Rat pulmonary circulation after chronic hypoxia: hemodynamic and structural features

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Rabinovitch, Marlene, Walter Gamble, Alexander S. Nadas, Olli S. Miettinen, and Lynne Reid. Rat pulmonary circulation after chronic hypoxia: hemodynamic and structural features. Am. J. Physiol. 236(6): H818-H827, 1979 or Am. J. Physiol.: Heart Circ. Physiol. 5(6): H818-H827, 1979.—In 55 Sprague-Dawley rats (mean wt, 277 ± 6.2 g) exposed to hypobaric hypoxia (air at 380 mmHg), and 23 weight-matched controls kept in room air, pulmonary and systemic artery pressures were measured daily for 2 wk via indwelling catheters. After each day of exposure, 1 or 2 hypoxic rats, to a total of 20, and 5 control rats were killed during the experiment. In these rats, the pulmonary arterial tree was injected post mortem with barium-gelatin and inflated with formaldehyde solution, and three structural features were quantified microscopically: 1) abnormal extension of muscle into peripheral arteries where it is not normally present (EMPA); 2) increased wall thickness of the normally muscular arteries, expressed as a percentage of external diameter (%WT); and 3) reduction in artery number expressed as an increase in the ratio of alveoli to arteries (A/a). Mean pulmonary artery pressure (Ppa) rose significantly after day 3 of hypoxic exposure (P < 0.05) and had doubled by day 14; the mean systemic artery pressure (Psa) of hypoxic rats and Ppa and Psa of control rats were unchanged. The level of Ppa correlated with the degree of structural changes; for EMPA, r = 0.84; for %WT, r = 0.64; and for A/a, r = 0.73 (P < 0.001 in all).

The structural changes produced by chronic hypoxia in the pulmonary vascular bed are most apparent in the small and peripheral pulmonary arteries (1–3, 6, 9, 10, 12, 13, 16, 22). In people living at 12,000 ft, abnormally increased muscularization of the small peripheral arteries is seen (3). In cattle native to high altitude and in those brought to high altitude that develop pulmonary hypertension, medial hypertrophy and intimal change is observed in arteries at all levels, but particularly in the small and peripheral ones (2). Mice exposed to chronic hypobaric hypoxia are observed to have increased medial mass in arteries less than 80 μm (16), in arteries accompanying terminal and respiratory bronchioles (13), and even in those associated with alveolar ducts and walls (12). Rats after similar hypoxic exposure periods are demonstrated to have a generalized increase in wall thickness in "small peripheral arterioles" (1), doubling of medial wall thickness in arteries 50–100 μm (22), and an increase in the proportion of muscular alveolar duct and wall arteries (12). The most detailed and quantitative morphological study of the peripheral pulmonary arteries after chronic hypoxic exposure was carried out by Hislop and Reid (9), in rats.

THE STRUCTURAL CHANGES produced by chronic hypoxia in the pulmonary vascular bed are most apparent in the small and peripheral pulmonary arteries (1–3, 6, 9, 10, 12, 13, 16, 22). In people living at 12,000 ft, abnormally increased muscularization of the small peripheral arteries is seen (3). In cattle native to high altitude and in those brought to high altitude that develop pulmonary hypertension, medial hypertrophy and intimal change is observed in arteries at all levels, but particularly in the small and peripheral ones (2). Mice exposed to chronic hypobaric hypoxia are observed to have increased medial mass in arteries less than 80 μm (16), in arteries accompanying terminal and respiratory bronchioles (13), and even in those associated with alveolar ducts and walls (12). Rats after similar hypoxic exposure periods are demonstrated to have a generalized increase in wall thickness in "small peripheral arterioles" (1), doubling of medial wall thickness in arteries 50–100 μm (22), and an increase in the proportion of muscular alveolar duct and wall arteries (12). The most detailed and quantitative morphological study of the peripheral pulmonary arteries after chronic hypoxic exposure was carried out by Hislop and Reid (9), in rats. After injecting the pulmonary arterial tree with a barium-gelatin mixture and then inflating the lung with formaldehyde solution, they recognized three structural changes that could each be quantified and that increased in severity with the continuing exposure of the rats to hypoxia. These features were 1) extension of muscle into smaller and more peripheral arteries than is normal, 2) an increase in the thickness of the medial muscular coat in the normally muscular arteries, and 3) a reduction in the number of peripheral arteries.

