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**Location: Periodical Collection 3rd
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Journal Title: Ethology.

Odyssey

Volume: 96 **Issue:** 3
Month/Year: 1994**Pages:** 213-232

Article Author: BRANTLEY,

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Article Title: ALTERNATIVE MALE SPAWNING
TACTICS AND ACOUSTIC-SIGNALS IN THE
PLAINFIN MIDSHIPMAN FISH PORICHTHYS-
NOTATUS GIRARD (TELEOSTEI,
BATRACHOIDIDAE)

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Alternative Male Spawning Tactics and Acoustic Signals in the Plainfin Midshipman Fish *Porichthys notatus* Girard (Teleostei, Batrachoididae)

RICHARD K. BRANTLEY & ANDREW H. BASS

BRANTLEY, R. K., BASS, A. H. 1994: Alternative male spawning tactics and acoustic signals in the plainfin midshipman fish *Porichthys notatus* Girard (Teleostei, Batrachoididae) *Ethology* 96, 213—232

Abstract

The plainfin midshipman *Porichthys notatus* has two male reproductive morphs, 'Type I' and 'Type II', which are distinguishable by their physical traits alone. Type I males are eight times larger in body mass than Type II males and have a six-fold larger relative sonic (vocal) muscle mass than Type II males. In contrast, the testicles of Type II males are seven times larger than those of Type I males. This study demonstrates morph-specific patterns of reproduction, including acoustic signals, for Type I and II males. Field censuses of nests showed that only Type I males maintained nests. Type II males and females transiently appeared in these nests in association with each other. Infra-red video and hydrophone recordings in aquaria showed that Type I males maintained nests and readily vocalized. Long-duration 'hums' and sequences of short-duration 'grunts' were produced during advertisement and agonistic contexts, respectively. Humming Type I males attracted females to their nests, pair-spawned, and then guarded egg clutches alone. By contrast, Type II males neither acoustically courted females nor maintained available nest sites, but rather 'sneak-' or 'satellite-spawned' at the nests of Type I males. Type II males infrequently produced low amplitude, short duration grunts that were similar in spectral, temporal and amplitude characteristics to the grunts of females. Type II males appear to be obligate sexual parasites of the nest-building, mate-calling, and egg-guarding Type I males. The dimorphic body and vocal muscle traits of the two male morphs in the plainfin midshipman are thus paralleled by a divergence in their reproductive tactics and the properties of their acoustic signals.

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Introduction

Alternative reproductive tactics are discontinuous male reproductive behaviours used to obtain fertilizations (CARO & BATESON 1986; GROSS 1984). Within a species, males may vary both in aggressive competition and in the active courtship of females. Non-courting males may attempt forced copulation, or in the case of externally fertilizing fishes, release sperm either at a distance from (satellite-spawning), or close to (sneak-spawning), a mating pair using stealth, speed, or female mimicry to gain proximity. In contrast to the numerous cases where males can facultatively change reproductive tactics, permanent male dimorphism with alternative tactics is known in few vertebrate groups, of which sunfishes are an example (DOMINEY 1980; GROSS & CHARNOV 1980), as are Pacific salmonids (GROSS 1985), swordtail fish (KALLMAN & BORKOSKI 1977; RYAN & CAUSEY 1989; ZIMMERER & KALLMAN 1989), and among birds, the ruff *Philomachus pugnax* (VAN RHIJN 1966).

The plainfin midshipman, *Porichthys notatus* Girard (Batrachoididae), is a marine fish of western North America long recognized for its sound-producing abilities. GREENE (1924) first described the sonic muscles that are attached to the swimbladders of nest-building males. These muscles contract rapidly to produce the 'whirring' sound described by COX (1905), earning this species the name California singing fish. Vocal behaviour in a social context has been reported in several dozen families of teleost fishes (FISH & MOWBRAY 1970; MYRBERG 1981). Yet *P. notatus* is remarkable for one unique sound — the hum — produced by nesting males and attractive to gravid females (IBARA et al. 1983). *P. notatus* is now known to have two reproductively mature male morphotypes, designated Type I and Type II, which are distinguishable by somatic and neurobiological features (Fig. 1; BASS 1990, 1992, 1993). Based on patterns of sonic muscle development, the Type I and II morphs appear to be endpoints of alternative developmental trajectories and are not simply part of an ontogenetic sequence (BRANTLEY et al. 1993a).

Here we demonstrate the reproductive behavioural correlates of male dimorphism in relative gonad size, relative sonic muscle size, and body size in *P. notatus*. Despite the difficulty of field work with this inter-tidal species (murky waters, nocturnal courtship, and nesting beneath large rocks), aquarium observations using infrared videotape and hydrophone recordings, together with field censuses of nests, permitted identification of distinct reproductive tactics for each of the male morphs. Portions of this work have been summarized in abstract form (BRANTLEY & BASS, 1988, 1990, 1991; BRANTLEY et al. 1989).

Methods

Field observations were made along the western shore of Tomales Bay, California, from late May to Aug., a period encompassing almost the entire spawning season at this locale. Midshipman (*P. notatus*) were collected from nests and transported to the Bodega Marine Laboratory for aquarium observation. Some pertinent morphological characteristics of the two male types and female *P. notatus* used in this study are summarized in Table 1.

