Fish Out of Water: How Environment Impacts Body Propulsion in an Elongate Fish (*Erpetoichthys calabaricus*)

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Abstract

The environment an organism travels in is dependent on the type of locomotion it performs. There are several species of highly elongate fish that can inhabit an aquatic environment while also making terrestrial excursions. In this study, we investigated substrate use in *Erpetoichthys calabaricus* (ropefish) during aquatic and terrestrial locomotion. Like other elongate fishes, *E. calabaricus* is known to push against a substrate through their pre-caudal and caudal regions in terrestrial environments and uses their caudal region to produce propulsive force in aquatic environments. The pushing force of four individuals of *E. calabaricus* was measured between the two environments using a pegboard array spaced at 5 cm with one peg instrumental with two uniaxial strain gauges. Force was recorded using *Acqknowledge* software. From this study, it was found that there was no difference of force production between the terrestrial and aquatic environments. This study will determine how certain fish are able to move out of water using only their axial skeleton.

Keywords: *Erpetoichthys calabaricus*, Force Production, Substrate-Based Locomotion

1. Introduction

The emergence of new behaviors and structural appendages has evolved with terrestrial locomotion in vertebrates[1]. It has been known that the earliest tetrapods derived from brackish to marginally marine deposits, with aquatic tolerance[2, 3]. Recently, *Tiktaalik roseae* was shown to have characteristics of both fish and tetrapods[4]. Environmentally induced phenotypes incorporate hereditary traits that could play an important role in evolution such as the introduction to novel traits to increase fitness[5, 6]. A previous study has focused on air-breathing mechanisms that attempt to elucidate the physiological demands for aerial exposure in amphibious fishes, however little is known about behavioral variation in emergence from an aquatic environment[7].

Terrestrial locomotion in fishes can be attributed to transient ballistic behavior as an escape response to predators or sustained periodic movements to pursue prey on land or to migrate to a different body of water[8, 9]. Mudskippers would use their pectoral fins and tail to move away from a negative stimulus, and capture prey[10]. There are some amphibious fishes that make terrestrial excursions to spawn in order for a better survival rate of the eggs to avoid aquatic hypoxia and allow exposure to warmer temperatures[11]. Amphibious fish would also relocate to a terrestrial environment in response to temperature or presence of water contaminants. Killifish leave water that is contaminated with hydrogen sulfide[8].

Snakes can undergo sidewinding, which is where horizontal and vertical planes of the body superimpose on one another with a difference of approximately 90˚. *Crotalus cerastes* (sidewinder rattlesnakes) can use shallow differential turning and sharp reversal turning, which allows for gradual turning and a change in path without body rotation, respectively[12]. Lateral undulation is where the body pushes against irregularities of a substrate to overcome

sliding resistance from friction[13]. Jayne (1986)[14] observed pure lateral undulation, a combination of lateral undulation and sidewinding, and pure sidewinding when *Cerberus rynchops* traveled on sand and led Jayne (1986)[14] to conclude that snakes may use a variety of locomotor modes that increase their velocity for a given substrate. The shift from lateral undulation to sidewinding is thought to generate external resistive forces from vertical vertebral flexion to allow for the sidewinding motion[15].

Phylogenetically disparate fishes had structures such as whole bodies, specialized pectoral fins, and head, to interact with their surrounding substrates[16]. Some fishes that are completely aquatic, such as sharks and stingrays, use appendage-drive locomotion[17, 18]. An aquatic fish that makes terrestrial excursions must utilize a musculoskeletal system to displace the viscous medium for the production of ground reaction forces (GRF) against a substratum while moving through a fluid environment under gravity[7]. When *Protopterus annectens* (African lungfish) is submerged in water, it uses its paired fins for propulsion against a substrate; however, in a terrestrial environment the thin, flexible fins of the lungfish are insufficient for propulsive forces and the lungfish relies on its ossified crania and trunk, using the head as an anchor point to move the body around[16, 19, 20]. Climbing gobiids from Hawaii, such as *Lentipes concolor*, vertically propel themselves by simultaneous adduction of the paired pectoral fins and subsequent forceful axial-body undulations in order to generate lateral movements at the head and tail[7].

