Cardiac physiology in tunas. I. *In vitro* perfused heart preparations from yellowfin and skipjack tunas

A. P. FARRELL, P. S. DAVIE,[1] C. E. FRANKLIN,[1] J. A. JOHANSEN, AND R. W. BRILL[2]
Biological Sciences Department, Simon Fraser University, Burnaby, B.C., Canada V5A 1S6

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An *in situ* heart preparation perfused with oxygenated saline was used to examine cardiac performance at 25°C in yellowfin tuna (*Thunnus albacares*) and skipjack tuna (*Katsuwonus pelamis*). Heart rates (91–172 bpm in skipjack tuna and 101–157 bpm in yellowfin tuna) were comparable to those measured *in vivo*, and physiological stroke volumes were possible in yellowfin tuna with subambient filling pressures. In yellowfin tuna, maximum stroke volume and cardiac output were similar to the values obtained *in vivo* with spinally blocked animals; mean output pressures (up to 145 cmH_2O, 1 cmH_2O = 0.098 kPa) could exceed *in vivo* values without a major decrease in the resting cardiac output (homeometric regulation). In contrast, saline-perfused skipjack tuna hearts could not develop physiological output pressures without compromising cardiac output, with cardiac output being only 63% of the *in vivo* value at an output pressure near the *in vivo* ventral aortic pressure. The poor performance of the skipjack tuna heart is attributed to limited oxygen diffusion through the thicker walled ventricle. We conclude that the tuna heart is more dependent on its coronary circulation for normal function than the hearts of other fishes examined thus far. The coronary circulation was perfused with saline at various flow rates in isolated hearts from skipjack tuna to develop a pressure–flow relationship for the intact circulation. Coronary resistance reached a minimum of 24 $cmH_2O \cdot min \cdot g$ ventricular mass/mL at a flow rate of 2 mL/(min · g ventricular mass) with perfusion pressure about 40 cmH_2O. *In vivo* coronary blood flow was estimated from the pressure–flow relationship as 0.67 mL/(min · g ventricular mass). Injections of adrenaline, noradrenaline, and phenylephrine into coronary circulation under constant flow conditions increased perfusion pressure, indicating the possibility of α-adrenergic vasoconstriction.

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La perfusion *in situ* de coeurs de Thons à nageoires jaunes (*Thunnus albacares*) et de Thons à ventre rayé (*Katsuwonus pelamis*) au moyen d'une solution saline oxygénée a permis de mesurer la performance cardiaque de ces poissons à 25°C. Les rythmes cardiaques, 91–172 bpm chez le Thon à ventre rayé et 101–157 bpm chez le Thon à nageoires jaunes, étaient comparables à ceux enregistrés *in vivo* et des volumes systolaires physiologiques pouvaient être obtenus chez le Thon à nageoires jaunes en appliquant des pressions de remplissage sousambiantes. Chez le Thon à nageoires jaunes, le volume systolaire maximal et le rendement cardiaque étaient semblables aux valeurs obtenues *in vivo* chez des poissons soumis à un bloquage spinal; les pressions moyennes à la sortie du coeur (jusqu'à 145 cmH_2O, 1 cmH_2O = 0.098 kPa) pouvaient dépasser les valeurs *in vivo* sans qu'il y ait de chute importante du rendement cardiaque au repos (contrôle homéométrique). En revanche, les coeurs de Thons à ventre rayé en perfusion dans la solution saline ne pouvaient pas fournir de pressions physiologiques sans compromettre le rendement cardiaque et le rendement cardiaque n'était que de 63% de la valeur *in vivo* à une pression à peu près égale à la pression obtenue dans l'aorte ventrale *in vivo*. La mauvaise performance du coeur du Thon à ventre rayé est attribuable à une diffusion d'oxygène diminuée par l'épaisseur importante de la paroi du ventricule. Nous concluons que la circulation coronarienne a plus d'impact sur le bon fonctionnement du coeur chez les thons que chez tout autre poisson examiné précédemment. La perfusion du système coronarien dans une solution saline à diverses vitesses de circulation dans des coeurs isolés de Thons à ventre rayé a servi à établir la relation pression–vitesse de circulation qui permet une circulation intacte. La résistance coronarienne a atteint un minimum de 24 $cmH_2O \cdot min \cdot g \cdot$ masse ventriculaire/mL à une vitesse de circulation de 2 mL/(min · g masse ventriculaire) (environ 40 cmH_2O) ou plus. La vitesse de circulation du sang *in vivo* dans le système coronarien a été estimée à 0,67 mL/(min · g masse ventriculaire) à partir de la relation pression–circulation établie précédemment. Des injections d'adrénaline, de noradrénaline et de phényléphrine dans la circulation coronarienne dans des conditions constantes d'écoulement ont augmenté la pression de perfusion, ce qui indique la possibilité d'une vasoconstriction sous contrôle α-adrénergique.

[Traduit par la rédaction]

Introduction

Tunas are amongst the most athletic of fishes. Their metabolic rates are almost an order of magnitude greater than those of rainbow trout (*Oncorhynchus mykiss*), approaching those of similarly sized mammals (see Dizon *et al.* 1978; Brill 1987). Many tuna species have body temperatures higher than the water in which they live (Carey 1973) as a consequence of specialized vascular countercurrent systems (Kishinouye 1923; Graham and Diener 1978) for conserving metabolic heat. Tunas generate higher arterial blood pressures than other fishes (Stevens 1972; Bushnell 1988) to drive blood through the relatively high vascular resistance of the branchial circulation (Muir and Brown 1971). Correspondingly, the relative heart mass in tunas is greater than in other fishes (Basile *et al.* 1976; Tota 1978; Poupa *et al.* 1981; Davie 1987). The ventricle has a thick layer of compact myocardium (>40% by mass; Santer and Greer-Walker 1980), and the coronary circulation supplies both the outer compact and inner spongy layers of myocardium (Tota *et al.* 1983; Tota 1989). The metabolic zonation between the compact and spongy layers has been

[1]Present address: Department of Physiology and Anatomy, Massey University, Palmerston North, New Zealand.
[2]Present address: Honolulu Laboratory, Southwest Fisheries Science Centre, National Marine Fisheries Service, National Oceanic and Atmospheric Administration, 2570 Dole Street, Honolulu, Hawaii 96822-2396, U.S.A.

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characterized (Maresca *et al.* 1976; Tota 1978; Gemelli *et al.* 1980; Tota 1983).

