

See discussions, stats, and author profiles for this publication at: <https://www.researchgate.net/publication/227661666>

# Feeding mechanism of *Epibulus insidiator* (Labridae; Teleostei): Evolution of a novel functional system

Article in *Journal of Morphology* · November 1989

DOI: 10.1002/jmor.1052020202

CITATIONS

129

READS

223

2 authors:



Mark W Westneat

University of Chicago

199 PUBLICATIONS 6,518 CITATIONS

[SEE PROFILE](#)



Peter C Wainwright

University of California, Davis

239 PUBLICATIONS 14,278 CITATIONS

[SEE PROFILE](#)

Some of the authors of this publication are also working on these related projects:



Collaborative Research: FishLife: genealogy and traits of living and fossil vertebrates that never left the water [View project](#)



2D geometric morphometrics of the avian wing and sternum [View project](#)

## Feeding Mechanism of *Epibulus insidiator* (Labridae; Teleostei): Evolution of a Novel Functional System

MARK W. WESTNEAT AND PETER C. WAINWRIGHT  
*Department of Zoology, Duke University, Durham, North Carolina 27706*  
 (M.W.W.); *Department of Developmental and Cell Biology University of California, Irvine, California 92717* (P.C.W.)

**ABSTRACT** The feeding mechanism of *Epibulus insidiator* is unique among fishes, exhibiting the highest degree of jaw protrusion ever described (65% of head length). The functional morphology of the jaw mechanism in *Epibulus* is analyzed as a case study in the evolution of novel functional systems. The feeding mechanism appears to be driven by unspecialized muscle activity patterns and input forces, that combine with drastically changed bone and ligament morphology to produce extreme jaw protrusion. The primary derived osteological features are the form of the quadrate, interopercle, and elongate premaxilla and lower jaw. *Epibulus* has a unique vomero-interopercular ligament and enlarged interoperculo-mandibular and premaxilla-maxilla ligaments. The structures of the opercle, maxilla, and much of the neurocranium retain a primitive labrid condition. Many cranial muscles in *Epibulus* also retain a primitive structural condition, including the levator operculi, expaxialis, sternohyoideus, and adductor mandibulae. The generalized perciform suction feeding pattern of simultaneous peak cranial elevation, gape, and jaw protrusion followed by hyoid depression is retained in *Epibulus*. Electromyography and high-speed cinematography indicate that patterns of muscle activity during feeding and the kinematic movements of opercular rotation and cranial elevation produce a primitive pattern of force and motion input. Extreme jaw protrusion is produced from this primitive input pattern by several derived kinematic patterns of modified bones and ligaments. The interopercle, quadrate, and maxilla rotate through angles of about 100 degrees, pushing the lower jaw into a protruded position. Analysis of primitive and derived characters at multiple levels of structural and functional organization allows conclusions about the level of design at which change has occurred to produce functional novelties.

*Epibulus insidiator* (Pallas), the sling-jaw wrasse, exhibits an extraordinary ability to protrude its jaws during feeding. *Epibulus* is a monotypic genus in the family Labridae and is widely distributed on coral reefs throughout the tropical Pacific, Indian Ocean, and Red Sea. The extreme protrusion of the mouth of this fish was noted by several early ichthyologists. Renard (1719) refers to *Epibulus* as Bedreiger (the deceiver) and Passer (the thief) and states that this fish "has a long snout hidden in the mouth that it throws out with great agility to capture any prey that comes too close." Bleeker (1862) refers to *Epibulus* as "ikan kakatua sumpit" (the parrot wrasse with the pea shooter), and Delsman ('25) cites a Malayan name "tagi utang" (the dunning creditor). These early references reflect interest in the feeding mechanism of *Epibulus*, which is unique among described fishes. Recent

widespread interest in the biomechanics of feeding in fishes (Alexander, '67; Barel, '83; Lauder, '82, '83; Liem, '78, '80; Motta, '84; Osse, '69) has resulted in the emergence of the teleost skull as one of the clearest examples of evolutionary change in a vertebrate functional system. The protrusion mechanism of *Epibulus* represents a major evolutionary change in this functional complex.

The evolution of major changes in complex functional systems is an issue of long-standing interest in evolutionary biology (Darwin, 1859; Goldschmidt, '40; Lauder, '81a; Simpson, '53; Wake and Larson, '87). How do drastic modifications in complex systems evolve? The unusual feeding mechanism of *Epibulus* provides an excellent opportunity to address this question. Our primary goal in this study is to describe the jaw mechanism of *Epibulus* as a mosaic of ancestral

and derived features. We ask the question, "At what level of structural and functional organization can the unique jaw mechanism of *Epibulus insidiator* be explained?" Our general approach to this question follows that of Lauder and Shaffer ('88) and involves description of character state change (or lack thereof) at multiple levels of musculoskeletal design. We examine cranial bones, ligaments, and muscles at three hierarchical levels of organization: 1) morphological structure, 2) patterns of activity during feeding, and 3) functional role in extreme jaw protrusion. Activity patterns of structures are obtained through high-speed cinematography and electromyography, and the functional role of a structure is defined as the product of a structure's morphology, activity pattern, and interaction with other structures.

Unique biological features may be sources of valuable functional information, yet organisms are combinations of both ancestral and derived features. For research in functional morphology to be understood in a phylogenetic context, functional questions should be addressed in terms of the characters that comprise functional systems. Thus, research on the functional morphology of a complex system should identify activity patterns and functions of features retaining a primitive condition as well as analysis of more recently evolved characters. This approach, which has been pursued by only a few recent studies (e.g., Bemis and Lauder, '86; Lombard and Wake, '86; Schaefer and Lauder, '86), requires the existence of a phylogenetic framework for *Epibulus* and allied genera because decisions about character state polarity are made for each structure of the feeding mechanism at each of the three levels described above. These decisions are made by using recent (Gomon, '79; Gomon and Russel, unpublished) and current (Westneat, unpublished) information regarding the phylogenetic relationships of *Epibulus*, tribe Cheilini, and several outgroups within the Labridae.

#### MATERIALS AND METHODS *Epibulus specimens*

Six live specimens of *Epibulus insidiator* (116–159 mm SL) were collected at Orpheus Island in the central region of the Great Barrier Reef, Australia (146° 33'E, 18° 46'S). Fish were captured with barrier nets in 3 to 5 m of water over the fringing reef crest or reef slope. The fish were maintained in marine aquaria for 2 to 3 weeks prior to being shipped to the University of California, Irvine. Specimens for anatomical study were obtained by hand spear. Dissection of five preserved (10% formalin) and seven fresh/frozen (50–220 mm SL) *Epibulus* specimens aided in morphological analysis. Five additional

specimens (30–80 mm SL) were cleared and double-stained (Dingerkus and Uhler, '77), and five dry skeletons (110–230 mm SL) were prepared by using dermestid beetles.

#### *Cinematography and film analysis*

High-speed cinematography and computer-aided acquisition of quantitative kinematic data were used to identify the activity patterns of bones and ligaments during *Epibulus* feedings. Live *Epibulus* were maintained in 200–400-liter laboratory aquaria on a diet of thawed shrimp (*Penaeus*) and live guppies (*Poecilia*). The natural diet of *Epibulus* on coral reefs is composed of small fishes and a variety of crustaceans (Randall, '83). Live *Epibulus* were trained to feed upon shrimp pieces held by forceps while camera lights were on. Feedings were filmed with a Redlake LOCAM model 51-0008 intermittent high-speed motion picture camera by using two tungsten-halogen 650-W flood lights and 16-mm Kodak 7277 (400 ASA) black and white 4X reversal film. Camera settings were 1/2,000-sec shutter speed, aperture 4.0, and 200 frames/sec frame rate. All feedings were filmed against a white background with a 1 cm<sup>2</sup> grid of lines for reference distance.

Four feeding events for each of four live *Epibulus* were selected from many films for kinematic analysis. The basis for film selection was clarity of focus, determination that the camera was at full speed, and the requirement that the fish maintain a lateral position during feeding. Film analysis involved measurement of angles and distances between moving structures during feeding. Because of the rapidity of the *Epibulus* strike, in most cases every frame of a feeding was analyzed. Figure 1 illustrates the measurement protocol for measured kinematic variables. The vertex point of each angle is listed first in each of the following descriptions.

#### Angles

1. Cranial elevation: first dorsal spine origin (vertex) to dorsal tip of rostrum (maxilla/ethmoid articulation) and to origin of first pectoral fin ray (Fig. 1, LDN).

2. Opercle rotation: anterodorsal tip of opercle at junction with preopercle and hyomandibula (vertex) to posterodorsal tip of opercle and to origin of first pectoral fin ray (Fig. 1, IMN).

3. Interopercle rotation: articulation of interopercle and opercle (vertex) to distal-most tip of interopercle and to origin of first pectoral fin ray (Fig. 1 JFN).

4. Quadrate rotation: quadrate/articular joint (vertex) to dorsal-most visible point of quadrate and to anterior tip of dentary (Fig. 1, EGB).

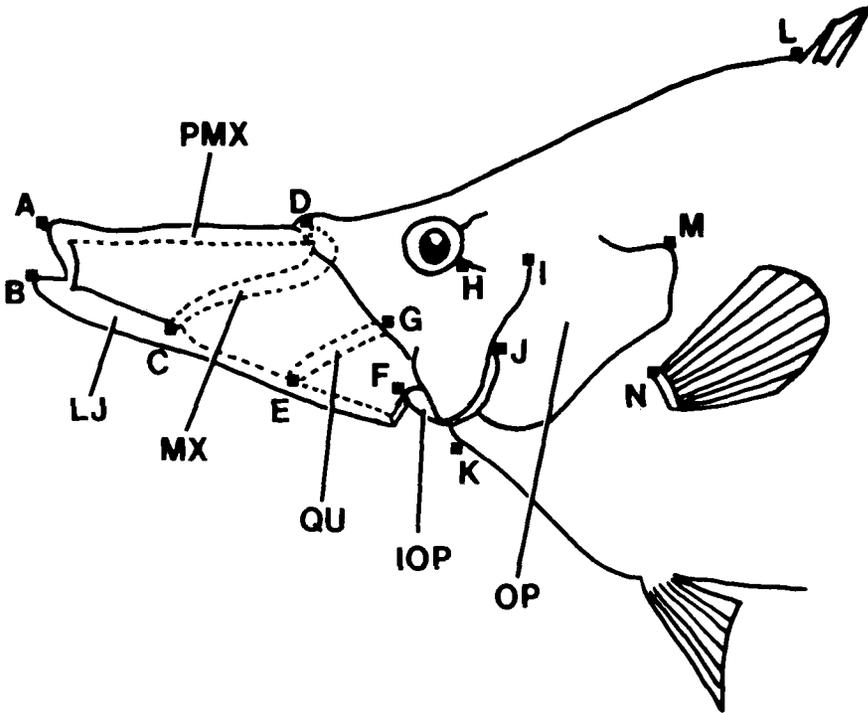


Fig. 1. Diagram of a feeding *Epibulus insidiator*, with points shown (A–N) for recording kinematic variables with a computerized digitizing system. See Materials and Methods (cinematography and film analysis) for list of variables and their measurement points. LJ, lower jaw; other abbreviations of skull bones as in Figure 2.

5. Maxilla rotation: maxilla/articular junction (vertex) to dorsal-most visible point of maxilla and to anterior tip of dentary (Fig. 1, CDB).

#### Distances

1. Gape: anterior premaxilla tip to anterior dentary tip (Fig. 1, AB).

2. Protrusion distance: lowest pigment line behind eye to anterior tip of premaxilla (Fig. 1, HA).

3. Hyoid depression: first dorsal spine origin to ventral-most point below interopercle (Fig. 1, LK).

4. Body-background distance: origin of first pectoral ray to point on background grid anterior to fish and prey (allows body velocity calculation).

5. Jaw-prey distance: premaxilla tip to prey item (allows protrusion velocity calculation).

The origin of the first pectoral ray is used in many of the above variables and it should be noted that change in position of this point could

produce changes in the measured angles. The movement of the first pectoral ray in relation to the fishes body was measured and variation in its position was found to be negligible.