Little is known of the correlation between the progressive alteration of the structure of the pulmonary arterial bed and the hemodynamic features of increasing pulmonary artery pressure and resistance. In rats, only mean right ventricular pressures (1, 22) obtained in anesthetized animals have been correlated with a generalized increased thickness of the medial muscular coat of the small arteries.

The object of our study was, therefore, to correlate in the rat the progressive pulmonary arterial changes described by Hislop and Reid (9) to the hemodynamic features of the circulation, measured via indwelling pulmonary artery and systemic artery catheters.

METHODS AND MATERIALS
Fifty-five rats, Sprague-Dawley (cesarean-derived), were maintained under conditions of chronic hypobaric hypoxia (air at 380 mmHg) from 1 to 14 days. Twenty-three rats served as controls and were kept in the same room in room air. Those of the hypoxic group had a mean initial weight of 277 ± 6.2 g (range, 217–385 g); those of the control group had a mean initial weight of 271.2 ± 9.3 g (range, 198–342 g). At the start of the experiment each animal was treated in a similar way. After anesthesia was induced with sodium pentobarbital (33 mg/kg ip) the pulmonary artery was catheterized and the catheter maintained in situ by a major modification of the tech-
nique of Herget and Palacek (7). A small incision was made in the neck, 0.5 cm to the right of the midclavicular line. The right external jugular vein was isolated. Two 2-0 silk ligatures were placed around the vein and the distal end was ligated. A small transverse cut was made proximal in the vein through which the introducer and catheter were passed. The introducer was a blunted 7.5-cm 19-gauge needle with the tip turned up 30° (Fig. 1A). The catheter was 12.5 cm long Silastic (ID 0.32 mm, OD 0.64 mm) with a notched end hole. It was passed through the introducer after it had been flushed with heparinized saline and was attached by a 25-gauge blunted needle to a pressure transducer (Ailtech MS10-B) and oscilloscope (Hewlett-Packard model 564). After the introducer had been positioned in the right ventricular cavity, identified by the pressure tracing, the tip was directed anteriorly. It was then possible to manipulate the catheter into the pulmonary artery. The catheter position was confirmed by the typical pressure tracing seen in the oscilloscope (Fig. 1B). The introducer was then slipped out over the catheter and removed, and the catheter was affixed to the vein at its entry point as well as to the tissue hole distally by basket-weave sutures. A 9-cm 18-gauge spinal needle was tunneled subcutaneously from the base of the skull to the neck anteriorly. The stylet was removed and the distal end of the catheter was inserted into the spinal needle and pulled through to the back of the skull where it was affixed to the skin with a third basket-weave suture. The anterior wound was then closed with 4-0 silk. The catheter was reattached to the transducer for final measurements of phasic and mean pulmonary artery pressures. The catheter was then flushed and filled with heparin (0.2 ml, 10,000 U/ml) and closed with a blunted wire plug.

The aorta was cannulated by the method of Weeks and Jones (21). A midline abdominal incision was made and the abdominal aorta was isolated just above the iliac bifurcation. Polyethylene tubing, PE-10 (ID 0.28 mm, OD 0.61 mm) and PE-20 (ID 0.38 mm, OD 1.09 mm) had been melted together to form a catheter that was then flushed with heparinized saline. The PE-10 part was inserted through a prior needle puncture into the artery and the PE-20 part was sutured to the psoas muscle. The catheter was then threaded subcutaneously along the back of the animal to the base of the skull where it was brought to the exterior and sutured with 4-0 silk. It was then attached by a blunted 25-gauge needle to the pressure transducer and the pressure was recorded. The catheter was subsequently flushed with heparin and closed with a blunted wire plug.

