FUNCTIONAL MORPHOLOGY OF RESISTIVE VESSELS AND CAPILLARIES OF THE LUNGS DURING ADAPTATION OF SPECIES AND INDIVIDUAL TO HIGH ALTITUDES

T. K. Kadyraliev and M. M. Mirrakhimov

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At high altitudes in the mountains the hypoxic form of pulmonary hypertension develops: it is based on spasm of the pulmonary arterioles, which maintains the regional distribution of blood and reduces the degree of arterial hypoxemia. Meanwhile an excessive rise of the pulmonary arterial pressure under these conditions may lead to high-altitude pulmonary edema or subsequent hypertrophy of the right ventricle, with the development of heart failure. The mechanisms lying at the basis of pulmonary arteriolar spasm have not yet been adequately explained [2-4].

The aim of this investigation was to study adaptive structural changes in the resistive vessels and capillaries of the lungs during specific and individual adaptation of man and animals to high altitude conditions.

EXPERIMENTAL METHOD

The effect of individual adaptation to high altitudes was studied in the experiments of series I. The lungs and heart of 62 noninbred male rabbits weighing 2.5-3 kg were studied after adaptation to high altitudes for 3, 7, 15, 30, 60, and 90 days. The lungs and heart of 18 rabbits, living in the foothills (760 m above sea level) served as the control. In series II morphological and functional changes in the pulmonary vessels and capillaries of animals indigenous to high altitudes were investigated, by studying the lungs and heart of 26 male yaks weighing 250-300 kg and 11 mountain goats weighing 50-60 kg. The results of a study of the identical organs of bulls and domestic goats of the same weight, living in the foothills, served as the control. Bulls and domestic goats were chosen as the control animals not by accident. Yaks and bulls are members of the genus *Bos,* whereas mountain and domestic goats belong to the genus *Capra.* The lungs and heart of indigenous inhabitants of the mountains and foothills, dying accidentally, also were studied. The former were nine, the latter five males aged from 18 to 36 years. Their average ages were comparable. Comparative biometry of the right ventricle was carried out by weighing parts of the heart separately by Muller's method in G. S. Kryuchkova's (1972) modification. Lung tissue for light microscopy was fixed by injecting 10% neutral formalin into the main bronchus. In sections stained with picrofuchsine-fuchselin the area of the media and of the lumen of the terminal arteries and arterioles was determined in three zones (lobes) of the right lung. The total number of arterioles and alveoli was counted and the concentration of peripheral arterioles, i.e., the ratio of arterioles to alveoli, was determined, starting from the number of arterioles per 100 alveoli. The area of the endothelial cells and of their nuclei in the large branches of the pulmonary artery was determined in two-dimensional membrane preparations, stained by silver impregnation and by hematoxylin-eosin. Tissue samples for ultrastructural investigation were fixed in 2.5% glutaraldehyde in phosphate buffer (pH 7.4). The material was dehydrated and embedded in a mixture of Epon and Araldite by the well-known methods. Ultrathin sections were examined in the

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TABLE 1. Morphometric Parameters of Pulmonary Arterioles 40-100 μ in Diameter in Rabbits During Adaptation for 3 Months to High Altitudes, Indigenous Mountain Dwellers and Inhabitants of the Foothills, and for Comparison, of Animals Living in the Mountains and Foothills (M ± m)

Lobe of lung	Rabbits		Mountain dwellers	Foothill dwellers	Yaks	Bulls	Mountain goats	Domestic goats
	control	90th day	Area of media, μ^2					
Upper	816±76	1837±112*	2041±156*	408±64	612±74*	1338±76	613±34*	1236±127
Middle	832±74	1756±132*	2134±132*	583±67	617±66*	1256±96	597±76*	1338±135
Lower	874±97	2037±126*	2178±147*	497±76	796±74*	1633±78	689±77*	1631±167
			Area of lumen, μ^2					
Upper	1837±76	1647±131*	2245±173*	2789±76	3625±194*	2267±271	2247±163*	2057±127
Middle	1793±112	1563±171*	2257±143*	2897±69	3576±136*	2365±176	2371±171	2371±134
Lower	1758±87	1429±132*	2178±143*	2567±73	3697±172*	2287±134	2131±137*	1973±121

Legend. Here and in Tables 2 and 3: *$p < 0.05$.