Cardiac output values at 25°C for normoxic spinally blocked yellowfin tuna (*Thunnus albacares*) and skipjack tuna (*Katsuwonus pelamis*) are 115 and 132 mL/(min · kg), respectively (Bushnell 1988; Brill and Bushnell 1991). These values are more than double the maximum cardiac output and 6 times the resting cardiac output reported for rainbow trout at 10°C (17 mL/(min · kg), Kiceniuk and Jones 1977). Similarly, ventral aortic blood pressure in tunas (115–119 cmH_2O, 1 cmH_2O = 0.098 kPa) is about twice that in rainbow trout (53 cmH_2O, Kiceniuk and Jones 1977) and 3 times that in sea raven (*Hemitripterus americanus*) (42 cmH_2O, Farrell 1986). Heart rates, measured in spinally blocked yellowfin tuna and skipjack tuna, are 97 and 126 beats per minute (bpm), respectively, and at these heart rates, stroke volumes are 1.3 mL/kg for yellowfin tuna and 1.1 mL/kg for skipjack tuna (Bushnell 1988). Maximum heart rates have been measured at 180–210 bpm in swimming tunas (Kanwisher *et al.* 1974; Bushnell 1988). The maximum heart rate reported in other lower vertebrates (fishes, amphibians, and reptiles) apparently does not exceed 120 bpm (Farrell 1991). Because the tuna heart clearly outperforms hearts of other fishes, we were particularly interested in increasing our knowledge of tuna cardiac physiology.

Our experimental approach for this study used an *in situ* perfused heart preparation. *In situ* perfused heart preparations have been used previously with a variety of fish species, providing useful information on the control of stroke volume and on the maximum capacity of the heart to generate flow and pressure (Farrell *et al.* 1983, 1985, 1988; Davie and Farrell 1991*a*). Rainbow trout hearts perfused with aerated perfusate and no coronary perfusion can perform work at levels comparable to the maximum *in vivo* level (Milligan and Farrell 1991). To provide additional information about the coronary circulation, pressure–flow relationships were determined by perfusing the coronary circulation of isolated skipjack tuna hearts with saline. In a companion study, the contractile properties of tuna cardiac muscle were examined with atrial strips isolated from skipjack tuna (Keen *et al.* 1992).

Materials and methods

Studies with in situ *saline-perfused hearts*

Data are reported for successful preparations from 10 skipjack tuna (mean body mass 1.32 ± 0.13 kg) and 8 yellowfin tuna (mean body mass 1.97 ± 0.08 kg). The fish were purchased from local fishermen and maintained in outdoor tanks at 24–25°C at the Kewalo Research Facility (Honolulu Laboratory, Southwest Fisheries Science Center, National Marine Fisheries Service, National Oceanic and Atmospheric Administration, Honolulu, Hawaii).

In situ *heart preparation*

The *in situ* perfused preparation was based on that developed for the rainbow trout in which the pericardium was left intact (Farrell *et al.* 1988). The basic procedure involved the following steps. Fish were anaesthetized with MS-222 (1 g/L plus 0.3 g/L $NaHCO_3$) in seawater and maintained in an anaesthetized state on an operating sling by irrigating the gills with cooled (15–20°C) MS-222 (0.1 g/L plus 0.03 g/L $NaHCO_3$). Sodium heparin (10 000 IU) was injected into the caudal vessels to prevent blood clotting in the heart. The peritoneal cavity was opened with a ventral incision. The intestinal vessels were ligated and the stomach and intestine removed to provide easier access to the hepatic veins. Two hepatic veins drain into the sinus venosus. After tying off the more dorsal one, the central hepatic vein was cannulated with a large-bore stainless steel tube to locate the tip of the cannula in the sinus venosus. This tube acted as an input cannula through which the heart received oxygenated saline at constant pressure from a Marriot bottle. Cardiac perfusion began immediately after the input cannula was secured. A gill arch was cut prior to opening the hepatic vessel, because air was aspirated into the heart chamber by its pumping action, and these air bubbles became trapped in the gill capillaries, creating a large back pressure on the heart. A large-bore stainless steel tube was secured in the ventral aorta to act as an output cannula. The ventral aorta was exposed in the isthmus region, and the cannula was advanced into the ventral aorta so that the tip was located in the region of the bulbus arteriosus. In some preparations the common cardinal veins were tied off. However, in later preparations this step was omitted because it reduced the volume of the sinus venosus. In all preparations the fish was transected near the pelvic fins so that the preparation could fit into the organ bath. In preparations in which the common cardinal veins were not tied off, a closed tube was inserted into the severed caudal vein. These surgical procedures lasted 10–20 min, after which the preparation was transferred to the organ bath. The preparation was fully immersed in saline at 25°C. The input cannula was connected to the perfusate reservoirs via an adjustable constant pressure device (Farrell *et al.* 1983) that set the filling pressure of the heart. The output cannula was connected to an adjustable constant pressure head that set the diastolic output pressure. Cardiac output was measured via an in-line, flow-through electromagnetic flow probe (Zepeda Instruments, Seattle, Wash.). The input and output pressures were continuously measured with U-Onics P-106 (Wayland, Mass.) pressure transducers that were connected to 23-gauge tubes which were inside the stainless steel cannulae and which opened at the orifice of the cannula.

Saline composition

The composition (in mM) of the saline used throughout this study was: 185.7 NaCl, 1.1 $MgCl_2 \cdot 6H_2O$; 7.0 KCl; 1.9 $CaCl_2 \cdot 2H_2O$, 7.1 *N*-tris[hydroxymethyl]methyl-2-aminoethanesulfonic acid (TES, Na salt), 2.8 TES (free acid), and 5.5 dextrose. The final pH at 25°C was 7.7. In previous studies with perfused hearts, tonic levels of adrenaline were found to be important in improving the durability of the preparation. Therefore, in the absence of other information for tunas regarding either perfused hearts or *in vivo* levels of adrenergic stimulation (neural or humoral), adrenaline was added to the saline to a final concentration of 0.1 μM. Aerated saline was used successfully with rainbow trout hearts in which the coronary circulation was not perfused (Farrell *et al.* 1988; Milligan and Farrell 1991). Moreover, rainbow trout hearts performed at work loads comparable to the maximum *in vivo* level for at least 40 min without deterioration, suggesting adequate oxygen delivery to the myocardium in the absence of coronary perfusion. The perfusate used in this study was oxygenated. Therefore, the partial pressure of oxygen (Po_2) of the tuna perfusate was approximately 5 times higher than that used in successful rainbow trout hearts and approximately 20 times higher than that of tuna venous blood (30 mmHg, Bushnell 1988). We anticipated that this supernormal luminal Po_2 would compensate for the fact that the coronary circulation was not perfused. In some experiments lactate and fatty acids were added to the saline.

Instrumentation, calibrations, and data analysis

The flow probe was calibrated with known flow rates of saline. The pressure transducers were calibrated daily with a water manometer. Pressures were referenced to the saline in which the preparation was fully immersed. The pressure and flow signals were suitably amplified and displayed on a Gould chart recorder (Cleveland, Ohio). In addition, an A/D converter (ADDA12, Flytech), interfaced with an IBM AT computer, recorded these signals. Mean pressures, cardiac output, stroke volume, and heart rate were calculated from representative records taken over a 10-s period. Cardiac power output was calculated as follows:

$$\text{power output (mW)} = \text{cardiac output (mL/s)} \times (P_o - P_i)\ (\text{cm } H_2O) \times 0.098$$

where P_o is output pressure, P_i is input pressure, and 0.098 converts mL · cmH_2O/s to mW. Power output is expressed per unit body mass (kg) and per unit ventricular mass (g).