After 24-48 h were allowed for recovery, measurements were taken of pulmonary and systemic artery pressure with the animals unanesthetized, and a blood sample was drawn for a hematocrit determination. If the hematocrit was below 35, the animal was transfused with 10 ml/kg of whole blood. The animals were then heparinized and the catheters were again filled with concentrated heparin (10,000 U/ml) and plugged.

![Diagram](https://example.com/diagram.png)
Rats in both groups were housed in the same room in individual cages, the only difference being that the experimental hypoxic group was kept in the hypobaric chamber. All animals were weighed initially after catheter placement and almost daily thereafter. The catheters were flushed daily with dilute heparin (20 U/ml) and the rats were heparinized (150 U/kg). On alternate days measurements of pulmonary and systemic artery pressure were recorded from the indwelling catheters with the animals quiet but unanaesthetized. It was therefore necessary to remove the hypoxic rats from the hypobaric chamber for up to 1 h each day to weigh them, replenish their food and water, record pressures, flush their catheters, and heparinize them.

Twenty hypoxic rats were killed by overdose of pentobarbital (300 mg/kg) between days 1 and 14 of exposure; that is, after each day of exposure one or more animals were killed. Between day 6 and 14 of the experiment, five control animals were also killed. Weights and pressure measurements were taken and a blood sample for a second hematocrit reading was drawn. The heart and lungs were removed en bloc. A transverse incision was made in the right ventricular outflow tract, the pulmonary artery catheter was removed, and a 3-in. polyethylene cannula (PE-90) (ID 0.86 mm, OD 1.27 mm) was inserted into the main pulmonary just below the bifurcation of right and left pulmonary arteries. The cannula was sutured tightly in place. The right ventricular free wall was then completely separated and removed; the left ventricle and septum were thereafter removed together according to the method of Fulton et al. (5). Right ventricular free wall (RV) and left ventricle together with septum (LV + S) were weighed separately and expressed as a ratio of alveoli to arteries. The features of the tissue were analyzed microscopically and expressed as a ratio of alveoli to arteries. The lung volumes were then averaged for the 15 fields. A minimum of 50 arteries per tissue section were then landmarked by their accompanying airway, and their external diameters were measured. In addition, each artery was identified as one of the three structural types: completely muscular (completely surrounded by muscle), partially muscular (incompletely surrounded, having only a crescent of muscle), or nonmuscular (no muscle). Wall thickness of the muscular arteries and of the muscular part of the partially muscular arteries was quantified as a percentage wall thickness, calculated by the formula:

\[
\text{percent wall thickness} = \frac{2 \times \text{wall thickness} \times 100}{\text{external diameter}}
\]

To be certain that the morphological changes seen were not secondary to thrombotic lesions induced by the catheters, eight additional weight-matched uncatheterized rats were studied. Four were kept with the catheterized rate in the hypobaric chamber for a 2-wk duration of exposure and four were kept in room air. At the end of 2 wk they were treated in a similar way to the catheterized animals.

All angiograms and all microscopic sections were studied without previous knowledge of whether the animals were hypoxic or catheterized. The hemodynamic findings were correlated with duration of hypoxia and with the morphological changes. Statistical significance was assessed from correlation coefficients established from linear regression plots. To assess the relative contribution of the individual morphological features to the level of mean pulmonary artery pressure and that of the pulmonary artery pressure and duration of hypoxia to the morphological changes, multivariate analyses were also performed. Comparisons between animals, whether hypoxic or catheterized, were based on the t test (unpaired).

