

## A COMPARATIVE STUDY OF CENTRAL BLOOD PRESSURES IN FIVE AMPHIBIANS

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(Received 19 April 1968)

### INTRODUCTION

A number of articles written in recent years about the pattern of blood flow through the amphibian heart have described results which differ quite widely (Simons, 1957, 1959; de Graaf, 1957; Johansen, 1963; Shelton & Jones, 1965*a, b*). One of the main difficulties in interpretation has been that all the work cited above has been done on different species of anuran and urodele. It is clearly possible to ascribe many of the major differences to variation between species in anatomical or physiological adaptation to different environments (Foxon, 1964). However, the doubt must remain that some of the differences may be due to experimental methods, involving as they do variation in anaesthesia, operative technique and recording method.

The problem of the amphibian circulation has been developed along two distinct lines. In one, recognizable materials (coloured dyes or particles, radio-opaque materials, oxygen) have been introduced into the blood stream and their route through the heart into the arterial arches followed (Foxon, 1947; Simons, 1959; de Graaf 1957; Johansen, 1963). In the other, pressures have been measured in various parts of the system and attempts made to assess blood flow from the gradients which are found (Simons, 1957; de Graaf, 1957; Johansen, 1963; Shelton & Jones, 1965*b*). Although the two methods are complementary and full analysis is impossible with either taken alone, it is to the second type of determination that the present paper relates.

In *Amphiuma tridactylum* Johansen (1963) claimed that the peak systolic pressure produced by ventricular contraction was very much greater when measured in the ventricle than a short distance downstream in the conus. Contraction of the conus produced a second pressure peak which was roughly the same size as the first, ventricular peak. Johansen thus assigned to the conus a major function of smoothing blood flow from the ventricle and extending the period of flow from the heart so that it was no longer intermittent. In *Rana pipiens*, on the other hand, Shelton & Jones (1965*b*) were unable to confirm the very considerable smoothing suggested by Johansen and found that pressures in ventricle and conus followed one another closely until the time of ventricular relaxation. Divergence of the two pressures then occurred because of continued conus contraction but outflow of blood from the conus was negligible and came to an end when the conus relaxed. Outflow from the heart was intermittent, with the conus playing a very small part in the total pumping activity when compared with the ventricle.

The contributions on *Amphiuma* and *Rana* are the only ones in which serial pressure

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recordings were made in all the main chambers of the heart but several authors have measured pressures in the arterial arches of Amphibia. Here again important differences have been reported. Simons (1957) found that the pressures in systemic and pulmocutaneous arches of *R. temporaria* and *Bufo bufo* were identical over the full pulse cycle. In *Xenopus laevis*, de Graaf (1957) described slight differences in systolic, and major differences in diastolic levels in the two arches, with the pulmocutaneous pressures being the lower. This type of relationship was also suggested by Shelton & Jones (1965*a*) after their experiments on *R. pipiens* and *R. temporaria*. Johansen (1963) found that sometimes there were differences of systolic and diastolic pressure in the arches to body and lungs in *Amphiuma* but that these were not consistent.

The aim of the present study is to determine how far five amphibian species differ under the conditions of our previous experiments and to assess how far interspecific differences are likely to exist.

#### METHODS

The experiments were carried out on the anurans *Rana pipiens*, *R. temporaria*, *Xenopus laevis* and *Bufo bufo* and the urodele *Salamandra salamandra*. The *Rana* species and *Bufo bufo* varied in weight from 20 to 40 g., *Xenopus* from 80 to 119 g. and the salamander from 22 to 48 g. Blood pressures were measured by inserting hypodermic needles directly into the exposed heart or arterial arches. Though working on the open-chest preparation has been found to give some anomalous results in the mammal (Rushmer, 1961) this technique makes cannulation unnecessary and interferes little with blood flow. It also permits rigid connexion to be made between the needles and manometers with the advantage of increased frequency response over connexions made with flexible tubing.

As much of the heart and arterial arches as necessary were exposed by cutting through the clavicles and coracoids and removing part of the sternum. The pericardium was left intact as far as possible, but when determinations were made in the ventricle the heart had to be exposed completely. During the experiments the animal was held lightly on a wax block with its ventral side uppermost. Three anaesthetics were used during the work: paraldehyde (0.02 ml./10 g.) was injected subcutaneously into the dorsal lymph sac, Sandoz MS 222 (300 mg./l.) and urethane (5 g./l.) were administered by immersing the animals in aqueous solutions. Initially the animals were deeply anaesthetized to a level at which the buccal breathing movements became feeble or stopped. The surgery was then carried out and the recording needles were put into position. The animal was allowed to recover to a more lightly anaesthetized state and the recordings were usually made when normal breathing movements began. Complete recovery was prevented by further injections or by moistening the skin with anaesthetic. Anaesthesia often has a profound effect on the circulatory system and in the present case paraldehyde at the higher levels used caused a marked fall in blood pressure. Some degree of sedation is essential, otherwise sudden changes of blood pressure can be caused by variations in the state of the animal, sudden movements, etc. Moreover with rigid connexions between needles and manometers, excessive movements were liable to tear the arterial walls. At the lower levels of anaesthesia the picture of blood flow and pressure was the same for all three anaesthetics and it is thought therefore that the results are representative of the resting animal.

