

Motor Basis of Suction Feeding Performance in Largemouth Bass, *Micropterus salmoides*

JUSTIN R. GRUBICH* AND PETER C. WAINWRIGHT

Department of Biological Science, Florida State University, Tallahassee, Florida 32306-3050

ABSTRACT We examined the relationship between cranial muscle activity and buccal pressure during suction feeding by the largemouth bass, *Micropterus salmoides*. Buccal pressure was recorded simultaneously with electromyograms (EMG) from four muscles that function prominently during the strike, including three expansive phase muscles (levator arcus palatini, epaxialis, and sternohyoideus) and the major compressive phase muscle, the adductor mandibulae. Feeding behavior was analyzed in 145 strike sequences from five individuals. EMG from each muscle was quantified with four variables (integrated area of rectified EMG, burst duration, intensity of activity, and onset time of activity), and pressure pulse was characterized with seven variables (area of subambient pressure curve, area of superambient pressure curve, minimum pressure, maximum pressure, pressure pulse duration, time to minimum pressure, and time to maximum pressure). Correlation, multiple regression, and a principal components analysis (PCA) were used to investigate the relationship between muscle activity and buccal pressure patterns. About 50% of the correlations among muscle variables were significant, while fewer than 25% of the correlations between muscle activity and buccal pressure variables were significant. Multiple regression models accounted for about 50% of the variance in each pressure variable, although substantial differences were found among individual fish in the success of these models. A PCA performed on the correlation matrix of EMG variables yielded a first principal component that accounted for 33% of the overall variance in strikes and was significantly correlated with the timing variables of buccal pressure. A general trend was apparent in which *Micropterus* modulated the magnitude of suction pressure and the timing of the pressure pulse during the strike by altering the extent of activity (i.e., integrated area and burst duration) in cranial muscles. This study shows that suction performance is moderately influenced by cranial muscle activity. However, extensive strike-to-strike and inter-individual variation suggest that the relationship between muscle activity patterns and buccal pressure is not precise, and bass are able to use a variety of motor strategies to generate strikes with similar suction pressures. *J. Exp. Zool.* 277:1-13, 1997. © 1997 Wiley-Liss, Inc.

Prey capture has been studied with combinations of kinematic, electromyographic, and other analyses in numerous vertebrate taxa (Lauder, '85; Bramble and Wake, '85; Nishikawa et al., '92; Schwenk and Throckmorton, '89). Fish feeding mechanisms are particularly well known, with published data available for over 50 taxa (Liem, '79; Lauder, '80, '85; Wainwright and Lauder, '86; Sanderson, '88; Westneat '91; Norton, '91). One prominent theme of this literature owes its origins to the observations of Liem ('78) who noted that several cichlid species were able to alter kinematic and motor patterns when feeding on different prey. Subsequently, several additional fish species have been shown to modulate feeding behavior in response to prey type, satiation, or subtle variation in the position of the prey in the water column (Lauder, '81; Wainwright and Lauder, '86; Norton, '91; Sanderson, '88; Wainwright, '89;

Wainwright and Turingan, '93; Sibbing et al., '86). The ability to alter functional properties of the strike in response to environmental stimuli appears to be a general property of teleost feeding mechanisms. However, the relationship between patterns of muscle contraction and kinematic or pressure patterns that are produced by the sequence of muscle contraction are not well understood. What motor strategies are used by fishes to modulate key aspects of feeding performance, such as suction pressure, or the speed of the strike?

The present study investigates the relationship between cranial muscle activity and pressure within the buccal cavity during suction feeding in

Contract Grant sponsor: NSF; Contract Grant number IBN-9306672.
*Correspondence to: Justin R. Grubich, Department of Biological Science, Florida State University, Tallahassee, FL 32306-3050.
Received 18 April 1996; Revision accepted 3 September 1996

largemouth bass (*Micropterus salmoides*). Suction feeding involves rapid expansion of the buccal cavity by simultaneous depression of the lower jaw and hyoid apparatus, elevation of the neurocranium, and lateral expansion of the suspensorium to create a sharp drop in buccal pressure that causes water to rush into the mouth, carrying the prey (Lauder, '85). Expansion is followed by immediate jaw adduction and buccal cavity compression. We measure standard EMG variables: burst duration, integration area of burst, intensity, and onset of muscle activity in four key muscles involved in the suction feeding mechanism, and we attempt to elucidate their effects on pressure changes within the buccal cavity during the strike.

MATERIALS AND METHODS

Research specimens

The largemouth bass, *Micropterus salmoides*, is the largest member of the endemic North American freshwater fish family Centrarchidae. We chose this species because its feeding mechanism has been well studied (Nyberg, '71; Lauder, '83) and because it is known to modulate motor activity and buccal pressure when feeding in different conditions and on different prey (Wainwright and Lauder, '86; Norton and Brainerd, '93). During prey capture, largemouth bass employ a range of kinematic strategies that span the "ram-suction continuum" (Norton and Brainerd, '93). All specimens used in this study were collected from Lake Jackson, Leon County, Florida, and transported directly to the laboratory, where they were housed separately in 100 L aquaria at room temperature ($21 \pm 2^\circ\text{C}$). The bass were maintained on a mixture of experimental prey species (ranging in size from 25 to 50 mm; sailfin mollies (*Poecilia latipinna*), mosquitofish (*Gambusia affinis*), bluefin killifish (*Lucania goodei*), least killifish (*Heterandria formosa*), and golden shiners (*Notemigonus crysoleucus*). The study individuals were numbered one through five and had standard lengths of 235, 253, 250, 300, and 245 mm, respectively. We intentionally selected a narrow range of body sizes to avoid the confounding effects of scale on feeding behavior, which have been described in this species (Richard and Wainwright, '95; Wainwright and Richard, '95).

Feeding experiments

Buccal pressure changes and cranial muscle activity were recorded simultaneously during feeding experiments. Pressure was measured with a

Millar SPR-407 microcatheter tipped pressure transducer that was threaded through a plastic canula leading into the buccal cavity. The plastic canula was implanted through the neurocranium along the dorsal midline just anterior to the eyes. The canula was flanged on the distal end, holding it in place on the superior wall of the buccal cavity.

Electromyographic recordings of muscle activity were made through bipolar electrodes constructed from paired 1.5 m sections of 0.051 mm diameter poly-insulated stainless steel wire (California Fine-Wire). The electrode ends were glued together and threaded through a 26 gauge, 0.5 inch hypodermic needle. The insulation was scraped away with a razor blade under a dissecting microscope to expose 0.5 mm tips of wire which were separated by approximately 30° . The tips were then bent back over the shaft of the needle to form hooks which anchored the electrode in the muscle upon percutaneous insertion of the needle into the fish. Four electrodes were implanted, tied to a suture on the dorsum of the fish, and glued into a common cable. During recording sessions, electrodes were connected to the high impedance probes of Grass P511 signal conditioners. Electrical signals were amplified 10,000 times, and a bandpass of 100–1,000 Hz was employed with a 60 Hz notch filter activated. Electrodes and the pressure canula were implanted while the fish were under anesthesia (about 0.7 g L^{-1} tricaine methanesulfonate). During feeding trials, EMG, buccal pressure, and a simultaneous voice track were recorded on a 14-channel TEAC XR-5000 FM recorder. Strike sequences were later played back to produce a hard copy on a Graphtec thermal array recorder for visual inspection of the recorded events.