**RESULTS**

**Growth**

From a total of 283 weighings of the 88 catheterized animals and 32 of the 8 uncatheterized, a striking difference in growth patterns was observed (Fig. 2). Control catheterized animals lost weight for a few days after the procedure, but by the end of 2 wk they were growing at a lower level but parallel to the uncatheterized control animals. The hypoxic uncatheterized animals lost weight initially; after the 1st wk they began to gain weight, but slowly. The catheterized hypoxic animals lost weight for a week; then although they stopped losing, they did not gain.

**Hemodynamics**

From pulmonary artery pressure measurements, recorded on 155 occasions from 55 hypoxic rats and on 84 from the 23 controls, a mean pulmonary artery pressure and standard error were calculated for each day of the experiment. It was observed that in control rats the mean pulmonary artery pressure (Ppa) did not change over a 14-day period. The mean initial measurement was 18.1 ± 1.6 mmHg and the mean final measurement was 20.8
The number of animals in each of these control and hypoxic groups in which blood gas samples had been taken was seven. Arterial blood gases could not be taken directly in the chamber. However, to simulate the effect of half an atmosphere, 14 weight-matched rats were given 10% oxygen by mask for 15 min (time to achieve a steady state): $P_AO_2$ fell from 92.6 ± 0.6 to 55 ± 3.8, ($P < 0.005$).

**Hematocrit**

Because the mean hematocrit value (Hct) for the uncatheterized control rate was 45 ± 1.0, but 35 ± 3.4 for the catheterized control group, we infer that blood loss during catheterization drops the hematocrit measurement by about 10%. For this reason, a less striking degree of polycythemia in response to hypoxia (Hct, 47.8 ± 1.6), developed in the catheterized as compared with noncatheterized animals (Hct, 60 ± 1.0) ($P < 0.005$).

**Right Ventricular Hypertrophy**

When compared with matched controls, groups of hypoxic rats (closed squares) and control rats (open squares). Growth curves for the catheterized rats are indicated (CATH). Best growth curve is that of control uncatheterized animals. It is followed by that of the control catheterized animals who show a growth lag for a few days, and then gain at the same rate as other controls; hypoxic uncatheterized rate lose weight only the 1st wk of exposure and then grow slowly and hypoxic catheterized rats lose weight for 1 wk, then stabilize but do not gain weight. ± 1.4 mmHg, ($P > 0.3$). On the other hand, $P_{PA}$ of the hypoxic group rose progressively with increase in duration of hypoxic exposure. From a base-line mean value of 17.7 ± 0.4 mmHg, this rise was significantly increased after day 3, ($P_{PA} = 23.7 ± 1.1$ mmHg, $P < 0.05$) and had more than doubled by day 14 ($P_{PA} = 44.6 ± 4$ mmHg) (Fig. 3).

**Systemic artery pressures were recorded on 150 occasions from the 55 hypoxic rats and on 72 from the 23 controls. The hypoxic rats had similar initial and final measurements ($P_{SA} = 104 ± 1.8$ mmHg and 110 ± 2.4 mmHg) as the controls ($P_{SA} = 102.9 ± 4.1$ mmHg and 108.5 ± 8.5 mmHg). Systemic artery pressure was therefore not influenced by exposure to hypoxia.**

Arterial blood gases were not obtained from either the hypoxic or control groups of animals. In previous experiments in our laboratory we had shown that $PO_2$, pH, and $PCO_2$ values were normal in animals that had been in room air for 1 h after 2-wk exposure to hypoxia and with the similar pulmonary artery pressures to animals in the present experiment: (pH = 7.4 ± 0.03, $PO_2 = 101.8 ± 5.4$ mmHg, $PCO_2 = 32.4 ± 1.4$ mmHg). Control animals kept in room air also had similar values (pH = 7.34 ± 0.04, $PO_2 = 106.5 ± 13.7$ mmHg, $PCO_2 = 33.7 ± 1.8$ mmHg).

**Fig. 2.** Growth curves of groups of hypoxic rats (closed squares) and control rats (open squares). Growth curves for the catheterized rats are indicated (CATH). Best growth curve is that of control uncatheterized animals. It is followed by that of the control catheterized animals who show a growth lag for a few days, and then gain at the same rate as other controls; hypoxic uncatheterized rats lose weight only the 1st wk of exposure and then grow slowly and hypoxic catheterized rats lose weight for 1 wk, then stabilize but do not gain weight.

