

LE COEUR AU COURS DE L'EVOLUTION ANIMALE

Annexe C, Chapitre 5

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Ce texte développe plus en détail les aspects phylogénétiques de l'anatomo-physiologie cardiaque mentionnés au Chapitre 5 sous la rubrique "Mécanique ventriculaire".
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Introduction

Facing hemorrhage, sepsis or metabolic stress, heart failure is an unfortunate but frequent event in operating rooms and intensive care units. Caregivers in charge of patients suffering from ventricular insufficiency might wonder sometimes if our heart is really a good pump ! Although it might appear conceited to judge the work of nature, it is possible to search for an answer at this question on two different levels. First, we can observe the mechanics of ventricular contraction and relaxation under the point of view of an engineer who would have to build such a device; this is developed in the Chapter 5 (Mécanique ventriculaire). Second, we can look at the attempts and successes of animal evolution, starting from the peristaltic tube of primitive worms and progressing up to the four-chamber heart of birds and mammals. Analyzing the pumping performances of the heart under this evolutionary angle might give a clue to the adaptation of the heart to the constraints imposed by the propulsion of blood in adequacy with the needs of the organism.

Human heart is a sophisticated and robust system, rather well adapted to the requirements of the body. But the question remains: is it a the best possible pump ? Is a four-chamber heart, connected to two

distinct circulations with the same output but different pressures, the most appropriate system to carry oxygen and nutrients ? Looking back at 800 million years of animal evolution, it appears that nature has experimented many different devices for propelling blood, has constructed various circulatory systems for feeding the organs, and has used several carriers for transporting oxygen. This review will be devoted to the examination of the most characteristic pumping systems in the different classes of animals, their advantages and failures, and the way they have been modified with evolution. Comparing human heart with these various achievements, we might be able to pronounce a more founded judgment on the pump we inherited from our animal ancestors.

The tree of evolution

The evolution of living animal species is usually represented as a ramified tree, which roots are anchored at the level of the bacteria and the first prokaryotic cells appeared 1.5 billion years ago. Multicellularity started about 800 million years ago. From the metazoa, which are heaps of cells, emerged the Poriphera (sponges) and the Cnidaria (coral, jellyfish), organized animals with a radial symmetry and a single central body cavity where occur gas exchange, food convection and reproduction. They consist of only two embryologic layers, the ectoderm and the endoderm. Stemming from the Cnidaria, remote common ancestors developed a bilateral symmetry, a clear dorsoventral axis, and added a third layer, the mesoderm [9]. These Bilateria gave two divergent types of animals according to their embryologic development (Figure C1).

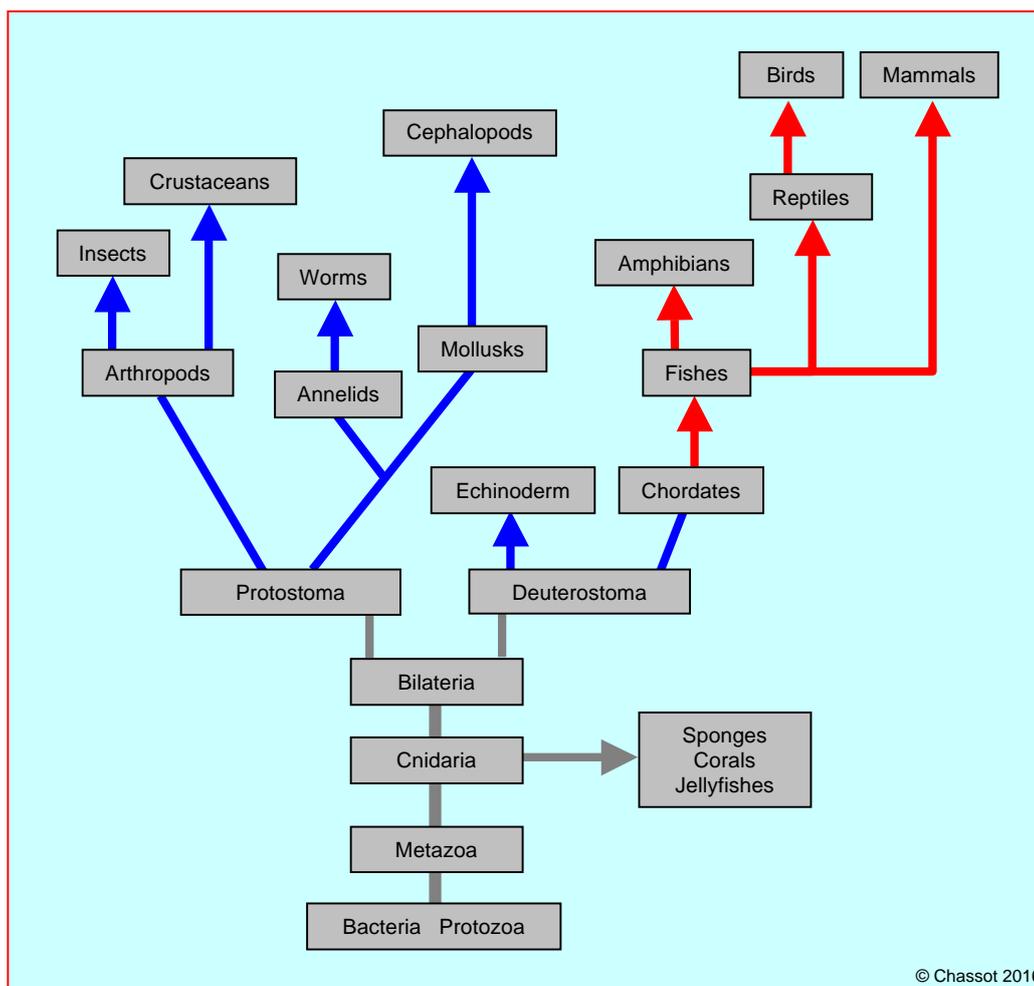


Figure C1. Example of a simplified tree of evolution. Fossils show that from Cnidaria (coral, jellyfishes, sponges), which have a radial symmetry, Bilateria developed a lateralized symmetry and two types of embryologic growth: in Protostoma, the first opening appearing in the embryo is the mouth, whereas in Deuterostoma it is the anus. Protostoma gave the annelids (worms), the arthropods (insects, arachnids and crustaceans) and the mollusks. The Deuterostoma gave the echinoderms (sea urchins, sea stars) and the chordates (tunicates, lancelets). From the chordates evolved the vertebrates (fishes, amphibians, reptiles, saurians, birds and mammals). All chordates have a backbone protecting a dorsal nervous cord, and use hemoglobin as an O₂-carrier pigment, whereas Protostoma may have hemoglobin, hemocyanin (copper-containing pigment) or only dissolved O₂. Blue arrows: invertebrates. Red arrows: vertebrates.

- The Protostoma (annelids, arthropods, mollusks) develop in such a way that the first opening appearing in their embryo (blastopore) is the mouth. The early cleavages of the morula lead to already committed cells. They have the general morphology of a "tube-within-a-tube". The space between the digestive tract and the outside wall is the coelom. It contains the different organs and vessels, and is filled with a fluid which assists circulation and respiration by diffusing nutrients and oxygen. The dorsal vessel carries blood anteriorly and the ventral vessel posteriorly. When it exists, the neural cord is ventral.
- Deuterostoma (echinoderms and chordates) develop the distal extremity of the digestive tract, the anus, as the first opening. The early cleavage cells of the morula are totipotent. Their dorsal vessel carries blood posteriorly, and their neuronal cord is on the dorsal side, usually protected inside a fibrous sheet [75].

Chordates, which are primitive beings represented nowadays by strange animals like tunicates and lancelets (see below), are the root on which emerged the vertebrates: fishes, amphibians, reptiles, saurians, and subsequently birds and mammals. This progressive development is usually represented by a "tree of evolution", which is depicted under its conventional shape in Figure C1, although new molecular analyses have modified our understanding of phylogeny [28]. With amphibians and reptiles, vertebrates started to leave the aqueous environment, to walk on earth and to breathe air with a more and more efficient respiratory apparatus. With birds and mammals, they became homeotherm, keeping a stable temperature at 37-38°C. The cardiovascular system adapted to this new modalities by developing multi-chamber hearts, high perfusion pressure, closed circulation with arteries, capillaries and veins, and sophisticated cardiovascular regulations. All Chordates use hemoglobin as an O₂-carrier pigment, whereas Protostoma may have hemoglobin, hemocyanin (copper-containing pigment) or only dissolved O₂.

The first tubular heart showed up probably in the bilaterian ancestors 600-700 million years ago, when appeared the mesodermal layer. Actually, the embryogenesis of animals as remote as insects, mollusks, fishes, birds and mammals shows the same common step of a primary cardiac tube derived from bilateral, anterior mesodermal precursors that converge at the midline [11]. Bilateralia initiated developmental pattern processes that have been easily used as a platform for further evolutionary variations when conditions made them possible [23]. These striking similarities speak for a common mechanism of cellular differentiation of cardiomyocytes transmitted all along the scale of animals, on which subsequent specific developmental programs lead to a variety of structures and functions [10]. The primitive cardiac tube, devoid of chambers or valves, had probably some kind of peristaltic movements moving fluid into pericellular spaces, without true vessels or unidirectional flow [9]. During the Cambrian era (from 550 to 480 million years ago), isoforms of actin, myosin and troponin appeared and permitted to build up a striated musculature, which was incorporated in the first pulsating bulges of the primary tubular heart [57]. The cardiomyocytes of all animals carry the same beta-adrenergic receptors, use cAMP as second messenger, and benefit from a similar Ca²⁺-sensitive contractile mechanism [70,91].

General principles

As long as living beings were only clusters of cells, simple diffusion of oxygen and nutrients was adequate, because the distances between cells were very short. Diffusion is still appropriate for gas exchange and transport if the body diameter is less than 1 mm, and if the envelope is freely permeable to gases [14]. But as soon as the organisms started to grow in size and multiplied the internal organs, a transportation system became mandatory to carry nutrients and oxygen (O₂) and to remove waste products and carbon dioxide (CO₂). There are two possible configurations for the circulation of a fluid through the organism.

- Open system with a high output but a low pressure, and a priority to flow. The mechanical analogue would be a fan in a room. This system has good performances over short distances. It is frequent among primitive organisms like annelids, non-cephalopod mollusks or tunicates, and in the peripheral circulation of most arthropods (insects, spiders). Blood and lymph are usually mixed together in these animals. This mixing, called *hemolymph*, circulates towards the periphery in arterial vessels, and then flows freely into the interstitial space, bathing the tissues and organs. It is directed by a network of sinuses into venous collectors, which bring it back to a central vein. This system requires a large volume of circulating fluid (50% of body weight in mollusks and tunicates). The circulation time is about 6 minutes for a few centimeter long animal [54].
- Closed system with high pressure allowing to carry blood rapidly over long distances inside elastic tubes (arteries), capillaries and veins. It is able to track reliably the variations in demand of the organism. It is similar to any mechanical pressure pump, and is used in all vertebrates, in cephalopods (octopus) and in echinoderms (sea urchins). Blood is separated from lymph, and is continuously enclosed in a vascular system lined with specialized cells (endothelium). The blood volume is much lower than in the previous case (6-8% of body weight), and the system is much faster (≤ 1 minute of circulation time in human).

There are basically three types of pumps in use among animals (Figure C2).

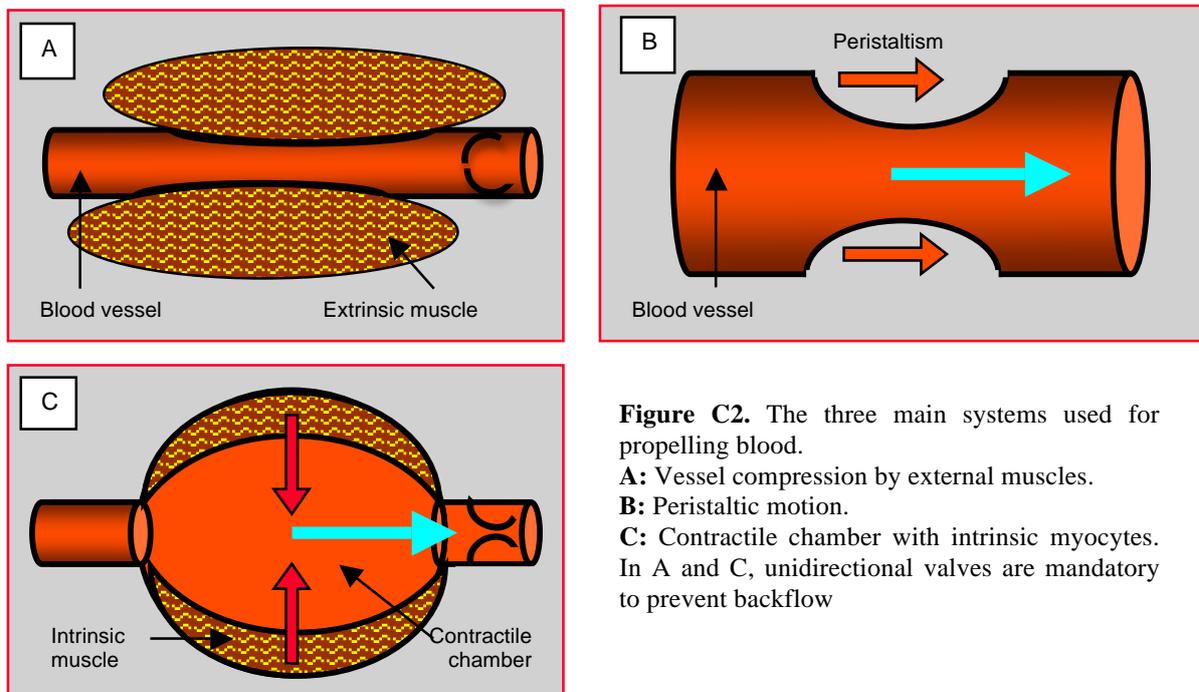


Figure C2. The three main systems used for propelling blood.

A: Vessel compression by external muscles.

B: Peristaltic motion.

C: Contractile chamber with intrinsic myocytes.

In A and C, unidirectional valves are mandatory to prevent backflow

- Peristaltic propulsion, similar to the digestive tract. The flow wave is continuous and slow, the pressure is low (< 20 mmHg). This system is in use among annelids (worms) and in the primary cardiac tube of the embryo. Because of the peristaltic wave in one single direction, valves are not necessary.
- Compression by extrinsic musculature, like in the caudal heart of hagfish (Figure C15, page 21) or in the venous circulation of lower limbs in humans. The pressure generated is low. Valves are necessary to maintain the flow in one single direction.
- Contractile chamber which pushes blood by compression into a vascular tree. This is the most commonly used system in nature. The chamber can be unique (vertebrates) or multiple (series of aortic bulbs in insects). This system requires valves on both extremities, an inflow valve to prevent backflow into the venous side in systole, and an outflow valve to prevent backflow from the arterial side in diastole.

In all animals, the blood is flowing in one direction only, excepted in tunicates. These chordates look like colored bags anchored at the sea floor or on the reef. They have a peristaltic tube-shaped heart, which moves the blood forwards. After several dozen of contractions propagating at 0.5 cm/s in one direction, the motion ceases and the contraction reverses direction, pumping the blood in the opposite direction [39]. This the only arrangement where the blood is conveyed in one sense for bringing nutrients and oxygen and in the reverse sense for removing waste and carbon dioxide.

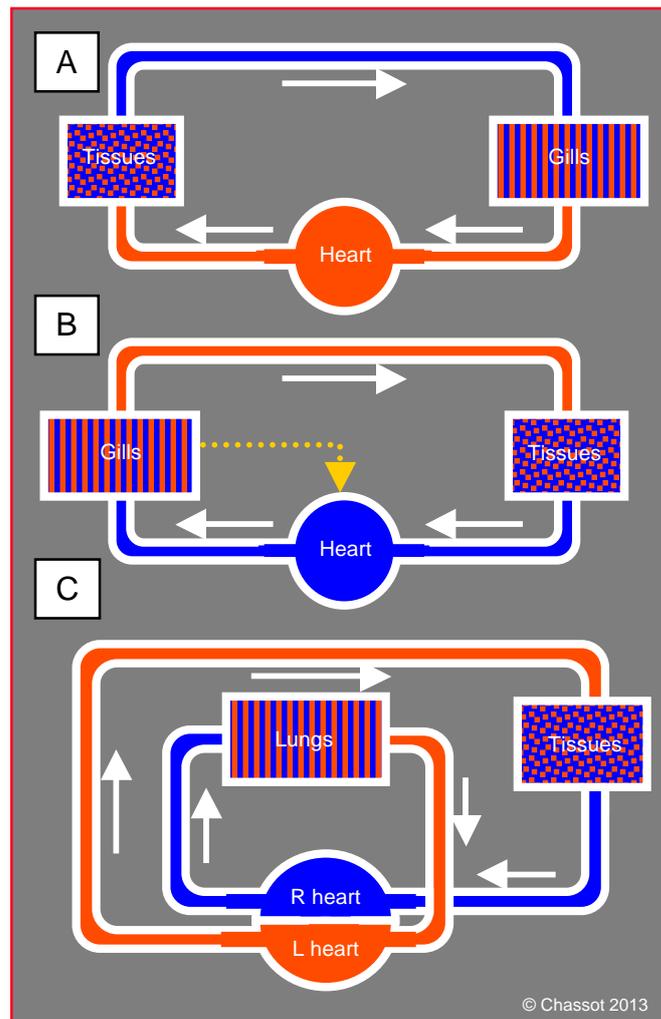
Several assemblies are possible to provide adequate metabolic and gaseous exchanges between the tissues and the surrounding world (Figure C3).

