Comparison of Plantaris Muscle Fibers from Rats and Rabbits during Postnatal Different Ages

ZHOU Dao-ke
(School of Life Sciences, Nantong University, Nantong, Jiangsu Province 226007)

Abstract: The distribution of muscle fibers classified on the basis of their content of different myosin heavy chain (MHC) isoforms was analysed in muscle samples from the plantaris muscles of postnatal different ages of rats and rabbits by correlating standard actomyosin ATPase and succinic dehydrogenase SDHase histochemistry. The percentage of type I, IIX fibers of rats and rabbits decreased whereas that for type II A, IIB fibers increased from postnatal 2 weeks to 24 weeks old. The histochemical characterization of larger number of single fibers was compared and correlated on a fiber-to-fiber basis. The results showed that the mean body weight and the average wet weight of the plantaris muscles was gradually increased with postnatal developments. Type I, IIX, IIA and IIB fibers were found in all muscles of postnatal different age groups except postnatal 2 days. Mean fiber type composition of type IIB fiber of males rats and rabbits was larger than that of female rats and rabbits during postnatal development. Three oxidative histochemically-classified fibers composition of type I, IIX, IIA fiber in male rats and rabbits were smaller than that of female rats and rabbits. The mean cross-sectional area of type I, IIX, IIA and IIB fiber in rats were significantly smaller than those of rabbits. There were evident sexual distinction of rats and rabbits. In rats and rabbits, Type IIX fiber had the smallest cross-sectional area. Type I, IIA fibers had the intermediate in area and type IIB fibers had the largest. This has important implications in attempts to study the physiological properties of fast fiber types adequately in rodents.

Key words: skeletal muscle fiber; rodent; postnatal development; histochemistry

大鼠与家兔生后各年龄阶段跖肌纤维的比较

朱道立
(南通大学生命科学学院、江苏南通 226007)

摘要: 应用建立在肌球蛋白重链异构体基础上的标准肌动球蛋白 ATP 酶和琥珀酸脱氢酶组织化学方法，分析大鼠和家兔出生后发育各年龄阶段跖肌纤维型分布。在出生后 2 周至 24 周龄的大鼠和家兔, IIX 型肌纤维百分比减少, 而 IIA, IIB 型纤维则增加。进行大量单肌纤维的组织化学特征的比较和相关性探讨。结果显示动物平均体重与肌纤维的平均湿重量生后发育逐渐增加。I, IIX, IIA 及 IIB 型纤维均在生后各年龄组的全部肌肉内被发现，但出生后 2 日龄组是个例外。在生后发育期间，雄性大鼠和家兔 IIB 型纤维的平均肌纤维型构成要大于雌性大鼠和家兔，而雄性大鼠和家兔 I, IIX, IIA 型三种氧化组织化学分类的肌纤维型构成均小于雌性大鼠和家兔。大鼠 I, IIX, IIA 及 IIB 型纤维的平均横切面明显要大于家兔的同类型肌纤维要小。大鼠和家兔可见性别的性别差异。大鼠和家兔的 IIX 型纤维横切面积是最小的。I, IIA 型纤维呈中等大小，IIB 型纤维最大。该重要的测试有助于我们深入研究啮齿类动物快慢肌纤维生理特征的适应。

关键词: 骨骼肌纤维; 啮齿动物; 生后发育; 组织化学

Skeletal muscles contain different fiber types characterized by specific myosin heavy chain (MHC) isoforms. The MHC composition of muscle fibers is the main determinant of important functional properties such as the velocity of contraction and fatigue resistance. In mammals, the embryonic and neonatal MHC isoforms expressed in immature muscles are gradually replaced after birth by adult isoforms. Four
major MHCs present in adult muscles of small mammalian species: I, IIX/IID (henceforth IIX), IIA and IIB\(^{[2,3]}\). These isoforms also have a close relationship with differences in muscle function. Consequently, the constitutional proportions of these isoforms change at different stages of growth and development. Based on their physiological types and contractile properties, type I, IIX, IIA and IIB fibers in many species are corresponded to slow-twitch fatigue-resistant (SO), fast-twitch with intermediate fatigue resistance (FI), fast-twitch fatigue-resistant (FR) and fast-twitch fatigue (FF), respectively\(^{[4]}\). In the normal rats and rabbits, type I, IIX, IIA and IIB could be identified\(^{[5,8]}\) and seem to be close to slow-twitch oxidative (SO), fast-twitch oxidative (FO), fast-twitch oxidative glycolytic (FOG) and fast-twitch glycolytic (FG) on the basis of histochemical staining\(^{[7]}\).

