Expression of lipoprotein lipase in rat muscle: regulation by feeding and hypothyroidism

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Abstract Lipoprotein lipase (LPL) is a key enzyme in lipid metabolism and is found predominantly in adipose tissue and muscle. We examined the mechanism of regulation of LPL in muscles composed of different fiber types (soleus, extensor digitorum longus, and heart) in fed, fasted, and hypothyroid rats. In all muscles, the detergent-extractable (EXT) fraction represented approximately 95% of total LPL activity and mass. LPL activity was similar in the heparin-releasable (HR) fractions of heart and soleus (predominantly type I fibers), while in the EXT fraction LPL activity in soleus was 418 ± 48 nEq/min per g, and in heart was 272 ± 30 nEq/min per g ($P < 0.05$). However, LPL activity in extensor digitorum longus (EDL, predominantly type II fibers) was considerably lower (7.9 ± 0.8 nEq/min per g in EXT, $P < 0.0001$ versus heart and soleus). LPL immunoreactive mass followed a pattern similar to LPL activity. LPL mRNA levels were quantitated by both Northern blotting and reverse transcriptase-polymerase chain reaction (RT-PCR), and were approximately equal in heart and soleus, and 5-fold lower in EDL. In response to feeding, LPL activity, mass, and mRNA levels in heart were 30% to 50% lower than in fasted rat heart, although feeding had no effect on soleus or EDL. In hypothyroid animals, muscle LPL activity was increased by 3- to 4-fold in the HR (but not EXT) fractions of heart and soleus ($P < 0.05$), with no change in LPL mass or mRNA. Thus, muscles with oxidative, type I fibers expressed higher levels of LPL mRNA than muscles containing glycolytic, type II fibers. Feeding affected heart LPL with no effect on soleus or EDL. Soleus and heart from hypothyroid rats demonstrated increased HR LPL, with no change in LPL mass or mRNA, suggesting posttranslational regulation by thyroid hormone.—**Ong, J. M., R. B. Simsolo, M. Saghizadeh, A. Pauer, and P. A. Kern.** Expression of lipoprotein lipase in rat muscle: regulation by feeding and hypothyroidism. *J. Lipid Res.* 1994. **35:** 1542–1551.

Supplementary key words muscle fibers • messenger RNA • reverse transcriptase-polymerase chain reaction • posttranslational processing

Lipoprotein lipase (LPL) is the rate-limiting enzyme in triglyceride-rich lipoprotein metabolism and is expressed in a variety of tissues. In adipose tissue, LPL hydrolyzes circulating triglyceride into free fatty acids (FFA) which are reesterified for subsequent lipid storage (1), while in muscle tissue, LPL delivers FFA which is catabolized in muscle tissue to provide energy (2). LPL is often regulated inversely in adipose tissue and muscle, such that increases in adipose LPL or decreases in muscle LPL may be important in lipid partitioning towards adipose tissue storage and the development of obesity (3). Because of the intimate relationship between adipose lipid storage by LPL and obesity, much research has been devoted to the regulation of adipose tissue LPL by hormones and other physiologic events, and regulation of LPL has been described at multiple levels of gene expression (4). However, comparatively less attention has been paid to the mechanism of regulation of muscle LPL.

All muscles are composed of a mixture of different fiber types, which can be classified according to contractile and/or metabolic characteristics (5). Fast-twitch fibers (type II) have a high rate of cross-bridge turnover, and generate energy quickly through glycolysis and anaerobic metabolism for forceful, short-term contractions. In contrast, slow-twitch fibers (type I) have a slow speed of contraction and a less developed glycolytic system, but large and numerous mitochondria. Type I fibers are fatigue-resistant, rely on aerobic metabolism for energy, and are particularly well suited for the oxidation of FFA. Previous studies have suggested that muscles composed predominantly of type I fibers have higher levels of LPL activity (6–11).

Among the hormones that are known to affect muscle metabolism is thyroid hormone. Although rats and humans tend to respond in an opposite manner to thyroid hormone (reviewed in refs. 1 and 2), the effects of thyroid

Abbreviations: LPL, lipoprotein lipase; HR, heparin-releasable LPL; EXT, extractable LPL; FFA, free fatty acids; SDS, sodium dodecyl sulfate; EDL, extensor digitorum longus; RT-PCR, reverse-transcriptase polymerase chain reaction; PBS, phosphate-buffered saline; ELISA, enzyme-linked immunosorbent assay.

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hormone on muscle LPL have been conflicting. For example, when LPL activity was examined in the hearts of hypothyroid rats, some studies found an increase in LPL activity (12, 13), whereas others found a decrease or no change (14, 15). The reasons for these differences, and the mechanism of change in LPL due to thyroid hormone, are unclear. The data are more consistent with adipose tissue, and a number of studies have demonstrated an increase in LPL activity with hypothyroidism in rats (reviewed in ref. 1). We have further examined the mechanism of this increase in adipose LPL and found that thyroid hormone decreases LPL translation in adipose tissue (16).

In this study, we examined the mechanism of LPL regulation in muscle composed of different fiber types. In addition, we examined the effects of hypothyroidism and feeding on LPL expression in these different muscles.