Pressure was measured by Ole Dich (type ATH), Sanborn 267 B, or Statham P 23 DC manometers connected by copper tubing to the hypodermic needles. Usually 26-gauge needles were used but in particular cases both finer and coarser needles have been tried. The needles were inserted into the blood vessels in the direction of blood flow. The characteristics of the whole pressure-recording system were measured by applying a sudden pressure change to the manometer through the connexions and the 26 G needles commonly used. Under these conditions the Sanborn instruments had a natural frequency of 35–40 cyc./sec. and 30–50% critical damping, the Statham a frequency of 40–45 cycles and 50% critical damping, and the Ole Dich a frequency of 60–70 cyc./sec. and 40–60% critical damping. The damping was slightly less than optimal for a uniform amplitude response and phase lag at frequencies up to the natural frequency. As expected, the long-term stability was greatest in the instrument with the lowest natural frequency. Since the amphibian heart rate is quite low and no very rapid changes appeared in the pressure traces it was clear that even the slowest of these systems was adequate. No differences could be seen in the characteristics of the traces whichever manometer system was used.

The pressure recordings were made on an A.E.I. three-channel pen recorder or a Sanborn 966 six-channel instrument, both writing on rectangular co-ordinates. The electrocardiogram (E.C.G.) was detected by a thin wire near the heart, amplified in a condenser-coupled amplifier, and recorded on the pens. The experiments were carried out at room temperature (18–20°C.).

## RESULTS

### (a) *Xenopus laevis*

Simultaneous recording of blood pressures measured in the ventricle, conus, systemic arch, and pulmocutaneous arch of *Xenopus* confirmed the general pattern seen in earlier results obtained from *Rana pipiens* by superimposing pairs of traces (Shelton & Jones, 1965*b*). Traces which show the general features of the pressure changes as well as some of the differences in detail are shown in Figs. 1, 2 and 3. They are taken from three animals; one was anaesthetized with paraldehyde (Fig. 1), the others with MS 222, and the toad of Fig. 3 had an intact pericardium whilst the others did not. In all other respects the animals were, as far as could be determined, in identical condition. The basic pattern emerges clearly. Following the P wave of the E.C.G., auricular contraction filled the ventricle. Usually very little blood flowed into the ventricle before this. In some cases auricular systole succeeded in filling the conus, producing in it a slight increase in pressure (Fig. 1) but this was not commonly seen. The QRS wave was followed by a latent period of approximately 80 msec. after which the ventricle contracted, causing a sudden increase in pressure in both ventricle and conus. As diastolic pressures in first the pulmocutaneous arch and then the systemic and carotid arches were exceeded so blood began to leave to these vessels and the rate of pressure increase fell off. The end of ventricular contraction and beginning of the T wave saw a steep fall in ventricular pressure. Continued contraction caused a much slower fall in pressure in the conus, the valves between this chamber and the ventricle being closed. Eventual relaxation of the conus musculature resulted in a greatly accelerated fall in pressure, the extent of the fall depending on several factors. The

most obvious and important of these was heart-rate. If the heart was beating slowly the conus pressure could fall almost to zero (Fig. 1). Usually this did not occur since the next cycle began before the pressure reached this level (Fig. 2). The rate of fall in pressure in the relaxed conus was never as great as that in the relaxed ventricle (Figs. 1, 2). After conus relaxation, pressures in the arteries were maintained solely