Highly elongate fishes, such as *Erpetoichthys calabaricus* (ropefish), can transition from an aquatic environment to a terrestrial environment. Ropefish inhabit a variety of aquatic habitats ranging from slow flowing rivers to seasonal flood plains in central Africa[21]. According to the IUCN (2010), *E. calabaricus* is Near Threatened and is jeopardized by habitat deterioration due to wetland drainage for agricultural development and deforestation[22]. Ropefish are polypterids that can tolerate low oxygen levels most likely due to their largely paired lungs that ventrally connect with the esophagus and well-developed gills that are highly vascularized[23]. Similarly, other elongate fishes such as *Polypterus senegalus* also have this similar respiratory anatomy that also allows them to have tolerance to low oxygen levels and make terrestrial excursions[24]. Anatomically, in both animals, there is a double circulatory loop that is not as developed as the lungfish, which is an obligate air-breathing fish[25]. In an experiment that studied air breathing in ten individuals of *E. calabaricus*, it was found that within a 24 hour period, each fish made an average of 6 terrestrial excursions lasting an average of 2 minutes each; it was also observed that during air exposure, *E. calabaricus* can maintain and increase oxygen uptake for short-term terrestrial excursions to consume insects found on land[26].

Ropefish are known to push against a substrate using their caudal region in an aquatic environment and their pre-caudal and caudal regions in a terrestrial environment[27, 28]. In a previous study, it was found that in an aquatic environment, the tail undulates and produces the most propulsive force, while other body regions do not and since the tail is laterally compressed in a terrestrial environment, the tail produces a temporal lag[27]. Similarly, in American eels (*Anguilla rostrata*), there is reduced muscle activation in the posterior regions of the animals in terrestrial trials than there is in aquatic trials[29]. With an intermediate environment, as water levels decreased, it was found that there was an increase in wave amplitude and lateral excursions in the anterior regions of *E. calabaricus*; as the ventral surface of the ropefish interacts with the substrate, there is reduced viscosity experienced by the dorsal region of the ropefish[27]. Most importantly, it is currently unknown as to what region of the body is the most important for pushing off a substrate.

A previous study determined that there was a difference found in peg contact for axial elongation between aquatic and terrestrial environments in *E. calabaricus* and *P. senegalus*[28]. While the previous study examined how fish contact pegs, in this study I will test the force produced by that contact. It was hypothesized that there would be differences in force production during pushing between aquatic and terrestrial environments. I predict that there would be greater force produced in a terrestrial environment.

2. Materials and Methods

2.1 Experimental Setup

Four individuals of *E. calabaricus* were obtained from commercial fish dealers: individuals and their respective body masses were weighed after each experiment. Each individual was kept in a 38-liter glass aquarium with freshwater that was maintained at a temperature range of 24–27˚C with standard aquarium heaters. All experiments performed were approved by the Adelphi University Institutional Animal Care and Use Committee.

A 30.5 cm x 76.75 cm pegboard was fitted with 77 pegs that were 5 cm in height. The distance between each adjacent peg was 5 cm (Figure 1a). The pegboard was placed in a plastic container 88.3 cm x 41.9 cm x 15.2 cm. With terrestrial

trials, water would be added to the pegboard for lubrication. With aquatic trials, water would be added to the height of each peg (5 cm). Filming was performed by a Panasonic LUMIX DMC-FZ1000 camera.

There was one peg (14.6 cm) with two drill holes towards the bottom and top that is 90° apart and placed within the last two rows of the pegboard toward the right side (Figure 1b). Force transducers were attached to a peg at a 90° angle with the top two screws placed in the drill holes. A ruler was used to measure the lengths of the strain gauges for consistency. One hook was added to one drill hole and one at a time, a 20 g and 100 g weights were added on top of the hooks for calibration prior to each trial. BIOPAC MP150 with DA100C amplifiers was set with a 1000 fold gain and stain gauges were 50g.

Individuals were allowed to acclimate and explore the pegboard environment 10 minutes prior to filming. All trials were filmed. Once acclimated, a trial began by encouraging individuals to move by gentle coaxing by hand or with a hand net. After trials were completed, the videos were observed and the time when the animal pushed along the large peg was recorded. The amount of time that the animal pushed along the peg was also recorded.