Data acquisition involved projecting high-speed films with an LW stop frame 16-mm film projector onto a Digipad 5 digitizing tablet (GTCO, Columbia, MD). A PC's Limited 286 computer and Sigma-Scan software (Jan del Scientific, Sausalito, CA) were used to record kinematic variables to data files. Data were analyzed and graphically presented by using an Apple MacIntosh IIcx computer and CricketGraph software.

#### Electromyography

To document the motor patterns in four cranial muscles we obtained electromyographic recordings of muscle activity during prey capture in two *Epibulus*. Stainless-steel bipolar electrodes were constructed from two 75-cm-long pieces of poly-coated wire (0.051-mm diameter)

threaded through the barrel of a 26-gauge hypodermic needle. The first 15 cm of the paired wires were glued together with a cyanoacrylate adhesive and 0.5-mm tips were exposed by scraping off the insulation with a razor blade under a dissecting microscope. These tips were bent back against the barrel of the needle to form hooks that helped to hold the electrodes in place through the course of the experiment. Anesthetized fish (tricaine methane sulfonate) were placed into a shallow dissecting tray that was filled half with fresh water and half with the anesthetizing solution and the electrodes were implanted directly into the four muscles. The electrodes were then bundled together and glued into a cable which was secured to a suture looped through the fish's mid-dorsum just anterior to the dorsal fin. Recordings were always made from the left side members of the same four muscles: the levator operculi (LOP), the epaxial muscles (EP), the sternohyoideus (SH), and the adductor mandibulae (AM). All of the muscles were superficial, allowing visual confirmation of electrode placement.

Within an hour of their recovery from anesthesia the fish were offered 1-cm<sup>3</sup> pieces of frozen shrimp (*Penaeus*), which were captured in mid-water. Electromyographic signals were recorded during prey capture on a Bell and Howell 4020AM-FM tape recorder. Signals were amplified by Grass P511J preamplifiers with a band-pass of 100 Hz at the low end and 3,000 Hz at the high end. The 60-Hz notch filter was always used. Electromyograms were recorded at a tape speed of 19 cm/sec and were later digitized with a Keithley 12-bit analogue-to-digital converter at a sample rate of 2,050 Hz.

The sampling rate used when digitizing electromyograms influences the degree to which the signal is faithfully reproduced. We used 2,050 Hz, because 1) a Fast Fourier Transform of representative signals showed that these electromyograms contained insignificant power (less than 5%) in the range above 1,025 Hz so the Nyquist sample criterion was met and aliasing could be eliminated as a source of signal bias, and 2) the variables that were measured from the digitized records were onsets and cutoffs of activity (see below), rather than spike amplitudes or integrated areas. Onsets and cutoffs of activity would not be affected by aliasing. A hard copy for visual inspection was made from the computer file of each feeding on a Gould 260 chart recorder.

The digital computer file from each feeding was then played into a Tektronix 4107 color graphics terminal. To quantify the time course

of muscle activity a computer program assisted in measuring nine variables from the electromyograms of each feeding. The duration of activity of each muscle (LOPDUR, EPDUR, SHDUR, AMDUR) and the onset time of each muscle relative to the levator operculi muscle (LOP-EP, LOP-SH, LOP-AM) were measured in milliseconds. We used the levator operculi as the reference muscle to facilitate comparisons with previously described motor patterns (see below).

To explore the possibility that the feeding mechanism of *Epibulus* is driven by a novel motor pattern we compared our EMG data to similar data obtained from two perciform taxa, *Micropterus salmoides* and *Lepomis macrochirus* (Centrarchidae) (Wainwright and Lauder, '86). The centrarchid data are from feedings on 1-2-cm pieces of earthworm (*Lumbricus*) but were otherwise collected following the same protocol used for *Epibulus*. We contrasted the seven common EMG variables among the two *Epibulus* (14 feedings), five *Micropterus* (50 feedings) and four *Lepomis* (40 feedings) in a nested analysis of variance experimental design, with individuals nested within species; the Systat statistical package (Wilkinson, '86) was used. An overall multivariate analysis of variance (MANOVA) with all seven variables was used to test the null hypothesis that the average motor patterns (defined by the seven EMG variables) of the three species were the same, and subsequent univariate ANOVAs were performed with each variable separately. Because several F ratios were examined in this analysis a conservative probability level of  $P < 0.01$  was used to establish statistical significance.

#### *Muscle stimulation experiments*

To aid our understanding of the mechanism underlying jaw protrusion in *Epibulus*, we observed the functions of five cranial muscles during electrical stimulation experiments on four live fish. All muscles examined were sufficiently superficial in location such that it was possible to observe their actions directly during stimulations. Each fish was first anesthetized (using tricaine methane sulfonate) and two widely spaced monopolar electrodes were implanted into each muscle. The monopolar electrodes were constructed by scraping the insulation from the last 1 mm of 75-cm-long pieces of 0.051-mm stainless-steel wire. The exposed tips were threaded through a hypodermic needle (26-gauge) and the last 3 mm were bent back against the needle's barrel to form a hook which held the electrode in place when the needle was inserted into the muscle belly. Left- and right-side muscles were always stimulated simultaneously. The

electrodes were attached to muscle stimulators (Grass S44 and S48) so that equal electrical stimulation could be delivered to a bilateral pair or simultaneously to several muscles. Muscles were given twitch (2-ms duration) and presumably tetanic stimuli (100 Hz, 2-ms pulse duration, 500-ms train duration, 2–15 V). Muscles were stimulated repeatedly with varying intensity and their actions on the elements of the jaws were recorded by a second observer as a percentage of the maximum action observed during live feedings.

## RESULTS

Our results begin with description of the cranial anatomy of *Epibulus insidiator*, followed by kinematic data from high-speed films and the results of electromyographic and muscle stimulation experiments. Descriptive morphology focuses on the feeding mechanism and the classification of structures as primitive (present ancestrally) or derived (unique to *Epibulus*) and the subsequent discussion draws conclusions regarding character polarity of functional characters as well. A phylogenetic framework is required for such statements of character polarity; therefore we state briefly our concept of *Epibulus* phylogenetic relationships and the outgroup structure for this analysis.

*Epibulus insidiator* is a member of the "cheiline" group within tribe Cheilini and is thought to comprise a monophyletic group with the genera *Cheilinus* and *Wetmorella* (Gomon, personal communication). Presently five probable synapomorphic characters unite this group relative to labrid outgroups (Gomon, '79; Westneat, unpublished data). These characters are: 1) vertebral count = 23 (9–14), 2) epipleural rib count reduced to ten, 3) mesethmoid extends caudad of preorbital process, 4) pectoral ray count = 12, and 5) frontal shelf and recess are lost. Evolutionary polarity decisions for *Epibulus* characters are primarily based on comparisons with character states of *Cheilinus* and *Wetmorella*. Additional outgroups used for comparison include other lineages within tribe Cheilini, the primitive labrid tribe Hypsigenyini (Gomon, '79), and the perciform family Centrarchidae.

### Osteology

#### Jaws

Figure 2 illustrates the major cranial bones and several ligaments that function during jaw protrusion in *Epibulus insidiator*. *Epibulus* exhibits several unique features of cranial osteology, while other features retain a primitive labrid condition. Figure 3 presents the cranial

osteology of *Cheilinus diagrammus* for comparative purposes.

The elongate upper and lower jaws are prominent derived features of *Epibulus* (Fig. 2). The premaxilla (Fig. 2A, PMX) has an extremely elongate ascending process which, when fully retracted, passes posterodorsally through a channel formed by the mesethmoid and frontal bones and extends past the posterior edge of the supraoccipital crest. The alveolar arm of the premaxilla forms the toothed portion of the upper jaw and is a flattened process connected to the dentary via fibrous connective tissue, and to the maxilla via the premaxillary-maxilla ligament. This fibrous connection allows movement of the premaxilla relative to the dentary during increase in gape. The maxilla (Fig. 2A, MX) is laterally flattened in *Epibulus*, and the maxillary head is similar to that of other labrids, except that the dorsal processes of the maxillae meet dorsally around the ascending processes of the premaxillae, forming a guide through which the premaxilla slides.

The lower jaw is also very elongate (Fig. 2), and is formed from a robust, toothed dentary, a long slender articular which makes up about two-thirds of mandibular length, and the angular, which is a bony cap for the posterior tip of the articular. (Bone names are as in Rognes, '73.) The dentary (Fig. 2A, DT) exhibits a well-defined coronoid process above its suture with the articular. The joint between articular and quadrate is located in *Epibulus* forward of the posterior tip of the articular by about 20% of mandibular length (Fig. 2). In *Cheilinus* (Fig. 3) and most perciform fishes, this joint is located at the posterior tip of the articular. In *Epibulus*, the posterior tip of the mandible is the site of insertion of the interperculo-mandibular ligament onto the angular, as is the case in other labrids.

#### Suspensorium

The suspensorium of *Epibulus* is modified in several respects. One key feature is the derived form of the quadrate. The primitive labrid quadrate (and that of most other teleosts) is a flattened bone shaped approximately like an equilateral triangle, with a thickened knob at the anteroventral end where it articulates with the lower jaw (Fig. 3). In contrast, the quadrate of *Epibulus* is very elongate and approximates the shape of a long, thin cylinder (Fig. 2A, QU). The bony knob on the ventral tip and the joint between the quadrate and articular are retained in *Epibulus* from the primitive condition. However, a second joint involving the quadrate is found in *Epibulus*, formed by a bony knob on the dorsal tip of

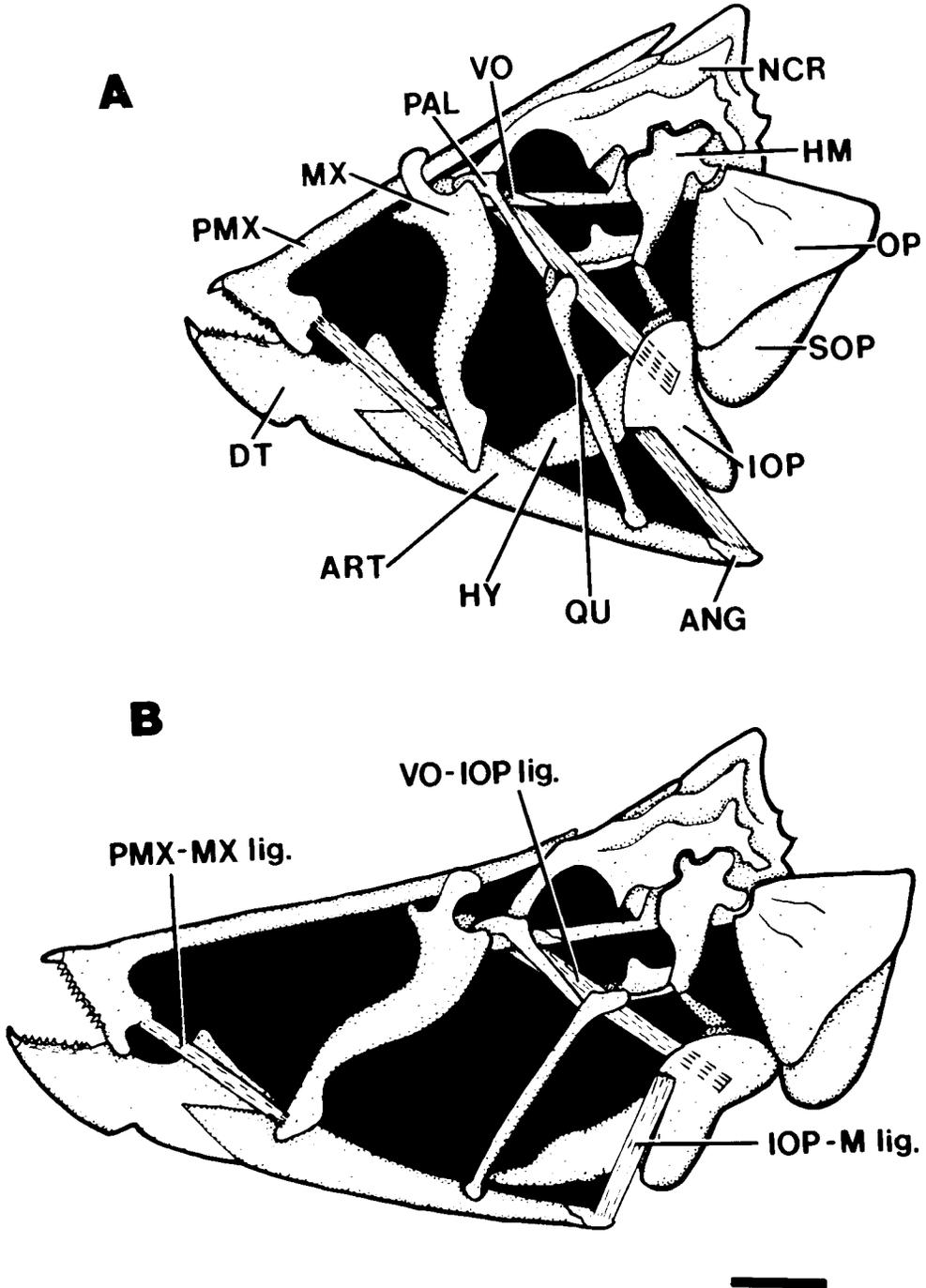


Fig. 2. Cranial osteology of *Epibulus insidiator*, with preopercle and circum-orbital bones removed. A: Jaws are slightly protruded. B: Jaws are almost fully protruded, and ligaments are labelled. ANG, angular; ART, articular; DT, dentary; HM, hyomandibula; HY, hyoid; IOP, interopercle; IOP-M

lig., interoperculo-mandibular ligament; MX, maxilla; NCR, neurocranium; OP, opercle; PAL, palatine; PMX, premaxilla; PMX-MX lig., premaxillary-maxilla ligament; QU, quadrate; SOP, subopercle; VO, vomer; VO-IOP lig., vomero-interopercular ligament. Scale bar = 1 cm.