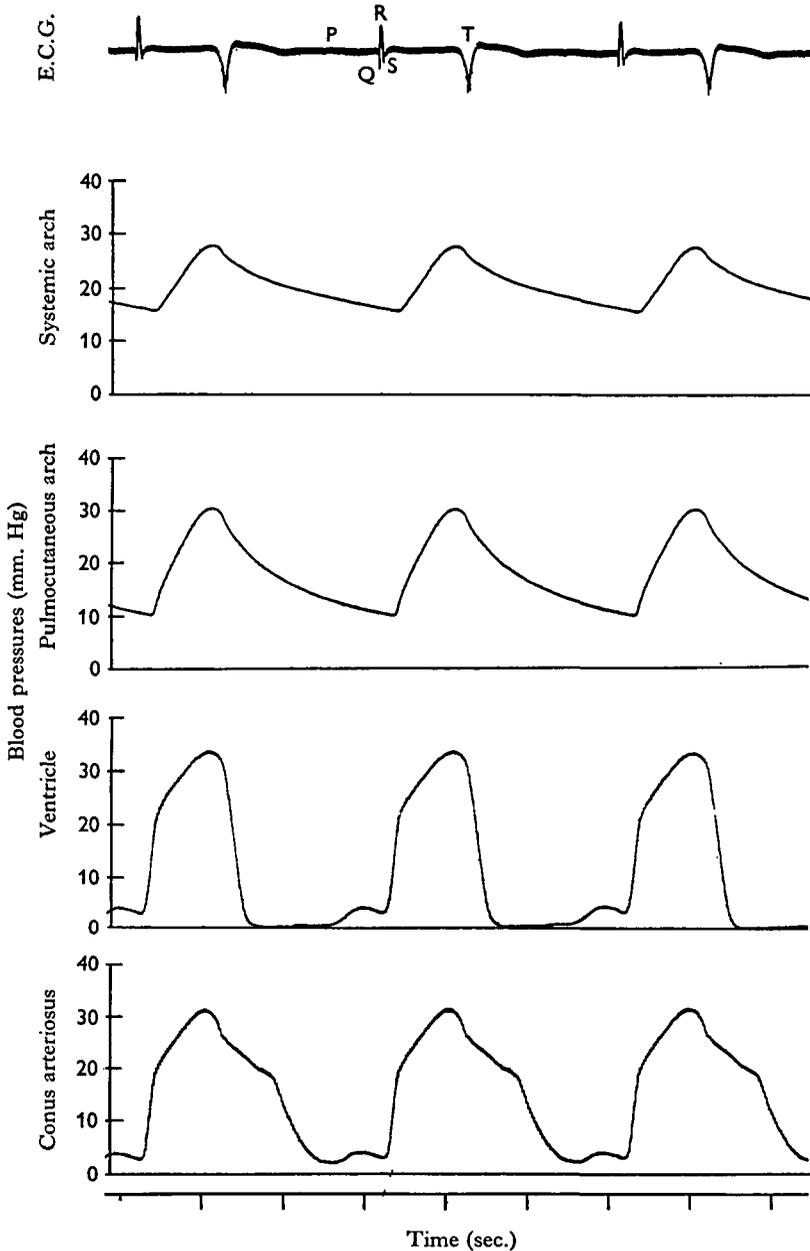


Fig. 1. *Xenopus laevis*. 95 g., paraldehyde anaesthesia. Blood pressures recorded simultaneously in ventricle, conus and arterial arches.

by elastic recoil of the vessel walls, valves between arterial arches and conus being closed. The rate of pressure fall was low, though different, in systemic and pulmocutaneous arches.

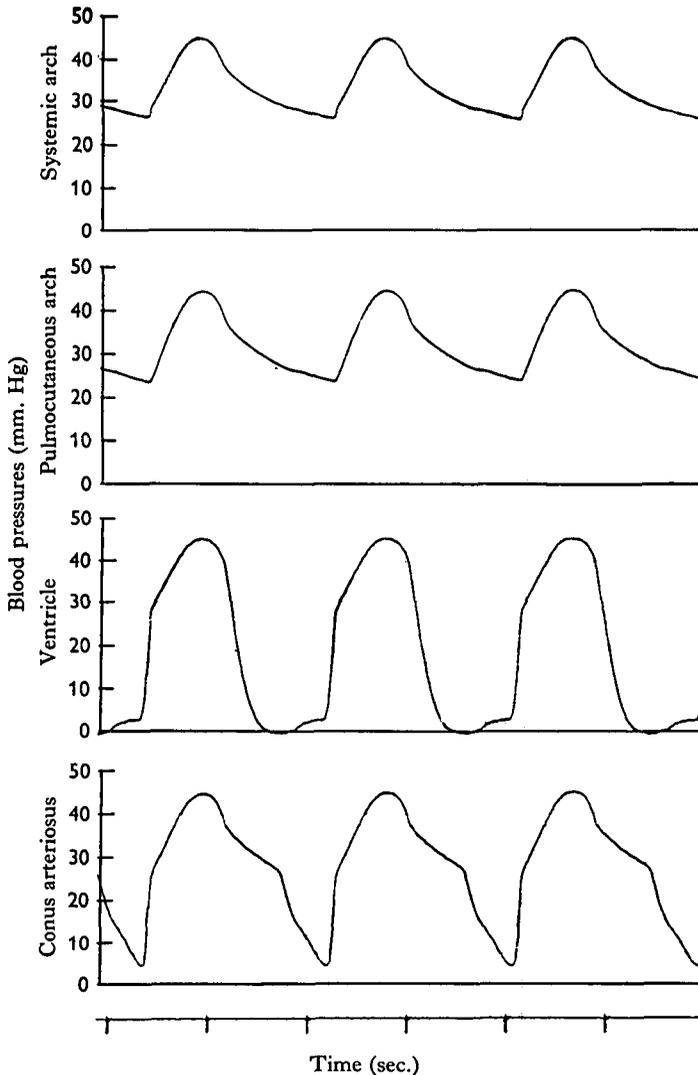


Fig. 2. *Xenopus laevis*. 80 g., MS 222 anaesthesia. Blood pressures recorded simultaneously in ventricle, conus and arterial arches.