2.2 Data Analysis

The exact time the animal pushed along the larger peg was coordinated with *Acqknowledge*. A full body push for the terrestrial environment and aquatic environment was considered as each individual using 100% and 75-100% of their body per push, respectively. There were a total of 119 terrestrial pushes and 55 aquatic pushes for all individuals combined. The aquatic environment pushes were more difficult to acquire due to all individuals not needing use the pegs for movement; in the aquatic environment the individuals would swim instead of pushing off the pegs, while the terrestrial environment lacked water, and individuals needed the pegs for movement. Therefore, the consideration of a full body push was more lenient for an aquatic trial than a terrestrial trial. To analyze this data, the peak is highlighted from the beginning to the very end of the push for one of the force transducers. The data was collected for peak-to-peak, integral, time, and change in time. The highlighted peak remained highlighted as the data for the other force transducer was collected. Using data points from the peak-to-peak values, Pythagorean Theorem was used to calculate the resultant force. Resultant force was standardized by body mass to account for difference in mass between individuals. The integral over time was calculated by dividing the orange (or green) integral by the change in time. All data was log-transformed prior to statistical analysis. Force was analyzed using a two-way ANOVA with individual and environment as factors with a Tukey Post-Hoc Test (JMP v.#7).
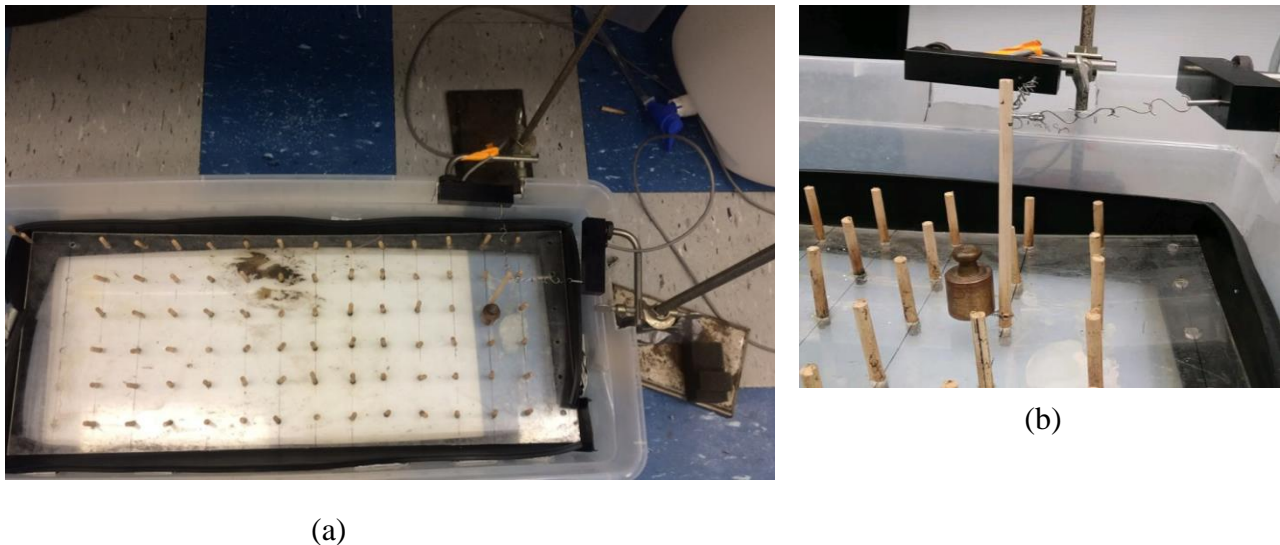

(a)

(b)

Figure 1. Experimental set-up. (a) Peg-board set up; (b) strain gauges force transducers

3. Results

There was a statistically significant difference (p-value= 0.011) between the standardized resultant force produced in aquatic and terrestrial environments for Individual 6 only (Figure 2). There were no other differences between

environment and individuals. Individuals 9 and 10 produced different resultant force than Individuals 6 in the terrestrial environment. In the aquatic environment, there were no differences between any individuals.

   The total force is the amount of force produced by an individual for each push. There was a statistically significant difference (p-value< 0.0001) between the log total force produced in aquatic and terrestrial environments for Individuals, 6, 9, and 10 (Figure 3). The Individual*Environment variable was statistically significant (p-value< 0.0001). This data was not standardized by body mass. There was a difference between aquatic and terrestrial trials for Individual 6. Individuals 10 and 11 were similar between aquatic and terrestrial trials. Individuals 10 and 11 were different from Individuals 6 and 9, where Individual 6 had a difference between both aquatic and terrestrial trials and Individual 9 only differed in terrestrial trials. Individual 6 was different from Individual 9 in the aquatic environment only.

