Walking in skates (Chondrichthyes, Rajidae): anatomy, behaviour and analogies to tetrapod locomotion

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Pelvic fin walking in skates is common. However, the structure and function of pelvic fins have not been analysed. Pelvic fins of skates of the genus *Psammobatis* and *Rioraja agassizi* are externally divided into an anterior leg-like lobe and a posterior fin-like lobe. Internally, the anterior lobes are supported by a compound radial, a proximal radial and distal radials that resemble a thigh, a calf and a foot, respectively, and three associated radials arising from the pelvic girdle. A highly developed radial condyle on the pelvic girdle enables broad 'limb' movements. The muscular arrangement of the anterior lobes is formed by protractor, retractor, flexor and extensor muscles, clearly departing from the generalized fin muscle arrangement of elasmobranchs. Walking is composed of propulsion and recovery phases. A backward movement of the compound radial in the horizontal plane characterizes the propulsive phase. The proximal radial connects vertically the compound radial with the foot-like distal radials, which are anchored on the bottom. During the recovery phase, the foot-like structure is lifted off the bottom and the entire limb-like anterior lobe is moved forwards for starting a new cycle. Walking in skates resembles the ancestral tetrapod sprawling locomotion seen in many salamanders and lizards. Pelvic fin anatomy and walking behaviour in skates and hemiscylliid sharks are compared. Ecological and evolutionary implications of walking locomotion in skates are also discussed. © 2002 The Linnean Society of London, *Biological Journal of the Linnean Society*, 2002, **77**, 35–41.

ADDITIONAL KEYWORDS: Elasmobranchii – biomechanics – vertebrate limbs – morphology – mosaic evolution.

INTRODUCTION

The basic functional difference between aquatic and terrestrial paired appendages (fins and limbs) of vertebrates arises from the contrasting physical properties of the media in which they act. The fins of fishes, aquatic turtles and penguins obliquely thrust against the water, the inertia of this dense medium causing it to push with equal force in the opposite direction (although these structures may also perform other functions besides thrust generation; see Davenport & Clough, 1986). The forward component of this force causes the animal to be propelled. In contrast, the thrust of a foot against the ground exerted by a terrestrial vertebrate, in the absence of the buoyancy of

water, should be shared between supportive (in opposition to the pull of gravity) and propulsive force components (Hildebrand, 1988; Walker & Liem, 1994). However, aquatic vertebrates that walk along the bottom combine structural and functional features of both locomotion modes. Although benthic fishes are slightly denser than sea water (Hildebrand, 1988), they are much less denser relative to their medium than terrestrial vertebrates; so relatively light 'limbs' can effectively propel them.

There are a number of ecological situations in which swimming is not the selected option and other locomotive specializations appear in fishes. This is true for several lineages of benthic fishes that walk on the bottom by using different specialized structures (Helfman *et al.*, 1997). Common examples of walking structures in bony fishes are the free pectoral radials of searobins (Scorpaeniformes, Triglidae) (Renous,

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Gasc, Bels & Davenport, 2000), the pelvic fins of flying gurnards (Scorpaeniformes, Dactylopteridae), and the paired fins of mudskippers (Perciformes, Gobiidae) (Moyle & Cech, 1982), and living Australian lungfishes (*Neoceratodus forsteri*, Sarcopterygii) (Pough *et al.*, 1989).

Chondrichthyans are less diverse than actinopterygians but they still have many different lifestyles (Compagno, 1990). Many cartilaginous fishes are benthic and some have locomotive specializations for walking. These include epaulette sharks (*Hemiscyllium* spp., Hemiscyllidae, Orectolobiformes) (Michael, 1993; Goto *et al.*, 1999), legged torpedos (*Typhlonarke* spp., Narkidae, Torpediniformes) (Compagno, 1999a) and legged-skates (Anacanthobatidae, Rajiformes) (Last, 1997; Compagno, 1999a,b). Many skate species have bilobed pelvic fins (Compagno, 1999b). There are some anecdotal comments on the use of pelvic fins for walking in skates (Berestovskiy, 1989; Compagno, 1999b). The showy pelvic fin division into two lobes, a distinctive feature of 'legged skates' of the genera *Cruriraja* and *Anacanthobatis*, leads to the hypothesis of sensorial/locomotor utilization of the anterior lobe (Bigelow & Schroeder, 1953; Last, 1997). However, the internal anatomy and function of pelvic fins have not been analysed among skates. In this paper, the walking behaviour of skates is documented and the anatomy and function of skeleton components and muscles involved in walking are analysed.