**Fig. 3.** Means and standard errors are given of pulmonary artery and systemic artery pressures in control and hypoxic animals on days of exposure in which these measurements were recorded. It is observed that mean pulmonary artery pressure becomes abnormally elevated by day 3 of exposure in hypoxic group and doubles in value by day 14. Systemic artery pressure is unchanged. Both pulmonary artery and systemic artery pressures in control group are unchanged over the duration of experiment. Control (open squares, broken line); Hypoxic (closed squares, solid line).
Hypoxic rats, whether catheterized or not, showed a significant increase in RV weight \( (P < 0.005) \). LV + S weighed less in the hypoxic rats, but this reflected their lower body weights (Table 1). Thus reduced LV + S/RV was the result of increased right ventricular rather than reduced left ventricular weight. The ratio was reduced after day 3 of hypoxic exposure \( (P < 0.005) \), and decreased progressively with increasing duration and hypoxia \( (r = 0.6) \) and level of mean pulmonary artery pressure \( (r = 0.7, P < 0.005) \) (Fig. 4). After 2 wk of hypoxia the severity of right ventricular hypertrophy was similar in catheterized and uncatheterized rats, LV + S/RV in the hypoxic animals being about 2:1 as compared with 3.3:1 in the controls \( (P < 0.001) \) (Table 1 and Fig. 4).

**Macroscopic Evaluation of the Lungs**

**Lung Volumes.** In uncatheterized animals, lung volumes measured by water displacement were larger in the hypoxic animals than in controls \( (P < 0.025) \). This was also true for the catheterized hypoxic rats if their smaller body weights were allowed for \( (P < 0.025) \). (Table 2).

**Angiography.** In hypoxic rats, the mean lumen diameter of the axial arteries was less along the whole pathway than in controls, and narrowed with progressive hypoxic exposure (Fig. 5). The filling of small arteries, assessed qualitatively as "background haze" was diminished in the hypoxic group, a change more striking as duration of hypoxia increased and pulmonary artery pressure rose.

**Microscopic Evaluation of the Lungs**

Microscopic examination of the lungs revealed three abnormal features of the pulmonary arterial circulation. The first to be considered was extension of muscle into smaller and more peripheral arteries than normal (Figs. 6 and 7). The degree of this abnormality correlated both with the level of pulmonary artery pressure \( (r = 0.84) \) and the duration of hypoxia \( (r = 0.75) \). Multivariate analysis showed that both features were necessary to explain the degree of abnormal extension of muscle; for each \( P < 0.02 \). Pressure may vary even over a short period of time, and individual animals vary in pressure response to a similar exposure to hypoxia.

After day 2 of hypoxia, when mean pulmonary artery pressure had risen only from 18 to 22 mmHg, 23% of arteries within the alveolar walls were partially muscularized. This represented a significant change \( (P < 0.05) \). By day 14, when the mean pulmonary artery pressure reached 37 mmHg, 75% of these alveolar wall arteries had become fully muscularized. Muscularization of alveolar wall arteries was similar in the hypoxic animals.

**TABLE 1. Right and left ventricle and body weight**

<table>
<thead>
<tr>
<th></th>
<th>FBW, g</th>
<th>RV, g</th>
<th>LV + S, g</th>
<th>RV/FBW, %</th>
<th>LV + S/ RV, %</th>
</tr>
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<tbody>
<tr>
<td>Control noncath, ( (n = 4) )</td>
<td>350±16 0.22 0.72 0.06 0.21 3.2</td>
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<tr>
<td>Hypoxic noncath, ( (n = 4) )</td>
<td>274±12 0.27 0.53±0.10 0.24 2.0±0.2</td>
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<tr>
<td>Control cath, ( (n = 5) )</td>
<td>286±21 0.19 0.68 0.07 0.24 3.5</td>
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<tr>
<td>Hypoxic cath, ( (n = 20) )</td>
<td>219±7 0.24±0.04 0.53±0.12 0.24 2.2±0.3</td>
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Values are means ± SE. FBW, final body wt; RV, right ventricle; LV + S, left ventricle and septum; cath, catheterized; \( n \), number of animals. * \( P < 0.01 \) when compared with matched control. † \( P < 0.005 \) when compared with matched control. ‡ \( P < 0.02 \) when compared with matched control.

