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## PECULIARITIES OF PRENATAL DEVELOPMENT OF LUNGS IN PLAIN-NOSED (VESPERTILIONIDAE) AND HORSESHOE (RHINOLOPHIDAE) BATS

I. M. Kovalyova

*Schmalhausen Institute of Zoology, vul. B. Khmelnyts'kogo, 15, Kyiv-30, MSP, 01601 Ukraine*

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**Peculiarities of Prenatal Development of Lungs in Plain-Nosed (Vespertilionidae) and Horseshoe (Rhinolophidae) Bats. Kovalyova I. M.** — For the first time the prenatal development of lungs and their definitive structure were studied in representatives of plain-nosed bats (*Vespertilionidae*) and horseshoe bats (*Rhinolophidae*). The results of this investigation show that in the studied bats the laying of lungs being at the 14<sup>th</sup> stage of the embryogenesis, as well as their subsequent development right up to the 18<sup>th</sup> stage were of the same mode. However after the 18<sup>th</sup> stage the development of lungs in the studied plain-nosed bats was sharply different from that in horseshoe ones when forming the structure of these organs. The explanation of such a phenomenon is given in connection with distinctions in spatial reciprocal location of the thorax organs, the body, and the head during embryogenesis. The supposition about the probable mediated relation between the bats echolocation (namely methods of radiating signals) and the peculiarities of lungs structures was suggested.

**Key words:** Vespertilionidae, Rhinolophidae, bats, prenatal development, lungs.

**Особенности пренатального развития легких у некоторых гладконосых (Vespertilionidae) и подковоносых (Rhinolophidae) летучих мышей. Ковалева И. М.** — Впервые исследовано пренатальное развитие легких и их дефинитивное строение у представителей рукокрылых: гладконосых летучих мышей (*Vespertilionidae*) и подковоносых летучих мышей (*Rhinolophidae*). Установлено, что у исследованных рукокрылых, начиная с 14-й стадии эмбрионального развития, закладка легких, а также дальнейшее их развитие вплоть до 18-й стадии происходит одинаково. Однако после 18-й стадии развитие легких гладконосых и подковоносых летучих мышей резко различается в части структурообразования рассматриваемых органов. Объяснение обнаруженному феномену дается в связи с различиями в ходе эмбриогенеза пространственного взаиморасположения органов грудной полости, туловища и головы. Высказано предположение о возможной опосредованной связи эхолокации рукокрылых (а именно: способов излучения сигналов) с особенностями в строении легких.

**Ключевые слова:** Vespertilionidae, Rhinolophidae, рукокрылые, пренатальное развитие, легкие.

### Introduction

There was no description of the embryology of bats' lungs in scientific literature, so the first results devoted to this question were published by the author of these work (Kovalyova, 1998 a, b; 1999; Kovalyova, Melnyk, 2000). The results on studies in the definitive structure of Bats' lungs showed that the lungs had the lobeless structure as well as the lobe one (Zhedenov, 1957; Torubarova, 1958; Kovalyova, 1995). V. N. Zhedenov marked in bats almost all kinds of transitions from typically lobe lungs to lobeless ones, and put forward the hypothesis that the presence of lobeless lungs in horseshoe bats is the secondary phenomenon realized by means of merging lobes. However the proper process of forming lobe and lobeless lungs in bats have not yet been investigated up to now in comparing aspects.

The present paper is devoted to the research in the prenatal development of lungs in representatives of two families of bats, taking into account the difference in the definitive structure of their lungs. Namely, the lungs in the Vespertilionidae representative are known being the lobe structure while those in the Rhinolophidae ones being the lobeless structure.

### Material and methods

The material consisted of the ontogenetic series of embryos and fetuses of bats, belonging to the species of *Rhinolophus hipposideros*, *Rhinolophus ferrumequinum*, *Rhinolophus bocharicus*, *Rhinolophus blasii*, *Myotis blythii* and *Nyctalus noctula* (tabl. 1).