MATERIAL AND METHODS

ANATOMICAL OBSERVATIONS

Frozen (−20 °C) specimens of smallthorn sand skate, *Psammobatis rudis*, blotched sand skate, *P. bergi*, shortfin sand skate, *P. normani* (*n* = 3), smallnose fanskate, *Sympterygia bonapartii* (*n* = 2), and Rio skate, *Rioraja agassizi* (*n* = 1) (Rajiformes, Rajidae) were dissected under a Wild Stereomicroscope (Heidelberg, Germany) to assess origin and insertion of pelvic fin muscles. Specimens were cleared and the cartilages were stained with alcyan blue, using a technique described by Wassersug (1976), to survey internal structure of pelvic girdle and fins. The pelvic girdle and fins of narrownose smoothhounds, *Mustelus schmitti* (Carcharhiniformes, Triakidae) were observed for comparative analysis. Skeletal terminology follows Compagno (1999a).

LOCOMOTOR BEHAVIOUR

Two individuals of *P. bergi* (12 cm total length) were kept in a tank of 145.2 L (i.e. $110 \times 33 \times 40$ cm). The fish were caught 2 days prior to the start of observations with a dredge during a research cruise at

38°18′S–57°35′W and 38°22′S–57°31′W, 32 and 44 m depth, respectively. Every observation session lasted at least 30 min. Skates were video-recorded during observations.

FUNCTION OF MUSCLES AND SKELETON DURING WALKING

Function of each muscle and skeletal element was inferred from analysing the correspondence between anatomical (i.e. origin and insertion of muscles, articulations of pelvic fin skeletal elements) and behavioural observations (i.e. movements of every part of the anterior lobe of pelvic fins relative to the others).

RESULTS

ANATOMICAL OBSERVATIONS

Pelvic fins of *P. rudis*, *P. bergi*, *P. normani* and *R. agassizi* are composed of an anterior and a posterior lobe (Fig. 1). The anterior lobe is functionally decoupled from the posterior lobe due to the shortness of radials 4 and 5. Pelvic fins of *Sympterygia bonapartii* were not as split as those from the former species because length of radials gently grade between anterior and posterior lobes.

The anterior lobe of pelvic fins is supported by a compound radial, a proximal radial, distal radials (Fig. 2A) and the three most anterior radials arising from the pelvic girdle (Fig. 1). The base of the posterior lobe is formed by a long basipterygium and the fin web is supported by basipterygial radials.

Pelvic fins are attached to the pelvic girdle by two joints; an antero-lateral joint articulates the radial condyle of the pelvic girdle with the compound radial (Fig. 2B). The semicircular radial condyle is greatly enlarged providing a long articulation surface to the compound radial (Fig. 2B). The proximal part of the compound radial has a long fossa that accommodates the radial condyle (Fig. 2B). The distal end of the compound radial has a large terminal posterior process behind the articulation face of the proximal radial (Fig. 2B). The articulation face of the compound radial is concave, thus accommodating the convex proximal part of the proximal radial. A posterior joint articulates the basal condyle of the pelvic girdle with the basipterygium, which supports the posterior lobe of the fin.

