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# THE ULTRASTRUCTURE OF PULMONARY ALVEOLI OF THE ONE – HUMPED CAMEL (CAMELUS DROMEDARIUS)

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## ABSTRACT

*The ultrastructure features of the pulmonary alveoli of the one-humped camel were basically similar to those described in other animals. The alveoli were lined by squamous type-I pneumonocytes interspersed with large irregular or cuboidal type-II pneumonocytes. They were resting on a thin basal lamina. The interalveolar septum was of variable thickness. It contained the gas exchange capillaries, fibroblasts, plasma cells and mast cells together with many collagenous and elastic fibrils. Alveolar macrophages appeared in most alveolar lumen. Intravascular active monocytes were seen.*

## INTRODUCTION

Among our live stock, the camel is considered as a very important animal. The camel had received little attention when compared with other species of animals. Several studies have been made on the lung in various mammalian species ***(Low; 1954, Epling; 1964, Atwal and Sweeny; 1971, Moussa and Heider; 1983, Moussa 1989b, Atwal, Singh, Staempfli and Minhas; 1992, Pries and kuebler; 2006 and Johnson; 2007)***. However, the available data regarding the lung of the camel was deficient ***(Moussa, 1989a)***. Therefore, the present work was intended to describe the fine structural features of the pulmonary alveoli of the one – humped camel (Camelus dromedarius).

## MATERIALS AND METHODS

The lungs of healthy adult two female and five male camels were collected from Cairo slaughter house of 6-8 years age after asking the owners. They were immediately cut into small pieces. For TEM examination, small blocks of lung tissue were immersed is 4% glutraldehyde solution of 0.1 M phosphate buffer (PH 7.2) at 4°C. they were postfixed in 1% osmium tetraoxide solution of 0.1 M phosphate buffer (PH 7.2) at 4°C, dehydrated, cleared and then embedded in epoxy resin ***(Hayat, 1989)***. Thick and thin sections were cut with an ultramicrotome. Sections of 1 um thick were stained with Toluidine blue. Thin sections were stained with uranyl acetate and lead citrate and then examined by electron microscope.

## RESULTS

The alveoli appeared to be lined by thin attenuated membranous pneumonocytes type-I with occasionally scattered pneumonocytes type-II epithelial cells resting on a thin basal lamina (Fig.1). The interalveolar septum was formed of connective tissue of variable thickness containing numerous blood capillaries housing different blood elements; RBCs, leukocytes and blood platelets (Fig.2). Many fibroblasts, macrophages and occasionally plasma and mast cells were encountered. In thin septum, only the basal lamina separated capillaries from the alveolar epithelium.

The type-I pneumonocytes had large, irregular nucleus which was surrounded by a thin rim of cytoplasm which extended to cover most of the alveolar surface. This surface was usually smooth but in some instances small invaginations of plasma membrane were seen especially along its basal surface (fig.3). Except for a few mitochondria and

ribosomes, not many other organelles were in the cell. Numerous minute pinocytotic vesicles, containing an electron translucent material, were evident in some areas of the cytoplasm (Fig.4). The huge number of such vesicles may cause displacing of the epithelial memberane into the alveolar lumen. However some vesicles of large diameter were also encountered.

Type-II pneumonocytes were cuboidal or irregular cells scattered along the epithelial surface forming a continuous epithelial lining with the type-I cells. The junctional area between type-I and type-II cells had the structural characterisitics of tight junction (zonula occludens). The alveolar surface of the type-II cell was distinguished by the presence of irregular microvili. Each cell contained irregular nucleus, the cytoplasm sometimes was condensed around the nucleus to form a thick layer. The cytoplasm usually contained many mitochondria with variable sizes and shapes, a compact Golgi apparatus and rER as well as accumulations of free ribosomes and polysomes (Figs.5 &6). Presence of large osmiophilic bodies which were specific features of type-IIpneumonocyte. These inclusions were randomly distributed within the cytoplasm but they were mostly found on one side of the cytoplasm and had no strict spatial relationship with mitochondria and other organelles. Occasionally some were in close opposition to the surface plasma membrane or even fused with it so that the contents were exposed to the alveolar space. The bodies appeared to consist of osmiophilic membranes; some of them appeared as electron dense homogenous ground that contained highly osmiophilic coarse lamellae. Large number of inclusions showed uniformly heavily osmiophitic nature while others appeared less dense and had fewer lamellae (Figs.6&7). In some sections inclusions showed lighter osmiophilic material accumulated around heavily osmiophilic core.