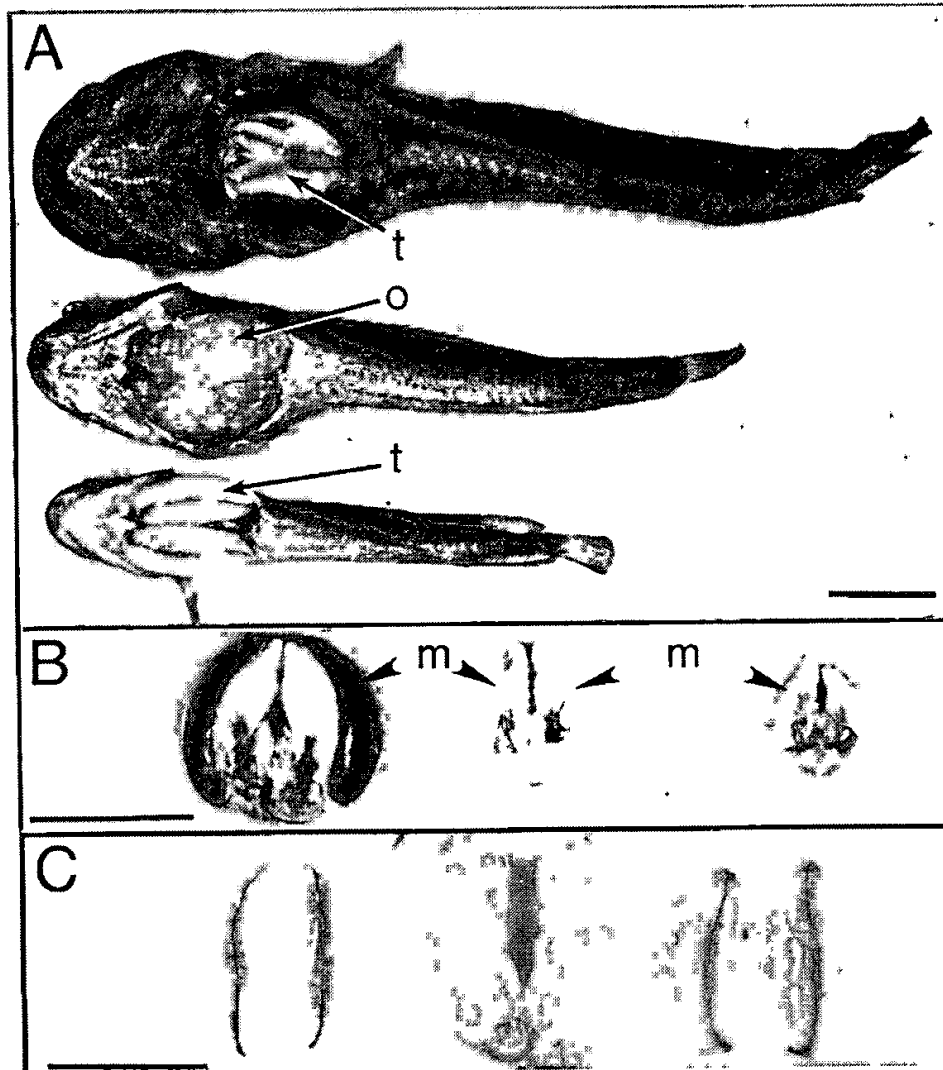


Fig. 1: Morphological differences between the *Porichthys notatus* male morphs and females (also see Table 1). A. Ventral view of three *P. notatus* taken from a nest, dissected to show gonads. Top to bottom: Type I male, gravid female, Type II male (t = testis; o = ovary). B. Swimbladders from the same specimens pictured above. Left to right: Type I male, female, Type II male (m = sonic muscle). Sonic muscle is difficult to see in the female and Type II male because of its small size and lack of pigmentation. C. Gonads from the same three specimens. The testes from the Type I male (left) were smaller in both absolute and relative size than those of the Type II male (right), despite a 5-fold difference in body weight between the pictured males. Note difference in size of scale bars, all of which represent 2 cm

Field Observations

During the lowest morning tides of the full and new moons (approx. 10 days per month), we located nests beneath 2–20 kg rocks exposed in the lower intertidal zone on sand or mud. At the time of lowest water (1–3 h daily), these nests contained only small pools of water or no water at all; *P. notatus* males, females and egg clutches apparently tolerate these conditions and remain moist beneath a rock.

From 1987 to 1990, 630 *P. notatus* nests were systematically noted and all 893 occupants were identified and, usually, returned to the nest, after which the rock was carefully replaced. Sex and morph were determined through a combination of ventral colouration, body size, the visibility of gonadal or egg profiles, and the shape of the urogenital papilla (Table 1; DEMARTINI 1988; GRAY & WINN 1961). The reliability of these characteristics in identifying age, sex and morph has been verified

Table 1: Morphological characteristics of breeding *P. notatus*. See BASS 1990, 1992; BASS & ANDERSEN 1991; BASS & MARCHATERRE 1989 and BRANTLEY et al. 1993a for detailed consideration of peripheral and neural features.

Characteristic	Type I males	Type II males	Females
Standard length (cm)	$\bar{X} = 15.3$ cm range = 12.1—21.0 cm SD = 2.5 cm; n = 23	$\bar{X} = 8.7$ cm range = 6.5—10.5 cm SD = 1.3 cm; n = 21	$\bar{X} = 12.2$ cm range = 9.6—15.2 SD = 1.3 cm; n = 22
Weight (g)	$\bar{X} = 70.1$ g range = 30.9—206.9 g SD = 42.8 g; n = 23	$\bar{X} = 9.2$ g range = 3.9—17.3 g SD = 3.7 g; n = 21	$\bar{X} = 28.7$ g range = 11.1—47.3 g SD = 12.0; n = 22
Ventral appearance	light to dark grey	mottled yellow; grey dots; belly very firm over testes	metallic golden belly grossly distended, eggs visible (gravid) or distended, flaccid (spawned)
Urogenital papilla ¹	elongate, posterior	elongate, posterior	small, conical, in vent
Sonic muscle color	red-brown	white	white
Sonic muscle relative size as % body weight	$\bar{X} = 1.2$ % range = 0.6—1.5 % SD = 0.2 %; n = 20	$\bar{X} = 0.16$ % range = 0.1—0.37 % SD = 0.07 %; n = 21	$\bar{X} = 0.12$ % range = 0.06—0.19 % SD = 0.04 %; n = 14
Relative gonad size as % body weight	$\bar{X} = 1.2$ % range = 0.69—1.7 % SD = 0.3 %; n = 23	$\bar{X} = 8.3$ % range = 2.2—16.6 % SD = 4.2 %; n = 21	$\bar{X} = 21.7$ % (gravid) ² range = 15.5—27.9 % SD = 4.4 %; n = 15
Motile sperm in testes	present	present	—
Fertile	yes	yes	—

¹ See also DEMARTINI 1988; GRAY & WINN 1961. ² 70—170 eggs, DEMARTINI 1990.

in extensive studies where morph type was confirmed by dissection and light or electron microscopy (BASS & ANDERSEN 1991; BASS & MARCHATERRE 1989; BRANTLEY et al. 1993a). The identity of fish used in aquarium observations was confirmed by dissection at the end of the season.

Daylight Observation Aquaria

In the summers of 1988 to 1990, three indoor observation aquaria (150—380 l) on natural photoperiod were furnished with a sand bottom. Four distinct nest sites were made from portions of broken cement blocks and tiles. Each tank was stocked with fish (two Type I males, two or three Type II males, and two females) identifiable both by markings of acrylic paint injected subcutaneously into fins and by measured body sizes. Type I males took several days to acclimatize, and then began to occupy and excavate gravel from the nest sites. Spawning, initiated in darkness, were observed the following day from morning until their completion hours later. Spawned females were then replaced with freshly collected gravid females. Periods of 12 aquarium spawnings in which Type II males participated were observed in these tanks.

Infrared Video and Acoustic Recordings

During the 1990 and 1991 seasons, a second set of aquaria (80—200 l) were established outdoors on thick styrofoam cushions for acoustic and electrical insulation. These tanks contained a single Type I male in his nest. Infrared video recordings with synchronized hydrophone recordings were used to determine male and female courtship behaviour, the acoustic repertoire of Type I males, and whether Type II males and females were capable of producing sounds. A neurophysiological study of the sonic motor pathway has predicted that if females and Type II males are capable of producing sounds, their signals will be of a similar fundamental frequency to, and distinct from that of Type I males (BASS & BAKER 1990, 1991). Therefore, the majority of videotapes contained one Type I male and one gravid female present, in order to first characterize any female courtship or spawning sounds. The nocturnal initiation of spawning behaviour and its preceding courtship were videotaped 11 times in 1990 and 1991. Portions of numerous other spawnings and social interactions were also recorded, including two

Table 2: Field censuses of *P. notatus* nests

Year of census	1987	1988	1989	1990
Number of nests in census:	217	120	233	60
Type I males nesting alone:	76 %	69 %	76 %	75 %
with eggs:	(56 %)	(55 %)	(50 %)	(42 %)
without eggs:	(44 %)	(45 %)	(50 %)	(58 %)
Type I males with female &/or Type II males present:	21 %	29 %	22 %	25 %
Type II males or females nesting alone:	0 %	0 %	0 %	0 %
Empty nest (eggs):	3 %	2 %	2 %	0 %
	100 %	100 %	100 %	100 %
Proportion of Type II males:	9.4 %	7.1 %	9.0 %	13.3 %
Probability of bias in Type II male distribution towards nests containing females:	$p = 0.0001$	$p = 0.003$	$p = 0.008$	$p = 0.15$
Estimate of % spawnings parasitized in Tomales Bay (% females in nests also containing Type II males):	38 %	19 %	26 %	30 %

lengthy spawnings in aquaria with multiple Type II males participating. Further recordings were made when nesting Type I males were faced with a newly introduced Type I male 'intruder'. Subsequently, hydrophone recordings were made from aquaria containing only Type II males or only females.

