourt College Publishers, Orlando, Florida. Pp. 330, 580-584
- Long, J. A. 1995. *The Rise of Fishes*. The Johns Hopkins University Press, Baltimore, Maryland. Pp. 66-73, 100-122.
- Lund, R., & E.D. Grogan. 1997. Relationships of Chimaeriformes and the basal radiation of Chondrichthyes. *Reviews in Fish Biology and Fisheries* 7: 65-123
- Lutz, B.R., & L.C. Wyman. 1932. Reflex cardiac inhibition of branchiovascular origin in the elasmobranch *Squalus acanthias*. *Biology Bulletin Marine biology Laboratory, Woods Hole* 62: 10-16.
- Mallatt, J. 1997 (a). Shark pharyngeal muscles and early vertebrate evolution. *Acta Zoologica* 78: 279-294
- Mallatt, J. 1997 (b). Crossing a major morphological boundary: the origin of jaws in vertebrates. *Zoology* 100: 128-140
- Marinelli, W. & A. Strenger. 1959. *Vergleichende Anatomie und Morphologie der Wirbeltiere. III. Lieferung (Squalus acanthias)*. Vienna: Franz Deuticke.
- Marion, G.E. 1905. Mandibular and pharyngeal muscles of *Acanthias* and *Raia*. – *The American Naturalist* 39: 891-924.
- Metcalfe, J.D., & P.J. Butler. 1984. On the nervous regulation of gill blood flow in the dogfish (*Scyliorhinus canicula*). *Journal of Experimental Biology* 113: 253-267.

- Metcalfe, J.D., & P.J. Butler 1986. The functional anatomy of the gills of the dogfish (*Scyliorhinus canicula*). *Journal of Zoology*, London 208: 519-530.
- Miles, R. S. 1971. The Holonematidae (placoderm fishes), a review based on new specimens of *Holonema* from the upper Devonian of Western Australia. *Philosophical Transactions of the Royal Society of London, Series B*, 263 (849): 101-234.
- Moy-Thomas, J.A. 1939. *Palaeozoic Fishes*. Chemical Publishing Co. Inc., New York, New York. 1939. Pp. 27-29, 57-61
- Paxton, J.R., & W.N. Eschmeyer. 1998. *Encyclopedia of Fishes*. Academic Press, San Diego, CA. Pp. 27-29
- Romer, A., & T. Parsons. 1986. *The Vertebrate Body*, 6th ed. W.B. Saunders Company, Philadelphia, Pennsylvania. Pp. 278, 307
- Rosenzweig, L.J. 1988. *Anatomy of the Shark: Text and Dissection Guide*. Wm. C. Brown Publishers, Dubuque, Iowa.
- Satchell, G.H. 1962. Intrinsic vasomotion in the dogfish gill. *Journal of Experimental Biology* 39: 503-512.
- Satchell, G.H. 1971. *Circulation in Fishes*. Cambridge University Press, Cambridge, Massachusetts.
- Summers, A.P., & L. A. Ferry-Graham. 2003. Respiration in elasmobranchs: new models of aquatic ventilation. Pp. 87–100 *in* *Vertebrate Biomechanics and Evolution* (Bels, V.L., J-P. Gasc, and A. Casinos). BIOS Scientific Publishers Ltd., Oxford, England.
- Taylor, E.W. 1985. Control and co-ordination of gill ventilation and perfusion. *Symposia of the Society for Experimental Biology*, 39: 123-161.
- Webster, D. & M. Webster. 1974. *Comparative Vertebrate Morphology*. Academic Press, Inc., New York, New York. P. 142
- Wilga, C.D., & P.J. Motta 1998. Conservation and variation in the feeding mechanism of the spiny dogfish *Squalus acanthias*. *Journal of Experimental Biology* 201(9): 1345-1358
- Wilga, C.D., P.J. Motta, & C.P. Sanford 2007. Evolution and ecology of feeding in elasmobranchs. *Integrative and Comparative Biology* 47(1): 55-69.
- Wingerd, B.D. 1988. *Dogfish Dissection Manual*. The Johns Hopkins University Press, Baltimore, Maryland.

7. Appendices

7.1 Appendix I: Tables

Table 1a-e. Compilation of earlier morphological descriptions of the superficial branchiomeric musculature of the Dogfish Shark. Only inferences with clear descriptions of the muscles are listed.

Table 1a. Dorsal superficial branchial constrictor muscles

Author(s) year	Synonymies	Origin	Insertion	Function
Andermann (this study)	Dorsal superficial branchial constrictor muscles	1 st , 2 nd , 3 rd muscles-vertebroseptal portion : vertebral column; Interseptal portion: myoseptum; 4 th muscle: vertebroseptal and interseptal portions: scapulocoracoid cartilage	Rostral myoseptum	Compress the pharyngeal chamber; partially compress the anterior cardinal sinus
Marion 1905	Constrictores superiores dorsales 3-6	Aponeurosis and extrabranchial cartilage; dorsal fibers converge at a tendon and originate among the fibers of the dorsal longitudinal bundle; last is attached to shoulder girdle	Aponeurosis in front; continuous with fibers of ventral constrictors	Compress the pharyngeal chamber, eject the water, and close the gill slits
Gilbert 1973	Constrictors 3-6	Connective tissue raphe that separate the constrictors	Connective tissue raphe [sic!] that separate the constrictors	Compress the pharyngeal chamber, eject the water, and close the gill slits
Romer & Parsons 1986	Superficial constrictor	Sheet of fascia on the back	N/A	N/A
Ashley & Chiasson 1988	Dorsal superficial constrictors	Fascia of dorsal myomeric muscle	Superficial tissue of gill septum	Constricts parabranial cavity
Rosenzweig 1988	Dorsal branchial constrictors	Fascia that covers the epibranchial musculature and the tendinous intersections between which it is interposed	Interbranchial septum	Compress the branchial pouches
Chiasson & Radke 1993	Dorsal branchial constrictors	Facia [sic!] of dorsal myomeric muscle	Superficial tissue of septum	Constrict parabranial cavity
Wingerd 1998	Dorsal constrictors	Connective tissue raphe	Extends towards the gill slits	Compress the pharyngeal chamber, close the gill slits
Fishbeck & Sebastiani 2001	Dorsal constrictors	Raphe	Raphe	Compression of the gill pouches
Kent & Carr 2001	Constrictors	Strong fascia above the gill pouch	N/A	Compress the gill pouches, expelling respiratory water
Homberger & Walker 2004	Dorsal superficial branchial constrictor muscles	Surface of cucullaris muscle	N/A	N/A
Kardong & Zalisko 2006	Dorsal constrictors 3-6	Vertical raphe	Vertical raphe	Compress gill pouches

Table 1b. Trematic branchial constrictor muscles

Andermann (this study)	Trematic branchial constrictor muscles	Posterior myoseptum	Posterior myoseptum	Close gill slit
Marion 1905	N/A	N/A	N/A	N/A
Gilbert 1973	constrictors 3-6	N/A	N/A	N/A
Romer & Parsons 1986	N/A	N/A	N/A	N/A
Ashley & Chiasson 1988	Septal constrictors	N/A	N/A	N/A
Chiasson & Radke 1993	Septal constrictors	Dorsal constrictors	Ventral constrictors	Compress septum
Wingerd 1998	N/A	N/A	N/A	N/A
Fishbeck & Sebastiani 2001	N/A	N/A	N/A	N/A
Kent & Carr 2001	N/A	N/A	N/A	N/A
Homberger & Walker 2004	Branchial trematic constrictor muscles	N/A	N/A	N/A
Kardong & Zalisko 2006	N/A	N/A	N/A	N/A

