Myosin heavy chain composition of skeletal muscles in young rats growing under hypobaric hypoxia conditions

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Bigard, A. X., H. Sanchez, O. Birot, and B. Serrurier. Myosin heavy chain composition of skeletal muscles in young rats growing under hypobaric hypoxia conditions. J. Appl. Physiol. 88: 479–486, 2000.—This study investigated the effects of voluntary wheel running on the myosin heavy chain (MHC) composition of the soleus (Sol) and plantaris muscles (Pla) in rats developing under hypobaric chronic hypoxia (CH) conditions during 4 wk in comparison with those of control rats maintained under local barometric pressure conditions (C) or rats pair-fed an equivalent quantity of food to that consumed by CH animals (PF). Compared with C animals, sedentary rats subjected to CH conditions showed a significant decrease in type I MHC in Sol (−12%, P < 0.01). Although strongly decreased under hypoxia, spontaneous running activity increased the expression of type I MHC (P < 0.01) so that no difference in the MHC profile of Sol was shown between CH active and C active rats. The MHC distribution in Sol of PF rats was not significantly different from that found in C animals. CH resulted in a significant decrease in type I (P < 0.01) and type IIa (P < 0.005) MHC, concomitant with an increase in type IIB MHC in Pla (P < 0.001), compared with C and PF animals. In contrast to results in Sol muscle, this slow-to-fast shift in the MHC profile was unaffected by spontaneous running activity. These results suggest that running exercise suppresses the hypoxia-induced slow-to-fast transition in the MHC expression in Sol muscles only. The hypoxia-induced decrease in food intake has no major influence on MHC expression in developing rats.

Skeletal muscle is a heterogeneous tissue composed of different muscle fibers. This diversity is expressed by the variety of myosin isoforms that result from the expression of several genes encoding for the myosin heavy chain (MHC) and myosin light chain isoforms (for review, see Ref. 15). Switching of the MHC profile occurs after hormonal changes, activity pattern, or perturbations in mechanical loading or during development.

Chronic exposure to hypobaric hypoxia has been shown to induce marked changes in skeletal muscle morphology, such as decreased mean fiber cross-sectional areas and increased capillary density (17). Furthermore, it has been previously reported that the percentage of type IIa fibers was higher in soleus muscles (Sol) and extensor digitorum longus (EDL) muscles of young rats exposed to chronic hypoxia than in control animals (5). These changes in fiber-type distribution, which are controversial in adults (1, 17), have been shown to be related to an inhibition of the shift of muscle fiber types that occurs during postnatal development in Sol muscles (4, 6). However, nothing is known about the mechanisms of this inhibition. Whether chronic hypoxia (CH) affects the MHC profile of fast-twitch muscles in young rats remains to be elucidated, and additional studies are needed to solve this issue.

Skeletal muscle is characterized by the ability to modify its structural and biochemical properties in response to increased functional load, and endurance training is known to result in fast-to-slow shift of the MHC profile of locomotor muscles (18). This was most clearly shown in fast-twitch red muscles and, to a lesser extent, in slow-twitch muscles, likely because they have a high percentage of type I MHC in sedentary rats. Voluntary wheel running has been widely used as a model of noninterventional exercise training that induces adaptive changes in biochemical properties of muscle fibers (7, 12). Moreover, repeated voluntary exercises have been shown to alter the fiber-type profile of fast-twitch muscles, a result that indicates a fast-to-slow transition of myosin expression (7). One of our hypotheses was that voluntary running activity could reverse the CH-induced inhibition of the growth-related shift of the MHC profile in slow-twitch skeletal muscles.

Long-term exposure of rats to CH is associated with a depressed growth rate mainly attributable to a decrease in food consumption in comparison with normoxic rats (2, 16). Although chronic energy restriction does not affect the fiber-type composition of locomotor muscles in adult rodents (9), a delayed maturation of muscle fibers has been reported in undernourished guinea pigs during the early postnatal period (20). Because alterations in the fiber-type composition of skeletal muscles have been reported in rats growing under hypobaric conditions (4, 6), determination of CH-induced changes in MHC expression requires a clear separation between the influences of hypoxia per se and the diet-related perturbations.