**FIG. 4. Right ventricular hypertrophy for 20 hypoxic animals (closed squares) and 5 controls (open squares) as it related to Ppa. Right ventricular hypertrophy was measured as a decreasing LV + S/RV; \( r = 0.6 \). LV + S/ RV = 0.05 (Ppa) + 3.9 ± 0.5, \( P < 0.001 \).**
whether catheterized or not.

The second morphological feature considered was thickness of the arterial medial muscular coat, expressed as a percentage of the external diameter (percent wall thickness) (Figs. 6 and 8). The increase in percent wall thickness (%WT) of small arteries correlated both with rise in pulmonary artery pressure \((r = 0.64)\) and with the duration of hypoxia \((r = 0.66)\) and was significant after day 3. For example, in vessels 50–100 \(\mu m\), a rise in percent wall thickness from 5.2 \(\pm 0.7\%\) to 8.0 \(\pm 0.1\%\) occurred \((P < 0.005)\). This was associated with a sustained rise in mean pulmonary artery pressure of 11 \(\pm 1\) mmHg. After 2 wk, in vessels of this diameter range, percent wall thickness had doubled to 10.8 \(\pm 0.7\%\) in association with 20 \(\pm 1\) mmHg rise in pressure.

The severity of the third morphological change, a reduction in the number of small arteries expressed as an increased ratio of alveoli to arteries correlated both with rise in mean pulmonary artery pressure \((r = 0.73)\) and with the duration of exposure to hypoxia \((r = 0.66)\) (Figs. 6 and 9). In control animals, catheterized or uncatheterized, the ratio of alveoli to arteries was approximately 22:1. This ratio rose significantly after day 3 of hypoxic exposure to 31 \(\pm 2\) \((P < 0.005)\) and continued to rise so that by day 14 it was 38:1. Because total number of alveoli per unit area did not differ in control and hypoxic rats \((P > 0.2)\), the increase in ratio of alveoli to arteries represented a reduction in artery number (Table 2). The increase in lung volumes observed in the hypoxic rats was therefore the result of an increase in alveolar size, rather than in number per unit area, i.e., relative to body weight.

With respect to each of the three structural features, not all animals with the same rise in pulmonary artery pressure had the same degree of morphological change. Each was the most striking in some animals. To assess
whether any individual morphological features correlated with rise in pulmonary artery pressure in response to chronic hypoxia when the other two remained fixed, multivariate analysis was applied to our data. The degree of abnormal extension of muscle into peripheral arteries proved to convey the totality of morphological information: when abnormal extension of muscle is taken into account, neither increased wall thickness of the muscular arteries nor increased ratio of alveoli to arteries correlates with pulmonary artery pressure (\(P > 0.7\) for each). These two features are informative only as surrogates for extension of muscle.

**DISCUSSION**

**Growth**

That rats kept in a hypoxic environment lose weight at first and then gain more slowly than age-matched controls kept in room air was observed both by Heath et al. (6) and Hislop and Reid (9). Our noncatheterized rats demonstrated a similar growth pattern to that reported by the previous investigators. The catheterized rats in each group grew less well than the uncatheterized. It seems that the anemia and trauma resulting from the catheterization operation caused the failure to thrive of the hypoxic rats and the poor initial growth of the controls. Because the animals were remarkably quiet during the blood pressure measurements, stress is unlikely to have been an important factor in causing the failure to thrive, although it cannot be excluded.