The difference between pulse pressures in systemic and pulmocutaneous arches is difficult to understand. It was variable in extent, sometimes being large (Fig. 1) and sometimes quite small (Figs. 2, 3) or even non-existent. The relationship varied in the same animal at different times. The pulmocutaneous pulse was larger because it reached a lower diastolic level. During ventricular systole the pressures in ventricle and conus increased very rapidly to the level in the systemic arch. The major change of slope in the rising phase of ventricle and conus pressure curves occurred at the diastolic

level of systemic and not pulmocutaneous pressures (Figs. 1-3). During the falling phase the two pressures became different after the inflexion marking closure of the valves between ventricle and conus. The difference appeared therefore whilst blood was still leaving the conus and the suggestion has already been made that, in *Rana pipiens* where a similar relationship appeared, the spiral valve was in some way capable

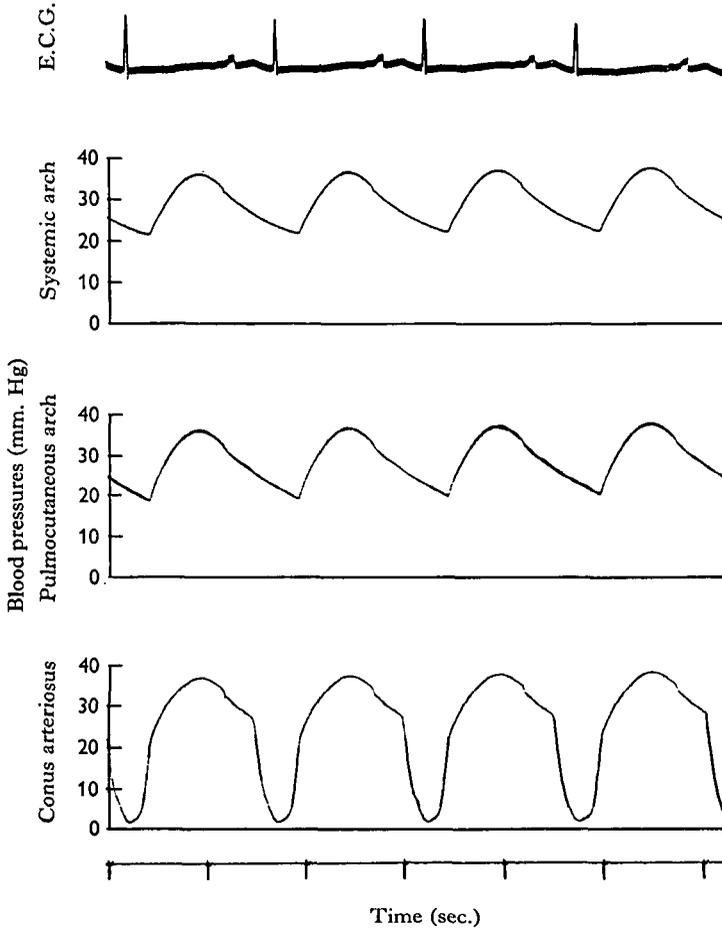


Fig. 3. *Xenopus laevis*. 114 g., MS 222 anaesthesia, intact pericardium. Blood pressures recorded simultaneously in conus and arterial arches.

of sustaining a pressure difference on its two sides (Shelton & Jones, 1965a). In most specimens of *R. pipiens* the pressure recorded from the contracting conus had a falling slope which was the same as that recorded in the systemic arch, but in a few animals the slope coincided with that in the pulmocutaneous arch. This was interpreted as support for the view that pressures were different in the two chambers of the conus and could be recorded by suitably placed needles. However, it never proved possible to record different pressures simultaneously with two needles in the conus no matter how they were placed, so the hypothesis was not confirmed by direct observation. The same relationships have been seen in other anurans and in Fig. 1 the falling slopes can be seen to be identical in conus and pulmocutaneous arch. Systemic arch

pressures are consistently higher during the falling phase. In Fig. 3, on the other hand, the falling slope in the conus is the same as that in the systemic arch and in Fig. 2 the two slopes are so similar that the precise relationship is impossible to determine.

(b) *Other anurans*

Comparison of results from *R. pipiens*, *R. temporaria* and *B. bufo* with those already described from *Xenopus* show that there were no major differences in the central blood pressures of these anuran species (Table 1). The differences in absolute pressure level are significant in a few cases (Table 2) but no consistent pattern emerges except in the case of the comparison between *Xenopus* and *R. pipiens*. All the *Xenopus*

Table 1. *Blood pressures (mm. Hg) and pulse ratios (pulse pressure/systolic pressure) measured in the arterial arches of four anurans and one urodele (standard errors are given)*

		<i>Xenopus laevis</i>	<i>Rana temporaria</i>	<i>Bufo bufo</i>	<i>Rana pipiens</i>	<i>Salamandra salamandra</i>
A. Blood pressures						
Systemic	Systolic	37.9 ± 1.98	33.8 ± 1.33	32.4 ± 1.88	29.6 ± 1.01	21.9 ± 0.81
	Diastolic	24.1 ± 0.96	22.1 ± 1.09	19.5 ± 1.49	19.6 ± 0.63	12.0 ± 0.68
	Pulse	13.8	11.7	12.9	10.0	9.9
Pulmocutaneous	Systolic	37.0 ± 2.01	33.2 ± 1.67	32.2 ± 1.85	30.2 ± 0.43	22.1 ± 0.81
	Diastolic	20.0 ± 1.59	14.1 ± 2.01	15.8 ± 1.54	14.0 ± 0.82	12.3 ± 0.65
	Pulse	17.0	19.1	16.4	16.2	9.8
B. Pulse ratios						
Systemic		0.36 ± 0.019	0.34 ± 0.019	0.40 ± 0.026	0.34 ± 0.016	0.44 ± 0.022
Pulmocutaneous		0.46 ± 0.029	0.57 ± 0.046	0.51 ± 0.027	0.53 ± 0.021	0.44 ± 0.021