Figure C3. Different assemblies of the cardio-circulatory system.

A: Single circulation, with the gills placed on the venous return (mollusks); the heart receives oxygenated blood and propels it to the tissues.

B: Single circulation, where the gills are placed between the heart and the tissues (fishes); the heart receives venous blood, but is itself perfused with oxygenated blood conveyed directly from the gills (dotted arrow).

C: Double circulation (systemic and pulmonary), which may have the same pressure and some shunts (frogs, turtles, most snakes) or different pressures and no shunt (birds, mammals). In the first case, the heart has only one ventricle (three-chamber heart), the pressure is identical in the pulmonary and systemic arteries; in the second it has two ventricles, with different pressures for the lungs and for the systemic organs.



- The gas exchange may take place through the skin (worms), abdominal pores (insects), gills (mollusks, fishes) or lungs (reptiles, birds, mammals).
- The gills can be mounted on the venous return (mollusks); the heart receives oxygenated blood and propels it to the tissues.
- The gills can be placed between the heart and the tissues (fishes); the heart receives venous blood, but is itself perfused with oxygenated blood conveyed by an arterial network coming directly from the gills.
- The higher animals have two separate circulations (systemic and pulmonary), which may have the same pressure (amphibians, turtles) or different pressures (birds, mammals). In the first case, the heart has only one ventricle with more or less septation and some shunting between the two circulations; in the second case, it has two ventricles and no shunt.

The amount of O₂ transported to tissues is proportional to the cardiac output, the blood volume (or the concentration of O₂-carrier pigment, when present) and its O₂ content (dissolved or fixed on a transporter). It is also dependent on the gas exchange system, which can be, by order of increasing efficiency, the skin (worms), the gills (mollusks, fishes) or the lungs (reptiles, mammals, birds). Improvement in gas exchange and the presence of an O₂-carrier pigment decrease 20 to 40 times the cardiac output necessary to provide the same amount of O₂ to the tissues. This is particularly important when O₂ requirements are increasing with high physical activity (octopus, reptiles) or with temperature control (homeothermia in birds and mammals). Slow and sluggish animals have low needs in O₂ and can avoid investing energy in the complicated synthesis of copper- or iron-based respiratory pigments.

Many different heme-binding proteins like cytochromes or peroxidases, are present among plants and animals. Hemoglobins are some of them. They are very common, and have been found in practically all classes of living beings from bacteria to man. They evolved from heme enzymes, and were probably used primarily to detoxify oxygen when it appeared in atmosphere with bacteria and plant photosynthesis. From an O₂-scavenging function, they became progressively involved in the transport of this gas once the aerobic respiratory chain was settled in the cells. The gene directing the expression of the heme ring and the globin chains can be traced back to the divergence between vertebrates and invertebrates 670 million years ago [29]. Invertebrate hemoglobins have different structures, frequently simplified, and are found free in the cytoplasm of cells, dissolved in various body fluids, or locked in circulating erythrocytes. The distribution of hemoglobin among nonvertebrate species is episodic, whereas it is constant among all vertebrates [88]. Hemocyanin is a copper-containing molecule giving a greenish color to the blood. It is the most widely used O₂-carrier after Hb, but it is found only in solution and never in concentrations that would transport more than 4-5 vol% of O₂ [89]. It is less efficient than Hb, since it carries 12 mL O₂ per liter instead of 210 mL O₂ L⁻¹ for Hb. It is found in mollusks, crustaceans and some arachnids.

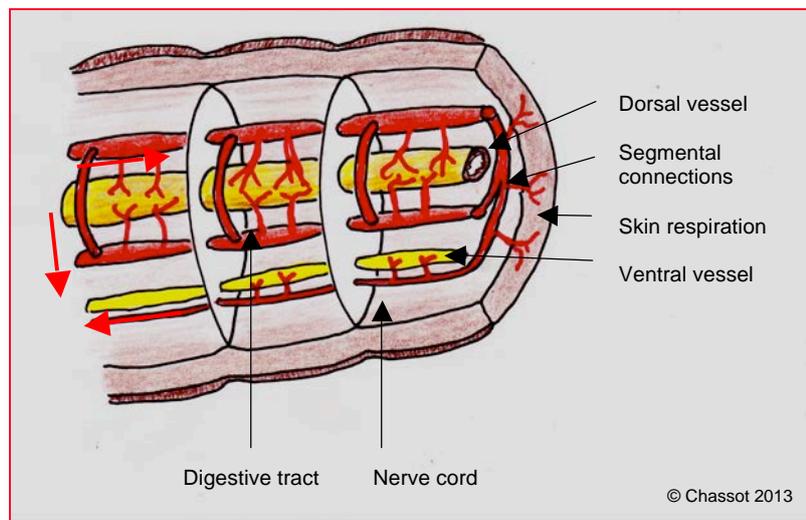
In order to understand why the human cardiovascular system is built the way it is, it will be necessary to follow the different types of hearts and circulations progressively selected during the chain of evolution.

Invertebrates

Lower invertebrates

Annelids (worms) evolved over varied systems in order to enhance convective transport between anatomical compartments. The most primitive worms have one single circulation made of two peristaltic tubes, one dorsal, where the blood is pumped forwards, and one ventral, where it flows in the opposite direction; they are linked together by lateral connections. In many species, the first 5 segmental connections, called aortic arches, function as accessory hearts. Distally, the system is open with no capillaries (Figure C4) [65]. These vessels carry hemolymph between the digestive tract, the tissues and the skin, where gas exchange takes place. The perfusion is slow (6-8 peristaltic waves per minute) and the hemolymph pressure is low (< 12 mmHg). Flow is continuous and flow variations are very restricted. This system is compatible only with a slow metabolism and with a low oxygen consumption (VO_2). Most worms breathe through the skin, but some have developed primitive gills integrated in the vascular system [68]. The most evolved among them, like the giant Brazilian earthworm (up to 120 cm and 500 gm), have an iron-containing pigment close to hemoglobin for carrying O_2 over rather long distances. This species belongs to a class of advanced worms (Polychaeta), which circulatory system consists of one peristaltic dorsal vessel pushing blood (pressure 20 mmHg) towards five pairs of segmental lateral hearts in the front part of the animal. When they contract, these accessory hearts push blood into the ventral vessel at a pressure up to 70 mmHg. Each segment is perfused by a pair of parapodial vessels arising from the dorsal and the ventral vessels and a rich network of capillaries. The ventral nerve cord is supplied by an independent longitudinal vessel [70].

Figure C4. Cardiovascular system in worms. In the three segments represented, the blood is pumped forwards by the peristaltic motion of the dorsal vessel and supplies the digestive tract and the nerve cord by segmental connections linking the dorsal and the ventral vessels together. Small vessels to the skin function for gas exchange. In many species, the first 5 segmental connections function as accessory hearts.

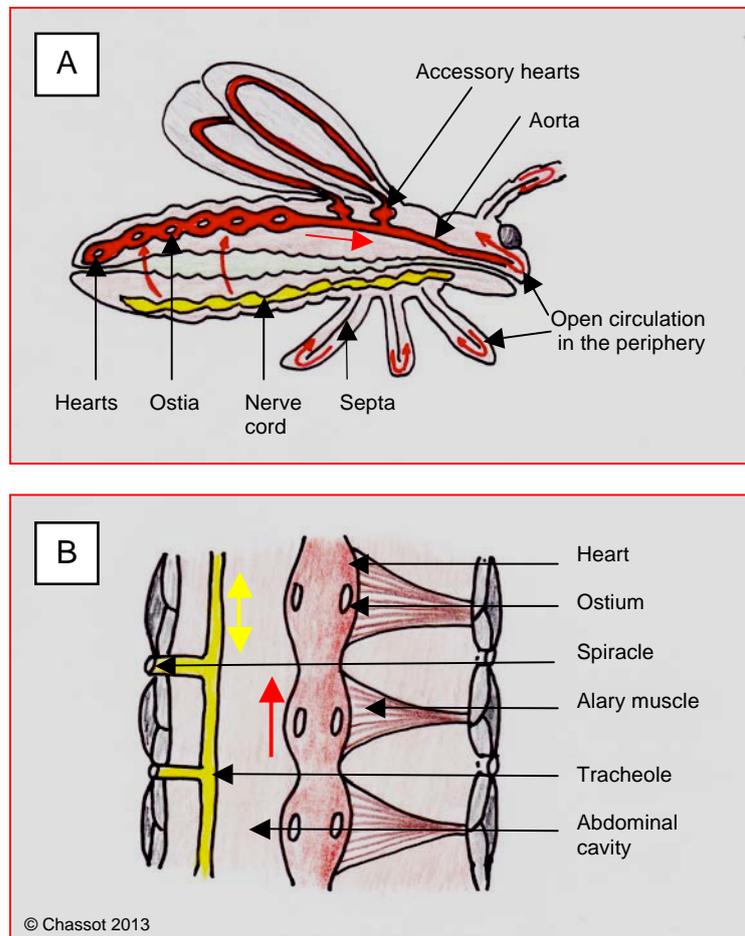


Arthropods

Among arthropods, insects (fly, beetle, butterfly, cockroach) have a series of pulsatile abdominal bulbs (hearts-in-series) along a large dorsal vessel (aorta). These contractile chambers are composed of a dense inner layer of myocytes, and a single-cell, epicardial layer of fibrocytes. The myofibrils are spirally or circularly oriented. This arrangement facilitates the wringing of the heart cavity and the ejection of the hemolymph. The synchronized contraction of the hearts propels the blood into the anterior aorta and its branches at a low pressure (20/10 mmHg). The wave of contraction is initiated near the blind caudal extremity, and propagates rapidly in the anterior direction (32 mm/s). Most insects have a fast heart rate (300-600 beats min^{-1}), with cyclical interruptions of a few seconds [76].

The filling of the hearts in diastole is achieved by suction, which is provided by the contraction of muscles (*alary muscles*) pulling externally on the heart walls (Figure C5). At the periphery, the circulation is open, without capillaries or endothelium. The blood flows freely into the interstitial space within the shell and the organs, where it is directed by a network of membranes and lacunae into venous sinuses which bring it slowly back to small lateral valved openings (*ostia*) of the abdominal hearts, where it is pumped inside.

Figure C5. Cardiovascular system of insects. **A:** Schematic drawing of a fly. Each abdominal segment has a contracting bulb functioning as a heart and propelling blood cephalad into the anterior aorta. The peripheral circulation is open and directed by septa. The blood is flowing back to the abdominal hearts where it enters through lateral openings (*ostia*). Accessory hearts at the root of the wings or the legs improve blood flow in these structures with high metabolic requirements (adapted from Schmidt-Nielsen 2002). **B:** Three abdominal segments (horizontal slice). In diastole, the hearts are pulled from outside by alary muscles inserted on the carapace. Insects breathe through small valved openings (*spiracles*) situated on each segment of the abdomen. The air is transported by a highly branched tree of small tubes (*tracheoles*) all the way to the tissues. Cyclic abdominal contraction and relaxation provide the ventilatory drive through these tubules (yellow arrow). Red arrow: blood direction towards aorta.



This peripheral open system limits the size of insects, spiders and scorpions to less than 15 cm, because the interstitial diffusion of gas and nutrients is efficient only over short distances. The number and identity of myocytes forming the cardiac system remain unchanged during the life of an insect, but the myocardial cells grow in size 200-500 times from the larva to the adult [52]. Although the lifespan of a fly like the *Drosophila* is only a few weeks, its heart grows old, and shows senescence as in mammals: loss of performance, decrease in heart rate, arrhythmias, myofibrils disorganization [55]. The cardiomyocytes of insects react to cardiovascular drugs as a mammalian heart, increasing heart rate and contractility with adrenaline and decreasing them with calcium-blockers [76].

Since the resistance to flow is rather high in appendages, and since the abdominal heart system is rather weak, insects have accessory hearts to assist the circulation of fluid in antennae, wings, limbs or abdominal cerci. The anatomy and function of these supplementary pumps vary according to the different species and to the degree of activity. Most of them are pulsatile structures functioning by suction of the hemolymph out of the wings (flies, bees), legs (locust) or antennae (coleoptera). The aspiration is obtained by different mechanisms. For example, the contraction dilates the ampullae at the base of the antennae because the muscle is inserted on the carapace, like the alary system. At the base of the wings, the accessory heart is a modification of the dorsal aorta enclosed beneath the dorsal

thoracic wall; the local contraction of the aorta decreases the surrounding pressure inside this rigid housing, and aspirates the hemolymph from the wings back into the body cavity. These accessory hearts work independently from the aortic central hearts and have their own beating rate (200-300 beats min⁻¹), which is directly related to the activity of the involved appendage [59]. Despite these clever mechanisms, the flow rate is slow: it takes 10 minutes to exchange the hemolymph of an antenna or a wing [60]. In arachnids (spiders, scorpions), the multiple abdominal hearts have fused into a single contractile chamber, which is a tube with several ostia, located in the dorso-abdominal area. It functions in the same way as the heart of insects and ejects the hemolymph into an anterior and a posterior aorta.

Insects breathe through small openings situated laterally on the abdomen (spiracles), and air is carried along a tracheal system consisting of very fine tubes extending to the tissues. Respiration is performed by expansion and contraction of the abdomen, which has also a major pumping effect on hemolymph, because the pressure raised by the abdominal contraction is higher than the pressure generated inside the heart. At low metabolic rate, a slow flow of hemolymph is sufficient, and the dorsal pulsating aorta is able to maintain an adequate output. But during physical activity, the abdominal pump is required to provide an accelerated flow through the organism [81]. The rate of the abdominal pump is around 60 beats/min [76]. Since the hemolymph percolates in an open system inside the carapace, the movements of the whole body participate in the flow propagation from and into the dorsal vessel: abdomen, appendages, wings, limbs, organs, they all ensure the circulation of gas, nutrients and wastes by their actions. In insects, the role of the central heart is much less important than in vertebrates, because the dorsal pulsating vessel cannot respond to the metabolic needs without the help of accessory hearts and abdominal pump. On the other hand, hemolymph circulation does not have to provide oxygen to the tissues, since ventilation is carried out by the tracheoles system and the abdominal motion. This mechanism, associated with the open circulation in the periphery, allows insects to withstand incredible accelerations when they fly or jump. A flea, for example, sustains accelerations of more than 150 “g” (gravity force, or acceleration of an object at the earth surface: 9.81 m/s²) when it springs. Its brain undergoes no “black veil” because oxygen is transported by air tubes and neurons are continuously surrounded by the hemolymph bathing the cells.

Blood is not involved in oxygen transportation in insects. In spiders and scorpions, however, blood carries O₂ because these animals have abdominal respiratory organs called *book-lungs* resembling sheets of tissue alternating with air clefts. They are perfused by venous blood on the way back to the heart. Some species have a copper-containing respiratory pigment called hemocyanin.

Crustaceans present a wide spectrum of cardio-vascular systems, extending from primitive conduits without any heart in small crustaceans of the plankton, to complex pulsatile circulation with variable amount of a respiratory pigment in large animals like decapods (crabs, lobsters). The latter have a single heart situated in the thorax. It is suspended inside a rigid pericardium by elastic alary ligaments, which provide diastolic expansion (Figure C6). The venous blood returns inside the pericardium and bathes the contractile mass. In diastole, it enters the ventricular cavity through channels (*ostia*) crossing the myocardial wall. In systole, the contraction closes these holes and blood is ejected into the anterior and posterior aortas. The main difference with arthropods is the presence of gills for gas exchange. They are placed on the venous return to the heart, which is therefore vascularized by arterial blood. The hemolymph of crabs and lobsters is greenish because of the Cu²⁺-containing O₂-carrier pigment hemocyanin. Crustaceans have a sophisticated control of heart rate (average 70 beats/min) and stroke volume by nervous input from the cardiac ganglion or the brain, and by direct action of neurohormones on the cardiac muscle [51]. The absence of smooth muscles in the arteries is compensated by the contraction or relaxation of muscular cardio-arterial semi-lunar valves situated at the base of each of the 5-7 arterial vessels originating from the heart. This system allows the flow and the blood pressure (30-40 mmHg) of arteries vascularizing different parts of the organism to be controlled independently from each other [49]. During walking or digestion, for example, the oxygenated hemolymph is directed preferentially to the limbs or to the digestive tract, respectively.

