

Circulation and Respiration in Lungfishes (Dipnoi)

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ABSTRACT This paper reviews the cardiorespiratory morphology and physiology of the living lungfishes, in the special context of their highly effective use of both air and water for gas exchange. Particular emphasis is placed on describing those features of the circulatory and respiratory systems that distinguish *Neoceratodus* from the Lepidosirenidae (*Protopterus*, *Lepidosiren*), and which, in turn, distinguish lungfishes from other aquatic vertebrates. Morphological and physiological characters that represent the plesiomorphic condition for the living Dipnoi are indicated (e.g., separate atrial chambers, vertical septum in ventricle, pulmonary veins, conal valves, twisting of bulbus cordis), as are those characters that may be shared derived features of the Lepidosirenidae (e.g., paired lungs, reduced anterior gill arches, well-developed spiral valve in conus). Morphological and physiological comparisons and contrasts with tetrapods are made to elucidate systematic relationships of the Dipnoi with other vertebrates.

The appearance of aerial respiration in ancestral fishes was a pivotal development in the evolution of terrestrial vertebrates (Johansen, '70; Gans, '70a; Johansen and Burggren, '80; Randall et al., '81; Little, '83). Since extremely little information on this respiratory transformation can be gleaned from the fossil record, physiologists interested in the evolution of aerial respiration must be content with the study of extant animals occupying a transitional position between an aquatic and terrestrial lifestyle. While these animals represent but a poor pseudophyletic progression (cf. Gans, '70a), investigation of the morphological and physiological adaptations for air breathing shown by these animals has, nonetheless, contributed greatly to our perception of how aerial respiration must have arisen in ancestors of the tetrapods.

The purpose of this review is to describe the cardiorespiratory adaptations for aerial respiration of extant lungfishes and to place these adaptations in the context of other vertebrates. Although the Dipnoi are a relict group only remotely related to other air-breathing fishes and separate from the amphibians by hundred of millions of years of independent evolution (Forey, this volume), we contend that ancestors of the tetrapods were probably very similar *physiologically* to the modern lungfishes.

AIR BREATHING IN FISHES—DIVERSE "SOLUTIONS" TO A COMMON PROBLEM

The unique morphological and physiological features for aerial respiration of lungfish are best appreciated when compared with those of other living fishes that breathe air. Aerial respiration has arisen independently numerous times in the evolutionary history of fishes, frequently (but not always) as a response to aquatic hypoxia (Bertin, '58; Gans, '70a; Munshi, '76; Graham, '76; Randall et al., '81); yet it is not a common evolutionary response to this environmental stress, since fewer than 70 of the more than 4000 genera of living fishes contain species that can actually breathe air (see Gans, '70b). While the vast majority of air-breathing fishes are tropical freshwater teleosts (see Gans, '70a; Dehadrai and Tripathi, '76), aerial gas exchange is remarkably prevalent amongst phyletically ancient gnathostome fishes (Burggren et al., '86), perhaps attesting to the value for survival of this innovative respiratory process. Whatever their evolutionary history, however, air-breathing fishes use a diverse range of structures to exchange respiratory gases with air. Organs

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for air breathing (arising both *de novo* and as adaptations of existing structures serving other functions) include the skin, the alimentary canal, the gills and structures derived from them, and diverticula of the branchial and buccal chambers, pharynx, and esophagus (for reviews, see Johansen; '70; Munshi, '76; Randall et al., '81). Virtually all air-breathing fishes still retain gills and gas-permeable skin functional in aquatic gas exchange. Thus, many air-breathing fish are so-called "facultative" air breathers, able to subsist entirely on aquatic gas exchange, but using the air-breathing organ (ABO) to facilitate respiration. Other fish are regarded as "obligatory" air breathers that will eventually drown if denied access to air. However, increasing metabolic rate or decreasing water O₂ availability, for example, can make air breathing obligatory for survival in fishes that under different conditions would be only facultative air breathers (Rahn et al., '71; Johansen et al., '70; McMahon and Burggren, '86).

In spite of considerable morphological and physiological diversity of air-breathing organs in fishes, these structural adaptations for air breathing are conceptually rather simple and not particularly effective by the standards of gas-exchange organs of tetrapods. The "effectiveness" of a particular respiratory system such as a gas bladder or lung depends greatly upon the source (and therefore gas content) of blood perfusing it and the consequent fate of blood draining it. In crocodilians, birds, and mammals, for example, the lungs are perfused exclusively by systemic venous blood that is relatively deoxygenated and high in carbon dioxide. In almost all nondipnoan fishes that breathe air, however, the blood supply to the ABO consists only partially of deoxygenated blood draining from the systemic tissues, with the remainder (and sometimes the major portion) consisting of blood that has most recently passed through a gas-exchange organ rather than the systemic tissues (Fig. 1). This situation can arise either when the ABO is downstream and in series with the gills (Fig. 1D,E) or in parallel with the gills (Fig. 1B,C). In either situation, some proportion of the circulation of blood to the ABO is "wasted," because little additional respiratory gas transport can be achieved by that proportion of oxygenated blood that has been shunted directly back into the afferent circulation to the ABO.

Why does the ABO (and for that matter the gills) of many air-breathing fish (e.g., Fig. 1B,D,E,F) receive blood that has already been partially oxygenated? In essence, the venous circulation of the ABO of most fishes is arranged just as that of any other systemic vascular bed, with the anatomical confluence of the efferent vessel from the air-breathing organ and the central systemic veins occurring well before the heart. This leads to substantial, if not complete, admixture of oxygenated blood from the ABO with deoxygenated blood from the systemic vascular beds on the venous side *before* the heart in almost all nondipnoan fishes.

As a direct consequence of the heart receiving a homogeneous venous input, in most air-breathing fishes there has been little selection for specializations leading to separate blood streams within the atrium and ventricle. Only with the evolution of an ABO with completely separate venous drainage directly to the atrium will any subsequent adaptation involving division of the heart or central arterial circulation confer any selective advantage to respiration. When preferential distribution of oxygenated blood to the tissues and deoxygenated blood to the ABO is facilitated by central cardiovascular division, gas exchange becomes more effective and can more readily be matched to changing internal and external conditions (see reviews in Johansen and Burggren, '85).

In considering the general phylogenetic progression in living vertebrates, there is a clear *physiological* progression toward a divided circulation with a separate, independently perfused circuit for gas exchange (Johansen, '85). The value is recognizing such a physiological progression, albeit in phylogenetically distant vertebrate lineages, is that it is strongly suggestive of the physiological transformations that we believe must have occurred in the actual ancestors of particular vertebrate lines as they evolved into terrestrial, air-breathing animals. In this context, the lungfishes are of vital importance, for they display morphological and physiological characters for respiration and circulation that were almost surely shared by the ancestral tetrapods going onto land.

The pulmonary circulation and the lung itself are crucial components of cardiorespiratory design in which the lungfishes appear both highly derived compared with other fishes and more closely affiliated with characters found in modern tetrapods. The follow-

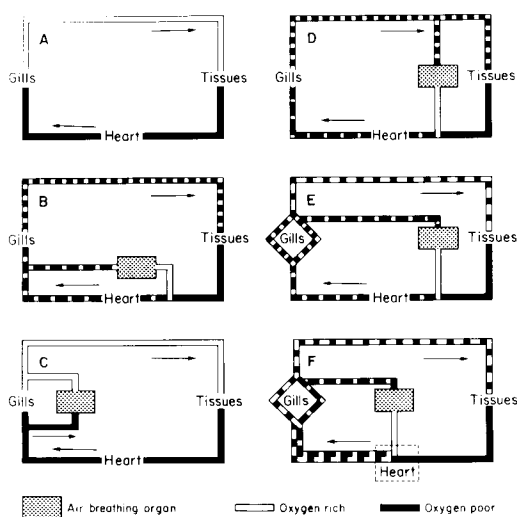
The pulmonary venous circulation

Fig. 1. Schematic representation of the flow of oxygenated (white) and deoxygenated (black) blood through the circulation of various air-breathing fishes. A. General arrangement of typical strictly aquatic fish breathing with gills. B. Air-breathing organ (ABO) derived from pharyngeal or opercular mucosa—e.g., *Monopterus*, *Ophiocephalus*, *Electrophorus*, *Amphipnous*, *Periophthalmus*, *Anabas*. C. Gills, buccal mucosa, or chambers extending from the opercular cavity serving as ABO—e.g., *Clarias*, *Saccobranchnus*. D. Intestinal tract used as ABO—e.g., *Hoplosternum*, *Plecostomus*, *Ancistrus*, *Misgurnus*. E. Air bladder serving as ABO—e.g., *Polypterus*, *Calamoichthys*, *Amia*, *Lepisosteus*. F. A lung homologous to that of tetrapods serving as ABO—e.g., *Neoceratodus*, *Lepidosiren*, *Protopterus*. (From Johansen, '70.)

ing discussion deals with both the morphology and physiology of gas exchange in the Dipnoi.

