Circulation during intermittent lung ventilation in the garter snake *Thamnophis*

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Received April 26, 1977

BURGGREN, W. 1977. Circulation during intermittent lung ventilation in the garter snake *Thamnophis*. Can. J. Zool. **55**: 1720–1725.

Systemic and pulmonary arterial and ventricular blood pressures have been recorded in both lightly anaesthetized and unanaesthetized, unrestrained garter snakes of the genus *Thamnophis*. Systemic blood pressure was 65/44 cmH_2O compared with only 52/21 cmH_2O in the pulmonary circulation. Peak systolic pressures throughout the ventricle were identical and were the same as systemic systolic pressures. Reduction of pulmonary arterial blood pressure arose solely from an impedance to blood flow located in a narrow region of the pulmonary ventricular outflow tract. Atropine greatly reduced this impedance. No changes in the blood pressure relationships within the central arterial circulation were observed during intermittent, voluntary lung ventilation. It is concluded that the ventricle of the snake *Thamnophis* functions as a single pump which perfuses both the pulmonary and systemic circulations, with pulmonary and systemic arterial pressure differences arising from the impedance characteristics of the ventricular outflow tracts.

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On a relevé les pressions sanguines systémiques ainsi que les pressions dans l'artère pulmonaire et le ventricule de couleuvres du genre *Thamnophis* légèrement anesthésiées ou non anesthésiées, gardées en liberté surveillée. La pression sanguine systémique s'établissait à 65/44 cmH_2O, alors que la pression dans la circulation pulmonaire n'atteignait que 52/21 cmH_2O. Les pressions systoliques maximales étaient uniformes et égales aux pressions systoliques systémiques. La réduction de la pression sanguine dans l'artère pulmonaire résulte de la seule entrave rencontrée par la circulation sanguine dans un passage étroit du tractus ventriculaire pulmonaire à sa sortie du cœur. L'atropine réduit considérablement l'importance de cette entrave. On n'a observé aucune modification de pression au sein de la circulation artérielle centrale lors de la ventilation intermittente et volontaire des poumons. On en a conclu que le ventricule de la couleuvre *Thamnophis* fonctionne à la manière d'une pompe unique qui alimente à la fois les voies des circulations pulmonaire et systémique, les différences entre les pressions artérielles pulmonaire et systémique résultant de l'opposition qu'offre à la circulation sanguine les voies d'écoulement entre le ventricule et l'artère pulmonaire.

[Traduit par le journal]

Introduction

The cardiovascular dynamics of the Ophidia, whose cardiac anatomy conforms in most respects to that of the other noncrocodilian reptiles (Mathur 1944; White 1968), has yet to come under close scrutiny. In one of the very few studies involving simultaneous measurements of intraventricular and arterial blood pressures, Johansen (1959) found systolic blood pressures to be superimposable in the ventricular chambers and in the systemic and pulmonary arteries of restrained grass snakes. The ventricle thus apparently served as a single pump ejecting blood into both the pulmonary and systemic circulations. However, no study since this time has addressed itself to determining both systemic and pulmonary arterial and ventricular blood pressures in snakes. The extent of functional ventricular separation in unrestrained, unanaesthetized snakes also remains to be established. Nor has the influence on snake cardiovascular function of intermittent lung ventilation, now known to be an important factor in the cardiovascular dynamics of other reptiles, been considered. To these ends, the present investigation reports measurements of arterial and intraventricular blood pressures during periods of apnoea and lung ventilation in both lightly anaesthetized and completely unanaesthetized, unrestrained garter snakes of the genus *Thamnophis*.

Methods

Experiments were performed on nine garter snakes of the genus *Thamnophis* ranging in body weight from 29 to 75 g. The three species *T. elegans*, *T. radix*, and *T. sirtalis* were represented. All experiments were performed at 28–30°C.

Blood pressure measurements were made in six acute, lightly anaesthetized preparations and in three unrestrained snakes after recovery from anaesthesia. All

snakes were initially anaesthetized with ether vapor. The animal was then carefully fastened ventral side up to the surgical table with strips of masking tape, being careful not to compress the rib cage.

