Coral Reefs
DOI 10.1007/s00338-015-1353-4

NOTE

Behavioural thermoregulation in a temperature-sensitive coral reef fish, the five-lined cardinalfish (*Cheilodipterus quinquelineatus*)

Tiffany J. Nay[1,2] · **Jacob L. Johansen**[3] · **Adam Habary**[1,4] · **John F. Steffensen**[4] · **Jodie L. Rummer**[1]

Received: 29 March 2015 / Accepted: 14 September 2015

Abstract As global temperatures increase, fish populations at low latitudes are thought to be at risk as they are adapted to narrow temperature ranges and live at temperatures close to their thermal tolerance limits. Behavioural movements, based on a preference for a specific temperature (T_{pref}), may provide a strategy to cope with changing conditions. A temperature-sensitive coral reef cardinalfish (*Cheilodipterus quinquelineatus*) was exposed to 28 °C (average at collection site) or 32 °C (predicted end-of-century) for 6 weeks. T_{pref} was determined using a shuttlebox system, which allowed fish to behaviourally manipulate their thermal environment. Regardless of treatment temperature, fish preferred 29.5 ± 0.25 °C, approximating summer average temperatures in the wild. However, 32 °C fish moved more frequently to correct their thermal environment than 28 °C fish, and daytime movements were more frequent than night-time movements. Understanding temperature-mediated movements is imperative for predicting how ocean warming will influence coral reef species and distribution patterns.

Communicated by Ecology Editor Dr. Michael Berumen

✉ Tiffany J. Nay
tiffany.nay@my.jcu.edu.au

[1] ARC Centre of Excellence for Coral Reef Studies, James Cook University, Townsville, QLD 4811, Australia

[2] College of Marine and Environmental Sciences, James Cook University, Townsville, QLD 4811, Australia

[3] Department of Marine Science, University of Texas, Marine Science Institute, Port Aransas, Texas 78373-5015, USA

[4] Marine Biological Section, Department of Biology, University of Copenhagen, Strandpromenaden 5, 3000 Helsingør, Denmark

Keywords Behavioural thermoregulation · Temperature preference · Temperature sensitivity

Introduction

Global mean sea surface temperatures are predicted to increase 2.6–4.8 °C by the end of the century (Collins et al. 2013) and may affect some locations and species differently. Given that extreme latitudes, such as the equator and poles, normally experience little variation in daily and seasonal temperatures (Somero 2002; Hoegh-Guldberg et al. 2007; Tewksbury et al. 2008; Lough 2012), resident species may be adapted to a narrow range of temperatures (Pörtner and Farrell 2008, Pörtner and Peck 2010; Tewksbury et al. 2008). Adaptation to and operating within a narrow temperature range can allow species to minimize maintenance costs and increase fitness (Pörtner and Farrell 2008), but operating outside these temperature ranges can come at a cost and may decrease overall fitness. With predicted temperature increases, more energy may be required to maintain daily processes, which may have deleterious effects on performance and survival (Pörtner and Peck 2010). Thus, populations near the equator and poles are expected to be at increased risk as sea surface temperatures rise. Species living at these latitudes will need to acclimate or adapt, redistribute to latitudes or depths where temperatures may be more forgiving, or risk disappearing from some areas completely (Perry et al. 2005).