We studied four bilaterally paired muscles which have been shown to function prominently in the feeding mechanism of *M. salmoides* and other centrarchids (Lauder, '85; Lauder et al., '86; Wainwright and Richard, '95). We recorded from three muscles that are active during the explosive expansive phase, the levator arcus palatini (LAP), the epaxialis (EP), and the sternohyoideus (SH); and the principal compressive phase muscle, section 2 of the adductor mandibulae complex (AM). The LAP, situated posterior to the orbit, connects the suspensorium to the neurocranium and laterally expands the suspensorium at the onset of the strike. EP elevates the head during expansion, and SH acts as the major depressor of the hyoid apparatus, dramatically lowering the floor

of the mouth (a major suction-generating action). The AM connects the suspensorium to the mandible and is the major jaw closing muscle. Electrodes were implanted into the left side members of these bilaterally paired muscles. Although we are unaware of data that demonstrate regional activity variation in muscles of feeding fishes, or other ectothermic vertebrates, we attempted to minimize any potential effects of electrode placement by taking special care to position them in the same portion of the muscle for all individuals. Electrode positions were the central portion of the LAP, the anterior-most mid-dorsal region of the EP, the anterior region of the SH, and the ventral-anterior region of the AM. At the end of the feeding experiments, individuals were euthanized in a solution of tricaine methanesulfonate, and electrode placement was confirmed by dissection of the fresh specimen.

Prior to feeding experiments, individual bass were starved for a period of 2 to 3 days to increase hunger level. Following electrode and pressure transducer implantation, each fish was returned to the aquarium where it recovered from anesthesia. Feeding trials began when the fish responded eagerly to prey. This was as early as 2

hours after implantation and as late as the following day. During each experiment, individuals were presented live fish prey and allowed to range freely to pursue and capture them within the 100 L aquarium. In order to elicit a broad range of strike efforts, bass were fed prey until they reached satiation. We analyzed both successful and unsuccessful prey capture attempts. In total, 145 strike sequences from five individuals were recorded and analyzed.

Data analysis

Analog recorded pressure and electromyographic data were digitized with a Keithley 500A system using a sampling rate of 8 kHz. A minimum of 20 strikes per individual were analyzed. A custom computer program was used to measure four activity variables for each muscle (Table 1): integrated area of the rectified EMG (LAP-area, EP-area, SH-area, and AM-area), duration of the burst (LAP-dur, EP-dur, SH-dur, and AM-dur), intensity of activity (calculated by dividing the integrated area by duration: LAP-int, EP-int, SH-int, and AM-int), and the onset time of activity (EP-on, SH-on, and AM-on). The onset of activity in the LAP was used as a reference time

TABLE 1. Descriptive statistics of EMG and buccal pressure variables measured during suction feeding in largemouth bass ($N = 5$ individuals; total number of strike sequences = 145)¹

Muscle/pressure	Variables	Mean	S.D.	Minimum	Maximum
EMG	LAP rectified integrated area	5.9	4.7	.19	24.8
	LAP duration	104.2	57.3	9.9	307.4
	LAP intensity	.06	.03	.008	.15
	EP-area	1.6	1.9	0	9.7
	EP-dur	46.6	38.4	0	148.5
	EP-int	.02	.02	0	.07
	EP-onset	17.5	29.6	-134.8	112.3
	SH-area	2.3	3.8	0	25.5
	SH-dur	60.4	41.8	0	167.5
	SH-int	.03	.03	0	.16
	SH-on	21.5	31.6	-151.9	112.3
	AM-area	4.2	4.9	0	32.4
	AM-dur	62.4	54.4	0	336.1
	AM-int	.06	.05	0	.24
	AM-on	58.1	46.3	-62.3	208.6
Buccal pressure	NEG-area	117.7	77.5	3.8	441.1
	POS-area	21.2	33.6	0	179
	NEG-peak	-3.4	2.6	-16.4	-44
	POS-peak	.38	.39	-.17	2.3
	PRESS-dur	179.9	91.0	25.6	475.3
	Time to NEG-pk	44.6	31.1	7.9	165.9
	Time to POS-pk	123.6	76.3	0	411.8

¹Units of measurement: onset and duration are in milliseconds (msec); intensity is in millivolts (mV); integrated area of EMG is in (msec × mV); peak pressures are in kilopascal (kPa); area under pressure curves is in (Pa × sec). See text for abbreviations.

to measure the relative onsets of activity in the other muscles. Seven pressure variables were also measured (Table 1): area of the subambient portion of the pressure curve (NEG-area), area under the superambient portion of the pressure curve (POS-area), lowest value of subambient pressure (NEG-peak), highest value of superambient pressure (POS-peak), and duration of the pressure pulse (PRESS-dur). Time to NEG-pk and Time to POS-pk were measured relative to the onset of LAP.

Statistical analysis

Pearson correlation matrices were calculated for the EMG and pressure variables from each fish and for the pooled data set of all individuals. Multiple regression models were constructed for each pressure variable using a priori selected sets of EMG variables as independent variables. To investigate the repeatability of relationships between muscle activity and buccal pressure, regressions were calculated separately for each individual fish.

A principal components analysis was performed on the overall correlation matrix of EMG variables. Subsequently, correlation and analyses of covariance were performed to investigate the as-

sociation of PC1 scores with each pressure variable. Design of the ANCOVAs included PC1 as the covariate and individual bass as a crossed factor. All correlations and multiple regressions were calculated with \log_{10} transformed data using Systat for Windows v. 5 (Wilkinson, 1992).

RESULTS

Bass showed a broad range of behaviors during prey capture attempts which appeared to relate directly to the "ram-suction continuum" identified by Norton and Brainerd ('93). Although we did not analyze the kinematic patterns of these strikes with film, bass often captured prey without simultaneous forward locomotion, suggesting the absence of a substantial "ram feeding" component in these strikes. In other sequences, prey were captured at the end of a rapid lunge by the bass, suggesting the use of "ram feeding."

