Heat Exchange in the Black Skipjack, and the Blood–Gas Relationship of Warm-Bodied Fishes

(thermal profile/hemoglobin/rete mirabile)

JEFFREY B. GRAHAM

Smithsonian Tropical Research Institute, P.O. Box 2072, Balboa, Canal Zone

ABSTRACT   The black skipjack, *Euthynnus lineatus*, uses a centrally located vascular heat exchanger to maintain core body temperatures warmer than ambient sea water. The heat exchanger is composed of the dorsal aorta, the posterior cardinal vein, and a large vertical rete. The dorsal aorta is embedded in the posterior cardinal vein and is completely bathed in venous blood. Skipjack hemoglobin appears similar to that of the bluefin tuna in that oxygen capacity is unaffected by changing temperature. Temperature-insensitive hemoglobin may function in warm-bodied fishes to prevent the premature dissociation of oxygen from hemoglobin as blood is warmed en route to the muscles.

Tuna fish (family Scombridae) and mackerel sharks (Isuridae), which are fast-swimming open-water species, maintain body temperatures that are warmer than ambient sea water by conserving the heat generated by their swimming muscles. To do this, these fishes use retia mirabilia as countercurrent vascular heat exchangers; the retia are located in and adjacent to their red muscles (1). One advantage of a high and fairly constant body temperature is that it facilitates high-speed swimming by increasing muscular contraction frequencies, and thus increases available swimming power. Warm-bodied fishes probably also achieve a marked independence from environmental temperature, and this enables a few species to make extensive vertical and latitudinal migrations (2, 3). It has been recently demonstrated that the bluefin (*Thunnus thynnus*) can thermoregulate over a wide range of water temperatures (4, 5).

PROBLEM

In their review of warm-bodied fishes, Carey *et al.* (1) describe two types of heat exchanger: lateral and central. Large tuna and mackerel sharks have lateral exchangers, while skipjack, and to some extent the yellowfin, which are smaller species, have central heat exchangers. The thermal profiles of fishes with central heat exchangers have not been determined nor has the structure of central heat exchangers been fully described.

Another area in warm-bodied fish physiology that has not received full attention is the effect of temperature changes on blood–gas relationships. In the gills, blood is in thermal equilibrium with ambient sea water but rapidly warms as it passes into the muscles. Temperature changes from gill to muscles can be as large as 21° for the bluefin and can range from 2 to 13° for most other species (1, 5). The prevailing *a priori* assumption is that the increase in blood temperature enhances the unloading of oxygen in the tissues by decreasing hemoglobin–oxygen affinity. Temperature would thus act, in addition to the lowered pH and increased CO_2 in the tissues, to increase the rate and the magnitude of oxygen dissociation.

A problem arises with this assumption due to the spatial separation of the rete and muscle. If heat exchange begins in the rete and oxygen dissociation commences as blood temperature increases, what prevents the blood from unloading a portion of its oxygen in the rete or in the periphery of the muscles? Further, given that hemoglobin can unload very rapidly (6–8), what ensures the uniform distribution of oxygen within the respiring muscle?

The present study relates these questions to the interesting and important discovery by Rossi–Fanelli and Antonini (9) that the capacity of bluefin hemoglobin for combining with oxygen is virtually independent of temperature. It is herein proposed that the function of temperature-independent hemoglobin is to ensure the efficient delivery of oxygen to muscle tissue by preventing the premature unloading of this gas in the rete or outer layers of the muscle mass.

A study of the black skipjack *Euthynnus lineatus* Kishinouye, a centrally-heated warm-bodied fish, was undertaken to determine its thermal profile, describe its vascular heat exchanger, and determine the effects of temperature on its blood–gas relationships. The black skipjack occurs throughout tropical and subtropical waters in the eastern Pacific Ocean from California to Peru, but it is most frequently found near the mainland or near off-shore islands where water temperatures are from 24 to 31° (10). All fishes used in this study were taken in the Gulf of Panama.