The capillaries network in the interalveolar septum was very extensive. The capillary had endothelial cells rest on a thin basal lamina. These cells were thin except in the region of their nuclei which protruded into the lumen. These nuclei appeared irregular and elongated with extensive euchromatine (Fig.2). The endothelial cytoplasm was less electrone dense than epithelial cytoplasm. Numerous pinocytotic vesicles were seen in both luminal and basal surfaces of these cells (fig.4). Perinuclear area of endothelial cell contained some organelles like mitochondria, endoplasmic reticulum and free ribosomes. Tight junctions were seen as lateral membrane thickening between adjoining endothelial cells, the intravascular features were blood plasma, RBCs, different leukocytes and blood platelets. In addition, a specific feature of camel's lung was the presence of large intravascular active monocytes. The cells were highly irregular in shape and were encountered through out the blood plasma, some of them had long cytoplasm processes. Their electron lucent cytoplasm was abundant and contained many mitochondria, rER, free ribosomes and many vacuoles. The thin side of the alveolar wall, where diffusion distances for gases are minimal, was formed by the close approximation of capillary endothelium and alveolar epithelium, the two being separated only by the fused basal lamina (Figs.2&8)

In addition to blood capillaries the interstitial tissue of the alveolar wall contained collagen fibers with clear periodicity and elastic fibers embedded in amorphous slightly electron opaque ground substance (Fig.9). Many connective tissue cells were seen; fibroblasts were scattered among the extracellular fibers. This cell had large irregular nucleus, its cytoplasm contained mitochondria, ribosomes and well developed rough endoplasmic reticulum. Many sections, showed a contact between the fibroblasts and type-II pneomoncytes (Figs.7& 10).

Mast cells appeared in some sections with their different morphological granules and with long surface microvilli (Fig.11). Many mitochondria and glycogen deposits were evident. Plasma cells were sparsely found in the interstitial tissue with their characteristic nucleus and Golgi zone (Fig.12). Alveolar macrophages appeared in most alveoli; they were usually seen to be close the alveolar epithelium or may be free in the lumen (Fig.13). These cells had many mitochondria and ribosomes in the form of free polysomes or as rER as well as many vacuoles.

## DISCUSSION

The lung alveoli of one humped camel (Camelus dromedarius) showed nearly the same ultrastructural features as those described in other mammals. Our findings came in agreement with many authors in that the pulmonary alveoli were lined by pneumonocytes type-I and type–II alveolar epithelial cells resting on a basal lamina . Beneath this basal lamina there were capillaries and connective tissue of different thickness (***Low; 1954, Karrier; 1956, Epling; 1964 Atwal and Sweeny; 1971, Rybicka, Daly, Migliore and Norman; 1974, Moussa and Heider; 1983, Moussa; 1989a, El-Nashar, Abd El Moneim and Saber; 2001 , Kelleny ; 2005 and Ibrahim , 2006)***.

The air alveoli, as we mentioned, were covered by membranous pneumonocytes (type-I) and granular pneumonocytes (type-II) which were joined by tight junctions. The membranous (agranular) pneumonocytes was the primary constituent of the alveolar lining; it was an endothelial like cell with attenuated cytoplasm it appeared with extending cytoplasm sheet to cover larger areas potentially available for gas exchange. Thus the susceptibility to be damaged by many inhaled

agents was greater than pneumonocytes type-II ***(Kuhn; 1978, Kawanami, Ferrans and Crystal; 1982 and Moussa; 1989a)***. ***Weibel; (1974), Kauffman , Burri and Weibel; (1974)***, ***Vaccaro and Brody; (1981) and Kawanami et al. (1982)*** stated that pneumonocytes type-II replace the damaged type-I cells by a process of spreading out of their cytoplasm and disappearance of their osmiophilic lamellar bodies. This was supported by the opinion of Evans, ***Cabral and Stephens ,(1973)*** in that the pneumonocytes type-I unable to undergo mitotic division . The process of compensation of type-I cell by means of type-II pneumonocytes was supported by experiments involving epithelial repair after exposure to toxic concentration of oxygen ***(Kapanci, Weibel, Kaplan and Robinson; 1969 and Gould, Tosco and Wheelis; 1972)***. Regarding the ion transport, the general theory of ion and fluid transport in the lung was that the alveolar type-IIcells known to contain ion channels governed ion transport and that the type-I cells believed to be incapable of ion transport, (only allowed passive movement of water); in this respect recent investigators demonstrated that type-I cells were capable of ion transport and played a role in regulating lung fluid balance ***(Johnson, Bao, Helms, Chen , Tigue, Jain, Dobbs and Eaton, 2006 and Johnson, 2007)***.