THE CARDIORESPIRATORY INNOVATIONS OF THE LUNGFISHES

With respect to circulatory and respiratory characters, *Neoceratodus* appears less highly derived from the plesiomorphic condition for lungfishes than either *Lepidosiren* or *Protopterus*. The Lepidosirenidae have many characters in common with tetrapods, in most instances likely representing convergence. This relationship appears to hold not only for morphological characters but also for what we presume to be the much more evolutionarily labile physiological processes.

The site of entry of oxygenated blood from the ABO into the central venous circulation is of crucial importance, for it portends further cardiovascular specialization. In *Neoceratodus*, the pulmonary vein from the single lung joins the systemic venous circulation prior to the heart at the level of the sinus venosus (Fox, '65) or left ductus Cuvieri (Satchell, '76). This shows a clear tendency for a more central termination of the pulmonary vein than of the veins draining the ABO of any phylogenetically ancient fish or teleost. Interestingly, a very similar arrangement of venous drainage from the gas bladder occurs in *Amia*, while in *Polypterus* and *Calamoichthys* the pulmonary vein enters the systemic circulation slightly more distally at the level of the hepatic vein (Purser, '26).

The greatest degree of separation of venous blood from the ABO and systemic blood from the tissues in any living fish is evident in *Protopterus* and *Lepidosiren*. In these genera the paired pulmonary veins draining the lungs fuse to form a single pulmonary vein lying dorsal and slightly lateral to the large vena cava. After passing anteriorly in the dorsal wall of the sinus venosus, the pulmonary vein empties *directly* into the left side of the atrium, rather than into the central systemic veins as in other fishes. Consequently, in *Lepidosiren* and *Protopterus*, oxygenated pulmonary blood and deoxygenated systemic blood are kept completely separate during passage to the heart. Thus, in comparison with *Neoceratodus* and other fishes, the central venous circulation of *Protopterus* and *Neoceratodus* much more closely resembles the condition in all tetrapods, where the pulmonary vein drains directly into a distinct left atrium.

Although beyond the scope of this paper, it is interesting to note that the systemic veins of the Dipnoi also show characters absent in other fishes (except *Latimeria*—Rosen et al., '81) but present in tetrapods, most notable being the prominent posterior vena cava and large sinus venosus (Goodrich, '30).

The heart and pericardium

Detailed accounts of the cardiac morphology of the Dipnoi have been given by Lankester (1878), Spencer (1898), Robertson ('13), Goodrich ('30), Bugge ('61), and Johansen et al. ('68). Generally, the hearts of living lung-

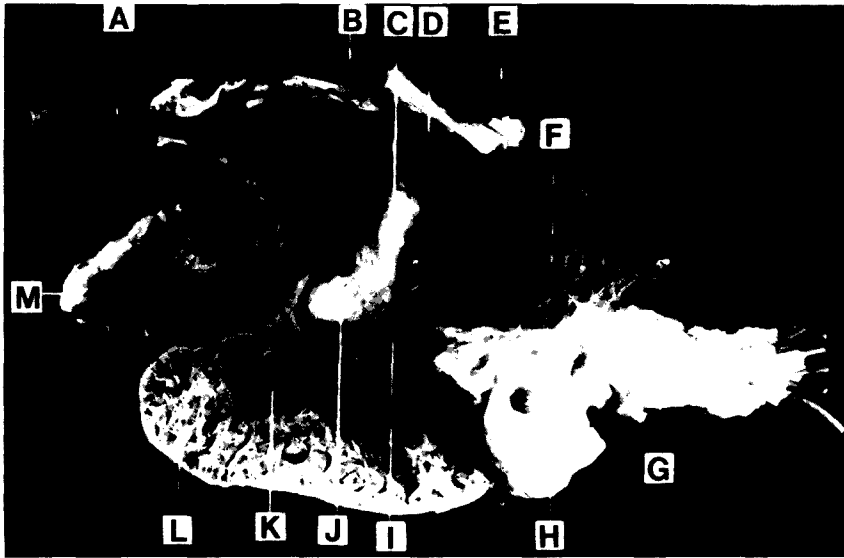


Fig. 2. Sagittal section through the heart of *Protopterus aethiopicus*. The arrow shows where the vena pulmonalis ends behind the pulmonalis fold. A. Probe in vena cava posterior; B. Probe in vena pulmonalis; C. Pulmonalis fold; D. Oblique fold separating the apertures of the two ducti Cuvieri; E. Probe in the right

ductus Cuvieri; F. Anterior unpaired part of the atrium; G. Distal section of the bulbus cordis; H. Spiral fold in the transverse section of the bulbus cordis, which has been opened; I. Atrioventricular aperture; J. Atrioventricular plug; K. Ventricular septum; L. Ventricular apex; M. Left auricular lobe. (From Bugge, '61.)

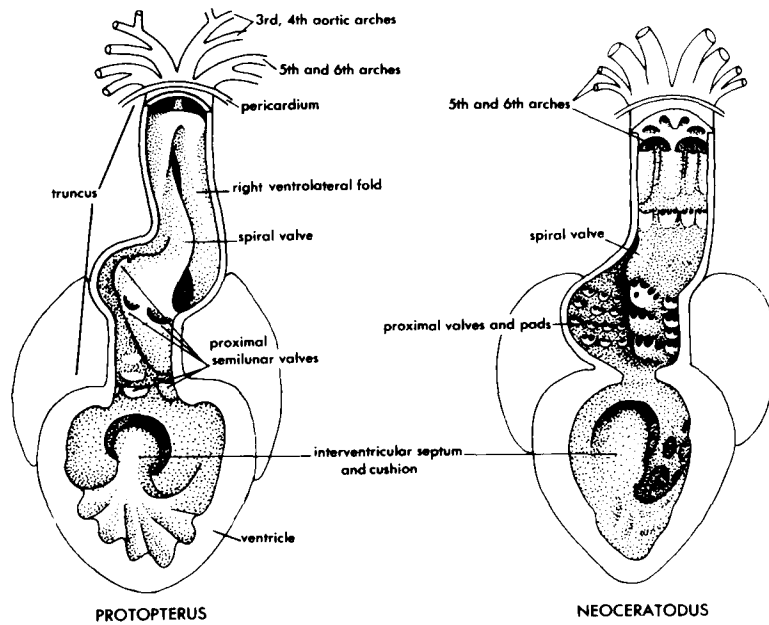


Fig. 3. Schematic sagittal sections through the heart of *Protopterus* and *Neoceratodus*. Hearts are shown from a ventral aspect. (From Jollie, '73, after Goodrich, '30.)

fishes are much more complex than the typical piscine heart, which functions only to propel deoxygenated systemic venous blood to the gills (Fig. 1A). The most notable specializations of the dipnoan heart are its more extensive S-folding, giving it an external appearance much more like that of a urodele amphibian, and the extensive internal septation, tending to subdivide the atrial and ventricular chambers.

In all three genera of lungfishes, the single atrium is partially divided into a larger right and smaller left side by a partial septum termed the pulmonalis fold, which arises from a deformation of the atrial wall caused by the overlying pulmonary vein, and by a unique structure termed the atrioventricular "cushion" (Goodrich, '30) or atrioventricular "plug" (Bugge, '61). The sinus venosus, conveying systemic venous blood, enters the atrium on its right side via a valved sinoatrial aperture. The pulmonary veins enter the atrium to the left of the pulmonalis fold, with this opening guarded by a tissue fold purportedly serving a valving function. Of the living lungfishes, *Lepidosiren* shows the greatest degree of atrial subdivision, possessing an almost complete interatrial septum. The atrioventricular orifice is not valved as in other fishes or in amphibians, but rather is guarded by the atrioventricular plug (Figs. 2, 3). This plug is raised out of the a-v orifice during atrial systole and lowered into it during atrial diastole, and thus it serves a valving function preventing regurgitation of blood into the atrium from the ventricle.

