

Thermal Tactics of Air-Breathing and Non Air-Breathing Gobiids Inhabiting Mangrove Tidepools on Pulau Hoga, Indonesia

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Air-breathing and non air-breathing gobiids inhabiting mangal tidepools on Pulau Hoga, Sulawesi, Indonesia, displayed differing types and degrees of behavioral and physiological adaptations to mitigate daily thermal and oxyc stress. Non air-breathing Dusky Frillfin Goby, *Bathygobius fuscus*, and an undescribed sandflat goby species, *Bathygobius* sp., had higher critical thermal maxima, CT_{Max}, (41.2 and 41.6 C, respectively) and lower critical thermal minima, CT_{Min}, (11.0 and 11.4 C, respectively) than amphibious Common Mudskipper, *Periophthalmus kalolo* (40.8 and 12.8 C, respectively). During hypoxia, Common Mudskipper CT_{Max} decreased significantly (38.8 C) and fish displayed a pronounced escape response, while Dusky Frillfin Goby showed no significant CT_{Max} decrease (40.8 C). Neither bathygobiid attempted to escape during CTM trials. Common Mudskipper, however, attempted escape at temperatures significantly lower than their normoxic and hypoxic CT_{Max} (36.4 and 35.0 C, respectively), and at temperatures higher than their normoxic CT_{Min} (18.7 C). Temperature quotients (ratio of metabolic rates or oxygen consumption at two temperatures) for all species approached unity and ranged from 1.00 to 1.37 when exposed to a temperature increase of 6 C. Oxygen insensitive CT_{Max} and a wide thermal scope allow bathygobiids to remain in tidepools during the most extreme conditions. In contrast, amphibious Common Mudskipper use a well developed behavioral escape response to avoid unfavorable environments. A sharply reduced temperature quotient response for these gobies diminishes the metabolic costs typically associated with diel temperature increases.

FISHES inhabiting the mangrove forest (mangal) tidepools around Pulau Hoga, southeast Sulawesi, Indonesia experience some of the most demanding diel temperature fluctuations seen in an aquatic habitat. Midday low tides isolate mangal fishes in shallow pools exposing them to rapid temperature increases from morning lows near 24 C to over 40 C by mid-afternoon (Dave Smith, pers. comm.). Challenges to fishes are further exacerbated as dissolved oxygen concentrations fall with rising temperature and increasing salinity. Despite these harsh conditions, the area supports a diverse gobiid fauna. Gobies, the most successful marine teleost family (Nelson, 1994), make up more than 60% of fishes inhabiting shallow intertidal habitats around Pulau Hoga (Taylor, unpubl. data). Two subfamilies of gobiids dominate these mangal tidepools. Subfamily Oxudercinae is represented by Common Mudskipper (*Periophthalmus kalolo*), an amphibious species that spends most of its time emerged and is resident to the mangal area. Subfamily Gobiinae is represented by several non air-breathing members of the genus *Bathygobius*; the two most common are the Dusky Frillfin Goby (*Bathygobius fuscus*), and a small, previously undescribed *Bathygobius* species (James Van Tassell, pers. comm.) commonly observed in mangal sandflats.

Both goby subfamilies are specialists possessing a number of tactics that allow them to tolerate the rapid and extreme temperature fluctuations common in their environment.

It may be expected that different physiological and behavioral adaptations exist to allow amphibious and non air-breathing gobies to coexist in Hoga's mangal habitats. The capacity to breathe air allows mudskippers to behaviorally exploit more benign habitats by shuttling between air and water (Clayton and Snowden, 2000) or perhaps take advantage of evaporative cooling during times when thermal conditions in pools become intolerable (Tytler and Vaughan, 1983). In addition to behavioral responses, some mudskippers may exhibit relatively high thermal tolerances (Stebbins and Kalk, 1961; Gordon et al., 1969; Clayton and Snowden, 2000) and the ability to offset metabolic costs (Gordon et al., 1978) as temperatures in their environment increase. Non air-breathing gobies, unlike mudskippers, have few behavioral options and may rely more heavily on physiological tactics to endure hyperthermic and hypoxic mangal tidepool conditions. Therefore, it is likely that metabolic and thermal tolerance responses are better developed in non air-breathing than air-breathing gobies from the tropics, but without direct comparative data, the type and extent of