TABLE 2. Changes in Components of Air–Blood Barrier in Rabbits During Adaptation for 3 Months, Compared with Indigenous Species of Animals and Man at High Altitudes (μ)

Components	Rabbits		Mountain dwellers	Foothill dwellers	Yaks	Bulls	Mountain goats	Domestic goats
	control	90th day	Area of media, μ^2					
Endothelium	0.25±0.01	0.15±0.03*	0.4±0.02	0.4±0.01	0.15±0.02*	0.23±0.02	0.15±0.03*	0.25±0.04
Epithelium	0.3±0.01	0.1±0.01*	0.3±0.03*	0.5±0.02	0.1±0.02*	0.21±0.01	0.13±0.01*	0.3±0.02
Basement membranes	0.1±0.02	0.2±0.01*	0.6±0.01*	0.3±0.01	0.1±0.04	0.1±0.04	0.1±0.02	0.15±0.03
Total thickness	0.65±0.01	0.45±0.01*	1.3±0.02	1.2±0.01	0.35±0.03*	0.54±0.01	0.38±0.02*	0.7±0.03

TABLE 3. Changes in Diameter of Capillaries of Rabbits during Adaptation for 3 Months Compared with Indigenous Species of Animals and Man at High Altitudes (μ)

Lobe of lung	Rabbits		Mountain	Foothill	Yaks	Bulls	Mountain goats	Domestic goats
	control	90th day	Area of media, μ^2					
Upper	7±0.7	11±0.6*	13±0.8*	9.3±0.4	16±0.3*	10.5±0.9	14.2±0.4*	8.7±0.5
Middle	8.1±0.2	10±0.3*	12±0.7*	10±0.3	17±1.2*	9±0.7	12±0.7*	7±0.5
Lower	7±0.6	10±0.7*	12±0.9*	9±0.6	15±0.9*	10±1.3	14±1.1*	8.1±0.9

PÉM-100 electron microscope. Morphometry of the cell ultrastructures was assessed in accordance with the principles of stereology. For morphometry of the components of the air–blood barrier (ABB) negatives were used under magnification of 20,000, 25 measurements being made on each block.

EXPERIMENTAL RESULTS

During adaptation to high altitudes for 3 months the experimental animals developed functional and morphological changes in the pulmonary resistive vessels and alveolar capillaries, reflecting the new level of the pulmonary circulation. These changes were expressed by the development of hypertrophy of the right ventricle: the ventricular index of the rabbits rose to 0.57 ± 0.02 (0.36 ± 0.03 in the control). Dilatation of the lumen of the terminal and intralobular arteries was observed in all zones of the right lung, but at the same time an increase in the thickness of the media also was noted. For instance, in the upper lobe of the right lung the area of the media of the terminal arterioles was 4576 ± 147 μ^2 (2338 ± 173 μ^2 in the control). As regards arterioles 50-70 μ in diameter marked thickening of the media and considerable reduction of their lumen were observed (Table 1).

Electron-microscopic investigation revealed considerable changes in ABB ultrastructure. In the initial stages of adaptation the basement membranes of ABB became thinner and ruptured, releasing erythrocytes into the lumen of the alveoli (Fig. 1b). In the stable phase of adaptation (60-90 days) the working zones of ABB became enlarged, with simultaneous thickening of the basement membranes (Fig. 1c; Table 2). Under these conditions the endothelial cells of the pulmonary arteries and alveolar capillaries also underwent some significant changes. In large pulmonary