The dipnoan ventricle typically is composed of spongy, highly trabeculate myocardium (Fig. 2), resembling the construction of the ventricular walls of amphibians. In all three genera of lungfishes, a vertical septum arising from the dorsal and ventral walls of the ventricle anatomically divides much of the ventricular lumen, particularly toward the apex of the heart (Figs. 2, 3). Once again, *Lepidosiren* shows the greatest degree of ventricular septation, while *Neoceratodus* shows the least (Johansen et al., '68). In all three genera, the arrangement of the ventricular septum with respect to the pulmonalis fold and atrioventricular plug is such that oxygenated blood on the left side of the atrium will tend preferentially to flow into and to collect in the left side of the ventricle prior to ejection into the bulbus cordis. Deoxygenated blood from the right side of the atrium will flow toward the right side of the ventricle. In

amphibians, there is during diastole a heterogeneous distribution of atrial blood within the ventricle (which, with the exception of the ventricle of *Siren*, lacks any septation). The ventricular walls are highly trabeculate and act to trap and immobilize blood during ventricular filling. Together, these features minimize intraventricular mixing of oxygenated and deoxygenated blood prior to systolic ejection (Shelton, '76). Effective separation of blood in passage through the heart occurs in *Protopterus*, *Lepidosiren*, and, to a lesser extent, *Neoceratodus* (see below), a condition doubtlessly aided by mechanisms comparable to those operating in amphibians.

In all lungfishes the heart is surrounded by a tough, semi-rigid pericardium through which the central veins enter and the bulbus cordis exits. Although teleost fishes lack a stiff pericardium, it is a highly characteristic feature of all elasmobranchs, and its integrity is essential for normal cardiovascular function in sharks (Johansen, '65; Shabetai et al., '85). The condition of the pericardium in most actinopterygians is unknown.

The bulbus cordis

Separation of systemic and pulmonary blood streams passing through the heart will not assist preferential channelling of oxygenated and deoxygenated blood distally in the arterial tree unless laminar streaming of discrete blood flows can be maintained in the proximal arterial circulation. In this respect, the complex architecture of the bulbus cordis (or "truncus") plays a highly important role.

The bulbus cordis extends from the undivided anterior end of the ventricle, generally to the left of the ventricular midline (Fig. 3). Its walls are invested with cardiac muscle for the first third of its length. In *Neoceratodus*, there are several proximal rows of small conal valves (Lankester, 1878; Spencer, 1898; Goodrich, '09). These valves are reminiscent of the selachian conus, suggesting that this is a primitive character of gnathostomes. The lumen of the proximal region of the bulbus cordis is quite wide, and Satchell's ('76) interpretation is that these valves could prevent regurgitation of blood into the ventricle only during systole of the bulbus cordis when the valves are brought into apposition. Blood pressure in the bulbus cordis of *Neoceratodus* remains briefly elevated after pressure in the ventricle has fallen toward diastolic levels (Johansen et al., '68), suggesting that func-

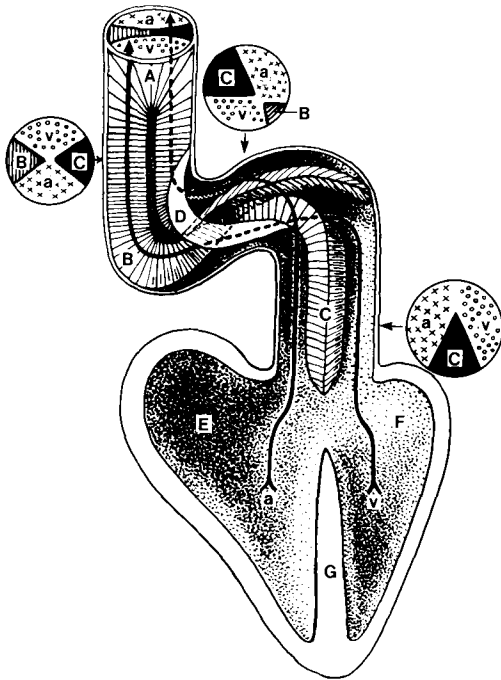


Fig. 4. Heart and bulbus cordis of *Protopterus aethiopicus* showing the structure of the ventricle and the bulbus cordis seen from the dorsal side. The arrows indicate the course of the arterial (a) and the venous (v) blood flow. A. The anteriorly joined part of the folds; B. The short left fold; C. The spiral fold; D. Cut surface where the spiral fold has been attached to the transverse section of the bulbus cordis; E. Left half of the ventricle; F. Right half of the ventricle; G. Vertical septum. (From Bugge, '61.)

tional valving at the base of the bulbus cordis does occur. The proximal base of the bulbus cordis of *Protopterus* and *Lepidosiren* contains a row of small "pocket valves" (Lankester, 1879; Boas, 1880; Robertson, '13; Bugge, '61), though these are considered largely "vestigial" (Johansen et al., '68). As discussed in detail by Satchell ('76), the hemodynamic function of the valves in the dipnoan bulbus cordis remains obscure and would be highly worthy of further anatomical and physiological investigation.

After leaving the heart, the bulbus cordis is sharply folded and twisted in all three genera (Fig. 3). This condition appears unique to lungfishes. However, a similar rotation of the truncus arteriosus occurs about its long axis in amphibians but is much more exaggerated in the lungfish, approximating 270° . In *Lepidosiren* and *Protopterus*, the lumen of the proximal region of the bulbus cordis is partially divided by the bulbar or spiral fold,

a fold of tissue arising from the ventral row of conal valves (Boas, 1880). This spiral fold continues down the length of the bulbus cordis, rotating 270° along with the bulbus cordis itself. In *Lepidosiren*, there is a spherical thickening of the distal end of the spiral valve that may serve a distal valving function (Robertson, '13). More distally, in *Lepidosiren* and *Protopterus*, a second smaller tissue fold arises from the inner bulbar wall directly opposite the spiral fold. The free edges of these two folds are nearly in apposition, thus partially dividing the lumen of the bulbus cordis into two outflow channels. In the most distal region of the bulbus cordis, the two folds fuse to form a horizontal septum that completely divides the lumen into dorsal and ventral channels. The ventral channel at the distal end of the bulbus cordis conveys primarily oxygenated blood that originates from the left side of the heart (Fig. 4). Comparatively deoxygenated blood from the right side of the heart is preferentially directed into the dorsal channel at the termination of the bulbus cordis.

Neoceratodus appears to show a less derived condition compared to other lungfishes, in that the bulbus cordis lacks a well-developed spiral valve (Fig. 3). Proximally, the spiral valve is evident, but within the bulbus cordis the distal pathway occupied by the spiral valve in the other lungfishes is instead marked by large semilunar valves in *Neoceratodus*. Distally, the spiral valve is increasingly prominent in *Neoceratodus*, dividing the bulbus cordis into a discrete dorsal and ventral channel as in the Lepidosirenidae.

Branchial and pulmonary arterial circulation

The aortic arch distribution of the Dipnoi shows the greatest modification from the basic piscine pattern of any living fish. In all lungfishes, the ventral aorta is virtually nonexistent, with the afferent branchial arteries arising almost directly from the distal end of the bulbus cordis (Fig. 3). In *Neoceratodus*, all four branchial afferents perfuse corresponding holobranchs of the branchial arches (Fig. 5). These arches bear numerous gill filaments and constitute a major site for respiratory gas exchange (Laurent, '84). There are no hemibranchs on the hyoid arch of *Neoceratodus*, which distinguishes this genus from *Lepidosiren* and *Protopterus* where the hyoid arch does bear a hemibranch. In these two genera, the two anterior-most holobranchs