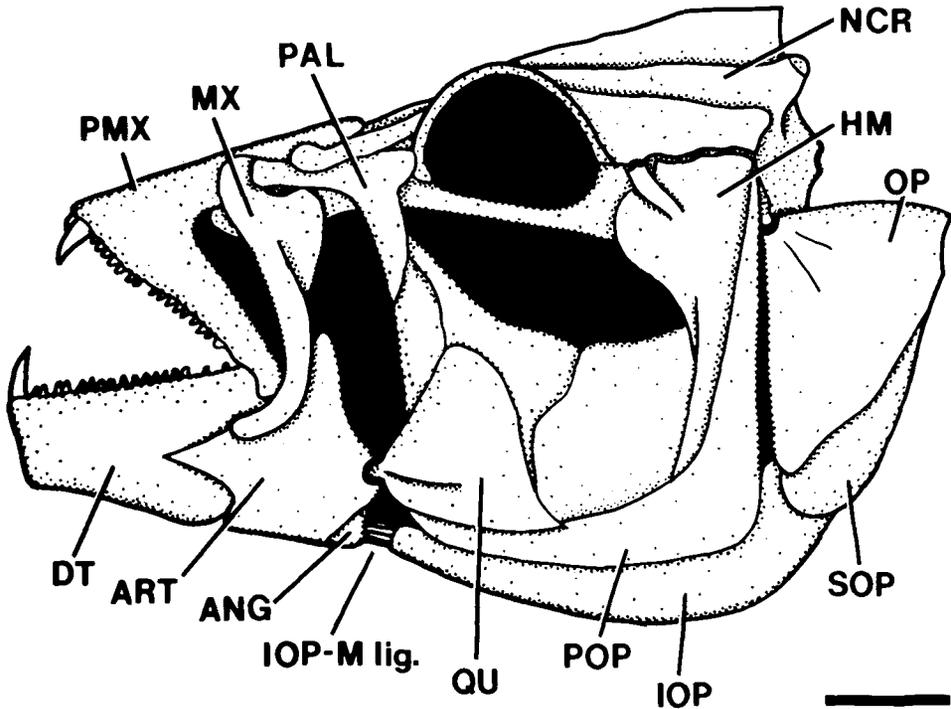


Fig. 3. Cranial osteology of *Cheilinus diagrammus*, a representative of labrid tribe Cheilini, and closely related to *Epibulus*. POP, preopercle; other abbreviations as in Figure 2. Scale bar = 1 cm.

the quadrate where it articulates with the metapterygoid. Such a joint allowing movement of the quadrate is absent in *Cheilinus* and other Perciformes. This derived feature of the *Epibulus* suspensorium frees the quadrate from its ancestral role of stationary support for the lower jaw and enables it to rotate anteriorly. Other aspects of the *Epibulus* suspensorium that deserve mention are the apparent loss of the entopterygoid and reduction in size of both the meta- and ectopterygoid elements.

#### Hyoid

The hyoid apparatus of *Epibulus* (Fig. 2) retains a structure similar to that of *Cheilinus* and other labrids. The hypohyal, ceratohyal, and epihyal form the body of the hyoid apparatus, located in the floor of the mouth. The interhyal extends dorsally from its joint with the posterior tip of the epihyal. The hyoid is involved in joints with the hyomandibula and the interopercle that function during feeding in *Epibulus*. The dorsal tip of the interhyal articulates with the ventral

tip of the hyomandibula. The hyoid bar may rotate around this joint during hyoid depression. The interopercle articulates with the postero dorsal tip of the epihyal and ventral tip of the interhyal via fibrous connective tissue. During jaw protrusion this joint is the axis of rotation of the interopercle during its extreme rotation of nearly 100 degrees.

#### Opercle series

The preopercle, opercle (Fig. 2), and subopercle in *Epibulus* exhibit forms similar to those of other labrids (i.e., *Cheilinus*; Fig. 3). The interopercle, however, exhibits several features apparently unique to *Epibulus* (Fig. 2A, IOP). The shape of the interopercle is atypical, with a thin process (visible in the adducted position; Fig. 2A) extending postero ventrally from the robust site of attachment of the interoperculo-mandibular ligament. In *Epibulus* the connection between interopercle and opercle is limited to a small region on the surface of each bone. The interopercle is thus free to rotate anteriorly.

## Neurocranium

The neurocranium of *Cheilinus* (Fig. 3) and that of primitive labrids such as the *Hypsigenyini* (Gomon, '79) is robust with thickened bony ridges around the large orbit, a ventrally directed vomer, a deep parasphenoid, and a thick supraoccipital crest. The *Epibulus* neurocranium (Fig. 2) is characterized by more delicate structures and a narrow profile. The vomer is directed anteriorly and only slightly ventrally and the parasphenoid is thin and has no ventral process. The supraoccipital crest is thin and reduced relative to the *Cheilinus* condition. A deep trough is present in the dorso medial surface of the *Epibulus* neurocranium, formed by the mesethmoid and frontal bones. The grooves in this trough guide the ascending processes of the premaxillae during protrusion. The flattened frontals and mesethmoid extend posteriorly to cover the anterior portion of the supraoccipital crest and in large specimens this often causes the supraoccipital to lean laterally, producing an asymmetry in the neurocranium.

### Connective tissues

Several connective tissues in the trophic apparatus of *Epibulus* exhibit derived conditions, including a ligament that apparently has arisen de novo in *Epibulus* and ligaments present ancestrally (in *Cheilinus* and other labrids) that are modified in *Epibulus*. An unusual ligament in *Epibulus* originates on the lateral processes and ventral surface of the vomer and inserts on the medial surface of the interopercle (Fig. 2B). This ligament is apparently unique to *Epibulus*, as we have found no evidence of its presence in other labrid taxa and have found no reference to such a ligament in perciform fishes. We refer to this ligament as the vomero-interopercular ligament (Fig. 2B, VO-IOP lig.). It is broad and flattened along its considerable length and the fibers pass beneath the palatine to contact the quadrate. A few fibers insert on the dorsal head of the quadrate, but most pass medial or dorsal to the quadrate to insert on the interopercle. Additional fibers originate on the quadrate and insert onto the interopercle. When tightened by cranial elevation, the vomero-interopercular ligament transfers an antero dorsal force from the neurocranium to the interopercle.

The interoperculo-mandibular ligament is present in *Cheilinus* (Fig. 3) as a fibrous connection between the anterior margin of the interopercle and the posteroventral tip of the lower jaw. In *Epibulus* this ligament (Fig. 2B, IOP-M lig.) retains the same origin and insertion points but is thickened and elongate, forming a broad

band of fibers whose alignment is generally dorso ventral as opposed to the primitive antero posterior orientation. The interoperculo-mandibular ligament of *Epibulus* transfers an antero dorsal force to the lower jaw as the interopercle rotates.

The premaxilla-maxillary ligament is the fibrous connection between the posterior tip of the alveolar process of the premaxilla and the medial surface of the maxillary shaft. The primitive condition (*Cheilinus*) of this ligament is short, allowing minimal movement of the alveolar process of the premaxilla independent of the maxilla. The premaxilla-maxillary ligament in *Epibulus* (Fig. 2B, PMX-MX lig.) takes the derived form of a long, flattened band of fibers that retain the ancestral sites of origin and insertion. The ligament is probably in tension during retraction of the jaws.

It should be noted that connective tissues in *Epibulus* that link bony elements at points of movement or rotation are restricted to fibrous connections at small points on each element, allowing the bones to rotate relative to one another. An example of this is the tight fibrous connection between the postero dorsal tip of the epihyal and the medial surface of the interopercle, around which the interopercle rotates.

### Myology

Figure 4 presents a lateral view of the cheek and head muscles of *Epibulus*. For the purpose of functional description these muscles can be divided into jaw-retracting muscles, jaw-protruding muscles, and those performing other functions. The adductor mandibulae complex of muscles functions in retraction of the jaws and has three divisions, each of which shows little change from the perciform conditions of Winterbottom ('74). The A1 component (Fig. 4) originates on the anterior edge of the preopercle and inserts via a long, continuous tendon onto the head of the maxilla dorsally, and onto the lateral face of the lower jaw (articular) ventrally. When the jaws are fully protruded, A1 contraction serves to rotate the maxilla posteriorly and pull the lower jaw posteriorly and dorsally, closing the jaws. The A2 division also originates on the antero dorsal edge of the preopercle and passes under the A1 medially. The A2 fuses with the A3 division below the origin of the A3 on the hyo-mandibula and pterotic. The combined A2 and A3 fibers (Fig. 4) are aligned primarily dorso ventrally as they continue below A1 as a thin band of muscle and insert tendinously on the medial face of the articular. Contraction of the A2/A3 muscle fibers after protrusion has occurred functions in retracting the lower jaw (see Results: Muscle Stimulation).

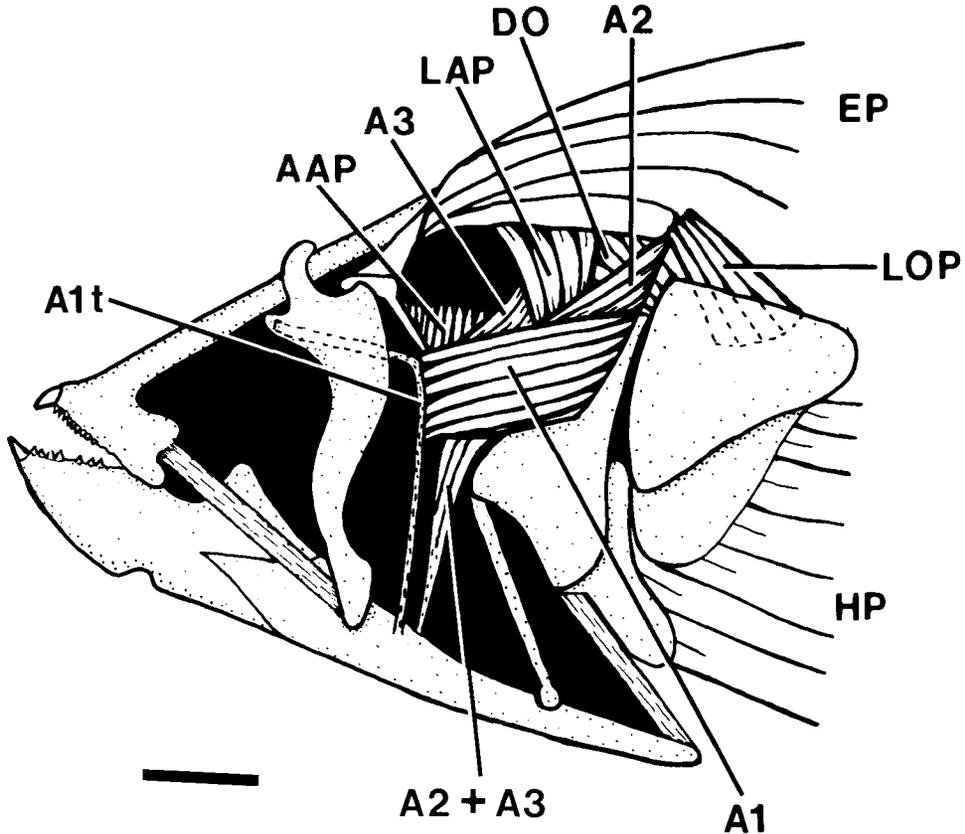


Fig. 4. Cranial muscles of *Epibulus insidiator*. AAP, adductor arcus palatini; A1, A2, A3, divisions of the adductor mandibulae; A2 + A3, combined A2 and A3 divisions; Alt, tendon from A1 muscle to maxilla and lower jaw; DO, dilatator operculi; EP, epaxialis; HP, hypaxialis; LAP, levator arcus palatini; LOP, levator operculi. Scale bar = 1 cm.