In some preparations, 1-mL saline samples were taken simultaneously from the input and output lines near the heart. The Po_2 in these samples was measured with a Radiometer PHM 73 analyzer (Copenhagen, Denmark) and associated oxygen probe maintained at 25°C. Some samples of perfusate were collected from the output head and used for analysis of lactate (Bergmeyer 1974).

Experimental protocols for yellowfin tuna

Diastolic output pressure was initially set to 60–70 cmH_2O, and the filling pressure in the sinus venosus was set so that cardiac output averaged 80–90 mL/(min · kg). The beating frequency of the perfused heart was set by the intrinsic rate of the sino-atrial pacemaker plus the adrenergic tone provided by the adrenaline in the perfusate. Filling pressure was increased in a stepwise manner to generate a Starling curve for each preparation ($N = 8$). A maximum stroke volume was established when a further increase in filling pressure failed to produce an increase in cardiac output. With cardiac output at this maximum level, diastolic output pressure was increased in a stepwise fashion to examine homeometric regulation by the heart (i.e., the ability of the heart to intrinsically adjust pressure development without compromising flow development). Diastolic output pressure was raised until the heart failed, as indicated by a significant decrease in cardiac output ($N = 6$). A value for maximum power output was established during this procedure. Heart rate decreased to a stable level (the reported mean heart rate) at the outset of the experiment. Maximum heart rate refers to the highest heart rate observed at the beginning of the perfusion.

Experimental protocols for skipjack tuna

The same experimental protocol was attempted for the skipjack tuna with limited success. Data are reported for 10 skipjack tuna heart preparations that performed with a mean output pressure between 70 and 80 cmH_2O, which is near the *in vivo* ventral aortic blood pressure (Bushnell 1988). However, cardiac performance deteriorated rapidly in another 20 preparations. In most of these cases, cardiac output was reasonably high (30–80 mL/(min · kg)) provided that the diastolic output pressure was subphysiological (20–30 cmH_2O). Cardiac deterioration was greatly accelerated when diastolic pressure was increased to a physiological level. Increasing the adrenaline concentration in the perfusate to 1.0 μM did not attenuate the decline. When the pericardium was opened to view the heart in some of these failing preparations, it appeared as if only the atrium was contracting forcefully, with the atrium and ventricle beating at different rates.

Cardiac morphology

The ventricle and atrium were removed after the experiment, blotted on paper towelling, weighed, and fixed in formalin. Compact and spongy myocardial tissues were dissected from the formalin-fixed ventricle and dried to a constant mass. Compact myocardium is reported as a percentage of the ventricle mass.

Pressure–flow relationships for the coronary circulation

Six skipjack tuna (body mass 1.61 ± 0.12 kg) were used. Fish were either stunned by a blow to the head or anaesthetized by immersion in buffered MS-222 (1 g/L). Sodium heparin (10 000 IU) in 0.8 mL of saline was injected into the caudal vessels. The heart was quickly removed via a midventral incision through the pericardial wall and was placed in a dish of ice-chilled saline. Care was taken not to tear the atrium when cutting through the sinus venosus. A PE 50 cannula (i.d. = 0.58 mm, o.d. = 0.97 mm, Clay Adams) was secured in the single coronary artery that lies on the dorsal surface of the ventral aorta. Ten millilitres of heparinized saline (1000 IU/mL) was injected from a syringe into the coronary artery to help clear the coronary circulation of blood and prevent clotting. The cannulated heart was immersed in saline at 25°C for the perfusion studies. Preparation time was approximately 10 min.

The coronary circulation was perfused with saline (see above) under constant flow conditions using a Haake MP2000 peristaltic pump (New Jersey). A windkessel was placed in the inflow line to reduce the large pressure oscillations produced by the pump (Farrell 1987). Perfusion (input) pressure was monitored by a sidearm in the input cannula connected to a U-Onics P106 pressure transducer (Wayland, Mass). Perfusion pressure was referenced to the saline in the organ bath and corrected for the flow-dependent pressure drop along the input cannula, downstream from the measurement site. The cannula resistance was determined at the end of each experiment by measuring the pressure drop at two known flow rates. The suture around the cannula was left in place for this calibration procedure. The pressure signal was suitably amplified and displayed on a Gould chart recorder (Cleveland, Ohio).

Experimental protocol

Flow was initially set at 0.5 mL/(min · kg body mass) to clear blood from the preparation and establish a stable perfusion pressure. The heart was not beating. Pressure–flow curves were generated by measuring stable perfusion pressures at 14 flow rates ranging from 0.01 to 3 mL/(min · kg body mass). The highest flow rate represented approximately 3% of the resting cardiac output measured in spinally blocked skipjack tuna (Bushnell 1988).

Flow was then set to 1 mL/(min · kg body mass) prior to measuring the effects of vasoactive drugs on the coronary circulation. Preparations were tested with no more than three vasoactive drugs, which were injected into the perfusion line as a bolus of 0.1 or 0.2 mL. Dose–response curves were established for L-adrenaline bitartrate, L-noradrenaline bitartrate, L-phenylephrine hydrochloride, DL-isoproterenol, acetylcholine hydrochloride, and adenosine diphosphate (ADP) (Sigma). Perfusion pressure returned to a stable level between successive drug injections. The maximum response to the drugs is presented as a percentage change from the initial coronary resistance.

Calculations

At the end of the experiment the ventricle was weighed and coronary flow rate was recalculated as mL/(min · g ventricular mass). In the absence of information on the drainage of tuna coronary veins and coronary venous blood pressure, we assumed that coronary venous pressure was zero when calculating coronary vascular resistance. Coronary vascular resistance = (input pressure)/(coronary inflow), where resistance is in cmH_2O · min · g ventricular mass/mL, input pressure is in cmH_2O, and coronary inflow is in mL/(min · g ventricular mass). An unknown proportion of the coronary circulation perfuses the spongy myocardium of the tuna heart and empties directly into the ventricle (Tota 1978; Tota 1983; Tota *et al.* 1983), but the effect of this anatomical arrangement on our calculations is unknown.

Mean values and standard error of the mean are presented. Statistical comparisons were made using a Student's *t*-test with the limit of significance set at $p < 0.05$.

Results

Cardiac morphology

The ventricular mass was 0.40 ± 0.01% of body mass in skipjack tuna and 0.29 ± 0.01% of body mass in yellowfin tuna (Table 1). This was a statistically significant species difference. The value for the relative ventricular mass of skipjack tuna is larger than previously reported values for tunas (northern bluefin tuna *Thunnus thynnus* (relative ventricular mass, 0.31% of body mass; Poupa *et al.* 1981) and southern bluefin tuna *Thunnus maccoyii* (relative ventricle mass, 0.29% of body mass; Davie 1987)). Other tunas, which were part of the physiological data set, had similarly sized ventricles. The compact layer of skipjack tuna hearts comprised a significantly greater proportion of the total ventricular mass (65.6 ± 1.2%) compared with yellowfin tuna (54.5 ± 1.2%). The estimates for compact myocardium are higher than other estimates for tunas using the same technique (e.g., 48.5% in southern