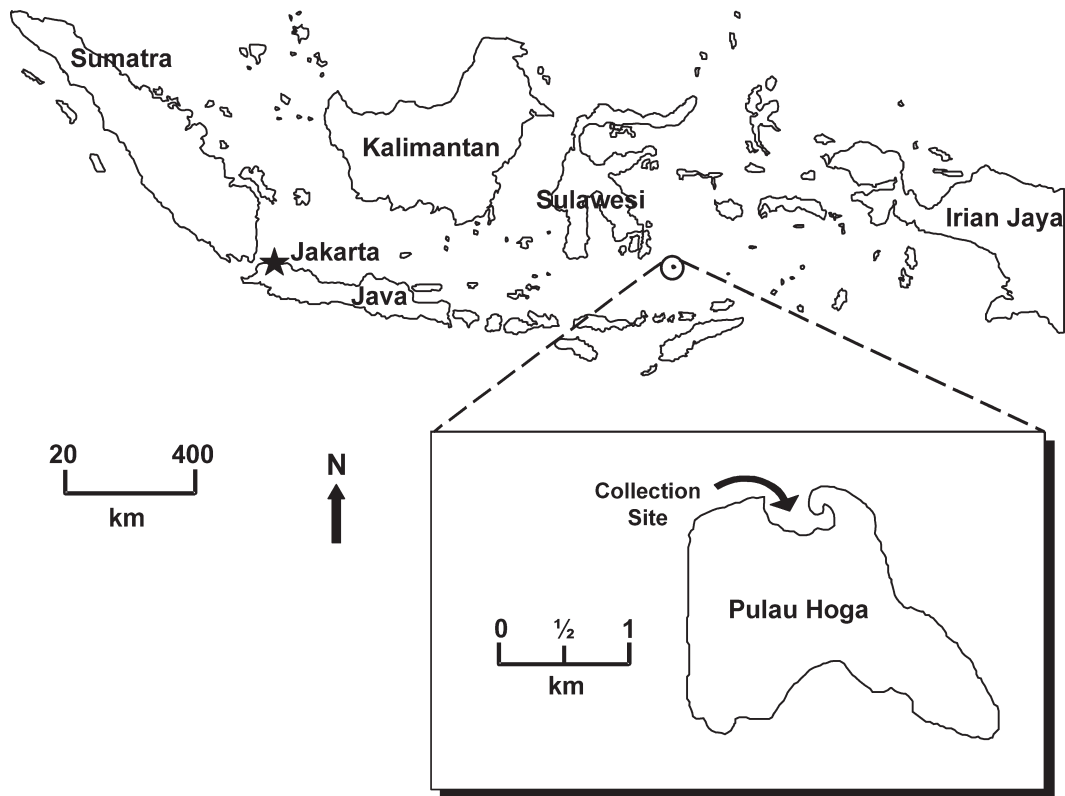


Fig. 1. Gobiid mangal tidepool collection site (exposed at low tide) Pulau Hoga, Republic of Indonesia.

physiological adaptation (i.e., short term adjustment to novel conditions) remains unknown.

Previous studies have quantified individual hyperthermal tactics of selected tropical gobiids but overlooked interactive physiological and behavioral adaptations that contribute to a species' overall thermal survival strategy (Stebbins and Kalk, 1961; Gordon et al., 1978; Clayton and Snowden, 2000). While both thermal tolerance and metabolic responses can provide useful insights into behavioral or physiological processes, alone they may not fully explain the complex relationship fish have with their thermal environment. Furthermore, attempts to integrate data from multiple studies can be confounded by differences in methodology or experimental endpoints. We tested the hypothesis that physiological thermal adaptations used by non air-breathing Dusky Frillfin Goby and sandflat goby (our common name for the undescribed species) differ markedly from those of air-breathing Common Mudskipper. Specifically, we quantified upper and lower temperature tolerance, escape behavior, and metabolic responses for these species and assessed the effects of hypoxia on upper tolerance and escape temperatures for

Dusky Frillfin Goby and Common Mudskipper. Our study is the first to utilize standardized methodology to compare the range of physiological thermal adaptations seen in sympatric gobies from equatorial mangal environments.