Jaw protrusion is effected by the epaxial (EP) and levator operculi (LOP) muscles (Fig. 4). Both of these muscles exhibit the structural condition found in *Cheilinus* and many perciforms. The epaxial muscles are extensive on the dorsal surface of the neurocranium, inserting onto the supraoccipital, parietal, frontal, and lateral ethmoid elements. Epaxial contraction elevates the neurocranium. The levator operculi originates on the pterotic as a flat sheet of muscle fibers and inserts on the dorsal and dorsomedial surfaces of the opercle. Levator operculi contraction functions in rotation of the opercle postero dorsally. Also pictured in Figure 4 are the dilatator operculi (DO: origin, sphenotic and pterotic; insertion, opercle), levator arcus palatini (LAP: origin, sphenotic; insertion, hyomandibula), and adductor arcus palatini (AAP: origin, parasphenoid; insertion, hyomandibula, and metapterygoid).

Figure 5 illustrates the geniohyoideus (GH; = protractor hyoideus, Winterbottom, '74) and the sternohyoideus (SH) muscles of the ventral surface of the head of *Epibulus*. The geniohyoideus is extremely elongate in *Epibulus*, extending from its anterior site of attachment on the dorsal surface of the dentary symphysis to a posterior attachment on the paired ceratohyals. This muscle occupies the ventral midline between the two halves of the lower jaw as a narrow column of fibers wrapped by a cylinder of connective tissue. This connective tissue appears to exhibit a crossed helical fiber arrangement. The geniohyoideus muscle becomes bifid at the anterior end of the urohyal and continues postero laterally to its paired attachments on the medial surface of each ceratohyal. Preliminary measurements of geniohyoideus length from its anterior attachment to the point where it becomes bifid indicate that the extended length of the muscle

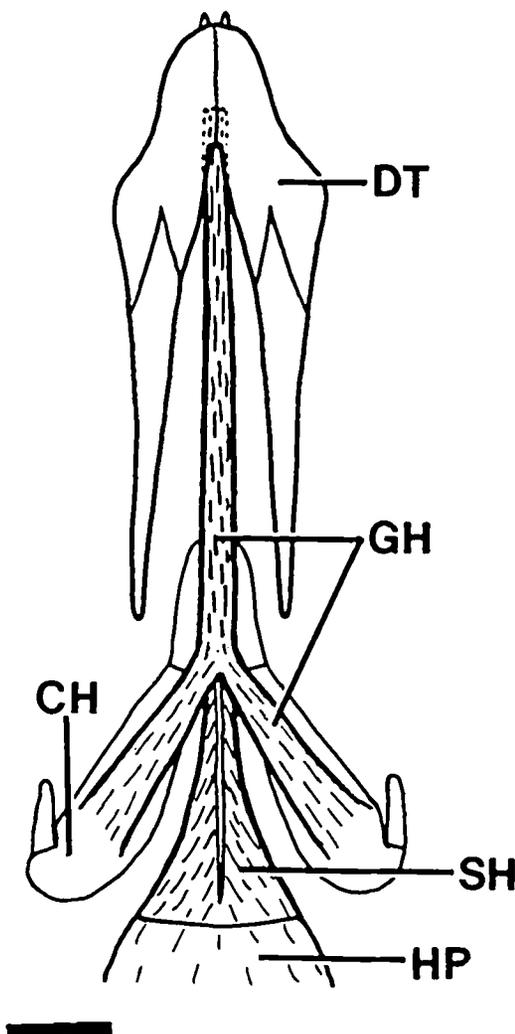


Fig. 5. Ventral head muscles of *Epibulus insidiator*, with dentary (DT) and ceratohyal (CH) bones labelled. HP, hypaxialis; GH, geniohyoideus; SH, sternohyoideus. Scale bar = 1 cm.

is approximately twice that of its contracted length.

The sternohyoideus (Fig. 5, SH) originates dorsally on the cleithrum and ventrally on the anterior face of the myoseptum separating the sternohyoideus from the hypaxialis (HP). The sternohyoideus inserts onto the urohyal anteriorly.

#### General feeding behavior

Prey capture in *Epibulus insidiator* may be divided into three components: 1) approach to

the prey, 2) the strike, and 3) prey processing/swallowing. Table 1 presents data on strike characteristics for *Epibulus*, and Figure 6 presents frames from a film of *Epibulus* during the strike phase. Prey items appeared to be detected visually and were approached to within 30–40 mm (about 25% of SL or 70% of head length) before strike initiation (Table 1). Body velocity slowed to near zero during the strike phase and maximum protrusion velocity of the jaws during the strike was always over 2 m/sec (Table 1). Prey were engulfed by the jaws within 25–40 ms of strike initiation. *Epibulus* employs suction during feeding, as can be seen in Figure 6 (frames 6–8) where the dark prey item can be seen moving into the buccal cavity at a speed of over 2 m/sec (Table 1). Retraction of the jaws was completed within 100 msec of strike initiation. Jaw retraction was accompanied by visible swallowing movements and by a forward sweep of the pectoral fins to back away from the site of prey capture.

#### Film kinematics

Description and analysis of the kinematics of cranial bones during feeding will allow interpretation of the mechanism of jaw protrusion in *Epibulus*. Figure 6 presents frames from a high-speed film of an *Epibulus* feeding. Figure 7 illustrates the mechanism of jaw protrusion during feeding in the form of schematic mechanical diagrams. The diagram numbers in Figure 7 correspond to the frame numbers in Figure 6. The most evident feeding mechanics in Figures 6 and 7 are extreme protrusion of both the upper and lower jaws, rotation of the neurocranium, opercle, interopercle, quadrate, and maxilla, and increase in gape. These variables can be divided into input and output kinematics. Input kinematics are those directly caused by muscle contrac-

TABLE 1. Characteristics of the strike of *Epibulus insidiator*<sup>1</sup>

Characteristics	Mean	SD
Body velocity during strike (m/sec)	0.05	0.04
Prey distance at strike (mm)	36.90	6.90
Time to prey capture (msec)	34.70	6.50
Time to jaw retraction (msec)	111.30	9.40
Maximum protrusion velocity (m/sec)	2.31	0.45
Maximum protrusion acceleration (m/sec <sup>2</sup> )	111.20	43.80
Suction velocity of prey (m/sec) <sup>2</sup>	2.34	0.66

<sup>1</sup>Data are chiefly calculated from analysis of 4 feedings from 4 individuals; tabulated as means and standard deviations (SD) of strike characteristics.

<sup>2</sup>Data are from 3 feedings from 1 individual.

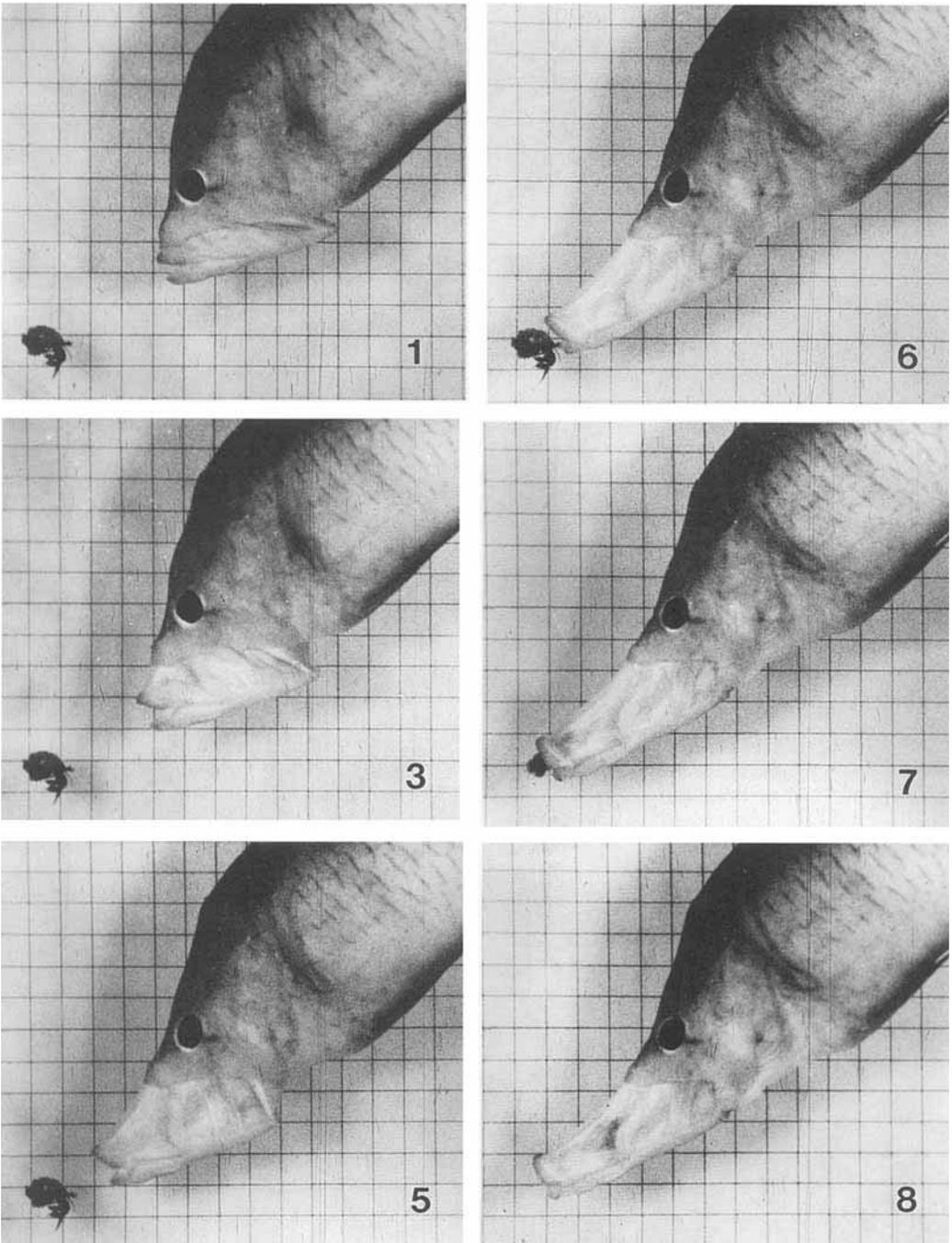


Fig. 6. **Frames 1, 3, 5, 6, 7, and 8:** From a high-speed film (200 frames/sec) of the strike of *Epibulus insidiator*. Successive frames are 0.005 sec apart. Note rotation of quadrate, maxilla, and interoperculo-mandibular ligament. Suction is apparent in frames 6, 7, and 8. See Figure 7 for corresponding mechanical diagrams.

