Myoglobin in the heart ventricle of tuna and other fishes

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Summary. The myoglobin content in the hearts of several fishes is positively correlated with the ecological physiology of the species. In the tuna heart, where the highest myoglobin values are found, the logarithmic relationship between myoglobin content and body weight is reported.

Myoglobin is a major determinant of the aerobic homeostasis in vertebrate striated muscles. During our studies on the functional biochemistry of tuna (*Thunnus thynnus* L.) heart, a very high cardiac content of myoglobin was found[1]. This prompted the present study on the developmental changes of cardiac myoglobin in this very specialized, high-speed and 'thermoregulatory'[2,3] pelagic teleost, whose survival and behaviour are dramatically O_2 dependent.

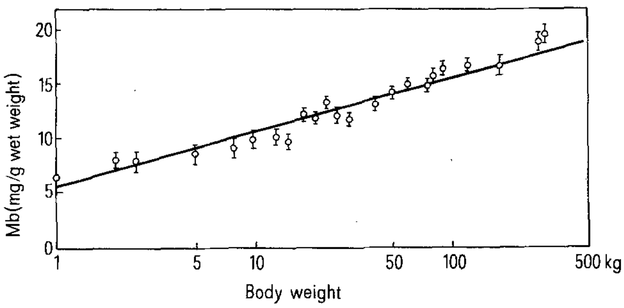
Quantitative surveys on myoglobin have been mostly focused on homoiotherms, and hence data concerning the occurrence of the pigment in lower vertebrates is very scarce. Therefore, for comparative purposes, the cardiac myoglobin content of some other fishes are here reported.

Materials and methods. Thunnus thynnus L., *Xiphias gladius, Lepidopus caudatus, Trachurus trachurus,* and the shark, *Alopias vulpinus* were obtained from commercial fishermen (Ganzirri, Sicily and Naples) soon after slaughter, and both heart ventricle and red and white muscle samples, dissected from the middle region of the myotome, were stored at −20 °C until used for analysis. All other species, obtained from the aquarium of the Zoological Station of Naples, were maintained for up 4–10 days in holding tanks with circulating sea water at 19–21 °C. The fish were killed by decapitation. Myoglobin content was assayed by the methods of Reynafarje[4].

Results and discussion. The table shows that a very close correlation exists between the content of myoglobin in the ventricular myocardium and the ecological physiology of the species. The amount of myoglobin increases from the most sedentary benthic fishes up to the pelagic, continuous high-activity animals. So far, the close relationship between high myoglobin concentration and vigorous, repetitive, continuous muscular activity, demonstrated in homoiotherms since the important studies of Millikan[5] and Lawrie[6], is also confirmed in fish by these data. This correlation may be of particular interest in the tuna. In this animal the myoglobin concentrations in the cardiac and red myotomal musculature is higher than in any other fish so far examined. It is also higher than that found in the majority of homoiotherms, higher values being reported only in diving birds and mammals. Unlike most other fishes, tuna ventilate their gills entirely by means of their movements through the water[7], and thus, by the continuous contraction of their red musculature, they must always cruise at high speed in order to survive. This depends on an adequate oxygen supply both to the heart and to the red myotomal musculature, where the vascular counter current heat-exchanger maintains the temperature higher than that of the environment[2,3].

This warmth in turn increases the rate at which myoglobin delivers oxygen to the muscle mitochondria[8]. Thus, the very high content of myoglobin provides a good example, at the molecular level, of environmental control of adaptive functions.

The quantitative developmental changes of myoglobin in tuna heart, reported in the figure, show the relationship between pigment concentration and body weight. At a very young stage (i.e. 1 kg b.wt) the cardiac myoglobin concentration of bluefin tuna is in general even higher than the average cardiac content of most adult homoiotherms (5 mg/g wet weight)[8]. The pigment increases at a high rate until the animal has reached the b.wt of about 50 kg; afterwards, the rate progressively decreases in a plateau-like fashion after 100 kg b.wt. This finding is in agreement with the knowledge that during growth, metabolism increases less than body mass[9]. These data might provide an



Semilogarithmic plot of cardiac myoglobin content (Mb) and b.wt in the tuna (*Thunnus thynnus L.*). Each point is the mean value of Mb in the heart ventricle of animals having the same b.wt. Vertical bars = SD. Total number of animals = 88. The equation is: (Mb) = a log (b.wt) + b, where b = 5.45; a = 4.97; R = 0.948.