The anterior lobe possesses a complex muscle arrangement that comprises flexor, extensor, protractor and retractor muscles (Fig. 1). Ventral muscles of the anterior lobe include the distal radials' flexor (anterior), originated from the ventral surface of the pelvic girdle and inserted on the antero-ventral part of the distal radials; the proximal radial flexor, originated from the ventral surface of the pelvic girdle
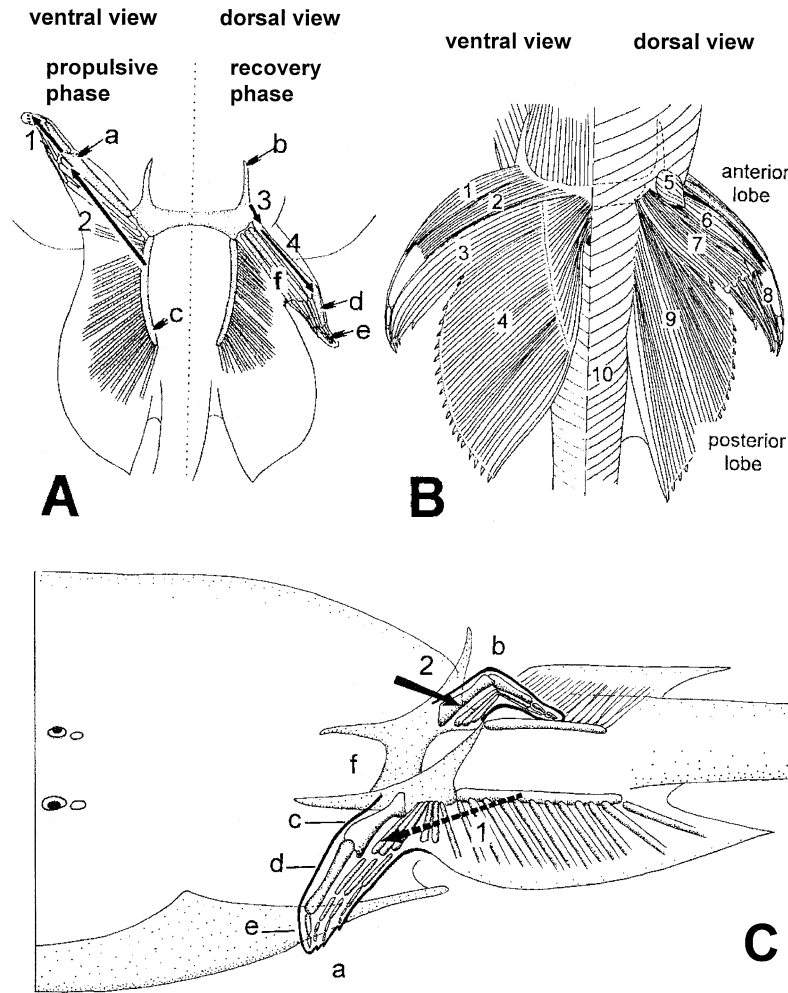
Figure 1. (A) Schematic view of function of muscles and skeletal components during propulsive and recovery phases of walking in skates. (1) Distal radial's flexor (posterior) muscle. (2) Compound radial retractor muscle. (3) Compound radial protractor muscle. (4) Proximal radial extensor muscle (a) Compound radial (b) Pelvic girdle (c) basipterygium (d) Proximal radial (e) Distal radials (f) Three associated radials. Not to scale. (B) Pelvic fin musculature of sand skates of the genus *Psammobatis*. (1) Distal radials' flexor (anterior). (2) Droximal radial flexor. (3) Compound radial retractor. (4) Ventral fin depressor. (5) Compound radial protractor. (6) Proximal radial extensor. (7) Extensor of the compound radial and three associated radials. (8) distal radials' flexor (posterior). (9) Dorsal fin levator muscle. (10) Axial musculature. (C) Dorso-lateral scheme of sprawling locomotion in skates. The anterior lobe of the pelvic fin is shown at the beginning (a) and at the end (b) of the propulsive phase (c) compound radial ('thigh') (d) proximal radial ('calf') (e) distal radials ('foot') (f) pelvic girdle. (1) Compound radial retractor muscle. (2) Compound radial protractor muscle.