Ectotherms, such as fish, will be at risk from changing environmental conditions because, for most, core body temperatures reflect local thermal environments. Behavioural thermoregulation, a form of phenotypic plasticity, may allow some species or populations to reduce or mitigate the deleterious impacts of changing environmental temperatures (Ward et al. 2010; Thumus et al. 2012; Johansen et al. 2014). Fish may use movement to maintain an internal temperature that closely resembles their preferred temperature (T_{pref}) (Neill et al. 1972; Schurmann and Steffensen 1991), which can reduce daily maintenance costs (Killen 2014) and therefore influence critical biological processes (Pörtner and Farrell 2008), potentially leading to increased performance, fitness, and survival. Indeed, an organism's T_{pref} may theoretically reflect its optimum temperature (T_{opt}) for aerobic performance as well as its distribution range. However, some tropical species already occur at latitudes where maximum temperatures are close to their T_{opt} (Rummer et al. 2014). These populations may temporally use deeper, cooler habitats to reduce metabolic costs. Thermal preference may therefore influence species' distribution patterns in several ways through habitat selection (Pörtner and Farrell 2008; Gardiner et al. 2010) and modification to depth and/or latitude ranges (Perry et al. 2005; Grebmeier et al. 2006; Pörtner and Peck 2010).

Temperature preference has been investigated in some temperate fish species (Fry 1947; Brett 1952; Kelsch and Neill 1990; Johnson and Kelsch 1998; Killen 2014), but no study to date has explored this trait in tropical coral reef fish species. Cardinalfishes (Apogonidae) are known as temperature sensitive (i.e., limited capacity for acclimation and/or limited temperature tolerance range) (Nilsson et al. 2009, 2010; Gardiner et al. 2010; Rummer et al. 2014) and thus may be good candidates for investigating temperature preference. It is already known that some cardinalfishes are unable to acclimate numerous morphological and/or physiological traits, even over days to weeks (e.g., gill morphology, see Bowden et al. 2014; aerobic scope, see Rummer et al. 2014; Gardiner et al. 2010; Nilsson et al. 2010), to the elevated temperatures expected under global climate change. Furthermore, cardinalfishes have repeatedly been shown to lose condition and reduce physiological performance with minor temperature changes (Gardiner et al. 2010; Nilsson et al. 2010; Rummer et al. 2014). Using the five-lined cardinalfish (*Cheilodipterus quinquelineatus*), we aimed to investigate whether temperature-sensitive coral reef fishes can utilize behaviour to move to areas of suitable temperatures, even after they have been pre-exposed to higher temperatures for prolonged periods of time. Most cardinalfishes, including *C. quinquelineatus*, exhibit nocturnal behaviours, actively foraging over sand and reef at night (Chave 1978; Marnane and Bellwood 2002) and inhabit specific resting sites during the day (Greenfield and Johnson 1990; Gardiner and Jones 2010) when temperatures in shallow reef habitats may rise due to sun exposure and tidal level (Craig et al. 2001). Therefore, a second aim was to determine whether a species' thermal profile reflects its daytime or night-time habitat.

Materials and methods

Animal care and experimental temperature treatments

Cheilodipterus quinquelineatus were collected using hand nets in shallow coral reef lagoons near Lizard Island (14°40′08″S, 145°27′34″E), Northern Great Barrier Reef, Australia, in January 2014 when temperatures were approximately 28 °C. Fish were then transported to the Marine Aquaculture Research Facilities Unit (MARFU) at James Cook University in Townsville, Queensland, Australia. Groups of cardinalfish of similar size (t test, $t_{17.24} = -1.01$, $p = 0.32$) were held at either 28 °C ($n = 8$) or 32 °C ($n = 10$) to account for end-of-century predictions (Collins et al. 2013). To reach 32 °C, aquarium temperatures were increased at a rate of 0.5 °C d^{-1} using a 5000 W heater until the target temperature was reached. Both groups were maintained at treatment temperatures for a minimum of 6 weeks to ensure that any acclimation processes were complete (Guderley and Gawlicka 1992). Fish were maintained under a 12:12 photoperiod and were fed commercial pellets and newly hatched *Artemia* spp. twice daily to satiation. However, fish were fasted for 24 h prior to experimental trials to ensure a post-absorptive state that maximized energy available for performance (Niimi and Beamish 1974).