TABLE 1. Cardiovascular variables for skipjack and yellowfin tunas

	Skipjack tuna	Yellowfin tuna
Maximum cardiac output (mL/(min · kg body mass))	84.5±5.4	108.0±4.0
Maximum stroke volume		
mL/g ventricular mass	0.17±0.01	0.31±0.02
mL/kg body mass	0.68±0.05	0.87±0.10
Maximum sustainable output pressure (cmH_2O)		
Systolic	109.0±4.1	178.5±6.3
Mean	84.7±4.0	145.1±5.8
Maximum power output		
mW/g ventricular mass	2.55±0.21	7.58±0.44
mW/kg body weight	10.1±0.88	21.9±1.04
Mean heart rate (bpm)	138.5±7.1	123.3±4.2
Maximum heart rate (bpm)	154.0±5.0	137.6±4.8
Body mass (kg)	1.32±0.13	1.96±0.08
Heart mass (g)	5.22±0.44	5.72±0.30
Compact myocardium (%)	65.6±1.23	54.5±1.21
Relative ventricular mass	0.40±0.01	0.29±0.01
N	10	8

NOTE: Values are given as means ± SEM. Maximum cardiac output, maximum stroke volume, and mean heart rate values were obtained from the response to increasing filling pressure. Maximum output pressures and maximum power output values were obtained from the response to increased output pressure. Maximum heart rate was derived from the highest value observed for each preparation. Relative ventricular mass is ventricular mass expressed as a percentage of body mass. All the variables reported for the skipjack tuna were statistically different from those for the yellowfin tuna, with the exception of heart mass.

bluefin tuna; Davie 1987). The atrial mass was 0.05−0.06% of body mass in both species.

Cardiac dynamics in yellowfin tuna

The individual responses of perfused yellowfin tuna hearts to changes in filling pressure are shown in Fig. 1. Two important points emerge. First, physiological stroke volumes were achieved at subambient filling pressures. Second, stroke volume increased with increased filling pressure in accordance with the Frank−Starling mechanism. Generally, the yellowfin tuna heart was most responsive to filling pressure at stroke volumes that produced subphysiological cardiac output values. We were reluctant to extend this relationship by decreasing cardiac output below 60 mL/(min · kg) because a low cardiac output usually led to irreversible cardiac failure. The largest stroke volume on each Frank−Starling curve was termed maximum stroke volume. Accordingly, the maximum stroke volume for perfused yellowfin tuna hearts was 0.87 ± 0.10 mL/kg body mass (Table 1). The maximum cardiac output (108 ± 4.0 mL/(min · kg body mass); Table 1) was 94% of that measured *in vivo* (115 mL/(min · kg body mass); Bushnell 1988).

Heart rates ranged from 101 to 157 bpm in yellowfin tuna. Beyond an initial decrease in heart rate from the maximum heart rate of 137.6 ± 4.8 bpm to a stable value of 123.3 ± 4.2 bpm (Table 1), heart rate was not significantly affected by normal manipulations in filling pressure and diastolic output pressure. However, excessive increases in diastolic output pressure resulted in arrhythmia. Normal cardiac pumping could be restored by immediately lowering the diastolic output pressure.

The individual responses of perfused yellowfin tuna hearts to increases in diastolic output pressure are shown in Fig. 2. The hearts were capable of homeometric regulation in that

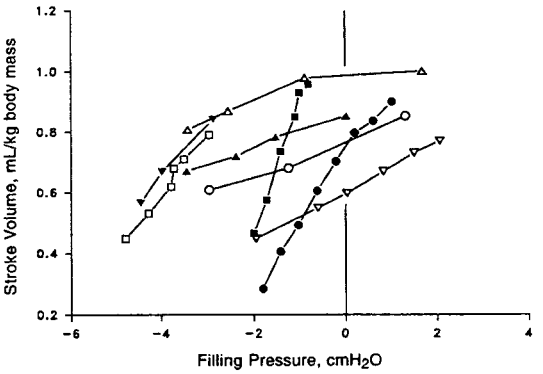

FIG. 1. Individual curves showing responses of cardiac stroke volume to increases in venous filling pressure in perfused yellowfin tuna hearts. There were no further increases in stroke volume beyond the maximum values shown for each curve. It is important to note that the majority of the curves were generated at subambient filling pressures, suggesting a *vis a fronte* filling mechanism for the yellowfin tuna heart.

generally there was little decrement in stroke volume when diastolic output pressures were raised to produce mean output pressures ranging between 130 and 170 cmH_2O (Fig. 2A). The mean maximum output pressure was 145.1 ± 5.8 cmH_2O (Table 1), which exceeds the mean ventral aortic pressure observed *in vivo* by 21% (120 cmH_2O; Bushnell 1988). At the maximum output pressure, cardiac output (97.5 ± 3.5 mL/(min · kg body mass)) was 10% lower than the maximum cardiac output.

Power output increased with output pressure (Fig. 2B). A plateau in power output with increasing output pressure was observed in only one preparation before arrhythmias developed. Therefore, the greatest power output on each curve was termed maximum cardiac power output. Maximum cardiac power output was 7.58 ± 0.44 mW/g ventricular mass or 21.9 ± 1.04 mW/kg body mass (Table 1). Comparisons of cardiac power output relative to body mass are valuable because ventricular mass varies significantly between fish species.

Pulse pressures were unusually large compared with the situation *in vivo*. The average maximum systolic pressure (178 cmH_2O; Table 1) was 33 cmH_2O higher than the mean pressure. This was primarily a result of the high peak flow rate through the output cannula. We minimized cannula resistance by using a tube with the largest bore and shortest length possible. Nonetheless, with peak systolic work being unusually high for a given mean output pressure, maximum power output, which we calculated from the mean output pressure, may underestimate the work performed by the perfused heart.

Cardiac dynamics in skipjack tuna

Although the surgical procedures used for the skipjack tuna and yellowfin tuna were identical, the perfused skipjack tuna heart performed poorly. Typically, hearts could generate a near physiological cardiac output but only at subphysiological diastolic output pressures. Moreover, increasing diastolic output pressure to physiological levels often produced rapid declines in cardiac output and irreversible cardiac failure. The ability of the perfused heart to generate pressure deteriorated

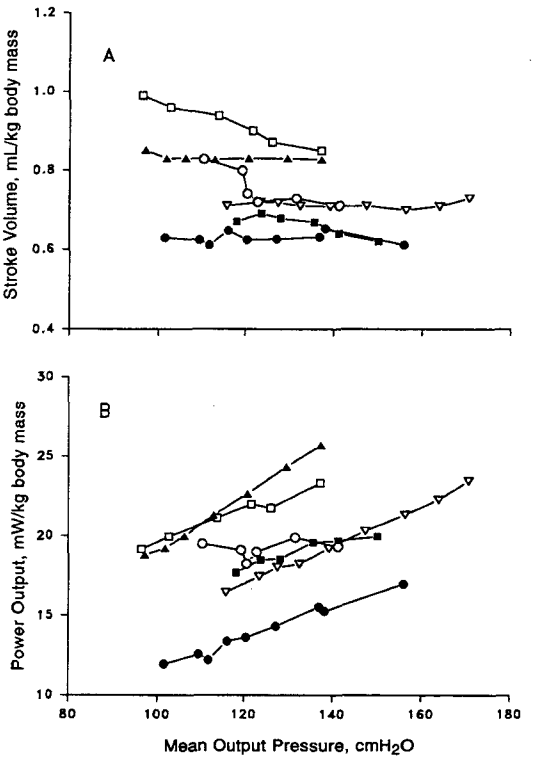

FIG. 2. Individual curves for the response of cardiac stroke volume (A) and power output (B) to increases in diastolic output pressure in perfused yellowfin tuna hearts. Stroke volume was largely independent of output pressure over the range 130–170 cmH_2O. As a result, increases in output pressure produced significant increases in myocardial power output up to the maximum indicated. Increases in output pressure beyond those shown resulted in cardiac arrhythmia. An incomplete range of output pressures was tested on two additional preparations, and these are not included in the figure.