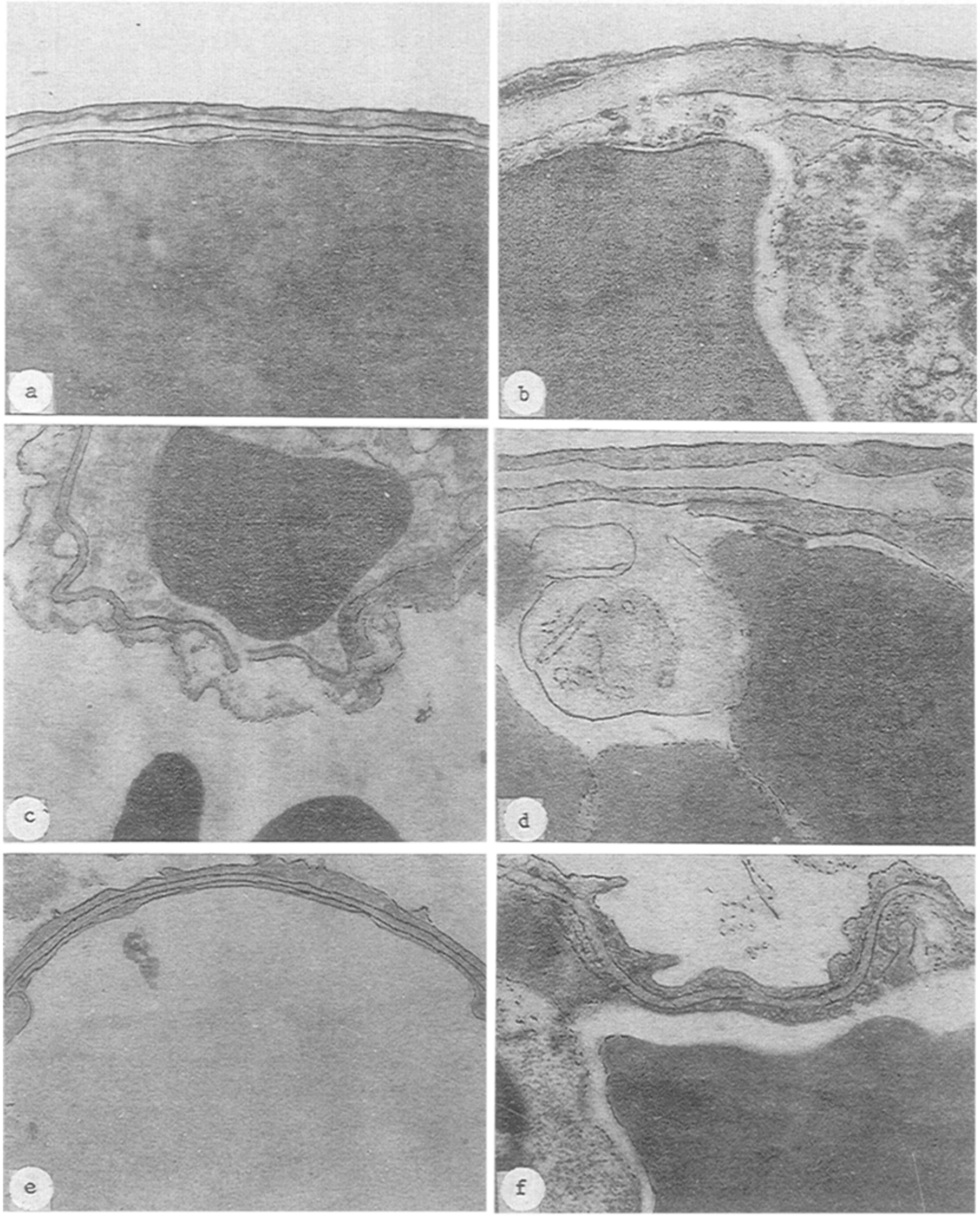

Fig. 1. Changes in air–blood barrier during specific and individual adaptation to high altitude conditions: a) air–blood barrier of control rabbit (20,000×); b) rupture of basement membranes of air–blood barrier of rabbits on 7th day of adaptation (release of erythrocytes into alveolar lumen, vacuolation and lysis of cytoplasm of endothelium and of type I alveolocytes (20,000×); c) air–blood barrier on 60th day becomes shaped like an elongated arch (20,000×); d) air–blood barrier of mountain dwellers is characterized by thickening of basement membranes and thinning of type II alveolocytes (20,000×); e) air–blood barrier of inhabitants of the valley (20,000×). f) Air–blood barrier of yak is characterized by thin basement membranes (20,000×).