Temperature preference equipment

The T_{pref} was determined for each fish by placing a single individual into a shuttlebox developed by Schurman and Steffensen (1991) and Peterson and Steffensen (2003). In brief, the shuttlebox is a two-chamber PVC aquarium with the bottoms of each chamber made from transparent plexiglass (Ø 35 cm). Each chamber is cylindrical (Ø 34.5 cm), and the two chambers are joined at the middle with a 50-mm-wide passage allowing the fish to move freely between chambers. One chamber was consistently maintained 1 °C cooler than the other using two chillers and a 5000 W heater. This differential was chosen because it is large enough to prompt movement between chambers, as determined from preliminary observations on this species. Prior to each trial, temperatures inside the chambers were set to the fish's treatment temperature (28 or 32 °C), and a single fish was placed in one chamber of the shuttlebox and permitted to familiarize with the system for 1.5 h. The allotted time for the fish to 'learn' the system was based on observed reaction times to the passageway between the chambers.

When a fish entered the 'warm' chamber, the temperature of the entire system increased at a rate of

1.5–2 °C h^{-1}, and when the fish entered the ‘cool’ chamber, the temperature of the entire system decreased at the same rate, while maintaining a 1 °C difference between chambers. By moving between tanks, each fish was able to control the temperature of its environment, and therefore its body temperature.

Throughout each trial, temperature sensors recorded values to thermostats linked to a computerized software system, and the fish’s position was analysed using LoliTrack (Loligo Systems, Tjele, Denmark). Depending on the real-time position of the fish, the computer and purpose-written software (Labtech Notebook Pro, Laboratories Technology Corp., Andover, MA, USA) would automatically turn on or off the corresponding heating or cooling reservoir loop. To ensure the fish was detectable by the tracking software during both daytime and night-time hours, infrared lights were used to illuminate the fish from underneath, which created a strong contrast between the chamber background and the fish. In addition, a small lamp was used at night-time to mimic moonlight and to allow the fish to navigate between chambers.