MATERIALS AND METHODS

Habitat characterization.—Fishes in our study inhabited mangal tidepools located on the northwest edge of Pulau Hoga ($05^{\circ}27.53'S$, $123^{\circ}46.33'E$, Fig. 1). The substrata were largely coral sand and rubble with no macrovegetation. Pools were characterized at low tide on 17 daytime and eight nighttime occasions between 20 June and 10 August in 2002 and 2003. On each occasion, depth (cm), temperature (± 0.1 C, NIST calibrated mercury thermometer), salinity ($\pm 1.0\%$, calibrated Fisherbrand[®] refractometer), and dissolved oxygen (± 0.1 mg/L, calibrated YSI 55-oxygen meter) were collected from each of ten pools selected at random.

Collection and care of fishes.—Common Mudskipper, Dusky Frillfin Goby, and sandflat goby were collected using aquaria dip nets and transported

to Hoga Marine Research Centre (HMRC) in 20-L buckets. A bright light was used to immobilize mudskipper during nighttime collections. Mudskipper were housed with pieces of palm fronds and rock to allow fish free access to air. Both non air-breathing fishes were kept at HMRC in 190-L holding tanks with filtered seawater ($33 \pm 2\%$) at average temperatures similar to those experienced in the mangal environment (approximately 24–34 C). All aquaria were biologically filtered, moderately aerated, and water was changed daily to assure good water quality. Fish were not fed during captivity because all experimental trials were completed within 48 h. Following experimentation, all fish were released at their site of capture.

Normoxic and hypoxic temperature tolerance.—The Critical Thermal Methodology (CTM) estimated temperature tolerance as critical thermal maximum, CTMax, or critical thermal minimum, CTMin, in experimental trials (Cowles and Bogert, 1944). During CTM trials, fish were exposed to constant water temperature increases or decreases slow enough to track body temperature, but fast enough to prevent thermal acclimation, until final loss of equilibrium (LOE; inability to maintain dorsoventral orientation for at least one minute) was reached (Cox, 1974; Becker and Genoway, 1979; Beitinger et al., 2000). Although some authors prefer to use onset of muscle spasms (OS) as the CTM endpoint (Paladino et al., 1980), not all fishes in our trials displayed OS during CTMax trials, and no fish exhibited OS during CTMin trials. For each CTMax or CTMin trial, fishes were randomly selected from holding tanks and placed into 250-ml Nalgene® beakers containing clean, fully aerated seawater at acclimation temperature (26–29 C). Beakers were then suspended in a 35-L recirculating water bath. Water bath temperature was increased or decreased at a rate of 0.21 C/min using a 1500 W immersible heater connected to a temperature controller (Electrothermal® MC242) or a chiller (New Ocean Model #AQ-E3, 1/4 HP) during CTMax or CTMin trials, respectively. Following LOE, water temperature (± 0.1 C, calibrated Fisherbrand® NIST mercury thermometer), standard length (± 0.5 mm), and wet mass (± 0.1 g) were recorded, and fishes were returned to pre-trial conditions to recover. In addition to final LOE, escape response temperatures were recorded if the fish attempted to escape the beaker and persisted in this activity for at least one min. Critical thermal maxima, minima, and escape response were quantified as the arithmetic mean of the collective endpoint temperatures for each species. Thermal scope for each species was then

calculated as the difference between mean CTMax and CTMin values (Bennett and Beitinger, 1997).