The present study was carried out to 1) investigate changes in the fiber-type and MHC composition of slow- and fast-twitch muscles in young rats growing under
hypobaric hypoxia and 2) evaluate the protective effect of voluntary exercise on the inhibition of the age-related shift of the fiber-type composition of Sol and plantaris (Pla) muscles. To differentiate the effects of hypoxia per se from those of the CH-induced decrease in food intake, groups of rats exposed to normoxia were pair-fed quantities of food equivalent to those consumed by animals subjected to CH.

**MATERIALS AND METHODS**

Treatment of animals. Forty-seven male Wistar rats [initial body wt 60.4 ± 2.2 (SE) g at the start of the experiment] were used for this study. Before the start of the experiment, all animals consumed a standard mixed diet available at all times. After 4 days of acclimatization to the animal facility, rats were randomly assigned to either sedentary control or exercise training groups. Sedentary rats were housed individually in hanging wire-mesh cages (30 cm width x 40 cm length). Exercising rats were placed in individual cages with wheels for voluntary exercise, providing the same living space as cages housing the sedentary rats. After 1 wk of acclimatization to individual cages and running wheels, both sedentary and active rats were randomly distributed into three groups for the remaining 4 wk of the experiment: chronically exposed to simulated high altitude (CH), maintained under local barometric pressure conditions (C), or pair-fed an equivalent quantity of food to that consumed by CH animals (PF). Thus the 47 rats were divided into 6 groups: sedentary or active rats exposed to CH (n = 8 CHs and n = 8 CHa, respectively); sedentary or active rats maintained at ambient pressure, fed ad libitum (n = 8 Cs and n = 8 Ca, respectively); or sedentary or active rats maintained at ambient pressure and pair-fed an equivalent quantity of food to that consumed by CH animals (n = 8 PFs and n = 7 PFa, respectively). All experiments were performed in accordance with the Helsinki accords for humane treatment of laboratory animals.

CHs and CHa groups were housed in a hypobaric chamber and exposed to barometric pressure that was reduced over 2 days until the equivalent of 5,500 m altitude was reached (barometric pressure = 370 mmHg; PIO2 = 79 mmHg). In the hypobaric chamber, rats were maintained at an ambient temperature of 22 ± 2°C with a 12:12-h dark-light cycle and had free access to food and water. The chamber was opened daily to measure food consumption and refill water dispensers. The retention period at the ambient pressure (~740 mmHg) did not exceed 30-45 min. The remaining four groups of animals were maintained at ambient pressure and at a temperature of 22 ± 2°C in individual cages with a 12:12-h dark-light cycle. They had free access to water. Cs and Ca animals were provided food ad libitum, whereas PFs and PFa rats were pair-fed on a daily basis using as a reference the food intake of CH rats. All animals consumed a standard mixed diet. Rats were weighed daily to monitor weight gain.

Each animal was individually surgically removed, weighed, mounted in an embedding medium (TEK OCT compound), and frozen in isopentane cooled to the freezing point (~160°C by liquid nitrogen). All samples were stored at −80°C until histochromic and biochemical analysis. Changes in MHC isoform expression were assessed by using immunohistochemical detection of MHCs within single muscle fibers and gel electrophoresis to separate MHC isoforms in whole muscle.

**Immunocytochemistry.** Six mouse monoclonal antibodies directed against specific MHC isoforms were used in this study. Serial tissue cross sections were cut at −20°C using a cryostat and were incubated for 30 min in an appropriate blocking solution at 37°C in a humid chamber. Sections were subsequently incubated for 1 h at 37°C in working solutions of mouse monoclonal antibodies that reacted with either 1) slow type I (Novoceastra, Newcastle-upon-Tyne, UK), 2) all adult fast and developmentally regulated epitopes but not with slow myosin (MY-32, Sigma Chemical, St. Louis, MO), 3) fast type IIA (clone SC-71), 4) fast type IIB (clone BF-F-3), 5) slow and fast type IIA and type IIB but not with type IIX MHC (clone BF-35), or 6) embryonic and neonatal MHC isoforms but not any of the adult isoforms (RNMY2/9D2, Novoceastra). Monoclonal antibodies SC-71, BF-F-3, and BF-35 have been previously characterized (14) and were taken from antibody-producing hybridomas (DSM, Braunschweig, Germany). Sections were rinsed with PBS and incubated in the appropriate peroxidase-conjugated secondary antibody. The avidin-biotin immunohistochemical procedure was used for the localization of the antigen-antibody binding (kit PK-4002, Vector Laboratories, Burlingame, CA). A sample of ~400 fibers that were free from artifact were randomly selected from fields equally distributed over the biopsy for single-fiber MHC composition. Fibers were classified according to their staining profile with the aid of a microscope linked to a computer-based image analysis system (Visiijlab 200, Nikon-France).