Over the past 35 years myosin polymorphism in rat and rabbit skeletal muscle has been extended by the recognition of fiber types I, IIX, IIA and IIB indicated by the actomyosin adenosine triphosphatase (mATPase) histochemical method based on acid and alkaline denaturation. In spite of the good agreement among histochemical, immunohistochemical and electrophoretic techniques for the identification of pure fiber types, the problems still persist regarding the accurate classification of hybrid fibers on the basis of histochemistry alone\(^{[6,8]}\). While hybrid fibers expressing similar amounts of two distinct MHC isoforms stain intermediately between the pure isoforms\(^{[9]}\), the predominance of one isoform can mask the contribution of another myosin present at low levels, i.e. their histochemical profiles will resemble those of the isoforms in dominance. However, hybrid fibers also occur under normal conditions\(^{[10]}\) and their significance is uncertain. If intermediate fibers also represent a significant proportion of the overall muscle fiber composition in rodents, it may have important consequences for the interpretation of results using aerobic enzyme succinic dehydrogenase (SDHase) histochemistry.

We aimed (1) to analyse the distribution of enzyme histochemistry of MHC-classified fibers in plantaris muscles of the postnatal growing rats and rabbits, with particular attention to the presence of fast hybrid fibers, and (2) to examine the possible existence of sexual distinction and regional differences within the muscle.

1 Materials and Methods

1.1 Tissue Preparation

Postnatal 2-day-old (body weight 14–10 g), 2 (40–32 g), 4 (93–81 g), 6 (182–160 g), 8 (224–195 g), 10 weeks old (260–234 g) male and female Sprague-Dawley rats, postnatal 2-day-old (body weight 125–100 g), 2 (234–212 g), 4 (422–394 g), 8 (766–733 g), 12 (1235–1186 g), 16 (1680–1600 g), 20 (2170–2080 g), 24 weeks old (2500–2410 g) male and female Chinese domestic rabbits. Six animals of each age group which came from Nantong University Laboratory Animal Center were used in this study. The rats were anaesthetized with an intraperitoneal (i.p.) injection of compound anaesthetic (0.1 ml / 100 g body mass) and rabbits were anaesthetized with an intravenous (i.v.) injection of 25 % urethane (1.0 g / kg body mass). Plantaris muscles were dissected bilaterally and weighed, then frozen by immersing in gelling isopentane and liquid nitrogen.

1.2 Enzyme-Histochemical Stain

Frozen muscle samples were raised to −20 °C in cryostat, serially sectioned at 10 μm and on coverslips for histochemistry. Serial transverse-sections were stained qualitatively for the demonstration of actomyosin adenosine triphosphatase (mATPase) activity after acid (pH 4.35) and alkaline (pH 10.3) preincubation by using a modification of the Brooke and Kaiser method\(^{[11,12]}\). Additional serial transverse and vertical-sections were also stained for aerobic enzyme succinic dehydrogenase (SDHase) using a modification of the Nachlas\(^{[13,14]}\) method. These serial sections were systematically photomicrographed and the fibers numbered at random and classified by both method.