METHODS

Animals and tissues

Male Sprague-Dawley rats weighing between 180 and 220 g were killed after an overnight fast (4 PM to 9 AM), and the muscle tissues were removed immediately. Fed animals were allowed free access to laboratory chow up until the time of killing. The muscles that were dissected included heart, soleus, which is composed of 84% type I, or slow-twitch oxidative fibers, and the extensor digitorum longus (EDL), which is composed of 97% type II, or fast-twitch glycolytic and intermediate fibers (5). Hypothyroid rats had had their thyroids removed surgically 6–8 weeks prior to being killed, and pooled serum uniformly demonstrated elevated TSH levels. Control rats were weight matched; because hypothyroid rats tend to gain weight more slowly, the control rats were slightly younger than the hypothyroid rats. There were no differences in serum glucose or lipids between control and hypothyroid rats.

Measurement of LPL activity

LPL catalytic activity was measured in the fraction released by heparin (HR), as well as in the fraction remaining after heparin release that was found in cell extracts (EXT) (17), as described previously. In brief, muscle was minced and incubated in phosphate-buffered saline (PBS) containing 13 μg/ml heparin (Fisher Scientific Co.) for 30 min at 37°C. An aliquot of this buffer was then assayed as described below. The cells were then washed and the EXT fraction was prepared by homogenizing the muscle tissue in buffer containing deoxycholate and heparin, as described previously (17). LPL activity was then measured in a 10-μl aliquot of the supernatant after centrifugation. Control experiments demonstrated that this method removed all inhibitory effects of the detergent.

LPL activity was measured using a [^{3}H]triolein-containing substrate emulsified with lecithin, and containing normal human serum as a source of apolipoprotein C-II (18). After incubating the samples with substrate for 45 min at 37°C, liberated [^{3}H]-FFA were separated and quantitated by liquid scintillation. Activity was expressed as nEq FFA released/min per g.

LPL immunoreactive mass

The measurement of LPL immunoreactive mass used an enzyme-linked immunosorbent assay (ELISA), which has been described previously (17, 19). In brief, HR and EXT samples for LPL immunoreactive mass were prepared using protease inhibitors (1 mM phenylmethylsulfonyl fluoride, 1 mM benzamidine, and 1 mM EGTA) in all the buffers. The sandwich-type ELISA uses affinity-purified chicken anti-bovine LPL antibodies (19) as a capture antibody. After addition of sample or bovine LPL standard, LPL is quantitated by the addition of biotinylated affinity-purified anti-LPL antibody, followed by streptavidin–peroxidase. The concentration of LPL in the samples was then calculated using the standard curve for bovine LPL, and expressed as ng/g for muscle.

RNA extraction and Northern analysis

RNA was extracted using the method of Chomczynski and Sacchi (20) with minor modifications (16). Equal amounts of total RNA were resolved on a 2.2 M formaldehyde–1% agarose gel, transferred to nylon membrane, and blotted with the ^{32}P-labeled cDNA probes for human LPL (21), γ-actin (22), or cyclophilin (23), as described previously (16). To further demonstrate that equal quantities of total RNA were loaded, samples of the RNA were resolved by electrophoresis and stained with ethidium bromide for quantitation of the 28S and 18S rRNA bands. To quantitate differences between Northern blots, autoradiographic images were quantitated by densitometry on a laser image analyzer.

Quantitative RT-PCR

Quantitative competitive reverse-transcriptase polymerase chain reaction (RT-PCR) was performed as described previously (24, 25). RNA was extracted and quantitated as described above, and equal quantities of RNA were added to increasing quantities of a rat LPL cRNA construct that contained primer sites for rat LPL. The cRNA was constructed by RT-PCR of a 306 base pair sequence (nucleotides 1303 to 1608) using primers containing restriction sites for cloning into the RNA vector pGEM-4Z (Promega). An internal deletion of 19 nucleotides was then made by cutting with PstI, followed by religation of the plasmid. This construct was confirmed by

double-stranded DNA sequencing (USB, Sequenase sequencing kit). When this construct was transcribed, the resulting LPL mRNA was 19 nucleotides shorter than native LPL mRNA, and therefore distinguishable on an agarose gel. To quantitate LPL mRNA from tissue, known amounts of the LPL cRNA were added to a fixed quantity of extracted RNA. For soleus and heart, 2–4 ng of total RNA was used for the reaction, and for EDL, 50 ng of total RNA was used. The primers sites for the RT-PCR reaction were located at nucleotides 1303 to 1322, and 1589 to 1608 of the rLPL cDNA, which spans an intron in the LPL gene (to avoid contamination with genomic DNA). Following the reverse transcriptase reaction, PCR was carried out for 35 cycles at 55°C. The resulting ethidium bromide-stained gel was imaged using an Imagestore 5000 scanner, and analyzed using the Gelbase/Gelblot software (Ultraviolet Products, Ltd., San Gabriel, CA). The ratio of LPL product/cRNA standard was plotted against the number of copies of cRNA added, to yield the equivalence point between cRNA and LPL mRNA. The data are expressed as the number of copies of LPL mRNA per ng total RNA.

Statistics

All data were expressed as the mean ± SEM, and differences between groups were analyzed using one-way analysis of variance (ANOVA). The n for each group, which represents the number of animals, is stated in the legends.