Pressure pulse in a typical strike sequence began with a rapid decrease in buccal pressure (Fig. 1). This portion of the pressure sequence was followed immediately by a return to ambient pressure and an occasional pulse of superambient pressure (Fig. 1A). NEG-area and NEG-peak dem-

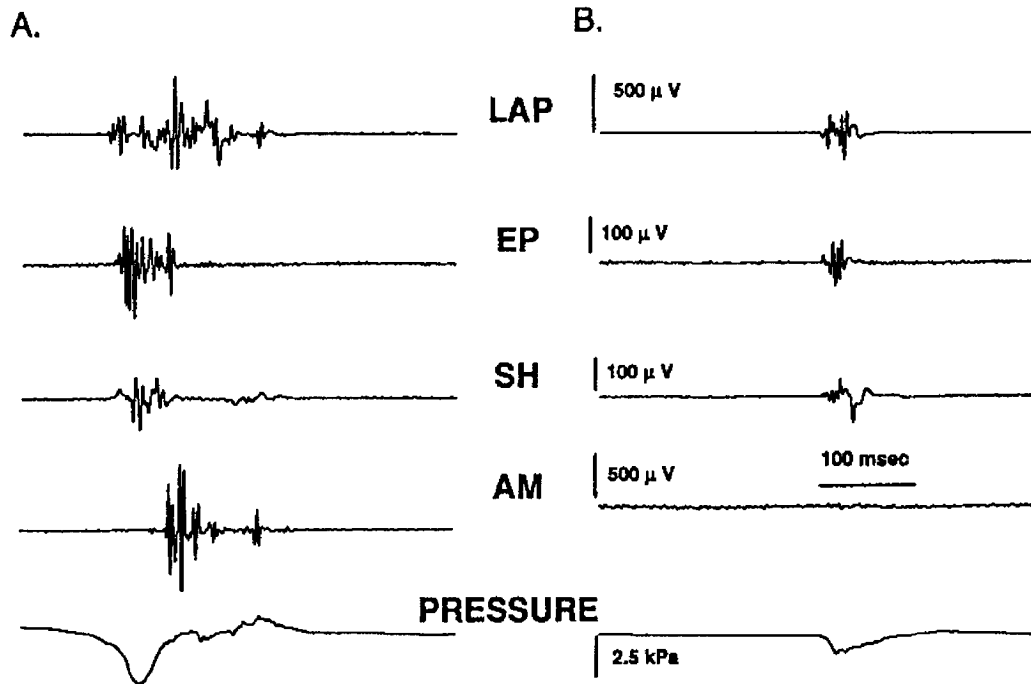


Fig. 1. Simultaneous EMG and buccal pressure recordings of two prey capture sequences from the same fish (Individual 3) a few minutes apart. Note the greater magnitude

in the sub- and superambient pressure in A is associated with higher levels of muscle activity. (See text for abbreviations.)

onstrated dramatically larger mean values and ranges than POS-area and POS-peak (mean NEG-area = 117.7 Pa × sec, mean POS-area = 21.2 Pa × sec; NEG-peak range = -0.44 to -16.4 kPa, POS-peak range = -0.17 to 2.3 kPa). The mean duration of the pressure pulse was 179.9 ms, but this variable was also characterized by a large range of values from 25.6 to 475.3 ms. Mean Time to POS-pk was almost three times as long as Time to NEG-pk (Table 1).

Muscle activity in a typical strike showed nearly synchronous activity in the three expansive phase muscles (LAP, EP, and SH) followed by activity in the AM (Fig. 1A). The similar onset times of the EP (17.5 ms) and SH (21.5), and strong correlations between them (0.76 Fig. 3B) indicate that these muscles initiated activity almost synchronously and shortly after the LAP. In contrast, the adductor mandibulae began activity an average of 62.4 ms after the LAP. The mean burst durations varied from 46.6 ms in the EP to 104.2 ms in the LAP (Table 1). Mean integrated areas of the LAP and AM were approximately double the areas of the EP and SH. Mean intensities showed a similar trend with LAP-int and AM-int equal to 0.06 mV while EP-int and SH-int were 0.02 and 0.03 mV, respectively. Virtually all EMG and pressure variables were characterized by a large range and standard deviation indicating that the fish actively modulated these features among strikes (Table 1).

Representative strike-to-strike variation is illustrated in Figure 1. A strike with approximately average subambient pressure is shown in Figure

1A. Note extensive activity in the three expansive phase muscles, LAP, EP, and SH. A considerable superambient pulse follows the subambient phase and is accompanied by strong activity in the AM. Figure 1B depicts a second strike recorded a few minutes later from the same individual. The subambient pressure pulse is smaller (NEG-area for Fig. 1A and B = 150 and 32 Pa × sec, respectively) as is the activity in the three expansive phase muscles. Further, lack of activity in the AM is associated with a gradual return to ambient pressure and the absence of a superambient pressure pulse.

The relationship described above between EMG and pressure was not always present. Considerable variation in muscle activity patterns occurred between strikes by the same fish even in cases where pressure profiles were very similar (Fig. 2). In Figure 2, the first strike lacks SH activity but shows high amplitude activity and long duration in the AM. The second strike shows a pressure pulse with about the same NEG-area and NEG-peak (first strike = 87.0 Pa × sec and 2.4 kPa below ambient; second strike = 107.6 Pa × sec and 2.5 kPa below ambient, respectively), but the SH is active and the AM shows markedly lower amplitude and duration than the first strike. Further, duration of the EP is about half that of the first strike.

Correlations calculated between EMG variables for the entire data set of 145 strikes revealed significant correlations in 43 out of 84 combinations. There were significant correlations between burst durations of all three expansive phase muscles

TABLE 2. Correlations between EMG and buccal pressure variables during suction feeding in largemouth bass, *Micropterus salmoides*¹

	NEG-area	POS-area	NEG-peak	POS-peak	PRESS-dur	Time to NEG-pkq	Time to POS-pk
LAP-area	-.09	.27	.04	.26	.41*	.48*	.38*
LAP-dur	-.13	.28	.05	.23	.53*	.56*	.46*
LAP-int	-.04	.16	-.01	.20	.10	.16	.09
EP-area	-.05	.10	.07	.09	.23	.24	.24
EP-dur	-.05	.16	.11	.14	.30	.31*	.32*
EP-int	.00	.10	.09	.11	.13	.21	.20
EP-on	-.16	.07	-.13	.07	.12	.09	.12
SH-area	-.09	.19	.03	.12	.32*	.34*	.33*
SH-dur	-.18	.26	.02	.22	.43*	.41*	.38*
SH-int	-.13	.23	-.04	.17	.30	.31*	.28
SH-on	.00	-.01	.01	-.03	.05	.06	.02
AM-area	-.11	.28	.00	.28	.37*	.31	.32*
AM-dur	-.23	.38*	-.12	.37*	.35*	.26	.33*
AM-int	.01	.26	.07	.30	.31*	.31	.30
AM-on	.04	.23	.09	.24	.33*	.31*	.43*

¹Correlations based on log₁₀ transformed data of all individuals. N = 145. See text for variable abbreviations.

*Bonferroni-corrected probabilities significant at $P < 0.05$ level.