**Hemodynamics**

**Pulmonary artery pressure.** After 2 wk, mean pulmonary artery pressure did not rise in control animals, whereas in the hypoxic rats the pulmonary artery pressure rose progressively. Acute intermittent hypoxia in anesthetized dogs is associated with potentiation of the rise in pulmonary artery pressure, but on return to room air is quickly and completely reversible (20). Our measurements were recorded after the animals had been out of the hypobaric chamber for an hour and show that under these conditions the pulmonary artery pressure does not return to normal. Although acute rise in pulmonary artery pressure is probably the result of vasoconstriction, the sustained elevation that we observed may be the result of morphological alteration of the pulmonary vascular bed. Correlation of regression of morphological changes with pulmonary artery pressure will elucidate this, although the possibility cannot be excluded, that in chronic hypoxia, a biological mediator alters the pressure flow characteristics of the pulmonary vascular bed.

Abraham et al. (1) kept rats in a hypobaric chamber for 33 days: within 24 h of removal from the chamber,
right ventricular mean pressures were double those of controls. A sustained rise of mean right ventricular pressure of similar degree was observed after rats had been exposed to intermittent hypoxia, 4 h daily for 70 days (22). Our measurements have the two advantages of being direct readings from the pulmonary artery and of

FIG. 8. A: an artery accompanying a terminal bronchiolus (TBA) and an artery accompanying a respiratory bronchiolus (RBA) from control rat of Fig. 7. These arteries are only partially muscularized and wall thickness expressed as a percent of external diameter is normal. B: a terminal bronchiolus artery (TBA) and a respiratory bronchiolus artery (RBA) from hypoxic rat of Fig. 6. These arteries are completely muscularized and the wall thickness expressed as a percent of external diameter is twice normal (magnification X100).

FIG. 9. A: low power field (x25) in control rat showing many alveolar wall and duct arteries (see arrows) and a normal alveoli/arteries ratio. B: same power field in hypoxic rat after 14 days of exposure. There is a striking absence of small peripheral arteries, i.e., no alveolar wall or duct arteries and only one respiratory bronchiolus artery (arrow) is seen.
being uninfluenced by anesthesia. In awake swine after a 4-wk exposure to hypobaric hypoxia, McMurtry et al. (14) measured pulmonary artery pressure while the animals were still in the chamber. Their values were twice as high as ours. This may reflect the effect of an additional 2 wk of exposure, species difference, or a strong acute vasoconstriction component because the measurements were recorded with the animals still in the chamber. Will et al. (23) exposed cattle to chronic hypoxia by bringing them from an altitude of 5,000 to 10,000 ft for periods from 6 wk to 6 mo. Some animals doubled their pulmonary artery pressures and others developed right-sided congestive heart failure. In human subjects acclimatized to high altitude, mean pulmonary artery pressure increased by 17% (18). This represented a smaller increase than that observed in men living at high altitude from birth where pulmonary artery pressure is double control sea level values (11, 17).

**Systemic artery pressure.** Measurements of systemic artery pressures have been obtained in previous studies from the carotid arteries of both anesthetized (7) and unanesthetized rats (4, 21). Herget and Palacek (7) recorded values lower than ours, perhaps because their rats were more deeply anesthetized. Our values were lower, however, than those reported by Coleman (4) and Weeks and Jones (21), perhaps because their animals were of a different breed (Wistar) or larger. Also, because of the double operation (the placing of a pulmonary artery catheter and a pulmonary artery angiogram) our animals may have been more hypovolemic, and as a result, more hypertensive. The response of systemic artery pressure to hypoxia in rats has not been previously studied. McMurtry et al. in swine (14) found no change after a month of hypobaric hypoxia. In human subjects acclimatized to high altitude, no significant difference in mean systemic artery pressure is found (18).