METHODS

Black skipjack (weight 1–3 kg; 500–700 mm fork length) were caught by trolling on the surface and quickly brought on board, usually within 40 sec of being hooked. Body temperatures were immediately measured with a fast-reading hypodermic thermistor probe (YSI no. 454); readings were rounded to the nearest one-half degree. Body temperatures changed after 5 min on the shaded deck and all temperature measurements were made within 5 min of capture. Blood samples were taken by cardiac puncture, heparinized, and stored on ice for laboratory analysis. Red-muscle distribution and the structure of the heat exchanger were determined by dissection of fresh and frozen specimens.

RESULTS AND DISCUSSION

Thermal profiles, compiled from measurements on 22 fishes, reveal a warm central core in *E. lineatus* and demonstrate the close relationship between elevated body temperatures and the distribution of red muscle in the fish (Fig. 1). The warmest body temperatures occur along the vertebral column between the first and second dorsal fins. For the 22 fishes, the average temperature in this region was 34.2° (range 33.5–36.5°). Surface water temperatures, recorded when fishes were caught, averaged 29.2° (range 28.5–30.0°). None of the fishes had elevated visceral temperatures, and heart temperatures averaged only slightly warmer than sea water. The brain temperature (measured through the pineal window) of seven specimens caught in 28.5° water averaged 30.0° (range 29.0–31.5°). Temperatures at the base of the eye in these seven fishes averaged 31.0° (range 30.0–31.5°). Elevated brain and eye temperatures have been reported in other warm-bodied fishes (5, 11), and it is assumed that elevated temperatures in these regions enhance neural integrative functions needed for rapid reaction during feeding and orientation (11). Linthicum and Carey (11) recently described a countercurrent heat exchanger in the carotid artery of bluefins that functions to maintain elevated brain and eye temperatures in this species.

The burst of rapid swimming required to catch the troll lure and the subsequent struggling to resist capture probably raised black skipjack body temperatures above their normal values (12). Different levels of activity apparently caused variations in body temperature of *E. lineatus*. On one occasion it was found that the temperatures of three fishes from one school, while in good agreement with each other, were warmer by 1.5° than those of three fishes from another school caught at the same water temperature. Carey and Teal (4) made similar observations for *T. thynnus*. Stevens and Fry (12) observed disparities in the body temperatures of ocean-caught and captive skipjack and suggested this was partially caused by different physiological responses to exercise (for captive specimens) and the feeding frenzy of fishes in the wild.

The central heat exchanger of *E. lineatus* consists of the dorsal aorta, the posterior cardinal vein, and their small vessels, which form a vertical central rete (Fig. 2). The exchanger occurs below the vertebrae in the expanded hemal canal and extends along the body from the first to second dorsal fin; its lineal distribution closely corresponding to that of red muscle (Figs. 1 and 2). Blood-vessel dimensions in *E. lineatus* agree with those measured in other warm-bodied fishes. The following are maximum diameters (in mm) of the vessels in the heat exchanger of a 580-mm (fork length) specimen: dorsal aorta, 2.5; posterior cardinal vein, 5.0; retial vessels, 0.05; cutaneous artery, 1.0; cutaneous vein, 2.0. At the edge of the rete, retial vessels coalesce to form larger vessels (0.2–0.3 mm in diameter) that penetrate the red muscle, via the longitudinal septa, at various positions along the vertebrae (Fig. 2). The dorsal aorta is embedded in the dorsal side of the posterior cardinal vein and is surrounded by the vast network of retial vessels that enter the posterior cardinal vein at its dorsal surface. Many of these vessels actually empty into the lumen of the cardinal vein above the dorsal aorta. Blood discharged into the vein at this level flows down around the aorta into the main cavity of the vein (Fig. 2). Through this structure and the position of the dorsal aorta in the dorsal side of the vein, the posterior cardinal vein in effect surrounds the dorsal aorta. This allows the rete to occur through a complete arc above the two vessels, and thus permits the maximum heat-exchanging area in the rete. The structure and the position of the vessels at the center of the fish suggest that some heat exchange may actually occur while blood is in the major vessels. The dorsal aorta is bathed in venous blood and blood flow in the two vessels is opposed. Also, the vessels are almost completely
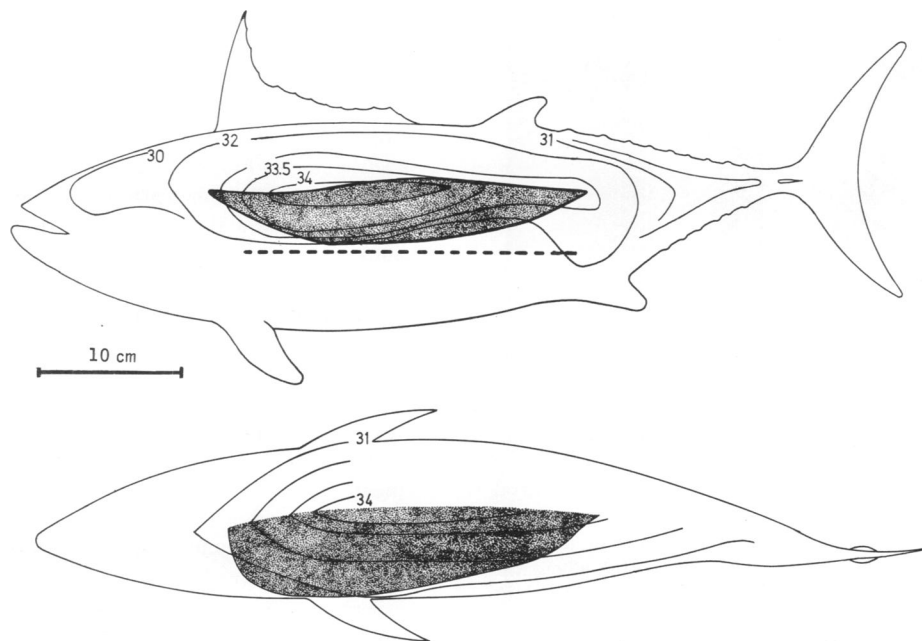