In acute experiments, a 3- to 4-cm-long median incision was made in the ventral body wall and the heart and arterial arches were exposed. Blood pressure cannulae (PP50 polyethylene tubing fitted with PP10 tips) were used to nonocclusively cannulate the pulmonary artery and the right aorta or left or right carotid artery. In all experiments, cannulae tips were positioned facing upstream towards the heart. The tips of arterial cannulae were not sutured in place as the arterial walls usually sealed tightly around them and hence the cannulae tips could be moved towards the heart and manoeuvered through the valves and into the ventricular chambers with negligible blood loss. In several experiments, PP20 cannulae were carefully introduced through needle stab wounds into the cavum venosum and cavum pulmonale of the heart to allow direct recording of intraventricular blood pressures. (The cavum venosum of the heart of the snake and other squamate reptiles receives blood from the right auricle and the cavum arteriosum and ejects blood directly into the systemic arteries; the cavum pulmonale receives all of its blood from the cavum venosum and ejects blood into the pulmonary artery (White 1968; Shelton and Burggren 1976).) Snakes prepared for acute experimentation were allowed to recover from anaesthesia to the point where ventilatory movements were spontaneously resumed and were thereafter maintained in this state of light anaesthesia with ether vapour.

In preparations for chronic blood pressure recording, a 2- to 3-cm-long median incision was made in the ventral body wall some 4 cm posterior to the apex of the ventricle. The lung was exposed and a small side branch of the pulmonary artery running over the lung parenchyma was cannulated with a 40-cm length of PP20 cannula. The cannula tip was advanced from the side branch slightly upstream into the larger pulmonary artery. The dorsal aorta was also cannulated with a 40-cm length of PP20 cannula implanted about 2 cm posterior to the confluence of the left and right aortae. Both pulmonary and systemic cannulation points were about equidistant from the heart. The cannulae were sewn into place in the blood vessel walls and then led out through the ventral incision in the body wall, which was finally closed with interrupted sutures. After a 2- to 5-h period of recovery from anaesthesia, the snake was placed in small aquarium containing 5 cm of water. Systemic and pulmonary blood pressures could be recorded in unrestrained animals and cannulae usually remained patent for 2–3 days. All cannulae were filled with heparinized saline and 20 IU/100 g body weight of heparin were injected into both acute and chronic preparations at the start of each experiment.

The cannulae were connected to Statham BB fluid pressure transducers whose outputs were recorded on an E and M Instruments Physiograph Six rectilinear chart recorder. Frequency and damping characteristics of the pressure monitoring and recording system were assessed at the beginning of each experiment to ensure that the system was adequate to record blood pressure profiles without significant phase or amplitude error.

Breathing frequency was monitored in both acute and chronic preparations by direct observation of body wall movements during inspiration and expiration, and an event marker was activated to indicate these events on the records.

Atropine in a dosage of 1.0 mg/kg body weight in a carrier volume of 0.3 ml saline was administered during the course of the experiments to five snakes.

Results

Measurements of cardiovascular performance in *T. elegans*, *T. radix*, and *T. sirtalis* revealed no differences between species, nor were there any differences evident between lightly anaesthetized snakes and those which had completely recovered from anaesthesia and were unrestrained. Hence, Table 1 presents pooled data on heart rate and pulmonary and systemic blood pressures recorded from nine *Thamnophis* under varying experimental conditions.

No variations in heart rate associated with voluntary intermittent lung ventilation were evident in any of the snakes when swimming or when in air. Mean systolic pressures in the systemic arterial circulation in *Thamnophis* were about 12–15 cmH_2O higher than in the pulmonary circulation (Table 1). This is clearly illustrated in Fig. 1, which presents blood pressure records from the right carotid and pulmonary arteries of a lightly anaesthetized *T. elegans*. Blood pressure began to rise in the pulmonary artery some 100 ms before it rose in the systemic circulation, but pressure began to rise in the systemic circulation well before a similar pressure was attained in the pulmonary artery. In fact, at no time during systole were systemic and pulmonary arterial blood pressures ever superimposable (Fig. 1B). Pulmonary pulse pressure in most *Thamnophis* was only 8–12 cmH_2O greater than systemic pulse pressure (Table 1). Nonetheless, the rate of fall of pulmonary arterial pressure during diastole was much greater than in the systemic circulation of the snake, suggesting that the pulmonary vascular bed had a greater compliance and (or) lower impedance than the systemic bed.