Table 1c. Ventral superficial branchial constrictor muscles

Andermann (this study)	Ventral superficial branchial constrictor muscles	Ventroseptal portion: midventral raphe, epimysium of coracoarcual muscle; Interseptal portion: posterior myoseptum	Rostral myoseptum	Compress the pharyngeal chamber
Marion 1905	Constrictores superiores ventrales	Aponeurosis between each and the next succeeding muscle; fascia between the gill cavities and the common coracoid muscles; median line ventral to common coracoids; last is attached to pectoral girdle and fascia covering ventral fin muscles	Aponeurosis in front [rostral] or continued dorsally and laterally over into the dorsal constrictors	Compress the pharyngeal chamber, eject the water, and close the gill slits
Gilbert 1973	Constrictors 3-6	Connective tissue raphes that separate the constrictors	Connective tissue raphes [sic!] that separate the constrictors	Compress the pharyngeal chamber, eject the water, and close the gill slits
Romer & Parsons 1986	Superficial constrictor	Sheet of fascia on the throat	N/A	N/A
Ashley & Chiasson 1988	Ventral superficial constrictors	Ventral myomeric muscle fascia	Superficial tissue of gill septum	Constricts parabranial cavity
Rosenzweig 1988	Ventral branchial constrictors	Fascia that covers the ventral musculature of the head and the tendinous intersections between which it is interposed	Interbranchial septum	Compress the branchial pouches
Chiasson & Radke 1993	Ventral branchial constrictors	Ventral myomeric muscle fascia [sic!]	Superficial tissue of gill septum	Constrict parabranial cavity
Wingerd 1998	Ventral constrictors	Connective tissue raphe	Extends towards the gill slits	Compress the pharyngeal chamber, close the gill slits
Fishbeck & Sebastiani 2001	Ventral constrictors	Raphe	Raphe	Compression of the gill pouches
Kent & Carr 2001	Constrictors	Strong fascia below the gill pouch	N/A	Compress the gill pouches, expelling respiratory water
Homberger & Walker 2004	Ventral superficial branchial constrictor muscles	Surface of the coracoarcual muscle	N/A	N/A
Kardong & Zalisko 2006	Ventral constrictors 3-6	Vertical raphe	Vertical raphe	Compress gill pouches

Table 1d. Cucullaris muscle

Andermann (this study)	Cucullaris muscle	Tendon sheet continuous with superficial fascia	Scapulocoracoid portion: scapulocoracoid cartilage; Pharyngoepibranchial portion: pharyngoepibranchial cartilage	Compress anterior cardinal sinus, raise the dorsal elements of the caudal two branchial arches
Marion 1905	Trapezius muscle	Fascia covering sides of the dorsal longitudinal muscles	Anterior edge of pectoral girdle, dorsal edge of last branchial arch	Raise the pectoral girdle and draw it forward
Gilbert 1973	Cucullaris muscle	Fascia of the dorsal longitudinal bundle	Epibranchial cartilage of the last gill arch and the scapular process of the pectoral girdle	Elevates the gill arches and pectoral girdle
Webster & Webster 1974	Cucullaris muscle	N/A	N/A	Raises the gill arches
Romer & Parsons 1986	Cucullaris muscle	Dorsal fascia	Shoulder girdle	N/A
Ashley & Chiasson 1988	Cucullaris muscle	Fascia of cranial epaxial muscles and dorsal occipital neurocranium	Scapular part of scapulocoracoid	Draws pectoral girdle and limb craniodorsally
Rosenzweig 1988	Cucullaris muscle	Connective tissue that covers the epibranchial musculature	Scapular part of the scapulocoracoid cartilage and dorsal skeletal elements of last branchial arch	Draws scapulocoracoid cartilage and pectoral fin rostrally. Elevates the gill apparatus
Chiasson & Radke 1993	Cucullaris muscle	Fascia of cranial epaxial muscles and dorsal occipital neurocranium	Scapular part of scapulocoracoid	Draw pectoral girdle and limb craniodorsally
Wingerd 1998	Cucullaris muscle	Fascia of the dorsal longitudinal bundles	Scapular process of the pectoral girdle, epibranchial cartilage of the last visceral arch	Elevates the last visceral arch and elevation of the pectoral girdle
Fishbeck & Sebastiani 2001	Cucullaris muscle	Fascia of the epaxial muscles	Scapular process of the scapulocoracoid bar	Elevates the scapular process
Kent & Carr 2001	Cucullaris muscle	N/A	N/A	Raises the pharyngeal wall assisted by the levator hyomandibulae of the second arch
Homberger & Walker 2004	Cucullaris muscle	Surface of the epibranchial musculature	Scapulocoracoid cartilage and the epibranchial cartilage of the fifth branchial arch	Compresses the anterior cardinal sinus when the mouth is being opened and the branchial arches are unfolded
Kardong & Zalisko 2006	Cucullaris muscle	Dorsal longitudinal bundle fascia	Epibranchial cartilage and scapular process	Elevates the gill arches and pectoral girdle
Kardong 2009	Cucullaris muscle	Dorsal body surface	Last branchial arch and scapula	N/A

Table 1e. Epibranchial musculature

Andermann (this study)	Epibranchial musculature	Myoseptum; occipital region of neurocranium	Myoseptum	Bilateral contraction: raise the chondrocranium; Unilateral contraction: undulation
Marion 1905	N/A	N/A	N/A	N/A
Gilbert 1973	N/A	N/A	N/A	N/A
Romer & Parsons 1986	N/A	N/A	N/A	N/A
Ashley & Chiasson 1988	Did not distinguish from epaxial musculature	Caudal surface of chondrocranium	N/A	N/A
Rosenzweig 1988	Epibranchial muscles	N/A	N/A	N/A
Chiasson & Radke 1993	N/A	N/A	N/A	N/A
Wingerd 1998	N/A	N/A	N/A	N/A
Fishbeck & Sebastiani 2001	N/A	N/A	N/A	N/A
Kent and Carr 2001	N/A	N/A	N/A	N/A
Homberger & Walker 2004	Epibranchial musculature	Caudal surface of the chondrocranium and vertebral column	N/A	N/A
Kardong & Zalisko 2006	N/A	N/A	N/A	N/A
Wilga et.al. 2007	Epibranchial musculature	N/A	N/A	raises the chondrocranium during feeding if prey orientation requires manipulation of the head