1.3 Determinations of Muscle Fiber Types and Their Distribution

719
With SDHase methods, the muscle fibers stained high darkly, darkly moderately or weakly. According to the reaction product densities for oxidative enzymes, the stained fibers were interpreted as highest, higher, intermediate, or low in oxidative capacity, respectively. A SDHase-stained section from the middle portion of each muscle sample was randomly selected and photographed using a digital camera (Spot) attached to a Leica light microscope and transferred to a computer with image analysis software. On the selected section, each of the fiber types was manually identified and marked using a computerized image analysis system Jandel Scientific, San Rafael, CA (Sigma Scan). Thus, the computer software recognized the marked fibers and automatically calculated the numbers of each fiber type. The photographs were taken using different magnifications. Low-power view for each section was used to identify the overall distribution patterns of the muscle fiber types. Higher-power view was used to demonstrate the differences in fiber type distribution between different regions.

1.4 Statistical Analysis

The statistical package SPSS. 1 was utilized for all statistical analyses. Descriptive statistics were used to derive mean ± SD for all variables. We appropriate, data were analyzed using student’s t test. Differences were considered significant at $P < 0.05$.

2 Results

2.1 General Morphology and Fiber Types

The mean bodyweight and the average wet weight of the plantaris muscles were gradually increased with postnatal developments. The mean bodyweight and the average wet weight of male rats and rabbits were heavier than those of female rats and rabbits in postnatal growth period (Table 1). The plantaris muscles fibers were relatively loosely packed and had in general more polygonal than strictly rounded contours (Figs. 1, 2). All muscle fibers showed a marked individual and intramuscular variability with regard to diameter and shape of the fibers. The digitorm superficialis flexor muscles were surrounded by a fibrous tissue capsule and contained little amounts of connective tissues and fat. Based on the actomyosin ATPase reaction after preincubation at pH 4.35 and pH 10.3, the muscle fibers could be divided into type I, IIX, IIA, IIB fibers that were found in all muscles of postnatal each age group except postnatal 2 days. According to the extent of their SDHase reaction, type I fibers generally contained a moderately high oxidative enzyme activity and stained dark blue, but less pronounced subsarcolemmal accumulation of reaction product; Type IIX fibers possessed a high level of SDHase activity and stained the darkest blue, the reaction product appearing in the fiber as a coarse network, preferentially distributed in subsarcolemmal regions; Type IIA fibers showed oxidative enzyme activity and stained blue, the reaction product for SDHase was in the form of an open network; Type IIB fibers were characterized by low oxidative enzyme and stained light blue, the homogeneous reaction product distributed in the sarcoplasm (Plate 1, 2).

<table>
<thead>
<tr>
<th>Postnatal ages</th>
<th>Body weight (g)</th>
<th>Wet weight (mg)</th>
<th>Postnatal ages</th>
<th>Body weight (g)</th>
<th>Wet weight (mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>Female</td>
<td>Male</td>
<td>Female</td>
<td>Male</td>
</tr>
<tr>
<td>2 days</td>
<td>14 ± 2</td>
<td>10 ± 1</td>
<td>45 ± 2</td>
<td>42 ± 1</td>
<td>2 days</td>
</tr>
<tr>
<td>2 weeks</td>
<td>40 ± 3</td>
<td>32 ± 2</td>
<td>79 ± 3.5</td>
<td>68 ± 2</td>
<td>2 weeks</td>
</tr>
<tr>
<td>4 weeks</td>
<td>93 ± 4</td>
<td>81 ± 3</td>
<td>118 ± 4</td>
<td>104 ± 3.4</td>
<td>4 weeks</td>
</tr>
<tr>
<td>6 weeks</td>
<td>182 ± 5</td>
<td>160 ± 4</td>
<td>141 ± 5</td>
<td>128 ± 4.5</td>
<td>8 weeks</td>
</tr>
<tr>
<td>8 weeks</td>
<td>224 ± 7</td>
<td>195 ± 6</td>
<td>162 ± 6</td>
<td>145 ± 4.4</td>
<td>12 weeks</td>
</tr>
<tr>
<td>10 weeks</td>
<td>260 ± 9</td>
<td>234 ± 7</td>
<td>188 ± 7</td>
<td>170 ± 5</td>
<td>16 weeks</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>20 weeks</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>24 weeks</td>
</tr>
</tbody>
</table>