with time. Preparations rarely lasted 30 min. To check whether or not extracellular substrate was a limiting factor, 0.7 mM palmitate in 10% bovine serum albumin, or 5 mM lactate, or both were included in the perfusate in 4 preparations. Addition of these substrates did not improve either the maximum performance or the longevity of the preparation. In an attempt to understand the proximate cause of cardiac failure, lactate levels were measured in the output perfusate. In 5 heart preparations, lactate release into the perfusate ranged from 0.4 to 1.2 μmol/(min · g ventricular mass), indicating substantial anaerobic metabolism. Lactate release into the perfusate could not be quantitatively related to cardiac work, but the rate of lactate release was comparable to the rate of release in severely hypoxic, working, perfused trout hearts (Arthur *et al.* 1992). The change in P_{O_2} between input and output perfusate samples was never more than 100 Torr (1 Torr = 133.3 Pa). This means that oxygen was not depleted from the perfusate and the diffusion gradient driving oxygen from the lumen into the myocardium was always at least 10 times greater than that normally provided by the venous blood P_{O_2} (Bushnell 1988). The most likely explanation for high lactate production, despite a very high luminal P_{O_2}, is that oxygen delivery to the myocardium was inadequate.

Attempts to improve oxygen delivery were limited because of the difficulty in cannulating the coronary artery (see below) and a dwindling supply of skipjack tuna. The coronary circulation was perfused with oxygenated saline in 2 preparations by means of an autoperfusion system (connecting a sidearm from the ventral aortic output cannula to the main coronary artery). This procedure did not improve cardiac performance appreciably. Constant pressure perfusion of the coronary circulation with a red blood cell suspension (hematocrit 10%) was attempted in one additional preparation. Red blood cells appeared in the ventral aortic perfusate after less than 1 min, but the coronary flow rate was not measured. Even though there were eventually problems with blood clots, this was the only skipjack tuna heart preparation that performed for longer than 30 min and at output pressures that were physiological.

In view of the above, the data for saline-perfused hearts from skipjack tuna (Table 1) have limited value. In the 10 successful preparations, maximum cardiac output (84.5 ± 5.4 mL/(min · kg body mass)) was 64% of the *in vivo* value (132 mL/(min · kg body mass); Bushnell 1988) when mean output pressure was 74 cmH_2O. The highest individual cardiac output was 112 mL/(min · kg body mass). The highest individual stroke volume was 0.86 mL/kg body mass and was lower than that *in vivo* (1.1 mL/kg body mass; Bushnell 1988). The heart responded to filling pressure, which was often negative, by increasing stroke volume. The range for heart rate in the perfused skipjack heart was 91.1–172.4 bpm. At a mean output pressure of 87.4 ± 4.0 cmH_2O, power output increased to a maximum value of 2.55 ± 0.21 mW/g ventricular mass or 10.1 ± 0.88 mW/kg body mass (Table 1) with only a slight decrease in cardiac output (82.9 ± 3.5 mL/(min · kg body mass)). Higher output pressures significantly decreased cardiac output. All of the maximum values for cardiac variables were significantly lower than those for the yellowfin tuna (Table 1), with the exceptions of the mean (138.5 bpm) and maximum heart rates (154 bpm), which were significantly higher.

Pressure–flow relationships for the skipjack tuna coronary circulation

The pressure–flow relationship for 6 skipjack tuna hearts is summarized in Fig. 3A. The curves for all fish were convex toward the pressure axis at low flow rates, becoming linear at higher flow rates. Vascular resistance decreased exponentially as flow rate (and perfusion pressure) increased. Coronary vascular resistance reached a constant minimum of approximately 24 cmH_2O · min · g ventricular mass/mL at a flow rate of 2 mL/(min · g ventricular mass) (with perfusion pressure approximately 40 cmH_2O) or higher (Fig. 3B). At high flow rates, 4 of the 6 hearts began to beat spontaneously and eject perfusate from the ventral aorta.

Adrenaline and noradrenaline (α- and β-adrenergic agonists) produced similar concentration-dependent increases in coronary resistance (Fig. 4). Phenylephrine (an α-adrenergic agonist) was a less effective vasoconstrictor. Hearts began to beat more vigorously at high concentrations of adrenaline and noradrenaline, but not with phenylephrine. The amount of perfusate ejected was greater than that being delivered to the heart via the coronary circulation, and we assume that the excess was drawn from the organ bath via the atrium. The highest

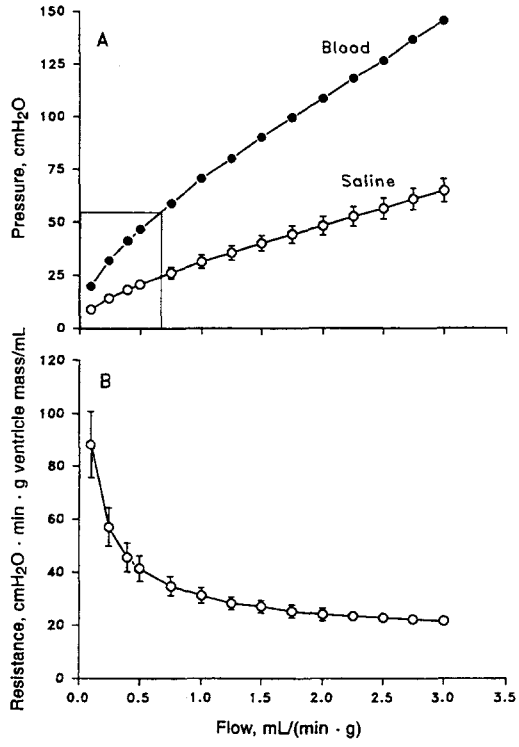

FIG. 3. The coronary perfusion pressure (A) and coronary vascular resistance (B) measured in skipjack tuna hearts perfused with saline at different flow rates. Each point represents a mean value for 6 preparations, and the standard error of the mean is indicated by the vertical bars. In panel A, a pressure–flow relationship is estimated for blood, by adjusting the saline line to account for the difference in the viscosities of tuna blood and the saline perfusate. There are no measurements of kinematic blood viscosity in tuna, and so a value of 2.5×10^{-3} Pa · s was assumed. This value is based on an assessment of known blood viscosities in fish (e.g., Wood 1974; Graham 1985; Graham and Fletcher 1983; Wells and Baldwin 1990; Baldwin and Wells 1990; Wells and Weber 1991). Saline was assumed to have a viscosity of 1×10^{-3} Pa · s at 25°C (Graham 1985). From the curve for blood an *in vivo* coronary blood flow was estimated for a dorsal aortic blood pressure of 55 cmH_2O (Bushnell 1988).