Infra-red light sources were constructed from a 2-w incandescent bulb and a 760 nm infra-red filter (Hoya Optics; Model IR-76). This wavelength is short enough to transmit through water, yet too long to be sensed by the retina of *P. notatus* (CRESCITELLI 1991; LOEW, Cornell Univ., personal communication). Video images were recorded from an infra-red-sensitive CCD camera (Cohu Model 4815-2000) onto a video cassette recorder (Panasonic AG-2400). Acoustic signals were recorded with a hydrophone (made by C. W. CLARK, Cornell Univ. Lab. of Ornithology) onto a cassette tape recorder (Sony WM-D6C with Maxell UDXLII tape since the frequency response is virtually flat from 50 Hz to 16 kHz), from which the amplified signal was also recorded onto the videotape. Acoustic signals on the cassette tapes were analyzed for temporal and spectral characteristics on a Kay DSP Sonograph.

Results

Reproductive Behaviour: Field Observations

Census data from 630 nests (Table 2) demonstrated distinct nest-site use patterns by Type I and Type II males. Type II males and females were only seen in nests containing a Type I male; neither Type II males nor females were found to nest alone. By contrast, Type I males were usually observed as sole nest occupants, often with single or multiple clutches of eggs. The presence of Type II males in nests was strongly biased towards those nests that contained females (Table 2). The reverse interpretation is also logically possible but unlikely considering the results from the aquarium observations (below). The average annual proportion of Type II males among all males in the census was 9.6 %. The proportion of females in nests which also contained Type II males, one estimate of the frequency of spawning parasitism, ranged from 19—38 % (Table 2).

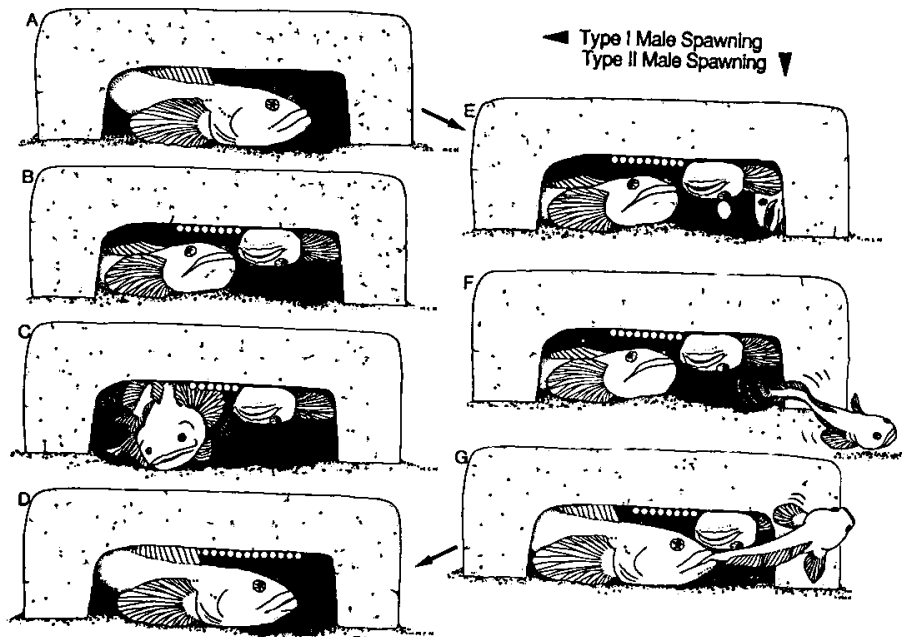


Fig. 2: Generalized spawning sequence of *Porichthys notatus* drawn from photographs. A. Type I male hums from his nesting territory after nightfall. B. A gravid female has entered the nest and deposits eggs on the ceiling of the nesting cavity. C. Type I male rolls and quivers, releasing milt near the eggs. D. After egg laying is completed, the female is ejected from the nest. Type I male guards the nest and hums to attract another female. E. Type II male inside the nest (far right) sneak-spawns and adopts the female egg-laying posture. F. Type II male outside the nest (far right) satellite spawns and releases milt while fanning water towards the nest opening. G. Nest-holding Type I male attacks a satellite-spawning Type II male

Reproductive Behaviour: Aquarium Observations

Aquarium observations allowed determination of Type I and Type II male patterns of nest occupancy, courtship, spawning behavior, and sound production. Figure 2 provides a pictorial overview of *P. notatus*' spawning behaviour that was taken from photographs of videotape frames such as are in Figure 3.

Nesting Behaviour. Type I males excavated cavern-like nesting territories beneath the tiles and cement blocks, expelling mouthfuls of sand and fanning sand out of the nest. While Type II males and females transiently appeared in vacant nest sites, they were never observed in nest sites for periods longer than 3 days. Type II males and females never excavated nests or excluded conspecifics from sites, even in aquaria without Type I males. When not swimming around the aquarium (typically at night) or directly engaged in spawning behaviour, Type II males and females remained buried in the sand.

Courtship and Spawning Behaviour of Type I Males. Shortly after dusk, Type I males moved to their nest entrances and, facing outward, began to repeatedly 'hum' (Figs. 2A, 3A). Both the rate and strength of gill contraction increased markedly during the vocalization, suggesting intense energy and respiratory demands. Gravid females were introduced to the observation tank either before dusk or after the night's humming began. Most gravid females swam rapidly about the tank when the male began humming (Fig. 3B). After appearing to locate the source of the hum, females freely swam towards, or charged into, the nest (Fig.

3C—E). Such 'willing' female nest entry was videotaped on 15 occasions. Spawning ensued in 8 cases; the female subsequently exited without spawning in the other 7 cases. After the female entered the nest, Type I males ceased humming, usually immediately, and always within 1 min. With the female inside the nest, the male then blocked the nest exit by extending his body sideways across the nest entrance, maintaining his head and body between the female and the entrance (Fig. 3F). Blocking ceased at the initiation of egg laying. Blocking was interspersed with lateral pressing: the Type I male aligned his body with the female's, and then pressed her against the side of the nest with a stereotypical, rhythmic bumping (1—3 s intervals) of his tail and body. Within minutes, the female assumed her distinctive egg-laying posture with her vent turned to the walls or ceiling of the nest cavity (Fig. 2B).

Type I males also attempted to drag or shove passing gravid females into the nest during brief periods of silence between hums. Many lunges at females resulted in misses, or in the female breaking free from the male's grasp. On four occasions, Type I males successfully forced females inside the nest, and then engaged in the usual pre-spawning behaviour including blocking of the nest entry. The female remained in the nest to spawn on one occasion and exited on three occasions.