All mean values had significant difference for each other ($P < 0.05$).
2.2 Fiber Type Composition

In general, there were systematic and significant differences in fiber type composition between type I, II A, IIB fiber of rats and type I, II X, II A, IIB fiber of rabbits during postnatal development (Table 2, 3). The percentage of type II A, IIB fibers of male rats increased respectively from 15±2 %, 62±3 % at 2 weeks old to 23±2 %, 69±5 % at 10 weeks old. That of female rats were too. The proportion of type IIB fibers in male rats at different stages of postnatal development were larger than that in female rats. The percentage of type I, II X fibers gradually decreased from 16±3 %, 7±2 % male and 20±3 %, 8±3 % female at 2 weeks old to 6±2 %, 2±1 % male and 6±1 %, 3±1 % female at 10 weeks old.

When type I, II X fibers of rabbits varied respectively from 17±2 %, 11±3 % male and 20±3 %, 9±2 % female at 2 weeks old to 7±2 %, 3±1 % male and 8±2 %, 4±1 % female at 24 weeks old. However, type II A, IIB fibers increased from 14±2 %, 58±3 % male and 21±2 %, 50±4 % female at 2 weeks old to 24±4 %, 66±5 % male and 29±3 %, 59±24 % female at 24 weeks old. Overall, we found that mean fiber type composition of type II X and IIB fibers of male rabbits at postnatal growth period were larger than that of female rabbits.

<table>
<thead>
<tr>
<th>Ages</th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>I</td>
<td>II X</td>
</tr>
<tr>
<td>2 d</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>
| 2 w  | 16±3
c | 7±2     | 15±2
d | 62±3
d | 20±3
d | 8±3     | 20±3
d | 52±4
d | 9    |
| 4 w  | 13±3
d | 6±2     | 16±3
d | 65±4   | 17±3
d | 6±2     | 22±4   | 55±3
d | 51±4
c |
| 6 w  | 10±2   | 5±1     | 18±2   | 67±3   | 14±2   | 5±1     | 23±4   | 58±5   | 55±3
d |
| 8 w  | 8±2    | 3±1     | 21±3   | 68±4   | 11±2   | 4±1     | 25±3   | 60±6   | 51±4
c |
| 10 w | 6±2
c | 2±1     | 23±4
c | 69±5
c | 6±1
c | 3±1     | 28±3
c | 62±5
c | 51±4
c |

Data are expressed as mean±SD. Means with different superscripts represent statistical difference (P<0.05).

Table 3 Mean fiber type composition in the plantaris muscles of the male and female rabbits at postnatal development

<table>
<thead>
<tr>
<th>Ages</th>
<th>Male</th>
<th>Female</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>I</td>
<td>II X</td>
</tr>
<tr>
<td>2 d</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2 w</td>
<td>17±2\c</td>
<td>11±3\bc</td>
</tr>
<tr>
<td>4 w</td>
<td>15±3\bc</td>
<td>10±2\c</td>
</tr>
<tr>
<td>6 w</td>
<td>13±3</td>
<td>8±2</td>
</tr>
<tr>
<td>12 w</td>
<td>11±2</td>
<td>7±1</td>
</tr>
<tr>
<td>16 w</td>
<td>9±3</td>
<td>6±2</td>
</tr>
<tr>
<td>20 w</td>
<td>8±2\bc</td>
<td>4±1\b</td>
</tr>
<tr>
<td>24 w</td>
<td>7±2\bc</td>
<td>3±1\bc</td>
</tr>
</tbody>
</table>

Data are expressed as mean±SD. Means with different superscripts represent statistical difference (P<0.05).

A mosaic distribution of type I, II X, II A, IIB fibers were seen in the specimens of the plantaris muscles of postnatal growing rats and rabbits. Any type grouping and enclosed fibers were not found in these muscles of each age group. Type I, II X, II A fibers were distributed in an area situated in the periphery of every muscular cluster.