The embryos and fetuses were sectioned in 10  $\mu\text{m}$  samples, and the sections obtained were stained with haematoxylin by the Mellory's method. The material was divided by the level of ontogenetic development and the Crown-Rump Length (CRL) into 29 groups according to the author's stages classification system. The system allows to classify materials from both embryonic and fetus periods and is based on the Carnegie system (O'Rahilly, 1972), the Sterba comparative ontogenetic stages system (1985, 1990) and the Ledenev, Likhotop system (1988). Also the lungs of 15 adult individuals of *Vespertilionidae* and *Rhinolophidae* families were prepared with a view to carry out the morphometrical analysis.

## Results

The primordium of the trachea was formed from the epithelium of the gut anterior wall. At first the primordium looked as a longitudinally situated protuberance in the level of the most rear pair of pharyngeal pouches (fig. 1, *a*, *b*). The protuberance appeared at the 14<sup>th</sup> stage of development (CRL 4.0 mm).

At the 16<sup>th</sup> stage the bifurcation of the trachea terminal end took place having appearance of laterally directed pouches — “bronchial buds” (fig. 2).

At the 17<sup>th</sup> stage the bronchial buds were transformed into primary bronchi (fig. 3).

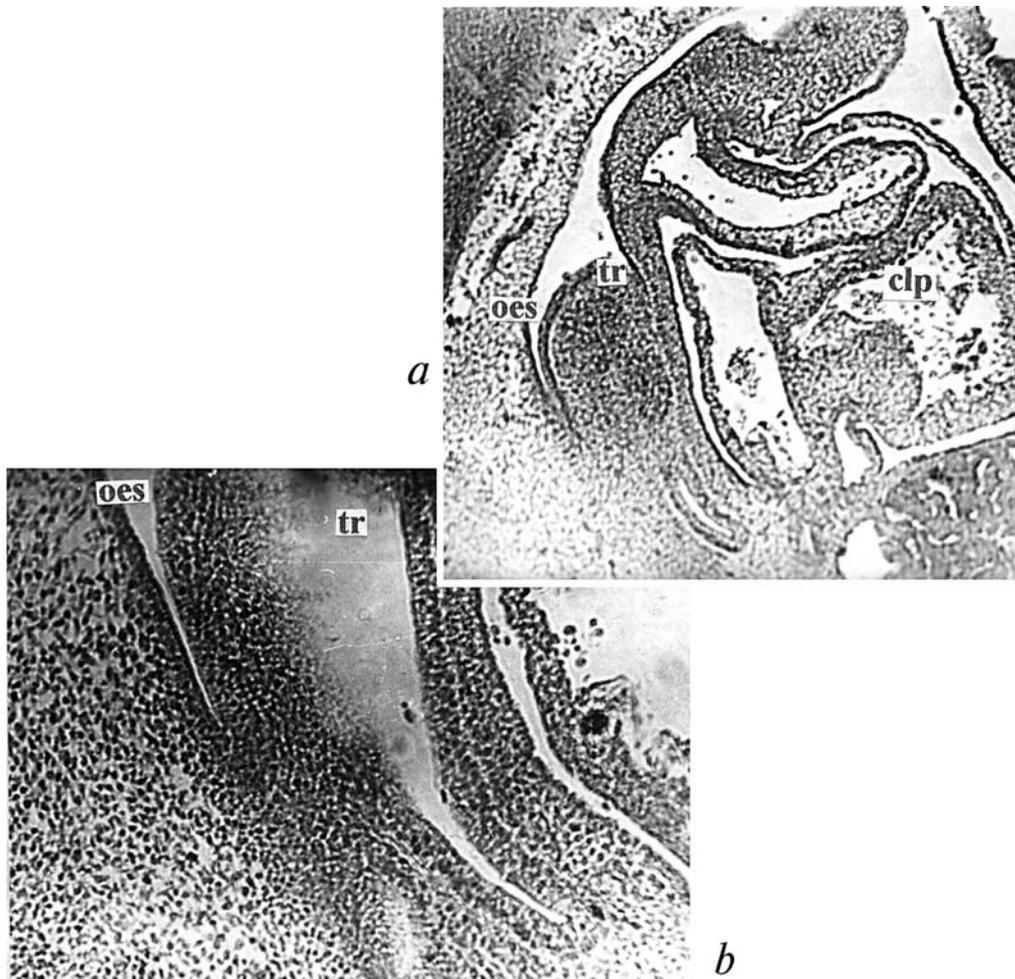


Fig. 1: *a* — embryo *Nyctalus noctula* CRL 4.0 mm; 14<sup>th</sup> stage; sagittal section through primordium of the trachea (tr), oesophagus (oes), cardiac-liver protuberance (clp); *b* — detail of preceding section.