RESULTS

Expression of LPL in different muscle types

To study the regulation of muscle LPL in rats, two different skeletal muscles, soleus and extensor digitorum longus (EDL), were dissected from male Sprague-Dawley rats, along with the heart. Heart and soleus had similar high levels of heparin-releasable (HR) and extractable (EXT) LPL activity (**Fig. 1, A and B**). However, LPL activity in EDL was considerably less than that of the other two muscle tissues. HR and EXT activities of EDL were 0.6 ± 0.1 and 7.9 ± 0.8 neq/min per g, whereas HR and EXT LPL activities of heart were 6.1 ± 0.8 and 272 ± 70 neq/min per g, respectively. To determine whether there was a similar difference in LPL protein between EDL, soleus and heart, LPL immunoreactive mass was measured by ELISA (Fig. 1, C and D). The levels of LPL immunoreactive mass paralleled the differences in LPL activity in both HR and EXT. The levels of both HR and EXT LPL mass were much higher in heart and soleus than in EDL. As shown in Fig. 1, all of these muscle tissues express approximately 95% of their LPL catalytic activity and immunoreactive protein mass in the EXT fraction.

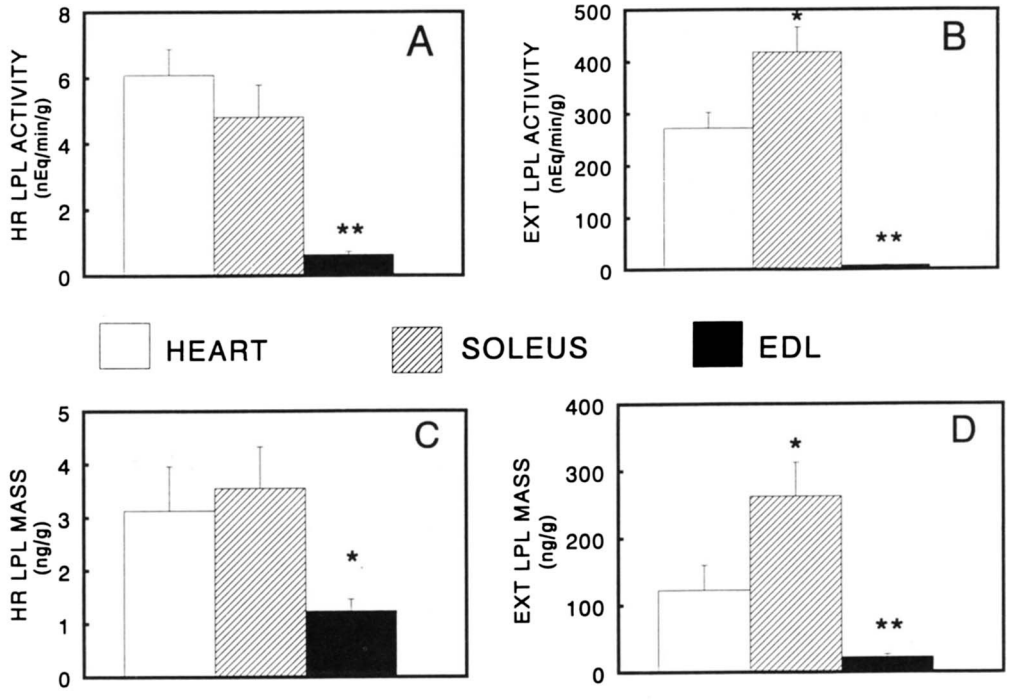


Fig. 1. Expression of lipoprotein lipase in different muscle types. Heart and skeletal muscles of soleus and extensor digitorum longus (EDL) were excised from male Sprague-Dawley rats followed by measurements of HR (heparin releasable) (A) and EXT (extractable) (B) LPL catalytic activity (A and B) and LPL immunoreactive protein mass (C and D). Data are expressed as the mean ± SE with n = 21–28; $^{*}P < 0.05$ vs. heart; $^{**}P < 0.0005$ vs. heart and soleus.

Total RNA was extracted from soleus, heart, and EDL and subjected to Northern blotting analysis for the expression of the LPL gene. Equal amounts of total RNA were added to the gels, and blots were hybridized with a cDNA probe to LPL, as well as cDNA probes to γ-actin and cyclophilin. LPL mRNA levels from soleus and heart were approximately equal, whereas EDL had much lower levels of LPL mRNA (**Fig. 2**). Although the level of LPL expression was similar in soleus and heart, the level of γ-actin expression was much lower in heart, in spite of equal loading of total RNA. To be certain of equal gel loading, the blot was rehybridized with the cDNA for cyclophilin, which demonstrated no difference between heart and skeletal muscle. Thus, the significantly decreased level of LPL catalytic activity in EDL was reflected in decreased levels of both LPL immunoreactive protein mass and LPL mRNA.

The level of LPL expression in EDL was low, requiring the development of a more sensitive method for measurement of LPL mRNA. Competitive RT-PCR was performed, as described in Methods, to more precisely determine the relative abundance of LPL mRNA in the different muscle tissues. As shown in **Fig. 3A,** increasing quantities of the rLPL cRNA standard were added to