medially to the origin of the distal radial flexor, and inserted on the base of the proximal radial; the compound radial retractor, which is originated on the basipterygium and is inserted on the proximal posterior part of the compound radial (deep fibres) and on the three associated proximal and distal radials (superficial fibres). Dorsal muscles of the anterior lobe (Fig. 1) are: the compound radial protractor, which originates from the epaxial musculature and is inserted on the proximal part of the compound radial and onto the fibres (proximal part) of the proximal radial extensor muscle; the proximal radial extensor originates from the base of the compound radial and inserts on the base of the proximal radial; the extensor of the compound radial and the three associated radials originate from the pelvic girdle (iliac process) and insert on the posterior margin of the compound radial and distal part of the three associated radials; the distal radials' flexor (posterior) originate from the postero-distal process of the compound radial and distal part of the three associated radials and insert onto the postero-ventral part of the distal radials.
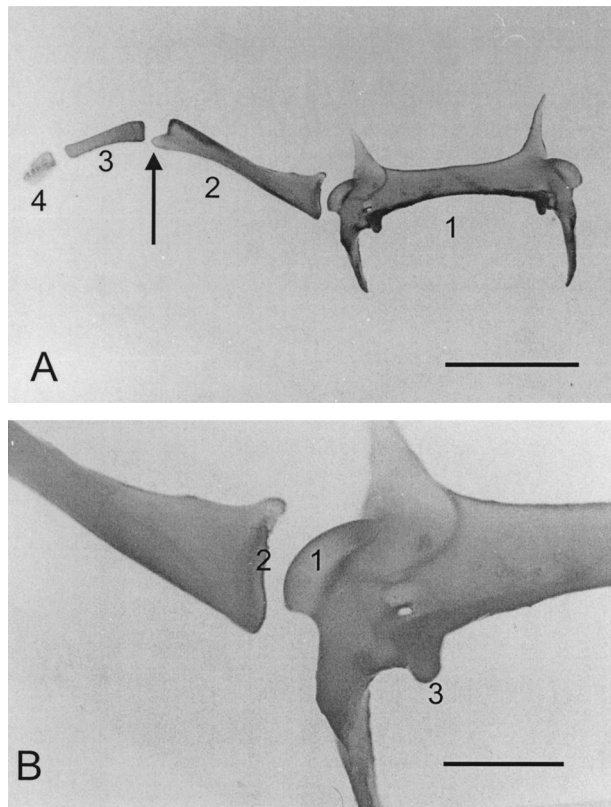
Figure 2. Skeleton of the anterior lobe of the right pelvic fin of a smallthorn sand skate, *Psammobatis rudis*. (A) Ventral view of the whole anterior lobe: (1) pelvic girdle. (2) Compound radial. (3) Proximal radial. (4) Distal radials, the arrow indicates the terminal process of the distal part of the compound radial. Scale bar: 20 mm (B) Detail of the pelvic girdle-compound radial articulation: (1) radial condyle of pelvic girdle. (2) Fossa on the proximal part of the compound radial. (3) Basal condyle of pelvic girdle. Scale bar: 5 mm.

The musculature of the posterior lobe is fairly simple (Fig. 1). It is composed by a dorsal fin levator muscle originated from the epaxial musculature and inserted on the radials, and a ventral fin depressor muscle that originates on the basipterygium and inserts on the radials (Fig. 1).

No substantial differences were observed regarding muscular and skeletal features of pelvic girdle and fins between the three species of *Psammobatis* and *R. agassizi*. Although pelvic fins of *S. bonapartii* are externally not split into two lobes, no internal differences regarding the musculature and skeleton of pelvic fins were found with respect to *Psammobatis* spp. and *R. agassizi*.

Pelvic fin skeleton and musculature of *M. schmitti* are not different from those of a generalized elasmobranch. That is, the anterior margin is supported by a flat compound radial, and the base by the basipterygium. The articulation between the compound radial and the distal radials is not movable. The musculature is fairly simple and is composed of a dorsal levator and a ventral depressor muscle.

LOCOMOTOR BEHAVIOUR

The analysis of video-recorded locomotion behaviour of aquarium-reared specimens of *Psammobatis bergi* (Rajiformes, Rajidae) show that walking is continuous and composed of two phases: propulsion and recovery (Fig. 3). A backward movement of the compound radial in the horizontal plane characterizes the propulsive phase. The proximal radial connects vertically the compound radial with the small foot-like distal radials, which are firmly anchored on the bottom. This anchorage provides the pivoting point needed to carry out a step. During the recovery phase, the foot-like structure is lifted off the bottom and the entire limb-like anterior lobe is moved forwards for starting a new cycle. Left and right anterior lobes were out of phase, i.e. when one was in the propulsive phase the other one was in the recovery phase. This mode of locomotion did not demand the undulation of pectoral fins (i.e. swimming), and the entire skate disk was kept parallel to the bottom. Skates were seen to walk when approaching food, when climbing the tank's walls, and in every short-distance and slow speed movement. Backward walking was also observed. They swam only when escaping at high speeds from a source of disturbance