Рис. 1: *a* — эмбрион *Nyctalus noctula* CRL 4,0 мм; 14-й стадии; сагитальное сечение через зачаток трахеи (tr), пищевода (oes), сердечно-печеночного выступа (clp); *b* — детализация предыдущего сечения.

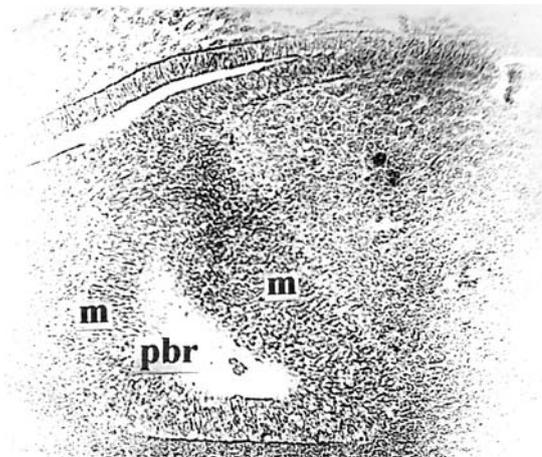


Fig. 2. Embryo *Nyctalus noctula* CRL 5.9 mm; 16<sup>th</sup> stage; sagittal section through the primordia of the principal bronchi (pbr) covered with dense mesenchyme (m).

Рис. 2. Эмбрион *Nyctalus noctula* CRL 5,9 мм; 16-й стадии; сагиттальное сечение через зачатки первичных бронхов (pbr), окруженных плотной мезенхимой (m).



Fig. 3. Embryo *Nyctalus noctula* CRL 6.2 mm; 17<sup>th</sup> stage; transversal section through bronchus (br), oesophagus (oes). The wall of bronchi consists of high epithelium (e).

Рис. 3. Эмбрион *Nyctalus noctula* CRL 6,2 мм; 17-й стадии; трансверсальное сечение через бронхи (br), пищевод (oes). Стенка бронхов образована высоким эпителием (e).

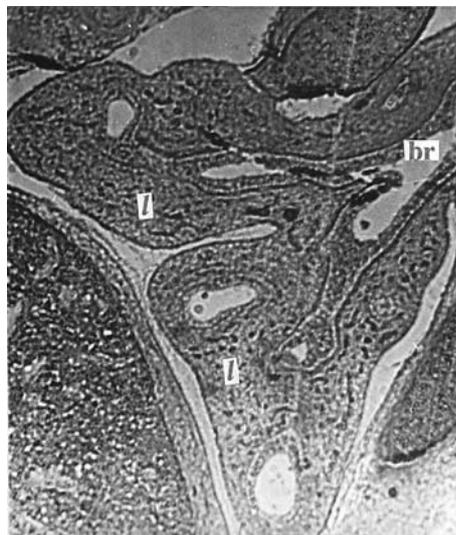


Fig. 4: a — Embryo *Nyctalus noctula* CRL 9.1 mm. The right lung is divided into lobes (l) according primary bronchi (br); b — Embryo *Rhinolophus hipposideros* CRL 8.8 mm. The right lung is divided into “lobes” (l).

Рис. 4: a — эмбрион *Nyctalus noctula* CRL 9,1 мм. Правое легкое разделено на доли (l), соответственно первичным бронхам (br); b — эмбрион *Rhinolophus hipposideros* CRL 8,8 мм. Правое легкое разделено на «псевдодоли» (l).

At the 18<sup>th</sup> stage the primordium of the right lung in *Nyctalus noctula* (fig. 4, a) was represented by a primary bronchus with 4 bronchial buds, while that in the left lung had only 2 bronchial buds.