We also quantified the effects of low dissolved oxygen on CTMaxima and escape response of Dusky Frillfin Goby and Common Mudskipper. Data were collected by exposing Common Mudskipper and Dusky Frillfin Goby to simulated hypoxic mangal conditions using a modified version of the counter-current oxygen depletion system described by Smale and Rabeni (1995). For these experiments, we placed up to five fish, one each, into 500-mL Erlenmeyer flasks filled with 100% saturated seawater. A Supreme Mag Drive® model MD12 pump continuously recirculated water between the test chambers and a polyvinyl chloride counter-current exchange column (180 cm \times 5.1 cm OD). Compressed nitrogen, forced through a fine-bubble diffuser stone at 1.5 L/min, generated a bubble stream in the exchange column. Counter-current nitrogen/water flow in the exchange column reduced dissolved oxygen in the system by approximately one-half every 18 minutes as oxygen diffused into the nitrogen bubble stream and was carried out of the system. Water was recycled through the system until an experimental hypoxic level similar to commonly observed minimum dissolved oxygen values (2.1 ± 0.60 mg/L) was reached. After reaching hypoxic conditions, CTMax trials were conducted as previously described and both LOE and escape response recorded.

Aquatic respirometry.—We used sealed-flask respirometry (Cech, 1990) to estimate resting routine oxygen consumption (expressed as mg O₂·h⁻¹) for Dusky Frillfin and sandflat goby at diel temperature extremes of 26 C (typical daily low temperature) and 32 C (typical daily high temperature). Each fish in a trial was transferred from the holding tank into an aerated 20-L plastic antechamber containing filtered seawater set in a 500-L water bath maintained at 26.4 C (SE \pm 0.06) with EboJager 300 W heaters. An Aquatic Ecosystems® powerhead vigorously circulated seawater ($33 \pm 2\%$) in the bath to prevent thermal stratification. Following a minimum one hour exposure at 26 C, fish were taken from the antechamber and individually sealed into 500-mL glass (Erlenmeyer) respirometry flasks filled with clean 26 C seawater from the water bath filtered with 63- μ m mesh plankton netting. Seawater from the same respirometry flask source was also collected in a 300-ml BOD bottle and initial dissolved oxygen content determined (± 0.05 mg/L at standard temperature and pressure) via standard Winkler titration (Cox, 1990). Respirometry flask volume always equaled or exceeded fifty times that of the fish

(Morris, 1961; Cech, 1990). Respirometry chambers containing fish were then suspended in the water bath until 10 to 30% of flask oxygen content was consumed—typically between 0.5 and 2.5 hours, depending on fish mass. At the end of each low temperature trial, the respirometry flask was removed from the water bath, 300 ml BOD of flask water collected, final dissolved oxygen content determined, and total elapsed time recorded. Fish were then returned to the antechamber and transferred to a second water bath heated to 32.2 C (SE \pm 0.11) for high-temperature trials. Fish remained in the 32 C antechamber for at least one hour to ensure their body temperature reached the higher temperature, after which data collection for sealed flask respirometry proceeded as described above. Fishes in both high- and low-temperature trials were carefully observed, and trials in which individuals displayed prolonged or abnormally high activity levels were omitted from data analysis.

Following respirometry trials, fish were weighed (wet mass \pm 0.1 g), standard length determined (\pm 0.5 mm), and resting routine oxygen consumption for high and low temperature trials calculated using the following equation:

$$\text{Oxygen consumption} = ((\text{Initial O}_2 - \text{Final O}_2) \div \text{Elapsed time}) \times \text{Flask volume}$$

Adjustments were made for bacterial and planktonic oxygen consumption by subtracting mean consumption estimates measured in control respirometry flasks (similar in all respects, but containing no fish) from total oxygen consumption calculated for each individual fish. Control respirometry flasks were used during each trial.

Aerial respirometry.—Resting routine oxygen consumption in air was estimated for two separate groups of Common Mudskipper at approximately 26 C and 32 C and values converted to standard temperature and pressure. Captured mudskipper were held between 24 and 72 hours prior to undergoing experimental trials. Oxygen consumption was determined using a modification of Cech's (1990) manometric respirometry method. The respirometer consisted of a 500-ml Erlenmeyer test flask containing a single fish and connected by a manometer tube to a 500-ml Erlenmeyer reference flask. The manometric apparatus was submerged in a constant-temperature water bath maintained at the set point temperature \pm 0.5 C using Penn-Plax® 50-Watt aquaria heaters. The apparatus was pressure-tested prior to each trial to identify and