Analysis of MHCs. Sol and Pla muscles were subjected to MHC isoform analysis by using previously described techniques (19). Frozen muscles were minced with scissors in 9 vol of a solution containing 20 mM NaCl, 5 mM sodium phosphate, and 1 mM EDTA (pH 6.5). Myosin was then extracted with 3 vol of 100 mM sodium pyrophosphate, 5 mM EDTA, and 1 mM dithiothreitol (pH 8.5). After 30 min of gentle shaking, the mixture was centrifuged at 12,000 g for 10 min, and the supernatant containing myosin was diluted twice with glycine and stored at −20°C. The separating gel solution contained 30% glycerol, 8% acrylamide-bis (50:1), 0.2 M Tris, 0.1 M glycine, and 0.4% SDS. The composition of the stacking gel was 30% glycerol, 4% bis-acrylamide (50:1), 70 mM Tris, 4 mM EDTA, and 0.4% SDS. Myofibril samples were denatured by using a sample buffer containing 5% β-mercaptoethanol, 100 mM Tris base, 5% glycerol, 4% SDS, and bromophenol blue. Electrophoresis was performed by using a Mini Protean II system (Bio-Rad Laboratories, Hercules, CA). Gels were run at constant voltage (70 V) for ~28 h and then
stained with Coomassie blue. The MHC protein isoform bands were scanned and quantified by using a densitometer system equipped with an integrator (GS-700, Bio-Rad Laboratories).

Statistical procedures. All data are presented as means ± SE. Determination of the statistical significance of environment (CH, C, or PF conditions) and voluntary running was accomplished using a two-way ANOVA. When appropriate, differences between groups were tested with a Newman-Keuls post hoc test. Statistical significance was accepted at P < 0.05.

RESULTS

Growth, body, and muscle weights. An initial period of weight loss occurred from the first day of exposure to hypoxia, and both CHs and CHa rats recovered their initial body weight within 3 days. After the 1-wk period of acclimation to the individual cages, the daily body weight gain of CH and PF rats was less than that of C animals (3.09 ± 0.15% and 2.84 ± 0.2%, respectively, in comparison with 3.95 ± 0.25%, P < 0.001, irrespective of the level of spontaneous activity. No significant difference was found between the body growth rate of CH and PF rats, either sedentary or active. Final body weight was significantly affected by the environment (P < 0.001) and was reduced by 23% and 19% in CHs and CHa rats, respectively (P < 0.01), and by ~20% in PFs and PFa rats (P < 0.01), in comparison with corresponding C animals (Table 1). High Hct values were consistent with hematological adaptation to CH.

Absolute Sol and Pla muscle weights were significantly altered by the environment (P < 0.001), with the activity status having no effect (Table 1). Exposure to CH decreased the absolute weights of Sol and Pla in active rats (P < 0.05, P < 0.01, respectively) and of Pla in sedentary animals (P < 0.01) compared with C animals. Moreover, the relative Pla muscle weight-to-body weight ratio in CHa rats was lower than that measured in PFa rats (P < 0.01). The ANOVA of the Sol and Pla muscle weight-to-body weight ratio showed a significant effect of voluntary running (P < 0.005). Between-group comparisons demonstrated that spontaneous running activity increased the Pla muscle weight-to-body weight ratio only in PF rats (+15%, P < 0.05).