The vascular tree of decapods is extremely complex. In large crabs, the seven arteries arising from the heart give small arterioles ramifying deeply within the tissues and splitting into fine capillary-like vessels (Figure C7) [50]. The arterial vascular system is closely applied to the central nervous system, which has therefore a priority in blood distribution. At their extremities, the vessels are "open", lacking a true capillary and venous system, which is replaced by a network of lacunae collecting fluid into venous sinuses. However, these lacunae are as fine as capillaries, and the sinuses form distinct units. Moreover, brain and antennal gland have an uninterrupted continuity between arteries and veins with fine channels of the same size as vertebrate capillaries [49]. Therefore, the system can be better termed "incompletely closed" [65].

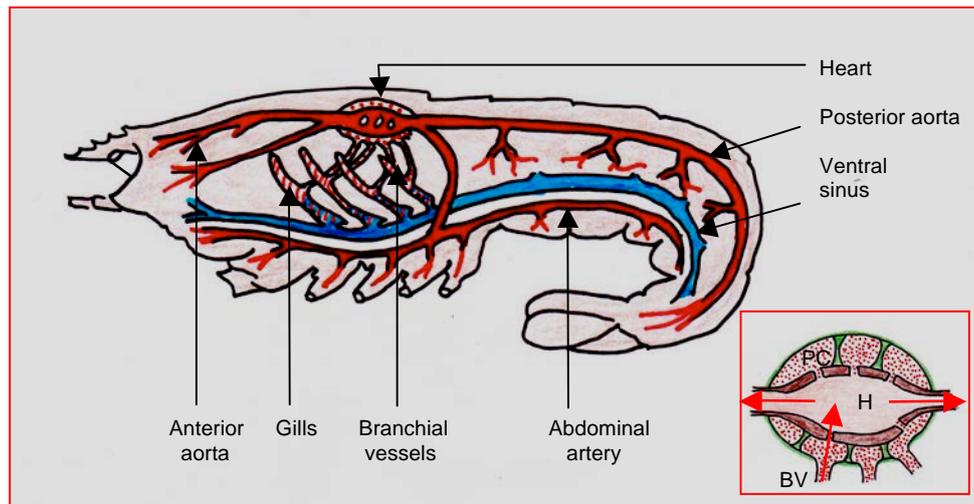
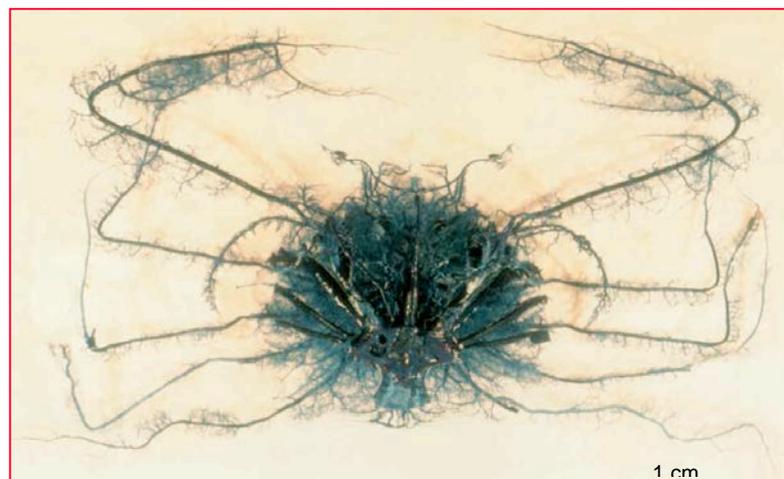


Figure C6. Cardiovascular systems of crustaceans illustrated by the longitudinal slice of a lobster. The unique heart (H) is one simple contracting chamber pushing blood in anterior and posterior aortas. The oxygenated blood (red dots) arrives from the branchial vessels (BV) inside the pericardium (PC, pericardial cavity), which functions as a large sinus. In diastole, it enters the heart cavity through ostia perforating the myocardial wall. These ostia close with the muscular contraction. The diastolic expansion is provided by the recoil of elastic ligaments suspending the heart to the pericardium (in green). In periphery, the circulation is open, and the blood returns from tissues to the gills through a large ventral sinus similar to a central vein (adapted from Schmidt-Nielsen 2002, ref 70).

Figure C7. Corrosion cast of the arterial vascular system of a crab (*Callinectes sapidus*). The seven arteries arising from the heart give small arterioles splitting into fine capillary-like vessels extending deep into the limbs, the claws, the antennae, the gills and the organs (McGaw 2005).

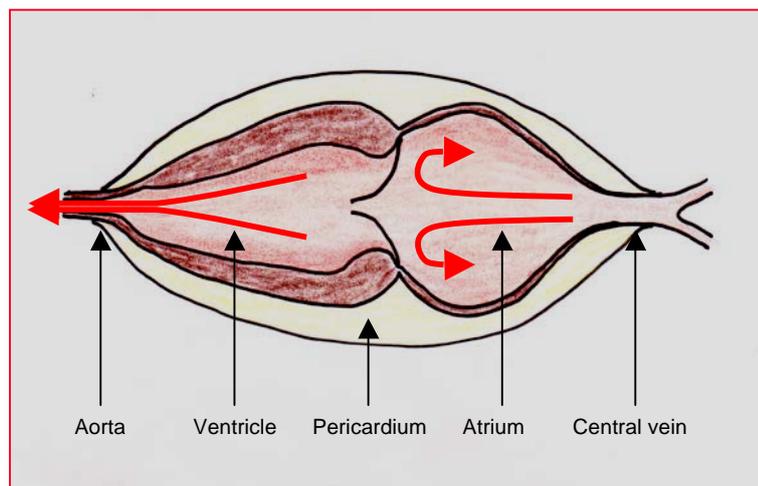


Mollusks

Gastropod mollusks (snails, mussels) appeared 500 millions years ago. They are the first animals in evolution to have a two-chamber heart consisting of a ventricle and an atrium, enclosed inside a pericardial envelope (Figure C8). The presence of an atrium has a double advantage: 1) it acts as a buffer reservoir between the continuous venous return and the pulsatile ejection, and 2) its contraction can raise the ventricular end-diastolic pressure, increasing the Frank-Starling effect without increasing the mean central venous pressure. This primitive, but individualized, ventricle increases its stroke performance when preload is elevated. It has also an autonomous rhythmicity, but with frequent pauses lasting up to 30 seconds (at 10°C). The average heart rate of snails and mussels is 20 beats min^{-1} , and their blood pressure is 10-20 mmHg [12]. Gas exchange takes place in gills or lung placed on the venous circuit, between the organs and the heart, which is therefore perfused with arterialized blood. The O_2 -carrier pigment is hemocyanin.

With the exception of cephalopods, mollusks have an open circulatory system in the periphery. Hemolymph and interstitial fluid are therefore mixed together and may constitute as much as 50% of mussel body weight without the shell [47]. Nevertheless, the O_2 -carrying fluid is distributed to the different organs according to their O_2 consumption and not to their respective mass. The kidney receives 8 times more hemolymph than the foot muscle, despite the fact that the latter represents 60% of the body mass [36]. Instead of the bath of interstitial fluid, the compact cerebral ganglia have an extensive bed of small vessels, first appearance of a capillary-like network. Besides gas exchange and nutrient transport, the circulatory system of gastropods has a secondary function: it serves as a pump for the head-foot, which has therefore a hydraulic mechanism on top of the muscular contraction to move the animal [12].

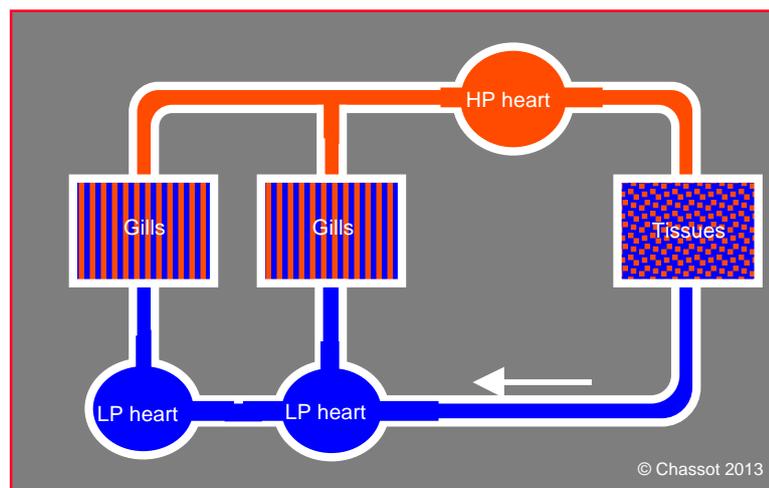
Figure C8. Heart of mollusks. The heart is made of two chambers in line, one ventricle and one atrium. At their junction, a unidirectional valve prevents backflow during ventricular contraction. The pericardium contains a discrete amount of fluid.



Cephalopods (octopus, squids) are the most sophisticated mollusks. They have a closed system of arteries, capillaries (diameter 10-20 μm) and veins lined by endothelial cells, and a double-pressure circulation. First, a two-chamber heart collecting blood from the gills propels it into the systemic circulation at a high pressure (50-70 mmHg). Cardiac output can be adapted to exercise by a 2-3 fold increase in stroke volume (from 1.5 to 5 mL kg^{-1}) and heart rate (from 40 to 100 beats min^{-1}) [12,89]. Fast-swimming squids have a much higher stroke work (4.2 J kg^{-1}) than the sedentary abalone (0.3 J kg^{-1}) [36,90]. Secondly, two accessory pulsatile cavities (right and left branchial hearts) push the blood through the gills on each side of the body under a lower pressure (20 mmHg) (Figure C9). Without these low-pressure hearts, the venous blood would not have enough kinetic energy to cross the gills with an adequate velocity. There is therefore a single circuit functioning in series, but two different pressure regimes. This is the first appearance of a “pulmonary” flow at low pressure to facilitate gas exchange, and a systemic circulation at high pressure to ensure more efficient blood distribution

according to the metabolic needs and the increased activity patterns associated with predatory behavior and active swimming. It is also the first appearance of a completely closed vascular system, with elastic arteries, capillaries and veins, although the endothelium is quite permeable and is not as a selective barrier as in vertebrates. But its predominant role is probably to maintain a laminar flow throughout the vascular tree, and to prevent the turbulences and eddies which appear when flow drops into lacunary spaces or when interior surfaces of vessels are not regular and smooth [15]. A closed vascular tree means also that intravascular and interstitial fluids are completely separated. In cephalopods, they represent 6% and 15% of body weight, respectively [47]. Moreover, cephalopods have sophisticated cardio-circulatory regulatory mechanisms, which coordinate cardio-ventilatory performance with metabolic needs [78]. Octopus and squids ventilate their gills by pumping water into the mantle cavity, and by ejecting the water through the siphon.

Figure C9. Circulatory system of cephalopods (octopus, squids). The systemic heart generates a rather high pressure (HP) (up to 70 mmHg). However, the gills are situated at the end of the venous system coming from the tissues. Two accessory branchial hearts generating low pressure (LP) (20 mmHg) provide the kinetic energy necessary to cross the gills.



Cardio-respiratory interactions are important in mollusks as well as in mammals. The ventilatory pumping activity of the mantle wall in cephalopods, of the shell muscle in snails, and of the gills cilia in mussels is a significant help for fluid circulation, because it increases the venous return. On the other hand, the systolic pulsation of the anterior aorta and the pericardial volume change with the beating of the ventricle push the blood alternately into and away from the branchial chamber. This rhythmic change in chamber volume favors gas exchanges across the gills or the lungs [12].

Vertebrates

Vertebrates are characterized by a central multi-chamber heart, and a completely "closed" vascular system with selectively permeable capillaries. These capillaries are entirely covered by an active endothelial cell lining with multiple functions, involved in gas diffusion, nutrients transport, vasomotor tone, hemostasis and cell growth. The blood contains red and white blood cells and coagulation factors, and is completely separated from the lymph. The O₂-carrier pigment is hemoglobin, locked up inside red blood cells. Myocardial mass varies from 0.2% of body weight in fishes to 0.3-0.5% in reptiles and to 0.8-1.2% in birds; in mammals, man included, it represents 0.6% of body weight [70,71]. With lungfishes, amphibians and reptiles, vertebrates moved from an aquatic to a terrestrial environment 350 million years ago, where they had to breath air and to stand up against gravitational forces. With birds, they have even conquered the air. Finally, birds and mammals have managed to keep their blood warm, which makes them more independent from the surrounding temperature. These multiple physiological challenges explain the wide variation in heart morphologies found among vertebrates, although heart structure is rather uniform inside each class (Figure C10).

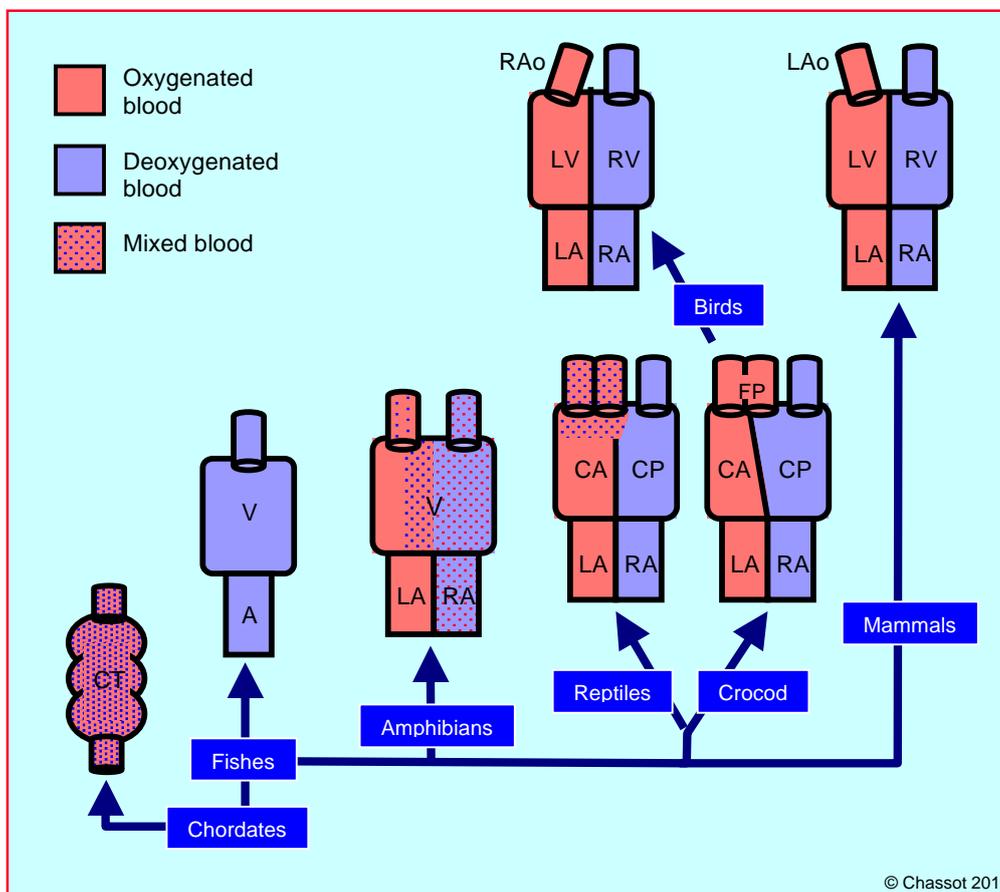


Figure C10. The different heart configurations among vertebrates. Tunicates are the closest animals to the chordate ancestor of all vertebrates ; they have a single contracting tube (CT) for blood propulsion. Fishes have a single atrium and a single ventricle ; it contains venous blood since the gills are placed between the heart and the tissues. Amphibians have two atria but a single ventricle with some degree of blood mixing between aorta and pulmonary artery ; the systemic venous blood is partially saturated in O₂ because the skin participates to gas exchange. Reptiles have two atria and one ventricle divided by a septum in two cavities (cavum pulmonale CP, cavum arteriosus CA) communicating through an interventricular foramen ; the two aortic arches receive arterialized blood with variable O₂ saturation. Crocodiles have a four-chamber heart and two aortic arches communicating together by the foramen of Panizza (FP). Birds and mammals have a true four-chamber heart with a single aorta, which is a right aortic arch (RAo) in birds and a left aortic arch (LAo) in mammals.

Anatomic evolution of the circulatory system

During the Cambrian era, the Deuterostoma, which were headless, eyeless, worm-like creatures, gave birth to the Chordates. Currently, all vertebrates stem from this common chordate ancestor (Figure C10). The extant chordate phylum comprises: 1) the urochordates or tunicates ("sea squirts"), 2) the related cephalochordates ("lancelets"), and 3) the vertebrates. As already mentioned, chordates are small, primitive, marine animals looking like bags or anchovies. In common with vertebrates, cephalochordates and larvae of tunicates share a dorsal neural cord protected inside a notochord made of densely packed cells, which is a primitive backbone. But cephalo- and urochordates still have an open circulation and a non-directional peristaltic heart, with several other vessels capable of rhythmic contractile movements and acting as accessory hearts. The heart of the first vertebrates evolved from the chordate main heart, which was a series of contractile chambers along a tube. In vertebrates, this cardiac tube made of primary myocardium differentiates into sequential segments during embryogenesis according to the flow direction: the *sinus venosus* and the atrium for the inflow, the atrio-ventricular canal, the ventricle and the *conus arteriosus* for the outflow. The primary myocardium is characterized by a poor contractility and a slow propagation of cardiac impulse; it will give essentially the AV canal and the heart valves. Under the transcriptional control of different genes, the inflow and outflow segments will differentiate in fast-conducting, well-contracting and highly Ca^{2+} -sensitive myocardium, which will constitute the main component of the ventricular and atrial chambers [11].