FIG. 1. Thermal profiles, red muscle distribution (*stippled*), and lineal distribution of the central heat exchanger (*dashed line*) in *E. lineatus*. Profiles are based on body-temperature measurements of 22 fishes. In the dorsal view, red muscle is shown on one side only. Water temperature, 29°.

surrounded by warm muscle, which would favor heat conduction inward.

In the heat exchanger the dorsal aorta appears highly segmented due to the large number of small arteries that emerge from it and pass into the rete (Fig. 2). Three to four such vessels leave each side of the aorta in each segment. One set of vessels, which does not enter the rete, leaves the aorta at its ventro-lateral side and descends along the inner surface of the vein before it penetrates its wall en route to lower portions of the body (Fig. 2). Vessel ports occur throughout the wall of the vein, as was previously discussed, even above the dorsal aorta.
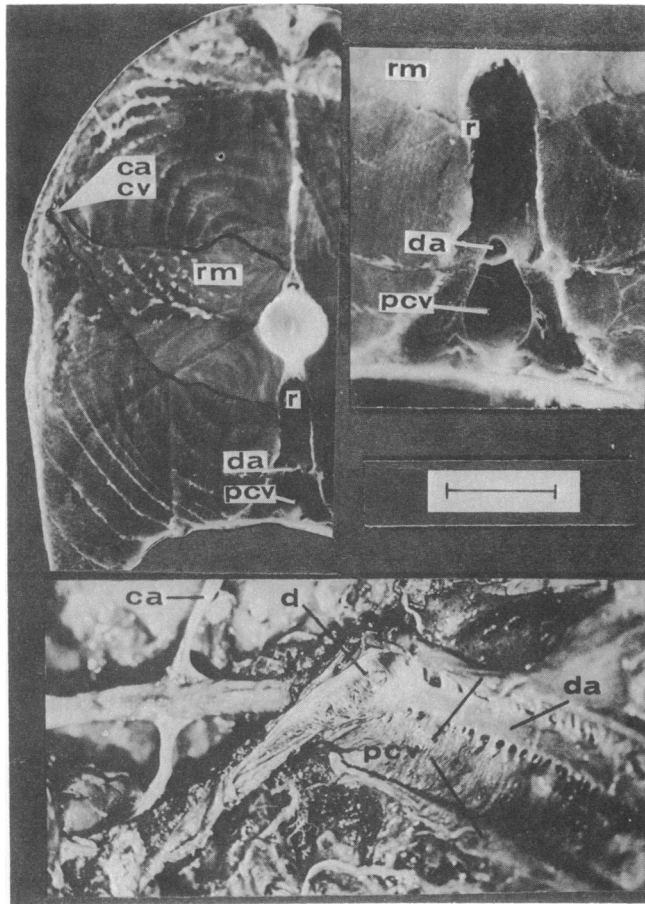