Food consumption. Compared with C rats, the food consumption of CH animals was markedly reduced (P < 0.001) (Table 2). In all groups, there was a progressive age-related increase in daily food intake, but values measured in CH rats were consistently lower than those measured in C animals (P < 0.001). When normalized to body weight, mean values of relative daily food intake show that both CH and PF rats ate less than C rats only during the first week of hypoxia (P < 0.001). There was no significant difference in daily food intake between sedentary and exercised animals.

Spontaneous running activity. The pattern of spontaneous running activity is shown in Fig. 1. An increase in running activity occurred in Ca and PFa rats during the first 2 wk of the experiment. In contrast, the daily distance run by CHa rats decreased markedly from the first day of exposure to hypobaric hypoxia, so that running activity was only ~28% that of Ca and PFa
animals. During the first week of food restriction, PFa rats ran a greater distance per 24-h period compared with Ca animals (32%, P < 0.05). After this initial period, no significant difference in average running distances was observed between PFa and Ca groups. As expected, Ca rats showed a nocturnal locomotor activity with a peak between 2200 and 0200. A 3-h phase advance of the nocturnal peak of locomotor activity was observed in PFa rats, associated with an advance of both the onset and the end of running. Because the hypobaric chamber was opened daily, the progressive reestablishment of ambient pressure affected the rhythm of running activity in CHa rats. These changes were clearly experimenter induced.

Immunohistochemistry. The majority of fibers in Sol muscles of C rats contained type I MHC isoform (Fig. 2). Most fibers labeled positively for type I MHC, whereas only a small percentage were pure type IIa fibers (~2%). All fibers labeling positively for fast MHC antibody contained type IIA MHC. The fiber-type composition of Sol taken from C rats was unaffected by voluntary running activity (Table 3). In the Pla muscle, the ANOVA of the fiber-type composition showed that CH resulted in significantly decreased percentages of type I (P < 0.01) and type IIA (P < 0.005) fibers and a concomitantly increased percentage of type IIb fibers (P < 0.001), compared with C and PF animals. The hypoxia-induced alterations in the percentages of type I and type IIb fibers were mainly observed in voluntary running rats (Table 3). The spontaneous activity in Ca rats resulted in significantly increased percentage of pure type I fibers and decreased percentage of type IIb fibers (P < 0.05). In contrast to Sol, the slow-to-fast shift in the fiber-type profile related to CH was unaffected by spontaneous running activity. The fiber-type composition of Pla in PF rats was nearly similar to that observed in C animals and an increase in the percentage of type I MHC was also observed as a result of voluntary running (38%, P < 0.05).

MHC phenotype expression. For all groups, only type I and type IIA MHC isoforms were identified by electrophoresis in Sol muscles (Fig. 4). The relative distribution of MHC isoforms was affected by the environment (P < 0.001) and by voluntary running (P < 0.001) (Table 4). Type I MHC was lower in CHa rats than in Cs animals (P < 0.01). Spontaneous activity increased the percentage of type I MHC (P < 0.01) so that no difference in type I MHC isoform was shown between CHa and either Cs or Ca rats. The MHC distribution in PF rats was similar to that found in C animals. Chronic hypoxia resulted in significant decreases in the percentage of type I and type IIA MHC in Pla muscles, associated with an elevation in the percentage of type IIB MHC in comparison with C and PF groups (Table 4). These alterations were mainly observed in voluntary wheel-running rats. Type I MHC in Pla muscles of both Ca and PFa groups exceeded that of Cs and PFs groups (P < 0.01, P < 0.05, respectively). Active rats in C and PF groups had a lower percentage of type IIB MHC than sedentary animals (P < 0.01).
The main purposes of this study were to determine whether voluntary running could reverse the inhibition of the developmental changes in MHC distribution that has been reported in young rats exposed to hypobaric hypoxia and to evaluate the role of CH-induced decrease in food intake on this inhibition. The results demonstrate that 1) in addition to the well-known decrease in the percentage of type I MHC in Sol muscles in CHs rats, a similar slow-to-fast transition was observed in Pla muscles; 2) voluntary running activity, although decreased in CH rats, was able to counteract the effects of hypoxia on the MHC content of Sol but not Pla muscles; and 3) the CH-induced reduction of dietary intake did not affect the MHC isoform profile of Sol and Pla muscles.