Egg Laying and Fertilization. Egg laying lasted for up to 20 h after nocturnal initiation. This long duration suggests that, under natural field conditions, egg laying initiated during the evening of one day must extend at least into the low-tide period of the subsequent day. During egg laying, each large, 4—5 mm egg was slowly extruded through the vent onto the spawning substrate. Every few minutes during egg laying, the Type I male 'rolled and quivered', first pushing against the female, and then rolling his body towards her (Fig. 2C). At the apex of rotation he paused, briefly quivered, and then returned to the resting position. This behaviour appeared to be part of sperm release (fertilization is external): on one videotaped 'roll and quiver', a small cloud of milt was visible at the male's midsection. Periodically, the Type I male strongly bit the female on the side, at which point she became rigid and a low amplitude, grunt-like acoustic signal was sometimes recorded. When spawning was finally completed, the female turned right-side-up in the nest and appeared buoyant. The Type I male then ejected the female from the nest and always remained to guard the nest site (Fig. 2D). One Type I male resumed his humming 75 s after the spawned female's departure. Type I males continued this pattern of humming and long-term nest occupancy, multiple matings and nest maintenance, for up to 10 wk in captivity (when observations ended).

Although bioluminescence has been well documented in *P. notatus*, including a suggested role in courtship (CRANE 1965), it was not observed in this study. Bioluminescent capability has been reported to vary geographically with diet and body size in *P. notatus* (THOMPSON et al. 1988).

Reproductive Tactics of Type II Males. When Type II males were present, they congregated around the nest where the spawning activity was taking place. Two types of spawning behaviour were observed among Type II males: 'sneak-spawning' and 'satellite-spawning' (GROSS 1982); the same individuals were

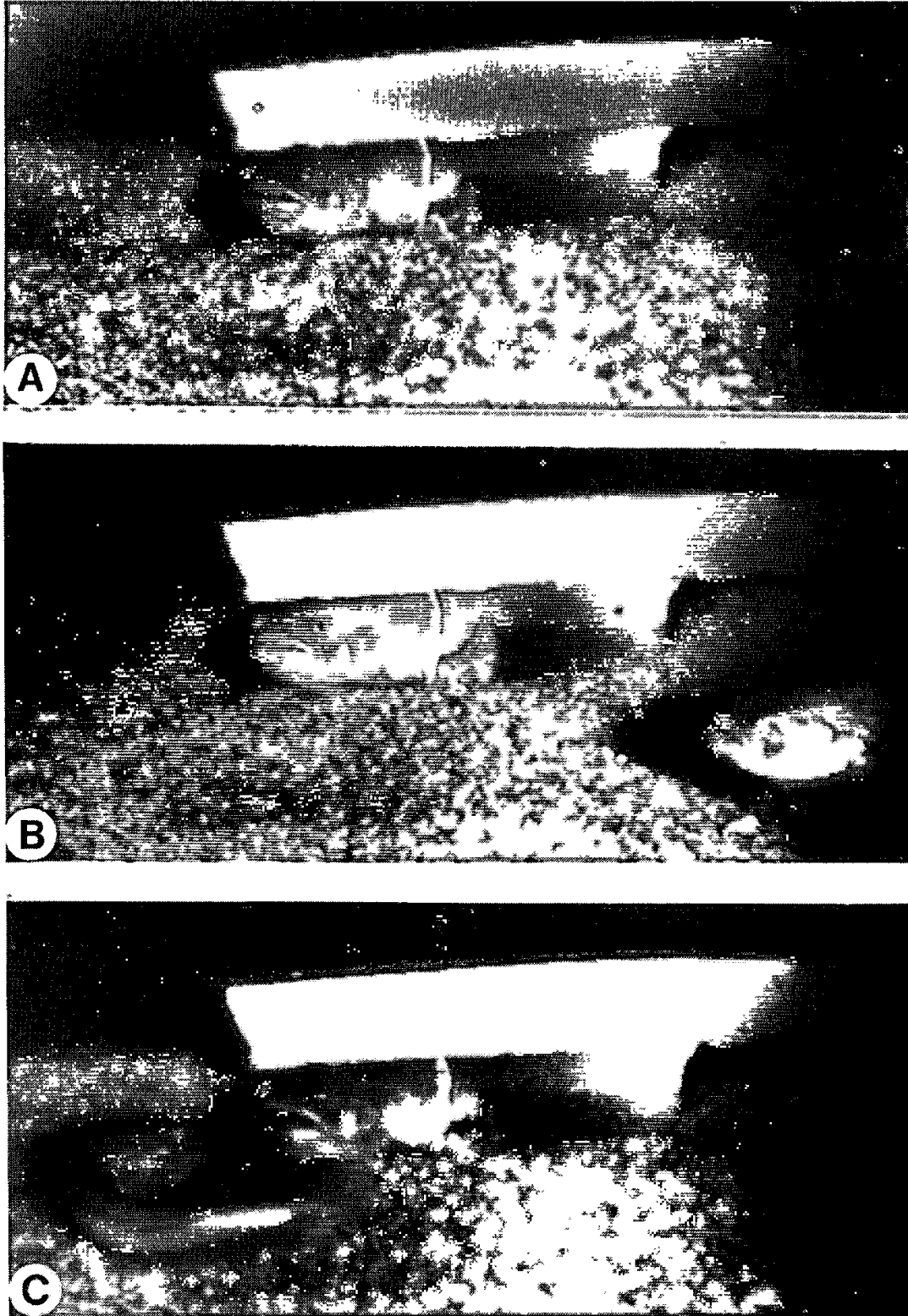
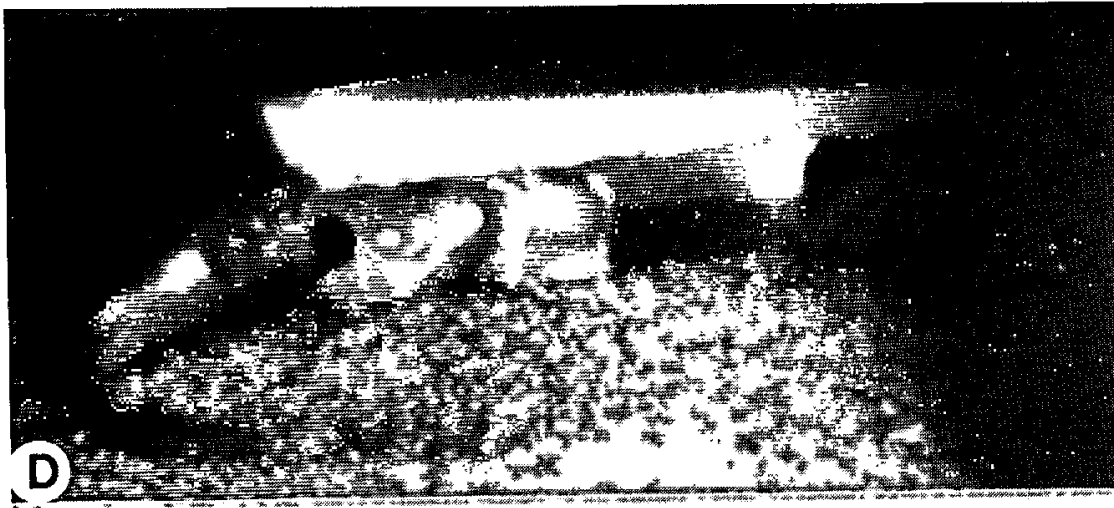


