Circulatory changes during experimental diving in the turtle

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WHITE, FRED N., AND GORDON ROSS. Circulatory changes during experimental diving in the turtle. Am. J. Physiol. 211(1): 15-18. 1966.—Blood flow (electromagnetic flowmeters) and pressures in pulmonary and systemic circuits were studied in intact, unanesthetized turtles (Pseudemys scripta) in both nondiving and diving states. Cardiac output (combined pulmonary, left and right arch flows) in the predive state was in the order of 50-60 ml/kg per min with 60% distributed to the pulmonary circuit (left-to-right shunt). During diving, bradycardia and a reduction and redistribution of cardiac output (right-to-left shunt) occurred after an interval varying from 8 to 180 min. The development of the shunt was accompanied by a decrease in the systemic and increase in the pulmonary resistances. These changes were rapidly reversed on surfacing into air or nitrogen, but persisted when the turtles surfaced into carbon dioxide, suggesting that hypercarbia is involved in their maintenance.

Methods
Specimens of Pseudemys scripta weighing 1-3 kg were used. Each animal was refrigerated at 5°C for 12 hr before operation which was performed in a tray of ice. No chemical anesthetic agent was used. After removing an 8 x 8-cm portion of the plastron, noncannulating electromagnetic flow probes (5) were then placed on the arteries under study. The flow signal was measured with a phase-sensitive sine-wave flowmeter (4). In all experiments flow in the left pulmonary artery was determined. In some, right and left aortic arch flows were also recorded, the probe on the right arch being placed just distal to the brachiocephalic artery. In other experiments subclavian and pulmonary arterial flows were simultaneously recorded. Figure 1 illustrates flow probe placements.

Pressure in the vessels was measured from indwelling heparin-filled catheters placed proximal to the flow probes and connected to Sanborn pressure transducers. The catheters and the flow probe leads were carried out through the ventral opening in the plastron and strapped to the carapace. The excised portion of the plastron was then replaced and secured by stainless steel sutures. The gap between the replaced section and the remainder of the plastron was sealed with acrylic denture material. The animal was then allowed to attain room temperature.

Pressure and flow measurements were made during the first four or five postoperative days when the animal's behavior and activity did not differ from that before operation or from that of nonoperated turtles kept under the same conditions. Later the condition of the animals deteriorated and death occurred 5-10 days after operation. Autopsy invariably revealed a massive blood clot in the area of the operation but no evidence of constriction, damage, or thrombosis of the vessels carrying the flow probes. Calibration was performed by allowing saline to pass through the lumen of the flow probe via a gravity feed. During diving very low heart rates were observed, e.g., 1-2/min. The portion of the flow trace immediately preceding one of these infrequent beats was considered to represent zero flow and was used as the reference level for flow measurements.

Diving experiments were performed in an aquarium. During the studies of prolonged diving, the turtles were weighted with lead.
RESULTS

Observations in Nondiving Turtles

Pulmonary arterial pressure and flow. Systolic pressure averaged 26 mm Hg and diastolic pressure 10 mm Hg. Figure 2 illustrates the similarity during each cardiac cycle of the pressure and flow contours of a typical animal with a heart rate of 33/min. Four main components are shown: a) a sharp upstroke reaching a peak after 0.2 sec; b) a rounded summit of 0.3 sec duration; c) a steep downstroke lasting 0.4 sec; and d) a gradual decline to end-diastolic values. Pulmonary flow continued throughout the entire cardiac cycle at this heart rate. Stroke flow in each pulmonary artery was 1.1 ml and minute flow was 36.3 ml. Total pulmonary flow was, therefore, 72.6 ml/min assuming equal flow in the two pulmonary arteries.

Aortic pressure. Systolic pressure averaged 30 mm Hg and diastolic pressure 26 mm Hg. The upstroke of the instantaneous aortic pressure curve began 0.13 sec after the commencement of pulmonary flow and was followed by a rounded summit. The downstroke was less steep than in the pulmonary artery and showed a definite dicrotic notch. At no time in the cardiac cycle were aortic and pulmonary pressure curves coincident.