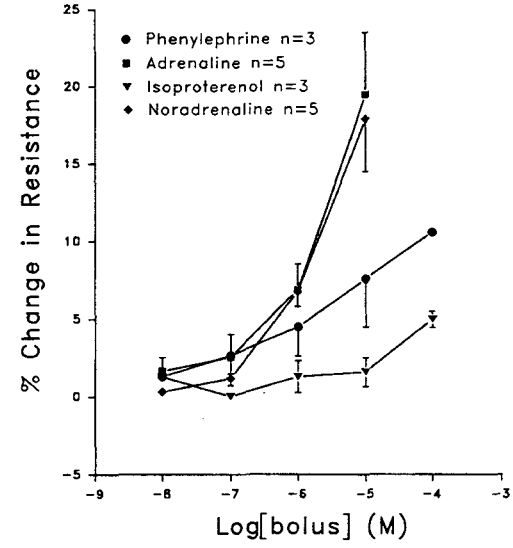

FIG. 4. The vascular reactivity of the coronary circulation in skipjack tuna perfused with oxygenated saline at a constant flow rate (1 mL/(min · g ventricular mass)). Each drug was injected as a bolus (0.1 mL) into the perfusion line. The concentration of the bolus is presented. Responses (percentage change from the baseline resistance) are presented as mean values for 3–5 preparations, and the standard error of the mean is indicated by the vertical bars.

concentration of isoproterenol (a β-adrenergic agonist) made the hearts beat more vigorously and also produced a modest increase in vascular resistance (+5%).

Acetylcholine produced a small increase in vascular resistance in 1 preparation and a small decrease in resistance in another. ADP produced a small decrease in vascular resistance in 1 preparation and no response in another. In the preparation that responded to ADP, vascular resistance was initially slightly elevated (a residual effect from an earlier adrenaline injection) and vascular resistance was reduced only to the normal baseline level. The preparation that did not respond to ADP also did not respond to acetylcholine or adrenaline.

In a preliminary experiment, noradrenaline (10 μM) and acetylcholine (10 μM) were injected together in 1 preparation. The resulting contraction was stronger and lasted several minutes longer than that normally associated with noradrenaline alone. Similarly, when ADP (10 μM) and noradrenaline (10 μM) were injected together in 1 preparation, the resulting constriction was more prolonged but was reduced compared with that normally associated with noradrenaline alone.

Discussion

The study of tuna cardiovascular physiology *in vivo* is hampered by the supply, holding, and handling of the fish, and the fact that tunas are normally continuous swimmers. Probably the best set of cardiovascular data are from spinally blocked tunas (Bushnell 1988), even though it is difficult to relate these data to a normal physiological state. Based on our past experience with perfused fish hearts (six fish species, including two active ones), we expected that an *in situ* preparation would provide information regarding maximum cardiac performance in tunas. In view of this expectation, perhaps the most important observation was that saline-perfused preparations of yellowfin and skipjack tuna hearts were unable to match all of the cardiac variables previously measured in live, spinally blocked tunas (Bushnell 1988). The yellowfin tuna heart preparations performed very near, but not above, the *in vivo* cardiac output, whereas the skipjack tuna heart preparations performed below the *in vivo* cardiac output. Intrinsic heart rates were, nevertheless, greater than the *in vivo* values. Interestingly, the maximum *in vivo* power output in rainbow trout is 7.2 mW/g ventricular mass and this can be simulated *in vitro* with aerated saline and no coronary perfusion (Milligan and Farrell 1991). The maximum power output obtained for perfused yellowfin tuna hearts was similarly 7.58 mW/g ventricular mass.

Several observations lead us to conclude that, in the absence of coronary perfusion, the relatively thick ventricle wall of the perfused skipjack tuna heart prevented the supernormal oxygen gradient from providing a sufficient rate of oxygen diffusion from the lumen to support a physiological work output. First, high levels of lactate appeared in the perfusate leaving skipjack tuna hearts. Second, perfused hearts were very sensitive to increases in diastolic output pressure (i.e., an increase in myocardial oxygen demand with no change in luminal oxygen supply). Third, perfused hearts were also very sensitive to reductions in cardiac output (i.e., a decrease in luminal oxygen supply). Fourth, a much higher cardiac performance was possible in yellowfin tuna hearts, which were 38% smaller than the hearts of skipjack tuna. In fact, we found a significant negative correlation between mass specific maximum power output and ventricular mass for skipjack tuna (power output = $15.2 - 1.18$(ventricular mass); $r = 0.502$, $p < 0.05$).

Thus, even though the relatively larger heart in tunas is advantageous in terms of cardiac performance, the heart appears to be necessarily more dependent on its coronary circulation to overcome increased diffusion distances from the lumen. It seems likely that tuna hearts, particularly those of skipjack tuna, cannot support resting levels of cardiac performance without a coronary circulation. This contrasts with salmonids; they survive after the coronary artery has been ligated (to eliminate the coronary route for myocardial oxygen supply) and then can swim quite well in normoxic and hypoxic water (Farrell and Steffensen 1987; Farrell *et al.* 1990; J. F. Steffensen and A. P. Farrell, unpublished observations).

We made limited attempts to perfuse the coronary circulation and thereby improve oxygen delivery to the *in situ* heart. Coronary cannulation was technically very difficult, and the results obtained were inconclusive. The coronary artery lies next to the thin-walled jugular veins. Thus, it was very difficult to tie the coronary cannula in place without puncturing the veins. Once punctured, leakage of perfusate from the sinus venosus via the jugular veins was substantial. Another technical problem related to the choice of fluid to perfuse the coronary circulation. The suggestion that air- or oxygen-equilibrated salines are adequate for perfusion studies with fish hearts (Perry and Farrell 1988) is clearly not valid for tuna hearts, given the present observations. Perfusion with a red blood cell suspension appears to be a more promising option, given the present observation of improved preparation viability. Even so, the difficulty in obtaining sufficient tunas to provide a pool of blood donors and preventing blood clotting may favour the use of perfluorinated carbon compounds in the future.

The observation that active fishes with higher blood pressures have more compact myocardium has lead to the suggestion that the compact layer is more important in pressure development (see reviews by Tota 1983, 1989). Physiological studies also support this idea. Coronary ligation in hypoxic, swimming rainbow trout reduces ventral aortic blood pressure (J. F. Steffensen and A. P. Farrell, unpublished observations). Furthermore, coronary perfusion with red blood cells in hypoxic dogfish and eel hearts improves the ability of the heart to develop pressure (Davie and Farrell 1991*a*; Davie *et al.* 1992). The present observation of a limited ability to generate high output pressures by perfused tuna hearts is also consistent with the idea of the compact myocardium being more important in pressure development.