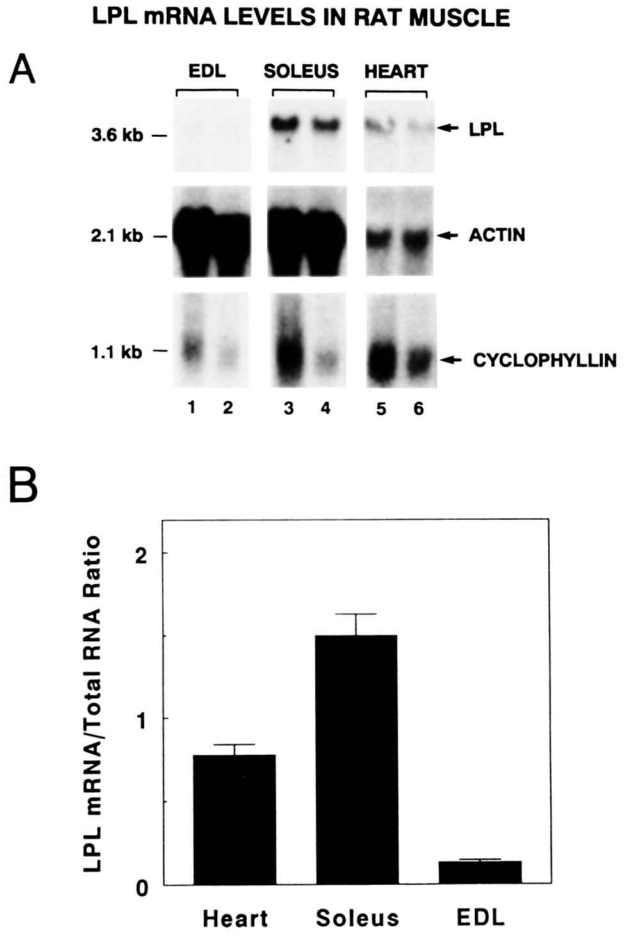


Fig. 2. Levels of LPL mRNA expression in different muscle types. Total RNA was extracted from each muscle tissue and analyzed by Northern blotting with ^{32}P-labeled cDNA probes for LPL, γ-actin, and cyclophilin. A: Representative Northern blots from each muscle type; B: LPL/total RNA ratios from all Northern blots; n = 6–8 for each muscle type.

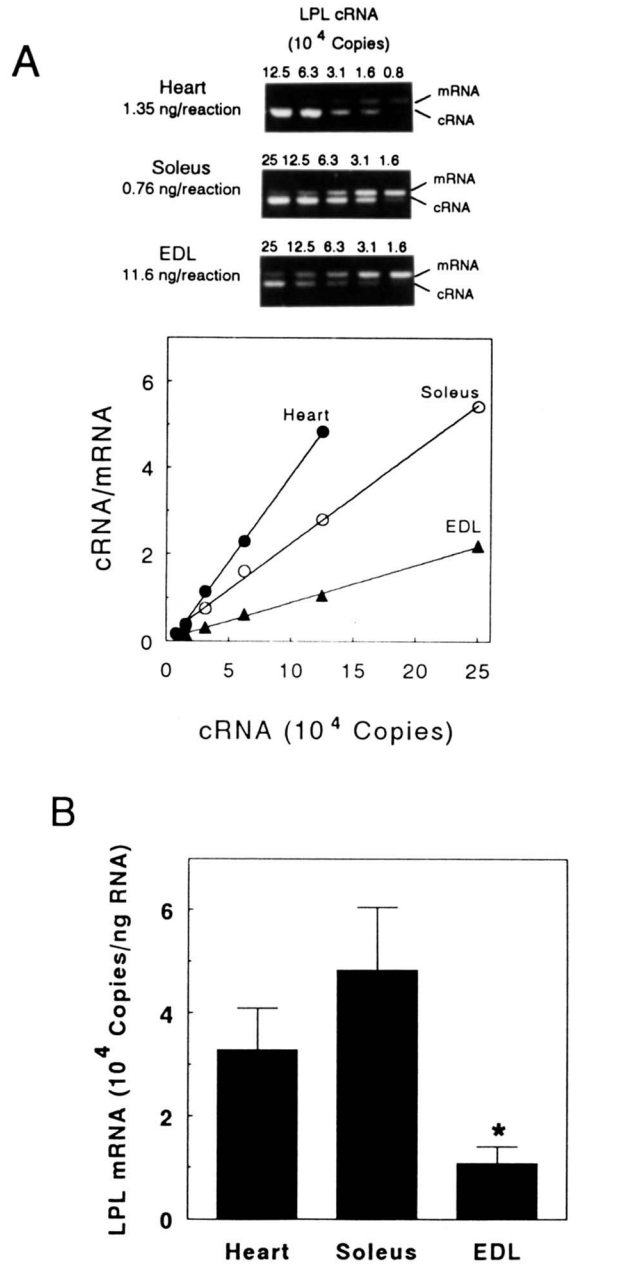


Fig. 3. Quantitative RT-PCR of rat muscles. A: Equal quantities of total RNA were added to increasing quantities of rLPL cRNA, followed by RT and PCR. The resulting image was quantitated, and the ratio of cRNA/mRNA was plotted against the amount of cRNA added (see Methods). B: Quantitation of LPL mRNA in rat muscles using quantitative RT-PCR. Data are from 4–9 animals, and are expressed as the mean ± SEM; $^*P < 0.02$ vs. heart and soleus.

samples of total RNA from each muscle, followed by RT-PCR. The ratio of cRNA/mRNA was plotted against the number of copies of cRNA added, and the equivalence point was taken as the number of copies of LPL mRNA present in the sample. Using competitive RT-PCR, LPL mRNA levels were quantitated in the EDL, soleus, and heart muscles from four to five animals, expressed as number of copies of cRNA/ng total RNA. As shown in Fig. 3B, heart and soleus had 3.3 ± 0.8 and 4.8 ± 1.2 × 10^4 copies/ng RNA, and EDL had 1.1 ± 0.3 × 10^4 copies ($P < 0.02$).

Effects of feeding

The above experiments were performed in fed rats, and feeding is known to result in a decrease in muscle LPL activity (2). To determine whether heart, soleus, and EDL respond differently to feeding, LPL activity and mass were measured in fasted animals, and compared to fed animals. As shown in **Fig. 4,** feeding resulted in a significant decrease in heart LPL activity and mass, although only in the HR fraction. In contrast, there were no significant changes in soleus or EDL activity or mass in either the HR or EXT fractions.