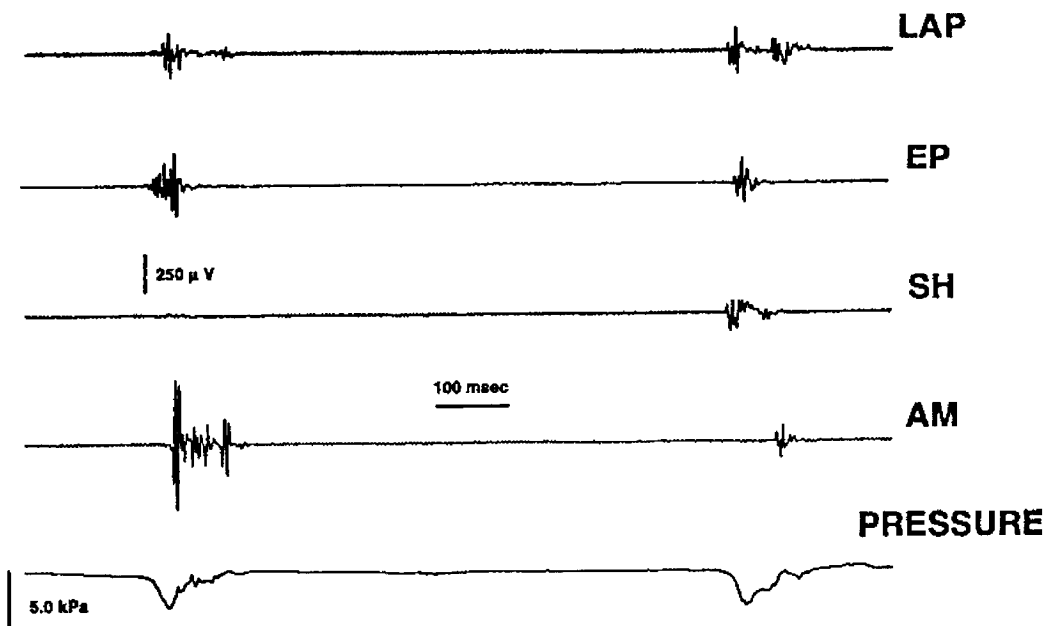


Fig. 2 Two consecutive strikes showing simultaneous EMG and buccal pressure recorded from Individual 3. Note that

the pressure pulses are similar in spite of quite different EMG patterns. (See text for abbreviations.)

(average $r = 0.45$). LAP-area and SH-area had the strongest correlation between integrated area variables ($r = 0.46$; Fig. 3C). Intensity variables did not correlate significantly with each other; however, intensity variables showed some correlations with other variables. For instance, SH-int and AM-int were correlated with LAP-area ($r = 0.42$ and 0.49 , respectively). Onset of activity in the AM was positively correlated with duration of activity in the LAP (Fig. 3A). This correlation indicates that initiation of muscle activity to begin the compressive phase was associated with cessation of activity in the LAP. The strong correlation between EP-on and SH-on (Fig. 3B) indicates that these muscles initiated activity nearly simultaneously, independently of the onset of activity in LAP.

There were fewer significant correlations between EMG and buccal pressure variables (25 out of 105; Table 2). There were no significant correlations between any of the EMG variables and NEG-area or NEG-peak. Only AM-dur was correlated with POS-area and POS-peak. In contrast, the pressure timing variables showed several significant correlations with EMG variables. For example, PRESS-dur was correlated with the expansive phase variables LAP-area, LAP-dur, SH-area, and SH-dur and all the AM

variables. Time to peak pressure variables showed similar trends with most expansive phase muscle variables being correlated with Time to NEG-pk and the AM variables being correlated with Time to POS-pk. Figure 3D, E, and F shows bivariate plots for data from individual bass. Patterns in Figures 3D, E and F illustrate that patterns seen within individual fish resembled those seen in the overall data set (Table 2). However, inter-individual variation in some correlations was extensive. For instance, the correlation between POS-area and AM-dur (Fig. 3D) ranged from 0.24 to 0.80 among the five individuals, and the correlation between LAP-dur and Time to NEG-pk varied from 0.07 to 0.71.

The multiple regression model explained about 60% of the variation in NEG-area across the five individuals (Table 3). However, R^2 values differed among fish, and there was no clear trend in which independent variables were significant in the regression with each fish. The multiple regression model for NEG-peak explained a similar amount of variation, average $R^2 = 55\%$ (Table 4). R^2 values varied among individuals from 19 to 81%. Individuals 1 and 4, which exhibited the highest R^2 values, had similar significant independent variables of LAP activity and AM-on. Three out of the five NEG-peak models showed integrated area