Only small increases in cardiac filling pressure are needed for two- to three-fold increases in stroke volume with perfused hearts from other fishes (sea raven, rainbow trout, ocean pout (*Macrozoarces americanus*), eel (*Anguilla dieffenbachii*), dogfish (*Squalus acanthias*), and kingfish (*Seriola lalandi*)) (Farrell 1984; Farrell *et al.* 1985; Farrell *et al.* 1986; Davie and Farrell 1991*a*; Davie *et al.* 1992; D. Hutchison, C. E. Franklin, and P. S. Davie, unpublished observations). In contrast, perfused yellowfin tuna hearts were less sensitive to filling pressure. There are two potential explanations for this weak response to filling pressure: either the maximum stroke volume observed here is close to the maximum *in vivo* value, or stroke volume was severely limited by insufficient oxygen delivery to the myocardium. We favour the former explanation for a number of reasons. First, stroke volume did not increase beyond the observed maximum level when diastolic output pressure was reduced (i.e., oxygen demand was lower). In fact, perfused yellowfin tuna hearts could maintain stroke volume at output pressures higher than those *in vivo*. Second, when yellowfin tuna and skipjack tuna were exposed to moderate environmental hypoxia (>80 Torr), stroke volume was unchanged and cardiac output declined in proportion with the bradycardia (Bushnell 1988; Bushnell *et al.* 1990). In contrast, other fish exposed to moderate levels of hypoxia typically developed bradycardia that was accompanied by an increase in stroke volume (Randall 1970; Farrell 1984; Chan 1986; Peyraud-Waitzenegger and Soulier 1989). Therefore, we propose that the maximum anatomical and physiological limit for stroke volume in yellowfin tuna is near the value of 1.3 mL/kg body mass, as determined for spinally blocked yellowfin tuna by means of a dye dilution technique (Bushnell 1988).

If this conclusion proves to be correct, tunas would necessarily alter heart rate to increase cardiac output appreciably, with stroke volume being relatively fixed. This type of cardiac pumping strategy resembles that in mammals rather than that in other teleosts, in which increases in stroke volume are the primary means of increasing cardiac output during exercise (see Farrell 1991). Brill and Bushnell (1991) analyzed the existing cardiovascular data for tunas and suggested that cardiac output would increase approximately twofold to account for the maximum oxygen uptake in skipjack tuna. Since the heart rate in tunas spans a twofold range (see Introduction), there is no need to postulate a major increase in stroke volume.

An interesting pattern may be emerging if the maximum stroke volume in tunas is indeed near 1 mL/kg body mass. Maximum stroke volumes are also near 1 mL/kg body mass in other fishes capable of impressive swimming feats, such as rainbow trout (Graham and Farrell 1989), kingfish (D. Hutchison, C. E. Franklin, and P. S. Davie, unpublished observations), and eel (Davie *et al.* 1992). This stroke volume may represent the upper limit for active teleosts that develop high levels of blood pressure and cardiac output. In contrast, hemoglobin-free icefish, with a high cardiac output and a low blood pressure, have a very high stroke volume (estimated at 4 mL/kg body mass; Hemmingsen *et al.* 1972). Further studies on the interrelationships between chamber size, pressure development, wall thickness, and oxygen delivery to the myocardium would be worthwhile to develop a general hypothesis regarding the maximum cardiac stroke volume in fishes.

Lai *et al.* (1987) observed subambient intrapericardial pressures in albacore (*Thunnus alalunga*) and suggested *vis a fronte* atrial filling. The subambient filling pressure required

to generate near normal stroke volumes in perfused yellowfin tuna hearts is consistent with *vis a fronte* atrial filling. Our most graphic observation in support of *vis a fronte* filling was the aspiration of air into the heart when the hepatic vein was cut. *Vis a fronte* cardiac filling is also found in elasmobranchs (Satchell 1971) and rainbow trout (Farrell *et al.* 1988). Farrell *et al.* (1988) note that the rigid pericardial cavity, which is necessary for *vis a fronte* filling, restricts any increase in the volume of the cardiac chambers, e.g., when stroke volume is increased. Elasmobranchs solve this problem by displacing pericardial fluid into the peritoneal cavity when stroke volume and intrapericardial pressure increase (Shabetai *et al.* 1985). In contrast, tuna may circumvent this particular problem by varying heart rate instead of stroke volume to increase cardiac output.

Myocardial power output has not been calculated previously for tunas. We used the ventral aortic blood pressure and cardiac output reported for spinally blocked yellowfin tuna and skipjack tuna (Bushnell 1988; Brill and Bushnell 1991), and assumed a relative ventricular mass as reported in Table 1 to estimate myocardial power output *in vivo*. In skipjack tuna, power output is 4.7 mW/g ventricular mass (or 18.8 mW/kg body mass), and in yellowfin tuna, power output is 5.6 mW/g ventricular mass (or 16.8 mW/kg body mass). Compared with these *in vivo* values, the maximum myocardial power output in perfused yellowfin tuna hearts (7.58 mW/g ventricular mass; Table 1) could exceed the *in vivo* value by 35%, whereas perfused skipjack tuna hearts produce only 54% of the *in vivo* value. Surprisingly, the *in vivo* power output per gram ventricular mass in spinally blocked tuna is less than the maximum value for rainbow trout (7.2 mW/g ventricular mass, see Farrell *et al.* 1988). To what degree cardiac performance in tunas increases beyond these levels is not clear. We measured levels 35% higher in yellowfin tuna. Brill and Bushnell (1991) suggest that cardiac output could double. If myocardial power output per gram ventricular mass can be twice that in rainbow trout, the biochemical and physiological organization responsible for a difference of this sort are worth investigating.

The truly exceptional performance of the tuna heart becomes apparent when the large ventricle is considered. The yellowfin tuna and skipjack tuna ventricles are over 4 times and almost 6 times larger, respectively, than the rainbow trout ventricle (0.07% of body mass at 15°C; Graham and Farrell 1990). When myocardial power output is expressed per kilogram body mass to account for the species difference in relative heart size, the power outputs for skipjack and yellowfin tunas (18.8 and 16.8 mW/kg body mass, respectively) are nearly 10 times greater than the maximum value for rainbow trout (2.0 mW/kg body mass, Graham and Farrell 1990).

The present study established a pressure–flow relationship for the coronary circulation of skipjack tuna. It is possible to estimate *in vivo* coronary flow from this relationship (Fig. 3A) by adjusting for the difference in viscosities for blood and saline and using a known value for dorsal aortic blood pressure in skipjack tuna (55 cmH_2O; Bushnell 1988; Brill and Bushnell 1991). Since there are no measurements of kinematic blood viscosity in tuna, a value of 2.5×10^{-3} Pa · s was assumed. This value is based on an assessment of known blood viscosities in fish (e.g., Wood 1974; Graham 1985; Graham and Fletcher 1983; Wells and Baldwin 1990; Baldwin and Wells 1990; Wells and Weber 1991). Obviously, the uncertainty regarding the exact blood viscosity in tunas has an important impact on the flow estimate. If tuna blood viscosity is 2.0×10^{-3} Pa · s, coronary blood flow *in vivo* would be 20% lower than that presented. Conversely, if tuna blood viscosity is 3.0×10^{-3} Pa · s, coronary blood flow *in vivo* would be 20% greater than that presented.