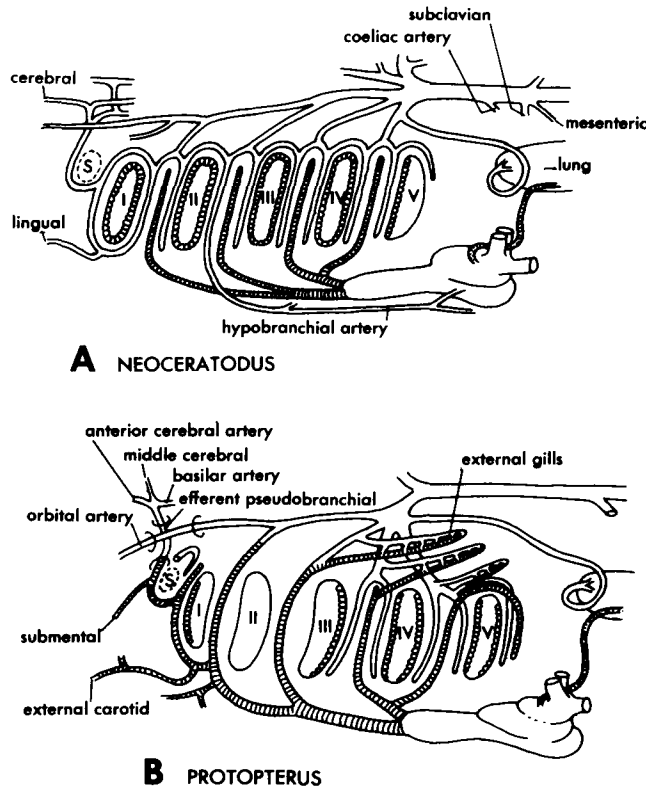


Fig. 5. Highly diagrammatic lateral view of the central circulatory morphology of *Neoceratodus* (A) and *Protopterus* (B). (Modified from Jollie, '73, after Goodrich, '30.)

are devoid of gill filaments (Johansen et al., '68; Laurent, '84). The afferent blood supply for these degenerate arches is derived from the ventral (oxygenated) channel of the bulbus cordis (as in *Neoceratodus*). The branchial efferents from these two anterior arches pass directly into the dorsal aorta. In essence, these two arches are nonrespiratory conduits shunting oxygenated blood from the left side of the heart directly into the dorsal aorta and on to the systemic tissues. In *Protopterus*, the two posterior-most arches possess highly modified gill filaments bearing secondary lamellae (Johansen et al., '68; Laurent et al., '78; Laurent, '85). Unlike the two anterior arches, these posterior arches can thus serve as respiratory surfaces. In *Lepidosiren*, however, the gill filaments on the two posterior arches are very sparse and coarse, and their respiratory role is equivocal (Fullarton, '31; Johansen and Lenfant, '67; Laurent et al., '78). Interestingly, equally well-developed gill filaments occur on all of

the branchial arches of the fossil lungfish *Griphognathus*, suggesting that the condition in *Lepidosiren* and *Protopterus* is derived rather than primitive (Campbell and Barwick, this volume).

In all three genera, the afferent blood supply to the posterior arches is derived from the dorsal channel of the bulbus cordis, which conveys relatively deoxygenated blood derived from the right side of the heart.

The course of the efferent vessels of the posterior arches of the Dipnoi is complex, particularly since many vessels merge and arise in a confined area dorsal to the gill arches (Fig. 5). Of the two posterior branchial arches, the third arch flows most directly into the dorsal aorta. The efferent vessel of the fourth branchial arch (embryonic arch VI) gives rise to the pulmonary artery in all three lungfish genera. This vessel is thus homologous with the pulmonary artery of tetrapods. In addition, however, efferent blood from this arch can bypass the lungs

and flow directly into the dorsal aorta via a highly specialized vascular segment variously termed the "ductus" (Fishman et al., '85) or "ductus arteriosus" (Laurent, '85). This structure has been considered analogous rather than homologous to the ductus arteriosus of mammals (Fishman et al., '85). Both the proximal pulmonary artery and the ductus are highly vasoactive and provide a mechanism by which blood draining the fourth gill arch can be preferentially directed into either the pulmonary or systemic circuits (see below).

In addition to the four branchial arches confined within the branchial cavity, all larval lepidosirenids possess external gills. *Neoceratodus* never has external gills as larva or adult (Kerr, '19). Some species of *Protopterus* retain external gills throughout the life cycle. These external gills are located downstream and in series with the three posterior gill arches (Fig. 5). Trewavas ('54) summarizes observations of many workers on the external gills of *Protopterus*. *Protopterus annectens*, *P. amphibius*, and *P. aethiopicus* all show external gills or their vestiges at even relatively advanced stages of development (e.g., 0.6-meter body length for *P. annectens*), although they are lost earliest in *P. aethiopicus*. However, the size of both these external gills and of the specimens bearing them is highly variable, and is an equivocal character on which to base species identification (Trewavas, '54).

The pulmonary circulation and lung structure

Many air-breathing fish carry out aerial respiration using a gas bladder connected via a pneumatic duct to the esophagus (Wilmer, '34; Johansen, '70; Randall et al., '81). In many species, these gas bladders are not just hollow bags but are structurally rather complex. Indeed, in fishes such as *Calamoichthys* or *Polypterus* the air-breathing organ should be regarded as a lung, in that 1) the pneumatic duct originates from a ventral evagination of the esophagus, and 2) its blood supply is derived from the sixth branchial arch, both conditions very similar to those of tetrapods (Goodrich, '30). Yet, amongst all air-breathing fishes, only in the Dipnoi does the structure of the lung so closely resemble that of extant amphibians and reptiles.

Neoceratodus possesses only a single lung, unlike both *Lepidosiren* and *Protopterus*, which have paired lungs. However, a small

vestige of a left pulmonary lobe has been described in the embryo of *Neoceratodus* (Neumayer, '04; Ballantyne, '27), suggesting that the adult condition of a single lung in *Neoceratodus* is a derived feature. In *Lepidosiren* and *Protopterus*, the lungs are fused anteriorly to form a common pneumatic duct that opens into the posterior region of the pharynx at the glottis. The pneumatic duct, which bears little resemblance to the tracheal-bronchiolar system of terrestrial vertebrates, is heavily invested with smooth muscle (Johansen et al., '68). This smooth muscle, as well as the considerable smooth muscle in the lung parenchyma, is probably important to the mechanics of lung ventilation and internal gas distribution (Grigg, '65; Johansen and Reite, '67).

Detailed accounts of the morphology of dipnoan lungs are given by Owen (1840), Hyrtl (1845), Gunther (1871), Parker (1892), Poll ('62), Grigg ('65), and Hughes and Weibel ('76). Internal septa, ridges, and pillars partition the lung into smaller lateral compartments opening into a central cavity (Fig. 6), similar to the arrangement in many amphibians and reptiles. The smallest pulmonary compartments in Dipnoi are approximately 1 mm in diameter, with the least degree of septation evident in *Neoceratodus* (Spencer, 1898; Poll, '62; Grigg, '65; Hughes and Weibel, '76). Although this degree of pulmonary septation exceeds that of some amphibians, Hughes and Weibel ('76) have estimated that *Lepidosiren*, for example, has a total pulmonary respiratory surface area only about one-tenth of that anticipated for a "typical" amphibian of similar body mass. However, mass-specific respiratory surface area of dipnoan lungs does considerably surpass that of the ABOs of many nondipnoan fishes (Munshi, '76). Diffusion distance between gas and blood in the lung capillaries of dipnoans is approximately 0.5–1.0 μm (Klika and Lelek, '67), which approaches the diffusion distance in mammal lungs.

The ventral surfaces of the lung(s) are perfused by the left pulmonary artery, which bifurcates about one-third of the way back from the cranial end of the lungs. The dorsal surfaces of the lung are perfused by the right pulmonary artery (Goodrich, '30).

GAS EXCHANGE IN LUNGFISHES

The preceding discussion has shown that extant lungfish show many cardiorespiratory morphological characters that distinguish

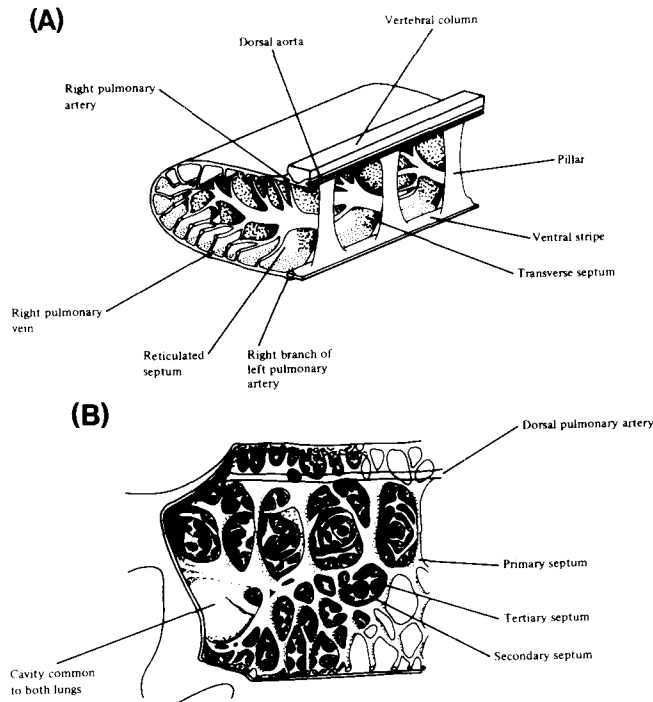


Fig. 6. Lung structure in Dipnoi. A. Reconstruction of a section of the right half of the single lung of *Neoceratodus forsteri*. (From Little, '83, after Grigg, '65.) B. Septa in the anterior region of one of the paired lungs of *Protopterus*. (From Little, '83, after Poll, '62.)

them from other air-breathing fishes and align them much more closely with tetrapods. While some of these characters clearly represent convergent evolution, others apparently are shared derived characters. What physiological advantages, if any, do these characters provide to the lungfishes, and what differences may exist among extant genera? To attempt to answer this question, it is necessary to examine how the cardiovascular and respiratory systems of lungfishes perform *physiologically*.