Fig. 3: 'Willing' female nest entry in *Porichthys notatus* as a Type I male hums; photographs from infrared videotapes. A. Type I male hums from entrance to his nesting cavity shortly after dusk. B. An introduced gravid female is visible at far right, approaching the nest. C. Female orients to the nest in darkness



D. Type I male opens his mouth and continues to hum. E. Type I male engulfs the head of the female in his mouth. His 'fangs' are embedded in the top of the female's head. His humming pauses as he bites down. F. After several more bites, the female has entered the nest. Type I male has ceased humming and blocks the female's exit

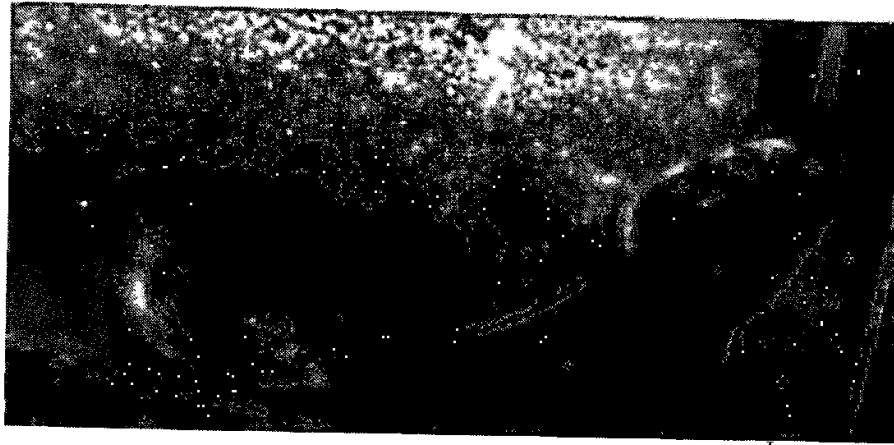


Fig. 4: Nest-holding Type I male attacks a satellite-spawning Type II male (see Fig. 2G). A spawning female (not visible) is inside the nest

observed to do both (Fig. 2E—G). A sneak-spawning, Type II male entered into the nest and either assumed a female-like posture with his own vent on the ceiling or side of the nesting cavity (Fig. 2E), or quickly burrowed into inaccessible nooks in the nest where he remained through the spawning or until his expulsion by the Type I male. By contrast, Type I males never assumed a female-like posture over four summers of observations. On 3 occasions, Type II males, while upside-down in the female-like posture, strongly bit the spawning female, as would a spawning Type I male.

Satellite-spawning Type II males lingered just outside the opening of the nest. Resting on the bottom of the tank, they swam backwards with their pectoral fins brushing against the sand as they moved close to the entrance of the nest. They then rolled slightly, vigorously rippled the anal fin, and fanned the tail (Fig. 2F). This behaviour momentarily set up a water current (a visible stream of aquarium detritus) into the nest. Release of a small amount of milt is believed to accompany this stereotyped fanning behavior. One observation, made at extremely close range, allowed a clear view of milt release — a faint line of dense white material was extruded from the vent, immediately dispersed by anal fin rippling, and fanned towards the nest. Such rippling and fanning was observed hundreds of times over the course of the study. Yet it occurred only when egg laying was taking place in the nest and only in proximity of the nest opening. Type II males also used any available openings in the sides or rear of the nest to insert their tails into the nest, and could remain in place for hours, with good proximity. When the Type II male was at close range, he was attacked by the Type I male (Figs 2G, 4).

Putative sneaking and satellite behaviour were seen in the field. Type II males were found both in the centre of the nest cavity with the spawning female(s) and the Type I male, and at the periphery of the nest rock, their bodies jammed into small nooks that were seemingly inaccessible to the Type I male. While these positions within the nest may change in response to falling water levels, the aquarium observations lead the authors to suspect that both sneak- and satellite-spawning tactics are used by Type II males in their Tomales Bay habitat.

When ripe females were held for days to weeks in aquaria lacking Type I males, they occasionally laid eggs inside an empty nest or on the tank wall. In all 3 of 3 such instances when Type II males were present, these males spawned (in 'female-like' posture, as described above) with the female, resulting in fertilizations and subsequent development of the young. Yet, in each instance, neither the Type II male nor the female remained to guard the eggs or nest afterward, in sharp contrast to the post-spawning, guarding behaviour that is typical of Type I males.

Male-Male Interactions. When a nest-holding Type I male faced either another Type I male or a Type II male, the nest-holding male made both open-mouth threats and repeated, regularly spaced sequences of grunts. Agonistic encounters between the Type I and Type II males during spawning were uniform in outcome. Nest-holding Type I males were observed during 17 spawnings to lunge at (37 times) and bite (53 times) spawning Type II males (Figs 2G, 4). By contrast, Type II males never bit or lunged at a Type I male. Type II males fled every attack, but promptly returned to the spawning.

There were two direct observations of mating competition between Type I males. In the first case, the spawning Type I male was forcibly ejected from his own nest by a second Type I male. The intruding male usurped the spawning male and the ejected male buried himself in the sand elsewhere in the tank. The intruder maintained his newly acquired nest and eggs until the experiment ended several weeks later, having abandoned his former egg-containing nest. In the second case from another aquarium, a Type I male left his own nest, entered a neighbouring nest with a spawning pair, pressed against the egg-laying female, and then rolled and quivered. In this case, the intruding male was expelled from the nest by the territory owner.

Acoustic Signals

Type I males. Two classes of signals were recorded from Type I males during behavioural observations (and also recorded from freeliving animals in Tomales Bay). These signals, hums and grunts (formerly called burps, BRANTLEY & BASS 1990), are distinguishable by their temporal characteristics. The hum is easily recognized by its extreme duration and unchanging spectral qualities (Fig. 5A–C) for a given temperature (Fig. 6A). This sound was easily attributable to Type I males because of the accompanying increase in gilling rate. The hum was produced almost entirely by those Type I males without females in their nests (Fig. 7A), and ranged in duration from 370 ms to almost 14 min (Fig. 5C).

Grunts – very regular, short duration (50–200 ms) sound pulses – were produced both singly and in regularly spaced sequences (Fig. 8A–C). Like the hum, production of sequences of grunts was also dependent on the context (Fig. 7B). Given the long duration of grunt sequences, determination of the identity of the individual producing the grunts was easily made because of the sounds' changing intensity as the Type I male moved about in the nest opening. In contrast, the identification of a particular individual with a single, short-duration grunt could not be made reliably.