Echinoderms (sea urchins, sea stars) are a separate branch originating from the Deuterostoma (see Figure C1). They have retained a peristaltic system for blood circulation, which contains hemoglobin in some species like the sea cucumber. The distal circulation is open and mixed with the fluid of the coelomic system, which fills the space between the body wall and the digestive tract, and is mainly involved in nutrient diffusion.

Cyclostomes, which are the most primitive fishes (see below Fishes) and the oldest extant vertebrates, have already modified the geometry of this primary heart: the inflow has left its caudal position and became dorsally placed, whereas the outflow stayed in a ventral position. Along the different classes of vertebrates, this heart will progressively alter its shape and become more complex (Figure C11). Different modifications take place [54,75].

- Looping of the heart structures with ventral displacement of the ventricular and outflow cavity, and progressive rotation of the inflow structures from a dorsal position to a cephalad position.
- Ballooning of the chambers, at the dorsal side for the atria and at the ventral side for the ventricles.
- Rotation of the heart from the antero-posterior axis into a left-right orientation; the original left lateral wall becomes the ventral wall.
- Transformation of the straight intracardiac flow into a "S" shape flow, and progressively into a "U" shape flow (Figure C12).
- Septation of the atrium in a right cavity including the sinus venosus and a left cavity receiving the veins from the lungs.
- Development of the right ventricle from the proximal part of the conus arteriosus, downstream the left ventricle, and trabecularization of this portion (Figure C13).
- Secondary septation of the ventricle starting at the interventricular groove and separating the left from the right chamber.
- Development of a high-pressure systemic left ventricle and a low-pressure, subpulmonary, trabeculated right ventricle.
- Disappearance of the sinus venosus and the conus arteriosus.

Figure C11. Progressive modification of the heart anatomy with evolution in vertebrates. From the cyclostomes (primitive fishes) to the amniotes (reptiles, birds and mammals), the inflow leaves its caudal position and comes in a dorsal and more frontal position, whereas the outflow stays in a ventral position ("looping"). The sinus venosus (SV) and bulbus arteriosus (BA) have less importance in air-breathing vertebrates and disappear progressively. Valves are present at the sinoatrial, atrio-ventricular, and ventriculo-bulbar junctions to prevent backflow. A: atrium. V: ventricle. Ao: aorta (adapted from Simoes-Costa 2005).

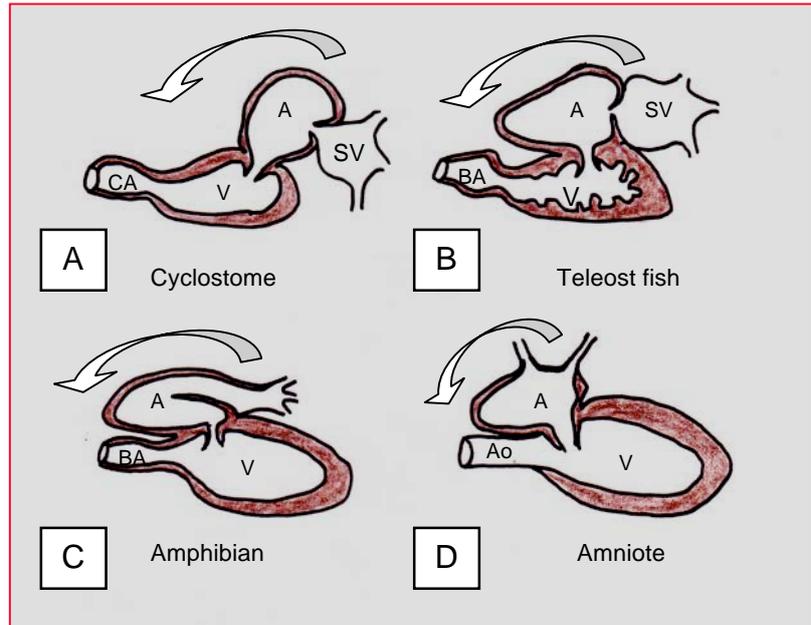
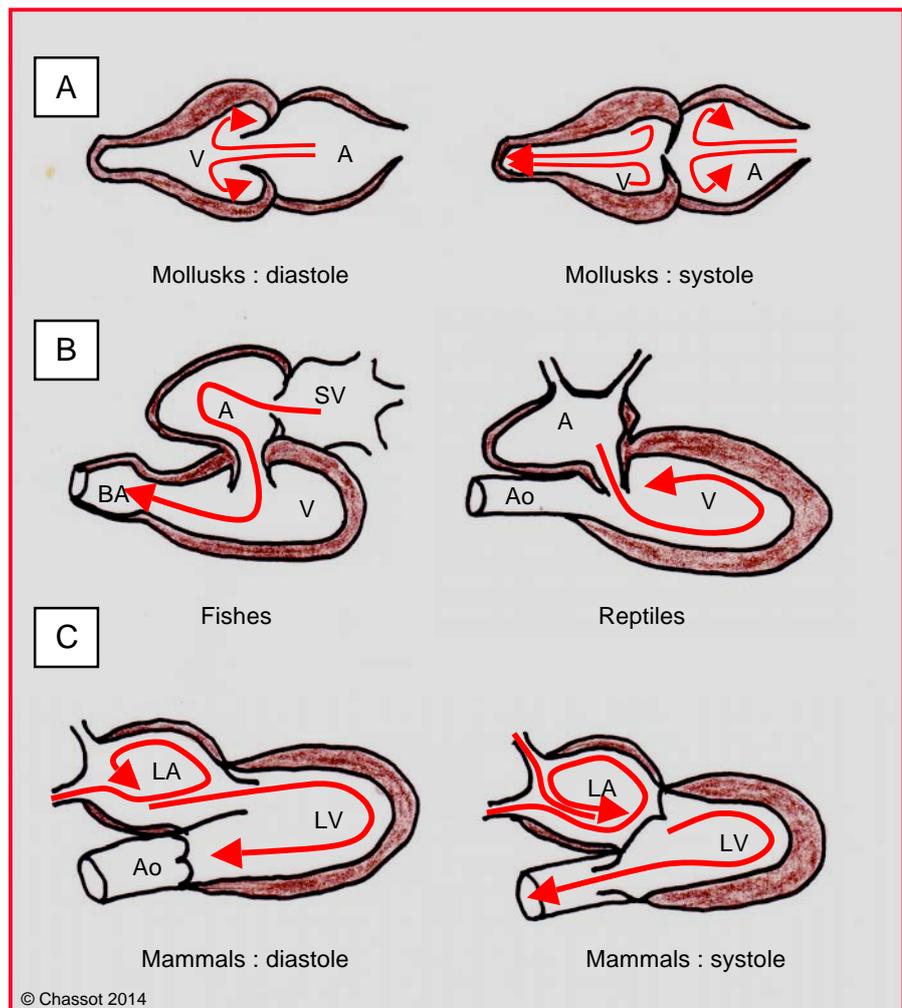


Figure C12. Modification of the intracardiac flow design with evolution. **A :** In mollusks, the flow is straight and creates turbulences which decrease efficiency. **B :** In fishes, the flow is « S »-shaped ; this configuration decrease the risk of turbulences. In reptiles, inflow and outflow are at the base of the ventricle; the flow is « U »-shaped, which provides better filling and ejection. **C :** In mammals, the flow is continuously whirling around inside atrium. In diastole, the flow into the ventricle makes a large vortex placing the blood volume in front of the outflow tract, ready to be ejected during systole. This system ensures the minimal loss in kinetic energy during the cardiac cycle. A : atrium. V : ventricle. SV : sinus venosus. BA : bulbus arteriosus. Ao : aorta.



The sinus venosus collects the venous return before the atrium. It is important for providing an adequate preload when the venous pressure is very low, like it is the case in fishes, amphibians and

most reptiles, which have a venous pressure of < 4 mmHg and a maximal arterial pressure of 40 mmHg. The conus arteriosus smoothes the systolic pulsatility. This is important for the fishes, because the gills are placed directly after the heart on the arterial system, and for the amphibians, where the pulmonary and the systemic circulations are issued from the same unique ventricle. These two structures have less importance in air-breathing vertebrates and have disappeared in birds and mammals because these animals have two completely separated circulations. Reptiles have two symmetrical aortic arches, one turning to the right and the other to the left. Higher vertebrates retain only one aortic root, the left in mammals and the right in birds.

All these changes in configuration resemble the progressive modification of the heart anatomy during the embryogenesis in human. According to Haeckel's law, "Ontogeny recapitulates phylogeny": during fetal life like during evolution, the primitive tube loops on itself, the primitive atrium moves dorsally and cephalad, the area between the primitive ventricle and the conus gives the right ventricle, the septation progresses and divides the two chambers into four chambers (Figure C13) [41]. This analogy is precious for comparing the different phyla. The comparison must be carried out on the embryos rather than on the adults, because specialized features have developed late in evolution, when the lineage was already detached from the common ancestors [54]. The fact that mammals share some parenthood with reptiles, for example, does not mean that they are linked to the present-day turtles and snakes, but that they did have some common precursor.

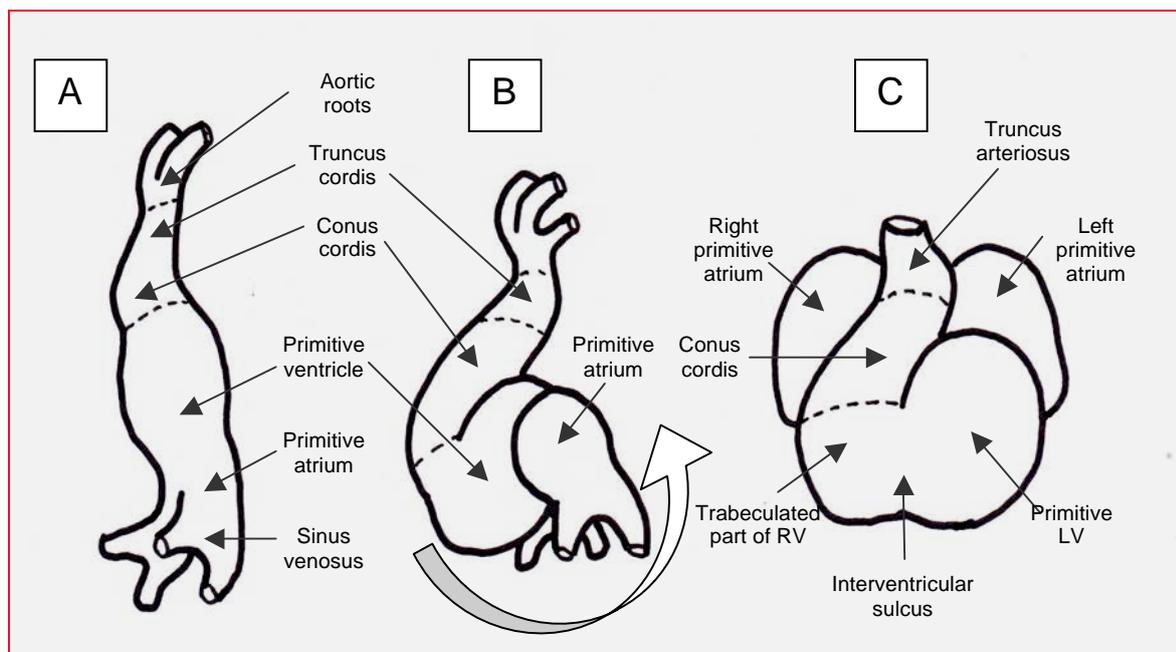


Figure C13. Progressive modification of the heart anatomy during the embryogenesis in human. **A** : At 3 weeks, the heart is a longitudinal tube ; the inflow (sinus venosus) is caudal. This stage corresponds to the primitive cardiac tube. **B** : A progressive rotation brings the primitive atrium posteriorly and cephalad. This stage corresponds to the heart of a fish. **C** : At 28 days, the right ventricle starts to appear. The progressive septation separates the primitive atrium in right and left chambers. The septation of the primitive ventricle starts at the interventricular sulcus. The spiral septation of the truncus cordis will give the right and left outflow tracts. This configuration is close to the three-chamber heart of amphibians and reptiles (adapted from Kussman 2001).

In vertebrates, the architecture of the myocardial tissue consists of two types of contractile fiber arrangements [69].

- The trabeculated, or non-compacted, myocardium consists of randomly distributed myocardial fibers forming large trabeculations at the endocardial surface. The arrangement is anarchic

except near the valvular orifices, where it is involved in the opening and closing mechanism of the leaflets. It has the spongy appearance of the non-compacted myocardium encountered in some human congenital heart diseases [84]. The O₂ supply is provided by diffusion from the ventricular cavity through the deep lacunae appearing in the intertrabecular spaces. This type of myocardium cannot generate significant blood pressure (≤ 25 mmHg) despite its good performance as a volume pump. It predominates in fishes with low levels of activity (flounder, icefish) and in amphibians; it represents less than 50% of the ventricular wall in reptiles, and has completely disappeared in birds and mammals.

- The compact myocardium has an orderly arrangement in well-organized fiber bundles able to develop high pressures. The subepicardial and subendocardial layers show mainly longitudinal fibers, whereas the innermost fibers have a more circular pattern. The compact myocardium is supplied by coronary vessels. This type of myocardium is visible on the 15-35% of the wall thickness of active fishes (mackerel, tuna), on most of the left ventricle in reptiles, and in the totality of the ventricles in birds and mammals.

Unfortunately, birds and mammals have partially lost the ability to produce new cardiomyocytes after birth and to regenerate dead myocardial cells, whereas mollusks, arthropods and amphibians have the possibility to renew 20-30% of their myocardium from undifferentiated precursor cells [44,48].

Intracardiac hemodynamics

Compared to the heart of invertebrates, the heart of the first fishes has already a more efficient hydraulic configuration (Figure C12). In snails, for example, the inflow and outflow are on a straight line. Therefore, the flow is interrupted during the alternating systole and diastole and eddies appear. This "stop-and-go" type of flow is a loss of energy; it reduces mechanical efficiency and does not allow a high heart rate. But snails are not particularly known for their frantic activity. On the contrary, the atrio-ventricular orifice of early fishes is almost at right angle with the direction of the inflow and of the outflow. This will remodel the flow configuration into a "S" shape, and establishes asymmetrical flow streams through winding circuits in atrium and ventricle. In four-chamber hearts of birds and mammals, the venous inflow contributes to a net rotational momentum inside the atria, with a tangential direction of the blood volume towards the atrio-ventricular valve. In diastole, the filling flow into the ventricle is directed dorso-posteriorly and makes a large vortex near the apex, pushing the blood volume ventro-anteriorly around the edge of the anterior leaflet of the mitral valve and towards the outflow tract. The inflow and the outflow are almost parallel ("U" shape flow), a configuration which provides the best filling of the ventricle and the most performing ejection by combining circumferential, longitudinal and rotational contractions [38]. This coherent and continuous swirling of blood preserves kinetic energy, maintains blood cells in motion, reduces dissipation of flow momentum, and redirects appropriately the volume for onward passage to the next cavity. The winding and reciprocal sling-like motion of blood and heart walls is particularly efficient at high heart rates, since it enhances the filling and the emptying of the ventricle [37]. Moreover, the torsion of the ventricle during systolic contraction imparts a rotational movement to the stroke volume in aorta, which reduces the frictional forces and improves the flowing of blood. The ability to increase cardiac output several-fold has been a determining factor in the success of the "fight-or-flight" behavior of higher vertebrates.

The stress of gravity

Buoyancy in water provides practical weightlessness, but standing on the ground means resisting to gravitational forces. The taller the animal, the more affected it is by gravity. This is a particular concern for snakes or mammals standing upright, for long-necked or long-legged birds and mammals, and for very large animals like the late dinosaurs. They all have to maintain the perfusion of the brain, which is positioned much higher than the level of the heart, and they have to keep a normal venous

return despite the lower position of the limbs. Animals have developed different adaptive measures against gravity [45].

- Increase in blood pressure. Whereas aquatic snakes have usually a blood pressure below 30 mmHg, terrestrial species have a pressure up to 80 mmHg; it is roughly proportional to the distance between the head and heart when standing upright [72]. The giraffe has a systolic pressure of 260 mmHg in order to keep a value of 100 mmHg in the brain [70]. A high blood pressure is obviously an advantage for changing postures, but it would impair gas exchange and increase dramatically transpulmonary filtration. Therefore, different systems have evolved to keep pulmonary arterial pressure lower than systemic pressure.
- Resistance to edema. With an upright position, blood pools in dependent vessels and peripheral edema supervenes. Birds and mammals have developed less compliant subcutaneous tissue and slender shape in lower legs in order to resist better to the distending forces. The contraction of muscles provides an active compression of the venous network.
- Baroreceptor reflexes. They quickly modify vascular resistances and heart rate according to the changes in position. Postural hypotension is a defect of these reflexes.
- Blood vessels adaptation. In mammals, vasoconstriction predominates in lower limbs whereas autoregulated vasodilatation predominates in the brain. Capillary basement membrane and medial smooth muscle are twice as thick in the legs compared to the neck in the giraffe [61].