FUNCTION OF MUSCLES AND SKELETON DURING WALKING

During the propulsive phase, the compound radial (and the three associated radials) acts as a driving lever propelling the skate forward as it swings. At this stage, retraction of the anterior lobe chiefly results from contraction of the ventral compound radial retractor muscle (Fig. 1). The compound radial moves back and forward close to the horizontal plane. The articulation between the semicircular condyle of the pelvic girdle and the fossa on the proximal part of the compound radial allows a broad movement in the horizontal plane. The proximal radial articulating with the compound radial extends downward at a nearly right angle to the compound radial due to the contraction of the proximal radial flexor muscle (Fig. 1). The contraction of the distal radials' (posterior) flexors presses the small distal radials against the substrate, preventing this foot-like arrangement from slipping backward (Fig. 1). During the recovery phase, dorsal extensors extend and protract the anterior lobe. The proximal radial extends due to the contraction of
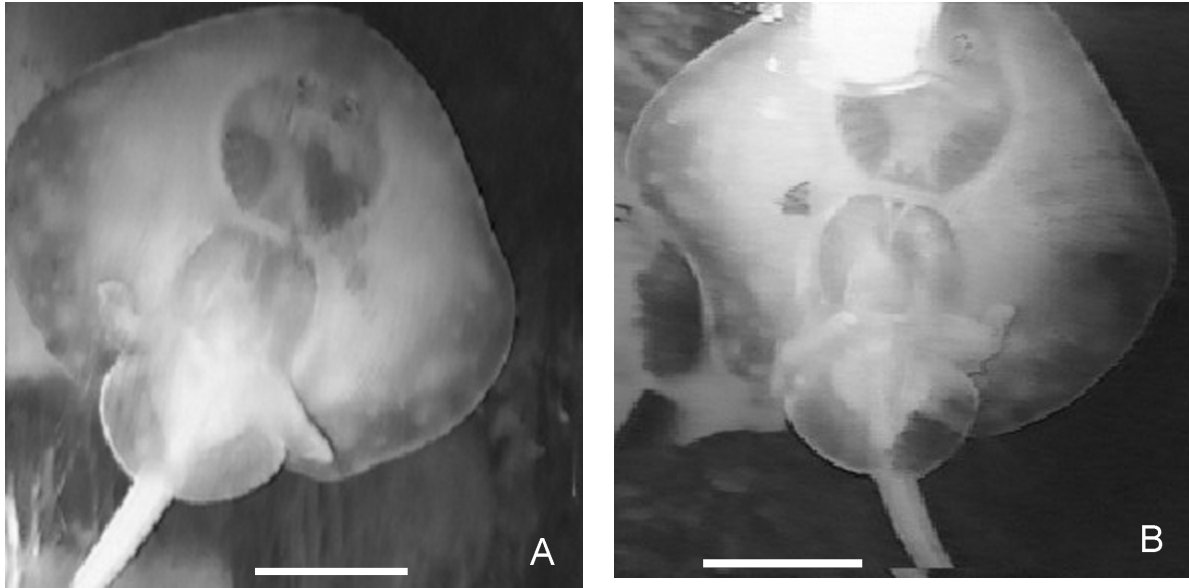
Figure 3. Ventral view of a step succession in a blotched sand skate, *Psammobatis bergi*. In (A) the anterior lobe of the right pelvic fin is in the propulsive phase, while the left anterior lobe is in the recovery phase. The opposite is shown in (B). Scale bars: 30 mm.

the proximal radial extensor muscle. Coincident protraction of the compound radial is accomplished by contraction of a short compound radial protractor muscle arising from the axial musculature (Fig. 1).

DISCUSSION

A walking terrestrial vertebrate advances by a succession of steps in which one foot (or hand) is first placed on the ground to develop a thrust that accelerates the body (propulsive phase of step), and then is removed from the ground and protracted (recovery or swing phase) (Walker & Liem, 1994). The structure and function of the anterior pelvic fin lobe of skates resemble this fundamental pattern of locomotion. In addition, because the proximal 'limb' segment (compound radial) moves back and forth close to the horizontal plane, the two phases of walking resemble the ancestral tetrapod sprawling locomotion seen in many salamanders and lizards (Duellman & Trueb, 1986; Walker & Liem, 1994).