In the present study, we investigated the MHC isoform content of slow- and fast-twitch muscles of 3-wk-old rats exposed to CH for 4 wk. As previously reported, the MHC composition of Sol muscles was...
altered by CH (2, 4–6). Our data, which demonstrate a CH-induced decrease in the percentage of pure type I fibers and in the relative content of type I MHC in Sol of CHs rats, are consistent with these previous results. Only type I and type IIA MHC isoforms were detected in Sol, and CH was not associated with the expression of fast MHC isoforms normally not found in that muscle. These changes in the fiber-type composition of the Sol muscle have been previously attributed to an inhibition of the age-related transition in the MHC isoforms (4, 5).

On the other hand, results of the present study show that prolonged hypobaric hypoxia induced changes in the fiber-type profile and MHC composition of Pla muscles. Overall decreases in type I and type IIA fibers as well as an increase in type IIB fibers were shown in CH rats. Consistent results were found in the MHC isoform distribution, and these findings indicate a CH-induced transition of the MHC isoform component toward a faster profile. In contrast, Itoh et al. (5) reported that EDL muscles in CH rats exhibited a greater relative distribution of type IIA fibers than in C animals. The discrepancy between these results and ours is unclear but may be explained by the difference in the function of locomotor muscles (i.e., ankle flexor for EDL, ankle extensor for Pla) and in the methods used for determination of the fiber-type composition [i.e., histochemical sensitivity of the actomyosin myofila-

Table 3. Fiber type composition of soleus and plantaris muscles

<table>
<thead>
<tr>
<th>Fiber Type</th>
<th>CHs</th>
<th>CHa</th>
<th>Cs</th>
<th>Ca</th>
<th>PFs</th>
<th>PFa</th>
</tr>
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<tbody>
<tr>
<td>Sol</td>
<td></td>
<td></td>
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<tr>
<td>Type I</td>
<td>83.8±2.7*</td>
<td>92.7±2.3§</td>
<td>94.8±2.6</td>
<td>97.6±1.9</td>
<td>90.1±2.6</td>
<td>96.6±1.5</td>
</tr>
<tr>
<td>Type I/IIa</td>
<td>4±1.1</td>
<td>3.3±1</td>
<td>3.2±1.2</td>
<td>1.5±1</td>
<td>2.5±0.7</td>
<td>1.9±0.9</td>
</tr>
<tr>
<td>Type Ila</td>
<td>12.2±2.9*</td>
<td>4±1.5§</td>
<td>2.1±1.8</td>
<td>0.9±0.9</td>
<td>7.4±2.1</td>
<td>1.4±1.1</td>
</tr>
<tr>
<td>Pla</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Type I</td>
<td>6.8±0.6</td>
<td>8.5±0.8†</td>
<td>8.5±1.1</td>
<td>13.1±1.2§</td>
<td>8.6±1.2</td>
<td>11.9±0.7§</td>
</tr>
<tr>
<td>Type I/IIa</td>
<td>1.3±0.2</td>
<td>1.4±0.3</td>
<td>1.7±0.4</td>
<td>0.6±0.2</td>
<td>1.3±0.2</td>
<td>0.8±0.3</td>
</tr>
<tr>
<td>Type Ila</td>
<td>19.6±1.2†</td>
<td>22.6±1.2†</td>
<td>30.6±2.0</td>
<td>34.2±2.3</td>
<td>26.5±2.3</td>
<td>30.3±1.9</td>
</tr>
<tr>
<td>Type I/Ix</td>
<td>6.8±1.3</td>
<td>7.5±0.8</td>
<td>4.7±0.6</td>
<td>5.4±1.1</td>
<td>6.4±0.7</td>
<td>6.7±0.9</td>
</tr>
<tr>
<td>Type Ix/Iib</td>
<td>18.4±1.8</td>
<td>18.6±1.1</td>
<td>16.4±3.1</td>
<td>24.9±1.7</td>
<td>23.9±1.6</td>
<td>25.8±3.6</td>
</tr>
<tr>
<td>Type I/IIb</td>
<td>4.4±0.7</td>
<td>3.2±1.0</td>
<td>4.9±1.2</td>
<td>3.5±0.6</td>
<td>4.9±0.4</td>
<td>4.2±0.5</td>
</tr>
<tr>
<td>Type IIb</td>
<td>42.6±2.2†</td>
<td>38.1±2.6‡</td>
<td>33.2±5.4</td>
<td>18.2±2.3§</td>
<td>28.3±2.6</td>
<td>20.3±2.6</td>
</tr>
</tbody>
</table>

Values are means ± SE. Different from corresponding C groups, *P < 0.01. Different from corresponding PF groups, †P < 0.05; ‡P < 0.01. Different from corresponding sedentary groups, §P < 0.05.