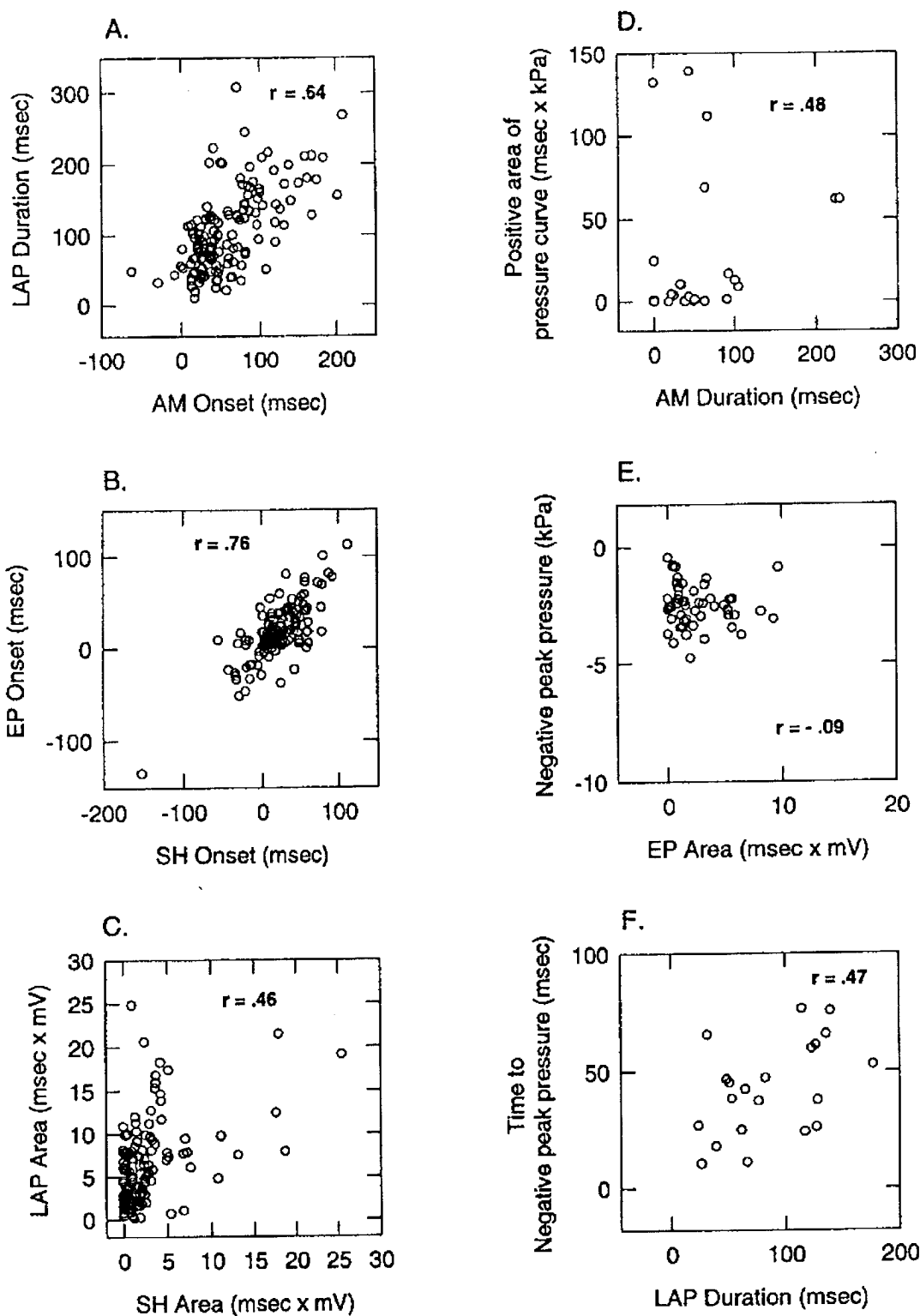


Fig. 3. **A, B, C:** Bivariate plots of correlations between muscle activity variables for the entire data set of 145 strike sequences. **D, E, F:** Bivariate plots of correlations between

muscle activity and buccal pressure variables for individuals 5, 3, and 2, respectively. (See text for abbreviations.)

TABLE 3. Multiple regression analyses with NEG-area and POS-area as the dependent variables¹

Pressure variables	Individual (N)	Multiple R ² (%)	Overall F (d.f.)	Probability	Significant variables*
NEG-area	1 (20)	82	10.05 (6, 13)	<.001	None
	2 (21)	64	4.22 (6, 14)	.012	None
	3 (54)	53	8.94 (6, 47)	<.001	SH-dur
	4 (20)	53	2.49 (6, 13)	.079	None
	5 (30)	47	3.43 (6, 23)	.015	None
POS-area	1 (20)	86	16.58 (5, 14)	<.001	AM-dur, EP-area
	2 (21)	19	0.69 (5, 15)	.641	None
	3 (54)	14	1.50 (5, 48)	.208	AM-dur
	4 (20)	63	4.81 (5, 14)	.009	AM-area
	5 (30)	57	6.42 (5, 24)	.001	LAP-area

¹N = number of strikes analyzed per individual. See text for variable abbreviations. NEG-area model's independent variables are: LAP-area, EP-area, SH-area, LAP-dur, EP-dur, and SH-dur. POS-area model's independent variables are: LAP-area, EP-area, SH-area, AM-area, and AM-dur.

*Independent variables within the model which were significant at the $P \leq .05$ level.

variables of expansive phase muscles to be significant.

Multiple regressions for POS-area and POS-peak revealed average R² values of 48 and 53%, respectively (Tables 3 and 4). Furthermore, with the exception of individual 2, similar R²s were found within the same individual for these two pressure variables. AM activity was significant in most individual models. As with all other dependent variables, inter-individual variation was substantial in the multiple regressions of POS-area and POS-peak.

Regressions on the pressure timing variables revealed R² values similar to those observed for the area and peak pressure models (Table 5). The models for PRESS-dur and Time to POS-pk explained an average of 60 and 51% of the variance, but the model for Time to NEG-pk explained only

39%. For PRESS-dur, EMG duration variables were significant for all individuals, but the specific muscles varied among individuals. EP-on and expansive phase duration variables were significant in two individuals for Time to NEG-pk. The model for Time to POS-pk revealed LAP-dur to be significant in two individuals. The Time to NEG-pk model exhibited a large range of variability between individuals, where the model explained as little as 2% of the variation for individual 4 and 81% for individual 1. In contrast, the Time to POS-pk model determined similar R² values for three out of the five individuals (1, 2, and 5).

A two-way ANOVA was calculated to compare the success of the models in accounting for variation in each of the pressure variables. In this ANOVA individual fish (a random factor) was

TABLE 4. Multiple regression analyses with NEG-peak and POS-peak as the dependent variables¹

Pressure variables	Individual (N)	Multiple R ² (%)	Overall F (d.f.)	Probability	Significant variables*
NEG-peak	1 (20)	81	7.35 (7, 12)	.001	LAP-dur, AM-on
	2 (21)	65	3.48 (7, 13)	.025	EP-area
	3 (54)	19	1.51 (7, 46)	.188	None
	4 (20)	74	4.28 (7, 12)	.009	LAP-area, AM-on
	5 (30)	35	1.67 (7, 22)	.169	SH-area
POS-peak	1 (20)	81	9.38 (6, 13)	<.001	None
	2 (21)	56	2.93 (6, 14)	.046	AM-area, AM-dur, AM-int
	3 (54)	14	1.25 (6, 47)	.301	AM-dur
	4 (20)	66	4.15 (6, 13)	.015	None
	5 (30)	50	3.76 (6, 23)	.009	LAP-area, AM-int

¹N = number of strikes analyzed per individual. See text for variable abbreviations. NEG-peak model's independent variables are: LAP-area, EP-area, SH-area, LAP-dur, EP-dur, and SH-dur, AM-on. POS-peak model's independent variables are: LAP-area, EP-area, SH-area, AM-area, AM-dur, and AM-int.

*Independent variables within the model which are significant the $P \leq .05$ level.

TABLE 5. Multiple regression analyses with Time to NEG-pk, Time to POS-pk and PRESS-dur as the dependent variables¹

Pressure variables	Individual (N)	Multiple R ² (%)	Overall F (d.f.)	Probability	Significant variables*
Time to NEG-pk	1 (20)	81	11.78 (5, 14)	<.001	SH-dur, EP-on
	2 (21)	51	3.11 (5, 15)	.040	None
	3 (54)	20	2.45 (5, 48)	.047	EP-dur, EP-on
	4 (20)	2	.059 (5, 14)	.997	None
	5 (30)	43	3.59 (5, 24)	.015	None
Time to POS-pk	1 (20)	61	3.42 (6, 13)	.030	None
	2 (21)	61	3.71 (6, 14)	.020	None
	3 (54)	38	4.74 (6, 47)	.001	LAP-dur, AM-dur, AM-on
	4 (20)	40	1.41 (6, 13)	.281	None
	5 (30)	56	4.94 (6, 23)	.002	LAP-dur
PRESS-dur	1 (20)	71	4.21 (7, 12)	.014	SH-dur
	2 (21)	77	6.12 (7, 13)	.003	AM-dur, SH-dur, EP-on
	3 (54)	36	3.65 (7, 46)	.003	EP-dur, LAP-dur, EP-on
	4 (20)	52	1.89 (7, 12)	.160	EP-dur
	5 (30)	65	5.94 (7, 22)	.001	AM-dur, LAP-dur

¹N = number of strikes analyzed per individual. See text for variable abbreviations. Time to NEG-pk model's independent variables are: LAP-dur, EP-dur, SH-dur, EP-on, and SH-on. Time to POS-pk model's independent variables are: LAP-dur, EP-dur, SH-dur, AM-dur, AM-int, and AM-on. PRESS-dur model's independent variables are: LAP-dur, EP-dur, SH-dur, AM-dur, EP-on, SH-on, and AM-on.