Type II Males and Females. Type I males frequently vocalized when handled

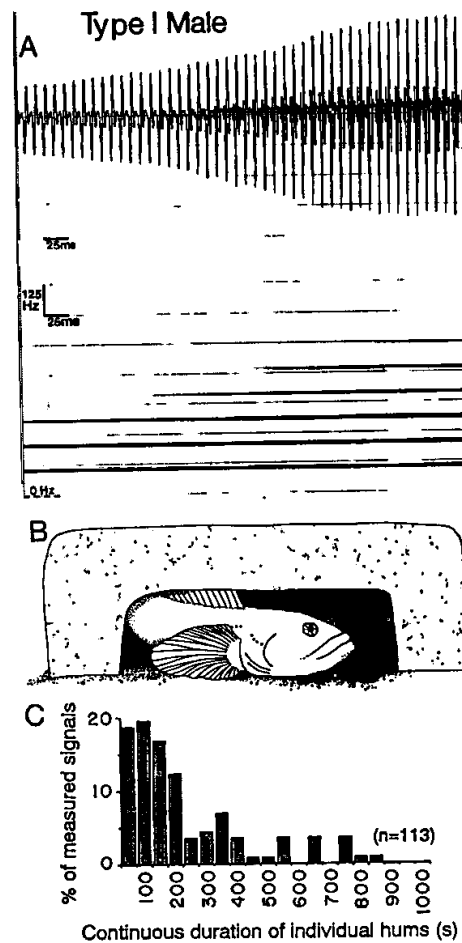


Fig. 5: An acoustic signal unique to *P. notatus* and to Type I males is the hum A. Oscillogram (above) and sonogram (below) records at the onset of a hum recorded in Tomales Bay. X-axis in both panels: 25 ms division; y-axis: relative amplitude for oscillogram and 125 Hz/division for sonogram; fundamental frequency of hum; 100 Hz. B. Hums are produced by unmated, nesting Type I males at night and appear to function as advertisement calls (see Fig. 7). C. Duration frequency histogram for pooled observations from multiple Type I males over portions of 14 nights

during a census (57 % of 120 fish sampled), but sounds were never heard from females (0 % of 116 fish) or Type II males (0 % of 75 fish). This observation originally led us to believe that these groups were incapable of generating sounds because of their small sonic muscles (BASS & MARCHATERRE 1989). However, recordings in aquaria demonstrated that both groups are indeed capable of vocalization. Hydrophones were placed in outdoor aquaria containing only Type II males or only females to permit attribution by sex. Video recordings were not made of these tanks because the electrical noise obscured the extremely low amplitude acoustic signals, hindering analysis. Low amplitude, single grunts (Fig. 9) were repeatedly recorded (typically 1–2 sounds/h from a tank containing 6–10 fish), about half of which had sufficient amplitude to permit determination of their fundamental frequency. At any given recording temperature, the grunts of Type II males and females had a similar fundamental frequency (Fig. 6B), as predicted by the earlier neurophysiological studies (BASS & BAKER 1990, 1991).

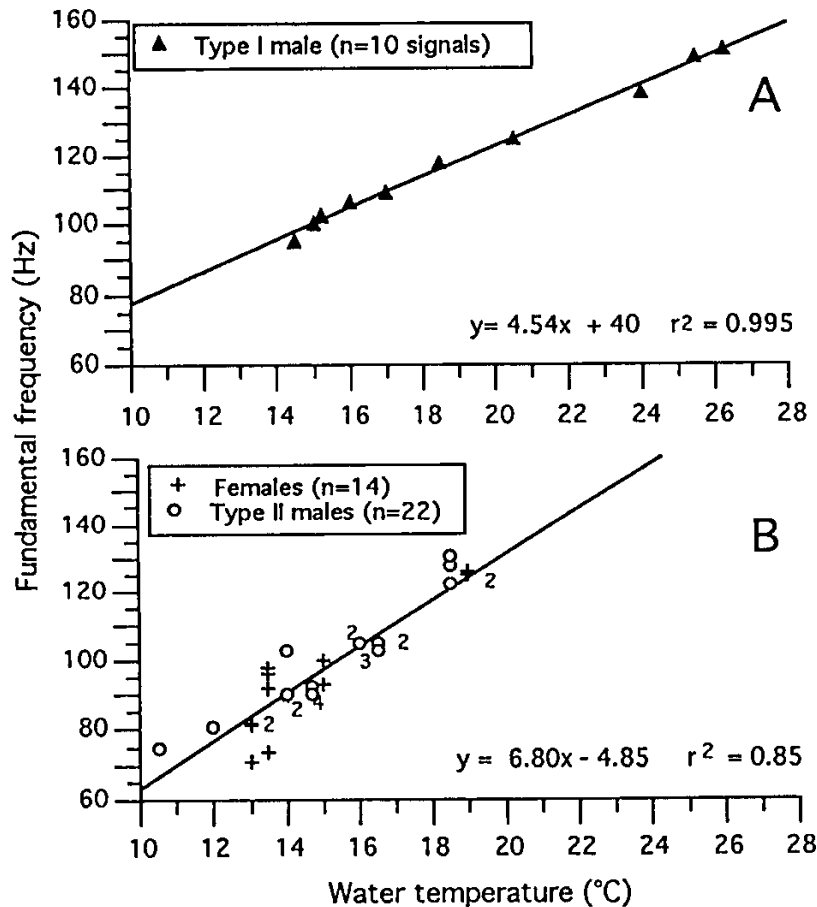


Fig. 6: Temperature affects the fundamental frequency of acoustic signals in *Porichthys notatus*. A. Fundamental frequency of the hum as water temperature was manipulated, based on recordings of one Type I male over a period of two nights. B. Temperature also affects the fundamental frequency of both female and Type II male grunts. These groups do not differ in temperature-adjusted fundamental frequency; regression line is for combined data sets. Numbers indicate multiple observations at the closest symbol

However, average grunt duration differed significantly (Student's *t*, $p < 0.0001$) between Type II males (143 ms, $n = 22$ signals) and females (230 ms, $n = 14$).

Videotapes of a Type I male and a female spawning contained occasional low amplitude grunts that sounded like those sounds recorded from isolated females. These grunts were produced when a female was bitten by a Type I male. However, their weak amplitude prevented the necessary spectral and temporal analyses to determine which individual was the sound source. No observations suggested that Type II males produced sounds while spawning.

Discussion

Dimorphic Males

This study now of the plainfin midshipman, *P. notatus*, shows that the divergence in somatic and neurobiological traits between Type I and Type II males is paralleled by a divergence in spawning tactics and acoustic signalling. The reproductive behaviour that characterizes Type I males — mate calling, pair-

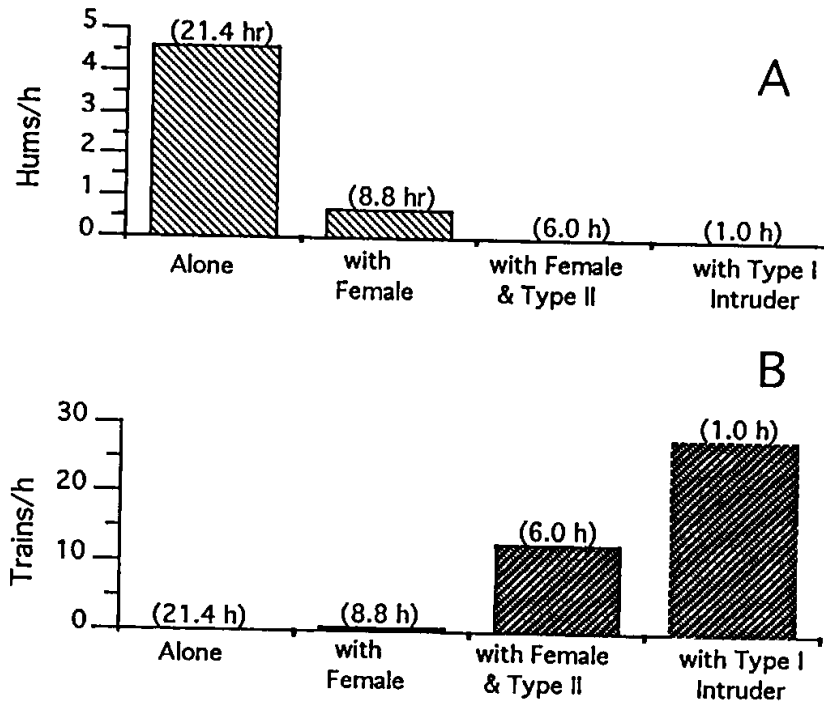


Fig. 7: The context of humming, A, along with the behaviour of gravid females, suggests its function as a Type I male mate call in *P. notatus*. By contrast, the context of sequences of grunts, B, and the interspersed open mouth threats (see Fig. 8), suggest a role in agonistic interactions. The hums in the category where there is only a female in the nest (with Female), briefly occurred moments after female nest entry. Numbers above bars: total hours of videotape analysed for each behavioural context. Type I male alone in nest; with Female = Type I male with female present in nest (pre-spawning and spawning); with Female & Type II = spawning with active Type II males present; with Type I Intruder = Type I male alone in nest while an unfamiliar Type I male has been introduced into the aquarium