To assess LPL mRNA levels in fed and fasted rat muscles, Northern blotting and competitive RT-PCR were performed. As shown in **Fig. 5,** Northern blotting of heart total RNA revealed a 30% decrease in LPL mRNA in the fed rats ($P < 0.001$). This decrease in LPL mRNA was consistent when expressed in relation to the γ-actin message or to total RNA. The level of expression of LPL was lower in EDL, and with Northern analysis it was difficult to quantitate small differences in mRNA level. Therefore, competitive RT-PCR was performed, as described in Methods, to examine the effects of feeding on soleus and EDL LPL mRNA levels. As shown in **Fig. 6A,** increasing quantities of the rLPL cRNA standard were added to samples of total RNA from each muscle, followed by RT-PCR. As described above, the ratio of cRNA/mRNA was plotted against the number of copies of cRNA added, and the equivalence point was taken as the number of copies of LPL mRNA present in the sample. Using competitive RT-PCR, LPL mRNA levels were quantitated in the EDL and soleus muscles from nine animals (five fasted, four fed), and the data are shown in Fig. 6B. Feeding had no significant effect on LPL mRNA levels in these muscles.

Regulation of muscle LPL by thyroid hormone

To assess the effects of thyroid hormone on muscle LPL, muscle LPL expression was measured in rats 6

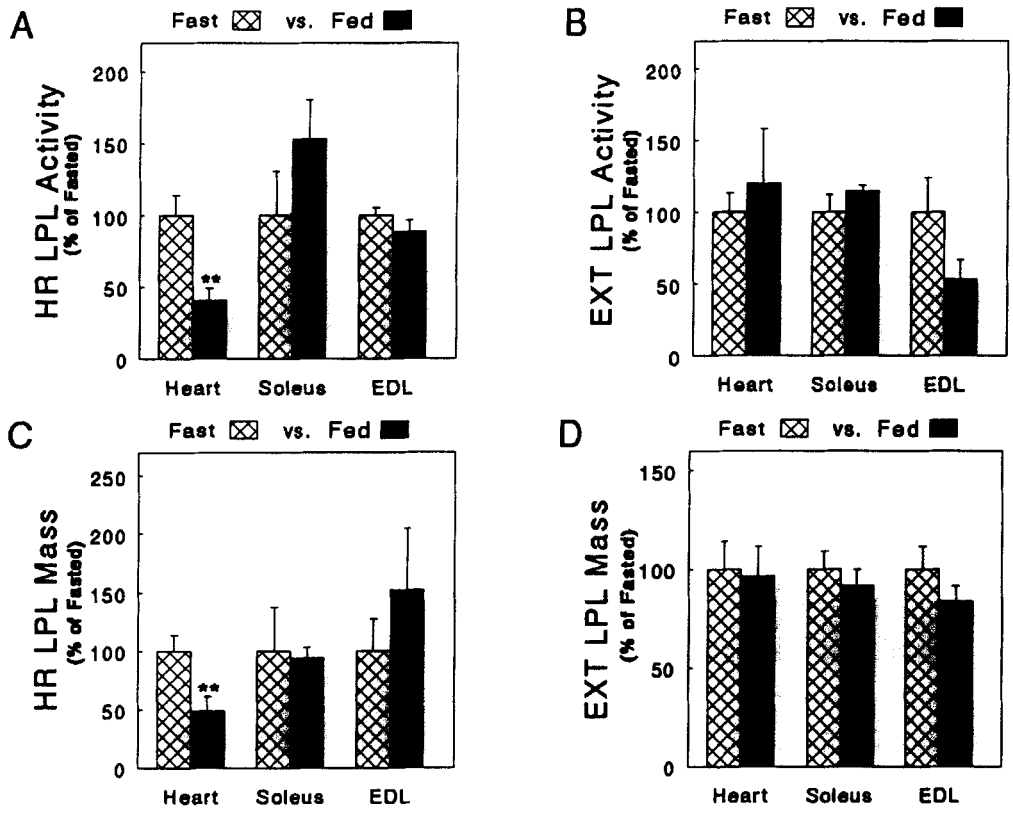
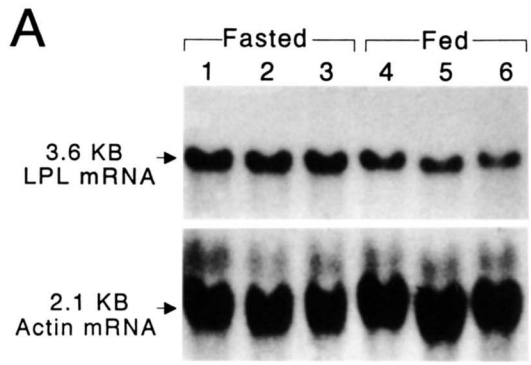


Fig. 4. Effects of feeding on muscle LPL. LPL activity and mass were measured in heart, soleus, and EDL from fed rats, and from 24-h fasted rats; $^{**}P < 0.005$; n = 6–8 animals for each measurement.