Subclavian flow. Maximum flow velocity was reached 0.12 sec after the beginning of aortic ejection and was maintained for a further 0.08 sec. The declining phase of the flow curve showed an initial gradual slope which became much steeper just before the appearance of the aortic pressure incisura. Forward flow was then rapidly halted and was followed by a period of back flow. During the remainder of the diastolic period, small low-frequency oscillations of flow occurred. The duration of significant forward flow occupied only 28% of the cardiac cycle. In some turtles a short period of back flow preceded the onset of forward flow (Fig. 2).

Right aortic arch flow (distal to brachiocephalic artery). The flow contour differed from that of the left arch only in showing little or no back flow. Stroke flow was 0.77 ml at a heart rate of 40/min (Fig. 3).

Left aortic arch flow. The instantaneous flow curve resembled that of the subclavian artery. Stroke flow was 0.69 ml at a heart rate of 40/min.

Changes During Diving

Heart rate. When allowed to swim freely, the turtles dived only for short periods and usually surfaced after less than a minute during which the heart rate did not change. During prolonged forced diving, the turtles made frequent vigorous efforts to surface. These efforts were characterized by neck extension, pharyngeal pumping, and forcible expulsion of water through the nares. Turtles which had not dived in the previous 24 hr developed intermittent bradycardia after an interval varying from 8 to 180 min. This bradycardia could be abolished by the administration of atropine. The periods of bradycardia at first lasted only a few seconds, but in most turtles they progressively lengthened and the heart rate fell to as low as 2-4/min (Fig. 4). Such rates were often maintained for over an hour after which the rate increased, although rarely to the predive value. In some turtles, the initial short periods of bradycardia did not lengthen even in dives lasting 3-4 hr. The time of onset of the bradycardia was influenced by the previous diving
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FIG. 3. Unanesthetized turtle, 2.5 kg; blood flow changes during diving. A: before diving, heart rate 43/min; B: during diving bradycardia, heart rate 4/min. From above down: right aortic arch flow (excluding brachiocephalic), left aortic arch flow, pulmonary flow.

FIG. 4. Unanesthetized turtle, 2.5 kg; changes in heart rate during diving (derived from left aortic flow record). A: before diving, heart rate 40/min; B: intermittent bradycardia; C: sustained bradycardia, heart rate 4/min.

history. If the turtle were forced to dive within a few minutes of a previous dive, bradycardia developed more rapidly and sometimes immediately. In all animals, attempts to surface produced a transient increase in rate. After surfacing, no change in heart rate occurred until the first breath was taken. The interbeat intervals then became progressively shorter until within 1 min the heart rate equaled or exceeded that before submergence. In the immediate postdive period, the respirations were deeper and more frequent than normal. During the next 15 min, both respiration and heart rate returned to the predive value.

When the turtle surfaced into 100% carbon dioxide, the bradycardia did not remit and became more profound although the predive rate could be rapidly restored by replacing the carbon dioxide with air. When the animal surfaced into an atmosphere of 100% nitrogen, the bradycardia remitted just as rapidly as when surfacing in air.

Pressure changes. Although pulmonary diastolic pressures changed little during diving, aortic diastolic pressure fell considerably. In a typical example, aortic pressure changed from 34/27 mm Hg to 36/16, while pulmonary pressure changed from 29/8 to 36/7 mm Hg. Comparison of the instantaneous pressure curves indicated that pulmonary pressure still began to increase before aortic pressure; but, in contrast to the predive situations, aortic and pulmonary pressures were identical in magnitude and contour from the opening to the closing of the aortic valves (Fig. 2B).

Flow changes. No consistent changes occurred until the onset of bradycardia. At this time, stroke flow in the pulmonary artery was reduced, whereas in both aortic arches and in the subclavian artery was increased (Figs. 2, 3). The duration of forward flow in all the vessels studied was increased. Cardiac output was strikingly reduced. In a typical turtle breathing air and with a heart rate of 40/min, the combined stroke flow of the pulmonary arteries and aortas (excluding brachiocephalic flow) was 3.7 ml and the minute flow was 148 ml/min. During diving, the heart rate fell to 2/min, the combined stroke flow to 3.3 ml, and minute flow to 6.6 ml. The distribution of blood flow between the pulmonary arteries and aortas also changed. Before diving, pulmonary flow was 60% of the combined flow, whereas during diving it fell to 40%.