**Hematocrit**

In the uncatheterized animals in our experiments that were exposed to 2 wk of hypoxia, blood hematocrit levels were similar to those reported in mice after 3 wk exposure (16). The catheterized hypoxic rats were not as polycythemic as the uncatheterized, perhaps because they started off with a lower hematocrit, as a result of blood loss at the time of catheter implantation. In the hypoxic catheterized rats in response to hypoxia, the correlation between the degree of elevation of pulmonary artery pressure or duration of hypoxic exposure and the hematocrit level was not close. Naeye (16) previously observed that bleeding prevented the polycythemic response to hypoxia, but that the morphological changes in the pulmonary vascular bed were unaltered. It is not surprising then, that the catheterized animals, in spite of relative anemia, showed the same degree of right ventricular hypertrophy and altered pulmonary vascular morphology as the uncatheterized animals.

**Right Ventricular Hypertrophy**

Our findings of right ventricular hypertrophy when expressed as the ratio (LV + S/RV) agree with those of others (1, 6, 9, 12), for rats under similar conditions of hypoxia. This ratio was similar in catheterized and uncatheterized animals in our series and is useful as it allows comparison of animals of different body weights.

**Macroscopic Evaluation of the Lungs**

**Lung volumes.** The lung volumes of the hypoxic animals, whether catheterized or uncatheterized, were larger than those of the controls. The difference was only evident in the catheterized animals if allowance was made for body weight. Our findings indicate that the basis for increase in lung volume was increase in alveolar size relative to body weight, which agrees with the observations of Hislop and Reid (9, 10). To our knowledge, this feature has not been studied either in animals or in the human and no satisfactory explanation has therefore been proposed.

**Lung angiography.** On angiography, the lumen diameter of the axial pulmonary arteries was smaller in the hypoxic than in the control animals. The presence of an indwelling catheter did not influence this. The thickening of the wall of the axial pulmonary artery contributes to the decrease in lumen diameter. The thickening of the wall is due to an increase in the adventitia, the muscle as well as the collagen and fibroblasts, and also to an increase in the alveolar size relative to body weight, which agrees with the observations of Hislop and Reid (9, 10). To our knowledge, this feature has not been studied either in animals or in the human and no satisfactory explanation has therefore been proposed.

**Microscopic Evaluation of the Lungs**

**Lung microscopy.** Microscopic examination of the lungs of the hypoxic rats revealed abnormal features in pulmonary arterial structure as first described by Hislop and Reid (9) under similar experimental conditions. They were slightly more pronounced in our series but not significantly (P > 0.05). No peripheral pulmonary emboli were found in the catheterized animals nor evidence of infection. The severity of each of the structural changes correlates both with the duration of exposure to hypoxia and with the level of mean pulmonary artery pressure.

It is difficult to be certain whether these structural changes occurred in any particular sequence. Although abnormal extension of muscle was significant after day 2 of exposure, increased wall thickness of the normal muscular arteries and the ratio of reduced alveoli to arteries also appeared as early, but because of the wider and more variable normal range in these measurements, were only significant after day 3 of exposure. This coincided with the earliest significant measurement of elevated pulmonary artery pressure.

Recently Hislop and Reid (10) have shown that both abnormal extension of muscle into small peripheral arteries and medial hypertrophy of the normally muscular arteries are at least partially reversible. It has been demonstrated that the basis of abnormal extension is the conversion of pericytes and intermediate cells into new muscle cells (15).

The reduction, in response to chronic hypoxia, of the number of arteries demonstrated by this technique has
previously been described only by Hislop and Reid (9). The loss is mainly in partially muscular and nonmuscular arteries, and it is possible that endothelial swelling, medial hypertrophy, and muscle constriction all contribute to the closing down of arteries. When rats are fed Crotalaria spectabilis seeds, pulmonary vascular changes similar to those described in response to chronic hypoxia develop, but they take longer and "ghost vessels" remain (8). Perhaps, in response to hypoxia the changes occur so rapidly that arterial remnants are not detected.

REFERENCES


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