A markedly lower pulmonary arterial blood pressure compared with that in the systemic circulation was evident not only in acute preparations (Fig. 1) but also in freely moving, unanaesthetized snakes. Figure 2 shows records of dorsal aorta and pulmonary artery blood pressure measured in a *T. elegans* which was breathing intermittently and also spontaneously swimming for short distances in the aquarium. Importantly, pulmonary arterial pressure was much lower than systemic pressure during both apnoea and

TABLE 1. Cardiovascular parameters measured in nine *Thamnophis*. Values for each snake were derived from five measurements made during a stable cardiovascular state. Mean values ± 1 SD are given for pooled data

Species	Body weight, g	Pulmonary blood pressure, cmH_2O		Systemic blood pressure, cmH_2O		Heart rate, beats/min
		Systolic	Diastolic	Systolic	Diastolic	
Thamnophis radix	75.3	50	31	82	61	51
Thamnophis radix	41.3	46	19	61	36	49
Thamnophis elegans	55.6	38	12	52	35	47
Thamnophis elegans	48.7	53	16	58	40	53
Thamnophis elegans	29.0	57	16	59	44	54
Thamnophis elegans	38.0	33	17	41	24	49
Thamnophis sirtalis	33.5	89	21	96	72	57
Thamnophis sirtalis	35.0	61	48	75	53	62
Thamnophis sirtalis	37.0	40	13	59	35	31
Mean values	43.8 ± 14.3	52 ± 17	21 ± 11	65 ± 17	44 ± 15	50 ± 9

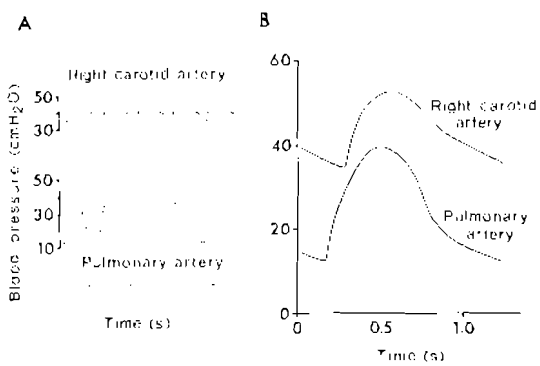

FIG. 1. (A) Records of systemic and pulmonary arterial blood pressures from a lightly anaesthetized, voluntarily breathing *Thamnophis elegans* (body weight 55.6 g). (B) Superimposed records of systemic and arterial blood pressures from Fig. 1A.

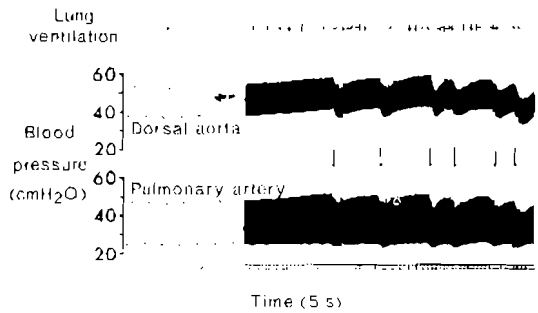

FIG. 2. Records of lung ventilation and systemic and pulmonary arterial blood pressure in a freely moving, unanaesthetized *Thamnophis elegans* (body weight 48.7). The arrows indicate the onset of brief periods of activity (swimming).

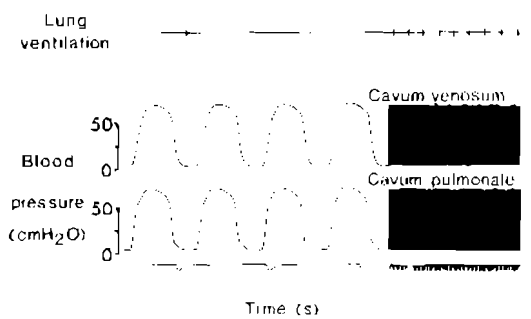

FIG. 3. Intraventricular blood pressures recorded directly through the ventricle wall of a lightly anaesthetized *Thamnophis radix* (body weight 41.3 g).

lung ventilation in all snakes. Ventilatory movements had a negligible effect upon absolute blood pressure levels and upon the pressure relationships between the two circulations. In all *Thamnophis*, active locomotor movements in water or air produced striking falls in systemic and pulmonary pressure followed by a slow return to inactive levels after activity ceased (Fig. 2), but the striking pressure difference between the pulmonary and systemic circuits remained essentially unchanged.