eliminate leaks in the system. Potassium hydroxide-saturated filter paper, suspended from the flask stopper, was used to absorb respired carbon dioxide from the test flask. After a one-hour holding period, oxygen uptake estimates were made every 0.5-hour for 2.5 hours. Respirometer flask pressure decreased as the fish consumed oxygen, causing manometer fluid displacement proportional to oxygen uptake. The absorbed volume was replaced by injecting air via a 10 cc gas-tight glass syringe. Air was added in small microliter volumes (100–200 μ l) at frequent intervals during the trial to allow any heat from gas compression to be dissipated quickly into the water bath heat sink. Injected air volume and elapsed time were recorded during each trial. Following respirometry trials, fishes were weighed (wet mass \pm 0.1 g, Mettler Toledo BD202), standard length (mm) determined, and resting routine oxygen consumption was determined for each fish using the following equation:

$$\text{Oxygen Consumption} = (\text{O}_2 \text{ consumed (mL)} \div \text{Time in flask (h)}) \div 0.698$$

Statistical analyses and Q₁₀ determinations.—Critical thermal maxima and minima were tested for statistical differences between species using one-way Analysis of Variance (ANOVA) or an independent t-test. Interspecific statistical comparisons of normoxic CTMax values were made by ranking LOE responses prior to ANOVA because data were non-normally distributed. Where significant differences were found, a Student-Newman-Keuls multiple range test (SNK) was used to identify statistical relationships between means. An independent t-test was used to test for statistically significant differences between normoxic and hypoxic CTMaxima and escape responses of Common Mudskipper and Dusky Frillfin Goby.

In order to account for possible body mass effects on oxygen consumption, Analysis of Covariance (ANCOVA) was used to interpret all respirometry data, with Log₁₀-transformed oxygen consumption as the response and Log₁₀-transformed body mass as the covariate. An ANCOVA was performed for Common Mudskipper respirometry data, whereas repeated-measures ANCOVA was performed for Dusky Frillfin and sandflat goby respirometry data. Least Square Mean (LSM) oxygen consumption values for all species were untransformed prior to calculating Q₁₀ values. Temperature quotients were calculated using the following equation, where untransformed LSM of oxygen consump-

TABLE 1. SUMMARY OF WATER CHEMISTRY VALUES OBTAINED FROM MANGAL TIDEPOLS ON HOGA ISLAND DURING LOW TIDE ON 17 DAYTIME AND EIGHT NIGHTTIME HABITAT CHARACTERIZATION EVENTS. During each characterization, water chemistry variables were collected from ten mangal tidepools selected at random. Mean values reflect the grand mean from all sampling events. Minimum and maximum values represent the highest and lowest values recorded during the entire study period.

Parameter	Day			Night		
	Mean (SD)	Min	Max	Mean (SD)	Min	Max
Temperature (C)	30.4 (1.90)	27.8	35.6	25.8 (1.18)	24.2	27.3
Dissolved oxygen (mg/L)	7.6 (2.63)	1.2	12.3	2.6 (1.11)	1.1	5.1
Salinity (‰)	35 (3.0)	30	44	35 (4.0)	31	47

tion estimates are designated R_1 and R_2 at ~ 26 C (T_1) and ~ 32 C (T_2):

$$Q_{10} = (R_2 \div R_1)^{10 \div (T_2 - T_1)}$$

All statistical decisions were based on an alpha equal to 0.05.

RESULTS

Habitat characterization.—Maximum differences between lowest and highest values measured for temperature, dissolved oxygen, and salinity observed during the study were 11.4 C, 11.2 mg/L, and 17.0‰, respectively (Table 1). Hoga experiences tidal amplitudes ranging from three to

four meters. As a result, a tidepool's depth could range from 0.002 m to 1.2 m over a single tidal cycle. Variations in depth were greatest during spring and lowest during neap tides.