Myoglobin content in the heart of eight species of marine fishes

Species	Habitat	Activity	Body weight (kg)	Myoglobin (mg/g wet weight) Heart ventricle	Red muscle	White muscle
Scorpaena porcus	Benthic	Sedentary	0.040-0.081	1.68 ± 0.31 (9)		
Scyliorhinus stellaris	Benthic	Sedentary	1-2.1	1.90 ± 0.40 (7)		
Trigla lucerna	Benthic	Low activity	0.138-0.230	2.45 ± 0.35 (7)		
Alopias vulpinus	Pelagic	Active	150; 160	3.80 (1); 3.91 (1)		
Lepidopus caudatus	Pelagic	Active	1.06-1.07	4.49 ± 0.12 (10)		
Xiphias gladius	Pelagic	Continuous high activity	35-40	3.50 – 4.28 (4)	4.7 – 6.2 (4)	0.9 – 2.11 (5)
			75-90	5.81 – 6.25 (5)		
Trachurus trachurus	Pelagic	Continuous high activity	0.155-0.162	6.10 ± 0.55 (6)	8.0 – 9.3 (5)	0.36 – 0.40 (5)
Thunnus thynnus	Pelagic	Continuous very high activity	10-20	9.64 – 12.0 (12)	15.1 – 21.2 (6)	1.30 – 2.10 (6)
			100-300	16.4 – 19.5 (14)	16.5 – 23.7 (5)	1.35 – 2.20 (5)

SEM calculated only for species having similar body weights; in all other cases the range of myoglobin is reported. The number of individuals sampled is given in parentheses.

insight into the oxidative processes related to the maintenance energetics and growth dynamics of this extraordinary fish.

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1 B. Tota, in: Ecological Physiology of Tunas. Ed. G. Sharp and A. Dizon. Academic Press, New York 1978.
2 F.G. Carey and J.M. Teal, Comp. Biochem. Physiol. *28*, 205 (1969).
3 F.G. Carey and K.D. Lawson, Comp. Biochem. Physiol. *44A*, 375 (1973).
4 B. Reynafarje, J. Lab. clin. Med. *61*, 138 (1963).
5 G.A. Millikan, Physiol. Rev. *19*, 503 (1939).
6 R.A. Lawrie, Biochem. J. *55*, 298 (1953).
7 G. Shelton, in: Fish Physiology, vol. 4. Ed. W.S. Hoar and D.J. Randall. Academic Press, New York 1970.
8 J.B. Wittenberg, Physiol. Rev. *50*, 559 (1970).
9 C.L. Prosser, in: Comparative Animal Physiology, p. 190. Ed. C.L. Prosser. Saunders Co., Philadelphia 1973.

Vascular reactivity in hypertension: Altered effect of ouabain[1]

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Summary. Ouabain inhibits the relaxing effect of Ca^{2+} (but not of Mn^{2+}) on contractile responses in tail artery strips isolated from spontaneously hypertensive and normotensive rats. The magnitude of ouabain inhibition was greater in vascular strips from hypertensive rats suggesting a significant difference in basic membrane function in hypertensive vascular smooth muscle.

The cause of the elevated arterial pressure in most forms of hypertension is an increase in vascular resistance. The mechanism responsible for this increase in resistance is not well defined. It could be a structural increase in wall thickness of the resistance vessels or a functional increase in contraction of the smooth muscle of these vessels. The latter could be caused by an increase in neurogenic or circulating humoral vasoconstrictor activity or by a change in the vascular smooth muscle itself which makes the muscle more sensitive to any constrictor stimulation. The present study gives insight into the mechanism responsible for a difference that occurs in vascular smooth muscle of spontaneously hypertensive rats (SHR).