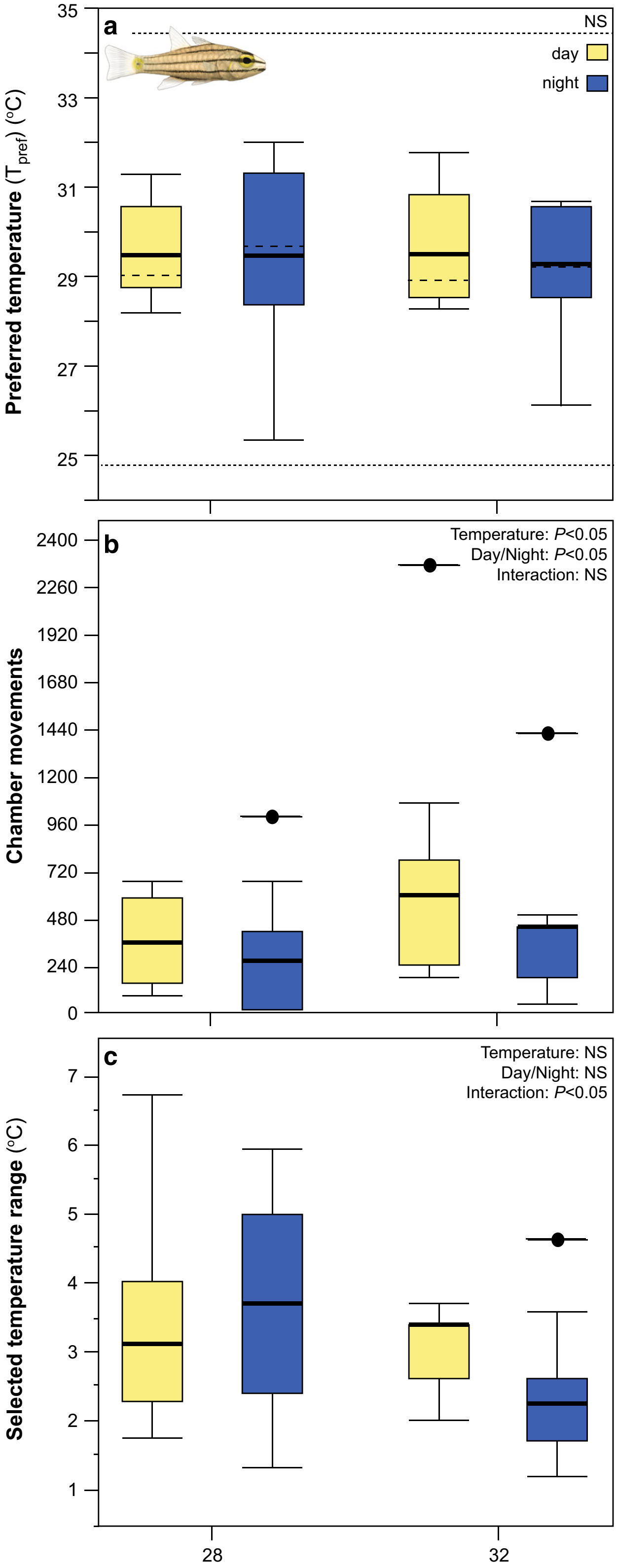
Data analyses

The T_{pref} data were analysed by calculating the proportion of time each fish spent at each temperature using one 5-h timeframe within each daytime and night-time period for each individual. The timeframes were chosen to ensure that calculations of night-time T_{pref} and daytime T_{pref} were adequately separated. One replicate of each daytime and night-time period was used for each fish in the analysis. Means were then compared between the fish from the 28 and 32 °C treatment temperatures, and given that this species exhibits nocturnal activity patterns, comparisons were also made between night-time and daytime periods. The differences in temperature preference (T_{pref}) between treatment temperatures, day/night-time periods, and interactions between the two factors were analysed using a two-way ANOVA and Holm–Sidak post hoc tests ($\alpha = 0.05$). This was also done for chamber movements and selected temperature ranges following $\log_{10}$ transformations. Chamber movements were defined as the movement from one chamber to the other, and the selected temperature range was characterized as the difference between the maximum and minimum temperatures experienced by each fish. All assumptions were met for analyses, and all results are presented as mean ± SE unless otherwise stated. Finally, to confirm that T_{pref} results were not a consequence of random or lack of movements, simulated trials were completed following identical system settings with a mock 28 °C fish and then compared to 28 °C fish data. Simulated trials confirmed significantly different T_{pref} between the mock fish and treatment fish (two-way ANOVA, $F_{1,15} = 6.35$, $p < 0.05$). Within the mock trial, as expected, there was no difference between day and night ($F_{1,15} = 0.00$, $p = 0.95$).

Results and discussion

Rising ocean temperatures are a major threat to thermally sensitive species, particularly those with little or no capacity for thermal acclimation. The temperature-sensitive *C. quinquelineatus* preferred 29.5 ± 0.25 °C (Fig. 1a), regardless of diel cycle ($F_{1,32} = 0.04$, $p = 0.85$) or prolonged exposure to present-day or elevated temperatures ($F_{1,32} = 0.06$, $p = 0.80$). This preferred temperature is close to the current average summer temperature that this population experiences in the wild, suggesting a possible evolutionary selection for temperatures that optimize performance and survival. Temperature adjustments (i.e., number of chamber movements, Fig. 1b) were more numerous in individuals from 32 °C ($F_{1,32} = 5.12$, $p < 0.05$) and during the daytime for both treatment groups ($F_{1,32} = 4.32$, $p < 0.05$). The interaction effect between time of day and treatment was significant, and post hoc tests revealed that the daytime selected temperature ranges were wider than night-time ranges within 32 °C fish (Fig. 1c, $p < 0.05$). Furthermore, the selected temperature ranges were wider in 28 °C than in 32 °C fish during the night-time periods ($p < 0.05$), but were similar between daytime periods. These results may indicate a greater urgency for fish to regulate their body temperature under thermal stress, when metabolic demands are elevated (see Rummer et al. 2014), and when they are further from their optimum temperature, and suggests that this species, and perhaps other thermally sensitive confamilials, may relocate to cooler habitats as temperatures continue to increase under ocean warming.