In the present work, type-II pneumonocytes were of variable occurrence; the number being vary from one to several per alveolus. They present where the alveolus walls unite and form angles. They were cuboidal or rounded cells which intermittently line the alveolar surface. Their cell bodies appeared foamy and project into the alveolar lumina. Their microvilli and osmiophilic lamellar bodies were the diagnostic features of these cells at the electron microscopic level. Similar findings were described in camel ***(Moussa, 1989a)***, in buffaloes (***Moussa and***

***Heider, 1983 and Moussa, 1983)***, in lambs ***(Moussa, 1989b)***, in goat ***(Atwal and sweeny , 1971 and kahwa, Atwal and Purton, 1997)*** and in rat ***(El Nashar et al 2001, Kelleny, 2005 and Schmiedl, Vieten, Hühfeld and Bernhard, 2007)***.

In the present investigation large number of mitochondria was encountered in camel type-II pneumonocyte . ***Tyler and Pearse, (1965)***, ***Said , Kiein, Norell and Maddox, (1966)*** and ***Said, Harlan, Burk and Elliot (1968)*** , have pointed out that the huge number of mitochondria reflect a high level of oxidative metabolic activity in their cytoplasm; moreover the glycolytic and the hexose monophosphate pathway enzymes may be another important source of energy and may play a role in the synthesis of pulmonary phospholipids and fatty acids. According to ***Hoffman (1972), Gill and Reiss(1973) and Kuhn (1978)*** the osmiophilic inclusions were considered as a storage form of surfactant due to their high content of phospholipids mainly dipalmitoyl phosphatidycholine; in this respect ***Clements and King (1976), Collet and Chevalier (1977), Meyrick and Reid (1977), Weibel and Gil (1977) and Junqueira and Carneiro (1983)***, demonstrated that the osmiophilic lamellar bodies contain a concenterate of the disaturated phospholipid dipalmitoyl lecithin; the same material that has been isolated from the alveolar surface lining. ***Banks (1981) and Schmiedle et al, (2007)***, reported that surfactant was a detergent – like material which reduces the alveolar surface tension and prevents alveolar collapse during expiration. Surfactant also facilitates the transport of gases between air- liquid phases in addition it has a bactericidal effect which aids in the removal of potentially dangerous bacteria that reach the alveoli (***Junqueira and Carneiro; 1983 and Gehr, Im Hof, Geiser and Schürch; 2000)***. The last authors added that surfactant is considered as primary immune barrier.

A contact between type-II pneumonocytes and interstitial cells, mostly fibroblasts and to some extent with mast cells, was seen in the examined sections of camel's lung; this was compatible with the results of ***Moussa, (1989a), Weibel , Gehr, Haies, Gil and Bachhofen, (1976) and Weinstein , Hogg, Nash and Mcnutt, (1977)***, They have pointed out that a fibroblastic pneumonocyte factor released by the fibroblast is necessary to enhace the differentiation and increase the production of surfactant by type-II cells

The present study revealed that, the features of the interalveolar septum of the camel were similar to those of ***Moussa, 1989a*** (in one humped camel), ***Moussa and Heidar, 1983*** (in buffalo) and ***Atwal and Sweeny, 1971*** (in Goat). The capillaries were arranged mostly in the centre of the interstitum; some bulge into the alveolar lumen. A close contact, between basement membrane of both endothelium of capillary and epithelium of alveolus, was apparent in some areas of the septum. Similar observations in which a thin blood gas barrier has been shown on both sides of a centrally located capillary by ***low (1952, 1961) Vaccaro and Brody (1981), Moussa (1989a) and Pries and Kuebler (2006)***.

This study revealed many plasma membrane invaginations and pinocytotic vesicles in both epithelial and endothelial cell, in this respect many authors have pointed out that the capillary endothelial invaginations have been identified as morphologic evidence of pinocytosis ***(Karnovsky ;1967, Bruns and Palade; 1968 and Rybicka et al. 1974)***. They may play an important role in fluid transfer across the cell ***(Schneeberger and Karnovsky, 1971 Gonzalez – Crussi and Boston***

***1972)***. ***Dermer, (1970)*** and ***Rybicka et al, (1974)*** mentioned that the epithelial pinocytosis, on the other hand, may serve mainly in the removal of surfactant in its transport from the lung to the lymphatic system.