Mechanics of gill and lung ventilation

In spite of some structural differences (e.g., reduction of opercular and hyoid bones in *Protopterus* relative to *Neoceratodus*), the mechanics of both gill and lung ventilation are qualitatively the same in *Lepidosiren* (Bishop and Foxon, '68), *Neoceratodus* (Grigg, '65), and *Protopterus* (McMahon, '69). The most extensive account of the ventilatory mechanics of a dipnoan has been provided for *Protopterus aethiopicus* by McMahon ('69), who used X-ray cinematography, electromyography, and pressure recording to describe

the mechanics of both air and water breathing. Ventilation of the gills with water occurs through the action of a positive pressure buccal pump anterior to the gills and an opercular suction pump posterior to the gills. These pumps operate in concert to generate a nearly continuous water pressure gradient favoring water flow in the mouth, through the gills, and out the opercular opening (Fig. 7). Such a ventilatory system is widely shared amongst aquatic vertebrates, occurring in phylogenically ancient fishes, highly derived teleosts, and is even preserved somewhat in larval and adult amphibians (Shelton, '70; Gans, '70a; Randall et al., '81; Burggren et al., '86).

Ventilation of the lungs with air in *Protopterus* is achieved by action of the same musculoskeletal elements involved in aquatic ventilation (Fig. 7). In essence, a single aquatic ventilatory cycle irrigating the branchial chamber is replaced by a modified cycle that generates a single air breath. The single-breathing cycle occurs immediately after the snout has been thrust above the water surface into the air. The glottis, which has

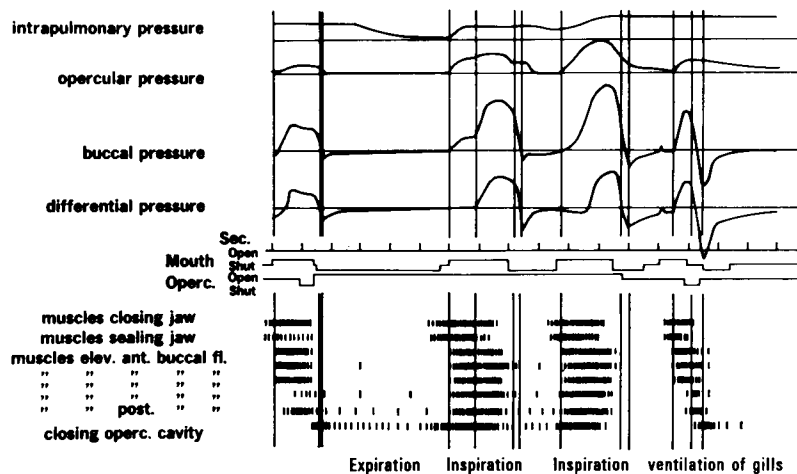


Fig. 7. Respiratory muscle activity and corresponding pressure changes during water- and air-breathing cycles in *Protopterus aethiopicus*. (From McMahon, '69.)

been tightly closed during water-breathing cycles, is opened, and pressure in the lungs decreases as expiration occurs. There follows a particularly tight closure of the opercular flaps, brought about by extended activity of the constrictor hyoideus muscle, and an exaggerated depression of the buccal floor drawing a large volume of fresh air through the opened mouth into the buccal cavity. The mouth is then closed, and the head reimmersed in water. A subsequent strong elevation of the buccal floor increases buccal pressure to approximately 20 mmHg, which is much higher than during a normal aquatic ventilatory cycle. This elevated buccal pressure drives inspired air past the open glottis into the lungs, completing the air-breathing cycle. The aquatic ventilatory cycles both preceding and following the cycle in which the lungs are ventilated show different patterns of muscle activation and the corresponding motor responses (Fig. 7). Lungfishes usually ventilate their lungs with a single air breath followed by a period of apnea of variable length, though under some experimental conditions tightly-grouped multiple breaths may occur (Bishop and Foxon, '68; McMahon, '69; Delaney and Fishman, '77). Estivation also causes changes in patterns and mechanics of lung ventilation (see Delaney and Fishman, '77; Fishman et al., this volume).

Lung ventilation in the Dipnoi thus incorporates a buccal force pump mechanism. Al-

though several workers have suggested that aspiration breathing involving rib movements for generation of subambient pleural pressures may occur in lungfish (Lomholt et al., '75) and other air-breathing fishes (Farrell and Randall, '78), these claims have not been substantiated (Greenwood and Liem, '84). It is most likely that the lung ventilatory mechanism seen in extant Dipnoi is similar to that of the earliest tetrapods, and that aspiration ventilation of the lung replaced buccal force ventilation only later as ribs took on increasing structural importance in fully terrestrial vertebrates (McMahon, '70).

Ventilatory patterns and oxygen uptake

As could be predicted from its elaborate and more typically piscine gill structure, *Neoceratodus* is an obligatory water breather. Laboratory observations indicate buccal/opercular movements at a frequency of about 30 cycles per min at room temperature (Johansen et al., '67). Observations of captive laboratory specimens at rest indicate that ventilation of the lung with air is comparatively rare when the fish is in oxygenated water. Thus, nearly 100% of both oxygen uptake and carbon dioxide elimination is aquatic via gills and skin (Table 1; Fig. 8). During prolonged swimming activity, when metabolic rate increases, air breathing becomes frequent and regular (Grigg, '65). Field observations of *Neoceratodus* (Kemp, this volume) indicate that air breathing may oc-

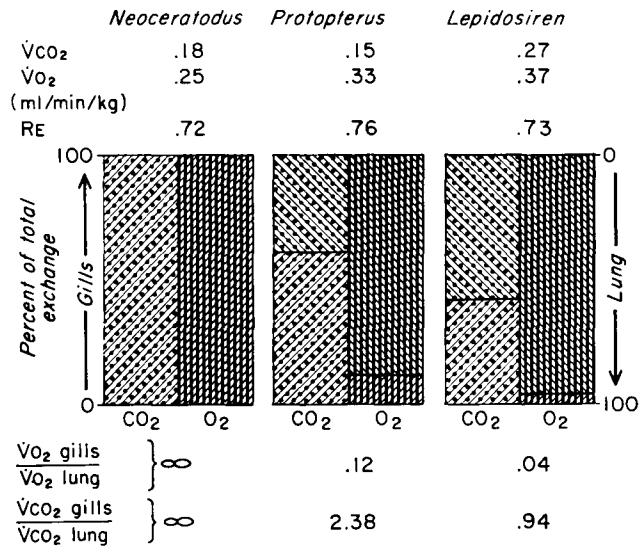


Fig. 8. Relative contributions of aquatic and aerial breathing to total gas exchange at 20°C in the three living genera of lungfishes. (From Lenfant et al., '70.)

cur more commonly during the breeding season, perhaps related to increased activity levels.