*Independent variables within the model which are significant at the $P \leq .05$ level.

crossed with the seven dependent pressure variables (also a random factor). Each cell in the analysis contained the R² values for multiple regression run on that individual and pressure variable combination. There was a significant individual effect, indicating that certain fish routinely produced higher R² values than other fish (F-ratio = 8.6; df = 4, 24; $P < 0.001$). The effect of the dependent variable was not significant (F-ratio = 1.0; df = 6, 24; $P > 0.05$), indicating that the regression models performed equally well for all seven pressure variables.

The principal components analysis on the correlation matrix of the entire EMG data set produced four principal components with eigen-values greater than one (Table 6). All EMG variables, with the exception of EP-on and SH-on, were positively correlated with PC1, the major multivariate axis in the data set, and most correlations were above 0.5. This PC contrasts strikes which have EMGs with low integrated areas, short durations, and small intensities with strikes that exhibit larger values of these variables. PC1 explained 33% of the variance in the EMG variable correlation matrix. PC1 was positively correlated with the timing variables of the pressure pulse (PRESS-dur, Time to NEG-pk, and Time to POS-pk > 0.5), but this axis was only weakly correlated with superambient pressure variables (POS-area and POS-peak = 0.35 and 0.32,

respectively) and was not correlated at all with subambient pressure variables (NEG-area and NEG-peak = -0.13 and 0.03, respectively). ANCOVAs were run to determine the effect of individual bass and PC1 on each of the pressure variables. Significant Interaction terms (Individual \times PC1 at $P < .05$) indicated dissimilar slopes among individual fish for the relationships between PC1 and most buccal pressure variables (F ratios = 6.79, 4.37, 2.73, 6.01, and 3.96 for NEG-area, POS-area, PRESS-dur, NEG-peak, and POS-peak, respectively, with df = 4, 135). However, the slopes of the lines describing the relationship of PC1 with Time to NEG-pk and Time to POS-pk did not differ among bass (F ratio of Interaction terms = 1.97 and 1.63, respectively with df = 4, 135) (see also Fig. 4).

DISCUSSION

Largemouth bass have been shown to alter muscle activity and buccal pressure patterns in response to different prey (Wainwright and Lauder, '86; Norton and Brainerd, '93) but the nature of the relationship between muscular modulation and the kinematic or pressure attributes of the strike have not been studied in this species and are poorly known in fishes (Lauder et al., '86). In this study, the electromyographic variables that we used to quantify muscle activity patterns were only moderately success-

TABLE 6. Factor loadings from a Principal Components Analysis performed on the correlation matrix of muscle activity variables¹

Muscle variables	PC 1	PC 2	PC 3	PC 4
LAP-area	.79	.39	.10	-.10
LAP-dur	.72	.34	-.27	.15
LAP-int	.45	.26	.34	-.34
EP-area	.57	-.48	-.60	.14
EP-dur	.65	-.36	-.52	.16
EP-int	.50	-.56	-.47	.20
EP-on	.09	.87	-.21	-.03
SH-area	.71	-.14	.16	-.52
SH-dur	.75	-.24	.25	-.11
SH-int	.70	-.14	.17	-.50
SH-on	.04	.74	-.51	.16
AM-area	.52	.06	.55	.59
AM-dur	.35	.07	.36	.66
AM-int	.62	.02	.46	.24
AM-on	.58	.56	-.31	-.08
Variance explained	33%	18%	15%	11%

¹Table entries are the correlations of each muscle activity variable with the first four principal components. Only principal components with eigen-values greater than one are shown. See text for variable abbreviations.

ful in accounting for the attributes of buccal pressure that we measured, with an average of 52% of the variance in each pressure variable explained by the regression models. A general trend was observed in which strikes with relatively low magnitudes and time-courses of subambient and superambient pressure were associated with shorter, less intense bursts of muscle activity (Fig. 1). However, R^2 values

from the regressions in the range of 50% indicate that considerable variation in EMG patterns was observed in association with any given magnitude of buccal pressure. Indeed, similar suction forces were generated between strikes despite considerable modulation of the motor pattern (Fig. 2). Thus, the relationship between EMG and buccal pressure was not tightly constrained.

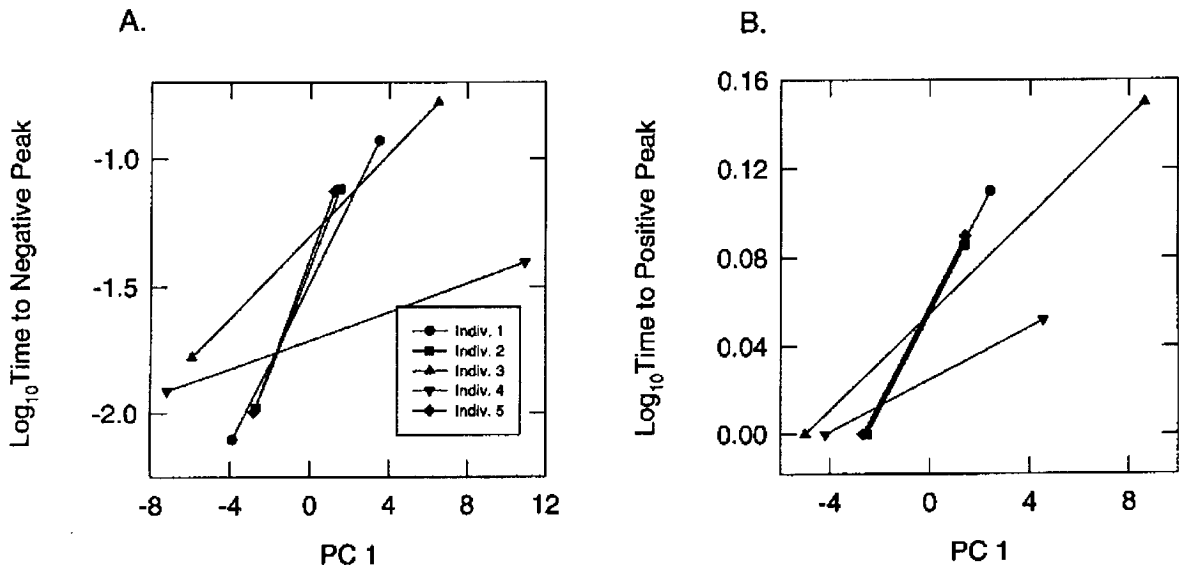


Fig. 4. Plots showing the relationships between PC1 and pressure variables, Time to NEG-pk (A) and Time to POS-pk

(B). Each line indicates the slope of the relationship for individual bass. (See text for abbreviations.)