7.2 Appendix II: Figures

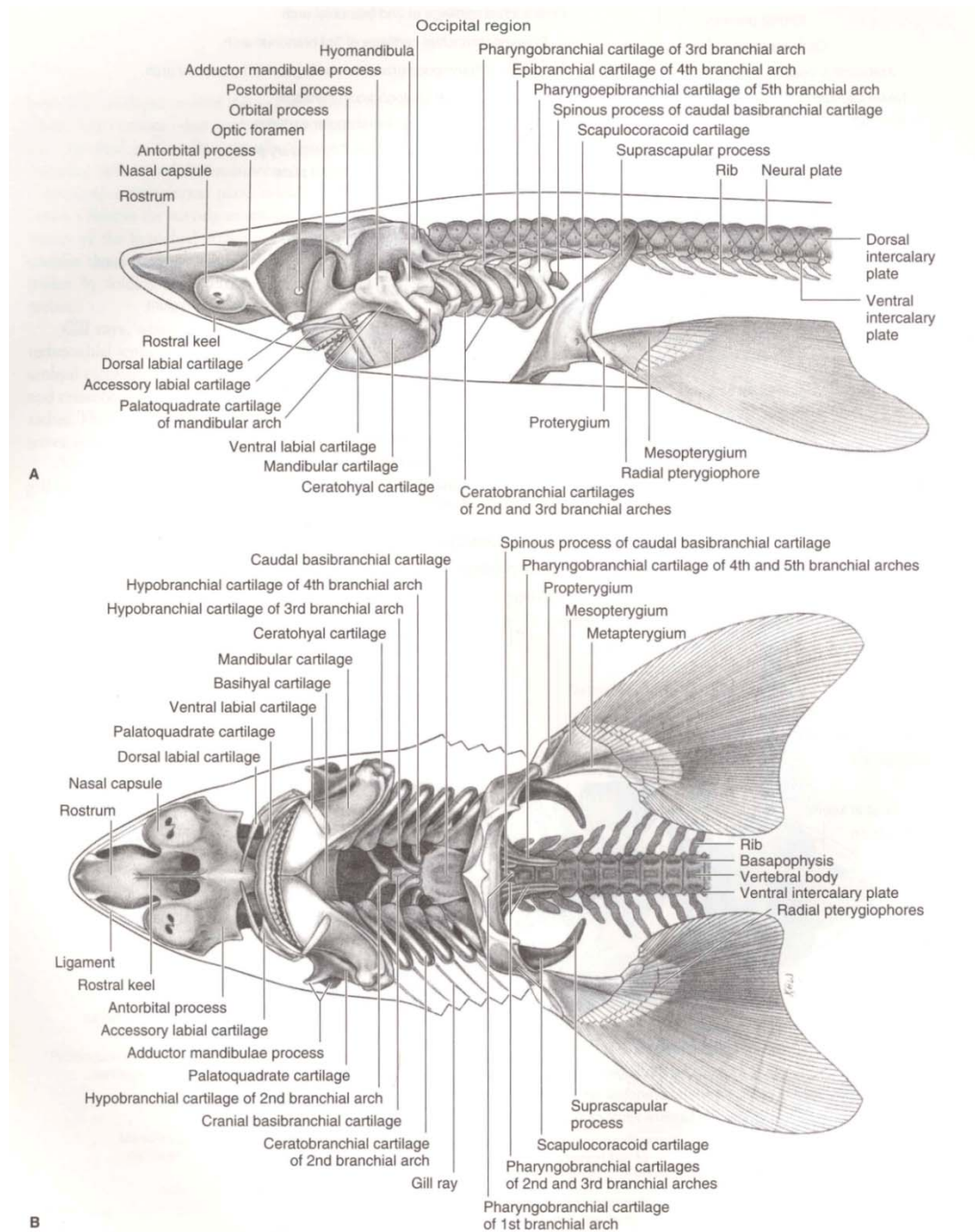


Figure 1. Cranial and visceral skeleton of *Squalus acanthias* with closed jaws and compressed gill arches. (A) sinistral view; (B) ventral view. (Adapted from Homberger & Walker, 2004: Figure 4-4, page 45), with permission.

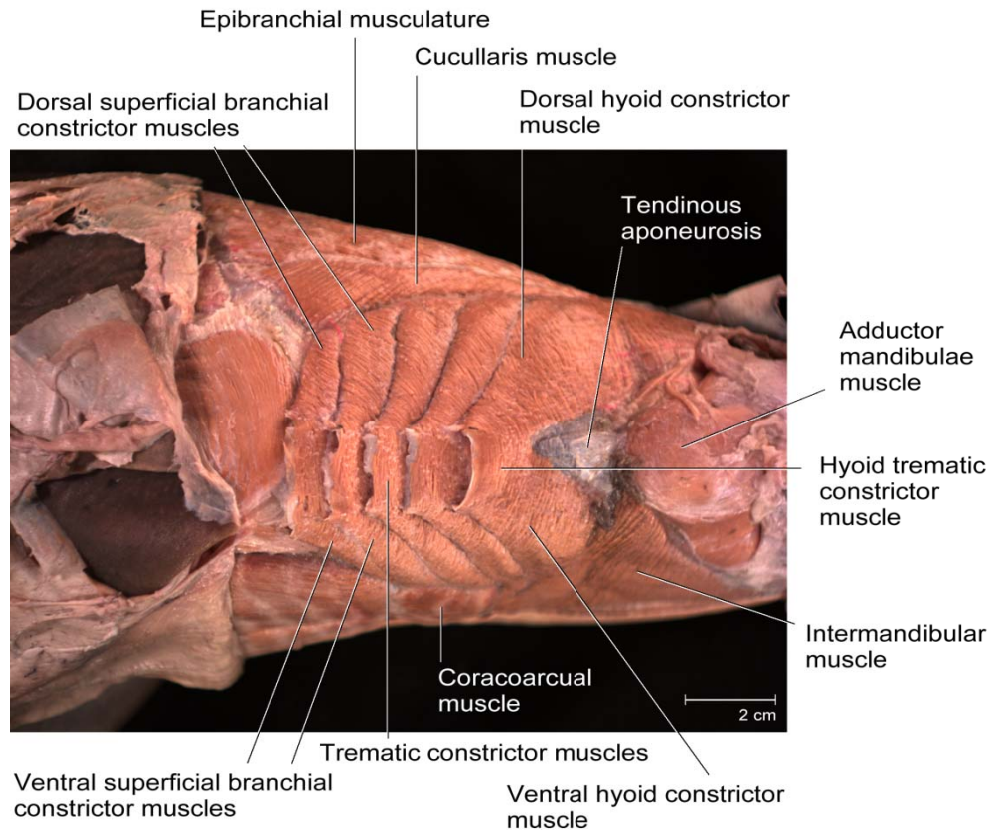


Figure 2. Dextral view of the branchiomeric muscles of the dogfish shark (*Squalus acanthias*) (RJA003) to show the contiguousness of the muscle fiber bundles of the trematic constrictor muscles with those of the dorsal and ventral superficial branchial constrictor muscles.

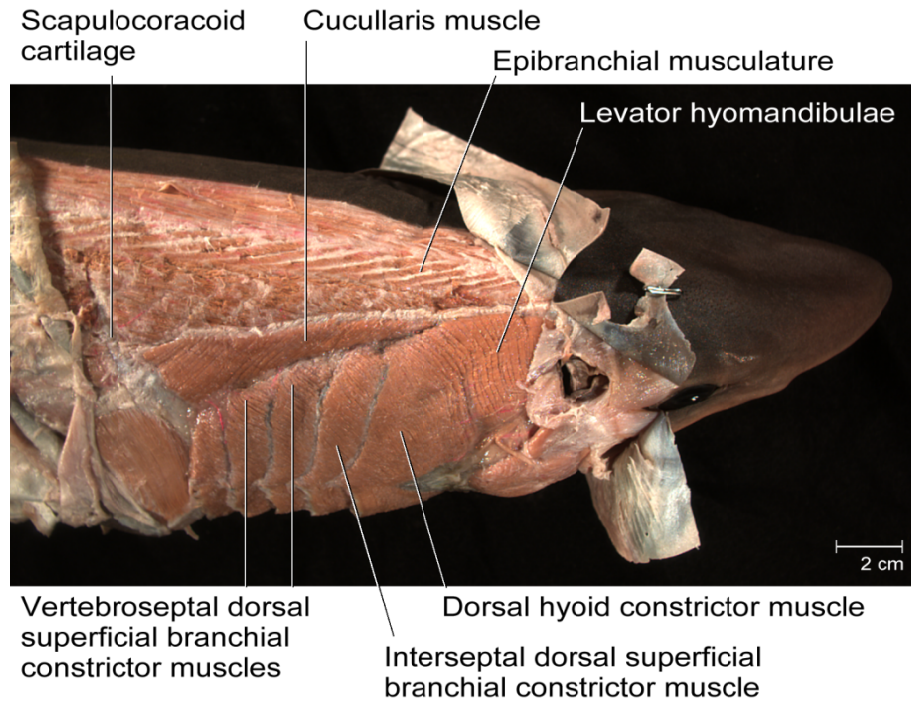


Figure 3. Dorsodextral view of the branchiomeric region of the dogfish shark (*Squalus acanthias*) (RJA003) with the skin and superficial fascia reflected, showing the topographical relationship of the dorsal superficial branchial constrictor muscles to the cucullaris and epibranchial muscles.