Table 1. Review of the material examined (prenatal stages)

Таблица 1. Перечень исследованного материала (пренатальные стадии развития)

Species	Stage	CRL, mm	Total weight, mg
<i>Nyctalus noctula</i>	14	4.0	8.0
	16	5.9	30.0
	17	6.2	39.5
	18	8.0	72.0
	19	9.5	112.0
	19	9.1	123.5
	20	11.8	232.0
<i>Myotis blythii</i>	22	14.4	393.0
	22	15.5	300.0
	18–19	8.8	115.0
<i>Rhinolophus hipposideros</i>	20	12.0	363.0
	25	15.3	500.0
	15	5.0	10.0
<i>Rhinolophus bocharicus</i>	17	6.4	31.0
<i>Rhinolophus blasii</i>	18	8.5	40.0
	20	—	—
	22	16.9	790
	25	19.3	1760.0
	fetus	22.5	2575.0

Notes. CRL stands for Crown-Rump Length.

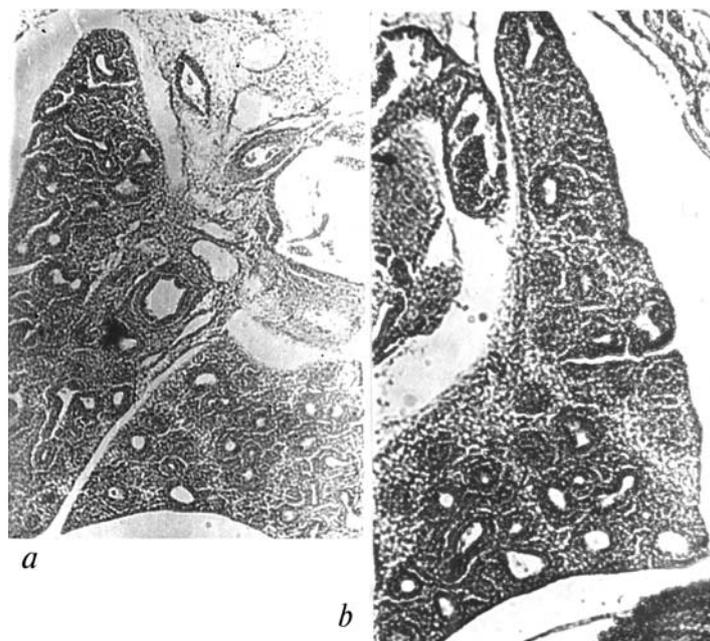


Fig. 5. Embryo *Nyctalus noctula* CRL 11.8 mm. The right (a) and the left (b) lungs are divided into lobes.

Рис. 5. Эмбрион *Nyctalus noctula* CRL 11,8 мм. Правое (a) и левое (b) легкие разделены на доли.

The forming bronchial tree in *Rhinolophus hipposideros* had 3 bronchial buds from the left and 4 ones from the right (fig. 4, b).

The further developments of lungs in *Nyctalus noctula* and *Rhinolophus hipposideros* differed sharply. So, at the 19<sup>th</sup> stage the lungs of *Nyctalus noctula* kept the lobe structure (fig. 5, a, b). The subsequent development of the lungs up to their adult state consisted in branching and lengthening bronchi, enlarging lungs dimensions, as well as differentiating cells of walls of both bronchi and alveoli.