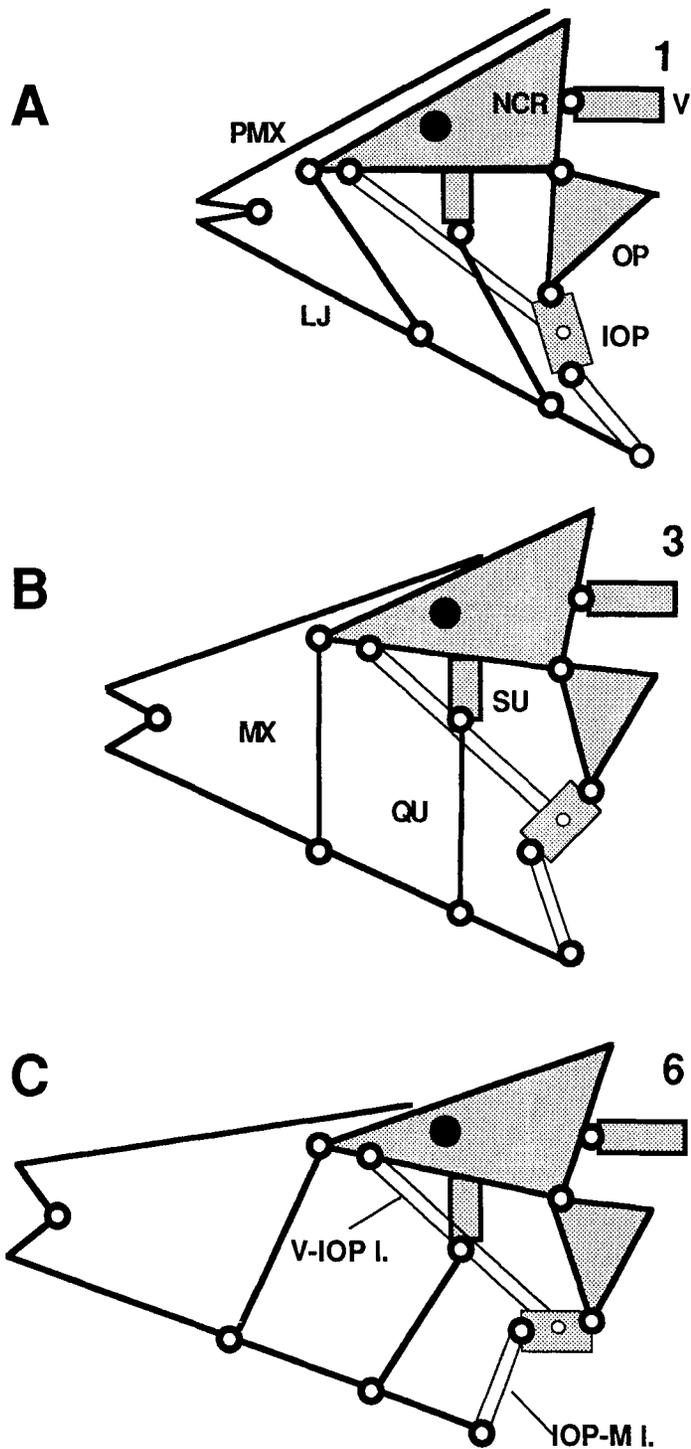


Fig. 7. Mechanical diagrams illustrating the feeding mechanism and kinematics of *Epibulus insidiator* during feeding. **A:** Retracted position, as in Figure 6, frame 1. **B:** Partially protruded position, as in Figure 6, frame 3. Note cranial elevation, opercle rotation, and interopercle rotation. **C:** Almost fully protruded position, as in Figure 6, frame 6. Note rotation of maxilla and quadrate. Abbreviations as in Figure 2.

tion (cranial elevation and opercular rotation), whereas output kinematics are caused by a transfer of force, through connective tissue or bone articulations, from one skeletal element to another.

Figure 8 presents quantitative kinematic data in graphic form for the variables of cranial elevation, opercular rotation, interopercular rotation, quadrate rotation, maxillary rotation, protrusion distance, gape distance, and hyoid depression. These kinematic profiles reveal the relative importance of cranial and opercular input to jaw protrusion in *Epibulus*. Cranial elevation and opercular rotation reach a maximum of around 10 degrees of arc. Cranial elevation (Fig. 8A) reaches its maximum arc within 35 msec of the start of the strike. The output movements of interopercular rotation (Fig. 8C), quadrate rotation (8D), maxillary rotation (8E), protrusion (8F), and gape (8G) are all virtually synchronous with cranial elevation. Peak opercular rotation (Fig. 8B), however, occurs about 15 msec before the synchronous movements of the other variables.

The highly synchronous pattern of movement among many of the variables depicted in Figure 8 is typical of every *Epibulus* feeding analyzed and was followed in most cases by peak hyoid depression approximately 15 msec later. The rotations of the interopercle, quadrate, and maxilla peak at approximately 100 degrees. Protrusion distance data (Fig. 8F) illustrate the extreme protrusion ability of *Epibulus*, with a maximum of over 3 cm, as well as the velocity of the strike (the slope of Fig. 8F is steep).

The mechanism of jaw retraction involves a reverse of the kinematic pattern of bone movements. (Note the bell-shaped appearance of the kinematic profiles in Fig. 8.) Muscle stimulation experiments (Table 3) indicate that jaw retraction is driven primarily by the adductor mandibulae muscles exerting posteriorly and dorsally directed forces upon the maxilla and the lower jaw. The A1 division (Fig. 4) pulls posteriorly upon the maxillary head, providing a posterior rotation in that element that serves as the input movement for jaw retraction. The A1 and the combined A2/A3 divisions of the adductor mandibulae muscle both insert on the lower jaw (Fig. 4) and transmit postero dorsal forces to the mandible that contribute to jaw retraction.

#### *Electromyography*

The motor pattern exhibited by *Epibulus* at the strike is summarized in Figure 9A, where the durations and the relative timing of activity are averaged for the 14 feedings from the two experimental fish. The strike is characterized by

broadly overlapping activity in all muscles, beginning with activity in the levator operculi and the epaxial muscles. This is followed by the onset of activity in the sternohyoideus and then the adductor mandibulae. Average durations of activity range from 33 msec for the epaxial muscle to 45 msec in the sternohyoideus.

The sequence of muscle activity in *Epibulus* appears to be the same as in *Lepomis* and *Micropterus*. The overall MANOVA comparison of the motor pattern in *Epibulus*, *Lepomis*, and *Micropterus* showed no significant difference among the three species (MANOVA comparison of species: Wilkes's  $\lambda = 0.026$ ,  $F = 1.48$ , d.f. = 14,4;  $P = 0.381$ ). This result is also reflected in the univariate ANOVAs presented in Table 2. Univariate ANOVA tests indicate that none of the seven EMG variables differed significantly among the three species (Table 2), although the test for significance of the relative onset time between the levator operculi and the adductor mandibulae had a very high F-ratio which approached significance at the 0.01 level (Table 2, LOP-AM). In contrast, six of seven variables showed significant among-individual variance.

#### *Muscle stimulation*

Anatomical analysis indicates that premaxillary protrusion may be effected by both the levator operculi (via the linkage from the opercle to the rotating interopercle) and the epaxial muscles (via the vomero-interopercle ligament). Stimulation of these muscles individually resulted in approximately 30% of maximum jaw protrusion (Table 3). However, when the two muscles were stimulated simultaneously up to 70% of maximal protrusion was observed. This was the greatest amount of protrusion attained during the muscle stimulation experiments.

Stimulation of the sternohyoideus caused depression of the hyoid but did not move the jaws. When the geniohyoideus was stimulated from an outstretched position (the jaws fully or partially protruded) the muscle shortened and tightened the intermandibular fascia but did not retract the jaws. Stimulation of the adductor mandibulae (A1) caused firm and complete retraction of the protruded jaws. This muscle effects retraction from any stage of jaw protrusion, even though the angle of insertion that this muscle makes against the dentary changes through about 90° during a full retraction stroke.

#### DISCUSSION

*Epibulus insidiator* exhibits the highest degree of jaw protrusion known among fishes. The feeding mechanisms of this fish is described be-

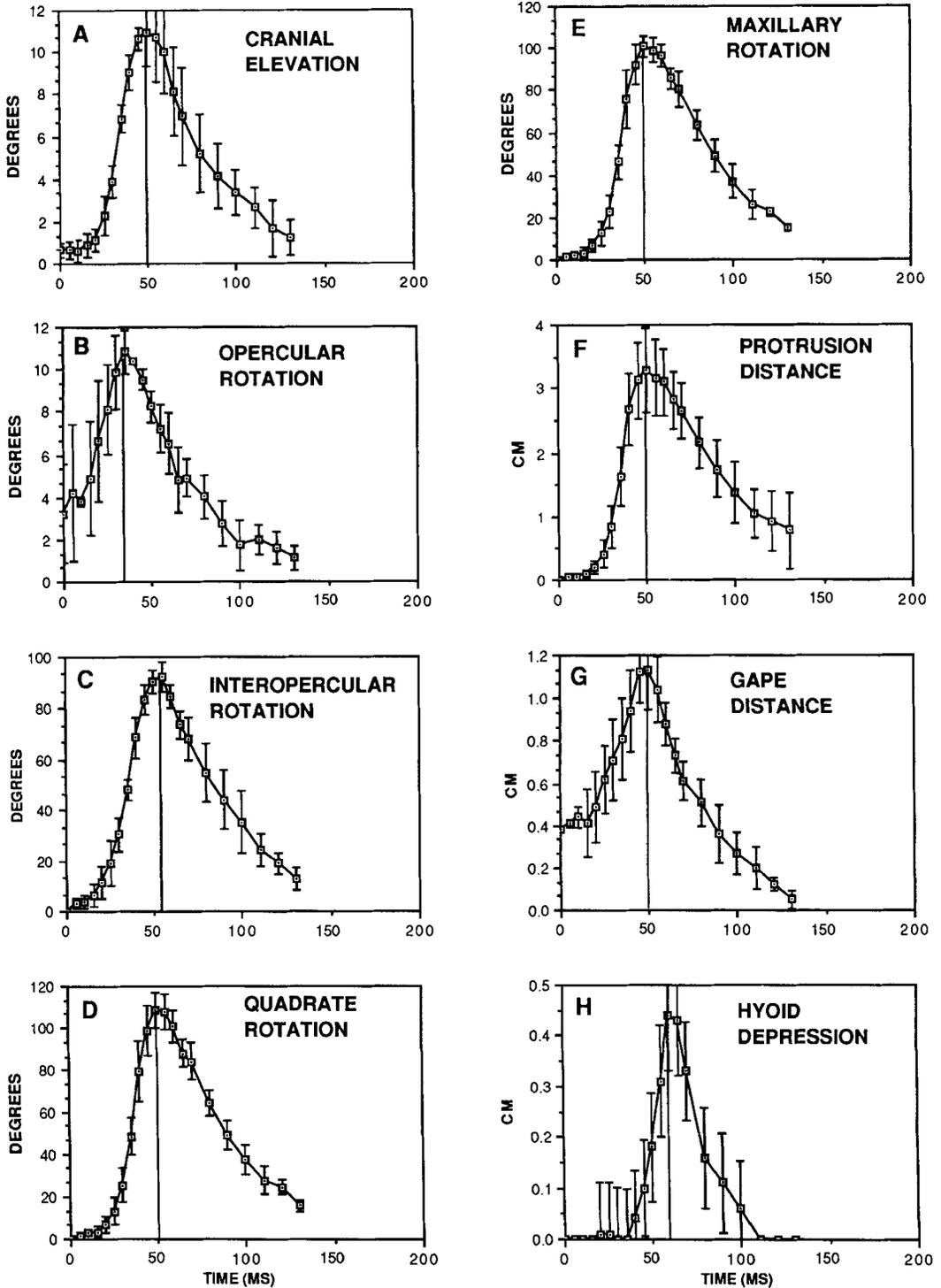


Fig. 8. Graphs of kinematic movement of cranial bones during feeding in *Epibulus insidiator*, resulting from measurements taken from four films each of four individuals (N = 16) feeding on shrimp (*Peneus*). Error bars are standard deviations and graph lines are not the result of fitting techniques.