TABLE 2. Predicted coronary flow rates in skipjack tuna and rainbow trout

Coronary flow rate	Skipjack tuna	Rainbow trout[a]	Ratio
mL/(min · g ventricular mass)[b]	0.67	0.38	1.76
mL/(min · g compact tissue)[c]	1.02	0.97	1.03
mL/(min · kg body mass)[d]	2.60	0.27	9.6
Proportion of resting cardiac output[e]	1.9%	1.5%	1.31

[a]Data taken from Farrell (1987).
[b]Value interpolated from Fig. 3A.
[c]The coronary circulation perfuses only the compact myocardium in rainbow trout. The exact perfusion pattern in tuna is unknown. The porportion of compact myocardium is greater in the skipjack tuna ventricle (65%) than in the rainbow trout ventricle (39%). The value was calculated as (mL/(min · g ventricular mass))/0.65.
[d]The skipjack tuna ventricle (0.4% body mass) is relatively larger than the rainbow trout ventricle (0.07% body mass). The value was calculated as (mL/(min · g ventricular mass)) × (% relative ventricular mass) × 10.
[e]Cardiac output in spinally blocked skipjack tuna (132 mL/(min · kg body mass)) is higher than in resting rainbow trout (17 mL/(min · kg body mass)). The value was calculated as 100 × (coronary flow, in mL/(min · kg body mass))/(cardiac output, in mL/(min · kg body mass)).

The estimate of coronary blood flow for the above conditions is 0.67 mL/(min · g ventricular mass) (Fig 3A; Table 2). From a physiological standpoint, this estimate probably should be regarded as a maximum flow rate for a given blood pressure because there was limited vascular tone in the coronary circulation (see below), and extravascular compression of coronary circulation was negligible (the afterload was minimal and the heart was not beating except at high flow rates). Extravascular compression, as a result of increasing heart rate, increased vascular resistance by 20–25% in the perfused rainbow trout hearts (Farrell 1987) and by 6–14% in the mammalian hearts (Feigl 1983).

The estimated coronary blood flow of 0.67 mL/(min · g ventricular mass) for skipjack tuna is 76% higher than the estimated value for rainbow trout obtained with an identical approach (Farrell 1987). The difference reflects a somewhat lower minimum vascular resistance in skipjack tuna (24 cmH_2O · min · g ventricular mass/mL; Fig. 3B) compared with rainbow trout (36 cmH_2O · min · g ventricular mass/mL; Farrell 1987) and a higher dorsal aortic blood pressure in skipjack tuna. The compact myocardium is proportionately larger in skipjack tuna. When coronary flow rate is expressed per unit mass of compact myocardium, the coronary flow rates are remarkably similar for the two species (Table 2). However, total coronary flow for skipjack tuna (expressed per kilogram body mass) is 9.6 times greater than that for rainbow trout (Table 2) because the tuna ventricle is approximately 6 times larger than that of the rainbow trout heart.

Cardiac output is also approximately 6 times greater in skipjack tuna than in rainbow trout. Therefore, the relative proportion of cardiac output going to the skipjack tuna heart is 1.9% (Table 2). Based on estimates with microspheres, coronary flow was 0.65% of cardiac output in the sucker (*Catastomus catastomus*) and 0.56% of cardiac output in the burbot (*Lota lota*) (Cameron 1975). Based on pressure–flow curves and similar assumptions to those used here, coronary flow in rainbow trout was estimated at 1.5% (Farrell 1987). Thus, even

though the proportion of cardiac output going to the coronary circulation is the highest so far reported for fish, the value is only 31% higher than for rainbow trout. Given the uncertainty regarding blood viscosity in tuna, the most important point to emerge from the above theoretical analysis is that the very high body mass specific cardiac output in skipjack tuna appears to be matched by a very high body mass specific coronary blood flow rate.

Based on the present study some tentative conclusions about the control of coronary flow in skipjack tuna can be made. From Fig. 3A the dorsal aortic blood pressures needed to double (85 cmH_2O) or triple (110 cmH_2O) coronary blood flow are high relative to *in vivo* values. While blood pressure is likely a major determinant of the coronary blood flow rate, a vasodilatory reserve could also contribute to hyperemia, as is the case in the mammalian heart (Feigl 1983). Adrenaline and noradrenaline constrict the coronary circulation of skipjack tuna, rainbow trout, and blue marlin (*Makaria nigricans*) (Davie and Daxboeck 1984; Farrell 1987). Thus, an α-adrenergic tonic vasoconstriction could be involved in the coronary vasodilatory reserve in fish. Adrenergic coronary vasoconstriction in rainbow trout is temperature dependent, being much less pronounced at 15 than at 5°C (Farrell 1987). Correspondingly, the adrenergic vasoconstrictions in skipjack tuna at 25°C were modest relative to those in rainbow trout. Synergistic interactions with catecholamines are worth further study. Our preliminary observations indicate that either ADP and noradrenaline, or acetylcholine and noradrenaline, prolong and alter the intensity of adrenergic vasoconstrictions. Synergistic actions of noradrenaline and adenosine have been reported previously for atrial and ventricular strips from the flounder (*Platichthys flesus*) (Lennard and Huddart 1990), and for mammalian cardiac tissue (Wilson and Broadley 1989). Pharmacological agents expected to significantly dilate the coronary circulation (isoproterenol and ADP) did not do so, probably because there was no vascular tone against which dilatation could be measured. The modest increase in vascular resistance with isoproterenol was a result of either extravascular compression as the heart began to contract, or a nonspecific action of isoproterenol which normally dilates coronary vessels in other fish species (Davie and Daxboeck 1984; Farrell and Graham 1986; Small *et al.* 1990; Davie and Farrell 1991*b*; Farrell and Davie 1991). While *in vitro* perfusion studies will continue to be useful in identifying potential vasoactive mechanisms, measurements of coronary blood flow *in vivo* are needed to fully understand coronary circulatory control in fish.

In summary, tunas are very active fishes and need high rates of oxygen delivery to their tissues. High levels of blood flow, ventral aortic blood pressure, and blood oxygen carrying capacity are needed to sustain this activity. High flow and pressure are generated by a large heart which has a relative mass comparable with that of mammalian hearts. Because of the ventricle's high myocardial oxygen demand and large diffusion distance from the lumen, venous blood is unlikely to ever satisfy the normal myocardial oxygen demand. Thus, tunas differ from other fishes examined so far in that venous blood apparently cannot support resting cardiac performance; a coronary circulation is essential. Furthermore, the very high body mass specific cardiac output in skipjack tuna appears to be matched by a very high body mass specific coronary blood flow rate. The outer compact myocardium and its coronary oxygen supply appear to be more important for pressure development by the heart than for flow output. Consequently, if dorsal aortic blood pressure is reduced, either through impaired cardiac performance or elevated gill resistance, then coronary flow will likely decrease. The effects of interruption of coronary flow in tunas are probably similar to those in mammals, where ischemia depresses cardiac performance which, in turn, further impairs coronary flow and infarction ensues.

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