FIG. 2. (*Top left*, scale = 2 cm): Transverse section of *E. lineatus* at the level of the pectoral fins, showing the position of the central heat exchanger, the red muscle (*outlined in black*) and the cutaneous vessels. (*Top right*, scale = 1.3 cm.): Enlarged view of the central heat exchanger showing the position of the dorsal aorta, posterior cardinal vein, and rete. (*Bottom*, scale = 1.3 cm): ventral view of the dorsal aorta and posterior cardinal vein in the anterior part of the body. In the heat exchanger (*right side*), the cardinal vein has been cut open along its ventral edge and laid back to expose the segmented dorsal aorta. From this vantage, the vessels that run along the inside of the vein before penetrating its wall are evident. At the point where the vessels diverge (*d*), the posterior cardinal vein slants to the right of the fish (*bottom of picture*) and descends towards the duct of Cuvier. The dorsal aorta continues anteriorly along the mid-line, and the left and right cutaneous arteries branch from it. Symbols: cutaneous artery, *ca*; cutaneous vein, *cv*; dorsal aorta, *da*; posterior cardinal vein, *pcv*; red muscle, *rm*; rete, *r*.

Anterior to the heat exchanger at about the level of the first dorsal fin, the dorsal aorta and posterior cardinal vein separate. The envelopment of the aorta by the vein and its retial vessels is dramatically evidenced at this level (Fig. 2), where the vein slants toward the right side of the fish's body and descends to the duct of Cuvier and the dorsal aorta continues along the mid-line of the body, losing its segmentation once outside the exchanger.

Small lateral retia are present in *E. lineatus*. These are formed by a cutaneous artery and vein that lie above the lateral line on each side of the fish. The cutaneous arteries arise from the dorsal aorta anterior to the central heat exchanger (Fig. 2); the veins follow the same path as the arteries and return blood to the sinus venosus (2). Some anatomical facts suggest that the lateral retia are not very important in maintaining the skipjack's thermal profile. The lateral retia are thin, consisting of only one layer of arterial vessels and two layers of venous vessels. Also, towards the posterior portion of red muscle, the diameters of the cutaneous artery and vein are reduced and many small vessels diverge into segments of white muscle.

The major differences between lateral and central heat exchangers are the location of the retia and the pattern of blood flow to them (1). In large tunas, such as the bigeye and bluefin, the retia occur on the sides of the body just under the skin at the surface of the red muscle (13). Blood is supplied through a double set of cutaneous arteries and veins that extend along the sides of the fish; one set dorsal and the other ventral to the horizontal midline. The dorsal aorta and posterior cardinal vein do not have any function in heat exchange (1). The heat exchanger in mackerel sharks (mako, porbeagle, and the great white) is basically similar, but these species have only one pair of cutaneous vessels on either side of their bodies (1, 14). Temperature profiles reveal the lateral position of the heat exchangers in the bluefin and the mako shark; temperatures are warmest in the cores of the muscle masses and cooler at the midline (1). This is fundamentally different from the profile of centrally-heated fishes where midline temperatures are the warmest.

Preliminary tests with whole blood did not indicate any pronounced effect of temperature on the oxygen binding of the blood. This is consistent with the results previously reported by Rossi–Fanelli and Antonini (9) for *T. thynnus*, and more recently for other warm-bodied fishes (8). The average hematocrit for nine fishes was 41% (range 40–44%). Investigations of the hemoglobin concentration in the blood of *E. lineatus* report levels from 18 to 20% (15).

With the exception of warm-bodied fishes, no vertebrates are known to have temperature-independent hemoglobin (8, 9, 16). For the bluefin tuna, it was suggested that this hemoglobin served as an adaptation for living in waters of very different temperatures (7, 9, 16), and the migrational patterns of this species would well support this (3). However, a similar hemoglobin in other warm-bodied fishes, including species that do not make extensive migrations, indicates that this adaptation probably has a more fundamental function.