The surrounding pressure does not seem to influence the physiology or the structures of the cardiovascular system. Cows living at the seaside or yacks living at 5'000 meters do not differ anatomically, but they show adaptation at the enzymatic and subcellular levels, particularly in the endothelium and in the alveolar epithelium. High-altitude mammals have an increased production of NO to prevent pulmonary hypertension, and a remodeling in the activity of the K^+_v and Ca^{2+} channels to prevent pulmonary edema [66]. Analogous biochemical transformations are found in deep-sea dwellers in order to adapt to the high water pressure.

Evolution of the respiratory system

Since circulation is intimately linked with respiration, a fast overview of the evolution of breathing among vertebrates may be appropriate. Aquatic species have to extract oxygen from water. But one liter of water contains only 7 mL of dissolved O_2 , whereas one liter of air contains 209 mL O_2 . Moreover, moving water in a gas exchange system requires much more energy than moving air in and out a respiratory tract. Compared to air-breathing animals, marine species have to hyperventilate in order to meet their O_2 requirements. Added to the higher diffusion of CO_2 in water than in air, this fact leads to a lower blood pCO_2 [14]. Therefore, breathing air is a prerequisite for full blood O_2 saturation, high metabolic rates, and intense physical activity. Different respiratory systems are in use among vertebrates.

- Fishes use gills for gas exchange. The vascular walls of gills are thicker than the capillary walls in the lungs, and gas exchange is not as efficient. Water is moved around the gill lamellae by the pumping effect of the mouth and the opercules, and by the swimming motion of the animal. In fast-swimming fishes like tunas or mackerels, the increased O_2 requirement is met by an increased area of the gills and by the active renewal of water around them with the displacement of the animal, which suffocates if it stops swimming. Between slow-moving and fast-swimming fishes, the dimension of the gills varies in a proportion of 1 to 50 [70].
- Amphibians and some sea snakes use moist skin respiration as a complement or substitute for lungs.
- Amphibians, reptiles and mammals have lungs with increasing degree of exchange surface. Ventilation is based on an in-and-out tidal displacement of air along a tracheal system into alveolae. The shorter the distance between blood and air, the better the gas exchange. The pressure in the pulmonary vascular tree must be low in order to have the thinnest possible

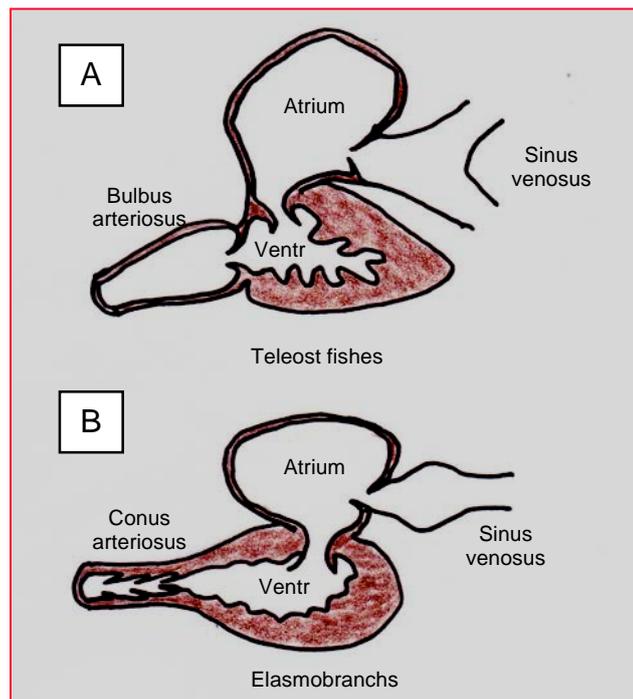
membrane between capillaries and alveolae, and the least possible exudate into the interstitial space.

- Birds have the most efficient system of gas exchange, since their O₂ extraction is 30-50% superior to mammals [13]. Their lungs consist of parallel tubes through which air flows in one direction only, intermingled with capillary vessels where blood is flowing in the opposite direction. This counter-current flow-through system is coupled at each extremity with air sacs functioning as buffering reservoirs between the alternating inspiration-expiration in the trachea and the continuous air flow in the lungs.

Fishes

Fishes have basically a single circulation, and the gills are situated between the two-chamber heart and the tissues (Figure C3-B). The heart itself, which receives venous blood, is supplied with oxygenated blood by an artery coming directly from the gill network, running in the middle of the ventral face of the ventricle like the left anterior descending artery in the human heart. A venous enlargement placed before the atrium (*sinus venosus*) provides a more continuous flow into the heart (Figure C14). A thickened elastic arterial cone at the root of the aorta (*bulbus* or *conus arteriosus*) buffers the systolic pulsatility of the flow ejected by the ventricle before entering the gills, which must resist systemic pressure. Unidirectional valves prevent backflow. On a functional basis, fishes can be grossly divided in four classes: cyclostomes, teleosts, elasmobranchs and lungfishes.

Figure C14. Fishes have a two-chamber heart (one atrium and one ventricle), preceded by the *sinus venosus* at the inflow and followed by the *bulbus* or *conus arteriosus* at the outflow. Unidirectional valves prevent any backflow at the atrio-ventricular and ventriculo-arterial levels. The bulbus is an elastic structure typical of the teleost fishes, whereas the conus is a partially fibrous and valved conduit found in elasmobranchs (sharks) (adapted from Schmidt-Nielsen 2002).



Cyclostomes (lamprey, hagfish) are jaw-less, primitive fishes resembling eels, appeared 450 million years ago. They have retained some of the characteristics of the chordate circulation, and differ from all other vertebrates. They have a partially open system, the blood flowing freely in large sinuses collected by veins. Their gills have striated muscles involved in the forward propulsion of arterial blood. In addition to their main 2-chamber heart, called *branchial* heart, cyclostomes have several accessory pumping chambers spread over the venous system: *cardinal* hearts helping venous return on the cardinal veins, *portal* heart pumping into the liver, and *caudal* heart near the tail. This caudal heart has a completely different design. It comprises two elongated parallel chambers around a central cartilaginous rod. The alternate contraction of two extrinsic muscles surrounding the chambers bows

the cartilage on one side or on the other; this alternate motion empties one chamber by compression and simultaneously fills the other by expansion (Figure C15) [70]. This system is unique to this prehistoric animal, and has obviously been abandoned by the natural selection because of poor performances.

Teleosts (tuna, salmon, haddock, and most fishes) have a muscular and elastic arterial cone stemming from the aortic wall (*bulbus arteriosus*) (Figure C14-A). The energy stored in the *bulbus* elastic wall during systole is released during diastole in order to maintain diastolic forward flow in the aorta. This elasticity decreases also the peak systolic pressure that the gills have to withstand. The closure of the ventriculo-bulbar bi-leaflet valve stops the flow in diastole like the aortic valve in mammalian heart.

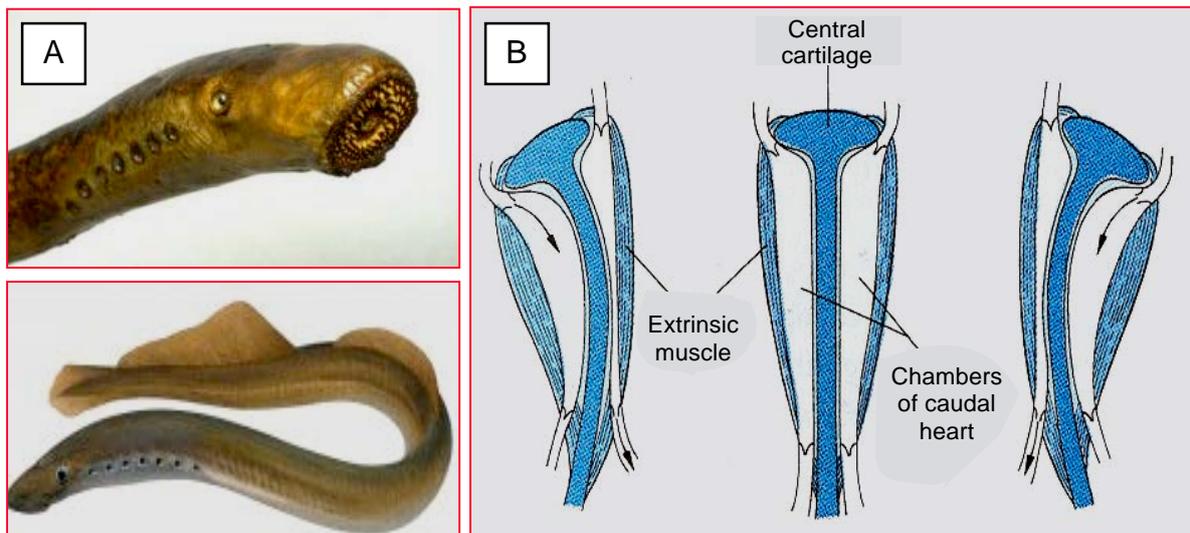


Figure C15. Cyclostomes. **A:** The hagfish (top picture) and the lamprey (bottom picture) look like eels, but they have a circular mouth made for sucking their preys instead of chewing them with jaws. **B:** The caudal heart of hagfish is centered on a cartilaginous rod bowing alternatively on each side by the contraction and relaxation of extrinsic muscles. The vascular chambers comprised between the rod and the muscle on each side are alternatively compressed or extended. This motion corresponds to a systole and a diastole, respectively (Reprint from Schmidt-Nielsen 2002).

Elasmobranchs (sharks, rays) and sturgeons have a slightly different heart. Their arterial cone (*conus arteriosus*) is developed from the heart muscle; it has a fibrous structure preventing its collapse, an actively contracting myocardium, and unidirectional valves preventing backflow. This is required because negative pressure can develop inside the rigid pericardium. Actually, sharks have a cartilaginous pericardium filled with fluid. During systole, the volume of the ventricle decreases and the pressure inside the pericardium becomes negative (- 1-3 mmHg). This phenomenon helps the filling of the atrium by a suction effect due to the decompression of the atrium with the displacement of the pericardial fluid around the ventricle (Figure C16). During diastole, the dilated ventricle pushes the fluid around the atrium. This compression assists the emptying of the atrium into the ventricle. Moreover, sharks can increase their stroke volume up to 40% by draining their pericardial fluid into the abdomen through a pericardioperitoneal canal, a remnant of the opening between pericardium and peritoneum appearing momentarily during embryogenesis in all vertebrates. This mechanism acts like the decompression of a tamponade [31]. Since cardiac sympathetic innervation does not exist in elasmobranchs, these fishes are dependent upon PPC-modulated pericardial volume change to rapidly increase stroke volume [26].

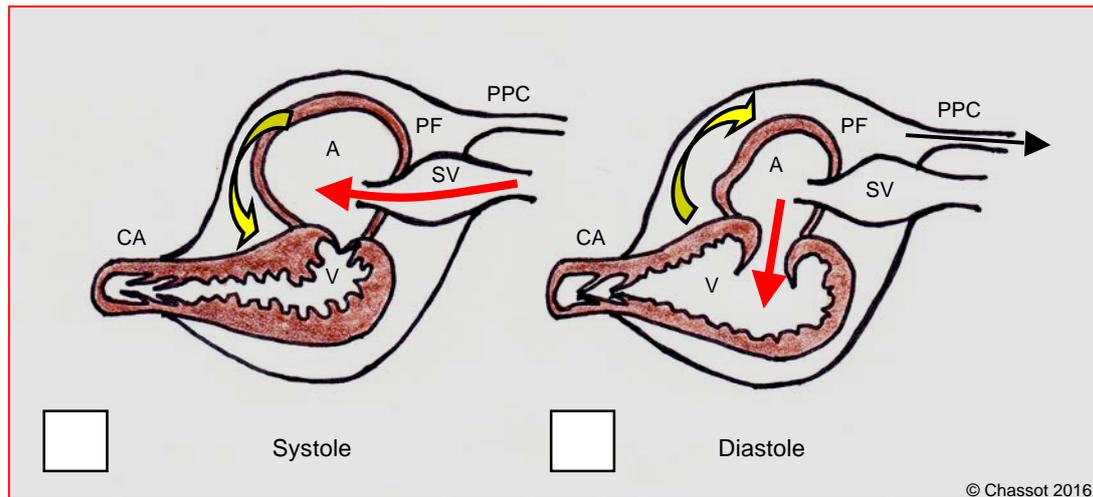


Figure C16. Elasmobranch fishes (sharks) have a cartilaginous pericardium filled with a clear fluid. **A:** In systole, the volume of the ventricle is reduced, the surrounding pressure becomes negative, the liquid is displaced and decompresses the atrium. This suction effect helps the filling of the atrium (red arrow). **B:** In diastole, the ventricle increases in volume and pushes the liquid around the atrium. This compression helps the ventricular filling. A: atrium. V: ventricle. SV: sinus venosus. CA: conus arteriosus. PF: pericardial fluid. PPC: pericardioperitoneal canal (black arrow: active drainage of pericardial fluid).

Lungfishes, or dipnoests, live in areas where long droughts may completely empty the lakes and stagnant ponds where they dwell. They developed two lungs, subdivided into numerous small air sacs, supplied by two pulmonary arteries branching on the vascular circuit after the anterior gill arches (Figure C17). Interestingly, lungfishes have a beginning of septation inside the atrium and the ventricle. Therefore, the oxygenated blood from the lungs arrives separately into the left part of the atrium, whereas the systemic vein returns into the right part of the same atrium. Blood flows with minimal mixing into the aorta, where the arterialized part goes preferentially to the first two, non-functional gills and then to the tissues, whereas the desaturated part goes to the posterior gills, where its CO_2 content decreases, and then to the lung, where its O_2 content increases [76]. The relative roles of lungs and gills vary according to the different species, since lungs are more efficient for O_2 transfer and gills for CO_2 removal. When breathing air, the arterioles to the gills become partially vasoconstricted and the pulmonary arteries completely open. Some species are already exclusive air-breathers. Since there is only one ventricle and one aorta, the blood pressure is the same in the systemic and in the pulmonary circulation.

Their relative predominance of trabeculated and compact myocardium varies among different species according to their more or less active behavior [82]. The trabeculated myocardium predominates in species with low levels of activity (flounder, icefish); the ventricle has a saccular shape and rather large end-diastolic dimensions. The compact myocardium, supplied by coronary vessels carrying oxygenated blood coming directly from the gills through the branchial arterial network, is typical of highly active, pelagic species (mackerel, tuna), where it represents up to 35% of the wall thickness. The ventricle has a pyramidal shape and an increased myocardial mass; it can develop much higher pressure (up to 80 mmHg in tuna) [82]. In sharks, the myocardium is always of the mixed type [69].

The heart of fishes displays a clear polarity of contraction in a posterior-to-anterior direction from sinus venosus to conus arteriosus. Pacemaker tissue is located in the inflow region and at the atrio-ventricular junction. The conduction velocity is fast in the atrium and ventricle, but it is much slower in the conus arteriosus, which acts as a valve preventing diastolic backflow into the ventricle [54]. The heart rate is proportional to the water temperature and inversely proportional to the animal size. At rest, it varies from 30 to 140 beats min^{-1} according to species. During exercise, the heart rate increases

only by 25%; the necessary increase in cardiac output is managed by a three-fold rise in stroke volume [40].

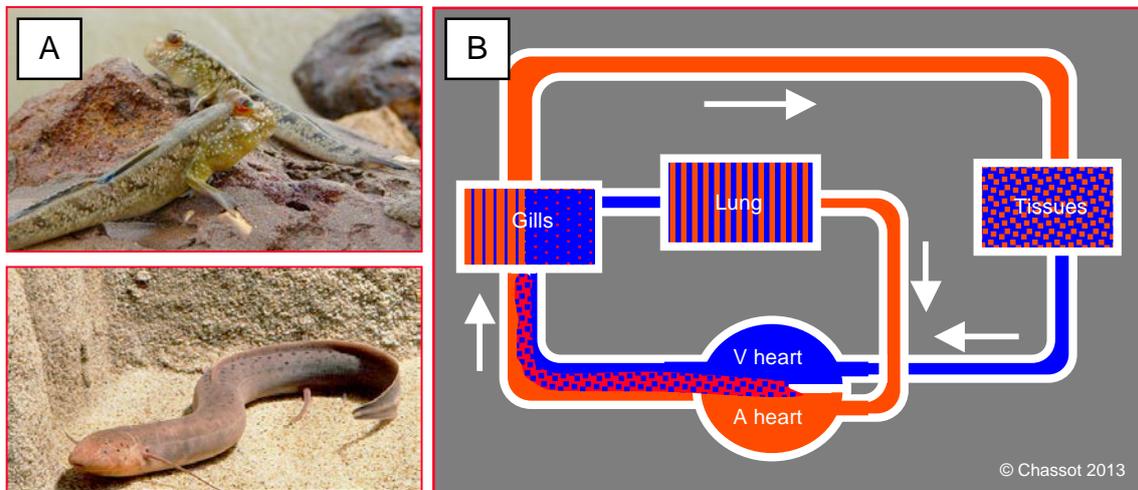


Figure C17. Lungfishes. **A:** Two different species of lungfishes breathing air outside water. **B:** Schematic illustration of the circulation in lungfishes. The partial septation of the atrium and ventricle allows some separation of flows inside the heart. Oxygenated blood from lung arrives in the left part of the atrium, and is ejected mainly in the first two, non-functional gills and then into distal aorta. Desaturated blood from tissues arrives in the right part of the atrium and is ejected mostly through the posterior gills and then to the lung. The relative role of gills and lungs in gas exchange varies according to species. V: venous. A: arterial.