Table 1. Results of the two-way ANOVA for the two variables considered in this study

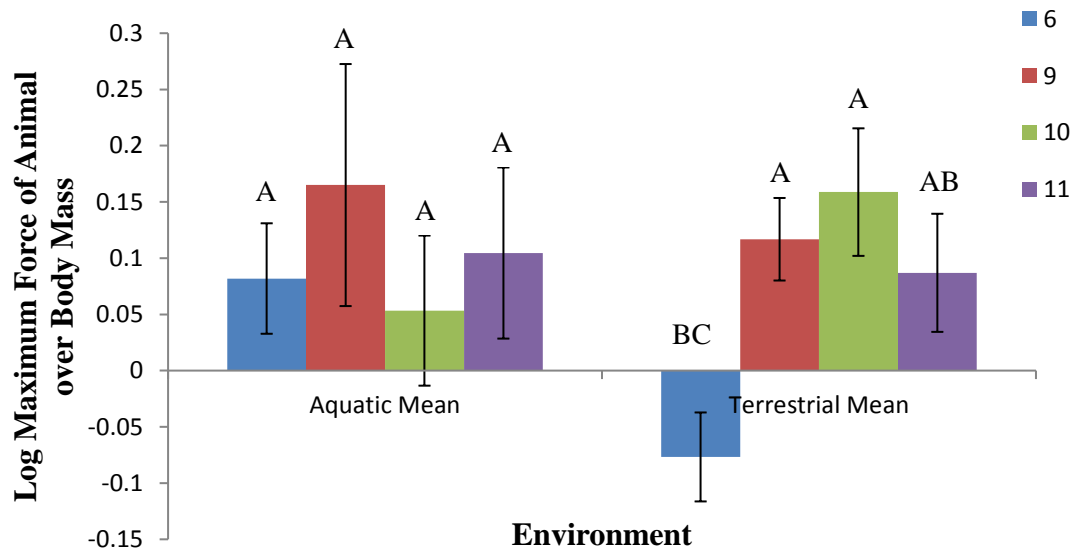| Variable	DF	F Ratio	P-Value	Eta-Squared
Standardized Resultant Force	170	2.7168	**0.0109**	10.54
Individual	3	2.34	0.075	0.41
Environment	1	0.43	0.51	0.02
Individual*Environment	3	2.07	0.11	0.36
Log Total Force	170	73.4258	**<.0001**	118.46
Individual	3	26.91	**<.0001**	14.13
Environment	1	94.77	**<.0001**	16.58
Individual*Environment	3	54.48	**<.0001**	28.60
Bolded p-values indicate significance <0.05				

Figure 2. Standardized Resultant Force of Animal over Body Mass (Standardized by Body Mass).

There was a statistically significant difference (p-value= 0.0109) in the Two-Way ANOVA, however, there were no differences between the aquatic and terrestrial environments, except with individual 6. There were no differences

between individuals in each environment with the exception of individual 6. Bars with the same letter are not significantly different from one another.
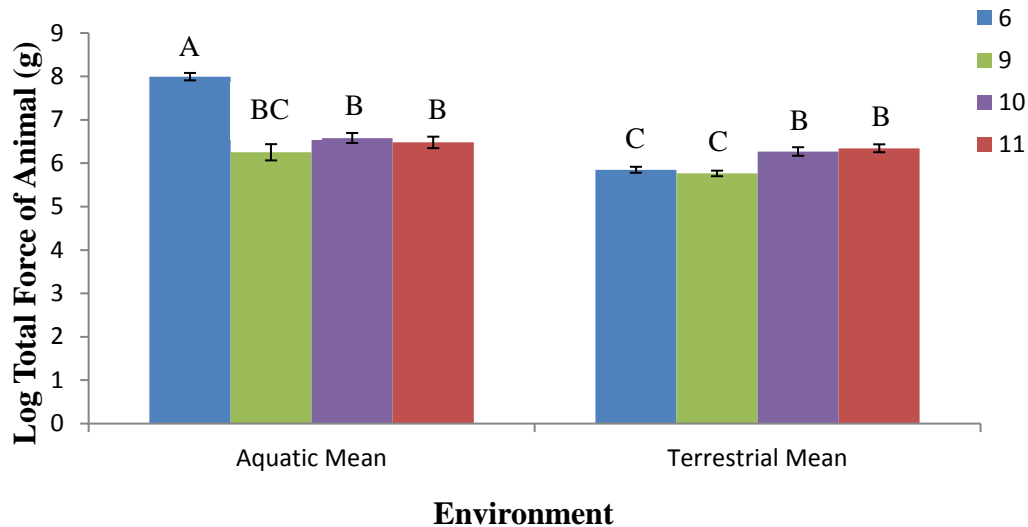

Figure 3. Log Total Force of Animal (not standardized by body mass).