Intraventricular blood pressures measured directly through the ventricle walls in four *Thamnophis* showed that systolic and diastolic blood pressures in the cavum venosum and cavum pulmonale were identical during both periods of apnoea and during active lung ventilation (Fig. 3). Although identical peak systolic blood pressures were always generated in these two ventricular chambers, their pressure profiles were not always completely superimposable. In two *Thamnophis*, the pressure rise in the cavum pulmonale preceded that in the cavum venosum by 20–40 ms and the rate of pressure rise during

systole was slightly greater in the cavum pulmonale of these particular garter snakes.

Since systolic pressures in the ventricular chambers of *Thamnophis* are always identical while pulmonary artery systolic pressure is much lower than systemic pressure, a large pressure drop (i.e., an impedance to blood flow) must occur between the pulmonary valves and the point at which pulmonary arterial pressures were measured. Experiments were performed on five *Thamnophis* in which cannulae in the right aorta and pulmonary artery were advanced upstream and maneuvered through the valves into the cavum venosum and cavum pulmonale, respectively. This operation had little or no effect on heart function as the valves closed tightly around the cannulae and no changes in diastolic or systolic pressure in the ventricle and arteries occurred. There was never a measurable pressure drop towards the base of the right aorta or across the systemic outflow tract of the ventricle; peak systolic pressures in the systemic circulation and the ventricular chambers were always the same. However, as Fig. 4 illustrates, a marked pressure drop occurred across the pulmonary outflow tract. Advancement of the tip of the cannula from the base of the pulmonary artery 3 mm forward through the pulmonary valves and into the cavum pulmonale resulted in a large rise in the measured systolic blood pressure. Withdrawing the cannula back through the valves into the pulmonary artery resulted in a drop of similar magnitude in the measured systolic blood pressure. This impedance to blood flow in the pulmonary outflow tract of the ventricle was demonstrated in all five preparations.

If atropine was injected into the cavum pulmonale of *Thamnophis*, the pressure drop across

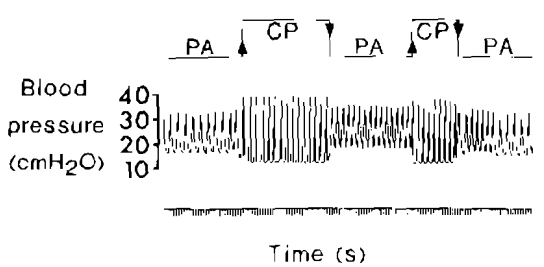

FIG. 4. Blood pressures recorded from the central pulmonary circulation of a lightly anaesthetized *Thamnophis sirtalis* (body weight 37.9 g). At the top of the record is indicated the positioning of the mobile cannula tip (see text for further details). CP, cavum pulmonale; PA, pulmonary artery.

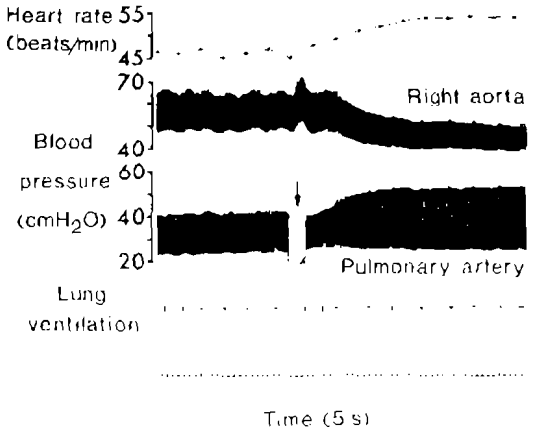

FIG. 5. Effect of atropine (1.0 mg kg body weight injected at the arrow) upon systemic and pulmonary blood pressure in an unrestrained, unanaesthetized *Thamnophis sirtalis* (body weight 37.9 g).

the outflow tract began to decrease within seconds. A few minutes after atropine injection, systemic and pulmonary systolic pressures were within 1–2 cmH_2O of each other (Fig. 5). The systolic pulmonary–systemic pressure difference was reduced from 25 cmH_2O in this undisturbed snake to only 1–2 cmH_2O after the injection of atropine, and heart rate rose from 46 to 56 beats/min. A fall in systemic mean and pulse pressure and a large increase in pulmonary pulse pressure also developed in this *Thamnophis*, the latter probably arising from blockade of cholinergic vasoconstriction in the pulmonary circulation.