Amphibians

After an evolutionary step with lungfishes, which have both gills and primitive lungs, amphibians (frogs, toads, salamanders) have abandoned gills and are exclusive air-breathers, but still on a bimodal pattern. The major part of the respiration takes place in the lungs, but a significant part of the gas exchange is performed through the moist skin. Amphibians have a three-chamber heart with two atria and one undivided ventricle, which myocardial structure is trabeculated and avascular (Figure C18). One atrium receives venous blood from the tissues and the other oxygenated blood from the lungs. The sinus venosus drains exclusively into the right atrium. The left atrium can be considered as a novel chamber, because it is a component of the pulmonary circulation [54]. In the unique ventricle, these two flows are parallel and practically do not mix, because there is a longitudinal, spiral baffle inside the conus arteriosus, which divides the flow in two laminar streams towards the two rows of semi-lunar valves [84]. The venous blood is ejected into the pulmonary artery and the oxygenated blood into the double aorta. The pulmonary artery gives branches to the skin ($\geq 25\%$ of pulmonary flow), where blood is also oxygenated (Figure C19). The blood returning from the lungs is highly saturated in oxygen (SaO_2 96%) like the blood returning from the skin, but the latter is mixed with the systemic venous blood.

During underwater diving, frogs develop an active vasoconstriction at the level of the artery supplying the lungs, and the pulmonary blood flow is diverted towards the skin, which becomes the major site of oxygen uptake [53]. Even if there is almost no pulmonary venous flow, mixed systemic venous blood returning to the heart has an increased O_2 saturation. This is sufficient to fulfill the low metabolic requirements of a frog hibernating in a cold swamp. The increased O_2 consumption in summer, when body temperature raises, is covered by additional uptake through the lungs. However, some toads and salamanders have no lungs and breathe exclusively through the skin. The heart rate of amphibians is rather low: the cardiac frequency of a frog is 30-50 beats min^{-1} . This average value is decreased by

hypothermia or diving, when the blood pO_2 is as low as 30 mmHg, and increased by the sympathetic stimulation of exercise or active ventilation, when the pO_2 increases to 100 mmHg [14].

Figure C18. Schematic illustration of the amphibian heart (frog, toad, salamander). The heart has three chambers: a right atrium, preceded by a sinus venosus, a left atrium, and one single ventricle. The alignment of the atrio-ventricular valves and the septation on the conus arteriosus are such that there is only a slight mixing of oxygenated and desaturated blood. The arterialized blood is propelled into the aorta and the desaturated blood into the pulmonary artery. However, both vessels have the same pressure. PA: pulmonary artery. Ao: aorta. SV: sinus venosus. CA: conus arteriosus.

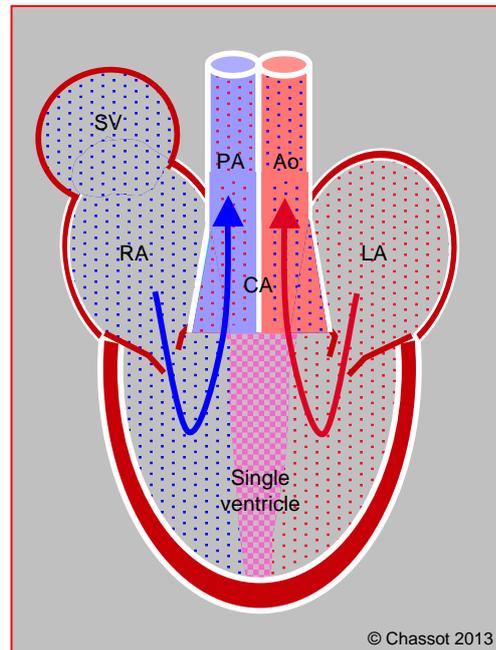
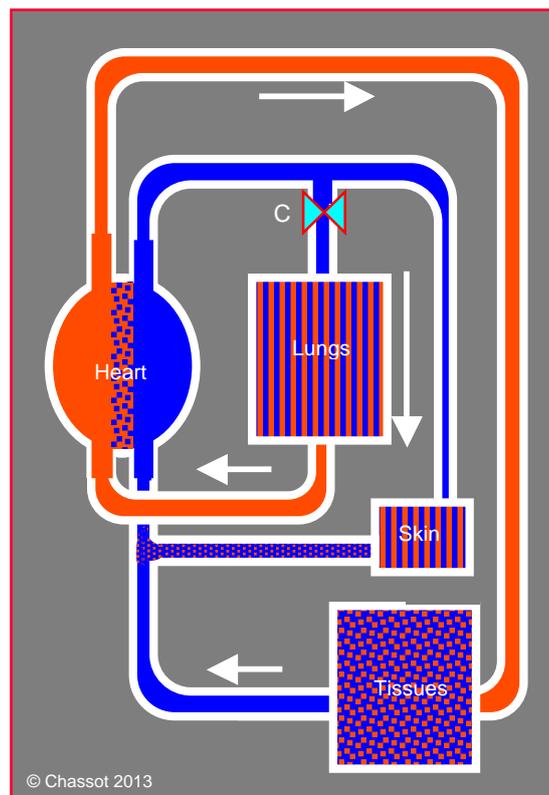


Figure C19. Schematic illustration of the amphibian circulation. The arterialized blood flows into the aorta, which supplies the organs and tissues. The blood comes back to the right atrium through systemic veins. The mixed venous blood is expelled into the pulmonary artery, which has a large branch for the cutaneous circulation, since skin participates for at least 25% to gas exchange. The saturated blood from the skin drains into the systemic veins and the right atrium, where the mixed venous has a relatively high SvO_2 . During diving, the pulmonary flow is reduced by vasoconstriction of the pulmonary artery (C) and the blood is preferentially diverted towards the skin.



Since there is only one ventricle, the blood pressure is identical in both pulmonary and systemic circulations. This is not a significant problem in amphibians for two reasons.

- The pressure of the circulatory system is rather low (systolic blood pressure 40 mmHg, diastolic 20-30 mmHg) ;
- The pulmonary artery sends branches to the skin, a circuit which has higher resistances than the pulmonary tree. It is therefore beneficial for the animal to have identical systemic and pulmonary pressures.

Reptiles

Reptiles represent a cardiovascular transition between the single circulation of fishes and the double circulation of birds and mammals. They are divided in non-crocodylians (snakes, turtles, lizards, varanids) and crocodylians (crocodiles). The former have a ventricle which is only partially divided into right and left cavities, whereas the latter have a true four-chamber heart with a complete separation between a larger left and a smaller right ventricle. Reptiles have one large pulmonary artery dividing in left and right branches, and two slightly smaller aortic arches, which bend dorsally and unite distally into a single aorta descendens supplying the systemic circulation. The brachiocephalic artery branches off the right aorta and supplies the head and the forelimbs. Like amphibians, reptiles have some mixing of venous and arterial bloods inside the heart. Functionally, the flows are more or less separated, because the partial septation of the ventricle directs the arterialized blood from the left atrium into the left and right aortic arches, whereas the desaturated mixed venous blood from the right atrium is directed into the pulmonary artery. The incomplete separation of the RV and the LV behaves like a ventricular septal defect (VSD) and has two major consequences.

- Variations in resistances of the pulmonary or systemic arterial tree modify the pulmonary-to-systemic ratio of the flows (Q_p/Q_s), and creates L-to-R or R-to-L shunts.
- With the possibility of exchanging blood between left and right ventricles, the stroke volume and the ventricular output on each side of the heart do not have to be equal.

At comparable body size, the O_2 consumption (VO_2) of the cool-blooded reptiles is 6-10 times lower than the VO_2 of the normothermic birds or mammals [71]. For diving animals, ectothermy is an advantage because it prolongs the duration of apnea. However, ectothermy and low metabolic rate imply to rely on anaerobiosis during intense activity. Well adapted to fight underwater, crocodiles can easily endure severe lactic acidosis; their blood lactate levels rise to 20 mmol L^{-1} during exercise, and even to 50 mmol L^{-1} in some species [5,8]. Varanid lizards, on the opposite, have a terrestrial lifestyle and sustain the highest metabolic rate of all reptiles during exercise. They rely essentially on aerobiosis because they have an almost complete separation of pulmonary and systemic circulations with non-significant shunting [87].

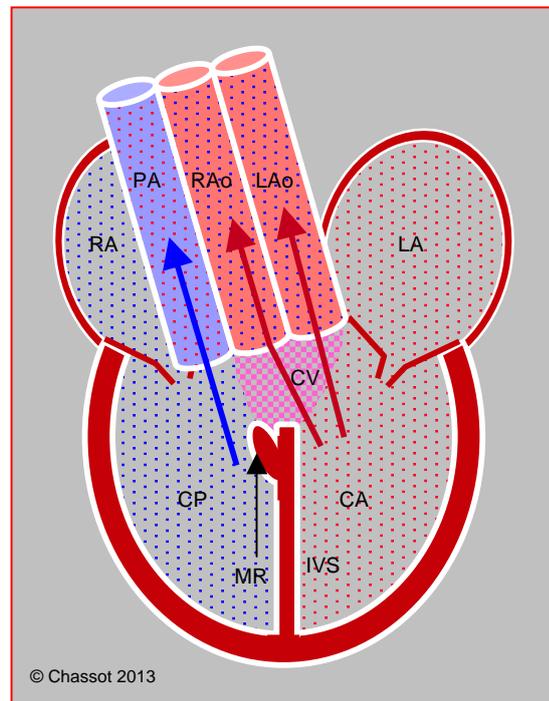
Reptiles deserve a detailed description of their different classes, because they show a wide spectrum of anatomical septation and shunting patterns between the right and the left side of the heart.

Turtles, non-varanid lizards, and most snakes have a three-chamber heart consisting of two atria and one ventricle (Figure C20). The ventricle is partially divided by a septum, and consists of three different parts: the *cavum pulmonale*, corresponding to a thin, ventrolateral, right ventricular chamber, the *cavum arteriosum*, corresponding to a thicker, dorsolateral, left ventricular chamber, and the *cavum venosum*, which is an intermediary chamber situated at the level of the interventricular communication (IC), under the outflow tract towards the two aortic arches.

The proximal extremity of the septum, close to the *cavum venosum*, is surmounted by a muscular ridge, which contraction in systole tends to decrease the size of the IC. Most of the ventricle is made of trabecular sponge-like myocardium, but the interventricular septum and part of the *cavum arteriosum* contain layers of compact, organized, pressure-generating myocardium. Compact myocardium is scarce in turtles and most snakes, but particularly developed in pythons and varanid lizards [35,84]. Despite the considerable separation between oxygenated and desaturated blood inside the heart, there is little or no pressure separation between the *cavum pulmonale* and the *cavum*

arteriosum in the majority of species. In turtles and in common snakes, the systemic blood pressure is 30-40 mmHg in average; the pulmonary arterial pressure is only slightly lower [19].

Figure C20. Schematic illustration of the reptilian heart. Non-crocodylians (common snakes, turtles, lizards) have a three-chamber heart consisting of two atria and one ventricle partially divided by an interventricular septum (IVS). The alignment of the septum and the flow inside the ventricles are such that oxygenated blood from the pulmonary veins is going towards the right and left aortic arches (RAo and LAo), and the systemic venous return towards the pulmonary artery (PA). The ventriculo-arterial valves are bicuspid. CP: cavum pulmonale. CA: cavum arteriosum. CV: cavum venosum with mixed blood. MR: muscular ridge.



Snakes and turtles are intermittent lung breathers. Brief periods of ventilation are interspersed among periods of apnea of variable duration, usually associated with diving. During apnea, the heart rate and the pulmonary flow (Q_p) decrease by 50-80%, with a 150% increase in pulmonary resistance. These phenomena are under cholinergic control. The result is a 50-90% R-to-L shunt [33]. During breathing, heart rate and Q_p increase two- and threefold, respectively. A net L-to-R shunt takes place. The magnitude of this shunting process can be very large in species like turtles, sea snakes and rattlesnakes, but is much reduced in other reptiles like pythons and lizards.

The anatomy is more subtly developed in pythons and varanid lizards. In these species, there is a higher degree of septation of the ventricle with an almost complete separation between left and right circulations. During diastole, the venous blood of the right atrium flows through the cavum venosum and enters the cavum pulmonale, whereas the oxygenated blood from the left atrium enters directly the thicker cavum arteriosum. There is no mixing of the blood through the IC, because the hanging septal leaflet of the left atrio-ventricular valve is occluding it (Figure C21).

In systole, the contraction of the prominent muscular ridge situated on the proximal interventricular septum occludes the IC and separates the flow into the pulmonary artery from the flow into the aortic arches [34,80]. The O_2 -poor blood of the cavum venosum is ejected first into the systemic circulation and causes a R-to-L shunt of about 10%, but the O_2 -rich blood left in the cavum venosum at the end of systole will flow back into the cavum pulmonale in diastole and increase the O_2 content of the pulmonary flow (L-to-R shunt) [34,85]. The cavum venosum is therefore a transit cavity for both systemic and pulmonary circulations. The shunt is non-significant and more likely to be a L-to-R than an R-to-L shunt, because the cavum pulmonale is weaker than the thicker cavum arteriosum [80]. The degree of shunting depends on the timing of the atrio-ventricular valves motion and on the contractile power of the muscular ridge. During rest, snakes have a low O_2 consumption and a significant bypass of the pulmonary circulation: the Q_p/Q_s ratio is about 0.5. When digesting (python) or exercising

(lizards), reptiles increase their O₂ consumption and increase the pulmonary flow ($Q_p/Q_s \geq 0.7$). The heart rate increases from 35 to 55 beats min⁻¹ [80].

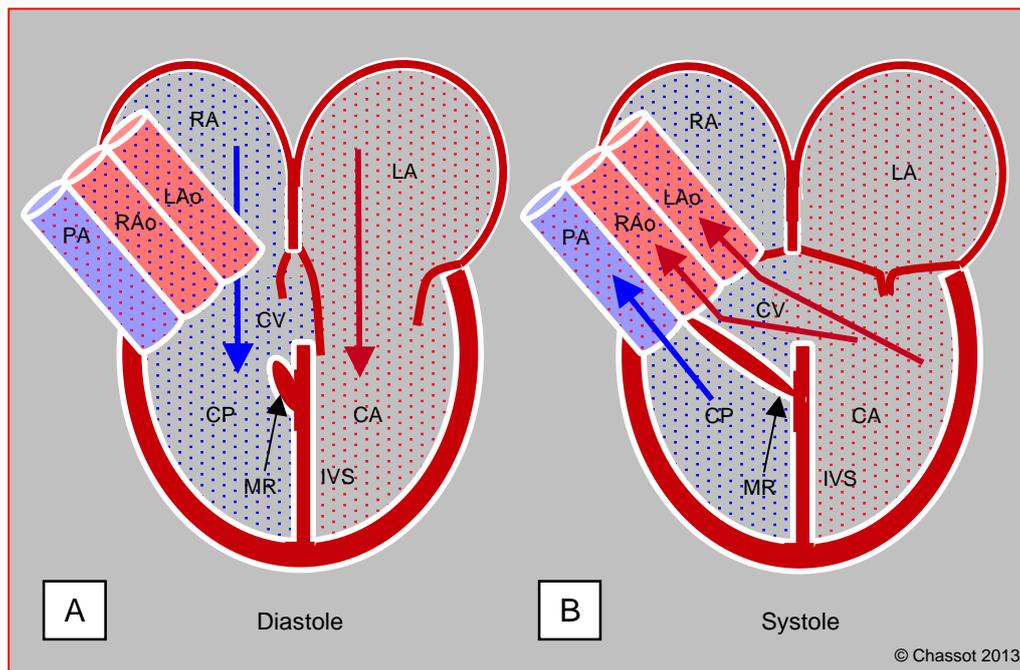


Figure C21. Schematic illustration of the intracardiac flows during a cardiac cycle in the python and varanid lizard. **A:** In diastole, the desaturated venous blood flows from right atrium (RA) into the cavum pulmonale (CP) through the cavum venosum (CV). The oxygenated blood from left atrium (LA) flows directly into the cavum arteriosum (CA). The long septal leaflet of the left atrio-ventricular valve occludes the interventricular communication. **B:** In systole, the contraction of the muscular ridge (MR) situated on the interventricular septum (IVS) separates the right ventricular cavity from the left. The cavum pulmonale ejects into the pulmonary artery and the cavum arteriosum into both aortic arches (RAo: right aortic arch, LAo left aortic arch). The pressure is higher on the left side than on the right side. The systemic ejection starts by pushing into the aortas the venous blood filling the cavum venosum. This represents an average 10% R-to-L shunt ($Q_p/Q_s = 0.9$). At the beginning of the next diastole, the oxygenated blood left in the cavum venosum will be washed into the cavum pulmonale. This increases the saturation of the venous blood and represents a small L-to-R shunt.