Vertical lines indicate time of maximum displacement. A: Cranial elevation. B: Opercular rotation. C: Interopercular rotation. D: Quadrate rotation. E: Maxillary rotation. F: Protrusion distance. G: Gape distance. H: Hyoid depression.

TABLE 2. Results of F-tests from nested ANOVAs comparing 7 electromyographic variables among *Epibulus*, *Lepomis*, and *Micropterus*<sup>1</sup>

Variable	Factor	
	Species (2, 8) <sup>2</sup>	Individuals (8, 74-92)
LOPDUR	1.40	3.48*
EPDUR	1.29	6.54*
SHDUR	3.16	3.73*
AMDUR	3.14	2.77*
LOP-EP	0.48	2.81*
LOP-SH	0.14	4.12*
LOP-AM	8.06	1.82

<sup>1</sup>Table entries are the F-ratios from the significance tests. See Materials and Methods (electromyography) for definition of abbreviations.  
<sup>2</sup>( ) = degrees of freedom for F-test.  
 \* =  $P < 0.01$ .

low, followed by a discussion of the levels of organization at which evolutionary changes have occurred to produce this unusual mechanism of jaw protrusion. Three levels of organization are examined for this purpose: 1) the structures involved in feeding, 2) the activity pattern of bones and muscles, and 3) the functional role of each element in the feeding mechanism. *Epibulus* clearly exhibits major changes in the structure of many bones and ligaments of the head, and films and electromyograms reveal the activity patterns of bones and muscles during feeding. These structural and functional data are interpreted in terms of the level of organization at which each element of the jaws exhibits a primitive or derived condition. Thus, each element of the jaws of *Epibulus* exhibits a character state at each of the three levels described above, and a decision of evolutionary polarity is made at each level.

*Mechanism of jaw protrusion*

Analysis of the kinematics of *epibulus* feeding (Fig. 8), consideration of morphology (Figs. 2, 4), electromyography (Fig. 9), and muscle stimulations (Table 3) are combined to propose a mechanism by which this fish captures prey. Figure 7 presents mechanical diagrams of the *Epibulus* jaw mechanism in retracted (7A), par-

Fig. 9. Bar diagrams showing the average motor pattern exhibited by three fish species, *Epibulus insidiator* (A), *Lepomis macrochirus* (B), and *Micropterus salmoides* (C), during prey capture. Mean duration of activity in each muscle is indicated by the length of each bar and average relative onset time is shown with reference to the levator operculi. Left-hand error bars are standard deviations of relative onset times and error bars on the right are standard deviations of muscle burst duration. See Materials and Methods (electromyography) for muscle abbreviations. Sample sizes: (A) 2 fish, 14 feedings; (B) 4 fish, 40 feedings; (C) 5 fish, 50 feedings. See Table 2 for statistical comparison of motor patterns.

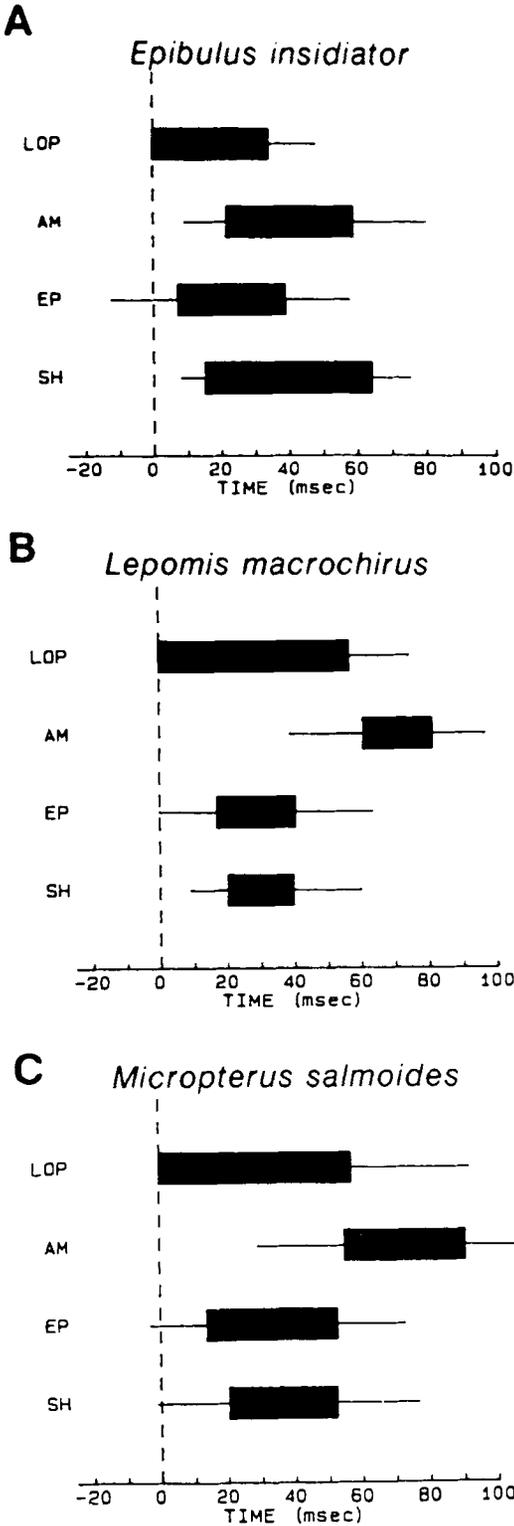


TABLE 3. Actions of cranial muscles during direct electrical stimulation<sup>1</sup>

Muscle	Action
Levator operculi	Opercular rotation, interopercular rotation, jaw protrusion (20% of maximum)
Epaxialis	Cranial elevation, interopercular rotation, jaw protrusion (30% of maximum)
Levator operculi & epaxialis	Opercular rotation, cranial elevation, interopercular rotation, jaw protrusion (70% of maximum)
Sternohyoideus	Hyoid bar depression, no jaw movement
Geniohyoideus	Tightening of intermandibular fascia, no jaw movement
Adductor mandibulae (A1)	Strong jaw retraction from all stages of protrusion

<sup>1</sup>See Materials and Methods for the experimental protocol.

tially protruded (7B), and nearly fully protruded (7C) views. Jaw protrusion is driven by the input motions of cranial elevation and opercular rotation, effected by contraction of epaxial and levator operculi muscles, respectively. Cranial elevation exerts an antero dorsal force along the length of the vomero-interopercular ligament (Figs. 2B, 7C, VO-IOP lig.). While opercular rotation pulls the dorsal tip of the interopercle posteriorly the vomero-interopercular ligament imparts an anteriorly directed force to the ventral portion of the interopercle. Evidence from stimulation experiments and EMGs shows that cranial elevation and opercular rotation supply input force for the protrusion mechanism. Stimulation of the levator operculi (LOP) and epaxialis (EP) muscles elicited partial jaw protrusion, both separately and simultaneously (Table 3). The LOP and EP also show strong motor activity patterns at the beginning of the strike (Fig. 9). These muscles impart forces to the interopercle serving to rotate it anteriorly.

The interopercle is a highly movable element that rotates around its contact with the epiphyal through an arc of nearly 100 degrees (Fig. 8C). Interopercular rotation transfers an antero dorsally directed force to the lower jaw via the interoperculo-mandibular ligament (Fig. 7C, IOP-M 1.). The interoperculo-mandibular ligament is always in tension during jaw protrusion. The direction of the force applied to the dentary via the interoperculo-mandibular ligament becomes largely dorsal as the interopercle rotates through the last part of its arc. This dorsally directed force continues to cause jaw protrusion because the forward movement of the lower jaw is guided by the rotation of the quadrate and maxilla, which also swing through large arcs (Fig.

8D,E). The premaxilla is pulled into protruded position by the lower jaw via ligamentous connections between the premaxillary alveolar process and the dentary coronoid process.

The kinematic data of Figure 8 reveal the relative importance of cranial and opercular input to jaw protrusion in *Epibulus*. Cranial elevation (Fig. 8A) peaks within 35 msec of the start of the strike and is virtually synchronous with the output movements of interopercular rotation (Fig. 8C), quadrate rotation (Fig. 8D), maxillary rotation (Fig. 8E), and premaxillary protrusion distance (Fig. 8F). However, peak opercular rotation (Fig. 8B) occurs about 15 msec before the synchronous movements of the other variables. Because of the synchrony of cranial elevation with jaw protrusion and other output variables, we conclude that cranial elevation is the principal movement driving the *Epibulus* jaw mechanism. Opercular rotation provides some input to the rotation of the interopercle, but probably serves to brace the interopercle and provide tension in the two important ligaments attached to the interopercle. Generation of ligament tension might also be achieved through hypaxial and sternohyoideus activity, which may tighten tissues connecting the hyoid and interopercle.

The mechanism of jaw retraction involves a reverse of the kinematic pattern of bone movements. (Note the bell-shaped appearance of the kinematic profiles in Fig. 8.) Muscle stimulation experiments (Table 3) indicate that jaw retraction is driven primarily by the adductor mandibulae muscles exerting posteriorly and dorsally directed forces upon the maxilla and the lower jaw. The A1 division (Fig. 4) pulls posteriorly upon the maxillary head, providing a posterior rotation in that element which serves as the input movement for jaw retraction. The A1 and the combined A2/A3 divisions of the adductor mandibulae muscle insert on the lower jaw (Fig. 4) and transmit postero dorsal forces to the mandible which contribute to jaw retraction.

The mechanism of jaw protrusion described above for *Epibulus* is unique in several respects. Extreme anterior protrusion of the lower jaw is a kinematic pattern never before documented in fishes. The role of the lower jaw in previous mechanical hypotheses of fish feeding is that of ventral rotation around a fixed fulcrum provided by the quadrate (e.g., Lauder, '82, '85; Liem, '80; Motta, '84). In these previous hypotheses, lower jaw depression is caused by a posteriorly directed force exerted on the lower jaw by the interoperculo-mandibular ligament or via a linkage involving the hyoid (Aerts et al., '87; Lauder, '79). Primitively for vertebrates, the mandibulo-

hyoid linkage functions in lower jaw depression, which is brought about when the sternohyoideus muscle retracts the hyoid. This posteriorly directed force is passed to the lower jaw through ligamentous connections between the epihyal and lower jaw.

Lower jaw movement in *Epibulus* is caused by anteriorly and dorsally directed forces exerted through the vomero-interopercular ligament to the interopercle and then to the lower jaw via interopercular rotation. These force vectors pull the lower jaw into protruded position, guided by the extreme rotation of the interopercle and the movable maxilla and quadrate (Fig. 7). The primitive hyoid-interopercle-lower jaw linkage is retained in *Epibulus*, but because of the unique nature of the rotating interopercle, retraction of the ceratohyals does not cause any motion of the lower jaws (Table 3, sternohyoideus). Thus the structural linkage for a primitive mechanism of jaw depression is retained in *Epibulus* but no longer functions in jaw movement. A reasonable hypothesis is that this linkage functions in orobranchial cavity expansion in *Epibulus*. Two other linkages that primitively produce jaw depression in teleosts, cranial elevation and opercu-

lar rotation (Lauder, '85), apparently play central roles in the *Epibulus* feeding mechanism as sources of input motion (Fig. 7).

#### Ancestral and derived features of *Epibulus*

Description of the mechanism of jaw protrusion in *Epibulus* reveals a mosaic of primitive and derived features that contribute to jaw function during feeding. Table 4 presents a list of structural features and a description of their activity patterns and their presumed functions with indication as to the phylogenetic polarity of each character as primitive or derived relative to *Cheilinus* and other labrids. The discussion of these features will focus on identifying the particular combination of primitive and derived conditions that has produced the unusual mechanism of jaw protrusion in *Epibulus*.