If (as blood moved from the gills to the muscles) increased temperature decreased the oxygen affinity of the hemoglobin, there would be a progressive unloading of oxygen, beginning as the blood was warmed in the vessels of the retia and continuing as blood flowed deeper into the muscle. The amount of oxygen in blood reaching the warmest muscle, where oxygen

demand is greatest, would be low. Also, changes in ambient sea-water temperature (such as those encountered while crossing a thermocline or on an extensive latitudinal migration) would markedly affect gas transport to the muscles not only by changing the gas equilibrium properties of hemoglobin in the gills, but also by changing the thermal profile of the fish as well. It is proposed here that temperature-insensitive hemoglobin has evolved in tuna to eliminate the problem of premature oxygen dissociation as blood crosses a thermal gradient en route to the muscles. The structure of the central heat exchanger and the warm central core in *E. lineatus* accentuate the importance of temperature-insensitive hemoglobin to this species, as blood temperature may actually begin to increase when it enters the warm central core. Tuna blood is very sensitive to low pH and elevated CO_2 tensions, and these factors are probably fundamental in regulating O_2 dissociation in the muscle (7–9, 16).

1. Carey, F. G., Teal, J. M., Kanwisher, J. W., Lawson, K. D. & Beckett, J. S. (1971) "Warm-bodied fish," *Amer. Zool.* **11**, 135–143.
2. Kishinouye, K. (1923) "Contributions to the comparative study of the so-called scombroid fishes," *J. Coll. Agr. Hokkaido Imp. Univ.* **8**, 293–475; Godsil, H. C. (1954) "A descriptive study of certain tuna-like fishes," *Calif. Fish Game Fish Bull.* no. **97**, p. 185.
3. Mather, F. (1962) "Transatlantic migration of two large bluefin tuna," *J. Cons. Perma. Int. Explor. Mer.* **27**, 325–327.
4. Carey, F. G. & Teal, J. M. (1969) "Regulation of body temperature by the bluefin tuna," *Comp. Biochem. Physiol.* **28**, 205–213.
5. Carey, F. G. & Lawson, K. D. (1973) "Temperature regulation in free-swimming bluefin tuna," *Comp. Biochem. Physiol.*, in press.
6. Prosser, C. L. & Brown, F. A. (1961) *Comparative Animal Physiology* (Saunders, Philadelphia), pp. 208–209.
7. Randall, D. J. (1970) in *Fish Physiology*, eds. Hoar, W. S. & Randall, D. J. (Academic, New York), pp. 270–276.
8. Anderson, M. E., Olson, J. S., Gibson, Q. H. & Carey, F. G. (1973) "Studies on ligand binding to hemoglobins from teleosts and elasmobranchs," *J. Biol. Chem.* **248**, 331–341.
9. Rossi-Fanelli, A. & Antonini, E. (1960) "Oxygen equilibrium of haemoglobin from *Thunnus thynnus*," *Nature* **186**, 895–896.
10. Calkins, T. P. & Klawe, W. L. (1963) in F.A.O. *Fisheries Reports*, ed. Rosa, H., Jr. (UN, Rome), pp. 130–146.
11. Linthicum, D. S. & Carey, F. G. (1972) "Regulation of brain and eye temperatures by the bluefin tuna," *Comp. Biochem. Physiol.* **43A**, 425–433.
12. Stevens, E. D. & Fry, F. E. J. (1971) "Brain and muscle temperatures in ocean-caught and captive skipjack tuna," *Comp. Biochem. Physiol.* **38A**, 203–211.
13. Carey, F. G. & Teal, J. M. (1966) "Heat conservation in tuna fish muscle," *Proc. Nat. Acad. Sci. USA* **56**, 191–195.
14. Carey, F. G. & Teal, J. M. (1969) "Mako and porbeagle: Warm-bodied sharks," *Comp. Biochem. Physiol.* **28**, 199–204.
15. Klawe, W. L., Barrett, I. & Klawe, B. M. H. (1963) "Hemoglobin content of the blood of six species of scombroid fishes," *Nature* **198**, 96.
16. Riggs, A. (1970) in *Fish Physiology*, eds. Hoar, W. S. & Randall, D. J. (Academic, New York), pp. 232–235.