Muscular basis of buccal pressure

EMG activity demonstrated moderate performance in accounting for the timing and magnitude of the pressure pulse during the strike. Multiple regressions on the two expansive phase variables, NEG-area and NEG-peak, accounted for 60 and 55% of the variance, respectively (Tables 3 and 4). Regressions on the compressive phase pressure variables, POS-area and POS-peak, had similar levels of success, with average R^2 values of 48 and 53%, respectively. Regressions on variables that represented the timing of major events in the pressure profile also showed similar R^2 values. Average R^2 values for PRESS-dur, Time to POS-pk, and Time to NEG-pk were 60, 51, and 39%, respectively.

The results of the regression models presented here for NEG-area are similar to findings with other fish taxa. For example, Wainwright and Turingan ('96) investigated the relationship between EMG activity and pressure characteristics during inflation behavior in striped burrfish (*Chilomycterus schoepfi*) and accounted for an average of 62% of the variance in NEG-area. Subambient pressure in suction feeding and inflation is generated by similar mechanisms. In a similar analysis of suction feeding in two other species of Centrarchids, the bluegill (*Lepomis macrochirus*) and the rockbass (*Ambloplites rupestris*), Lauder et al. ('86) generated regression models which used solely duration, onset, or amplitude variables of cranial muscle activity, and explained proportions of variance in NEG-area (avg. R^2 's = 38, 55, and 58%, respectively) similar to those reported here. In suction feeding, the magnitude of the subambient pulse has been considered a crucial aspect of buccal pressure which draws the prey into the mouth (Norton and Brainerd, '93; Muller et al., '82). The pressure variables NEG-area and NEG-peak represent the suction forces which are generated during the strike. Thus, with regard to NEG-area, the results of these three studies suggest that standard EMG measures of cranial muscle consistently explain around 50 to 60% of the variance in suction performance.

Although the multiple regressions indicated considerable variability in EMG patterns, some trends were apparent. The clearest pattern was a positive association between the time course of events within the pressure pulse and EMG integrated area and burst duration of all muscles and longer AM onset time (Figs. 1, 3; Table 2). This

pattern of association was confirmed by the multiple regression models for the pressure timing variables that were composed of burst duration and onset time EMG variables. With the exception of the results from individual number 4, the regression models were significant in each case. There was also a consistent trend for AM activity to account for superambient pressure (Tables 3 and 4) demonstrating a positive association between activity in this jaw-closing muscle and the pressure produced during buccal compression.

The contrast between strikes with high levels of EMG activity and others with much lower muscle activity levels appeared to be the major axis of variation in the data set. This could be seen in the principal components analysis, where PC1 (Table 6) was positively correlated with the integrated area and burst duration of all four muscles in addition to the onset time of the AM (but not the onset time of the other muscles). Factor scores on PC1 were positively correlated with pressure timing variables, and with the exception of PRESS-dur whose interaction term was barely significant, ANCOVA demonstrated that these associations were consistent across the five experimental fish (Fig. 4). Thus, in the strike of a bass, there is a consistent and positive association between the temporal features of muscle activity (i.e., onsets and burst durations) and the time course of the pressure pulse produced.

Associations between EMG variables and pressure magnitude variables (NEG-area, POS-area, NEG-peak, and POS-peak) were not as clear as seen with pressure timing variables. Of the 60 bivariate correlations among EMG and pressure magnitude variables, only two were significant: correlations between AM-dur and POS-area and POS-peak (Table 2). Nevertheless, multiple regression showed about the same level of success in accounting for these variables as was achieved with the pressure timing variables. Our inspection of raw data (e.g., Fig. 1) suggested that, like the timing variables, pressure magnitude was positively associated with higher levels of EMG activity. However, ANCOVAs that were run to examine the effect of factor scores from PC1 on the pressure magnitude variables indicated significant differences among individual fish in the shape of this effect (i.e., significant interaction terms). When correlations were inspected in the data sets of individual fish, we noted that those fish that demonstrated higher R^2 values in the multiple regressions also tended to have more significant bivariate correlations. For example, among the 105

correlations between EMG and pressure variables for individual number one, 56 were above an absolute value of 0.5. This contrasts markedly with the overall correlation matrix in which two of 105 correlations were above 0.5. Thus, the tendency for higher magnitudes of buccal pressure to be associated with longer muscle activity bursts and higher integrated area was quite strong in some individuals, but was not apparent in all fish.

Although EMG activity accounted for a significant proportion of the variation in pressure variables, a large fraction of the variation went unexplained in each case. Why might this be so? The simplest interpretation of this pattern is that a given magnitude of buccal pressure can be produced by a variety of muscle activation patterns (Fig. 2). In addition, we see at least two factors that may have contributed to this apparent trend. First, the functional link between the muscles that we studied and buccal pressure may be sound, but the parameters that we used to measure motor activity may not be robust indicators of the appropriate mechanical properties of the muscles. Rectified integrated area has been shown repeatedly to strongly reflect muscle tension (reviewed in Basmajian and De Luca, '85). Although quantitative measures of the strength of this relationship are not reported in these previous studies, graphical data presented by Lawrence and De Luca ('83) suggest a very strong relationship with R^2 values above 0.9. Furthermore, the relationship between muscle activity and the magnitude of positive buccal pressure recorded during oral compression behaviors has been shown to be quite strong, with R^2 values in excess of 0.9 (Wainwright and Turingan, '96). The relatively poor performance of EMG in accounting for subambient buccal pressure in this and other studies (Lauder et al., '86; Wainwright and Turingan, '96) may arise because muscular tension is less directly related to suction pressure than it is to superambient pressure. The magnitude of suction pressure is a complex function of the rate of buccal expansion, the magnitude of buccal expansion, and the size of the oral aperture through which water enters the mouth (Alexander, '67; Muller et al., '82). This more complex basis of subambient pressure may be reflected in the looser relationship observed between EMG and suction pressures.

Second, it is possible that muscles crucial to the modulation of buccal pressure were not included in our study. The feeding mechanism of *Micropterus* involves several muscles that were not included in our study, such as the hypaxialis,

levator operculi, and adductor arcus palatini. However, previous work with this species indicates that the four muscles studied are the primary effectors of the principal motions of the strike: cranial elevation, suspensorial abduction, hyoid depression, jaw depression, and jaw adduction (Lauder, '83, '85; Wainwright and Lauder, '86). Theoretical considerations indicate that during suction feeding, the magnitude of the subambient pressure pulse will be a function of (1) the rate and magnitude of the volume change as the buccal cavity expands, and (2) the area of the oral opening (Muller et al., '82). Variation among feeding events in pressure patterns is therefore expected to be largely due to variation in the net effects of the primary buccal expansion muscles, such as the LAP, SH, and EP.

An additional source of variation in our data could have arisen from slight regional variation in the pattern of buccal pressure during the strike. By locating the pressure canula in the same region of the buccal cavity in each experimental fish, we minimized this potential effect. Regardless of exactly where in the buccal cavity the pressure transducer was located, we expect that the general relationships between muscle activity and the magnitude of buccal pressure would be the same. Thus, we view the possibility of regional buccal pressure variation as an unlikely contributor to the relatively low explanatory power of EMG with regard to pressure, but it is possible that such variation contributed to quantitative differences between individual fish in pressure values.