blood pressures are significantly higher than the equivalent pressures measured in *R. pipiens*. The systolic pressures in *R. temporaria* are also significantly higher than those in *R. pipiens*. The temporal relationships of pressures recorded in ventricle and conus were exactly the same as described for *Xenopus*, with the pulse in the ventricle extending from zero to maximum systolic pressure and that in the conus extending over a variable but usually smaller range.

To eliminate differences due to absolute pressure level so that more direct comparisons can be made it is useful to consider the arterial pulse as a fraction of the peak systolic pressure. These figures are also given in Table 1. The ratios obtained from the same arch in any of the four anuran species do not differ significantly. The systemic arch ratio is always significantly lower than that of the pulmocutaneous arch in the same species but the difference is less marked in *Bufo* and *Xenopus* than in the *Rana* species. As already described for *Xenopus*, the systemic and pulmocutaneous pulses in the other species were occasionally found to be identical. It is difficult to specify the precise conditions under which this occurred but deep anaesthesia with no breathing movements was frequently associated. In some cases, and particularly in *Bufo*, the similarity in traces persisted during recovery when breathing was quite normal, though usually progressively greater differences appeared. Work is now going on to examine the reasons for this relationship.

(c) *Salamandra salamandra*

The main anatomical features of the salamander which differ from those of anura and which are relevant to the dynamics of blood flow through the heart are:

(1) Normally four arterial arches persist in the adult, the extra one being a fifth visceral arch running from the truncus arteriosus and joining the systemic arch just before it enters the dorsal aorta. This vessel is usually smaller than the preceding one.

Table 2. Comparison of systolic (S) and diastolic (D) pressures in the arterial arches of four anurans and one urodele to show incidence of significant differences (indicated by +; P greater than 0.05)

		<i>Rana temporaria</i>				<i>Bufo bufo</i>				<i>Rana pipiens</i>				<i>Salamandra salamandra</i>			
		Sys-temic		Pulmo-cutaneous		Sys-temic		Pulmo-cutaneous		Sys-temic		Pulmo-cutaneous		Sys-temic		Pulmo-cutaneous	
		S	D	S	D	S	D	S	D	S	D	S	D	S	D	S	D
<i>Xenopus laevis</i>	Systemic	S	-	.	.	-	.	.	.	+	.	.	.	+	.	.	.
		D	.	-	.	.	+	.	.	.	+	.	.	.	+	.	.
	Pulmo-cutaneous	S	.	.	-	.	.	-	.	.	.	+	.	.	.	+	.
		D	.	.	.	+	.	.	-	.	.	.	+	.	.	.	+
<i>Rana temporaria</i>	Systemic	S	-	.	.	+	.	.	.	+	.	.	.	+	.	.	.
		D	.	-	.	.	.	-	.	.	.	.	.	+	.	.	.
	Pulmo-cutaneous	S	.	.	-	.	.	.	+	.	.	.	.	.	.	+	.
		D	.	.	.	-	.	.	.	.	.	-	.	.	.	.	-
<i>Bufo bufo</i>	Systemic	S	-	.	.	.	.	.	.	+	.	.	.	+	.	.	.
		D	.	-	.	.	.	.	.	.	.	.	.	.	+	.	.
	Pulmo-cutaneous	S	.	.	-	.	.	.	.	.	.	-	.	.	.	+	.
		D	.	.	.	-	.	.	.	.	.	.	-	.	.	.	+
<i>Rana pipiens</i>	Systemic	S	+	.	.	.	.	.	.	+	.	.	.	+	.	.	.
		D	.	.	.	.	.	.	.	.	.	.	.	.	+	.	.
	Pulmo-cutaneous	S	.	.	+	.	.	.	.	.	.	.	.	.	.	+	.
		D	.	.	.	.	.	.	.	.	.	.	.	.	.	.	-

(2) The ductus Botalli connects the pulmonary and systemic arches together in their dorsal regions. It is a vessel of considerable size and is about half the diameter of the vessels it connects.

(3) The spiral valve is greatly reduced and hardly projects into the lumen of the conus, whereas in most anurans it seems likely that the valve can divide the conus into two chambers. Dissection of the living conus and paraffin sections of fixed material (after allowance is made for distortion and shrinkage) would confirm this.

(4) There is no major division of the pulmonary arch which corresponds with the cutaneous artery of anurans.

The systolic pressures recorded in ventricle, conus and aortic arches were significantly lower than in the anurans examined (Tables 1, 2). The diastolic pressures in the arterial arches were also significantly lower than equivalent pressures measured in the anurans except in the case of those from the pulmocutaneous arches of the two *Rana* species.