The examined camel's lung in the present study showed many connective tissue cells, specially fibroblasts, in the interalveolar wall as it was also described by ***Weibel et al. (1976); Kuhn, (1978)*** and ***Moussa (1989a)***. They mentioned that fibroblasts directly influence alveolar structure simply by their abundance. Also fibroblasts regulate gas exchange efficiency by affecting capillary diameter. Therefore the air and blood flow was regulated at the alveolar level through their contractile elements ***(Kapanci, Assimacopoulos, Irle, Zwahlen and Gabbiani, 1974)***. In agreement with ***Moussa (1989 a&b)*** in camels and lambs respectively and ***Atwal and Sweeny (1971)*** in Goat; mast and plasma cells were sparsely present in the interstitium. Degranulation of mast cells release histamine which acts as mediator of increased capillary permeability which is associated with the adult respiratory distress syndrome ***(Wilson, 1974)***. The study revealed presence of active intravascular monocytes which appeared irregular in its outline with cytoplasmic processes. The characteristic features of these cells made us to name it macrophage-like cells. ***Atwal et al., (1992)*** demonstrated that the equine lung has pulmonary intravascular macrophages as resident phagocytes in its microvasculature. Immunocytochemistry showed more pulmonary intravascular monocytes / macrophages in rats infected with E.Coli compared with the control group ***(Charavaryamath, Janardhan, Caldwell and Singh, 2006)***.