One striking pattern that emerged from this study is that the LAP was active in every strike we analyzed. In contrast, activities in the other muscles, EP, SH, and AM, were absent in nearly 20% of the strikes including both successful and unsuccessful attempts. One possible interpretation is that activity in the LAP provides an essential mechanical movement in the strike. It may be that lateral expansion of the suspensorium is a necessary component in the onset of any magnitude suction strike. In contrast, EP and SH activity were not always observed during successful prey capture, suggesting a less critical role in effective suction feeding.

One potentially important component of the strike that we did not address is the relationship between the buccal and opercular cavities. During suction feeding, the skeletal elements of the gill apparatus mechanically separate the buccal cavity from the opercular cavity (Lauder, '83), and work with other species of the Centrarchidae

found movements of the gill bars to be better predictors of buccal pressure during suction feeding than EMG variables (Lauder et al., '86). Although the difference between buccal and opercular pressure is sharper in species more specialized for suction feeding (Norton and Brainerd, '93), the possibility that gill bar motion plays a significant role in modulating buccal pressure somewhat independently of the muscles studied here, cannot be discounted.

ACKNOWLEDGMENTS

We thank J. Friel, K. Ralston, and C. Morrison for their constructive comments during the preparation of this manuscript. Financial support was provided by NSF grant IBN-9306672 to P.C.W.

LITERATURE CITED

- Alexander, R. McN. (1967) Functional Design in Fishes. Hutchinson, London.
- Basmajian, J.V., C.J. De Luca (1985) Muscles Alive. Butler J. and Eckhart C. eds. Williams & Wilkins, Baltimore, MD.
- Bramble, D.M., and D.B. Wake (1985) Feeding mechanisms of lower vertebrates. In: Functional Vertebrate Morphology. M. Hildebrand, D.M. Bramble, K.F. Liem, D.B. Wake, eds. Harvard University Press, Cambridge, MA, pp. 230-261.
- Lauder, G.V. (1980) Evolution of the feeding mechanism in primitive actinopterygian fishes: A functional analysis of *Polypterus*, *Lepisosteus*, and *Amia*. *J. Morphol.*, 163:283-317.
- Lauder, G.V. (1981) Intraspecific functional repertoires in the feeding mechanism of the characoid fishes *Lebiasina*, *Hoplias*, and *Chalceus*. *Copeia*, 1981:154-168.
- Lauder, G.V. (1983) Prey capture hydrodynamics in fishes: Experimental tests of two models. *J. Exp. Biol.*, 104:1-13.
- Lauder, G.V. (1985) Aquatic feeding in lower vertebrates. In: Functional Vertebrate Morphology. M. Hildebrand, D.M. Bramble, K.F. Liem, and D.B. Wake, eds. Harvard University Press, Cambridge, MA, pp. 210-229.
- Lauder, G.V., P.C. Wainwright, and E. Findeis (1986) Physiological mechanisms of aquatic prey capture in sunfishes: Functional determinants of buccal pressure changes. *Comp. Biochem. Physiol.*, 84A:729-734.
- Lawrence, J.H., and C.J. De Luca (1983) Myoelectric signal versus force relationship in different human muscles. *J. Appl. Physiol.: Respirat. Environ. Exercise Physiol.* 54:1653-1659.
- Liem, K.F. (1978) Modulatory multiplicity in the functional repertoire of the feeding mechanism in cichlid fishes. *J. Morphol.*, 158:323-360.
- Liem, K.F. (1979) Modulatory multiplicity in the feeding mechanism of cichlid fishes, as exemplified by the invertebrate pickers of Lake Tanganyika. *J. Zool. Lond.*, 189:93-125.
- Muller, M., J. Osse, and J.H.G. Verhagen (1982) A quantitative hydrodynamic model of suction feeding in fish. *J. Theor. Biol.*, 95:49-79.
- Nishikawa, K.C., C.W. Anderson, S.M. Deban, and J.C. O'Reilly (1992) The evolution of neural circuits controlling feeding behavior in frogs. *Brain Behav. Evol.*, 40:125-140.
- Norton, S.F. (1991) Capture success and diet of cottid fishes: The role of predator morphology and attack kinematics. *Ecology*, 72:1807-1819.
- Norton, S.F., and E.L. Brainerd (1993) Convergence in the feeding mechanics of ecomorphologically similar species in the Centrarchidae and Cichlidae. *J. Exp. Biol.*, 176:11-29.
- Nyberg, D.W. (1971) Prey capture in the largemouth bass. *Am. Midland Nat.*, 86:128-144.
- Richard, B.A., and P.C. Wainwright (1995) Scaling the feeding mechanism of largemouth bass (*Micropterus salmoides*): Kinematics of prey capture. *J. Exp. Biol.*, 198:419-433.
- Sanderson, S.L. (1988) Variation in neuromuscular activity during prey capture by trophic specialists and generalists (Pisces: Labridae). *Br. Behav. Evol.*, 32:257-268.
- Schwenk, K., and G.S. Throckmorton (1989) Functional and evolutionary morphology of lingual feeding in squamate reptiles: Phylogenetics and kinematics. *J. Zool. Lond.*, 219:153-175.
- Sibbing, F.A., J.W.M. Osse, and A. Terlow (1986) Food handling in the carp (*Cyprinus carpio*): Its movement patterns, mechanisms and limitations. *J. Zool. Lond.*, 210:161-203.
- Wainwright, P.C. (1989) Functional morphology of the pharyngeal jaw apparatus in Perciform fishes: An experimental analysis of the Haemulidae. *J. Morphol.* 200:231-245.
- Wainwright, P.C., and G.V. Lauder (1986) Feeding biology of sunfishes: Patterns of variation in the feeding mechanism. *Zool. J. Linn. Soc.*, 88:217-228.
- Wainwright, P.C., and B.A. Richard (1995) Scaling the feeding mechanism of the largemouth bass (*Micropterus salmoides*): Motor pattern. *J. Exp. Biol.*, 198:1161-1171.
- Wainwright, P.C., and R.G. Turingan (1993) Coupled versus uncoupled functional systems: Motor plasticity in the queen triggerfish, *Balistes vetula* (Teleostei, Balistidae). *J. Exp. Biol.*, 180:209-227.
- Wainwright, P.C., and R.G. Turingan (1996) Muscular basis of buccal pressure: Inflation behavior in the striped burrfish, *Chilomycterus schoepfi*. *J. Exp. Biol.*, 199:1209-1218.
- Westneat, M.W. (1991) Linkage biomechanics and evolution of the jaw protrusion mechanism of the sling-jaw wrasse, *Epibulus insidiator*. *J. Exp. Biol.*, 159:165-184.
- Wilkinson, L. (1992) Systat, for Windows, Version 5. Systat, Inc., Evanston, IL.