In the majority of animals the time courses of the pressure curves as well as absolute pressure levels were identical in the systemic and pulmocutaneous arches (Table 1, Fig. 4), and in these respects also the salamander differs markedly from anurans. Such differences as were observed were almost certainly attributable to slight variations in anatomy and in position of recording needles. The relative pulse size in the salamander was intermediate between the systemic and pulmocutaneous pulses of the anurans as the pulse:systolic pressure ratios show (Table 1). The *Bufo* ratios and

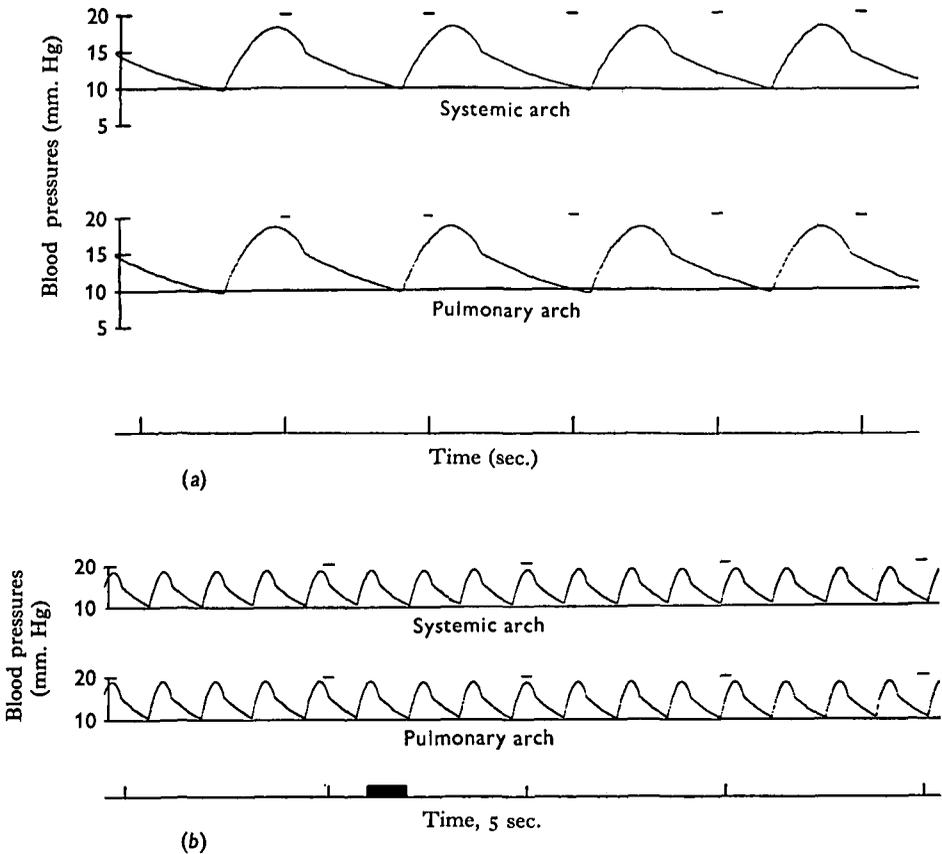


Fig. 4. *Salamandra salamandra*, 33 g., paraldehyde anaesthesia. Blood pressures recorded in the systemic and pulmonary arches. (a) To show pulse waveform; (b) to show effect of occlusion of the ductus Botalli (the ductus was closed at the beginning of the record and the artery clip was removed at the mark on the lower trace).

*Xenopus* pulmocutaneous ratio were not significantly different from the salamander ratios, however. The values from *Salamandra* were intermediate between, and significantly different from, the small systemic and large pulmocutaneous ratios of the *Rana* species. The presence of a fifth aortic-arch vessel had no effect on pulse patterns, which were identical in animals with or without the extra connexion.

Since these urodeles have an open connexion between systemic and pulmonary systems it was perhaps unremarkable that the pressures were identical. In a number of cases the ductus Botalli was approached from the ventral side in order to assess its

importance in equalization of the two systems. Observation confirmed that it was a large vessel which was slightly pulsatile and in the majority of cases there was flow of blood from systemic to pulmonary arch. Flow was also pulsatile but not rapid, corpuscles being clearly visible during the slower parts of the pulse cycle. On some occasions flow in the opposite direction was seen to occur and eventually it was found that slight alterations in the position and relationship of the major arches could affect rate and even direction of blood flow. For example, if the pulmonary arch was lifted clear of the surrounding tissues blood invariably flowed down the ductus Botalli to the systemic arch. These observations confirmed the equality of pressures in the two sides of the system and suggested that the ductus Botalli had in fact little part to play