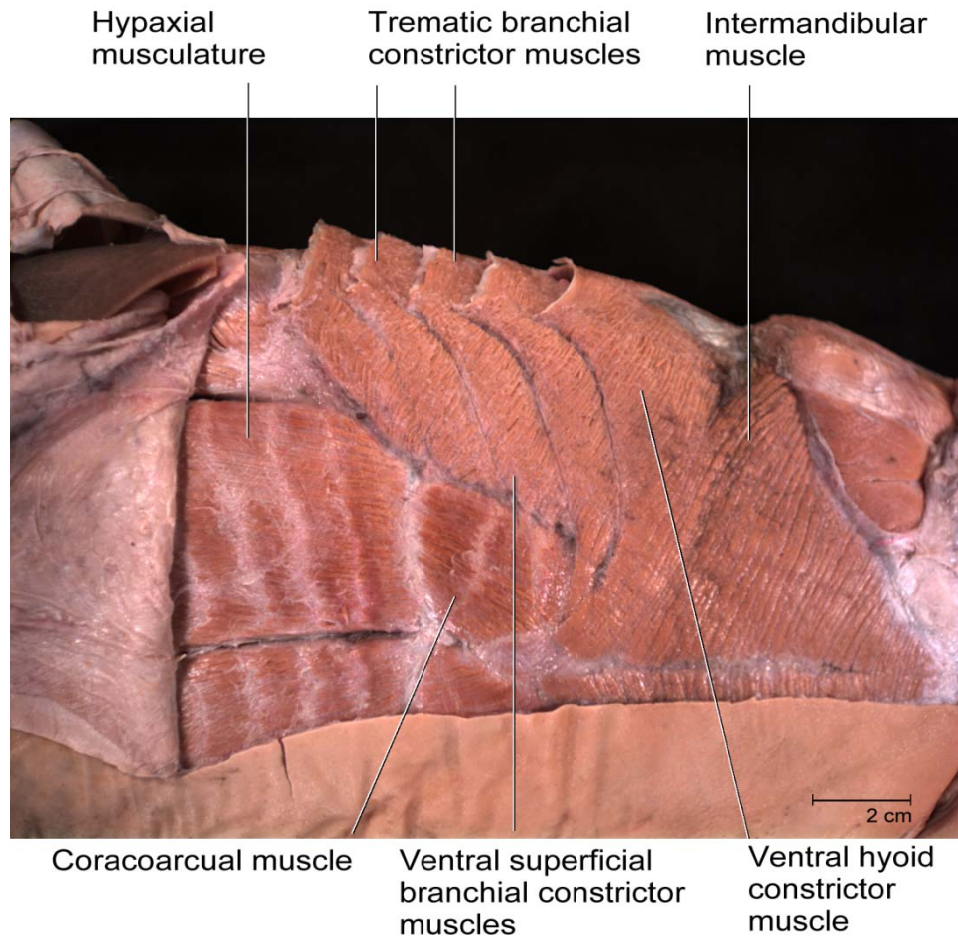


Figure 4. Ventral overview of a skinned dogfish shark (*Squalus acanthias*) (RJA003). Rostral is to the right.

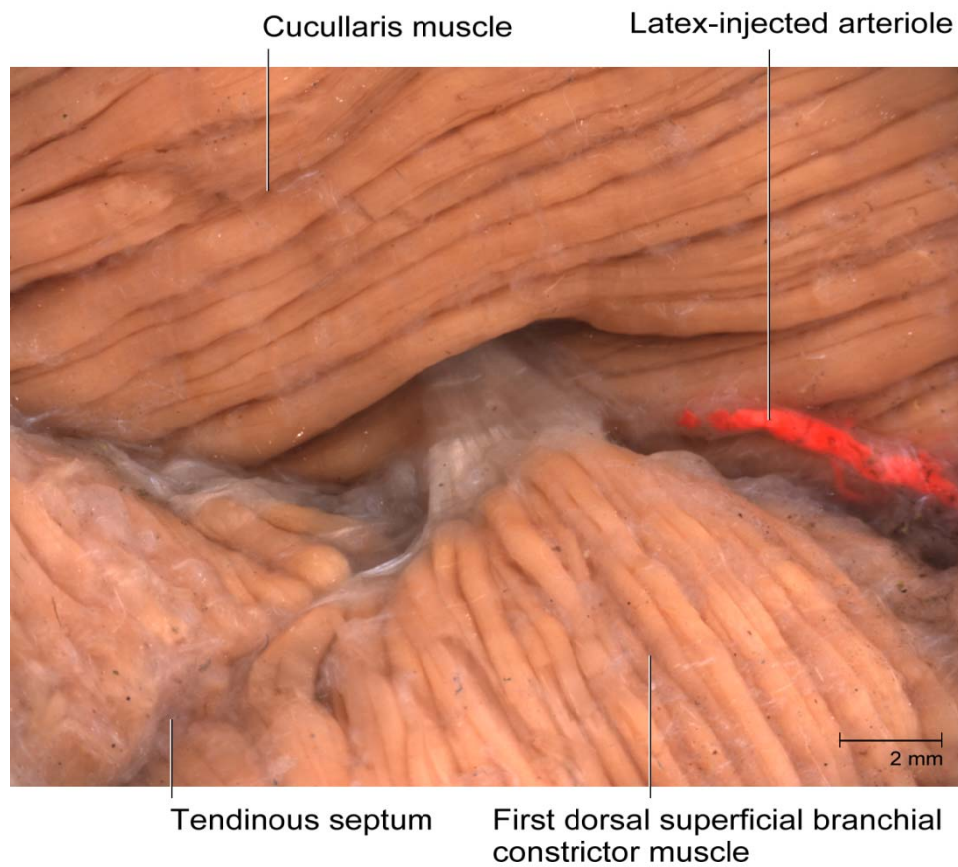


Figure 5. Magnified view of the vertebroseptal portion of the first dorsal superficial branchial constrictor muscle and the cucullaris muscle on the right side of the dogfish shark (*Squalus acanthias*) (RJA003), to show the vertebroseptal tendon passing between muscle fiber bundles of the cucullaris muscle on its way towards its attachment on the vertebral column. Some remaining portion of the perimysium of the cucullaris muscles was reflected and the muscle fiber bundles of the cucullaris muscles were slightly spread apart to reveal the tendon.