This dynamic separation of pulmonary and systemic flows during the cardiac cycle varies with heart contractility, preload and systemic versus pulmonary afterload. It is directly correlated with the increased systemic pressure encountered in pythons and varanid lizards: the systolic pressure is raised up to 80 mmHg in these reptiles, whereas the pressure in the pulmonary artery is 20 mmHg [2,86]. The cavum arteriosum of pythons, which accounts for 75% of the total ventricular mass, can eject against arterial resistance up to 95 mmHg, although the ventricular output decreases significantly, whereas the cavum pulmonale ceases to eject when the pulmonary pressure is above 60 mmHg [35,85]. With this high systemic pressure, pythons and varanid lizards need to separate systemic from pulmonary blood flows, otherwise they undergo a severe risk of pulmonary edema.

Crocodiles have a more complex anatomy and a more sophisticated physiology, because they are probably terrestrial animals secondarily returned to an aquatic lifestyle. Right and left atria open into right and left ventricles, which are totally separated from each other by a complete interventricular septum (IVS): the muscular IVS is prolonged by a membranous part inserted between the two aortic valves. The pulmonary artery exits from the RV, the right aortic arch (RAo) comes from the LV, and the left aortic arch (LAo) from the RV (Figure C22). The LAo supplies mainly the splanchnic organs and the RAo vascularizes the head, the tail and the four limbs. At their root, immediately after the bicuspid aortic valves, the aortic arches communicate with each other through an opening called

foramen of Panizza [58]. The systolic pressure developed by the RV is 20-30 mmHg, whereas the systolic pressure in the LV might reach 60 mmHg in some species. The heart rate of crocodiles is low and depends on the temperature; it is 15-20 beats min⁻¹ at 18°C and 25-40 beats min⁻¹ at 28°C.

During air-breathing through the lungs, the pressure in the RV of crocodiles is lower than the pressure in the LV, because the pulmonary arterial resistances are low. The higher-pressure LV pushes blood into the RAo and into the LAo through the foramen of Panizza. Since the pressure in the root of the LAo is above the pressure in the RV, the bileaflet aortic valve of the LAo stays closed and there is no mixing of blood. The situation changes completely when the crocodile is diving. Blood flow to the lungs decreases dramatically because of the closure of a cog-tooth valve, made of nodules of connective tissue resembling knuckles, situated in the right ventricular subpulmonary outflow tract. The pressure increases in the RV to the point that it ejects its mixed venous blood into the LAo, whereas RAo continues to receive mainly saturated blood from the LV. The result is a bypass of the lung circulation and a venous admixture in the systemic arterial tree. The presence of a complete septum precludes a L-to-R shunt from developing. But the arterial desaturation is asymmetrical and concerns essentially the liver, the stomach and the intestines. This shunting is controlled by the neurovegetative system. Diving reflex induces a parasympathetic stimulation and a cholinergic-mediated bradycardia. This situation is associated with a contraction of the cogwheel valve and a relaxation of the foramen of Panizza promoting a R-to-L pulmonary shunt. Adrenergic stimulation relaxes the cog-tooth valve and decreases the diameter of the foramen of Panizza; β -blockade has the opposite effect [25].

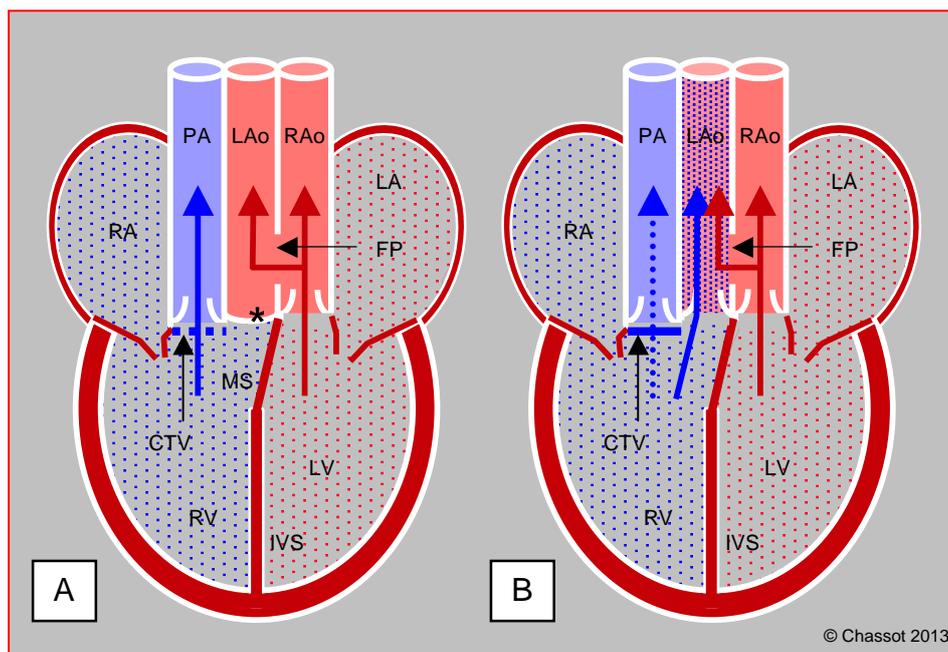


Figure C22. Schematic illustration of the crocodilian heart. The ventricles are completely separated by the muscular (IVS) and the membranous (MS) septa. The right aortic arch (RAo) is connected to the LV, but the left aortic arch (LAo) is connected to the RV. The RAo is turning rightwards and supplies the head, the tail and the four limbs. The LAo turns left and supplies the splanchnic organs. The foramen of Panizza (FP) is an opening between the roots of the RAo and the LAo, which allows the LAo to be supplied by oxygenated blood from the LV. The cog-tooth valve (CTV) is situated in the RV outflow tract and can modify the pulmonary flow. **A:** In air-breathing situation, the LV pressure is higher than the RV pressure. Therefore, the LAo valve stays closed (*) and the desaturated blood flows exclusively into the PA. The cog-tooth valve (CTV) is open. **B:** During underwater diving, the situation changes completely. The cog-tooth valve closes and interrupts almost totally the pulmonary flow. The pressure rises enough in the RV to equalize the pressure in the LV, and can therefore open the LAo valve. Mixed venous desaturated blood is flowing into LAo. CP: cavum pulmonale (right ventricle). CA: cavum arteriosum (left ventricle).

There have been many speculations on the potential adaptive value of the R-to-L shunting in reptiles and crocodilians, but its advantage remains elusive [32,33]. Perhaps some physiological processes linked to shunting may be beneficial, particularly for diving species.

- The decrease in arterial PO₂ tends to keep aerobic metabolic rate below its normal value. This downregulation may extend the duration of dives [71].
- The reduction in pulmonary blood flow decreases the amount of interstitial fluid filtered into alveolae, which has to be drained back by the lymphatic system [79].
- In crocodiles, the O₂ store in the lungs are larger than the store of the blood, in contrast to mammals. Reducing pulmonary flow shifts the burden of O₂ delivery to the blood and saves this larger reserve.
- The hypercarbia associated with lungs shunting increases the amount of peripheral CO₂ and the level of tissue acidity. This in turn will augment O₂ delivery (Bohr effect).
- The retention of CO₂ by the R-to-L shunt can be used by the gastric proton-pumps to increase the acid secretion during digestion, since reptiles swallow their preys without chewing them. This is particularly relevant for crocodiles eating very large meals [24].

The main advantage of this shunting capacity is most probably the possibility to vary the left and right ventricular outputs independently from each other. This kind of uncoupling gives a wide margin of safety when pulmonary or systemic vascular resistances vary in opposite direction, like it is the case during hypoxia, apnea and diving. These periods are characterized by a parasympathetic stimulation with bradycardia and increased pulmonary vascular resistance, leading to a net R-to-L shunt. Nevertheless, the shunting capacity might also be viewed as an ancestral heritage or an embryonic character that is useless but tolerable, because the hypoxia induced remains within the range tolerated by reptiles and has no negative impact on their performances [32].

Dinosaurs, the ancestors of crocodiles and birds, were the first large animals to stand upright on the ground and to adapt to significant gravitational stress. They had probably some kind of four-chamber heart, although the fossilized proofs for this assertion are very weak. They had certainly a high systolic blood pressure, since the head of some species was situated 6-8 m above ground. Comparatively, the giraffe has a systolic pressure of 260 mmHg in order to keep a value of 80-100 mmHg in the brain, which is 2 m above the level of the heart and 4 m above the ground. With these levels of systemic pressure, an independent, lower-pressure, pulmonary circulation was mandatory in order to facilitate gas exchange through thin-walled capillaries and to prevent pulmonary edema. A four-chamber heart with two separate circulations is the only possibility to fulfill this requirement [64]. The hemodynamics of a 40-tons walking animal is still a mystery. A fin whale of the same weight swimming on a horizontal plane has a blood pressure of 100 mmHg. Its left ventricle has a mass of 135 kg, a thickness of 10 cm and an end-diastolic diameter of 52 cm. With the same dimensions, a *Barosaurus* standing on the ground should have a blood pressure above 500 mmHg and a heart weighing 1-2 tons with a 50 cm-thick left ventricle. This seems unrealistic. Different hypotheses have been advanced to circumvent this problem: alternative pumping arrangements such as accessory cervical hearts, siphon effect on the venous side, position of the neck not erected but kept on a more or less horizontal plane, low metabolic rate due to a non-homeothermic temperature regulation. The best estimates of the average dinosaurs blood pressure is between 100 and 190 mmHg, and possibly up to 250 mmHg in some sauropods [73].

This rather long description of reptilian hemodynamic shows that a clear evolution takes place inside the phylum of reptiles, starting with a three-chamber heart, high degree of shunting and almost equally low systemic and pulmonary pressures, and ending with a four-chamber heart, very low degree of shunting, separated circulations and high systemic arterial pressure. This evolution corresponds to the move from an aquatic to a terrestrial lifestyle, from a plant-feeding to a predatory behavior, and from a low-speed locomotion to a high level of physical activity. Despite what seems a defect in the separation of systemic and pulmonary circulations leading to a relative hypoxemia, reptiles have

managed to build up a highly reliable hemodynamic system, which persisted without failure for 240 million years.

Birds and mammals

Birds and mammals are the only homeothermic animals: they maintain a constant body temperature around 37°C. A cold or a warm surrounding does not affect their activity. This is an evolutionary advantage, but at the price of an increased basal metabolic rate, which means a higher cardiac output and a higher blood pressure. They have a four-chamber heart, with the membranous part of the interventricular septum inserted between the aortic and the pulmonary valves. This is the best solution to keep two completely separated circulations with completely different blood pressures [17,76].

- A high-pressure systemic circulation responding to a high metabolic demand. The average blood pressure is 120/60 mmHg in mammals and 160-200/50-70 mmHg in birds; some avian species have a systolic pressure above 200 mmHg and a left ventricle with a typical concentric hypertrophy. This allows rapid transfer of oxygen and nutrients to the tissues and selective repartition of blood flow according to the needs (increased flow to the brain, the heart and the muscles during fighting or hunting, for example). The thick LV is sensitive to preload through the Frank-Starling mechanism, but maintains its output on a wide range of afterload.
- A low-pressure pulmonary circulation devoted to gas exchange: 25/15 mmHg in mammals and 20/10 mmHg in birds. Low blood pressure facilitates gas transfer, allows thinner capillary membranes, and reduces fluid filtration into air spaces. High pulmonary pressure would mean low oxygen transfer from air to blood, thicker membranes preventing easy gas exchange, and a constant threat of interstitial fluid leakage. The thinner RV is highly sensitive to afterload but can put up with large variations in preload.

The major hindrance here is the complete interdependence of both circulations. Left and right ventricles must beat synchronously and must have a strictly equal flow per minute. The average stroke volume and output must be the same, even if the preload of each ventricle is reciprocally modified by respiration. This requires a sophisticated system of hemodynamic regulation. On the other hand, heart and lungs should be close together because the pulmonary circulation has a pressure too low to accommodate long tubes. The loss of pressure which occurs along pipelines would not leave enough residual pressure in the venous system if the pulmonary tree were of the same size as the systemic vascular arborescence. The four-chamber heart and the ability to separate the low-pressure pulmonary circulation from the high-pressure systemic circulation evolved independently in birds and mammals. The phylum of mammals took off from early terrestrial vertebrates about 300 million years ago, whereas birds are descendants from early reptiles (Figures C1 and C10). It means that evolution followed parallel paths according to identical drives and responded to similar constraints with identical solutions.

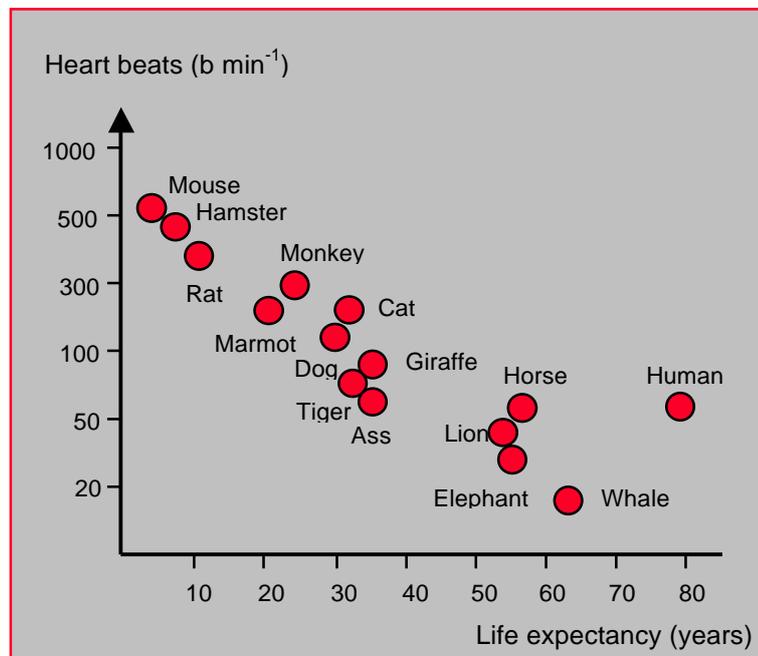
Birds have astonishing hemodynamic performances. Their stroke volume is identical to the stroke volume of mammals with the same body mass, but their cardiac output is six times higher because of a much higher heart rate at rest and during exercise: respectively 85 and 335 beats min^{-1} in the swan, 130 and 625 beats min^{-1} in the seagull, and 520 to 1260 beats min^{-1} in the hummingbird [16,46]. Larger birds have a lower heart rate, but their increase in frequency is 400% during exercise instead of 200% in smaller birds [46]. The high blood pressure of birds (systolic BP ≥ 200 mmHg) allows them to withstand high accelerations when they fly hunting preys or escaping enemies. They sustain easily 5-10 “g” without fainting. Birds are largely superior to mammals for their gas exchange capacity. Instead of a tidally-ventilated alveolar tree like mammals, they have highly-vascularized sponge-like lungs perforated with many parallel tubes through which air flows in one direction only. This counter-current flow-through system is coupled with air sacs functioning as buffering reservoirs. It permits an O_2 -extraction 50% higher than in mammalian lungs [62,63].

In mammals, the resting heart rate is also inversely proportional to the size: 600 beats min^{-1} for the shrew, 25 for the elephant and 6-12 for the whale. This is related to three phenomena [22].

- The maintenance of the diastolic pressure. A large ventricle requires a long diastole to fill properly, and the size of the ventricle increases with the size of the body, since the heart mass represents 0.6% of body weight in all mammals, man included.
- The velocity of the conduction system. The action potential would be far too slow for maintaining a high rate in large hearts.
- The O_2 consumption. Since surface varies to the square but volume to the third power, the body surface area increases relative to the body mass when the size decreases. In small animals, the heat loss is increased due to a relatively large surface area compared to body size. Maintaining homeothermy is more energy-consuming in small than in big animals, and requires higher cardiac output and heart rate, as it is the case in human neonates.

The life expectancy of mammals is inversely proportional to their resting heart rate, and the relationship is very tight (Figure C23). Large animals with slow rates live much longer than small animals with fast rates, but the average number of heart beats per lifetime is surprisingly constant, since it is $6\text{-}12 \cdot 10^8$ (average 1 billion), despite large variations in heart rates and longevities (Figure C24) [18,43]. In humans, the life expectancy of the population living 5000 years ago was around 40 years, as in the primitive tribes during the 20th century. This data fits with a total heartbeat count of 1.1 billion per lifetime. Nowadays, life expectancy is close to 80 years in industrialized countries with the improvements in medicine, sanitation and technology. This has doubled the total heartbeat count per lifetime [21]. The heart weight is 12 mg for a shrew (2-5 gm) and 600 kg for a blue whale (100 tons), but its ratio to the body weight is 0.6% in both animals as in all mammals. The stroke volume is 12 mL and 350 L for the shrew and the whale respectively, but the ratio of the SV to the body weight is of the same order of magnitude. Their total oxygen and ATP consumption per unit of weight during their lifespan is almost identical, since the basal O_2 consumption per heart mass is practically the same for all homeothermic animals [21].