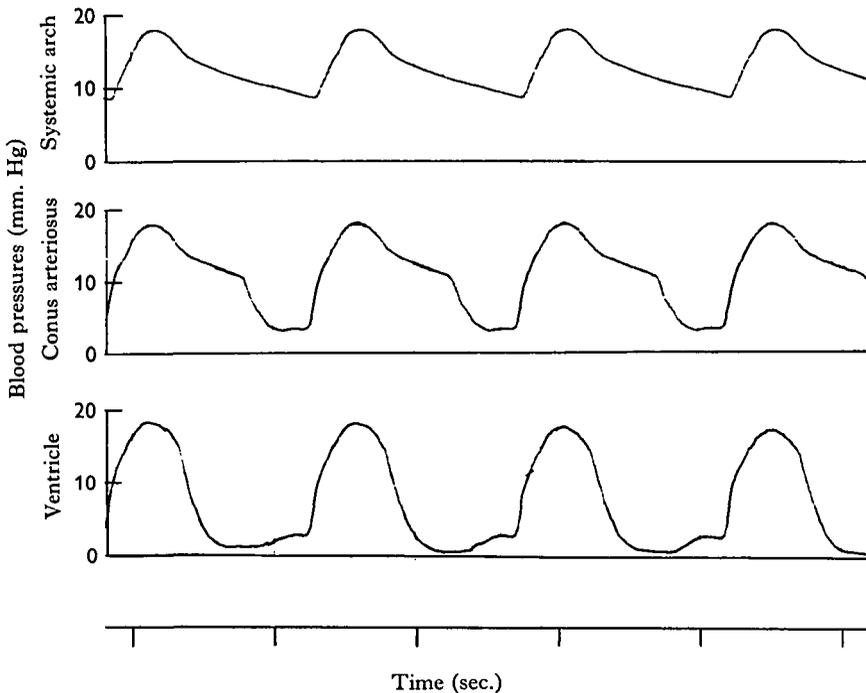


Fig. 5. *Salamandra salamandra*, 38 g., paraldehyde anaesthesia. Blood pressures recorded in the heart and systemic arch.

since the flow rate in it was low. Experiments were also carried out in which the ductus Botalli on one side was closed by an artery clip and the pressures were measured in pulmonary and systemic arches. The pulmonary arch on the clipped side was thus made independent of all the other arches except for its central connexion at the conus arteriosus. In spite of this the pressure remained identical for long periods and no change occurred when the clip was removed (Fig. 4*b*).

Apart from these differences the pressure recordings obtained from the heart of the salamander resembled very closely those found in anurans. The pulse in the ventricle was large, rising from zero to the maximum systolic level as the ventricle contracted (Fig. 5). The pulses in conus and arches followed this pressure closely until ventricular

relaxation, when continued conus contraction and arterial recoil caused the usual divergence after closure of the ventricle-conus valves. Divergence between conus and arterial pressures then followed as the conus relaxed and the conus-arterial arch valves closed (Fig. 5). There can be little doubt that, so far as blood pressures are concerned, the system functions in the same way as the anuran heart and that blood is propelled through the heart by the same sequence of events in both groups of animals.

## DISCUSSION

None of the results described in this paper is contrary to the suggestions made earlier (Shelton & Jones, 1965*b*) about the movement of blood through the heart of *Rana pipiens*. The description given here for *Xenopus laevis*, derived from simultaneous recording of pressures in the heart and arterial arches, could apply equally to the other anurans and, with certain differences in the arterial arch pattern, to the urodele. In all the animals studied, pressure differentials between ventricle, conus and arterial arches were small as blood was moving forward through these regions. The pulse waveforms during these phases of forward blood movement were also the same in the various regions. Large pressure differentials and considerable change in the outline of the pulse waveform, such as those reported by Johansen (1963) and by Sanchez-Cascos & Foxon (1963), can be explained in terms of high-resistance connexions between the various recording sites, very low pulse velocities, or complications due to pulsatile flow. It is interesting to look at these possibilities in the light of information collected during this and previous investigations.

In all the anurans, and in the urodele, the time courses of pressures in ventricle, conus and arterial arches were indistinguishable during the ejection phase. Peak systolic pressure was reached simultaneously in all regions. Pulse-wave velocity ( $c$ ) can be calculated from measurements of pressure-volume distensibility using the formula

$$c = \sqrt{(V\delta P/\rho\delta v)},$$

where  $V$  is the initial volume,  $\delta v$  and  $\delta P$  are increments of volume and pressure and  $\rho$  is the density of the fluid. In *Rana pipiens* figures obtained from photographs of a small section of the arterial arch region just before division into three separate vessels indicate an increment of 1.20 mm<sup>3</sup> on a volume of 3.75 mm<sup>3</sup> for a pressure increase of 15,700 dynes/cm<sup>2</sup>. The density of frog blood is approximately 1.05. The wave velocity in this case would be 2.2 m./sec. Other specimens have shown smaller elastic increases in volume and therefore somewhat higher wave velocities up to 4.0 m./sec. These are lower velocities than the 4–5 m./sec. in the thoracic aorta and 8–10 m./sec. in the femoral artery of the dog as quoted by McDonald (1960). Wave velocities in the conus based on similar determinations of pressure-volume distensibility are little more than half the values found in the truncus. This region is largely muscular and contains relatively little elastic tissue, added to which the muscle becomes active some time during the rising phase of the pulse. There are therefore obvious difficulties in a simple treatment but it is clear that pulse-wave velocity is sufficiently high for there to be little or no detectable effect with the recording speeds used here. Recording points in ventricle and arterial arches would be separated at the most by 1.5 cm.; the maximum paper speed was 25 mm./sec.