Thermal physiological responses to hypoxia.—Fishes in our study exhibited CTMax values exceeding 40.6 C and CTMin values of less than 12.9 C (Table 2). Critical thermal maxima values of Dusky Frillfin and sandflat goby were not significantly different (ANOVA on ranked data; $df = 2,48$; $F = 4.59$; $P = 0.015$) however, both species were significantly more tolerant of high temperatures, i.e., had a higher CTMaxima, than Common Mudskipper (SNK, $\alpha = 0.05$). Likewise, the CTMin value for Common Mudskipper was

TABLE 2. DYNAMIC TEMPERATURE TOLERANCE (CTMAX; CTMIN) VALUES UNDER NORMOXIA AND HYPOXIA FOR FIELD-ACCLIMATED COMMON MUDSKIPPER, DUSKY FRILLFIN GOBY, AND AN UNDESCRIBED SANDFLAT GOBY. Standard lengths, wet masses, and escape temperatures are also reported.

Species	N	Standard length (mm)	Wet mass (g)	CTM (C)	Escape response (C)
Common name	Mean (SD)	Mean (SD)	Mean (SD)	Mean (SD)	
Normoxic CTMax					
<i>Periophthalmus kalolo</i> Common Mudskipper	17	41.2 (31.02)	4.1 (3.10)	40.7 (0.48)	36.4 (1.63)
<i>Bathygobius fuscus</i> Dusky Frillfin Goby	25	29.9 (8.97)	0.8 (0.50)	41.2 (0.87)	None
<i>Bathygobius</i> sp. undes. sandflat goby	11	24.6 (4.35)	0.3 (0.19)	41.6 (0.70)	None
Normoxic CTMin					
<i>Periophthalmus kalolo</i> Common Mudskipper	15	71.7 (13.12)	6.5 (3.86)	12.8 (0.33)	18.7 (2.93)
<i>Bathygobius fuscus</i> Dusky Frillfin Goby	10	31.7 (5.63)	0.9 (0.41)	11.0 (0.56)	None
<i>Bathygobius</i> sp. undes. sandflat goby	8	26.6 (3.86)	0.3 (0.07)	11.4 (0.54)	None
Hypoxic CTMax					
<i>Periophthalmus kalolo</i> Common Mudskipper	17	71.3 (12.25)	6.2 (3.17)	38.7 (0.79)	35.0 (1.41)
<i>Bathygobius fuscus</i> Dusky Frillfin Goby	6	24.3 (6.56)	0.6 (0.53)	40.8 (0.94)	None

TABLE 3. OXYGEN CONSUMPTION RATES FOR DUSKY FRILLFIN GOBY (*Bathygobius fuscus*), AN UNDESCRIBED SANDFLAT GOBY (*Bathygobius* sp.) AND COMMON MUDSKIPPER (*Periophthalmus kalolo*) DETERMINED AT 26.4 (SE \pm 0.06) AND 32.1 (SE \pm 0.11) C. Oxygen consumption estimates reflect Least Square Mean (LSM) values obtained from ANCOVA (Common Mudskipper) or repeated-measures ANCOVA (bathygobiids) using Log₁₀-transformed oxygen consumption rates. Q₁₀ values were calculated using untransformed LSM values.

Species	n	Wet mass (g)	Standard Length (mm)	mg O ₂ ·hour ⁻¹ at ~26 C	mg O ₂ ·hour ⁻¹ at ~32 C	Q ₁₀
Common Name		Mean (SE)	Mean (SE)	LSM (SE)	LSM (SE)	
<i>Bathygobius fuscus</i>						
Dusky Frillfin Goby	25	0.91 (0.087)	31.2 (1.00)	-0.473 (0.0252)	-0.471 (0.0212)	1.00
<i>Bathygobius</i> sp.						
undes. sandflat goby	23	0.34 (0.047)	27.1 (1.05)	-0.621 (0.0270)	-0.599 (0.0228)	1.10
<i>Periophthalmus kalolo</i>						
Common Mudskipper	24	6.63 (0.564)	67.20 (2.292)	0.158 (0.0352)	0.228 (0.0418)	1.37

significantly higher than values determined for Dusky Frillfin and sandflat goby, which did not differ significantly from each other (ANOVA on ranked data; $df = 2,30$; $F = 53.61$; $P < 0.0001$; SNK, $\alpha = 0.05$). Dusky Frillfin and sandflat goby had a thermal scope (30.2 C) over 2 C greater than Common Mudskipper (27.9 C; Table 2).