2.3 Cross-sectional Areas

The cross-sectional areas of muscle fibers are dependent upon the overall size and age of the rat and rabbit. In the plantaris muscles, the mean cross-sectional areas of type I, II X, II A, IIB fiber in rats were significantly smaller than the same type fiber in rabbits. Type II X fiber had the smallest cross-sectional area. Type I, II A fibers had the intermediate in size and type IIB fibers had the largest. The mean cross-sectional areas of type I, II X, II A, IIB fiber in male rats and rabbits were larger than that in female rats and rabbits (Table 4, 5).
Table 4  Mean muscle fiber cross-sectional area (μm² ± SD) of the plantaris muscles of the rats at postnatal development

<table>
<thead>
<tr>
<th>Ages</th>
<th>1</th>
<th>IX</th>
<th>IA</th>
<th>IIB</th>
<th>1</th>
<th>IX</th>
<th>IA</th>
<th>IIB</th>
</tr>
</thead>
<tbody>
<tr>
<td>2d</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2w</td>
<td>210 ± 35</td>
<td>158 ± 18</td>
<td>267 ± 46</td>
<td>358 ± 52</td>
<td>186 ± 31</td>
<td>138 ± 17</td>
<td>248 ± 45</td>
<td>324 ± 48</td>
</tr>
<tr>
<td>4w</td>
<td>312 ± 47</td>
<td>262 ± 21</td>
<td>406 ± 55</td>
<td>537 ± 59</td>
<td>287 ± 39</td>
<td>244 ± 20</td>
<td>376 ± 51</td>
<td>503 ± 49</td>
</tr>
<tr>
<td>6w</td>
<td>488 ± 49</td>
<td>399 ± 27</td>
<td>651 ± 63</td>
<td>896 ± 63</td>
<td>466 ± 45</td>
<td>372 ± 26</td>
<td>611 ± 57</td>
<td>861 ± 56</td>
</tr>
<tr>
<td>8w</td>
<td>894 ± 56</td>
<td>714 ± 33</td>
<td>998 ± 72</td>
<td>1388 ± 84</td>
<td>871 ± 48</td>
<td>688 ± 29</td>
<td>962 ± 62</td>
<td>1330 ± 76</td>
</tr>
<tr>
<td>10w</td>
<td>1198 ± 61</td>
<td>988 ± 39</td>
<td>1458 ± 77</td>
<td>1868 ± 89</td>
<td>1173 ± 54</td>
<td>967 ± 36</td>
<td>1405 ± 72</td>
<td>1827 ± 79</td>
</tr>
</tbody>
</table>

The mean cross-sectional area of four types of fibers were significantly different (P<0.05).

Table 5  Mean muscle fiber cross-sectional area (μm² ± SD) of the plantaris muscles of rabbits at postnatal development

<table>
<thead>
<tr>
<th>Ages</th>
<th>1</th>
<th>IX</th>
<th>IA</th>
<th>IIB</th>
<th>1</th>
<th>IX</th>
<th>IA</th>
<th>IIB</th>
</tr>
</thead>
<tbody>
<tr>
<td>2d</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2w</td>
<td>244 ± 21</td>
<td>192 ± 16</td>
<td>298 ± 32</td>
<td>396 ± 47</td>
<td>221 ± 19</td>
<td>171 ± 15</td>
<td>272 ± 29</td>
<td>346 ± 44</td>
</tr>
<tr>
<td>4w</td>
<td>409 ± 27</td>
<td>357 ± 19</td>
<td>495 ± 36</td>
<td>655 ± 59</td>
<td>385 ± 21</td>
<td>331 ± 18</td>
<td>449 ± 33</td>
<td>612 ± 51</td>
</tr>
<tr>
<td>8w</td>
<td>566 ± 31</td>
<td>496 ± 24</td>
<td>789 ± 32</td>
<td>998 ± 79</td>
<td>531 ± 30</td>
<td>448 ± 23</td>
<td>741 ± 30</td>
<td>946 ± 68</td>
</tr>
<tr>
<td>12w</td>
<td>988 ± 36</td>
<td>748 ± 26</td>
<td>1286 ± 52</td>
<td>1588 ± 86</td>
<td>954 ± 32</td>
<td>702 ± 22</td>
<td>1238 ± 48</td>
<td>1512 ± 77</td>
</tr>
<tr>
<td>16w</td>
<td>1198 ± 42</td>
<td>987 ± 29</td>
<td>1497 ± 54</td>
<td>1884 ± 94</td>
<td>1157 ± 37</td>
<td>936 ± 28</td>
<td>1455 ± 49</td>
<td>1827 ± 89</td>
</tr>
<tr>
<td>20w</td>
<td>1399 ± 51</td>
<td>1162 ± 32</td>
<td>1768 ± 61</td>
<td>2165 ± 87</td>
<td>1349 ± 99</td>
<td>1107 ± 29</td>
<td>1719 ± 57</td>
<td>2105 ± 85</td>
</tr>
<tr>
<td>24w</td>
<td>1655 ± 48</td>
<td>1457 ± 35</td>
<td>2064 ± 68</td>
<td>2538 ± 93</td>
<td>1610 ± 42</td>
<td>1406 ± 34</td>
<td>2019 ± 62</td>
<td>2488 ± 86</td>
</tr>
</tbody>
</table>