Although the respiratory surface of the gills of *Protopterus* is considerably reduced compared to *Neoceratodus*, the former nonetheless ventilates the gills with water, using slow, rhythmic buccal/opercular movements at a frequency of 0.5–10 cycles per min at 20–24°C (Lenfant and Johansen, '68; McMahon, '69, '70). Branchial respiratory movements in resting *Lepidosiren* are very shallow and almost imperceptible at rest, also varying widely between 1 and 20 breaths per min (Johansen and Lenfant, '67). Air-breathing frequencies in both genera range from 2–30 breaths hr⁻¹ at water temperatures of 20–24°C (Johansen and Lenfant, '67, '68; McMahon, '69, '70). Both *Protopterus* and *Lepidosiren* in postjuvenile stages are obligatory air breathers at normal environmental temperatures, and will drown if denied access to air. In adult *Protopterus* and *Lepidosiren*, 90% or more of total O₂ uptake occurs via the lungs, while a lower but still substantial proportion of CO₂ elimination occurs via this route (Table 1). This partitioning of O₂ and CO₂ exchange between air and water is also typical of larval amphibians that are similarly equipped with gills, permeable skin, and lungs (Burggren, '84).

While most experimental data have been collected from adult lungfishes, a limited but compelling number of observations indicate that developmental stage exerts a strong influence on patterns of gas exchange in lungfishes. In *Protopterus amphibius*, for example, the dependence upon aerial respiration increases dramatically as body mass increases (Fig. 9), and a similar trend appears to exist for *Lepidosiren* (Table 1). The influence of development on both cardiorespiratory morphology and physiology is one of the most neglected yet promising fields for future research.

At rest, *Neoceratodus* is clearly much less dependent upon aerial respiration than *Protopterus* or *Lepidosiren* (Table 1). What if more severe respiratory demands occur? A comparison of the ability to use aerial respiration can be assessed in more detail by comparing the respective abilities of lungfishes to survive brief periods of complete air exposure. Although this experiment is ecologically irrelevant for *Neoceratodus*, which, unlike *Protopterus* and *Lepidosiren*, does not estivate, laboratory air exposure makes specific and equivalent respiratory demands of all three lungfishes. The physiological responses to these demands will reveal the existence of mechanisms crucial to the evolution of a terrestrial capability. Figure

TABLE 1. Oxygen uptake, carbon dioxide elimination, and the gas exchange ratio in Dipnoi¹

Species	Temperature (°C)	Oxygen uptake (ml O ₂ g ⁻¹ h ⁻¹)			Carbon dioxide elimination (ml O ₂ g ⁻¹ h ⁻¹)			Gas exchange ratio			References
		Aquatic (%)	Aerial (%)	Total	Aquatic (%)	Aerial (%)	Total	Aquatic	Aerial	Total	
<i>Neoceratodus forsteri</i>	18	15.0 (100)	0 (0)	15.0	18.6 (100)	0 (0)	18.6	1.24	—	1.24	Lenfant et al., '67
<i>Lepidosiren paradoxa</i> (juvenile, 150 gm)	18	54.0 (64)	30.6 (30)	84.6	72.6 (77)	22.2 (23)	94.8	1.34	0.73	1.12	Johansen and Lenfant, '67
<i>Lepidosiren</i> (adult)	20	1.8 (4)	40.2 (96)	42.0	—	—	—	—	—	—	Sawaya, '46
<i>Lepidosiren paradoxa</i>	20	0.7 (3)	21.5 (97)	22.2	6.7 (41)	9.5 (59)	16.2	9.57	0.44	0.73	Lenfant et al., '70
<i>Protopterus aethiopicus</i>	20			52.2							Smith, '30
<i>Protopterus aethiopicus</i>	24	5.0 (8)	57.5 (92)	62.5	31.8	15.6	47.4	5.40	0.27	0.73	McMahon, '70
<i>Protopterus aethiopicus</i>	20	1.3 (11)	10.1 (89)	11.4	6.1 (70)	2.6 (30)	8.7	4.69	0.26	0.76	Lenfant and Johansen, '68

¹All values recorded at the indicated temperature in air-saturated water with free access to air.

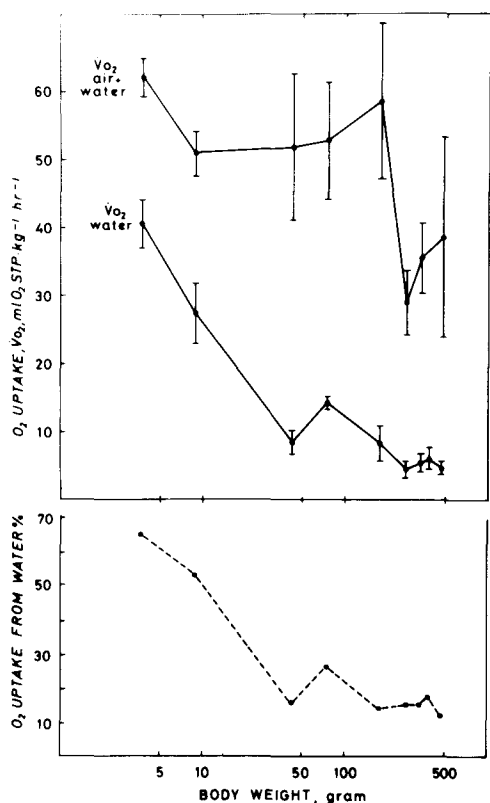
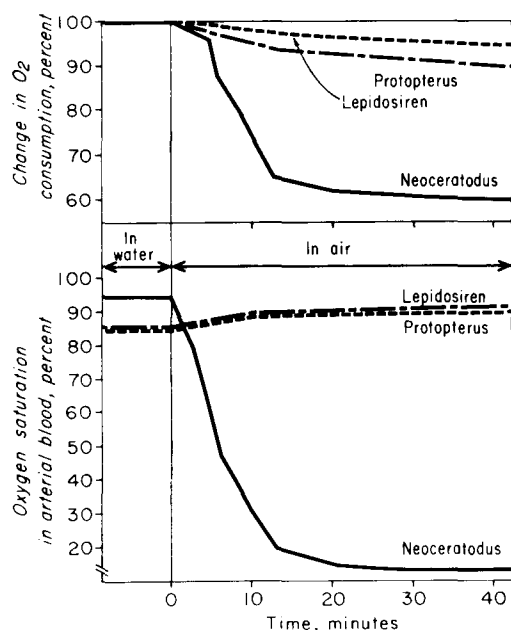


Fig. 9. Mass-specific oxygen uptake in *Protopterus amphibius*, expressed in absolute amount (top) and as a percentage of total O_2 uptake (bottom). (From Johansen and Lomholt, '76.)



10 compares total O_2 consumption and ability to saturate systemic arterial blood with oxygen in *Neoceratodus*, *Lepidosiren*, and *Protopterus* before and during 40 min of complete air exposure. When removed from water, all three genera of lungfishes show highly elevated rates of lung ventilation. Both *Protopterus* and *Lepidosiren* can maintain blood O_2 saturation and total O_2 uptake by shifting entirely to pulmonary gas exchange. *Neoceratodus*, in contrast, shows a serious insufficiency in blood gas transport within just 5–10 min of air exposure, reaffirming the much more aquatic nature of the Australian lungfish.

Neoceratodus can be further distinguished from *Protopterus* and *Lepidosiren* on the basis of its respiratory responses to environmental O_2 availability. When exposed to moderately hypoxic water, *Neoceratodus* responds with an immediate and large increase in rate of gill ventilation (Johansen et al., '67). Air breathing is initiated and progressively increases in frequency, though the onset of lung hyperventilation is much later than that of increased gill ventilation. Given that *Neoceratodus* has elaborate gills and depends heavily on aquatic exchange under normoxic conditions (Table 1), it is not surprising that this lungfish should show responses to aquatic hypoxia very reminiscent of strictly aquatic fishes (see Shelton '70). Adult *Protopterus*, on the other hand, shows virtually no branchial ventilatory response to aquatic hypoxia (Johansen and Lenfant, '68). This is consistent with the almost total lack of reliance on water for O_2 uptake shown by this fish (Table 1; Fig. 8). Exposure to aerial hypoxia, however, induces rapid increases in air-breathing frequency, indicating that *Protopterus* does, in fact, have chemoreceptors sensitive to changes in internal O_2 levels. Interestingly, juvenile *Protopterus* respond to exposure to hypoxic water by increasing rates of both water and air breathing (Jesse et al., '67). Juvenile *Protopterus* are more dependent on aquatic gas exchange (Johansen and Lenfant, '68; Johansen et al., '76a; Fig. 9), and a ventilatory response to hypoxic water in these less mature lungfish is more appropriate.

Fig. 10. Effect of air exposure upon O_2 uptake and O_2 saturation of arterial blood in *Neoceratodus*, *Lepidosiren*, and *Protopterus*. (From Lenfant et al., '70.)