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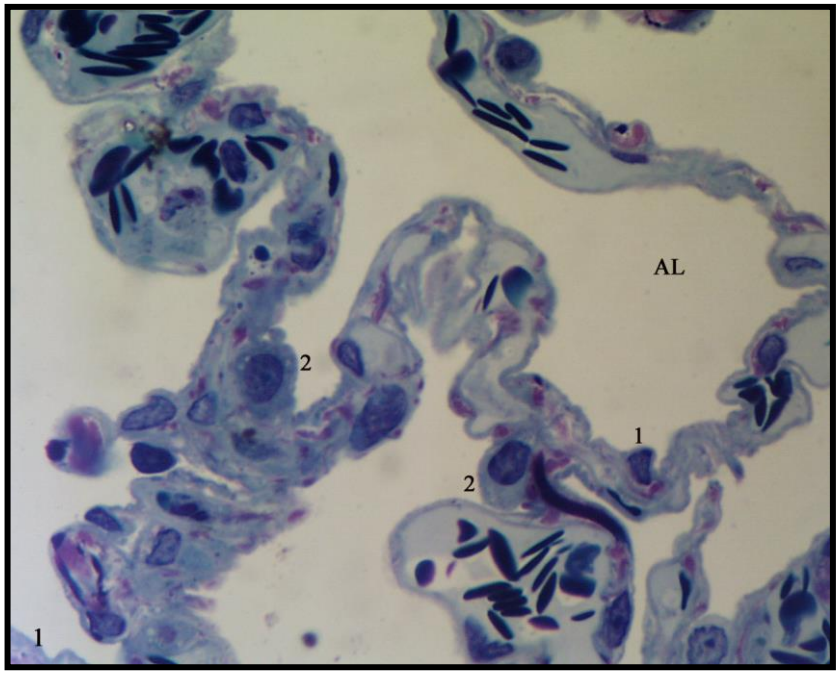
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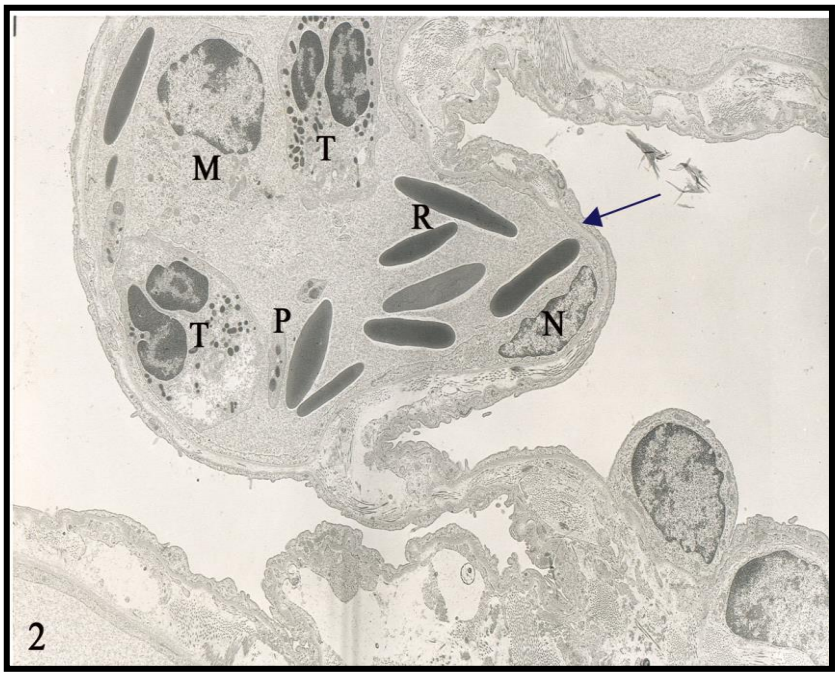
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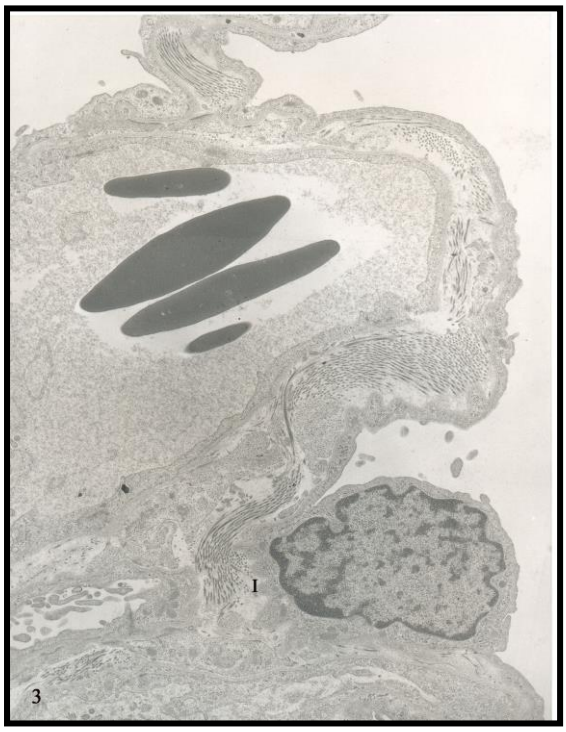
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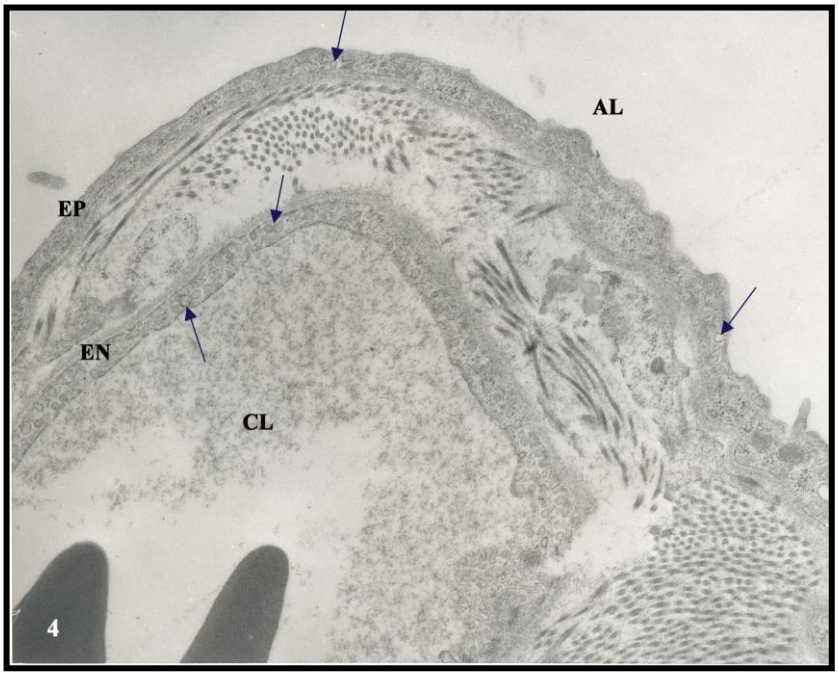