Common Mudskipper were more sensitive to hypoxia than Dusky Frillfin Goby. Under low oxygen conditions, Dusky Frillfin Goby exhibited a significantly higher CTMax than Common Mudskipper (independent t-test, $P < 0.0001$; Table 2). Furthermore, the CTMax of the strictly aquatic Dusky Frillfin Goby showed no response to perturbations in dissolved oxygen level (independent t-test, $P = 0.3492$); whereas, amphibious Common Mudskipper CTMax decreased significantly when fish were exposed to hypoxic conditions (independent t-test, $P < 0.0001$; Table 2).

Not surprisingly, the strictly aquatic gobiids made no attempt to escape the test chambers during CTMax trials as temperatures neared lethal limits, even when oxygen tensions were reduced (Table 2). Common Mudskipper displayed a marked escape response at 36.4 ± 1.63 C during normoxic CTMax trials, a response that occurred at significantly lower temperatures (35.0 ± 1.41 C) when the fish were exposed to hypoxic conditions (independent t-test, $P < 0.001$). Interestingly, Common Mudskipper exposed to progressively decreasing temperatures during CTMin trials also showed an obvious escape response at 18.7 ± 2.93 C, conditions that are probably infrequent in equatorial intertidal habitats.

Respirometry.—No gobiids in our study showed a marked increase in metabolic expenditure when exposed to a common increase in daily

temperature (Table 3). Dusky Frillfin, sandflat goby, and Common Mudskipper displayed Q₁₀ responses of 1.10, 1.00, and 1.37, respectively. All gobies in the experiment appeared to respire normally with no obvious decrease in ventilatory rates during experimentation. No large changes in dissolved oxygen were observed in control respirometry flasks during aquatic respirometry trials. In aerial respirometry trials, no abnormal or rapid fluctuations of manometer fluid were observed that would indicate pressure or temperature effects on experimental measures.

DISCUSSION

Hoga's resident tidepool gobiids experience dramatic fluctuations in depth, temperature, salinity, and oxygen. Diel thermal shifts greater than 10 C as well as oxygen saturations of less than 20% were common during the course of our studies (Table 1). Intertidal fishes found in seagrass and mudflat habitats outside the mangrove forest may experience similar abiotic changes; however, these fishes have access to deeper, cooler environments. Dusky Frillfin and sandflat gobies continuously occupy exposed, shallow pools with no reprieve from the highest midday temperatures. Even the amphibious Common Mudskipper, which has the ability to leave tidepools, may encounter potentially lethal high air temperatures when emerged. Successful exploitation of mangal habitats by these fishes clearly depends on behavioral and physiological responses to effectively ameliorate concomitant thermal and hypoxic stress.

Non air-breathing gobies rely primarily on well developed physiological adaptations, as they are unable to abandon hyperthermic tidepools dur-

ing low tide. Bathygobiids in our study showed remarkably high CTMax values approaching the biokinetic limits for vertebrate life. Extreme CTMaxima are rare in fish, with only 14 other species known to have values exceeding 41 C (see reviews by Clayton, 1993; Beitinger et al., 2000). The highest CTMax values recorded to date are reported for Belize pupfish, *Cyprinodon artifrons*, from mangal habitats (45.3 C; Heath et al., 1993) and sheephead minnow, *C. variegates*, from south Texas salt marshes (45.1 C; Bennett and Beitinger, 1997). In both cases, pupfish were acclimated to diel cycling temperatures peaking at approximately 40 C. Field-acclimatized Dusky Frillfin and sandflat goby each had considerably lower CTMaxima (> 41 C) than those reported for pupfish (Heath et al., 1993; Bennett and Beitinger, 1997), but higher CTMax values than the subtropical Shimofuri Goby (37 C; Matern, 2001). The differing responses are due in part to dissimilar thermal acclimation regimens; however, thermal ecology likely plays an important role in dictating final tolerance levels as well. The relatively low CTMax observed in Shimofuri Goby is indicative of fish living in cooler habitats where water temperatures never approach 40 C (Beitinger et al., 2000). In contrast, the remarkably high tolerance levels seen in pupfishes protect them from extreme and often unpredictable temperature spikes that are common in their environment. Likewise, the high CTMax and broad thermal scope are effective survival tactics for Dusky Frillfin and sandflat goby living in hyperthermic tidepools, but the predictable nature of environmental temperature changes minimizes the need for tolerance values much beyond the highest daily peaks. It is also interesting to note that marked hypoxia, a condition these fishes must experience daily, has no effect on their high temperature tolerance. Thermal physiology in these bathygobiid fishes reduces dependence on behavioral thermoregulatory adaptations and explains the lack of an escape response as water temperatures were increased during CTM trials.