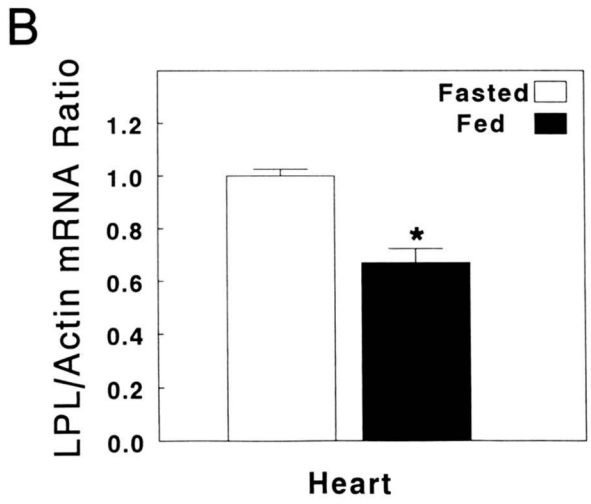


Fig. 5. Effect of feeding on muscle LPL mRNA levels. A: Heart LPL mRNA levels were determined in fed and 24-h fasted rats using Northern blotting. The representative blot is from three fed animals and three fasted animals from the same group. B: The LPL/γ-actin ratio from all animals (n = 7–8 for each group) was quantitated, and expressed in relation to the LPL/γ-actin ratio in fed animals; $^*P < 0.001$.

weeks after thyroidectomy. LPL activity and LPL immunoreactive protein mass of these thyroidectomized animals were compared with those of normal control rats (**Fig. 7**). In the hypothyroid rats, there were significant increases in HR LPL activity of soleus and heart. Although there was a trend towards an increase in HR activity of EDL, this did not reach statistical significance. In addition, there was no significant increase in EXT LPL activity for any of the tissues examined as a result of the hypothyroidism. Upon analysis of the LPL immunoreactive protein mass of thyroidectomized rats, there were no significant differences in the three tissues examined for both HR and EXT fractions when compared to control rats.

LPL mRNA levels were assessed in hypothyroid and normal rats by Northern blot analysis. The agarose gel was stained with ethidium bromide prior to transfer of the RNA in order quantitate the amount of total RNA loaded onto the gel. Based on the levels of the 28S and 18S rRNA bands, there were no significant differences in LPL mRNA between hypothyroid and normal animals in all three muscle tissues examined (**Fig. 8**).

DISCUSSION

Lipoprotein lipase (LPL) is an essential enzyme in lipid metabolism. In addition to its role at regulating plasma lipemia, LPL provides FFA substrate for the tissues in

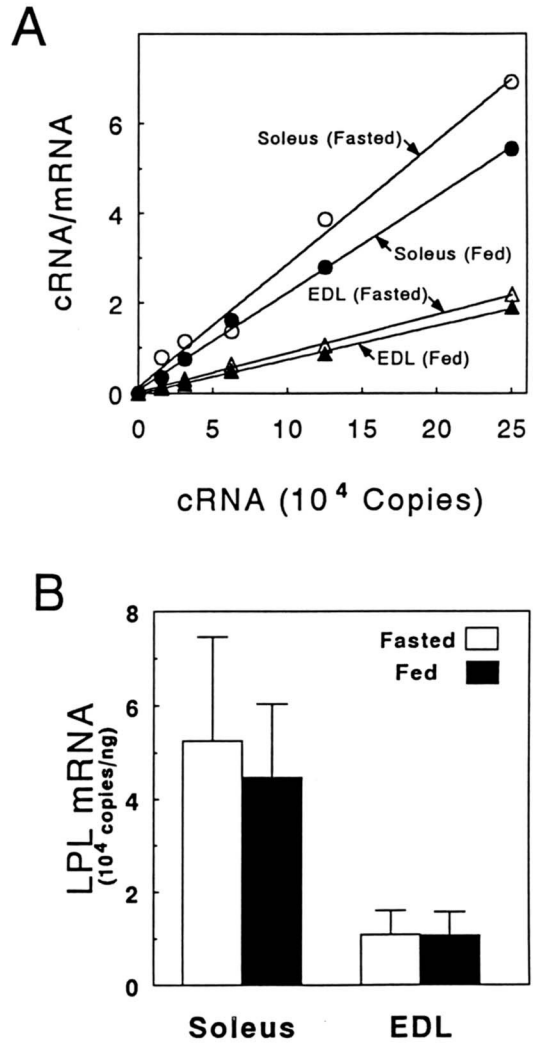


Fig. 6. Effect of feeding on soleus and EDL. Competitive RT-PCR was used to measure LPL mRNA levels in soleus and EDL. RNA was extracted from fed and fasted rat muscle and was added to increasing quantities of the rat LPL cRNA construct containing primer sites for LPL. The ratio of LPL cRNA/mRNA was plotted against the number of copies of cRNA added, to yield the equivalence point between cRNA and LPL mRNA. A: Representative RT-PCR data showing the reaction with RNA from the muscles of fed and fasted animals. B: Summary of all the RT-PCR data on soleus and EDL. The data are expressed as the number of copies of LPL mRNA/ng total RNA. Bar graph: each bar represents the data (mean ± SEM) from 4–5 animals.