The difference between movement of schools of fish and lone fish may provide an explanation for our observation of more frequent movements in the individual fish examined in this study during the daytime, as many factors affect an organism’s movement and activity patterns within their natural habitats and ecosystems. During the day, nocturnal species such as *C. quinquelineatus* may be more vulnerable to predation if conditions require them to move away from their sheltered microhabitats (Marnane and Bellwood 2002). Indeed, fish may have to make trade-offs between their preferred thermal environments and other important factors such as hypoxia, resource availability (e.g., shelter or forage), as well as predation risk. Species’ social structure may also play a role in determining activity movement patterns. This has been demonstrated in the wild for *C. quinquelineatus* during both day and night, as grouped cardinalfish remain at single resting sites and



◀**Fig. 1** Boxplots representing **a** temperature preference (T_{pref}), **b** chamber movements per 5-h trial, and **c** the selected range of temperatures for fish from the 28 and 32 °C temperature treatments. The *dotted lines* represent the minimum and maximum temperatures any one fish experienced during all trials. The *boxes* represent first and third quartiles, and the *whiskers* (errors) represent the minimum and maximum values outside of outliers. Outliers are *solid lines* with a *closed circle*. Within each box, median (*dashed line*) and mean (*solid line*) values are included. Daytime values are in light (*yellow*) boxplots. Night-time values are in dark (*blue*) boxplots

display high site fidelity during the daytime (Kuwamura 1985; Marnane 2000), but lone cardinalfish move frequently between sites, even over 20–50-m distances during daytime hours, potentially searching for a future mate (Rueger et al. 2014). In the wild, ecosystem interactions (biotic and abiotic) and social structure of the species can influence behaviour; however, temperature will undoubtedly affect the fish activity and movement.

Evidence suggests that many coral reef fish species have limited capacity for acclimation to increasing temperatures, which may result in greater stress on physiological processes impacting body size, condition, growth, swimming performance, and fecundity. By moving to more suitable habitats by latitude or at a local scale (i.e., around the reef, within microhabitats, or with depth), behavioural thermoregulation offers species an alternative strategy to ease some of the constraints that future elevated temperatures may have on physiological processes. However, temperature-mediated movements could shift species distributions and ultimately limit acclimation and selection of thermal physiology, as suggested in other ectotherms such as lizards (Buckley et al. 2015). Over 360 tropical coral reef fish species from 55 families have already been shown to be expanding their distribution ranges to higher latitudes (Feary et al. 2014). While this is a more positive alternative to species disappearing altogether, an influx of new species into an established ecosystem may also cause dramatic changes in ecosystem function, species abundance and diversity, and resource availability (Verges et al. 2014; Feary et al. 2014).

As climatic changes persist, many more tropical species may begin to expand their latitudinal distribution ranges. Our results demonstrated that this species exhibited the same preferred temperature regardless of exposure to increased temperature. This provides a compelling explanation for the numerous species already showing distributional range shifts. With a greater understanding of temperature preference, we can use this knowledge as a tool to predict how species distributions will change and respond to ocean warming.

Acknowledgments Thanks are due to C. Gervais for editorial assistance, the staff of the Marine and Aquatic Research Facilities Unit (MARFU) at JCU for help with infrastructure and logistical support, SOLEX Townsville for technical support, and Erin Walsh for fish illustrations. We are also appreciative to the anonymous reviewers for their critical and helpful comments and suggestions. This work was supported by an ARC Super Science Fellowship and ARC Centre of Excellence for Coral Reef Studies research allocation to J.L.R. All animal collections were conducted under Marine Parks Permit #G10/33239.1. All animal care and experimental protocols were approved by JCU ethics approval A2089.