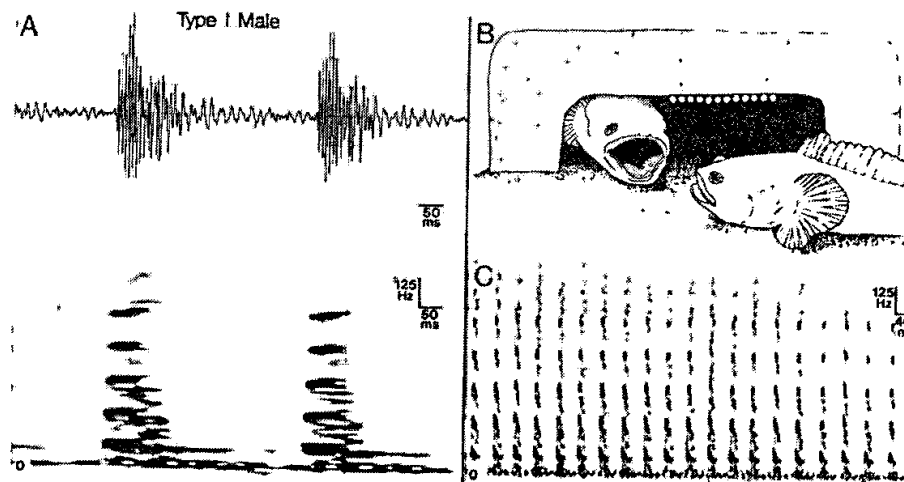


Fig. 8: A. Sequences of grunts generated by Type I male *P. notatus* appear to function as a threat display. Oscillogram (top) and sonogram (bottom) of Type I male *P. notatus* grunts taken from a repetitive sequence of grunts. X-axis for both panels: 50 ms/division. Y-axis: relative amplitude for oscillogram and 125 Hz/division for sonogram; fundamental frequency for the grunt is 125 Hz; some 60 Hz background noise is present. B. Nesting Type I male gives an open-mouth threat to an intruding Type I male. Sequences of grunts are interspersed with such threats and appear associated with defense of mating and/or territory. C. Grunt sequences: the same grunts as shown in A but on a larger x-axis at 400 ms/division. Sound pulses and intervening intervals are characteristically regular within a train sequence

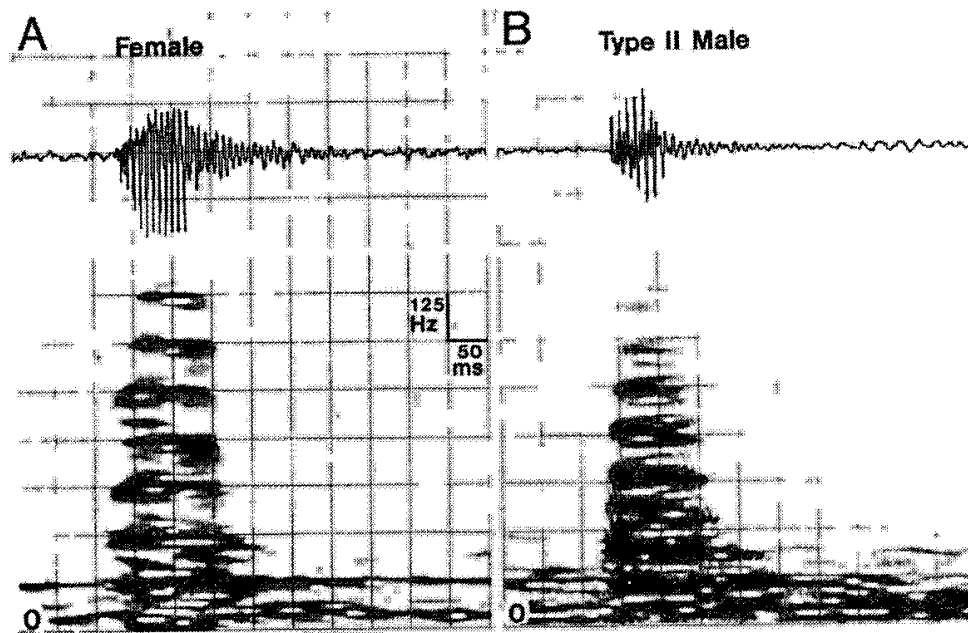


Fig. 9: Both Type II male and female *P. notatus* are capable of producing acoustic signals. A single, low-amplitude grunt was infrequently recorded at night in outdoor aquaria containing only females or only Type II males. Oscillogram (top) and sonogram (bottom) records of a female (A) and a Type II male (B) grunt

spawning, and nest-guarding — is absent in Type II males, as are the enlarged sonic muscles and increased body size associated with Type I male tactics. The non-courting, satellite- and sneak-spawning tactics exhibited by Type II males occur in other teleost fishes (BRANTLEY et al. 1993b). The apparently permanent behavioural differentiation of two male morphs in midshipman is comparable to that reported for courting and non-courting male morphs in sunfish, Pacific salmonids, and swordtails. These species also share a number of morphological, endocrinological and neurobiological traits (BASS 1993; BRANTLEY et al. 1993b). In other fishes with alternative tactics, individual males may freely switch tactics in response to the intensity of mating competition (BARLOW 1967; SIGURJONSDOTTIR & GUNNARRSON 1989; WIRTZ 1978), or may display age-dependent changes in tactics (JONES 1959; MAEKAWA 1983; ROBERTSON & WARNER 1978; WARNER & ROBERTSON 1978).

The authors have previously proposed that Type I and Type II male midshipman represent distinct ontogenetic pathways rather than part of a developmental sequence (BRANTLEY et al. 1993a). Developing into a Type II male may thus preclude mate calling for life if Type II males never express those specializations of sonic muscle fibres and central nervous system that permit prolonged humming (BASS & BAKER 1990; BASS & MARCHATERRE 1989; BRANTLEY et al. 1993a). Furthermore, Type II males lack the body size that increases the spawning success of Type I males through direct female preference and nest-site competition (DEMARTINI 1988; we recognize the nest-building males in his study as Type I males). The divergence in body size may reflect sexual selection for increased

body size on Type I males (DEMARTINI 1988) and the possible relaxation of such selection on Type II males that apparently neither compete for nest sites nor court females. Alternatively, small body size may be favoured among Type II males to avoid detection by Type I males or to spawn from small crevices within the Type I male's nest. Lastly, small body size may have no advantage *per se* and may merely reflect the life-history advantages of beginning reproduction at an earlier age and consequently undergoing maturation-related decreases in growth rate (BRANTLEY et al. 1993a; GROSS & CHARNOV 1980; WARNER & HOFFMAN 1980). Type II males appear to be obligate sexual parasites: even in tanks lacking Type I males, Type II males did not show parental nest-site fidelity to eggs they had fertilized. Becoming a Type I male, by contrast, does not preclude non-calling tactics such as forcing females into the nest or nest piracy. Only the potential for female mimicry is irrevocably lost by Type I males, due to increased body size, darkened colour, and increased fundamental frequency of acoustic signals.