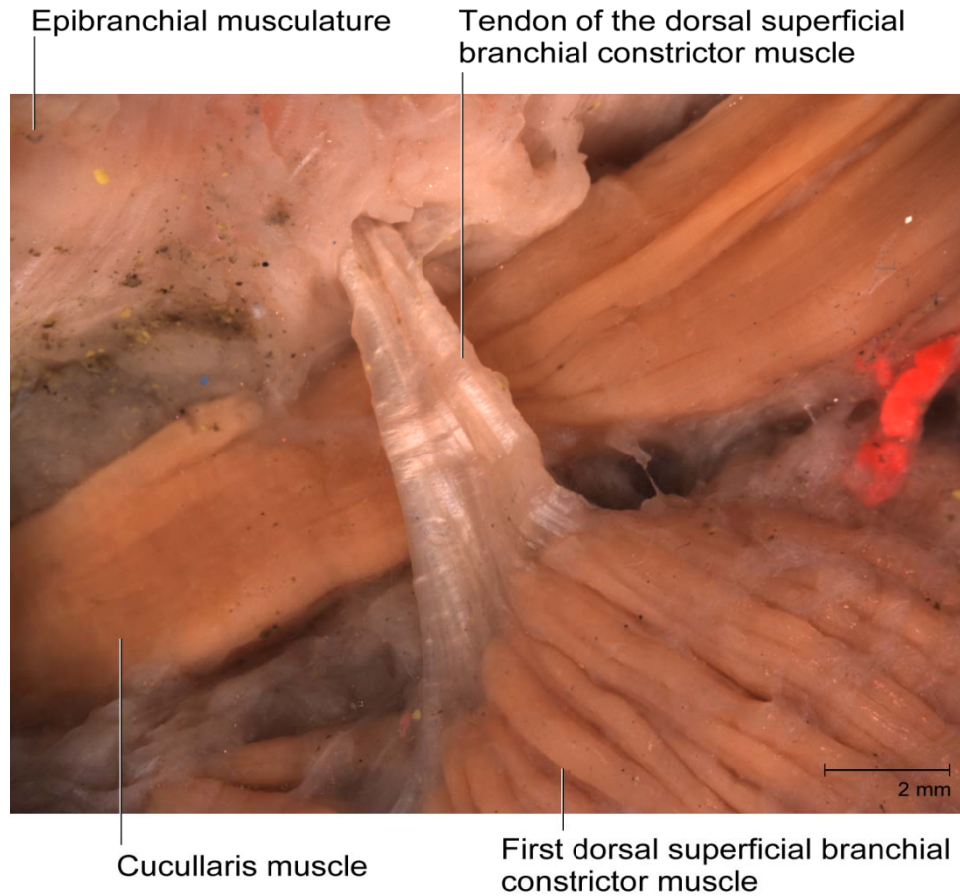


Figure 6. Magnified view of the vertebroseptal portion of the first dorsal superficial branchial constrictor muscle of the dogfish shark (*Squalus acanthias*) (RJA003) with the cucullaris muscle reflected. The tendinous portion of the muscle fibers of the cucullaris that cross the collagenous fibers of the dorsal constrictor tendon were also reflected, revealing an unhindered view of the dorsal constrictor tendon.

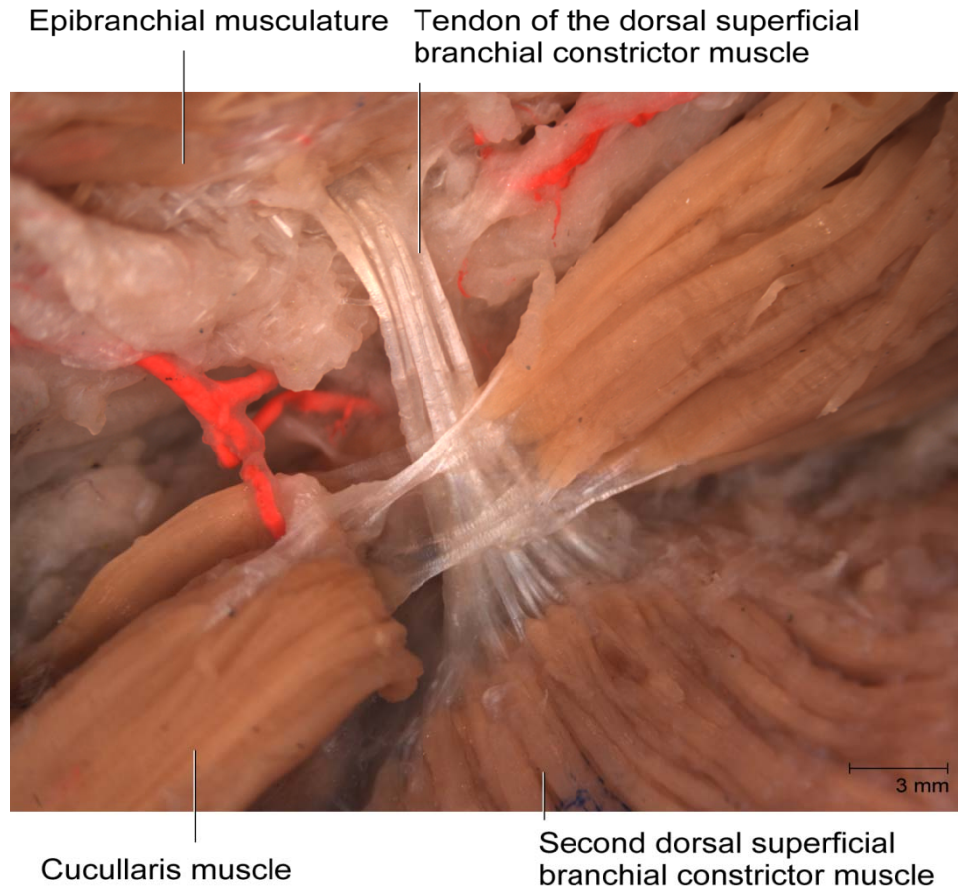


Figure 7. Magnified view of the vertebroseptal portion of the second dorsal superficial branchial constrictor muscle of the dogfish shark (*Squalus acanthias*) (RJA003) with the cucullaris partially reflected. The muscle fibers of the cucullaris muscle become tendinous within close proximity to the dorsal constrictor fiber, and they become muscular again with distance from the constrictor fiber.

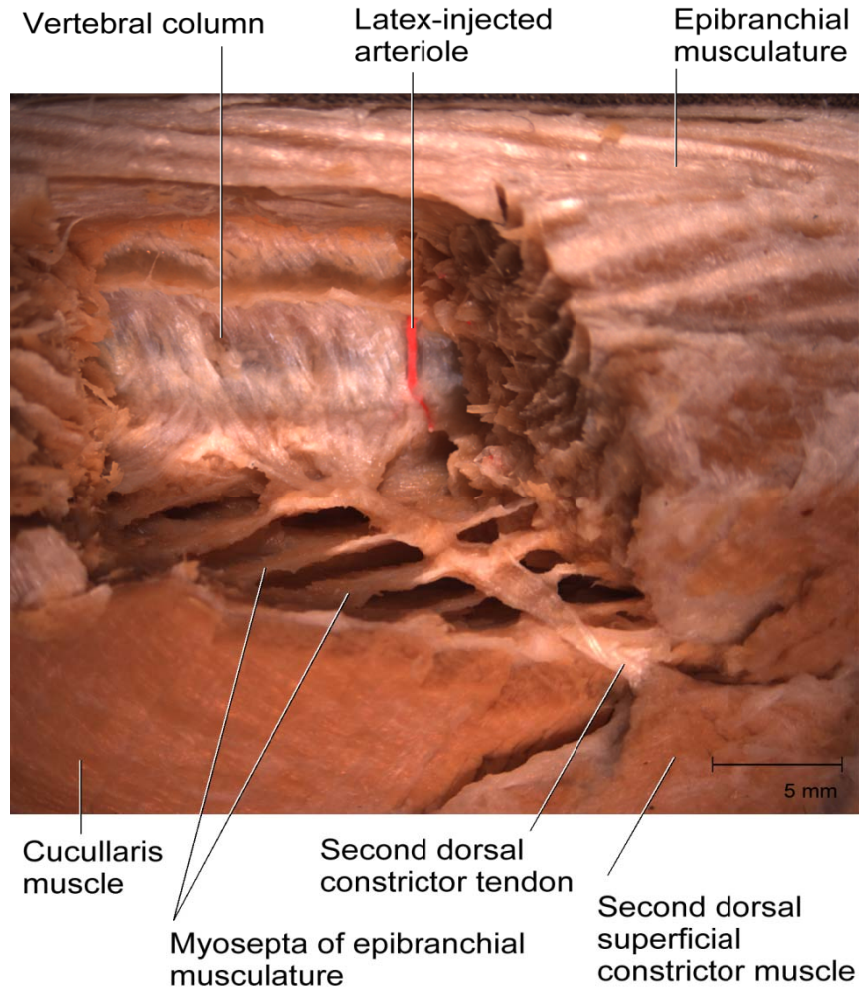


Figure 8. Overview of the vertebroseptal portion of the second dorsal superficial branchial constrictor muscle of the dogfish shark (*Squalus acanthias*) (RJA002). Portions of the cucullaris muscle and epibranchial musculature that overlaid the dorsal constrictor tendon were removed.