References

Bowden AJ, Gardiner NM, Couturier CS, Stecyk JA, Nilsson GE, Munday PL, Rummer JL (2014) Alterations in gill structure in tropical reef fishes as a result of elevated temperatures. Comp Biochem Physiol A Mol Integr Physiol 175:64–71

Brett JR (1952) Temperature tolerance in young Pacific salmon, genus *Oncorhynchus*. J Fish Res Board Can 9:256–323

Buckley LB, Ehrenberger JC, Angilletta MJ (2015) Thermoregulatory behaviour limits local adaptation of thermal niches and confers sensitivity to climate change. Funct Ecol 29:1038–1047

Chave EH (1978) General ecology of six species of Hawaiian cardinalfishes. Pac Sci 32:245–270

Collins M, Knutti R, Arblaster J, Dufresne JL, Fichefet T, Friedlingstein P, Gao X, Gutowski WJ, Johns T, Krinner G, Shongwe M, Tebaldi C, Weaver AJ, Wehner M (2013) Long-term climate change: projections, commitments and irreversibility. Cambridge University Press, Cambridge

Craig P, Birkeland C, Belliveau S (2001) High temperatures tolerated by a diverse assemblage of shallow-water corals in American Samoa. Coral Reefs 20:185–189

Feary DA, Pratchett MS, Emslie MJ, Fowler AM, Figueira WF, Luiz OJ, Nakamura Y, Booth DJ (2014) Latitudinal shifts in coral reef fishes: why some species do and others do not shift. Fish Fish (Oxf) 15:593–615

Fry FEJ (1947) Effects of the environment on animal activity. Pub Ont Fish Res Lab 68:1–62

Gardiner NM, Jones GP (2010) Synergistic effects of habitat preference and gregarious behaviour on habitat use in coral reef cardinalfish. Coral Reefs 29:845–856

Gardiner NM, Munday PL, GrE Nilsson (2010) Counter-gradient variation in respiratory performance of coral reef fishes at elevated temperatures. PloS One 5:e13299

Grebmeier JM, Overland JE, Moore SE, Farley EV, Carmack EC, Cooper LM, Frey KE, Helle JH, McLaughlin FA, McNutt SL (2006) A major ecosystem shift in the Northern Bering Sea. Science 311:1461–1464

Greenfield DW, Johnson RK (1990) Heterogeneity in habitat choice in cardinalfish community structure. Copeia 1990:1107–1114

Guderley H, Gawlicka A (1992) Qualitative modification of muscle metabolic organization with thermal acclimation of rainbow trout, *Oncorhynchus mykiss*. Fish Physiol Biochem 10:123–132

Hoegh-Guldberg O, Mumby PJ, Hooten AJ, Steneck RS, Greenfield P, Gomez E, Harvell CD, Sale PF, Edwards AJ, Caldeira K, Knowlton N, Eakin CM, Iglesias-Prieto R, Muthiga N, Bradbury RH, Dubi A, Hatziolos ME (2007) Coral reefs under rapid climate change and ocean acidification. Science 318:1737–1742

Johansen JL, Messmer V, Coker DJ, Hoey AS, Pratchett MS (2014) Increasing ocean temperatures reduce activity patterns of a large commercially important coral reef fish. Glob Chang Biol 20:1067–1074

Johnson JA, Kelsch SW (1998) Effects of evolutionary thermal environment on temperature-preference relationships in fishes. Environ Biol Fishes 53:447–458

Kelsch SW, Neill WH (1990) Temperature preference versus acclimation in fishes: selection for changing metabolic optima. Trans Am Fish Soc 119:601–610

Killen SS (2014) Growth trajectory influences temperature preference in fish through an effect on metabolic rate. J Anim Ecol 83:1513–1522