arteries over 2000 μ in diameter an increase was observed in the surface area of the endothelial cells to 1378 ± 76 μ^2 (651 ± 66.4 μ^2 in the control) and in the area of the nuclei to 841 ± 37 μ^2 (239 ± 17 μ^2 in the control). Morpho-

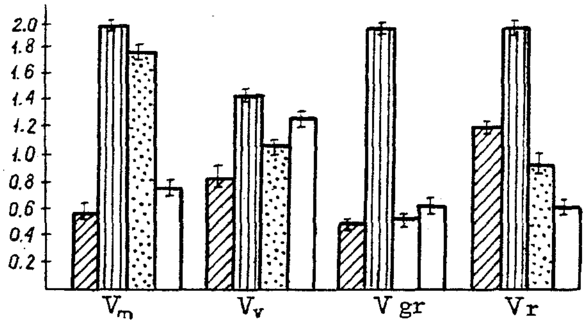

Fig. 2. Morphometric analysis of alveolar capillaries of rabbits during adaptation to high altitudes, and of mountain goats, compared with domestic goats. Oblique shading – control, vertical shading – 90 days of adaptation, dotted columns – mountain goats, unshaded – domestic goats.

metric analysis of the ultrastructure of the endothelium of the alveolar capillaries revealed an increase in bulk density of the mitochondria, ribosomes, structures of the endoplasmic reticulum, and granules (Fig. 2).

Characteristic differences, classed as adaptive, were discovered in the terminal arteries and arterioles of the pulmonary resistive vessels in indigenous mountain animals (yaks and mountain goats), namely a thin media and large lumen. The ratio of the total thickness of the wall to the lumen was 1:14 (1:8 in the control), a characteristic feature of vessels with low pulmonary vascular resistance. The capillary bed of the lungs in yaks and mountain goats resembled distended loops or long channels with enlarged caliber (Fig. 3d; Table 3). Parameters of the alveolar capillaries, allowing for the large volume of the lung parenchyma, indicate an increased capacity of the capillary bed. The endothelial cells of the large pulmonary arteries, arterioles, and alveolar capillaries of animals indigenous to high altitudes showed hypertrophy, as also did the endothelial-cells of the experimental animals, as a result of an increase in bulk density of the mitochondria, of structures of the rough endoplasmic reticulum, ribosomes, and secretory granules (Fig. 2). The ABB of yaks and mountain goats is distinguished by thinness of its components (Fig. 1f; Table 2), indicating maximal facilitation of gas diffusion.

Identical adaptive changes observed during these experimental studies could be found also in the pulmonary resistive vessels and alveolar capillaries of the indigenous mountain dwellers. Hypertrophy of the right ventricle was found in indigenous mountain dwellers living at an altitude of 2500-3600 m above sea level, and their ventricular index rose to 0.67 ± 0.3 (0.41 ± 0.3 in the control). In the upper and middle zones of the right lung of the mountain dwellers a tendency was observed for the lumen of the intralobular and terminal arteries to widen, but in this case the area of the media was increased also. In the upper zone (lobe) the area of the media of the terminal arteries was 6276 ± 123 μ^2 (5247 ± 154 in the control), whereas the area of the lumen was 12,697 ± 113 μ^2 (10,247 ± 154 in the control). In individual pulmonary arteries with a diameter of 100-500 μ, the longitudinal muscular layer was developing, and it delayed the dilatation of their lumen a little on account of "cushion" compression (Fig. 3a). In the lower zone (lobe) growth of the media of the terminal arteries was observed (6687 ± 214 μ^2 compared with 5279 ± 126 μ^2 in the control) without any appreciable dilatation of their lumen. Arteries 40-70 μ in diameter, formed by arterialization of the pulmonary capillaries, appeared in all zones (lobes) of the lungs, i.e., hyperplasia of the smooth-muscle cells of the media took place, with the appearance of an outer elastic layer (Table 1). The concentration of these arteries in all zones was 12 per 100 alveoli (three arterioles to 100 alveoli in the control). Proliferation of connective tissue took place around the adventitia. The diameter of the capillaries was increased. Most capillaries projected into the lumen of the alveoli and formed the boundary of the lumen of two alveoli (Fig. 3c; Table 3). Investigation of the components of ABB revealed thickening of the basement membranes and thinning of the cytoplasm of the type I alveolocytes (Fig. 1; Table 2). We interpreted thickening of the basement membranes as an adaptive process, aimed at increasing the strength of the capillaries should there be an excessive increase of intracapillary pressure. An increased number of ribosomes and polysomes could be seen in the pericytes of the capillaries and precapillaries of the lungs, together with large mitochondria. It can be tentatively suggested that the increase in functional activity of