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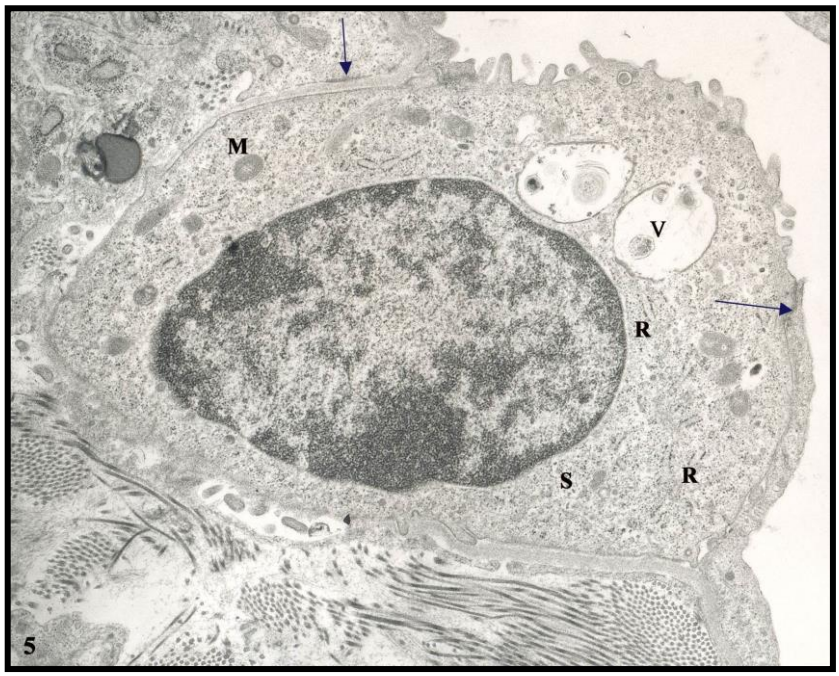
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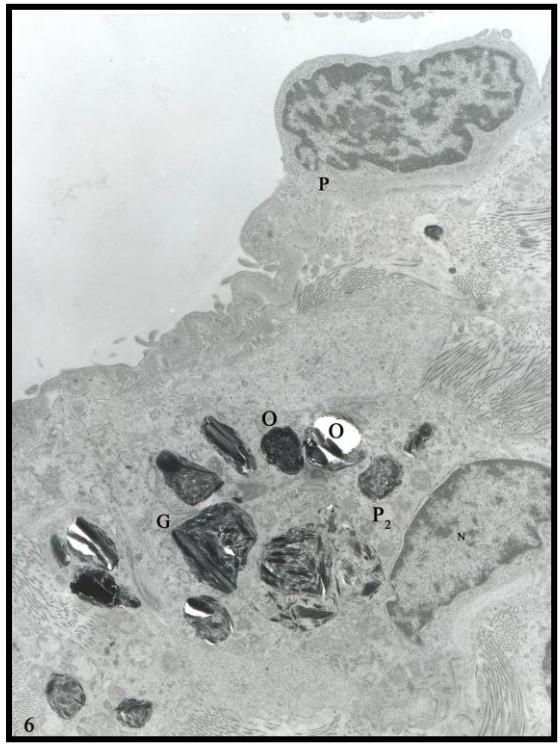
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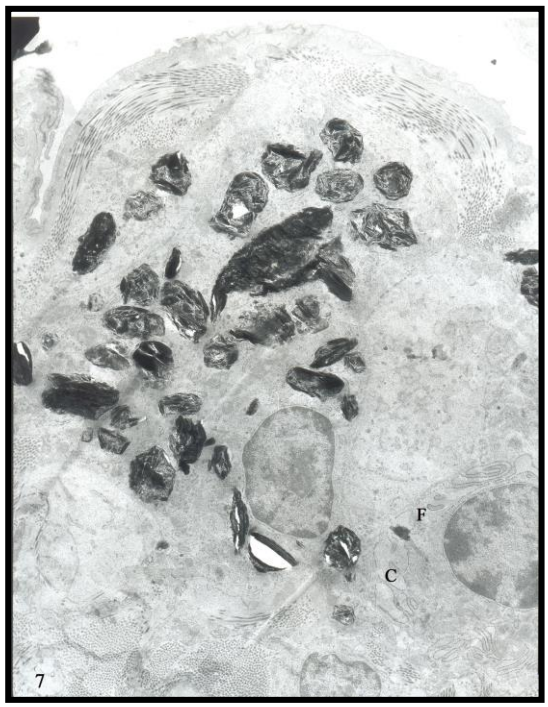
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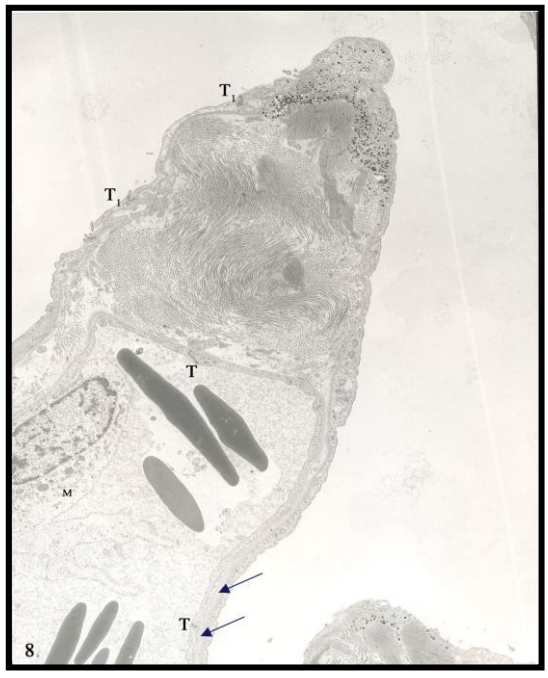
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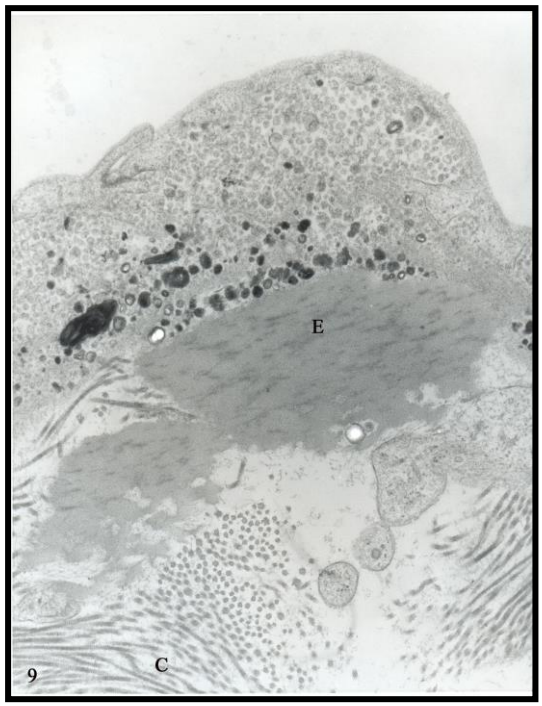