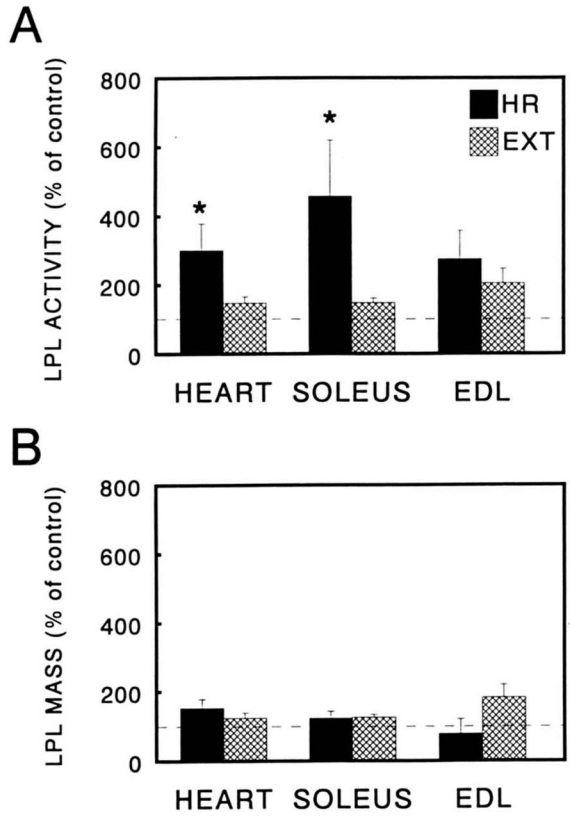


Fig. 7. Effect of hypothyroidism on muscle LPL activity and LPL mass in rats. LPL catalytic activity (A) and LPL immunoreactive protein mass (B) were measured in heart, soleus, and EDL from thyroidectomized rats, and compared to that of control animals. Data are expressed as the percentage (mean ± SE) of LPL in control rats. Dashed line represents the mean value of control animals normalized to 100%; n = 11–23; $^*P < 0.05$ vs. control rats.

which it is expressed. In adipose tissue, FFA are generated and then used for storage in the fat cell lipid droplet, while in muscle, LPL generates FFA that are used predominantly for energy. The regulation of LPL has been examined in a number of different cells and tissues. In response to different hormonal and metabolic influences, regulation has been demonstrated due to changes in LPL mRNA levels (26–31), translation (16, 32–35), and posttranslational processing (32, 33, 36–38). Therefore, the present studies were performed to examine the expression of LPL in different muscle types and in response to thyroid hormone.

To examine LPL expression in muscles of different fiber types, we examined soleus, EDL, and heart. Soleus and EDL were chosen because they are very different in fiber type. Soleus contains approximately 84% type I fibers, whereas EDL contains 97% type II fibers. Type I fibers are also referred to as red fibers or slow-twitch oxidative fibers that are rich in mitochondria and especially adapted for FFA oxidation and endurance functions (5). Type II fibers are referred to as fast-twitch or glycolytic fibers and are adapted for rapid movements. It is logical that muscle containing a high proportion of type I fibers would express a high level of LPL, which provides the tissue with FFA. When LPL catalytic activity was measured, soleus and heart tissue were about equal, but both were tenfold higher than the LPL catalytic activity of EDL. The lower amount of LPL activity in EDL was reflected in both its LPL immunoreactive mass as well as its LPL mRNA level, as demonstrated by both Northern blotting and competitive RT-PCR. Similar observations were made by Ladu, Kapsas, and Palmer (11), who examined LPL activity and mRNA levels in rat heart, soleus, and red and white vastus lateralis. Therefore, there are large differences in LPL expression between skeletal muscles composed of different fiber types, and this difference likely results from differences in LPL transcription or mRNA stabilization.

In response to feeding, heart LPL activity and mass in

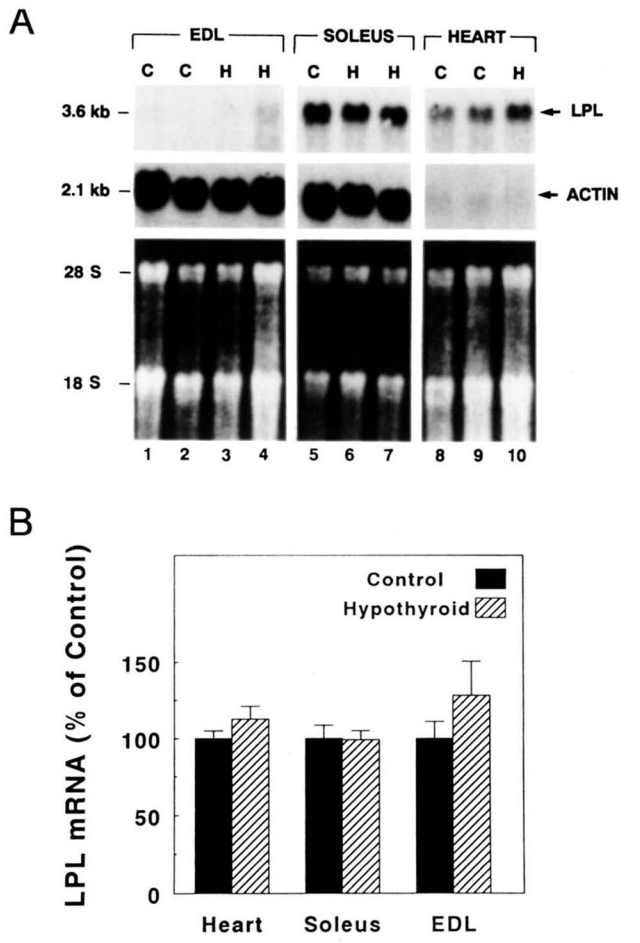


Fig. 8. Effect of hypothyroidism on rat muscle LPL mRNA levels. A: Total RNA was extracted from EDL, soleus, and heart tissue of control (C) and hypothyroid rats (H) and subjected to Northern blot analysis with ^{32}P-labeled cDNA probes for LPL and γ-actin. The bottom panel shows the 28S and 18S ribosomal RNA bands of the ethidium bromide-stained gel. B: LPL/total RNA ratios from all Northern blots; n = 6–8 for each muscle type.

the HR fraction decreased to about 50% of the values in fasted rats, and there was no change in the EXT fraction (non-heparin-released LPL). Northern blotting RNA revealed parallel changes in heart LPL mRNA levels. These data are consistent with the data of Ladu et al. (11), who observed parallel increases in heart LPL activity and mRNA with fasting, but different from the data of Doolittle et al. (38), who observed no change in heart LPL mRNA levels.