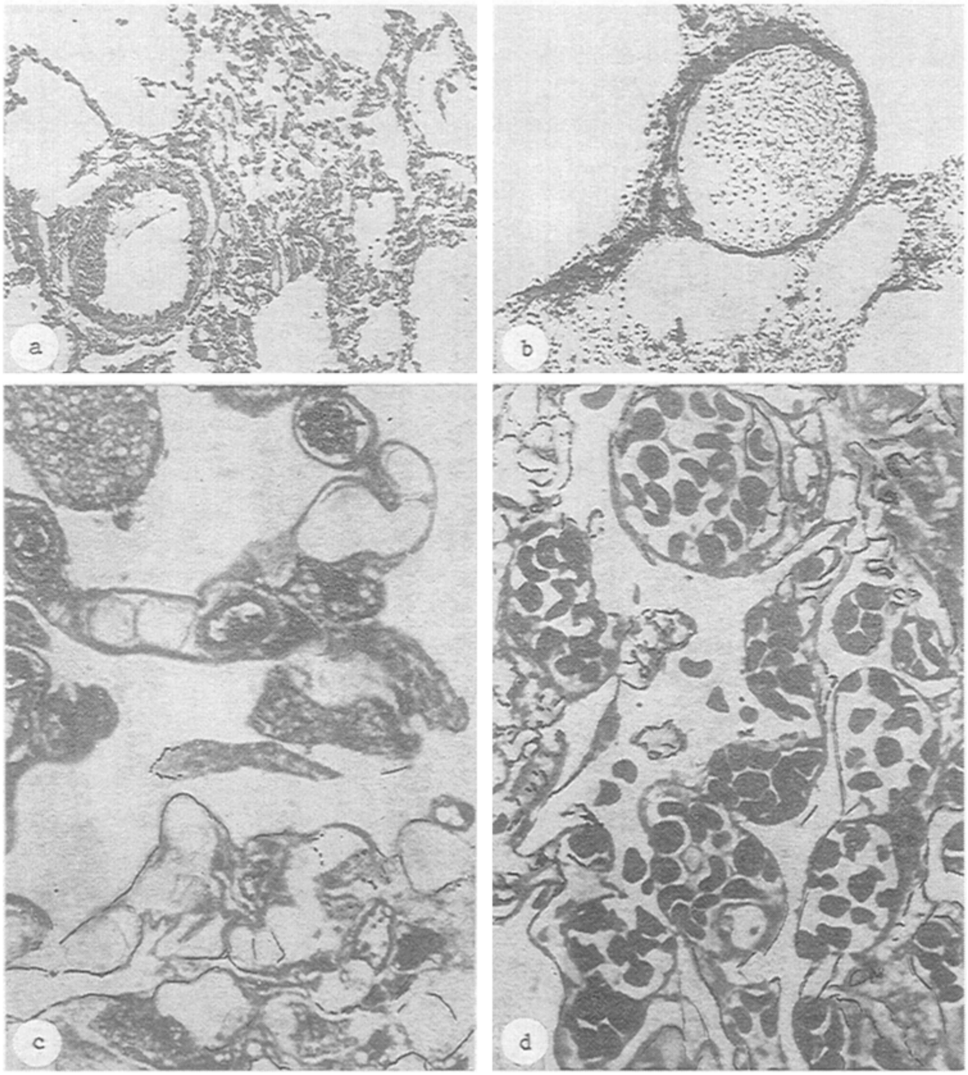

Fig. 3. Morphological changes in pulmonary resistive vessels and alveolar capillaries of permanent mountain dwellers and of animals indigenous to high altitudes: a) small pulmonary artery of a mountain dweller (Murgab). Development of longitudinal muscular layer can be seen. Picrofuchsine-fuchselin, 320×; b) pulmonary arteriole of yak. Vessel wall consists mainly of elastic fibers. Picrofuchsine-fuchselin, 320×; c) pulmonary capillaries of mountain dwellers are characterized by formation of wide capillary loops. Toluidine blue, 900×; d) pulmonary capillaries of mountain goat form dense capillary network. Capillaries of large caliber are filled with erythrocytes. Large pericytes visible in capillary wall. Toluidine blue, 900×.

the pericytes reflects their more intensive biosynthesis of the material of the basement membranes of the pulmonary capillaries. The endothelial cells of the pulmonary vessels and capillaries of the indigenous mountain dwellers also underwent hypertrophy (Fig. 4). In connection with the discovery in recent years of a new metabolic function of the lung endothelium, namely that these cells are involved in the metabolism of catecholamines, prostaglandins, adenine nucleotides, peptides, prostacycline, and so on, hypertrophy of the pulmonary endothelium can be regarded as the morphological manifestation of the strengthening of their metabolic activity, aimed at local regulation of vascular tone in response to the new level of the pulmonary circulation.

These investigations thus enable a differential approach to be made to the evaluation of adaptive changes taking place at the morphological and functional level in the pulmonary circulatory system.

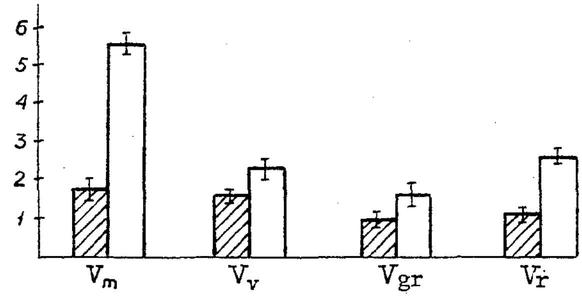