Blood respiratory properties

Blood and its circulation represent an indispensable link in the transport of respiratory gases between the ambient environment and the metabolizing cells. The most important respiratory properties of vertebrate bloods are the O₂ affinity of the blood, expressed as the partial pressure of O₂ at which the blood is half saturated with O₂ (the P₅₀), O₂ capacity, which is the maximum possible amount of O₂ contained per unit of blood, and the influence of blood pH on the O₂ affinity (commonly referred to as the "Bohr shift"). Blood is also important in CO₂ transport and in acid-base balance of the body fluids.

The respiratory properties of blood show a remarkable evolutionary plasticity or adaptability to environmental and behavioral situations in lower vertebrates. In regard to air-breathing fishes breathing bimodally with gills and/or an air-breathing organ, blood O₂ affinity tends to decline with increased dependence on aerial breathing (Johansen and Lenfant, '71). Since atmospheric air has a stable and high concentration of O₂, blood in the air-breathing organ can become fully O₂ saturated even in the face of a reduced blood O₂ affinity. Yet, at the level of the body tissues, the lower O₂ affinity will enhance unloading of oxygen from the blood (Johansen and Lenfant, '71). In air-breathing fishes that depend predominantly on aquatic breathing for oxygen uptake, there is an equally clear tendency for blood O₂ affinity to be increased, particularly if the species lives in oxygen deficient water. Thus, even though the major respiratory medium is oxygen deficient, blood of a higher O₂ affinity may still become O₂ saturated before leaving the gills or skin. The blood O₂ affinity of fishes thus appears to show evolutionary adaptation to the O₂ availability of the external medium most important to respiration. However, it is important to emphasize that an adaptation that may assist O₂ unloading at the tissues may be maladaptive with respect to loading in the gas-exchange organ, and *vice versa* (see Johansen and Lenfant, '71). Thus, each species in each environmental condition has to be examined individually to assess whether changes in blood properties are assisting blood oxygen transport.

A comparison of blood respiratory properties in lungfishes is difficult, primarily due to a shortage of data and to the different techniques used to measure blood O₂ affinity. Johansen and Lenfant ('67) found a very high

blood O₂ affinity in juvenile *Lepidosiren*. Using different techniques, Johansen et al. ('78) studied blood from mature *Lepidosiren* and obtained similar high affinities (P₅₀ 8 mmHg at pH 7.4, 29°C). *Neoceratodus* shows a lower blood O₂ affinity than *Lepidosiren* or *Protopterus* if compared at similar blood pH to take into account the Bohr shift. However, when compared at the values for pH occurring in vivo in arterial blood, *Neoceratodus* has a higher affinity for O₂ than the obligate air breathers *Lepidosiren* and *Protopterus* (Lenfant et al., '67). The blood of *Neoceratodus* also shows a larger CO₂ combining power and buffer capacity than the blood of either *Lepidosiren* or *Protopterus*. However, there appears to be little difference in blood oxygen capacity between lungfish genera (Johansen and Lenfant, '71).

Protopterus shows a low O₂ affinity expressed by a P₅₀ of 33 mmHg at pH 7.5 and 26°C (Johansen et al., '76b). Blood O₂ affinity of estivating *Protopterus* shows a dramatic increase to 9 mmHg when measured at the same temperature and pH as typical for an active, hydrated lungfish. However, since the blood pH of the estivating fish is much lower, the very large difference in blood oxygen affinity will be diminished by the Bohr shift when considering the in vivo situation. No satisfactory explanation has been advanced to explain the marked increases in O₂ affinity with estivation. Its mechanism, however, is clearly related to a large reduction in the red cell organic phosphates GTP and ATP, both of which act as important cofactors (or competing ligands) for the O₂ binding to hemoglobin. A higher Bohr factor in *Neoceratodus* than the other lungfishes must also be considered adaptive as long as this species maintains a much higher blood pH than the other lungfishes.

Much additional research on the blood respiratory properties of lungfishes is required to understand fully to what extent the differences in blood properties between genera represent important adaptive changes to differing demands on blood gas transport.

Cardiorespiratory regulation

A fundamental requirement of effective gas exchange is that the O₂ capacitance and other respiratory properties of blood on one side of the gas-exchange membranes should at all times be closely matched with the oxygen capacitance of the respiratory medium on the other side of the membrane (Piiper

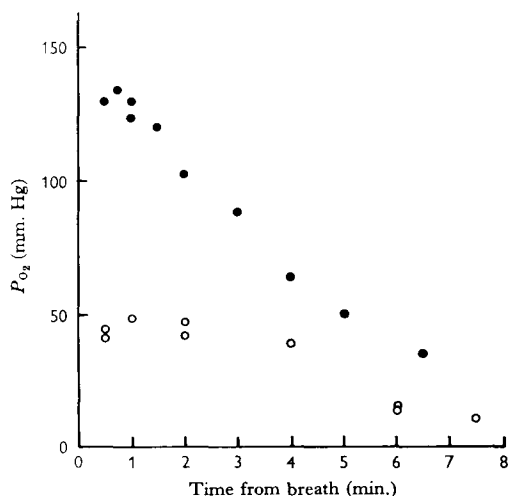


Fig. 11. Changes in O_2 partial pressure of pulmonary venous blood (open circles) and pulmonary gas (solid circles) following a spontaneous breath in *Lepidosiren paradoxa*. (From Johansen and Lenfant, '67.)

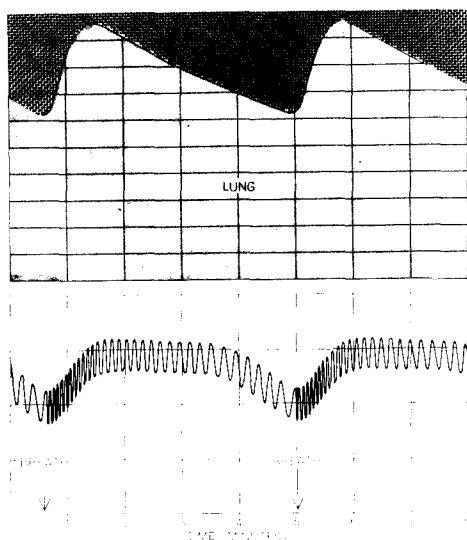


Fig. 12. Influence of voluntary, intermittent breathing upon source of systemic arterial blood (top) and upon pulmonary blood flow and heart rate (bottom) in *Protopterus aethiopicus*. (From Johansen, '68.)

and Scheid, '84). Stated differently, the ability of the blood to convey O_2 away from the gas-exchange organ should ideally equal the ability of the ventilatory pump to supply O_2 to that organ. Thus, hyperventilation of a gas-exchange organ with respiratory medium will achieve little increased gas exchange unless there is a corresponding increase in blood flow to that organ.

This concept of "ventilation-perfusion" matching takes on particular importance in

animals in which the ABO is ventilated intermittently rather than continuously. In animals such as the Dipnoi, the partial pressure of O_2 (PO_2) in pulmonary gas is typically high during and immediately after an air breath, but then falls progressively during breath holding until a subsequent breath renews the gas in the ABO (Fig. 11). Thus, both the total O_2 stores of the lungs and the gas partial pressure gradient driving O_2 from the gas into blood in the pulmonary capillaries are greatest in the early moments of breath holding. For most effective gas exchange, perfusion of the lungs with blood should be highest during this period when the potential for pulmonary gas exchange is highest, and should then diminish as breath holding progresses.

Physiological investigations of the last two decades have revealed that lungfishes possess effective mechanisms for matching blood perfusion of the lung to the intermittent pattern of lung ventilation. The most extensive hemodynamic studies have been performed on *Protopterus*, with limited observations on *Neoceratodus* and *Lepidosiren* (for reviews see Johansen, '70; Johansen and Burggren, '80; Johansen, '85; Fishman et al., '85; Burggren et al., '86). These experiments, often performed on intact, unanesthetized lungfish, have combined radiographic studies, direct measurement of blood flows and pressures, and estimations of cardiac output distribution based on O_2 concentrations of blood sampled from indwelling catheters located at various sites in the circulation. During the early minutes following an air breath in *Protopterus aethiopicus*, heart rate and cardiac output increase (Fig. 12). Blood leaving the two posterior-most gill arches is directed primarily into the pulmonary artery rather than the ductus arteriosus, and so pulmonary blood flow rises considerably. Since the PO_2 of lung gas is highest at this time, there is a major transfer of O_2 from lung gas to pulmonary venous blood. However, as breath holding progresses and the gas exchange potential of the lung decreases, then heart rate, cardiac output, and total lung blood flow decrease.