#### Anatomy

The general pattern of structural modification in the *Epibulus* feeding mechanism is extensive change in bone and ligament structure with comparatively little modification of the cranial muscles. Table 4 illustrates the distribution of derived osteological features in *Epibulus* including

TABLE 4. Primitive and derived characters of *Epibulus* morphologic structure, kinematic or electromyographic pattern, and function in jaw protrusion<sup>1</sup>

Structure	P/D	Kinematic or EMG pattern	P/D	Function	P/D
<b>Bones</b>					
Neurocranium	—	Elevation	P	Force to V-Iop lig.	D
Ethmoid/frontal	D	—	—	Guide premaxilla	P
Other elements	P	—	—	—	—
Premaxilla	D	Extreme protrusion	D	Protrusion	P
Maxilla	P	Extreme rotation	D	Prey capture	P
Mandible	D	Extreme protrusion	D	Brace premax., etc.	P
Quadrate	D	Extreme rotation	D	Protrude LJ	D
Opercle	P	Rotation	P	Protrusion	D
Interopercle	D	Extreme rotation	D	Prey capture	P
Palatine	P	—	—	Protrude LJ	D
Hyoid app.	P	—	—	Anterior force to LJ	D
<b>Ligaments</b>					
V-Iop lig.	D	—	—	Depress mouth floor	P
Iop-M lig.	D	Rotation	D	Anterior force to IOP	D
Pmx-Mx lig.	D	—	—	Anterior force to LJ	D
Operc-Iop lig.	D	—	—	Connect Pm to Mx	P
Hyoid-Iop lig.	D	—	—	Rotation point	D
<b>Muscles</b>					
Levator operculi	P	Early contraction	P	Opercle rotation	P
Epaxialis	P	10–20 ms after LOP	P	Cranial elevation	P
Sternohyoideus	P	15–20 ms after LOP	P	Hyoid depression	P
Geniohyoideus	D	—	—	Hyoid protraction	P
Adductor mandibulae	P	20–25 ms after LOP	P	Jaw retraction	P

<sup>1</sup>P = primitive, D = derived, relative to closely related and other labrid species or perciform fishes. Ligaments: V-Iop lig. = vomero-interopercular; Iop-M lig. = interoperculo-mandibular; Pmx-Mx lig. = premaxilla-maxillary; Operc-Iop lig. = operculo-interopercular; Hyoid-Iop lig. = hyoid-interopercular.

the premaxilla, lower jaw, quadrate, interopercle, and aspects of the neurocranium. It should be noted that derived conditions range in magnitude from extreme modification in structure (such as the quadrate) to modification in size (premaxilla) or general shape and orientation (interopercle). Most other cranial bones are similar in size and shape to the condition found in *Cheilinus* and many other perciform fishes.

Derived conditions found in connective tissues (Table 4) range from a completely unique ligament that plays a major role in the function of protrusion (i.e., the vomero-interopercular ligament) to reduced elements (i.e., between opercle and interopercle) or enlarged ligaments (i.e., premaxilla-maxillary). Many ligaments of the head are not listed in Table 4 and these appear to retain a primitive condition.

The muscles examined in *Epibulus* (Table 4) retain the origin, insertion, and general shape found in most labrids and in the perciform muscles described by Winterbottom ('74). An exception to this is the geniohyoideus muscle (Fig. 5), which is extremely elongate in *Epibulus*. Other muscles that retain a primitive condition (such as the adductor and levator arcus palatini muscles) are diagrammed in Figure 4 but are not included in Table 4 because their function was not examined.

#### Kinematic pattern

Several key elements of the feeding kinematics in *Epibulus* are conserved from the primitive condition widely present in perciform fishes. Previous work (Alexander, '66; Lauder, '79, '81b; Lauder and Liem, '81; Liem, '80; Osse, '69) reveals a biomechanical pattern typical of teleost feeding that consists of nearly simultaneous cranial elevation, gape, and jaw protrusion followed by hyoid depression. Peak opercular rotation has been shown to precede the simultaneous maxima of the above variables in several taxa (Liem, '70; Aerts et al., '87). The kinematic pattern described above is the same pattern found in *Epibulus*. Figure 8 illustrates the synchrony of peak cranial elevation, protrusion distance, and gape, preceded by opercular rotation and followed by hyoid depression. The magnitude of these variables (degrees of rotation or measured distance), except for protrusion distance, is also similar to literature values for other fish species of comparable size.

The notable differences in kinematic pattern between *Epibulus* and other teleosts are more numerous than the similarities (see Table 4). Interopercular and quadrate rotation (Fig 8C,D) for *Epibulus* cannot be compared to literature values because these movements do not occur in

other fishes. In fact, mechanical models of fish jaw movements published by Barel et al. ('77) and Aerts et al. ('87) suggest that only a small posterior translation of the interopercle can occur and allow for no independent movement of the quadrate relative to other jaw elements. Maxillary rotation in *Epibulus* (Fig. 8E) peaks at over 100 degrees. Lauder ('81b) demonstrated a 40–60-degree maxillary rotation for *Lebiasina boruca*, and maxillary rotation in three species of *Cheilinus* exhibits a peak value of approximately 20 degrees (Westneat, unpublished data). The high level of maxillary rotation is thus a derived activity pattern in *Epibulus*. These results demonstrate again that the timing and pattern of input movements (cranial elevation and opercular rotation) are similar to those described for other teleosts, but the elements that transfer this movement to the highly protrusive jaws exhibit derived kinematic patterns (Table 4).

#### Muscular motor pattern

No significant differences were found among *Epibulus*, *Lepomis*, and *Micropterus* in the average activity pattern of the four muscles, but the relative onset time of the adductor mandibulae (Fig. 9, AM) did approach significance (Table 2). Although we have not compared the *Epibulus* motor pattern quantitatively to that of other cheiline labrids, it conforms qualitatively to published accounts of *Cheilinus unifasciatus*, other labrids (Sanderson, '88), and other labroid taxa feeding on immobile prey (Liem, '78, '80). Thus, with the possible exception of the nearly significant relative onset of the adductor mandibulae, the evolution of the *Epibulus* jaw mechanism does not appear to have involved changes in the timing of activity of the primary feeding muscles (Table 4).

It is important to note that our motor pattern analysis is restricted to the seven duration and relative onset variables that were measured. Other levels of EMG signal analysis (e.g., signal amplitude, intra-burst heterogeneity, spike frequency, spike number times amplitude) were not performed so that our findings of a conserved motor pattern in *Epibulus* only relate to the timing variables that were measured. A more detailed analysis of muscle activity would be needed to fully determine if all aspects of the *Epibulus* motor pattern remain unchanged from the primitive labrid condition. We emphasize, however, that our statistical analysis was unable to falsify the null hypothesis that the average motor patterns were the same in the three species.

One factor that made finding significant differences among species less likely was the high levels of variation that were found among individuals within species (Table 2). High within-group variance decreases the likelihood that a statistical test will find significant differences among group means. High intraspecific variation in EMG variables has been found widely in aquatic feeding vertebrates (Shaffer and Lauder, '85; Bemis and Lauder, '86; Wainwright and Lauder, '86; Sanderson, '88; Wainwright, '89), indicating that there can be considerable plasticity between feedings in some aspects of the motor pattern. One practical consequence of high intraspecific variation is that it is important to base comparisons among species on adequate estimates of the within-species variance component. Thus, experimental designs should permit the partitioning of variance among species and among individuals within species, as exemplified by the nested analysis of variance design used in this study.

The lack of evidence of evolutionary change in the motor pattern of *Epibulus* agrees with several recent studies (Lauder and Shaffer, '88; Shaffer and Lauder, '85; Wainwright and Lauder, '86; Sanderson, '88; Wainwright, '89) that compared muscle activity patterns among closely related aquatic species during feeding. In each case fewer than 10% of the EMG variables that were compared were found to be significantly different among taxa. *Epibulus* presents a particularly striking example of drastic evolutionary changes in the morphology of the feeding mechanism, in contrast to an apparent lack of change in the muscular motor pattern of feeding.

#### *Functional roles and character states*

Consideration of the functional role (the product of a structure's activity pattern and its interaction with other structures in a mechanism) of each element within the jaw mechanism of *Epibulus insidiator* carries the analysis of primitive and derived character states to an additional level. Table 4 demonstrates the importance of considering each element of the jaw mechanism at the three levels of structure, activity, and functional role, because the distribution of primitive and derived conditions among these levels is rarely the same for any two elements. For example, the premaxilla exhibits a structure (Fig. 2A) and activity pattern (Fig. 8F) that is derived relative to *Cheilinus* and other labrids, yet its functional role (that of protrusion and prey capture) is primitive within teleosts. The maxilla, however, shows no derived structural condition (Fig. 2A) and yet its activity pattern (Fig. 8E, extreme rotation) is derived compared

to *Cheilinus* and other teleosts. Of the two functional roles described for the maxilla, bracing and guiding the premaxilla is a primitive one but guiding protrusion of the lower jaw is uniquely derived (Table 4). There are elements of the mechanism that exhibit derived conditions at all levels (the quadrate and interopercle) and some that are ancestral at each level (the opercle and most muscles).

Whole organisms are complex combinations of ancestral and derived features, a fact that forms the basis of currently accepted phylogenetic reconstruction techniques. We have used a phylogenetic framework of *Epibulus*, its related genera, and labrid outgroups to show that a functional complex within an organism is also likely to be comprised of both primitive and derived features. Furthermore, we demonstrate that an individual element within a functional unit may be both primitive and derived when that element is studied at successive levels of structural and functional analysis.

There is some argument over whether functional characters play a role in phylogenetic analysis (see Bock, '81), but functional characters have been used in constructing phylogenies and have played a prominent role in evolutionary interpretations of structural changes within lineages (Liem and Greenwood, '81; Schaefer and Lauder, '86). We suggest that experiments in functional morphology be designed, if possible, to include consideration of structural and functional features of organisms at the multiple levels discussed above and that they be performed within a phylogenetic framework that allows interpretation of character state evolution at each of these levels.

#### *Attack strategy using extreme protrusion*

The feeding biomechanics of many fish taxa have been examined, several of which protrude the upper jaw to a high degree. Table 5 compares the strike characteristics of *Epibulus* to those of several other protrusive fishes for which data are available (Alexander, '67; Liem, '70; Nyberg, '71; Lauder and Liem, '81). *Epibulus* protrudes its jaws to 65% of head length, which is twice the extent of protrusion measured in other fishes. *Stylephorus chordatus* (not shown in Table 5) exhibits a derived jaw mechanism that is very different from *Epibulus*, in which the premaxilla is protruded extremely far ventrally and the neurocranium shows extreme elevation (Pietsch, '78).

A frequently cited hypothesis for the function of jaw protrusion is that of increase in velocity of attack (Lauder and Liem, '81; Motta, '84), and the capture of elusive prey occurs in each of the

TABLE 5. Comparison of *Epibulus* prey capture data with data from previous studies<sup>1</sup>

Species	Premaxillary protrusion	Prey distance	Body velocity	cm/sec		Reference
				Premaxillary protrusion velocity	Total strike velocity	
<i>Epibulus insidiator</i>	65	80	5	230	235	This paper
<i>Luciocephalus pulcher</i>	33	90	130	51	181	Lauder and Liem ('81)
<i>Cheilinus chlorurus</i>	14	31	94	20	114	Westneat, unpublished
<i>Micropterus salmoides</i>	7	50	78	39	39	Nyberg ('71)
<i>Monocirrhus polyacanthus</i>	33					Liem ('70)
<i>Lepomis macrochirus</i>	17					Lauder and Liem ('81)
<i>Pterophyllum</i> sp.	23					Alexander ('67)

<sup>1</sup> Values for premaxillary protrusion and prey distance are given in percentage head length.

species listed in Table 5. Excepting *Epibulus*, the fishes listed in Table 5 primarily use body velocity during the strike, a strategy widespread among predatory teleosts. In *Epibulus*, an alternative strategy for attaining high strike velocity and capturing elusive prey has apparently evolved. The total strike velocity of *Epibulus* (Table 5) is high although time to maximum protrusion and gape is about 35 msec, a value comparable to the strike of other fishes. This velocity is achieved through jaw protrusion velocity only, the body velocity being near zero in every *Epibulus* feeding recorded (Table 5). This difference in the source of strike velocity may be reflected in general feeding behavior. *Epibulus* forages slowly, inspecting heterogeneous coral reef substrates and striking rapidly at small fishes and crustaceans. *Luciocephalus* and *Micropterus* are typical sit-and-wait predators which exhibit rapid acceleration of the body and combine suction and ram feeding strategies.