In contrast to the changes in heart, we found that LPL in soleus and EDL were generally unresponsive to feeding/fasting, and there was no significant increase in LPL activity, mass, or mRNA. In another study (11), there were increases in skeletal muscle LPL mRNA (but no change in activity) with 24-h fasting, and increased activity and mRNA with 6 days of fasting. Numerous previous studies have examined LPL activity in skeletal muscle in response to starvation. As reviewed by Borensztajn (2), most studies have observed increases in LPL activity with starvation, but several well-documented studies have failed to observe such changes, suggesting that the length of the starvation, and perhaps other factors, are important. These data suggest that heart LPL is more sensitive to feeding/fasting than is skeletal muscle.

Thyroid hormone affects many aspects of muscle metabolism, and previous studies have examined LPL activity during hyper- or hypothyroidism. Hyperthyroidism has been shown to increase heart LPL (15, 39–41); however, hypothyroidism has been shown to increase (12, 13), or not affect (14, 15) heart LPL. Fewer studies have examined the effects of thyroid hormone on skeletal muscle LPL, and again the data are somewhat conflicting. In hypothyroid rats, LPL activity in soleus and vastus lateralis was increased (15), whereas hyperthyroidism either increased (40) or decreased (15) skeletal muscle LPL. The data regarding thyroid hormone and adipose tissue LPL are more consistent; hypothyroidism results in an increase in LPL activity in rats (reviewed in ref. 1). In humans, however, thyroid hormone has been reported to regulate LPL in a manner opposite to that in rats in both muscle and adipose tissue LPL (1, 2, 39, 42).

In a previous study (16), we examined adipose tissue LPL in hypothyroid rats and found that LPL was increased in hypothyroidism due to an increase in LPL translation: an increase in LPL activity, immunoreactive mass, and LPL synthetic rate, but no change in LPL mRNA level. In this study, we found that muscle LPL activity was also increased in response to hypothyroidism, however this occurred through a different mechanism. Thyroidectomized rats had significantly higher levels of LPL activity in the HR fractions of soleus and heart, although there was no difference in the EXT fractions, when compared to control animals, and no change in LPL mass or mRNA levels. LPL activity of EDL did not change significantly due to the hypothyroid state. Because there was an increase in HR LPL activity but not in LPL mass in soleus and heart, the subsequent increases in LPL specific activity suggest that hypothyroidism exerts posttranslational regulation of LPL activity.

The exact mechanism for this posttranslational increase of LPL specific activity remains to be elucidated. Among many possible mechanisms, hypothyroidism may alter the ability of heparin to release LPL in muscle. This could be accomplished by increasing the number or affinity of glycosaminoglycan binding sites. In contrast to the distribution of LPL in adipose tissue, HR is only a small component of muscle LPL: only $< 30\%$ of LPL activity, and $< 5\%$ of LPL mass. These data suggest that only a small fraction of muscle LPL is active enzyme that is bound to capillary glycosaminoglycans. The majority of LPL is retained within muscle tissue where it remains largely catalytically inactive. A number of studies have recently characterized LPL posttranslational processing, and have suggested that the removal of terminal glucose residues from N-linked oligosaccharides is a critical step in the development of LPL activity and for LPL secretion (43–45). In addition, a recent study suggests that muscle LPL posttranslational processing is similar to adipose tissue (46). Therefore, these data on hypothyroidism suggest that this process of intracellular transport and posttranslational oligosaccharide processing is subject to regulation in muscle, as it is in adipose tissue.

When studying hypothyroidism in rats, it is difficult to control for nutritional state. Hypothyroid rats do not gain weight as rapidly as normal rats, and therefore presumably eat less. One possible explanation for the increase in LPL could be chronic decreased food intake. Although both fasting and hypothyroidism resulted in an increase in LPL in heart, fasting had no effect on soleus. Therefore, it is unlikely that underfeeding would explain the effects of hypothyroidism, although it is possible that some of the effects of hypothyroidism are due to nutritional influences.

Numerous other studies have demonstrated an inverse relationship between muscle and adipose tissue LPL in response to nutritional and hormonal influences (47). However, this study, coupled with previous studies (1, 16), demonstrated that hypothyroidism increased LPL in both adipose tissue and muscle. Therefore, inverse regulation of LPL in adipose tissue and muscle is not always present.

In summary, this study examined the mechanism of regulation of LPL in muscles composed of different fiber types in response to feeding/fasting, and in muscles of control and hypothyroid animals. Muscle LPL was increased in muscles containing predominantly type I fibers due to an increase in LPL mRNA, whereas the increase in LPL activity in heart in response to fasting, and in hypothyroid rat muscles was due to posttranslational changes. Together, these data suggest that the regulation of muscle LPL occurs through different mechanisms, and is likely as complex as the regulation of LPL in adipose tissue.

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