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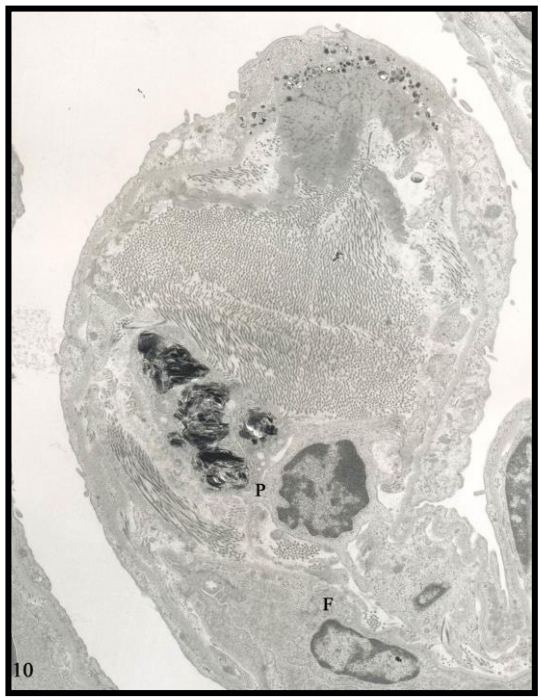
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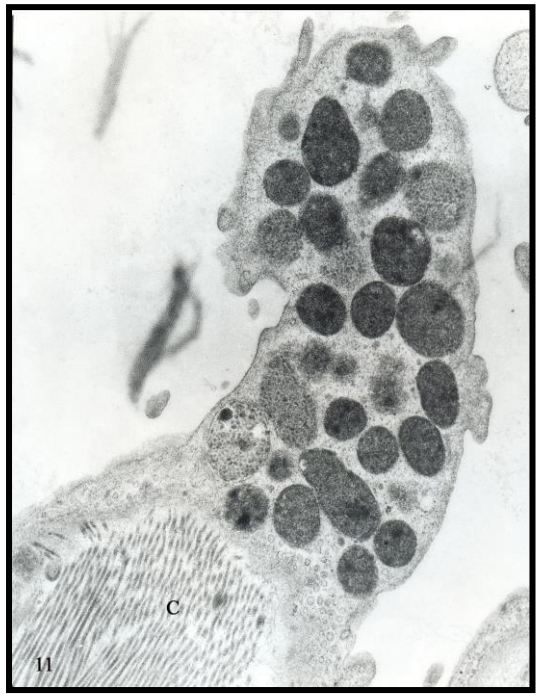
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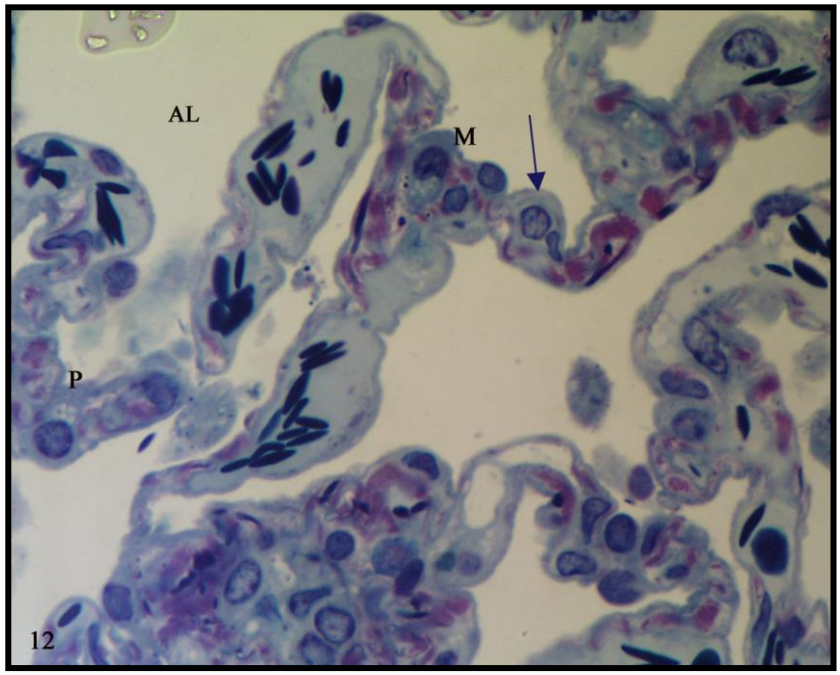
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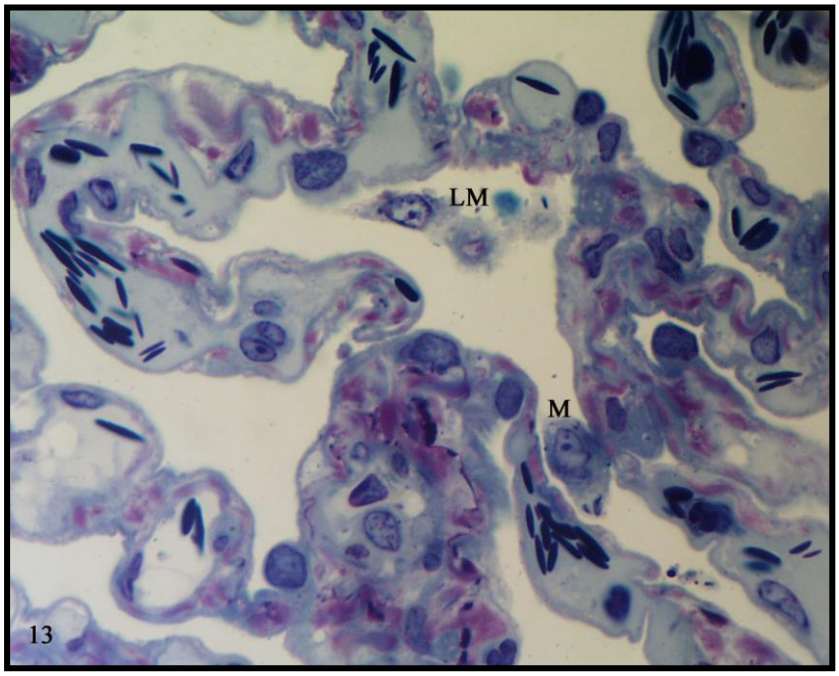