Clearly, *Protopterus* regulates cardiovascular function to match lung perfusion to lung ventilation. For several reasons, however, simply increasing pulmonary blood flow when lung PO_2 is highest is not sufficient for effective gas exchange. It must be emphasized that those gills that bear filaments and



Fig. 13. Ventro-dorsal (left) and lateral view of the passage of radio-opaque dye injected into the pulmonary vein. da. dorsal aorta; v. ventricle. (From Johansen and Hol, '68.)

secondary lamellae can be a site of O_2 loss as well as gain. Since lungfishes may encounter water with low O_2 levels (see Greenwood, Kemp, both in this volume), there is a very real potential that O_2 transferred from lung gas into pulmonary venous blood may be lost immediately back to hypoxic water flowing over the gills. Such a reversal of O_2 flow has been well documented for both air-breathing fish (Burggren, '79) and larval amphibians (West and Burggren, '82). Thus, it is essential for lungfish that, at least immediately following an air breath, oxygenated pulmonary venous blood en route to the systemic tissues not come into contact with branchial water. This separation can be achieved in one of two ways. In *Protopterus aethiopicus*, each secondary lamella of the gills has, in addition to the distal blood channels exposed to water, a thick-walled and muscular basal blood channel that directly connects afferent and efferent branchial arterial vessels (Laurent, '85). A decrease in the vasomotor tone of this basal channel (which has yet to be experimentally established) would, at the level of each secondary lamellae, shunt blood around rather than through the respiratory membranes (Fishman et al., '85; Laurent, '85). A second mechanism preventing oxygenated blood from contacting branchial water potentially low in O_2 involves a preferential distribution of pulmonary venous blood into the two anterior-most branchial arches bearing few (*Lepidosiren*) or no (*Protopterus*) secondary lamellae. Perfusion of the anterior arches with primarily pulmonary

venous blood will achieve a functional bypass of the respiratory membranes of the gills. Importantly, it will also prevent oxygenated blood from being needlessly recirculated back to the lungs before perfusing systemic tissues.

While the anatomical basis for channelling of systemic and venous blood during its passage through the central circulation has long been appreciated (see discussion above), physiological evidence for this separation has been provided only in the last few decades. As indicated in Figure 12, immediately following an air breath nearly 100% of the blood perfusing the anterior arches (and thus passing directly into the dorsal aorta without exposure to branchial water) is derived from the pulmonary vein. The extent of separation can be seen quite clearly in Figure 13, which shows the course of radio-opaque dye directed via an indwelling catheter into the pulmonary vein of *Protopterus aethiopicus*. As breath holding progresses, the partial pressure of O_2 in pulmonary venous blood decreases (Fig. 11) and approaches that of systemic venous blood. Under this condition, separation of pulmonary and systemic venous blood becomes less important and could even become detrimental if there is an energetic cost associated with maintaining this separation (see Burggren, '87). In fact, as breath holding continues, the proportion of pulmonary blood perfusing the anterior branches diminishes (Fig. 12), while the proportion perfusing the posterior, gill-bearing branches increases.

The actual mechanisms by which these labile and constantly changing patterns of gill and lung perfusion are achieved has been investigated in some detail, once again primarily for *Protopterus aethiopicus* (see Fishman et al., '85). Pharmacological investigations employing cholinergic and adrenergic agonists and antagonists have revealed that the ductus and the extrinsic segment of the pulmonary artery are highly vasoactive. Interestingly, vasoactivity in the extrinsic segment of the pulmonary artery is characteristic of amphibians (De Saint-Aubain and Wingstrand, '79; Malvin, '85) and is retained in some reptiles (Burggren, '77; Milson et al., '77). Importantly, in *Protopterus* the pharmacological responses of the ductus are somewhat opposed to those of the extrinsic pulmonary artery. For example, perfusion of isolated arterial segments with dilute acetylcholine solutions induces vasodilation of the ductus but vasoconstriction of the pulmonary artery (Fishman et al., '85). Norepinephrine, on the other hand, induces intense vasoconstriction of the ductus but has no effect on the extrinsic segment of the pulmonary artery. How might these mechanisms operate in vivo? An increase in catecholamines circulating in blood plasma will cause vasodilation of the ductus relative to the pulmonary artery, favoring a pulmonary bypass in which blood from the fourth branchial arch flows through the ductus into the dorsal aorta, rather than into the pulmonary artery (Fig. 5). Parasympathetic activity, on the other hand, will cause vasoconstriction of the ductus and vasodilation of the extrinsic pulmonary artery, tending to direct blood draining the fourth branchial arch preferentially into the pulmonary rather than the systemic circuit.

As evident from the above discussion, the mechanisms by which perfusion is matched to intermittent lung ventilation, where they have been determined, are known primarily for *Protopterus*. Although *Neoceratodus* increases lung ventilation during exposure to hypoxic water (Johansen et al., '67; Lenfant et al., '67), very little is known about how cardiac output is redistributed and how oxygenated blood circumvents the branchial respiratory membranes.

PHYLOGENETIC CONSIDERATIONS

As abundantly evident from other presentations in this volume, most considerations of the systematics of lungfishes have been

based on characters of dentition, cranial structure, etc. The major reason for this is not that these characters are inherently "more reliable" than soft tissues, but rather that the fossil record contains more evidence of such structural elements of phylogenetically primitive lungfishes, allowing direct comparisons with the surviving genera.

Although somewhat beyond the scope of this paper, it is important to emphasize that characters based on cardiorespiratory morphology (and the physiological processes which these structures support) may be of great value in describing systematic relationships of living fishes, including the Dipnoi. To give but a few examples, *Neoceratodus*, with less septate lungs, more fully developed gills, and a less partitioned central circulation, appears considerably less derived from what is generally interpreted to be the plesiomorphic condition for lungfishes. *Lepidosiren* and *Protopterus*, with cardiorespiratory structures more heavily modified for aerial respiration and terrestrial survival, possess many structures comparable to those of the tetrapods. At the same time, *Lepidosiren* and *Protopterus* also exhibit a number of differences in cardiorespiratory characters, such as the degree of anatomical and functional atrial and ventricular division, branchial structure, and persistence of external gills.

It is important to emphasize that any phylogenetic analysis of morphological or physiological characters should include not only consideration of characters for the ingroup in question (in this instance, the Dipnoi), but also in more primitive fish taxa. Such a cladistic analysis of lungfish systematics based on respiration and cardiovascular characters is currently being attempted.

CONCLUSIONS

Perhaps no vertebrates so recently discovered, with so few living representatives, and with such remote distributions, have received so much attention from so many biologists as have lungfishes. While many contributions in this volume attest to the fascinating morphology and systematics of the Dipnoi, we wish to emphasize that, after all, they were named LUNGfish! Their successful exploitation of aerial respiration, culminating in the ability to tolerate months or years of air exposure during estivation, surely ranks among the most remarkable features of vertebrates.

In our attempt to present a coherent overview of the cardiorespiratory physiology of living lungfishes, we have made many generalizations. Perhaps most notable of these is our position that *Neoceratodus* is less highly derived from the plesiomorphic condition for lungfishes than either *Lepidosiren* or *Protopterus*, which have many morphological and physiological features similar to those of tetrapods. However, it is important to indicate that, just as the fossil record of lungfishes has gaps, so too does the "morphological and physiological record" pertaining to circulation and respiration in living lungfishes. Particularly important areas of lungfish cardiorespiratory physiology that require much additional investigation include:

- 1) effects of embryonic and larval development and maturation
- 2) environmental/ecological influences
- 3) mechanisms for reflex regulation
- 4) more intensive investigation of *Lepidosiren*, which is poorly represented in the physiological literature
- 5) influence of estivation on metabolism

Finally, it is somewhat ironic that, in attempting to better our understanding of the cardiorespiratory morphology and physiology of lungfishes relative to other air-breathing fishes and amphibians, in some instances we now know more about lungfishes than the animals to which we compare them. Only by thoroughly understanding the general processes by which aquatic organisms came to exploit the aerial/terrestrial environment will we be able to use the Dipnoi to help us interpret the evolutionary transition from fishes to tetrapods.

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