Even though the pulse wave is transmitted fairly rapidly so that the main temporal features appear simultaneously when recorded from points close together, the absolute levels of pressure may still be influenced by a variety of factors. For example, the damping of a wave propagated in a viscous system results in a gradual attenuation of sharp peaks. Thus the incisura eventually disappears some 30–40 cm. from the aorta in the dog. There are no high-frequency components in the amphibian pulse and it is most unlikely that damping effects would appear in the main arterial trunks within a centimetre or so from the heart. Reflexions from all branches of the mammalian arterial tree, and particularly from the ends of the tree which behave as though they were closed, increase the amplitude and ‘peaking’ of the pulse wave. Again the effects are seen over distances of 10 cm. or more. Neither damping nor reflexion were evident in our pressure records and it will be interesting to see whether they appear in more peripheral pressure recordings from the amphibia.

In a central arterial system where the ventricle pumps blood into a tubular conus from which three or four major trunks ultimately arise on either side, it is clear that the velocity of blood flow may change considerably. Pressures may be affected in a way which, for ideal fluids, is described by Bernouilli’s theorem. In *Rana*, for example, a reasonable figure for stroke output would be 0.15 ml. pumped through a conus of 0.024 cm.<sup>2</sup> average cross-sectional area. If the ejection phase lasts for 0.9 sec. the average blood velocity would be 6.9 cm./sec. The kinetic energy in such a blood stream would be 25 ergs/ml. and equivalent to a pressure of the same number of dynes/cm.<sup>2</sup>. Assuming the blood in the ventricle to have no appreciable kinetic energy, the difference in pressure between ventricle and conus due to motion of the blood could not be more than 0.1 mm. Hg even during maximum velocity flow. The differences due to kinetic energy between conus and arterial arches would be equally small and impossible to detect with the recording system used.

It ought, in the absence of further complications, to be possible to estimate the pressure gradient necessary to produce the observable flows in conus and truncus. The fact that the flow is oscillatory, however, makes a direct application of the formula of Poiseuille somewhat suspect, as the extensive analyses of Womersley (1955) and McDonald (1960) have shown. Flow lags behind changes in pressure gradient because of inertia of the blood, and at high oscillation frequencies flow amplitude is no longer linearly related to pressure gradient. The deviation from Poiseuille’s formula and the extent of the phase lag is determined by a non-dimensional constant  $\alpha$ :

$$\alpha = R\sqrt{(2\pi n f \rho / \mu)},$$

where  $R$  is the radius of the tube,  $\mu$  and  $\rho$  the viscosity and density of blood,  $f$  the pulse frequency in cyc./sec. and  $n$  is the order of the harmonic component. When  $\alpha$  is 0.5 or less for the fundamental frequency, the phase lag is negligible and the flow approximates to the formula of Poiseuille. Up to a value of 1.0 the approximation is still quite good. Calculations for the major vessels leaving the heart of *Rana* give values of  $\alpha$  between 0.5 and 1.0 for the fundamental frequency (based on conus and truncus radii between 0.055 and 0.10 cm., pulse rates between 36 and 48 beats/min., a viscosity of 0.04 P. and density of 1.05 g./ml.). These values compare with 1.38–3.5 for the rat, 8.27–10.68 for the dog, and 13.5–16.7 for man, all referring to the root of the aorta (McDonald, 1960).

In the frog central arterial system, therefore, flow and pressure gradients are approximately linearly related. If it is assumed that flow is uniform throughout the ejection phase then a direct application of the Poiseuille equation shows that pressure gradients of 0.1–1.0 mm. Hg/cm. are necessary. Since flow is obviously not uniform there will be variations outside these limits perhaps, but in general one would expect gradients of this order of size. The experiments demonstrate that this is the case. There is no theoretical or practical evidence for substantial pressure gradients between chambers of the heart and the arterial arches during the blood-ejection phase.

## SUMMARY

1. The temporal relationships of blood pressures recorded from the ventricle, conus and arterial arches are identical in *Rana*, *Bufo* and *Xenopus*. Pressure levels in the arterial arches are very similar in all the species.

2. The pulse in the ventricle is large and the pressure falls to zero during diastole. Continued conus contraction after ventricular relaxation causes a divergence of pressure in the two chambers. Divergence between conus and arterial pressures follows as the conus relaxes. Pressure gradients between the various recording sites during forward movement of the blood is small. Theoretical considerations show that pressure-flow relationships approximate to the formula of Poiseuille. They also produce no evidence for substantial pressure gradients during blood ejection.

3. Blood pressures recorded in *Salamandra* are significantly lower than in the anurans. The systemic and pulmocutaneous pulses are identical, unlike the condition in anura where the pulmocutaneous pulse is almost invariably the larger. In all other respects the pressure relationships in *Salamandra* are identical to those in anurans.

The authors wish to thank the Science Research Council for financial support.

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