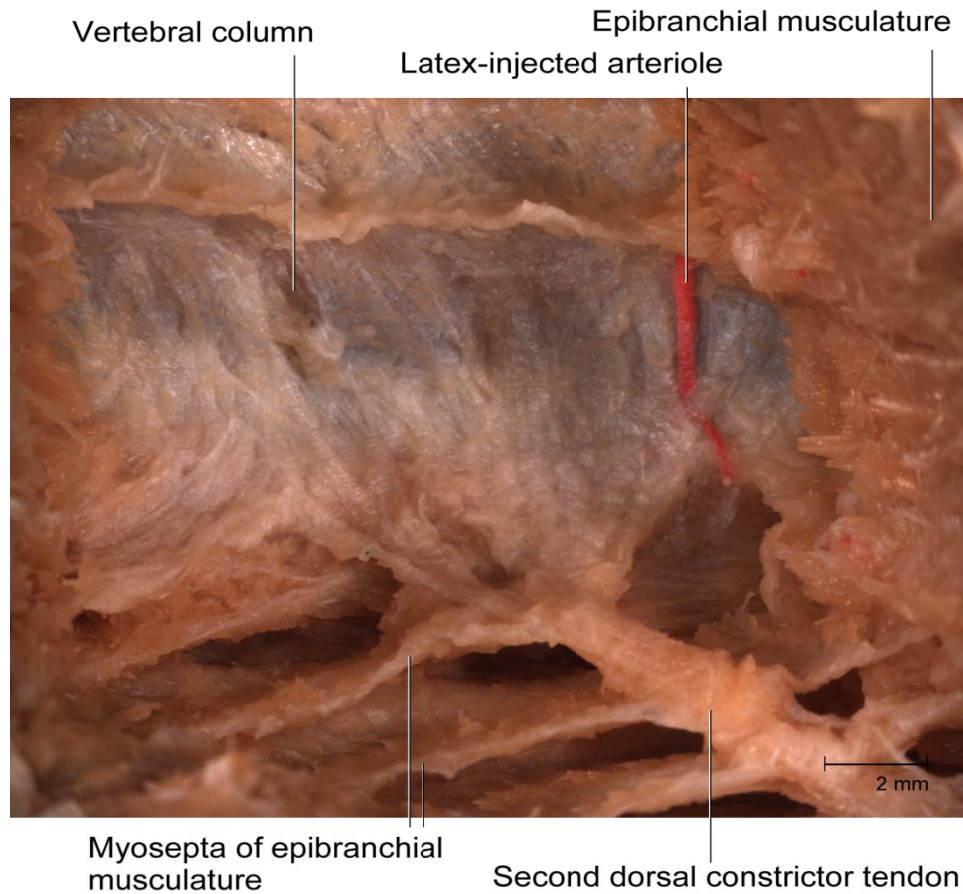


Figure 9. Magnified view of the second dorsal constrictor tendon of the dogfish shark (*Squalus acanthias*) (RJA002). The tendon fans out in its attachment to the vertebral column.

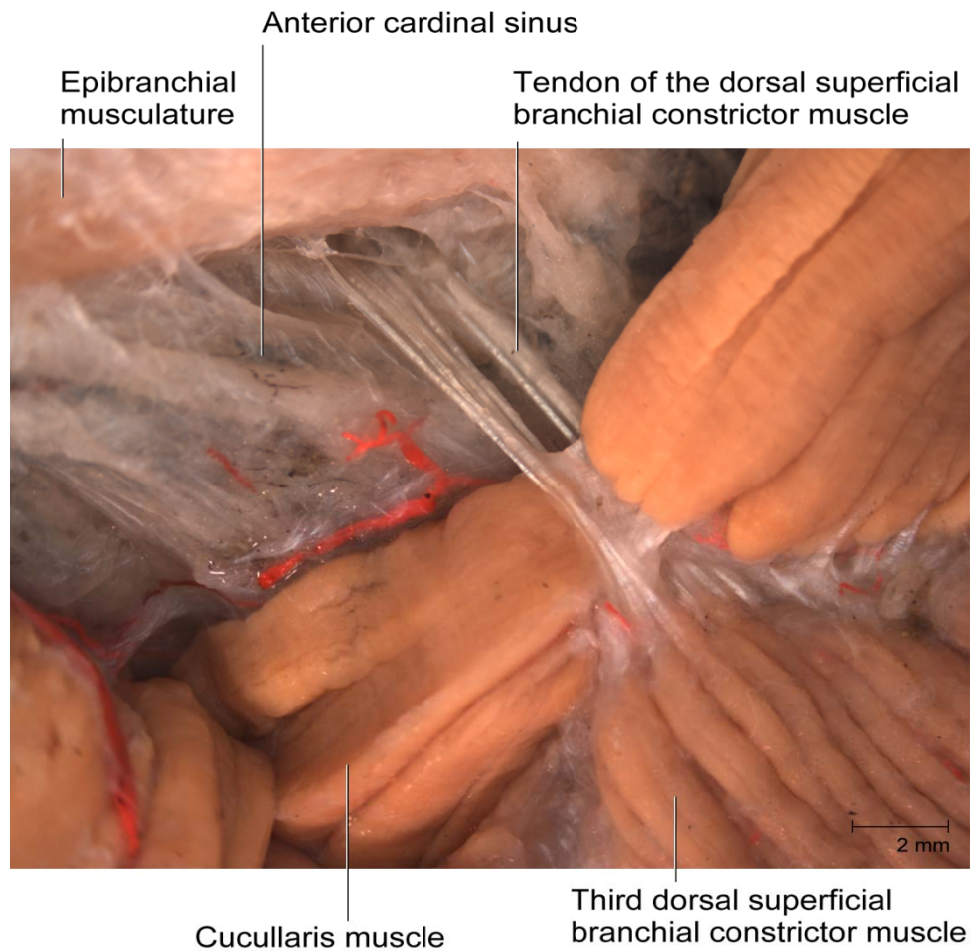


Figure 10. Magnified view of the vertebroseptal portion of the third dorsal superficial constrictor muscle of the dogfish shark (*Squalus acanthias*) (RJA003) with the cucullaris muscle partially reflected. The dorsal constrictor tendon is separated into two portions by the tendinous fibers of the cucullaris muscle, but they rejoin as they enter the fascia sheet overlying the epibranchial musculature. The third dorsal constrictor fiber directly overlies the fluid-filled (*in vivo*) anterior cardinal sinus.