Kuwamura T (1985) Social and reproductive behaviour of three mouthbrooding cardinalfishes, *Apogon doederlini*, *A. niger* and *A. notatus*. Environ Biol Fishes 13:17–24

Lough JM (2012) Small change, big difference: sea surface temperature distributions for tropical coral reef ecosystems, 1950–2011. J Geophys Res Oceans 117:C09018

Marnane MJ (2000) Site fidelity and homing behaviour in coral reef cardinalfishes (family Apogonidae). J Fish Biol 57:1590–1600

Marnane MJ, Bellwood DR (2002) Diet and nocturnal foraging in cardinalfishes (Apogonidae) at One Tree Reef, Great Barrier Reef, Australia. Mar Ecol Prog Ser 231:261–268

Neill WH, Magnuson JJ, Chipman GG (1972) Behavioral thermoregulation by fishes: a new experimental approach. Science 176:1443–1445

Niimi AJ, Beamish FWH (1974) Bioenergetics and growth of largemouth bass (*Micropterus salmoides*) in relation to body weight and temperature. Can J Zool 52:447–456

Nilsson GE, Östlund-Nilsson S, Munday PL (2010) Effects of elevated temperature on coral reef fishes: loss of hypoxia tolerance and inability to acclimate. Comp Biochem Physiol A Mol Integr Physiol 156:389–393

Nilsson GE, Crawley N, Lunde IG, Munday PL (2009) Elevated temperature reduces the respiratory scope of coral reef fishes. Glob Chang Biol 15:1405–1412

Perry AL, Low PJ, Ellis JR, Reynolds JD (2005) Climate change and distribution shifts in marine fishes. Science 308:1912–1915

Peterson MF, Steffensen JF (2003) Preferred temperature of juvenile Atlantic cod *Gadus morhua* with different haemoglobin genotypes at normoxia and moderate hypoxia. J Exp Biol 206:359–364

Pörtner HO, Farrell AP (2008) Physiology and climate change. Science 322:690–692

Pörtner HO, Peck MA (2010) Climate change effects on fishes and fisheries: towards a cause-and-effect understanding. J Fish Biol 77:1745–1779

Rueger T, Gardiner NM, Jones GP (2014) Relationships between pair formation, site fidelity and sex in a coral reef cardinalfish. Behav Process 107:119–126

Rummer JL, Couturier CS, Stecyk JA, Gardiner NM, Kinch JP, Nilsson GE, Munday PL (2014) Life on the edge: thermal optima for aerobic scope of equatorial reef fishes are close to current day temperatures. Glob Chang Biol 20:1055–1066

Schurmann H, Steffensen J (1991) Fisk foretrækker lavere temperatur under iltsvind. Vand and Miljø 9:434–437

Somero GN (2002) Thermal physiology and vertical zonation of intertidal animals: optima, limits, and costs of living. Integr Comp Biol 42:780–789

Tewksbury JJ, Huey RB, Deutsch CA (2008) Putting the heat on tropical animals. Science 320:1296–1297

Thumus M, Meekan M, Stevens J, Wilson S, Polovina J (2012) Evidence for behavioural thermoregulation by the world's largest fish. J R Soc Interface 10:20120477

Verges A, Steinberg PD, Hay ME, Poore AG, Campbell AH, Ballesteros E, Heck KL, Booth DJ, Coleman MA, Feary DA, Figueira W, Langlois T, Marzinelli EM, Mizerek T, Mumby PJ, Nakamura Y, Roughan M, Sebille E, Gupta AS, Smale DA, Tomas F, Wernberg T, Wilson SK (2014) The tropicalization of temperate marine ecosystems: climate-mediated changes in herbivory and community phase shifts. Proc R Soc Lond B Biol Sci 281:20140846

Ward AJW, Hensor EMA, Webster MM, Hart PJB (2010) Behavioural thermoregulation in two freshwater fish species. J Fish Biol 76:2287–2298