DISCUSSION

In the intact unanesthetized turtle, pulmonary systolic pressure was 2-6 mm Hg below aortic pressure throughout systole. This finding differs from that of Woodbury and Robertson (8), who found this difference only during the last half or two-thirds of systole. Steggerda and Essex (7) found no consistent differences in the simultaneously recorded systolic pressures of the pulmonary and brachio-
cephalic arteries of Chelydra serpentina, although their illustration of simultaneously recorded aortic and pulmonary pressures shows that pulmonary systolic pressure is slightly lower than aortic systolic pressure. In our experiments pulmonary and aortic systolic pressures were equal only in cooled animals or during diving.

Aortic diastolic and mean pressures were invariably higher than pulmonary diastolic and mean pressures. Pulmonary flow and pressure consistently began to rise before the beginning of aortic ejection. This difference in timing was not commented on by Steggerda and Essex (7), although it can be seen in their records. Woodbury and Robertson (8) state that the pulmonary systolic pressure increase always precedes the aortic systolic pressure increase by the length of time required to raise pulmonary pressure to the level of aortic diastolic pressure. Our observations do not support this view. Figure 2 shows that in the intact nondiving animal at 20 °C the aortic and pulmonary pressure curves fail to coincide at any time during the cardiac cycle. Our findings agree with those of Woodbury and Robertson only during the bradycardia of diving.

Scholander (6) has shown that mammals rapidly develop bradycardia on diving. Millen et al. (5) did not observe this in fresh-water turtles. In contrast, we have consistently observed bradycardia in these animals during diving. Since this bradycardia could be reversed by atropine while the dive continued, it was almost certainly due to increased vagal activity, as is the bradycardia which occurs in diving mammals and birds. Anoxia alone cannot be the cause of the increased vagal activity since the bradycardia was reversed just as rapidly when the animal surfaced in nitrogen as when it surfaced in air. Hypercarbia, however, does appear to play an important role in maintaining bradycardia since surfacing into carbon dioxide prevented reversal of the bradycardia and the pulmonary and systemic blood flow changes. Similar conclusions have been made by Johansen (2) in snakes. Whatever its mechanism, the threshold for eliciting bradycardia remains lowered after surfacing since cardiac slowing developed much more rapidly when a turtle was resubmerged within a few minutes of a previous dive.

In air, the stroke output of the ventricle (excluding brachiocephalic flow) was approximately 1.6 ml/kg body wt. Sixty percent of this output was distributed to the lungs and the remainder to the systemic circuit. Although brachiocephalic flow was not measured, the magnitude by which pulmonary flow exceeded aortic flow suggests the presence of a left-to-right shunt and supports the similar conclusions of the dye-dilution studies of Millen et al. (5) and Steggerda and Essex (7).

During the bradycardia of diving, cardiac output fell dramatically to approximately 5% of the predive value. This low output could be maintained for over an hour. Although during this time the animal was sluggish and showed muscular hypotonia and diminished withdrawal responses, no irreversible damage seemed to occur since a complete recovery of cardiac output, muscle tone, and general reactivity occurred if the animal were allowed to surface. With the development of bradycardia during diving, a redistribution of the cardiac output occurred and the aortic flow exceeded pulmonary flow. Millen et al. (5) also observed this redistribution and stated that pulmonary blood flow ceases during diving. Complete cessation of pulmonary flow did not occur in our experiments.

Since stroke aortic flow increases while aortic pressure decreases, it is likely that systemic resistance decreases during diving. On the other hand, in the pulmonary circuit, stroke flow is reduced while pressure is maintained, suggesting that pulmonary vascular resistance is increased. The redistribution of cardiac output during diving appears therefore to be due to decreased systemic and increased pulmonary vascular resistance.

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REFERENCES