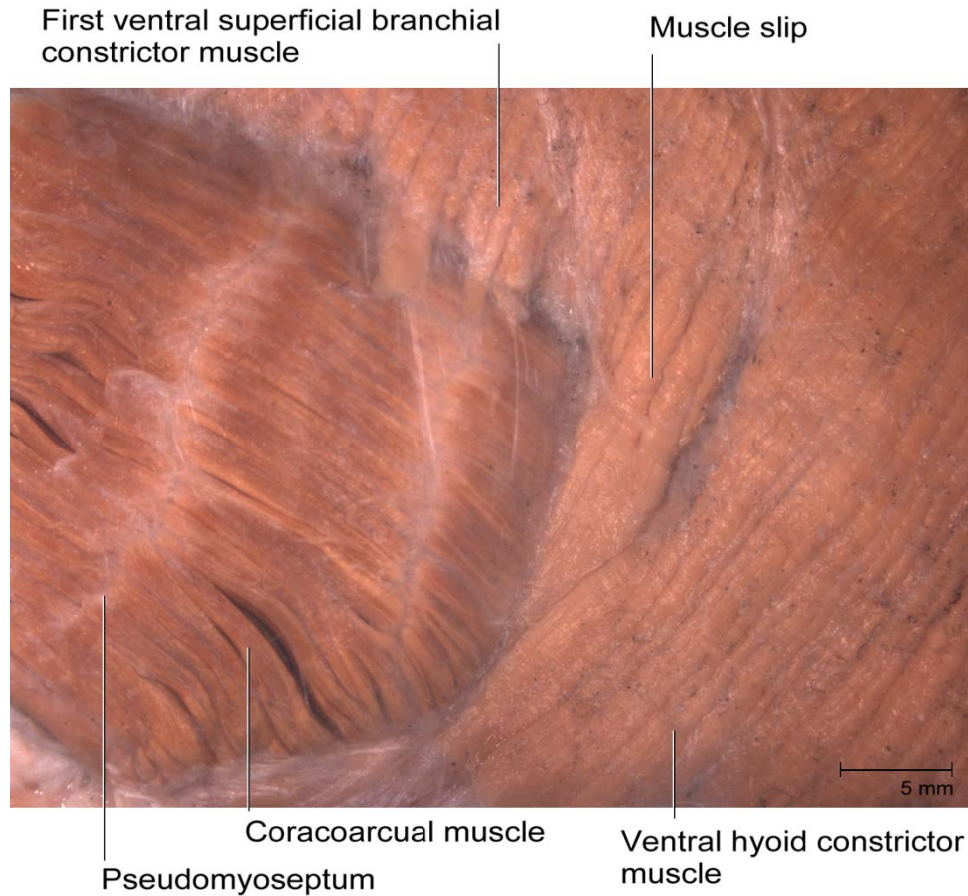


Figure 11. Magnified view of the ventroseptal portion of the first ventral superficial branchial constrictor muscle and the coracoarcual muscle on the right side of the dogfish shark (*Squalus acanthias*) (RJA003) to show the longer cranialmost muscle fiber bundles of the ventral superficial branchial constrictor muscle. The muscle fiber bundles do not uniformly end in myotendinous junctions. Rather, a the slip of muscle fiber bundles passes superficially and rostrally to part of the body of the coracoarcual muscle, while other muscle fiber bundles either terminate on the epimysium of the coracoarcual muscle or continue into the pseudomyosepta of the coracoarcual muscle.

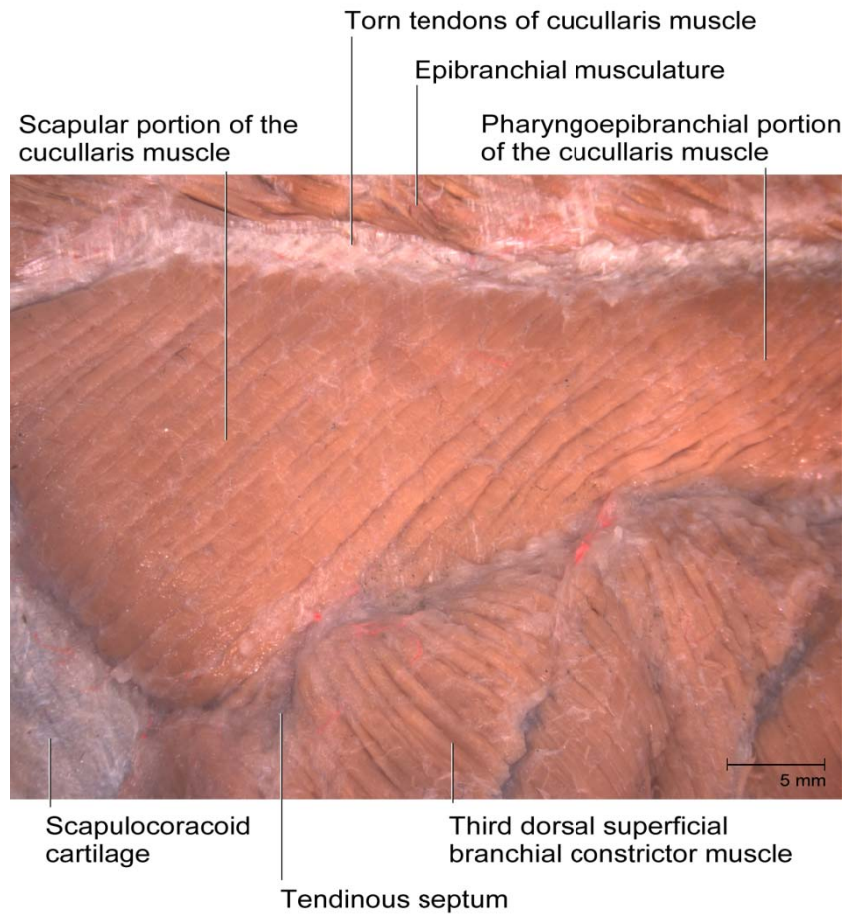


Figure 12. Magnified view of the border between the cucullaris muscle and the vertebroseptal portion of the caudal three dorsal superficial branchial constrictor muscles on the right side of the branchial region of the dogfish shark (*Squalus acanthias*) (RJA003). The dorsalmost muscle fiber bundles of the dorsal superficial branchial constrictor muscles converge towards their caudo-dorsal edge. The torn tendons at the dorsal edge of the cucullaris insert on the superficial fascia, but they were cut during removal of the overlying fascia.

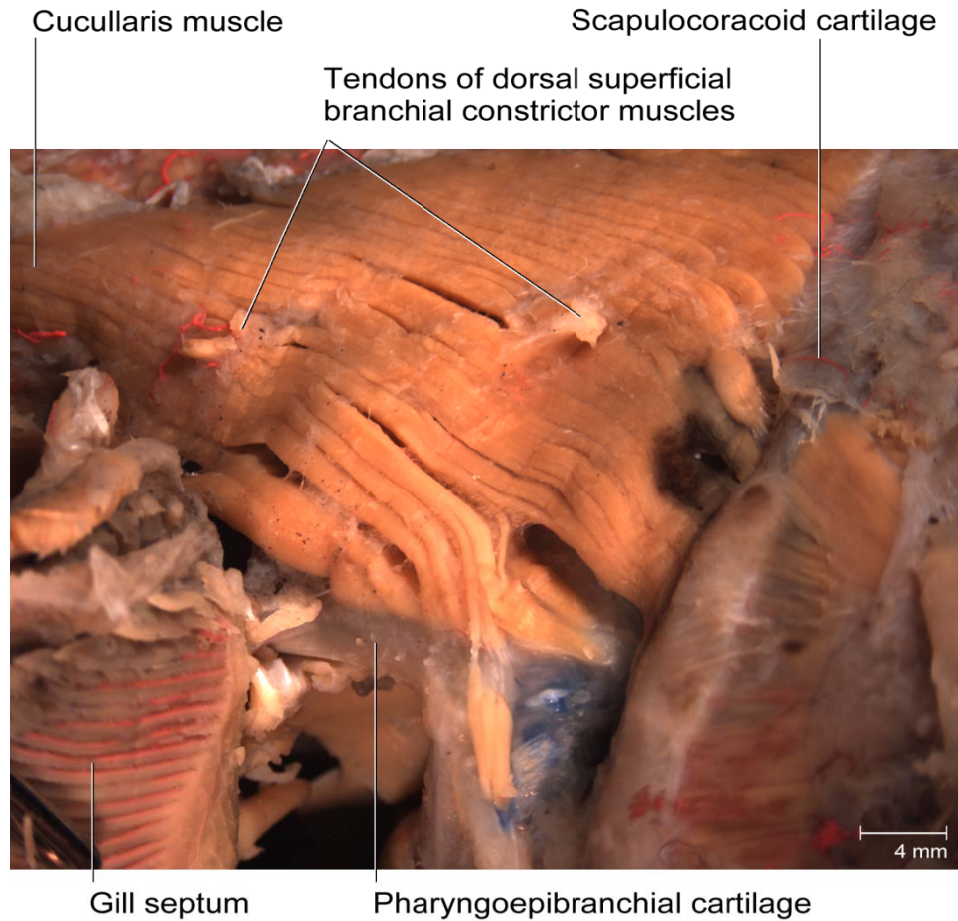


Figure 13. Overview of the cucullaris muscle of the dogfish shark (*Squalus acanthias*) (RJA004). The pharyngoepibranchial part of the cucullaris inserts on the pharyngoepibranchial cartilage, while the scapular part inserts on the scapulocoracoid cartilage. Cut edges of the dorsal superficial constrictor fibers can be seen in the body of the cucullaris muscle.