#### GENERAL CONCLUSIONS

The protrusion mechanism of *Epibulus insidiator* is an example of major evolutionary change in a vertebrate functional system. The central conclusion of this study is that the extreme protrusion ability of this fish is due to a multitude of structural and functional novelties occurring primarily in the bones and ligaments of the head. Our data reveal no definitive evidence of evolution in the patterns of muscle activity during feeding. The unusual function of extreme jaw protrusion is performed by using the forces exerted by a previously existing muscle architecture and pattern of activity, but this primitive pattern is transformed in terms of output behavior by a drastic reorganization of the linkage system of the jaws.

The implications of the reorganization of the jaw mechanism in *Epibulus* are thought-provoking when one considers the extent to which *Epibulus* differs from all other described fishes,

including its probable sister group (one or a combination of species within tribe Cheilini; Gomon, pers. comm.; Westneat, unpublished data). The evolution of the *Epibulus* jaw mechanism involves the derivation of a unique ligament (Fig. 2A, the vomero-interopercular ligament) and the change of structure, activity, and role of the quadrate, the interopercle, the lower jaw, and the interoperculo-mandibular ligament (Table 4). Further derived characteristics have evolved in the premaxilla, maxilla, neurocranium, and additional ligaments, each of which appear to play critical roles during *Epibulus* jaw protrusion.

Major modifications within a lineage of organisms, such as those described in *Epibulus*, often challenge evolutionary theory for an explanation of their occurrence. One source of puzzlement attending the identification of such phenotypes is a lack of intermediate or transitional forms from which to infer the evolutionary changes leading to the observed feature. Examples of functional innovations in feeding structures for which evolutionary explanations have been elusive are fairly common. These include the specialized jaw hinge of bolyerine snakes (Frazzetta, '66, '70) and the transition of the masticatory apparatus from a reptilian to a mammalian level of organization (Bramble, '78; Crompton and Parker, '78). The uniquely derived tongue projection behavior of chameleons, long considered an evolutionary anomaly, has recently been shown to be a specialization of a basic agamid lizard trophic mechanism (Schwenk and Bell, '86; Smith, '88).

There are no described perciform or labrid fish species with a feeding mechanism that might be considered intermediate between that of *Epibulus* and other labrids. Fossil remains of labrids are scarce, although Patterson ('64) places the radiation of labrids in the Eocene, 53–37 million years ago. Apparently the unusual *Epibulus* trophic apparatus will be relegated to the

status of "puzzling evolutionary phenomenon" for the present. Continued focus on the functional morphology and phylogenetic interrelationships of labroid fishes and other perciform clades is critical to our understanding of evolution in these highly diverse vertebrate groups.

## ACKNOWLEDGMENTS

The authors would like to express their friendship with the following people for indispensable help in this project: J. Resing, C. Matthews, H. Choat, G. Russ, K. Clements, and others at James Cook University, Australia, and the Lizard Island Research Station; R. Withrow and the Waikiki Aquarium; S. Wainwright, J. Lundberg, K. Smith, and the Zoology Dept. of Duke University; G. Lauder, S. Reilly, C. Sanford, and B. Jayne of the University of California, Irvine. S. Reilly, G. Lauder, J. Lundberg, S. Wainwright, and two anonymous reviewers offered critical comments on the manuscript. Discussions with M. Gomon helped clarify the phylogenetic position of *Epibulus*. This project was funded by a Duke University travel grant, a research assistantship under S. Wainwright, and a federal student loan to M. Westneat. Equipment and facilities were provided by K. Smith, B. Nicklas, and by NSF grants BSR 85-20305 and DCB 87-10210 to G. Lauder.

## LITERATURE CITED

- Aerts, P., J.W.M. Osse, and W. Verraes (1987) Model of jaw depression during feeding in *Astotilapia elegans* (Teleostei: Cichlidae): Mechanisms for energy storage and triggering. *J. Morphol.* 194:85-109.
- Alexander, R.McN. (1966) The functions and mechanisms of the protrusible upper jaws of two species of cyprinid fish. *J. Zool. (Lond.)* 149:288-296.
- Alexander, R.McN. (1967) The functions and mechanisms of the protrusible upper jaws of some acanthopterygian fish. *J. Zool. (Lond.)* 151:43-64.
- Barel, C.D.N. (1983) Towards a constructional morphology of cichlid fishes (Teleostei, Perciformes). *Neth. J. Zool.* 33:357-424.
- Barel, C.D.N., J.W. Van Der Muelen, and H. Berkhoudt (1977) Kinematischer Transmissionskoeffizient und Vierstangensystem als Funktionsparameter und Formmodell für Mandibulare Depressionsapparate bei Teleostiern. *Anat. Anz.* 142:21-31.
- Bemis, W.E., and G.V. Lauder (1986) Morphology and function of the feeding apparatus of the lungfish, *Lepidosiren paradoxa* (Dipnoi). *J. Morphol.* 187:81-108.
- Bleeker, P. (1862) *Conspectus generum Labroideorum analytici*. Verslag. Akad. Amst. 14:94-109.
- Bock, W.J. (1981) Introduction to the symposium: Functional-adaptive analysis in systematics. *Am. Zool.* 21:3-20.
- Bramble, D.M. (1978) Origin of the mammalian feeding complex: Models and mechanisms. *Paleobiology* 4:271-301.
- Crompton, A.W., and P. Parker (1978) Evolution of the mammalian masticatory apparatus. *Am. Sci.* 66:192-201.
- Darwin, C. (1859) *On the Origin of Species*. Cambridge, Massachusetts: Harvard University Press (1964).
- Delsman, H.C. (1925) Fishes with protrusile mouths. *Treubia* 6:98-106.
- Dingerkus, G., and L.D. Uhler (1977) Enzyme clearing of alcian blue stained whole vertebrates for demonstration of cartilage. *Stain Technol.* 52:229-232.
- Frazzetta, T.H. (1966) Studies on the morphology and function of the skull in the Boidae (Serpentes). II. Morphology and function of the jaw apparatus in *Python sebae* and *Python molorus*. *J. Morphol.* 118:217-296.
- Frazzetta, T.H. (1970) From hopeful monsters to bolyerine snakes? *Am. Nat.* 104:55-72.
- Goldschmidt, R. (1940) *The Material Basis of Evolution*. New Haven, Connecticut: Yale Univ. Press.
- Gomon, M.F. (1979) A Revision of the Genus *Bodianus* (Family Labridae) With a Phylogenetic Hypothesis of Related Genera in Tribe Hypsigynini. Ph.D. Dissertation, University of Miami, Florida.
- Lauder, G.V. (1979) Feeding mechanics in primitive teleosts and the halecomorph fish *Amia Calva*. *J. Zool. (Lond.)* 187:543-578.
- Lauder, G.V. (1981a) Form and function: Structural analysis in evolutionary morphology. *Paleobiology* 7:430-442.
- Lauder, G.V. (1981b) Intraspecific functional repertoires in the feeding mechanism of the characoid fishes *Lebiasina*, *Hoplias* and *Chalceus*. *Copeia* 1981:154-168.
- Lauder, G.V. (1982) Patterns of evolution in the feeding mechanism of acanthopterygian fishes. *Am. Zool.* 22:275-285.
- Lauder, G.V. (1983) Functional and morphological bases of trophic specialization in sunfishes (Teleostei, Centrarchidae). *J. Morphol.* 178:1-21.
- Lauder, G.V. (1985) Aquatic feeding in lower vertebrates. In M. Hildebrand, D.M. Bramble, K.F. Liem, and D.B. Wake (eds): *Functional Vertebrate Morphology*. Cambridge: Belknap Press, pp. 210-229.
- Lauder, G.V., and K.F. Liem (1981) Prey capture by *Luciocephalus pulcher*: Implications for models of jaw protrusion in teleost fishes. *Environ. Biol. Fish.* 6:257-268.
- Lauder, G.V., and H.B. Shaffer (1988) Ontogeny of functional design in tiger salamanders (*Ambystoma tigrinum*): Are motor patterns conserved during major morphological transformations? *J. Morphol.* 197:249-268.
- Liem, K.F. (1970) Comparative functional anatomy of the Nandidae (Pisces: Teleostei). *Fieldiana Zool.* 56:1-166.
- Liem, K.F. (1978) Modulatory multiplicity in the functional repertoire of the feeding mechanism in cichlids. I. Piscivores. *J. Morphol.* 158:323-360.
- Liem, K.F. (1980) Adaptive significance of intra- and interspecific differences in the feeding repertoires of cichlid fishes. *Am. Zool.* 20:295-314.
- Liem, K.F., and P.H. Greenwood (1981) A functional approach to the phylogeny of the pharyngognath teleosts. *Am. Zool.* 21:83-101.
- Lombard, R.E., and D.B. Wake (1986) Tongue evolution in the lungless salamanders, family Plethodontidae. IV. Phylogeny of the plethodontid salamanders and the evolution of feeding dynamics. *Syst. Zool.* 35:532-551.
- Motta, P.J. (1984) Mechanics and functions of jaw protrusion in teleost fishes: A review. *Copeia* 1984:1-18.
- Nyberg, D.W. (1971) Prey capture in largemouth bass. *Am. Midl. Nat.* 86:128-144.
- Osse, J.W.M. (1969) Functional morphology of the head of the perch (*Perca fluviatilis*): An electromyographic study. *Neth. J. Zool.* 10:289-392.
- Patterson, C. (1964) A review of Mesozoic actinopterygian fishes with special reference to those of the English chalk. *Philos. Trans. R. Soc. Lond. [Biol.]* 247:213-482.
- Pietsch, T.W. (1978) The feeding mechanism of *Stylephorus chordatus* (Teleostei: Lampridiformes): Functional and ecological implications. *Copeia* 1978:255-262.
- Randall, J.R. (1983) *Red Sea Reef Fishes*. London: IMMEL Publishing.
- Renard, L. (1719) Poissons, Ecrivisses et Crabes, de Diverses Couleurs et Figures Extraordinaire, que l'on Trouve Au-

- tour des Isles Moloques, et sur les Cotes des Terres Australes... Histoire des Plus Rares Curiositez de la Mer des Indes. 2 vols. in 1. Amsterdam: Louis Renard.
- Rognes, K. (1973) Head skeleton and jaw mechanism in Labrinae (Teleostei: Labridae) from Norwegian waters. *Univ. Bergen Arb. Mat. Naturv. (Ser. 1971)*4:1-149.
- Sanderson, S.L. (1988) Variation in neuromuscular activity during prey capture by trophic specialists and generalists (Pisces: Labridae). *Brain Behav. Evol.* 32:257-268.
- Schaefer, S.A., and G.V. Lauder (1986) Historical transformation of functional design: Evolutionary morphology of feeding mechanisms in loricarioid catfishes. *Syst. Zool.* 35:489-509.
- Schwenk, K., and D.A. Bell (1986) Chameleon-like tongue protrusion in an agamid lizard. *Am. Zool.* 26:65A.
- Shaffer, H.B., and G.V. Lauder (1985) Aquatic prey capture in ambystomatid salamanders: Patterns of variation in muscle activity. *J. Morphol.* 183:273-284.
- Simpson, G.G. (1953) *The Major Features of Evolution*. New York: Columbia University Press.
- Smith, K.K. (1988) Form and function of the tongue in agamid lizards with comments on its phylogenetic significance. *J. Morphol.* 196:157-171.
- Wainwright, P.C. (1989) Prey processing in haemulid fishes: Patterns of variation in pharyngeal jaw muscle activity. *J. Exp. Biol.* 141:359-376.
- Wainwright, P.C., and G.V. Lauder (1986) Feeding biology of sunfishes: Patterns of variation in the feeding mechanism. *J. Linn. Soc. (Zool.)* 88:217-228.
- Wake, D.B., and A. Larson (1987) Multidimensional analysis of an evolving lineage. *Science* 233:42-48.
- Wilkinson, L. (1986) *SYSTAT: The System for Statistics*. Evanston, Illinois: Systat Inc.
- Winterbottom, R. (1974) A descriptive synonymy of the striated muscles of the Teleostei. *Proc. Acad. Nat. Sci. Phila.* 125:225-317.