Acoustic Signals

Vocal signals from several dozen families of fishes typically last for a few ms (FISH & MOWBRAY 1970). In contrast, Type I male midshipman generate continuous hums of up to 14 min which, at a fundamental frequency of 100 Hz at 12°, is equivalent to 84,000 sonic muscle contractions. This is a remarkable duration for a single acoustic signal in any vertebrate taxon. While IBARA et al. (1983) stated that individual midshipmen hum continuously for up to 1 h, their observation was based on a sampling of males at 2-min intervals for sound production. Observations of the context of male sound production and male-female courtship behaviour in our study support IBARA et al.'s (1983) hypothesis that the hum functions as a mate call. Shorter duration boatwhistles (about 500 ms) are generated by nesting male oyster toadfish (*Opsanus tau*) and apparently serve the same function (GRAY & WINN 1961).

How information is encoded in these acoustic signals is not clear. As fundamental frequency of the sounds varied with temperature, a temperature-coupling mechanism in the receiver is probably present, as found in the acoustic systems of other vertebrate and invertebrate species (BRENOWITZ et al. 1985; PIRES & HOY 1992). The fundamental frequency of the acoustic signals produced by several other teleosts has a systematic relationship to body size (LADICH et al. 1992) and so may provide information regarding an individual's identity. This, however, may not be the case for midshipman since neurophysiological studies do not demonstrate a comparable relationship (BASS & BAKER 1990). By contrast, temporal features such as the duration of the hum, grunt and grunt sequence or the inter-hum and inter-grunt intervals, may indicate a signaller's identity or behavioural state and thereby influence female mate preference or male-male interactions. Hums differ by 2–3 orders of magnitude in duration (see Fig. 5C), while grunt duration, inter-grunt interval, and grunt-sequence duration are also likely to vary markedly (unpublished observation). Alternatively, signals such as the hum may function only as beacons that indicate an individual's location, with mate choice determined by proximal interactions.

Grunts have been reported in midshipman from handled fish in a bucket, and from unseen fish in the field (COHEN & WINN 1967; fish we now recognize as Type I males). Given the observations that regular sequences of grunts are produced when an unfamiliar Type I male is present or when Type II males approach the nest during spawning, these sequences probably function as an acoustic threat. Similarly, nesting male *Opsanus tau* make single grunts in response to the presence of conspecifics, most often towards other males (GRAY & WINN 1961). COHEN & WINN's (1967) 'buzzes', which have an irregular duration between grunts and hums, but only under stressful circumstances, e.g., when Type I males were grasped and pulled from their underwater nests by hand, were also recorded.

Why are Type II males and females convergent in acoustic function and sonic muscle traits? Similarity of signals may result from deceptive mimicry of the female signal by Type II males or from a common role in agonistic encounters with conspecifics. Convergence of acoustic signals may also arise from an absence of selection in either females or Type II males for mate-calling ability — the energy costs of building and maintaining a large, red muscle unnecessary for Type II male reproduction are patently high.

Testis Size

Increased testicular size has been supposed to evolve in response to sperm competition (HARCOURT et al. 1981; WARNER & ROBERTSON 1978). Fertilization success in a spawning fish should reflect milt volume, proximity of release to the eggs, and the duration of sperm motility. Among many externally fertilizing fishes, the duration of sperm motility is extraordinarily brief (STOSS 1983), lasting seconds to minutes in *P. notatus* (BASS, BRANTLEY & CLARK unpubl. data). Thus, the frequent sperm release observed by both male morphs during spawning may reflect sperm competition, short sperm lifespan, and/or a time limit on the fertilization of each egg as it is laid. The authors' field observations indicate Type I males generally pair spawn, and, as seen in aquaria, they release milt in close proximity to the female. By contrast, Type II males appear to be obligate sperm competitors with the nesting Type I male, and, furthermore, with any other Type II males present. Given poorer proximity to the female, satellite-spawning Type II males also face sperm dilution and sperm mortality due to the time of travel and distance to each egg, hence an increased production of milt, suggested by larger testicular size, may help offset sperm mortality and sperm dilution. The sperm morphology of the *P. notatus* male types appear qualitatively similar (BASS & ANDERSEN 1991). There also appears to be no difference in the ability of Type I or II male sperm to fertilize eggs as both give rise to viable offspring.

Quantitative gonad size data are not available for other toadfishes. Qualitatively, however, the male gonadosomatic index in *Opsanus tau*, *Opsanus beta*, and *Porichthys myriaster* (all Batrachoididae) is similar to that of Type I male *P. notatus* (unpubl. data). The difference between relative testes size in Type II males (on average, 8.3 % of body weight) and Type I males (1.2 %) parallels those reported for the parasitic and courting morphs in other diandric species, e.g.,

Lepomis macrochirus: 4.6 % vs. 1.1 % (GROSS 1982); *Thalassoma duperrey*: 4.0 % vs. 0.18 % (HOURIGAN et al. 1991).

In conclusion, this study demonstrates the behavioral correlates of intrasexual dimorphism in body size, testis size, and vocal motor traits in *P. notatus*. The large sonic muscles of Type I males, but not Type II males (or females), suggested that acoustic signals would be elaborate only in the Type I male and would function in reproduction. By contrast, the enlarged testes of Type II males suggested that these males would exhibit an alternative reproductive tactic (BASS & MARCHATERRE 1989). Both predictions were confirmed. As male type appears constant throughout the life of the fish (BRANTLEY et al. 1993a), it is expected that reproductive tactics may also hold throughout the life of the fish. This raises the question of whether mixed evolutionarily stable strategies that are genetically determined might exist in *P. notatus*. Further study is necessary to identify the precise selective forces that maintain intrasexual divergence in male morphology, spawning behavior, and acoustic signals.

Acknowledgements

We thank Dr J. C. CLEGG, P. SIRI and the staff of the University of California Bodega Marine Laboratory for facilities and support; Dr V. CHOW and J. MARON for logistical and field support throughout the entire study; B. HORVATH for the videotape analyses for Figure 7; J. TSENG for assistance with acoustic analyses; M. MARCHATERRE for photographic assistance; Dr C. W. CLARK for providing hydrophones and technical support; M. NELSON for line drawings, and Drs E. ADKINS-REGAN, R. C. HARRISON, G. A. NEVITT, and P. W. SHERMAN for suggestions and improvements on earlier versions of the manuscript. Supported by pre-doctoral training grant NIMH 5T32MH15793, a Cornell Continuing Graduate Fellowship, Sigma Xi, The Raney Fund of the American Society of Ichthyologists and Herpetologists, NSF research grants BNS 8708559 and 9021563, and New York State Hatch Act NYC 1916423.

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Received: July 27, 1993

Accepted: January 7, 1994 (J. Brockmann)