Fig. 3. Serial transverse sections of plantaris (Pla) muscles from CHs (A, B, and C), CHa (D, E, and F), Cs (G, H, and I), and Ca rats (J, K, and L): sections incubated with antibody specific for type I MHC. B, E, H, and K: sections incubated with antibody specific for type IIA MHC. C, F, I, and L: sections incubated with antibody specific for type IIB MHC. Note decrease in percentage of fibers expressing type IIB MHC as a result of spontaneous running activity in control rats (L vs. I). Calibration bar in C, 200 μm.
brillar ATPase (mATPase) or MHC analysis using both immunohistochemical and electrophoretic methods]. The effects of CH on the fiber-type composition of fast-twitch muscles in adult rats are controversial. A slight decrease in the percentage of type IIA fibers, as determined by the mATPase reaction, has been reported in the deep portion of Pla muscles (1), whereas other studies failed to show any change in rats acclimatized to CH (17). Whether the changes in the MHC profile of Pla muscles recorded in the present study are age related remains to be elucidated. A slight type IIB-to-IIX MHC switching has been reported in tibialis anterior muscles during the aging process (8). However, these changes appeared at a late stage of life and it is not clear whether an inhibition of age-related changes in MHC composition could explain the alterations observed in Pla muscles of hypoxic rats.

The exact mechanisms of the inhibition of postnatal maturation of the fiber-type profile of Sol muscles have not been determined. Neural influences, contractile activity, and mechanical factors, in addition to thyroid status, are known to determine muscle fiber-type composition (15). To our knowledge, the possibility that exposure to CH may be associated with changes in spontaneous physical activity and in muscle contractile activity has not been fully investigated. Perhonen et al. (11) reported that rats exposed to moderate hypoxia showed increased spontaneous activity, but a decrease in physical activity is clearly expected in rats exposed to severe hypobaric conditions. Although the effects of CH on the spontaneous activity of animals within cages remain to be determined, we showed for the first time in the present study a marked decrease in the spontaneous running behavior of CH rats.

Voluntary exercise induced a fast-to-slow transition of the fiber-type profile of Pla muscles in C and PF rats, and this finding is consistent with previous results in EDL muscles (7). These changes have been closely related to the total amount of activity, and the imposition of a known load during the running activity (i.e., 0.04 N·m torque on the wheel) can be considered as a favorable factor for determining adaptive responses to voluntary exercise.

The novel and major finding of this study is that voluntary running was able to counteract the specific effect of hypoxia on the age-related transition in fiber-type composition and MHC content of Sol muscle. However, this effectiveness of voluntary running activity on the developmental changes in the fiber-type distribution and in the MHC component in Sol was not observed in Pla muscles. Clearly, running activity did not significantly alter the MHC isoform composition of Pla muscles in CH rats. This finding may be interpreted as (1) a different pattern of muscle recruitment with respect to the level of spontaneous running activity or (2) a different responsiveness of Sol and Pla muscles to running activity.

First, our results clearly suggest that Sol and Pla muscles are activated during wheel-running activity in Ca rats; however, we cannot exclude the possibility that under CH exposure the low level of running activity was associated with a lower recruitment of Pla, a fast-twitch locomotor muscle that is mainly recruited during phasic movements. On the other hand, why Sol muscles would be more responsive to running activity than Pla muscles in CH rats remains to be elucidated. Many studies have indicated that the heterogeneity of myogenic cells contributes to the determination of muscle plasticity and response to endurance training. Because slow- and fast-twitch muscles comprise myofibers that arise from discrete populations of myoblasts (13), the different responsiveness of Sol and Pla muscles to running activity could be at least partly explained by the developmental history of myofibers.