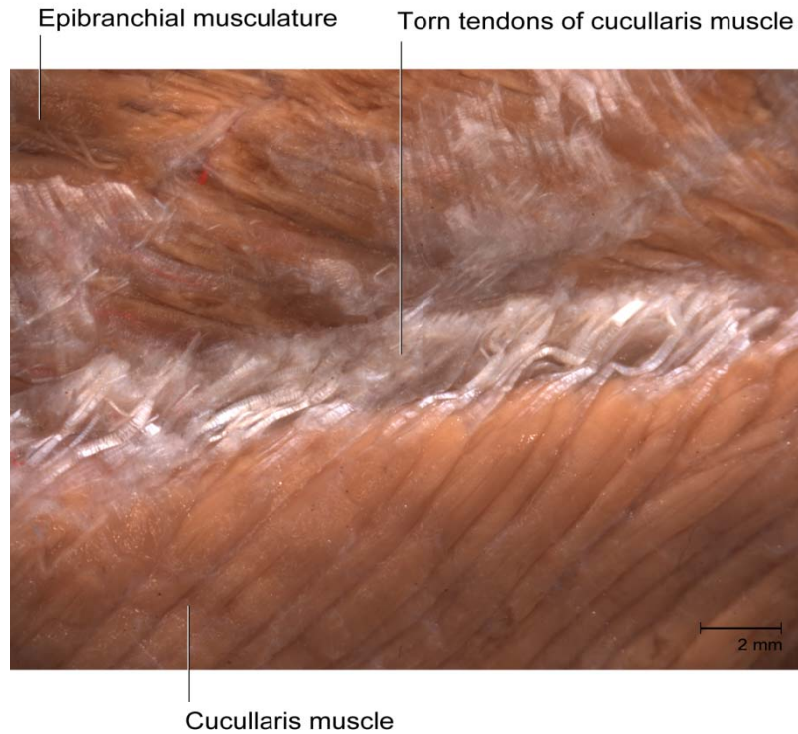


Figure 14. Magnified view of the border between the cucullaris muscle and the epibranchial musculature of the dogfish shark (*Squalus acanthias*) (RJA003). The muscle fiber bundles of the cucullaris muscle fibers end in tendons that continue into the superficial fascia underneath the lateral line organ.



Figure 15. Dorsal view of an undulating dogfish shark (*Squalus acanthias*) to show the angular deviation of the rostrum from the midline. The vertical red line is parallel to the scapulocoracoid cartilage of the pectoral girdle; the horizontal red lines represent perpendicular extrapolations from the pectoral girdle near the left and right glenoid cavities. The horizontal green line marks the halfway point between the horizontal red lines: the position where the rostrum would be if the head did not swing side-to-side during locomotion. The angled green line segment connects the tip of the rostrum with the halfway point between the bases of the pectoral fins. The angle between the green lines is approximately 4.2 degrees. The video used to capture the still images was provided by Anabela Maia and Dr. Cheryl Wilga from the University of Rhode Island (Kingston).

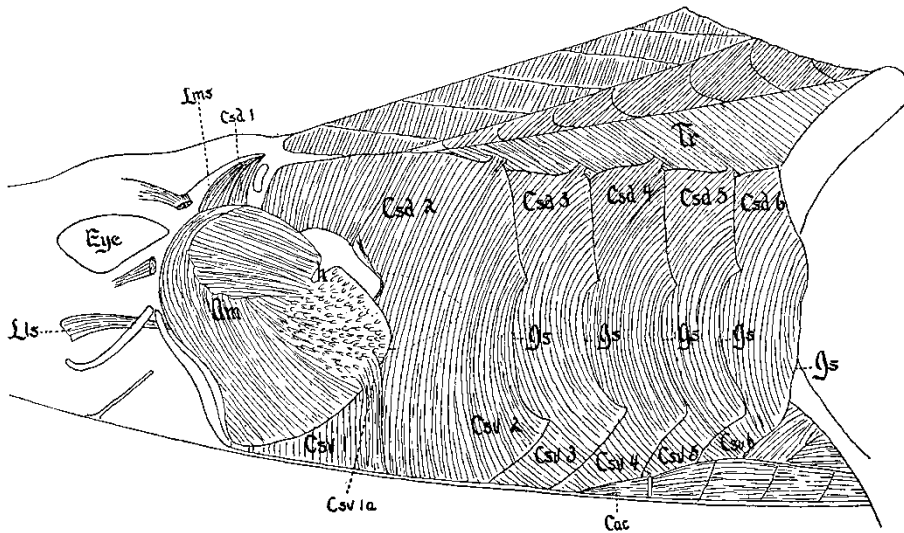


Figure 16. Diagram of a sinistral view of the superficial branchiomeric musculature of the dogfish shark (*Squalus acanthias*) (Marion 1905: Figure 14, page 919). Marion's (1905) observation of the penetration of the vertebroseptal portion of the dorsal superficial branchial constrictor muscles through the cucullaris muscle is evident from his sketch. At the caudo-dorsal corner of each dorsal superficial branchial constrictor muscle, the muscle fiber bundles can be seen to project between the muscle fiber bundles of the cucullaris muscle. Abbreviations: Am = adductor mandibularis; cac = coraco-arcualis communis; Csd 1-6 = constrictores superiores dorsales; Csv 1-6 = constrictores superiores ventrales; Csv1a = constrictor superioris ventralis, anterior fibres; Gs = gill slits; h = hinge of jaws; Lls = levator labialis superioris; Lms = levator maxillae superioris; Tr = trapezius muscle (i.e., cucullaris muscle).

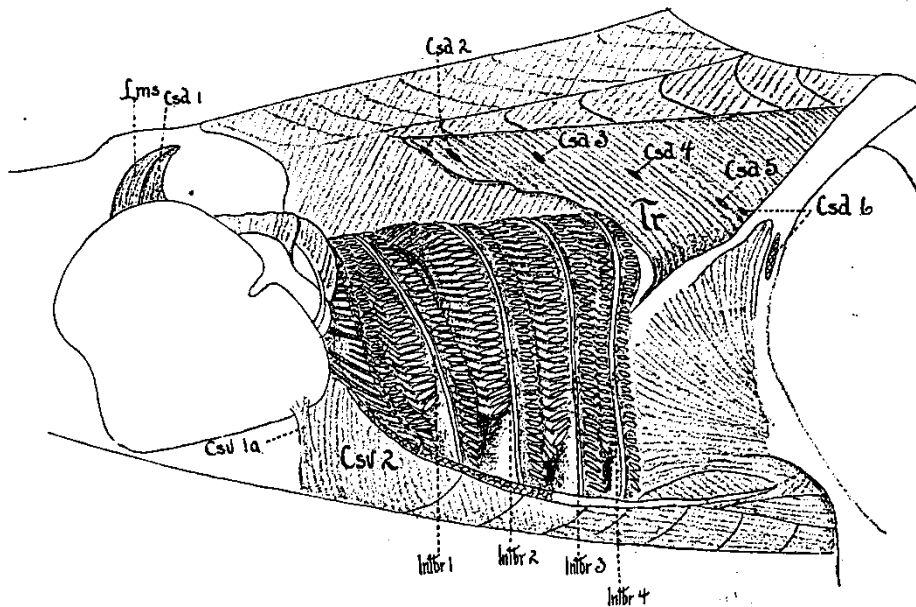


Figure 17. Diagram of a sinistral view of the deep branchiomeric musculature of the Spiny dogfish *Squalus acanthias* (Marion 1905: Figure 11, page 912). The dorsal superficial branchial constrictors have been removed; the positions for the penetration of each successive vertebroseptal dorsal superficial branchial muscle tendon through the cucullaris is indicated. Abbreviations: Csd 1-6 = constrictores superiores dorsales; Csd 1a = constrictor superioris ventralis, anterior fibers; Csd 2 = constrictor superioris ventralis 2 (i.e. ventral hyoid constrictor muscle); Intbr 1-4 = interbranchiales; Lms = levator maxillae superioris; Tr = Trapezius (i.e. cucullaris muscle).