An increase in extracellular Ca^{2+} concentration of vascular smooth muscle has been shown to have a triphasic effect on the contractile response[2]: a) small increases in free Ca^{2+} above physiological levels (1.6 mM) produce an augmentation of contraction; b) larger increases in Ca^{2+} depress the

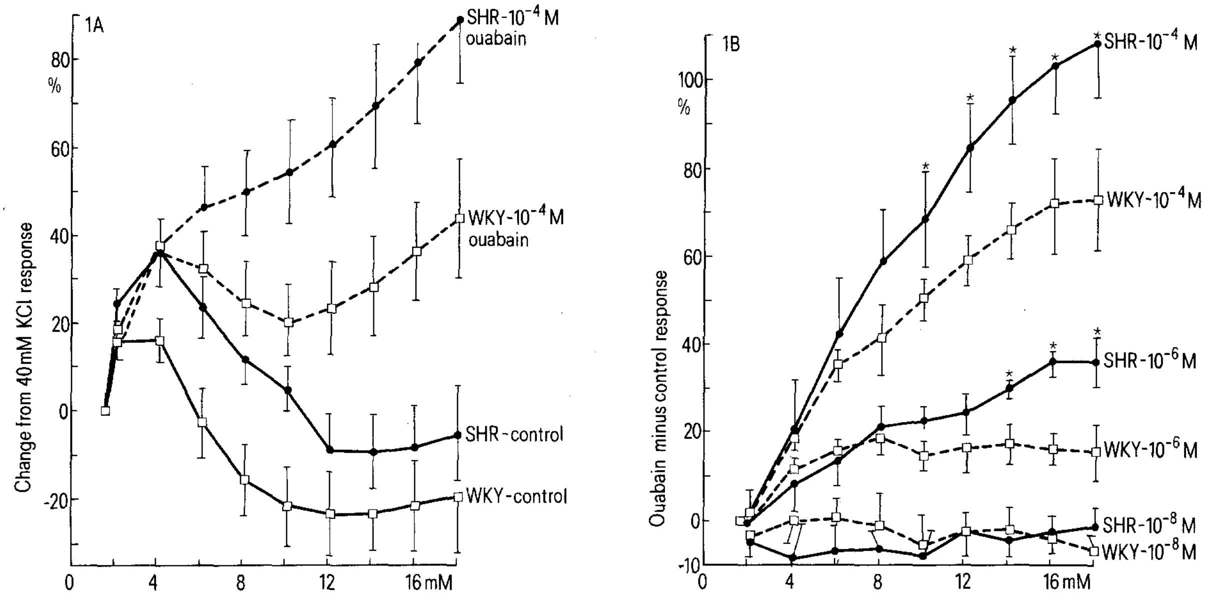

Fig. 1. *A* Effect of Ca^{2+} concentration on KCl contraction of tail artery strips from SHR and WKY. Helical strips of tail arteries from SHR and WKY were made to contract in response to 40 mM KCl in the presence and absence of ouabain. The bath concentration of $CaCl_2$ was then increased to the levels indicated. Changes in contractile tension are expressed as percent change from the 40 mM KCl response with 1.6 mM $CaCl_2$ in the bath. The mean contractile response to 40 mM KCl in each of the 4 groups before the addition of extra Ca^{2+} was: 1. SHR-control = 54 ± 6 mg; 2. WKY-control = 61 ± 8 mg; 3. SHR-10^{-4} M ouabain = 111 ± 10 mg; and 4. WKY-10^{-4} M ouabain = 121 ± 11 mg. Values are the mean ± SEM for 8 SHR and 8 WKY (2 helical strips per rat). *B* Ouabain inhibition of Ca^{2+} relaxation including some data from A. The measure of ouabain inhibition shown on the ordinate was determined by subtracting the control responses (no ouabain) of each animal from their respective treated responses (ouabain present). Statistical analysis (Student's t-test) of the responses at each Ca^{2+} concentration indicate that SHR responses were significantly ($p < 0.05$, marked by asterisk) greater than WKY at ouabain concentrations of 10^{-6} and 10^{-4} M. Integration of experimental values to determine a mean area for each curve showed SHR responses were different from WKY at the $p < 0.05$ level when 10^{-4} M ouabain was added, and at the $p < 0.10$ level when 10^{-6} M ouabain was added. Values are the means ± SEM for 4 SHR and 4 WKY (2 strips per rat.).