Food restriction led to a slight increase in wheel activity but only during the first week of exercise. Higher increases in wheel-running activity have been reported in rats submitted to severe food restrictions (10) or after a few months of food intake restricted to

Table 4. Myosin heavy chain composition of Sol and Pla muscles

<table>
<thead>
<tr>
<th></th>
<th>CHs</th>
<th>CHa</th>
<th>Cs</th>
<th>Ca</th>
<th>PFs</th>
<th>PFa</th>
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<tbody>
<tr>
<td>Sol</td>
<td>MHC I, %</td>
<td>88 ± 1†</td>
<td>96 ± 1.4§</td>
<td>98.6 ± 0.5</td>
<td>99.7 ± 0.3</td>
<td>94 ± 1.3</td>
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<tr>
<td></td>
<td>MHC IIA, %</td>
<td>12 ± 1†</td>
<td>4 ± 1.4§</td>
<td>1.4 ± 0.5</td>
<td>0.3 ± 0.3</td>
<td>6 ± 1.3</td>
</tr>
<tr>
<td>Pla</td>
<td>MHC I, %</td>
<td>4.6 ± 0.7</td>
<td>4.9 ± 0.7†</td>
<td>4.5 ± 0.6</td>
<td>9.4 ± 1.3§</td>
<td>4.5 ± 0.9</td>
</tr>
<tr>
<td></td>
<td>MHC IIA, %</td>
<td>14.8 ± 1.2</td>
<td>14.1 ± 1.2†</td>
<td>18.2 ± 2.4</td>
<td>25.9 ± 1.9€</td>
<td>17 ± 2.2</td>
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<tr>
<td></td>
<td>MHC IIX, %</td>
<td>29.2 ± 1.1</td>
<td>32 ± 1.3</td>
<td>29.9 ± 2.6</td>
<td>33.4 ± 1.6</td>
<td>35.5 ± 1.7</td>
</tr>
<tr>
<td></td>
<td>MHC IIB, %</td>
<td>51.4 ± 2.2</td>
<td>49.1 ± 1.8†</td>
<td>48.3 ± 4.2</td>
<td>31.3 ± 1.5§§</td>
<td>42.9 ± 1.9</td>
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</table>

Values are means ± SE. MHC, myosin heavy chain. Different from corresponding C groups, †P < 0.05. Different from corresponding PF groups, ‡P < 0.01. Different from corresponding sedentary groups, §P < 0.05, §§P < 0.01.
~30% below ad libitum (3). The only slight effect of food restriction on wheel-running activity could be explained by the mild caloric restriction and the short period of spontaneous running with food restriction in comparison with previous studies. A delay in the maturation of muscle fibers has been reported in undernourished guinea pigs, whereas no change in the fiber-type composition of SOL and PLA muscles occurred in adult rats (9, 20). In the present study, no significant changes in the expression of MHC isoforms were observed in PF rats in comparison with C animals. Only a trend to increased content of type II A MHC was observed in SOL muscles, without statistical significance. This finding suggests that changes in MHC expression shown in CH animals are closely related to hypoxia per se and not to the CH-associated decrease in energy intake. Interestingly, the MHC composition of PLA muscles of PF animals was affected by spontaneous running activity. As in C rats, voluntary activity elicited an increase in type I MHC and a decrease in type IIB MHC in PF rats, consistent with the high level of wheel-running activity.

In conclusion, our results clearly show that a slow-to-fast shift of the MHC profile was observed not only in SOL but also in PLA muscles of rats exposed to hypobaric hypoxia during development. The mechanisms responsible for these observations are unclear but could be related to the low levels of locomotor activity recorded in hypoxic rats. Although markedly decreased in CH rats, voluntary running was able to reestablish a profile of MHC isoforms in SOL muscles, similar to that of normoxic rats. Chronic hypoxia induced an increased expression of the fastest MHC isoforms in PLA muscles, and this transition in the myosin profile was unaffected by voluntary exercise. Finally, undernutrition associated with hypoxia does not appear to be responsible for the changes observed in the MHC composition of hindlimb muscles of CH rats.

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REFERENCES