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## التركيب الدقيق للاسناخ الرئوية في الجمل وحيد السنام (الجمل الدروميديري)

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تتشابه صفات التركيب الدقيق للاسناخ الرئويه في الجمل وحيد السنام مع التركيب الدقيق الموصوف في الحيوانات الأخرى حيث تتكون الخلايا المبطنة للاسناخ من نوعين من الخلايا: النوع

الأول هو الخلايا السنخية الحرشفية البسيطة (خلايا الرئة – I) و هي خلايا مسطحة للغاية ذات نواه غير منتظمة والسيتوبلازم خارج منطقة النواه يكون رقيق جدا. والنوع الثاني من الخلايا هو عبارة عن خلايا مكعبة سنخية (خلايا الرئة – II)، وهي خلايا تتميز باحتوائها علي اجسام صغيرة محبة لحمض الازميك وتحمل علي سطحها خملات دقيقة.

وللاسناخ غشاء قاعدي رقيق مشترك مع الغشاء القاعدي للشعيرات الدموية الموجودة في النسيج البيني بين الاسناخ أو بالقرب منه. يتميز هذا النسيج البيني باحتوائه علي ألياف غروية ومرنة بالإضافة إلي وجود خلايا ليفية، خلايا بلازمية وبلاعم. هذا وقد لوحظ وجود بعض البلاعم الطليقة في تجاويف معظم الاسناخ الرئوية.