Unlike the bathygobiids, Common Mudskipper depend heavily on behavioral responses to ameliorate hypoxic and hyperthermic stress. The strong escape response observed in Common Mudskipper complements the fishes' ability to move to cooler pool environments or exploit evaporative cooling on land (Tytler and Vaughan, 1983). When high temperature stress occurs in conjunction with low oxygen levels, escape and CTMax response temperatures are significantly reduced (Table 2). Interestingly, decreases in both values are approximately equal, allowing the fish to maintain similar margins of

safety in oxygenated as well as hypoxic water. Prominent decreases in CTMax and escape response temperatures argue that oxygen level is a major directing factor for Common Mudskipper resident to tidepool environments.

Previous mudskipper thermal tolerance studies report non-standardized tolerance endpoints, or resistance rather than tolerance measures. Consequently, upper tolerance limits in this group are vague (Clayton, 1993). Normoxic CTMax of Common Mudskipper in our experiments (40.7 C) were slightly lower than values reported by Stebbins and Kalk (1961) for Barred Mudskipper, *P. argentilineatus* (41.5 C). Common and Barred Mudskipper have a sympatric distribution, and one might expect similar CTMax values. The differing tolerance values between Common Mudskipper in our trials and Barred Mudskipper may reflect the fact that cessation of ventilation, which typically occurs at temperatures higher than LOE, was used as an endpoint for the Barred Mudskipper studies (Stebbins and Kalk, 1961). At low oxygen levels, Common Mudskipper show a 2 C decrease in CTMaxima from normoxic estimates, suggesting they are not as physiologically well suited to hypoxia as bathygobiids in our study. Gordon et al. (1978) likewise report that Shuttles Hoppfish, *Periophthalmus modestus*, displays a limited hypoxia tolerance indicating that pronounced sensitivity to low oxygen levels might be a widespread phenomenon in this group.

Diel temperature changes similar to those occurring in Hoga's tidepools (approximately 10 C) typically result in a doubling of fish metabolic rate; indeed, Q_{10} values below 2.0 are rare in fish (Bullock, 1955; Vondracek et al., 1982; Miklos et al., 2003). Both air-breathing and non air-breathing gobies in our experiment violated this principle by displaying unusually low temperature quotient responses when exposed to acute temperature increases. Dusky Frillfin Goby, sandflat goby, and Common Mudskipper displayed Q_{10} s ≤ 1.37 in response to an acute temperature increase. While one study has reported mudskipper Q_{10} values markedly less than 2.0 (Gordon et al., 1978), none of these values were measured at temperatures above 20 C. Unfortunately, no Q_{10} data are available for non air-breathing tropical gobies; however, Bridges (1988) reviewed oxygen consumption data for subtropical goby species and concluded that they show no special metabolic adaptations for surviving intertidal environments. In fact, all gobiids in Bridges' (1988) review had Q_{10} values above 2.2. Likewise, the amphibious Shuttles Hoppfish tested in seawater and air exhibited Q_{10} responses of 2.7 and 2.5,

respectively, when temperatures were increased from 20 to 30 C (Gordon et al., 1978). Unlike hopppfish, Common Mudskipper showed a Q_{10} response of 1.37 when exposed to temperature increases of 6–7 C. Our finding of three gobies with Q_{10} values of approximately 1.0 appears to be unique in the literature and more than coincidence. It may be reasonable to assume that tropical intertidal gobies exposed to predictable temperature increases are subject to strong evolutionary pressure to keep Q_{10} responses low. Thus, the ability to avoid metabolic rate increases would be a useful energy-saving physiological adaptation in a resource-limited environment such as mangal tidepools.

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