Crawling locomotion in epaulette sharks is performed by undulating the body and moving alternately the slightly modified paired fins (Pridmore, 1995; Goto *et al.*, 1999). Paired fins of *Hemiscyllium* spp. are not split into proximal, mesial and distal parts (i.e. thigh, calf and foot) (Goto *et al.*, 1999) as in skates. As a consequence, the use and internal anatomy of walking fins of epaulette sharks and skates is completely different. Skates do not bend the body while walking: their vertebral column is kept straight

during walking contrasting with the bending reported in hemiscyllid sharks. Most differences in the musculature of both pectoral and pelvic fins between *Hemiscyllium ocellatum* and generalized (i.e. swimming) forms were in the size of muscles rather than in the number and position of muscles (Goto *et al.*, 1999). Goto *et al.* (1999) describe only one additional muscle, the levator pectoralis inferior, in relation to generalized pectoral fins of other sharks. In contrast, our data show that a very different and specialized musculature occur in the pelvic fins of skates as compared to the generalized pelvic fin of a generalized elasmobranch or teleost fish (see Hildebrand, 1988).

Regarding skeletal features, one of the most striking structures of the anterior lobe is the highly developed radial condyle of the pelvic girdle and the deep compound radial fossa (Fig. 2), which greatly enable limb movements in the horizontal plane during both propulsive and swing phases. This condyle is absent or slightly developed in nonwalking elasmobranchs, but it is large and conspicuous in walking elasmobranchs (Goto *et al.*, 1999; this study). As indicated by the structure of the postcranial skeleton in osteolepiform fishes and basal labyrinthodonts (Jarvik, 1980), the articulation between girdles and paired appendages was one of the most important features in the evolution of the appendicular skeleton of walking ancestral vertebrates. Hence the functional importance of this trait in walking skates.

Walking has evolved many times among different lineages of benthic fishes (Helfman *et al.*, 1997). In

epaulette sharks, walking is said to be adaptive for a living in a structurally complex habitat such as coral reefs (Michael, 1993; Goto *et al.*, 1999). Most skates live on sandy or muddy bottoms and are uncommon in reefs (McEachran & Miyake, 1990a; Last, 1997). This implies that, in addition to anatomical and functional differences, walking locomotion has also ecological differences between walking sharks and skates. Our observations indicate that most movements of skates along the bottom are made by walking. Walking may be energetically less expensive than swimming by undulating the massive pectoral fins, because of the smaller muscular mass employed. Also, water displacement could be minimized precluding detection by potential prey and predators. Berestovskiy (1989) observed that captive young thorny skates, *Amblyraja radiata*, use the anterior lobes of pelvic fins as pivots when hunting live prey. This suggests that 'legs' could also be used during hunting.

Our anatomical observations from five species of skates indicate that the internal anatomy (both skeletal and muscular) of pelvic fins is not much variable between species. Although we have examined species from only one clade (the subfamily Arhynchobatinae) of the skate lineage, literature records suggest that walking is widespread within the entire families Rajidae and Anacanthobatidae. All species of the subfamily Rajinae have a skeletal anatomy of pelvic fins similar to that of the arhynchobatine species investigated here (Compagno, 1999a). This suggests that skates of the subfamily Rajinae walk on the sea bottom as observed in arhynchobatine species. The family Anacanthobatidae is more extreme in the division of the pelvic fins into an anterior and a posterior lobe (McEachran & Miyake, 1990b), and it is very likely that these structures are used in walking (Bigelow & Schroeder, 1953; Compagno *et al.*, 1989; Last, 1997). The occurrence of walking in other batoids (e.g. legged torpedos, *Typhlonarke* spp.; Compagno, 1999a) suggests that selective pressures are strong enough as to evolve this mode of locomotion more than once in the batoid lineage.

It is intriguing what developmental processes led to the differentiation of the pelvic fins into an anterior lobe specialized for walking while the posterior lobe remained unchanged, a fact that could be regarded as a 'mosaic evolution' event. The mosaic nature of vertebrate evolution (see examples in Raff, 1996) is the outcome of selection acting on modular body organization to dissociate developmental processes among modules. Nevertheless, the function of the posterior lobe during walking (such as production of either positive or negative lift) could not be rejected until performing a quantitative analysis of walking. It has been shown that the pectoral fins of another walking shark, the white-spotted bamboo shark, *Chiloscyllium plagio-sum*, produce a negative lift while keeping a position on the bottom (Wilga & Lauder, 2001).

The developmental pattern of paired appendage musculature in chondrichthyans differs from the pattern shared by teleosts and tetrapods (Neyt *et al.*, 2000). However, our observations show that both lineages may build up very similar limb structures, regardless the developmental pattern. Skate 'legs', as a strong analogy to tetrapod limbs, argues for a limited number of structural designs responding to functional requirements.

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