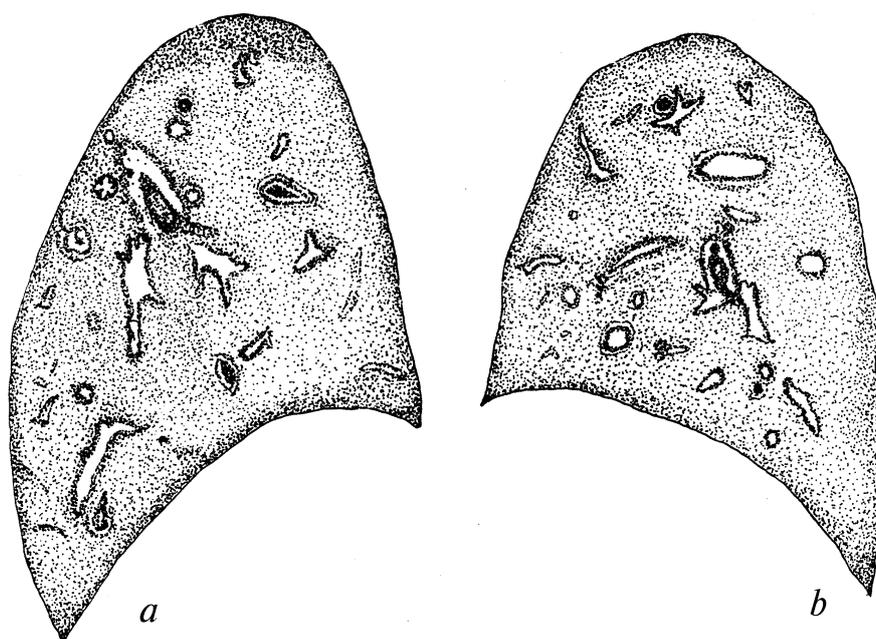


Fig. 6. Embryo *Rhinolophus hipposideros* CRL 12.0 mm. The right (*a*) and the left (*b*) lungs are the semblance of symmetrical undivided into lobes structure.

Рис. 6. Эмбрион *Rhinolophus hipposideros* CRL 12,0 мм. Правое (*a*) и левое (*b*) легкие представлены едиными неразделенными на доли структурами.

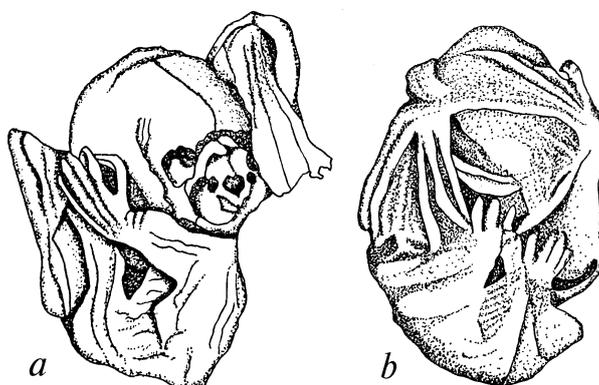


Fig. 7: *a* — embryo *Rhinolophus bocharicus* 20<sup>th</sup> stage; the head of the embryo turned to the left; *b* — embryo *Nyctalus noctula* 20<sup>th</sup> stage.

Рис. 7: *a* — эмбрион *Rhinolophus bocharicus* 20-й стадии, голова повернута влево; *b* — эмбрион *Nyctalus noctula* 20-й стадии развития.

In *Rhinolophus* at the 20<sup>th</sup> stage of development the lungs had no lobe structure being represented as unbroken formations (fig. 6, *a*, *b*).

The lungs in adult horseshoe bats individuals had the aspect of symmetrical undivided into lobes structures. The heart occupied the central position in the thoracic cavity. The symmetry of definitive lungs in horseshoe bats was also observed in weight proportions. So, the left lung and the right one have almost equal weights (tabl. 1).

The lungs of adult plain-nosed bats individuals were found to be divided into lobes, while the top of the heart displaced noticeably to the left. The left lung and the right one had very different weights.

## Discussion

Thus, if judging merely by ramifying a bronchial tree, one can decide that at the 18<sup>th</sup> stage of development the formation of 3–4 lobes of the right lung and 2 lobes of the left one is predetermined in horseshoe bats.

However, not only the bronchial tree determines the development of lungs. It is well-known, that only an epithelial cover and glands of bronchi and bronchioles are developed from the gut epithelial primordium. Forming in a hollow branching bronchial tree, the epithelial primordium grows into the mesenchyme (fig. 2). The latter is condensed around epithelial tubes, being differentiated into the connective tissue and smooth muscles cells, which form walls of bronchi and bronchiole.

Probably, the formation of a more branched bronchial tree in horseshoe bats, caused by the process of forming bronchi and bronchioli, accompanies with the active participation of mesenchyme cells, which fill all kinds of spaces among bronchi of the 1<sup>st</sup> and 2<sup>nd</sup> orders, excluding any their division.