The mean cross-sectional area of four types of fibers were significantly different (P<0.05).

3 Discussion

The size of the muscle fibers is depend upon the age, sex and body weight of the animal. Hence, larger rats and rabbits will have muscle fibers with a greater cross-sectional area. Although it is difficult to make comparisons between studies, cross-sectional area data gathered from the present investigation are similar to previously published data from rats and rabbits of comparable age, sex, body weight, and strain \cite{15,16}. Various strains of rats and rabbits used in the present study have yielded cross-sectional areas and individual muscle wet weights paralleling the increase in body weight. The sexual distinction in rats and rabbits were evident during postnatal different growth stages. The body weight and muscle wet weight of male rats and rabbits were heavier than those of female rats and rabbits during postnatal development.

In the plantaris muscles of rats and rabbits, the percentage of type I and IIX fibers had a low percentage, while the proportion of type IIA and IIB fibers was higher than that in the postnatal other ages. Additionally, in the medial gastrocnemius, digitorum superficialis flexor muscle and extensor digitorum longus \cite{17}, regional differences in fiber composition were detected along the muscle length. As muscle fibers in different regions do not play a similar role in these muscles, they are recognized to be highly compartmentalized. Though muscle compartments seem to be established early in development, probably prior to the differentiation of actual patterns of fast or slow fibers, a recent study in rabbits and rats demonstrated that innervation was a determining factor for differentiation into slow-twitch muscle \cite{17,18}. Therefore, neuromuscular activity through innervation could be important for developing muscle compartments as well as regional differences in fiber types of fast- twitch muscle.

As regards fiber types, knee and ankle muscles showed the greatest specialization. Plantaris muscles, the major flexor muscle investigated in the hindlimb, was made up primarily of type IIB fibers, with a small amount of type IIA and a negligible percentage of type I, IIX. Such near absence of slow fibers in the plantaris muscles is a general pattern in small quadruped mammals such as the rat, mouse and rabbit. In male and female rats the content of type IIB
fiber was separately increased from 62 ± 3% and 52 ± 4% of postnatal 2 weeks to 69 ± 5% and 62 ± 5% of postnatal 10 weeks, and yet in male and female rabbits that was respectively increased from 58 ± 3% and 50 ± 2% of postnatal 2 weeks to 66 ± 5% and 59 ± 4% of postnatal 10 weeks. Such a high percentage of glycolytic fibers (IIB), able to produce bursts of acceleration, can related to running and jumping biomechanics during postnatal development\(^{[19]}\).