Discussion

Direct measurement of intraventricular blood pressure has shown the heart of *Thamnophis* to be incapable of generating large internal pressure differentials between ventricular chambers, and in fact, identical peak systolic pressures are measured in the cavum pulmonale and the cavum venosum (Fig. 3). This situation also arises in the morphologically similar turtle heart, where a functionally single ventricular pump operates to perfuse both systemic and pulmonary circulations during apnoea, as suggested by White and Ross (1966), or during both apnoea and lung ventilation, as more recently demonstrated by Shelton and Burggren (1976) and Burggren, Glass, and Johansen (unpublished). Certainly, in the varanid lizards, the occurrence during systole of a functional division of the anatomically undivided ventricle into two distinct pumps is manifested in the generation of

very large systolic pressure differences between the systemic and pulmonary sides of the heart (Millard and Johansen 1974). In the garter snake *Thamnophis*, the identical systolic and diastolic pressures and the great similarity in pressure profiles recorded in the cavum pulmonale and cavum venosum strongly suggest that the systemic and pulmonary circuits are being perfused by a functionally single ventricular pump, as Johansen (1959) has similarly reported for a viper (*Vipera*) and a grass snake (*Tripodonotus*).

A most striking difference between cardiovascular dynamics in the garter snake and in other reptiles is the comparatively large reduction in pulmonary arterial blood pressure relative to that in the ventricular chambers and in the systemic arterial circulation. The depression of pulmonary arterial systolic blood pressure in *Thamnophis* arises solely from the large blood pressure drop which develops across a narrow region of the ventricular outflow tract of the pulmonary but not the systemic circulation. In chelonian reptiles, a discrete band of vascular smooth muscle underlying the bulbus cordis can affect large increases in the pressure drop measured across the pulmonary outflow tract in vivo when cholinergically excited or when the vagus nerve is stimulated (Burggren 1977). The present observation that pulmonary outflow tract impedance in *Thamnophis* is eliminated by atropine suggests both that smooth muscle vasomotor activity might similarly influence pulmonary arterial impedance in this snake and that there is a considerable vasomotor tone in the pulmonary outflow tract.

While the haemodynamic mechanisms resulting in different pulmonary and systemic arterial blood pressures have been demonstrated in *Thamnophis*, the physiological function of a large pressure drop across the pulmonary outflow tract is less obvious. The pulmonary and systemic circulations of *Thamnophis* are effectively linked in 'parallel' through intraventricular connections, so any variation in the impedance balance between these two circulations will result in a redistribution of blood flow between the arterial arches. A significant decrease in pulmonary outflow tract impedance, for example, could be expected to shunt blood into the pulmonary from the systemic circulation (a so-called 'left-to-right' shunt). However, neither voluntary intermittent lung ventilation nor spontaneous locomotor activity, both of which greatly influence blood distribution in the arterial arches of other reptiles (White and Ross 1966; Shelton and Burggren 1976), had any effect upon the pressure drop across the pulmonary outflow tract of the garter snake. In turtles, only a very small proportion of the total pressure drop from the cavum pulmonale of the heart to the pulmonary venous circulation occurs at the outflow tract. In *Thamnophis* (assuming a negligible venous pressure), about 20–25% of the ventricular–pulmonary venous pressure difference arises solely from passage through the ventricular outflow tract. That this considerable pressure drop can be pharmacologically eliminated in the snake clearly reveals the potential of this region to affect pulmonary impedance and so redistribute blood in the arterial arches.

Why then was pulmonary outflow tract impedance apparently fixed at a constant high level in these snakes? It may transpire that conditions which were not simulated during these studies, such as severe hypoxia and hypercapnia arising from extended periods of apnoea, perhaps in conjunction with locomotor activity, are in fact accompanied by variations in pulmonary outflow tract impedance. The location in the snake of a high and variable impedance at the pulmonary outflow tract rather than at the level of the distal pulmonary arteries and arterioles as found in turtles could allow a more effective control over pulmonary arterial blood volume and the systemic–pulmonary impedance balance. This especially would be so if the central pulmonary arteries of the snake were highly compliant as they have been reported to be in other reptiles (Burggren 1977). Additionally, the establishment of a higher blood pressure in the systemic circulation relative to that in the pulmonary circulation may favor systemic functions served by high arterial pressures (e.g., renal ultrafiltration) while at the same time not inducing pulmonary edema in the snake, which might conceivably develop if pulmonary arterial pressures were allowed to rise to the comparatively high systemic levels.

Acknowledgements

The author acknowledges the loan of equipment by Drs. H. C. Nordan and D. R. Jones. This investigation was performed while the author was a recipient of a joint NRCC–Killam postdoctoral fellowship.

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