Beginning from the 14 stage of their development, embryos of plain-nosed bats have the head pressed to the cardiac-liver protuberance. The embryos keep such a S-form curved body up to the 19<sup>th</sup> stage (fig. 7, b).

Horseshoe bats have the similar position of the head with respect to the trunk. However, beginning from the 18 stage, the head of the embryo turns to the left, pressing to the left shoulder (fig. 7, a). Such a position of the head keeps by embryos up to the 21<sup>st</sup> stage, and then the head takes the symmetrical position with respect to the trunk.

Such relative disposition of the head with respect to the trunk in horseshoe bats excludes any pressure onto the ventral surface of the trunk, in contrast to plain-nosed bats. It creates conditions of keeping a middle position by the heart in the thorax and does not cause the asymmetrical development of lungs.

The lungs in horseshoe bats are not the secondary unlobed structures realized by means of merging lobes, as other morphologists supposed before.

Differences in the head posture with respect to the trunk may be explained particularly by demands of vocalization during echolocation (Pedersen, 1991, 1993). In the period of the formation of brain, sensoric capsules, and pharynx, the disproportionate growth of these structures takes place, that led up to turning the cranium with respect to *columna vertebralis* in nasal-emitting bats, in particular, in horseshoe ones. Similar changes does not take place in the animals emitting echolocation signals through the mouth, for instance, in oral emitting plain-nosed bats.

- Kovalyova I. M.* Bats evolution in the light of adaptational transformations of the respiratory system // *Myotis*. — 1994–1995. — **32–33**. — P. 9–19.
- Kovalyova I. M.* Features of the development of horseshoe bats (Rhinolophidae) lungs in the prenatal ontogenesis // *Vestn. Belozerkovskogo Derzh. agrarnogo un-tu*. — Bila Zerkva, 1998 a. — **6**, part 1. — P. 154–156. (In Russian).
- Kovalyova I. M.* Features of the prenatal development of lungs in horseshoe bats (Rhinolophidae) // *Vespertilio*. — 1998 b. — N 3. — P. 45–50.
- Kovalyova I. M.* The prenatal development of Lungs in Bats // Abstracts VIII<sup>th</sup> European Bat Research Symposium. — Krakow, 1999. — P. 33–34.
- Kovalyova I. M., Melnik O. P.* The prenatal development of *Nyctalus noctula* (Chiroptera) Lungs // *Nauk. Vestn. Nacional. Agrarn. un-ta*. — Kyiv, 2000. — N 28. — P. 58–61. (In Russian).
- Ledenev S. Yu. & Lichotop R. Yo.* The stages of normal development of *Nyctalus noctula* (Vespertilionidae) // *Preprint Inst. Zool.* — Kyiv : Nauk. Dumka, 1988. — P. 3–15. (In Russian).
- O’Rahilly R.* Guide to the staging of human embryos // *Anat. Anz.* — 1972. — **130**. — P. 556–559.
- Pedersen S. C.* Echolocation and the ontogeny of head posture in Microchiroptera // *Amer. zool.* — 1991. — **31**, N 5. — C. 53.
- Pedersen S. C.* Cephalometric correlates of echolocation in the Chiroptera // *J. Morphol.* — 1993. — **218**, N 1. — P. 85–98.
- Sterba O.* Prenatal development of *Myotis myotis* and *Miniopterus schreibersi* // *Folia Zoologica*. — 1990, — **39** (1). — P. 73–83.
- Sterba O.* Ontogenetic levels in mammals. Evolution and morphogenesis / Ed. J. Mlikovsky, V. J. A. Novak. — Praha : Academia, 1985. — P. 567–571.
- Torubarova L. M.* Materials to zonal anatomy of lungs // *Tr. 2 Ukr. conf. anat., hystol. and ebyrologists.* — Kharkov, 1958. — P. 84–95. (In Russian).
- Zhedenov V. N.* Anatomico-comparative characteristic of lung form and lobe structure within different groups of Mammals // *Tr. Odessk. Agricult. Inst.* — 1957. — **12**. — P. 113–141. (In Russian).