Fig. 4. Morphometric analysis of ultrastructure of alveolar capillary endothelium of permanent inhabitants at high altitudes in the mountains compared with inhabitants of Bishkek. Shaded columns indicate control, unshaded columns – mountain dwellers.

It can be asserted that the evolutionary and adaptive changes which we have described in animals indigenous to mountain conditions give a more reliable protective effect than the acquired adaptive changes which we observed in experimental animals and in the indigenous population of mountain dwellers.

In indigenous animals, for instance, during evolutionary adaptation the most complete structural changes took place in the pulmonary resistive vessels and capillaries, ensuring maximal facilitation of the diffusion capacity of the lungs through hypervolemia, low pulmonary vascular resistance, an increase in capillary volume, and an increase in area and decrease in thickness of the working zones of ABB. In the experimental animals and indigenous mountain dwellers the adaptive changes which we found frequently border on deviations from the normal, in the form of a marked increase in pulmonary vascular resistance, followed by hypertrophy of the right ventricle and thinning of the basement membranes of ABB. This reduction in the thickness of the basement membranes of ABB discovered in indigenous mountain dwellers, in our opinion, is a protective-adaptive phenomenon against the development of pulmonary edema.

Adaptive changes in the wall of the pulmonary vessels and alveolar capillaries during specific and individual adaptation of man and animals to high-altitude hypoxia include hypertrophy of the pulmonary endothelium, a reflection of the intensification of its metabolic activity, which is evidently aimed at the local regulation of vascular tone.

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