Myosin heavy chain (MHC) isoform are one of the most important of the myofibrillar proteins that define the contractile characteristics of muscle fibers. The existence of several MHC isoforms, each with their own special characteristics, enables the skeletal muscles to fulfill different physiological demands during postnatal different development. In small mammals (mouse, rat, guinea pig, rabbit) three fast MHC isoforms are expressed (MHCIIIX, IIa, IIB) as well as one slow MHC I isoform. There is a high correlation between the MHC isoform content and the contractile properties of a muscle fiber. Studies of the isolated skinned fibers of rodents showed that the maximal shortening velocity of fiber types increased in the following manner: IIIX < I < IIa < IIB. But in larger mammals, like the horse, cattle and humans, only two fast isoforms, MHCIIa and MHCIIIX, have been demonstrated\(^{[20]}\). It has been hypothesized that if the fastest MHCIIb isoform were to be expressed in large mammals, due to their length, the shortening of the MHCIIb fibers would produce such a high intrinsic velocity that it would lead to the development of an unsustainable force. However, there must be other factors beside body mass that determine MHCIIb expression, as three fast isoforms have been demonstrated in the domestic pig and the llama\(^{[21]}\). Recently, gene coding for MHCIIb has even been discovered in humans; thus, its expression in skeletal muscles remains to be confirmed.

The muscle fiber types in rats and rabbits change with age and spontaneous activity from birth until adult. The relative frequency of SDHase I, IIIX fibers in the plantaris muscle of rats and rabbits decreases over the postnatal growth period and that of IIa, IIB fibers increases after birth. This study showed that there was a high correlation between MHC content and SDHase reaction, except when the 2 fast MHCs coexist within the same fiber\(^{[22]}\). Some recent quantitative histochemical and biochemical studies showed that fibers with homogeneous MHC content varied greatly with their SDHase reactivity. This suggests that some other factors, in addition to MHC content, may contribute to the SDHase histochemical reaction and explain the limitations of this technique in objective differentiation between fast fibers based on MHC content. A suggestion was made of the following transition pathway of MHC isoforms: 1 → II1/11A → IIIX → IIIX/IIIB → IIb. This observation means that oxidative capacity of a fiber cannot be used as a firm predictor of its MHC content. In fact, there is enough evidence that the contractile protein (e.g., MHC) genes and metabolic (e.g., SDHase) genes are not necessarily under the control of the same mechanism\(^{[23]}\). Nevertheless, since sequential transitions between MHC isoform seems to be dictated by energetic requirements of postnatal development, the energy potential of myofibers seems to play a regulatory role in determining MHC isoform profile.

Some genes expressed only in fast fibers contain a regulatory element, known as the fast intrinsic regulatory element (FIRE), which contains elements that appear to be necessary for fast fiber gene expression. Lakich et al. (1998) indicated that the mouse MHCIIb proximal promoter contained two distinct A/T rich regions in which MEF2 and OCT-1 bound separately, but in a cooperative manner which determined activation of this fast muscle specific promoter. Allen et al. (2001) also found that the mouse MHCIIb promoter was activated by MyoD, however they also found that myogenin and myf-5 could activate the promoter. Additional data indicate that MyoD is not the only factor involved in activating expression of MHCIIb, as suggested by the observation that some MyoD-null muscles express MHCIIb mRNA and protein at greater levels. Nevertheless, MyoD plays a very important, but not a singular, role in
modulating expression of MHCIIb\(^{24}\).

In the plantaris muscle, all types of muscle fibers of rats were significantly smaller than the same type fibers of rabbit. We found that in the plantaris muscle the mean cross-sectional area fell in rank order of type IIB > IIA > I > IX fibers. This was in accordance with other studies of the plantaris muscle and other leg muscles\(^{17,19}\). The fact that type IX fibers were considerably smaller in diameter than other fiber types can therefore be included in subtype IIA. Type IIX and I fibers tended to be more concentrated in deep limb muscles and deep portions of muscles clusters. Whereas type IIA and IIB fibers tended to be most concentrated in superficial muscles and periphery portions of muscles clusters. A reduction in fiber size of type IIX fibers in skeletal muscles is said to be associated with denervation and also with oxidative activity. The result showed that both the intra- and inter-individual variability of the cross-sectional area of type I fibers was relatively small, while the variability of the cross-sectional areas of type IIA and IIB fibers was relatively large (Table 4, 5). The unique great size of type IIB fibers in the plantaris muscle of rats and rabbits is probably related to hopping locomotion, requiring long-lasting resting posture on the hindlimbs and braking action at landing during long postnatal growth. This suggests that type II and hybrid fibers types are more prone to change than type I fibers.