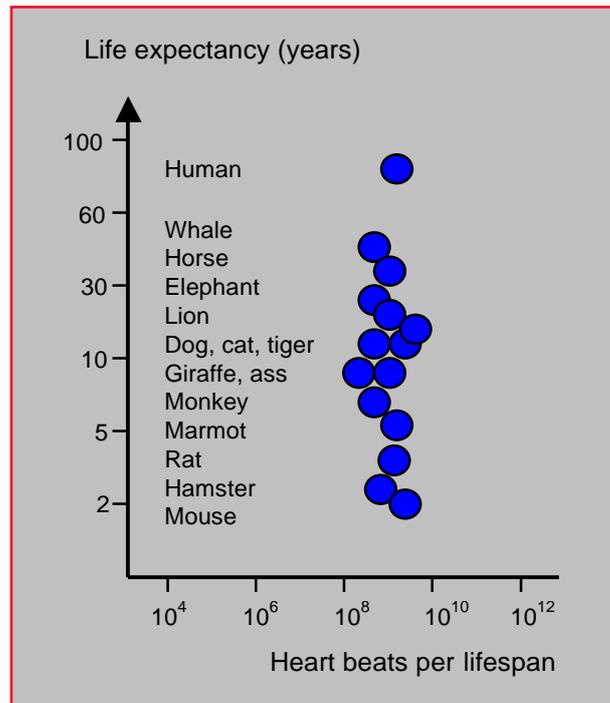
Figure C23. Relationship between resting heart rate and life expectancy in mammals. With a semilogarithmic scale for heart beats, the relation is inversely proportional and almost linear. Humans diverge mainly because they use artificial means for life prolongation, at least in industrialized countries (adapted from Cook 2006 & Levine 1997).



When they swim underwater, birds and mammals have the same diving reflex and parasympathetic response as other vertebrates: reduction of heart rate of 15-40%, even below $12 \text{ beats min}^{-1}$ during long apneas, pulmonary vasoconstriction, and reduced cardiac output. This diving response has an O_2

sparing effect, which prolongs the time duration before hypoxic damage [1]. In the blue whale, the diving heart rate is as low as 6 beats min^{-1} . In order to maintain an adequate blood pressure during the long diastole, the aortic arch is extremely compliant. It stores the stroke volume during ejection and releases it during diastole, providing a normal diastolic pressure [74]. Contrary to terrestrial mammals which main oxygen reserve is in the lungs, diving mammals use mainly blood as O_2 reserve for extending the duration of apnea. Their blood volume represents 200-260 mL kg^{-1} (instead of an average of 65 mL kg^{-1}); most of the oxygenated erythrocytes are stored in the spleen.

Figure C24. Relationship between life expectancy and total number of heart beats per lifespan (semilogarithmic scale). It is practically constant, with an average value of one billion ($10 \cdot 10^8$) (adapted from Cook 2006 & Levine 1997).



We tend to see animal evolution as leading to mammals, but mammals are far from being the kings of nature. Birds have much better lungs, dinosaurs were heavier than elephants, and insects have societies more organized than monkeys. Mammals don't even prevail by their number: they are just over 4'000 species, while there are 7'000 species of reptiles, 9'700 of birds, 45'000 of fishes, and about one million and a half of arthropods [42]. The largest part of biomass is made of bacteria, protozoa and cnidaria; they represent a volume around one thousand times the volume of the vertebrates. The single most important species in the world is the *krill* (> 800 million tons), a small shrimp which is part of the plankton in cold water. The success of higher mammals is probably due to their versatility and the intelligence of their brain more than to their physiological achievements. They are only a fragile epiphenomenon in the history of life.

The evolution of hemodynamics

Despite the large number of different circulatory systems adopted in the animal kingdom, some identical genes are crucial for the cardiac development from arthropods to mammals, although evolutionary lines of insects and vertebrates have diverged 700 million years ago. Homology, the inheritance of a character from a common ancestor, is typical of cardiomyocytes, which are derived from the coelomic epithelium. These contractile cells have basically the same structure in all types of hearts since the enigmatic Bilateria which have preceded the division between Protostoma and Deuterostoma (Figure C1) [9,75,91]. The primitive peristaltic heart evolved towards a chambered heart independently among protostoma in mollusks, and among deuterostoma in vertebrates. The heart-in-series of the arthropods (Figure C5) are initially designed as a single peristaltic pump. When challenged with the necessity of increasing blood pressure, insects created accessory pumping organs instead of modifying their central pump design, as did mollusks and chordates [91]. Therefore, there is a logical continuum between the primitive but widespread peristaltic heart and the sophisticated but uncommon multi-chambered heart.

Hemodynamics in animals show similar trends and convergent evolution, even within orders as diverse as arthropods, mollusks, reptiles and mammals. Cardiac output increases from $25 \text{ ml min}^{-1} \text{ kg}^{-1}$ in fishes to $75\text{-}100 \text{ ml min}^{-1} \text{ kg}^{-1}$ in mammals and $100\text{-}150 \text{ ml min}^{-1} \text{ kg}^{-1}$ in birds, proportionally to the gradual increase in metabolic demand [70]. This is accompanied by a steady increase of the systemic arterial pressure, because a high-pressure delivery system is required to sustain the augmenting metabolic needs and the higher peaks of physical activity. However, gas exchange in the respiratory organs are facilitated by a low blood pressure, because it allows the membranes between capillaries and alveolae to be thinner and because it improves the gradient for oxygen uptake. Colloidal osmotic pressure of the plasma varies with peak blood pressure in order to minimize the fluid leakage into the alveolae. The average values are 5 mmHg in frogs, 6 mmHg in turtles, 8 mmHg in fishes, 10 mmHg in crocodiles, 20-24 mmHg in mammals, and 25 mmHg in humans. Birds are an unexplained exception, since their osmotic pressure is only 8-10 mmHg, when their blood pressure is 150-200 mmHg [3].

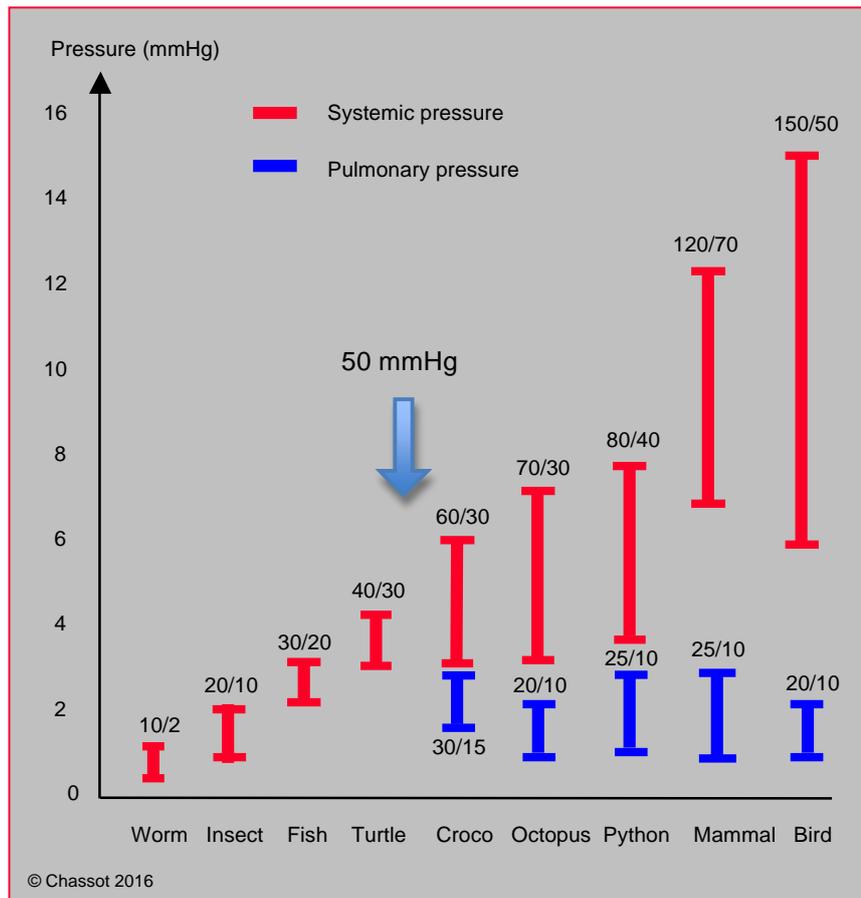
Therefore, we see the progressive differentiation between two separate circulations, one low-pressure system devoted to gas exchange and one high-pressure system required for tissue metabolism (Figure C25). This differentiation appears first in octopus and squids. It is most pronounced in birds: their average systolic pressure is 160-200 mmHg, while their pulmonary pressure is only 20/10 mmHg. From this point of view, the birds are the most accomplished animals [20].

Critical pressure beyond which pulmonary circulation loses efficiency appears to be between 40 and 50 mmHg. Indeed, animals with systemic pressure below 40 mmHg (arthropods, mollusks, fishes) do not have a differentiated pulmonary circuit. A separate low-pressure circulation for gas exchange appears when the systemic pressure exceeds 50 mmHg, as it is the case in cephalopods, some reptiles, mammals or birds. In humans, it is interesting to note that 50 mmHg is also the limit of pulmonary arterial pressure beyond which appear the serious clinical problems related to pulmonary hypertension. It is probably a physiological limit common to all animals.

The progressive augmentation of blood pressure raises also an unexpected question: what level of arterial hypertension is a disease, and what level is just the continuation of a natural process? An increase in arterial pressure, with its apparent decrease in longevity, might be the drawback of an increased physical activity and a full-blown cerebral efficiency. However, birds live longer than same-weight mammals despite their higher systemic blood pressure, probably because their mitochondria produce less free radicals [7]. Cardiovascular diseases seem evenly distributed among animals. Atherosclerosis is a genetic- and diet-dependent wearing process of arterial tree present in all mammals, but found also in certain species of birds (prevalence 8-75% in parrots), reptiles, and even active fishes with higher blood pressure like tuna [6,83]. Heart failure has been described in reptiles, birds and mammals (prevalence around 1%) [30]. Unfortunately, the data on the incidence of cardiovascular diseases among wild animals are scarce. Most data come from animals living in

captivity, where the situation is biased by stress, sedentarity and abnormal feeding, or from animal models of human disease used in research, where the situation has been experimentally manipulated.

Figure C25. Evolution of blood pressure. The increase in metabolic requirements and the stress of gravity for animals walking on earth have led to a progressive raise in systemic arterial pressure. Since a high blood pressure is not compatible with adequate gas exchange, the flow to gills or lungs have been maintained at a lower pressure within the same circulation or in parallel independent circulations. The critical pressure for pulmonary circulation seems situated around 50 mmHg. Animals with systemic pressure < 40 mmHg (arthropods, amphibians, reptiles) do not possess separate pulmonary circuits, but animals with systemic pressure > 50 mmHg (cephalopods, birds, mammals) have another low-pressure system for gas exchange.



Coronary arteries appear in the heart when ventricular pressure and oxygen consumption start to increase in the compact myocardium. In fishes, reptiles and sauropods, for example, the trabeculated myocardium is perfused by transparietal channels originating in the ventricular cavity. The flow inside these channels is provided by the compression and relaxation of the surrounding myocardial fibers. The progressive increase in the proportion of compact myocardium leads to the development of a coronary tree. In birds and mammals, where the whole myocardium is compacted, the coronary network is fully developed. The coronary tree of birds is mostly embedded inside the myocardium, whereas the large arteries are mostly epicardial in mammals. Speed running mammals and good flyer birds have a highly interlinked coronary network.

A pulsatile ventricle is the most commonly used pumping system among animals, because it affords high output under high pressure and can tolerate high vascular resistance. Nevertheless, pulsatility does not seem to be a requirement for the perfusion of peripheral organs, as proven by the excellent long-term survival rate and the low morbidity of patients receiving a continuous-flow, non-pulsatile ventricular assist device accompanied by a long-lasting anticoagulation [77]. Pulsatility certainly prevents vascular thromboses and improves lymphatic drainage [4], but its main significance is at the level of the heart: the pump must perfuse itself. A period of low pressure inside the ventricular wall is an absolute requirement for an adequate coronary flow bringing oxygen and nutrients to the myocardial cells. Without a diastole, the intraparietal pressure would be always slightly higher than the aortic pressure and no myocardial perfusion could occur. The system would work only in a very low-pressure regime and in very small ventricles, where wall perfusion could be provided by simple

diffusion from the ventricular cavity. However, a pulsatile system is flawed by two major hindrances for the circulation, which have been astutely resolved during evolution.

- At the preload level, the switch from a continuous venous return to a cyclic arterial ejection. This has been solved by the presence of an atrium and a sinus venosus acting as a buffering reservoirs.
- At the afterload level, the high pulse pressure in the peripheral circulation (pulse pressure = systolic pressure minus diastolic pressure). The elasticity of the great arteries serves to smooth out the systolic impulse, and the high-resistance arterioles decrease the distal pulsatility, in order to obtain a capillary flow, which is as continuous as possible. In humans, an elevated pulse pressure is an independent predictor of cardiovascular complications and of a reduced life span [56]. It seems that arterial tree has evolved in a way to absorb systolo-diastolic pulsatility, as it was increasing with the progressive elevation of blood pressure. Primitive animals with low blood pressure like snails have already a mechanism to decrease arterial pulsatility: they possess an ampula, which is an elastic sac connected to the aorta close to the ventricle, and devoted to buffer the raise in pressure during ventricular contraction. Octopus, which have a much higher pulse pressure, have developed elastin-like proteins in the walls of their large arteries in order to increase their pressure-absorbing capacity. Birds and mammals have a high elastin content in the wall of aorta and large arteries [91].

Therefore, the degree of pulsatile stress is neatly controlled. By combining the elasticity in aorta and the high resistance in small arteries, the flow into the capillary network is practically continuous.

Conclusions

Animal evolution does not resemble a long linear river flowing slowly from amoeba to primates, leading irreversibly to humans. It looks more like a huge and thick bush with a few diverging branches giving sometimes nice flowers. Despite their disparities, all branches seem to aim at more complexity and more activity, driving constantly towards new solutions to the challenges of the environment. They seem to evolve in response to the same constraints and to find strangely similar answers, a phenomenon called *convergence* (Figures C1 and C10) [91]. Numerous anatomical structures performing analogous function have been independently created by convergence, like the eyes of insects, cephalopods and vertebrates, or the wings of insects, birds and bats. In each phylum, the progression continues towards the same goals of increased metabolism, increased physical activity, and independence from the surroundings. Once separated from the main trunk, even the primitive classes evolve independently towards highly sophisticated animals. Advanced worms have a central nervous system, relatively high blood pressure and an almost closed circulatory system with capillaries. Octopus have a fine neurohumoral control of circulation, and show complex, clever and emotional behavior. Birds have the most efficient gas exchange system ever built and are the top animals for oxygen transport. Even if the solutions are different, they all tend to improve the performance of the cardio-circulatory system. "The evolution of most major groups is not the story of ecological variations..., but a history of mechanical improvement" [27]. Each type of heart seems the best fit for the needs of each phylum. The top performer is probably the bird's heart, but human heart appears well-adapted for a medium-sized creature, which main achievement is its brain.

This drive towards complexity does not imply an idea of progress, an organized plan or a directing intelligence. Random modifications and natural selection may lead to more sophisticated animals from the basic model of protozoa, because the evolution towards less complexity is not conceivable: it is not possible to be simpler than the simplest animal [27]. On the other hand, complex beings are an exception among the huge amount of bacteria, metazoa and cnidaria, which are many thousand times more numerous than vertebrates. Simple forms of life are surviving without modifications since a billion years, and still represent the vast majority of the living systems on earth. Sophisticated beings are more an epiphenomenon than an achievement. A vast proportion of the organic world evolved in directions which do not lead towards human beings [67]. Moreover, mankind can be considered more as a failure than a success of evolution, since it is the only species ever appeared on earth which is able to be threatened, and maybe destroy, the whole living system.

However, complexity and specialization might decrease robustness and flexibility. To be well adapted to a peculiar niche is also a dead-end when climatic conditions are modified, because most of the evolutionary potential is lost. After each mass extinction which happened during the history of life on earth, the evolution restarted with rather primitive beings, not with the survivors of the most advanced species. Early mammals were insignificant mice when the dinosaurs disappeared with the catastrophe due to a meteorite fall and a freezing winter lasting for years. If normothermia was a survival advantage under cold conditions, it is also a drastic constraint to refuel constantly the organism with oxygen and food [67]. The best strategy is therefore an adequate balance between sophistication and resistance.

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