There was a statistically significant difference (p-value= <0.0001) in the Two-Way ANOVA, however, there were no differences between the aquatic and terrestrial environments. There was no difference between the individuals, except for Individual 6, which was greater from the other individuals in the aquatic environment. Terrestrially, Individuals 6 and 9 were different from Individuals 10 and 11. Bars with the same letter are not significantly different from one another.

4. Discussion

The purpose of this experiment was to determine if there were any differences in force production between aquatic and terrestrial environments in *Erpetoichthys calabaricus*. I found only a few cases where force production differed between environments (Individual 6). A probable reason for significant differences in force production between aquatic and terrestrial environments seen in Individual 6 is due to Individual 6 using fewer pegs in a terrestrial environment than any other individual; Individual 6 had lower terrestrial forces (Figure 2). There was also more data collected for Individual 6 than any other individual (Individual 6: 37 terrestrial and 24 aquatic pushes, Individual 9: 43 terrestrial and 5 aquatic pushes, Individual 10: 19 terrestrial and 13 aquatic pushes, Individual 11: 20 terrestrial and 13 aquatic pushes).

During terrestrial trials, it was noticed that the ropefish would undulate their entire body using both their pre-caudal and caudal regions. However, there were many terrestrial trials where individuals would use their pre-caudal region, and the midsection slips until the caudal region thereby not using the entire midsection of their bodies. These pushes were not included in the data analysis. In the aquatic trials, all individuals tended to not use the pegs for movement, similarly seen in Ward et al. 2015[28]. The individuals in an aquatic environment swam between the pegs using their fins. Aquatic trial pushes were shorter in duration than terrestrial trial pushes. For the two-way ANOVA seen in Figure 2, the significant differences for Individuals 9 and 10 could be due to the fact that these specific individuals moved faster in the aquatic environment, having a smaller change in time for a full-body push.

4.1 Factors for Performance

With undulatory kinematics, Newton's Third Law states that for every force, there is an equal and opposing force. A force applied to a fluid essentially has two components, where the resistive component is the friction from the fluid-solid interface and the inertial component is from the acceleration of the fluid away from the moving individual[30]. It

has been shown that anguilliform swimmers have lateral drag and thrust, which uses more of the resistive component for slender bodies[30]. In an aquatic environment, *E. calabaricus* would produce the same forces when moving through the water and pegboard.

Since there is no difference of force production between aquatic and terrestrial environments (with the exception of individual 6), this could potentially be due to each individual touching the same amount of pegs before and after touching the peg that measures force. Individual 6 could have been touching more pegs in the aquatic environment, thus having more support for movement and a greater force than the terrestrial environment (Table 1). A previous study found that by increasing the amount of contact points increases lateral and forward forces, as well as increasing friction; thereby, suggesting that lateral undulation with more waves during movement is less efficient for terrestrial locomotion[31]. However, studies involving snakes traveling on a smooth surface reveal that contacting multiple pegs cancels the lateral force vectors on the body to avoid slipping and also reduces the curvature of each peg, which allows the animal to travel faster[32]. Snakes also seem to touch more pegs as the spacing of pegs increases, although previous studies have not yet quantified the amount of pegs touched in relation to peg spacing[33]. Since snakes share a similar elongate body to *E. calabaricus*, *E. calabaricus* would most likely travel similarly on a pegboard to snakes.

The movement and force production of *E. calabaricus* applies to the understanding of biophysics in robotics. Snake-like technologies (i.e. snake robots) are designed to find survivors in collapsed buildings or be used for less-invasive and more diagnoses for diseases[34]. Currently, procedures for designing swimming micro-robots are proposed and could travel in low velocity fluids[35] and could be used for military purposes. Undulatory locomotion has many functional uses in the medical and military fields.

4.2 Future Work

In the future, I would like to track the velocity and acceleration of the fish during pushes both aquatically and terrestrially. It would be very interesting to see if the animals have a preference for going faster in certain environments over others, especially from escaping predators. I could also standardize the force data by velocity. Another possible correlation is observing the amount of energy expended by each of these individuals; I would test how much oxygen and carbon dioxide each individual is inhaling and releasing, respectively, in each environment to observe if there are similar amounts of energy expended, since there is a similar force exerted in each environment.

5. Conclusions

Generally, there were no differences in force production between aquatic and terrestrial trials, with the exception of a few individuals due to the various forces exerted by individuals in each environment. It would be interesting to observe the velocity, acceleration, and energy exerted by each individual in each environment. In observing these factors, I could see if there is a preference in expending energy and velocity in a specific environment for given circumstances (i.e. if there was a predator or source of food nearby).

6. Acknowledgements

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