4 References


[20] Strithe M, Smarac V, Zupec M, Taton N and Fazmayne G. Pat-
赤狐华南亚种与东北亚种上毛髓质指数的比较研究

唐福全 1, 张伟 2
(1. 四川农业大学都江堰分校, 四川都江堰 611830; 2. 东北林业大学)

摘要：选取赤狐华南亚种和东北亚种 8 个部位上毛，用测试格测定上毛的髓质指数。结果表明，赤狐华南亚种和东北亚种上毛髓质指数在同一亚种的不同部位及亚种间差异显著（P < 0.01）。无论在总体平均值上，还是在不同亚种个体同一部位上，赤狐东北亚种上毛的髓质指数均大于华南亚种。两亚种躯干部上毛髓质指数的平均值均明显大于前肢。这种差异性表现在从前肢末端到动物躯干部呈梯度递增，其递增梯度在两个亚种的个体中表现出同步变化的趋势。

关键词：赤狐；上毛；髓质指数

中图分类号：Q959.8 文献标识码：A 文章编号：1000－7083（2006）04－0725－05

Comparative Study on Medulla Exponent of Upper Hair between South and Northeast Subspecies of Vulpes

TANG Fu-quan 1, ZHANG Wei 2
(1. Dujiangyan Campus of Sichuan Agricultural University, Dujiangyan, Sichuan Province 611830; 2. North-East Forest University)

Abstract: After choosing upper hair of eight positions from the south and northeast subspecies of Vulpes, we tested its medulla exponent by using test lattice. The result indicated that there existed significant difference between different positions of the same subspecies and between different subspecies (P < 0.01). The medulla exponent of upper hair of northeast subspecies was bigger than that of the south either in total average or in the same position of different subspecies. The average of medulla exponent from the bodies of two subspecies was also bigger than that of their forelegs. The difference increased progressively from the end of the foreleg to the body of Vulpes. The individuals of these two subspecies had the same gradient increasing tendency.

Key words: red wolf; upper hair; medulla exponent

毛发的研究已经有一段相当长的历史，20 世纪 70 年代以后，主要借助于光学显微镜观察和研究毛的外部形态，毛的髓质、皮质[1～5]。随着科学技术的进步，电子显微镜的诞生，为毛发的研究提供了更为优越的条件，人类可更进一步观察毛表面的鳞片类型[6～7]，使研究的结果进一步客观、精确。纵观这些研究成果，有关毛髓质方面的研究都比较笼统，研究报道仅限于反映髓质的类型是否发达等方面，既没有系统地进行比较，也没有将髓质的发达程度数量化。后来有人把毛横截髓质所占百分比作为毛的髓质指数进行了有关比较研究，将描述性研究转变为数量化研究[8,9]。这些数量化研
Plate 1. Serial transverse sections and vertical section of the plantaris muscles of postnatal 2 days (1,2), 2 weeks (3, 4), 4 weeks (5, 6), 6 weeks (7, 8), 8 weeks (9, 10), 10 weeks (11, 12) old male SD rats stained for succinate dehydrogenase (SDHase) activities. Type I: dark blue, Type IIIX: darkest blue, Type IIA: blue, Type IIB: light blue. ×100~400

Plate 2. Serial transverse sections and vertical section of the plantaris muscles of postnatal 2 days (1,2), 2 weeks (3, 4), 4 weeks (5, 6), 8 weeks (7, 8), 12 weeks (9, 10), 16 weeks (11, 12), 20 weeks (13, 14), 24 weeks (15, 16) old male CD rabbits stained for succinate dehydrogenase (SDHase) activities. Type I: dark blue, Type IIIX: darkest blue, Type IIA: blue, Type IIB: light blue. ×100~400