Muscle Protein Metabolism during Compensatory Growth with Changing Dietary Lysine Levels from Deficient to Sufficient in Growing Rats*

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Summary Livestock and laboratory animals show compensatory growth when they are fed ad libitum following a period of restriction feeding. Lysine is a major limiting essential amino acid in the diets both for humans and animals. We hypothesized that changing dietary lysine levels from deficient to sufficient induced compensatory growth in young rats. We elucidated the effect of lysine sufficiency on the dynamics of hormones, relevant to muscle protein synthesis and degradation, insulin-like growth factor-I (IGF-I) and corticosterone, and on the expression of proteolytic-related genes in skeletal muscle during compensatory growth. Lysine sufficiency where the dietary lysine level was increased from 0.46% to 1.30% after 2 wk of subjecting the rats to the lower lysine level induced 80% enhancement of growth rate of rats. During compensatory growth with the lysine sufficiency, fractional muscle protein synthesis rates were higher whereas fractional muscle protein degradation rates were lower than those of the control group (p<0.05). After lysine sufficiency, the expression of atrogin-1/MAFbx mRNA was decreased in gastrocnemius muscle (p<0.05). With the lysine sufficiency, serum IGF-I concentration increased (p<0.05) whereas serum corticosterone decreased (p<0.05). These findings suggest that compensatory growth with lysine sufficiency is due to a change of hormone levels before and after changing diets, resulting in incrementation of protein synthesis and suppression of protein degradation of skeletal muscle.

Key Words compensatory growth, lysine, rat, IGF-I, corticosterone

One of the important subjects in nutritional science is to elucidate how humans and animals respond and adapt to transitions of nutritional status and changes of metabolic reaction that occur during the adaptation.

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Abbreviations: AIN-93, American Institute of Nutrition-93; C, rats that consumed the control diet; GH, growth hormone; IGF-I, insulin-like growth factor-I; IGFBP-3, insulin-like growth factor binding protein-3; LC, rats that consumed the low lysine diet for the first 2 wk, and after that consumed the control diet; LL, low lysine.

There are numerous reports about adaptation of animals to excess or deficiency of nutrients or energy in diets. Compensatory growth is an example of adaptation to nutritional status. Livestock and laboratory animals show compensatory growth when they are fed ad libitum following a period of restricted feeding or fed adequate energy following a period of energy restriction (1–3). Previous studies in rats and pigs showed that when animals that had been fed a protein-free or a low protein diet were refed a diet containing a sufficient amount of protein, they developed faster than animals fed a control diet particularly for the first few days after the diet replacement (4–6). Promoted protein accumulation during compensatory growth is due to a larger difference between the rate of muscle protein synthesis and the rate of muscle protein degradation compared to normal growth. The phenomenon has been considered a transitional state in adapting to the nutritional status. Both anabolic and catabolic hormones may be involved in this response. All of these previous studies indicate that sufficiency of dietary protein and amino acid levels plays a role as a trigger for compensatory growth in animals. However, these studies provide no information...
about the role of a specific amino acid because levels of multiple amino acids were different in the diets fed to the animals (7, 8). To our knowledge, the effect of a single amino acid in an iso-energetic and iso-protein diet has not yet been elucidated.

Lysine is a major limiting essential amino acid in the diets both for humans and animals. From an estimation of plasma amino acid network structure, lysine is located at the top control level and has effects on almost all of the other plasma amino acids while no other amino acids can affect the concentration of lysine (9). Similar to animals with protein or multiple amino acid deficiencies, rats and pigs fed low lysine diets grew more slowly and less efficiently than those fed control diets (10, 11). These responses of animals to the shortage of a single amino acid allow us to hypothesize that changing dietary lysine levels from deficient to sufficient induces compensatory growth in young animals.

We elucidated the effect of lysine sufficiency on the dynamics of hormones relevant to muscle protein synthesis and degradation, IGF-I and corticosterone, and on expression of proteolytic-related genes of skeletal muscle during compensatory growth in young growing rats.

In the present study, we found distinct compensatory growth with changing dietary lysine levels from deficient to sufficient, particularly for the first 3 d after the sufficiency.

**MATERIALS AND METHODS**

**Diets.** We formulated two diets for this study: a control diet and a low lysine (LL) diet. The composition of the diets is shown in Table 1. Diets were based on the AIN-93 recommendations for rodents. For the control diet, additional lysine, HCl-L-lysine 10.8 g/kg, was then added to give a level of 13.0 g/kg diet to meet the requirement in the AIN-93. This added lysine was replaced by added glycine and L-glutamic acid, which are non-essential amino acids, in the LL diet because these amino acid do not antagonize lysine. Therefore, the control diet contained 1.30%, whereas the low lysine diet contained only 0.46% of total lysine. The two diets were approximately isoenergetic and iso-protein.

**Animals and experimental design.** Sixty Wistar rats (Japan SLC, Inc.) aged 4 wk were randomly allocated into two groups: a control group (this group is abbreviated as C) and an experimental group (this group is abbreviated as LC). The animals were individually housed in metabolic cages in an air-conditioned room at 25°C. The average initial body weight of the rats was 78.5 ± 2.8 g. The control rats were continuously fed the control diet throughout the 21-d experimental period. The LC rats were fed the low lysine diet for the first 2 wk; thereafter they were fed the control diet until the end of experiment. Thus the LC rats experienced a shift of nutritional status for lysine from deficiency to sufficiency. The day when the diet given to the LC rats was replaced was defined as 0 d. All rats had free access to food and water. From day -3 to 7, daily urine samples were collected in flasks containing 1 mL 1 M HCl, and stored at −20°C until analysis of 3-methylhistidine concentrations. On the day 0, 1, 3 and 7, six animals from each group were anesthetized with sodium pentobarbital (Dainippon Sumitomo Pharma). Following laparotomy, whole blood was collected from the abdominal aorta and placed into a serum separator tube (Terumo). Serum was obtained by centrifugation at 1,200 × g for 15 min at 4°C. These specimens were stored at −80°C until analysis of concentrations of hormones and amino acids. The gastrocnemius muscle samples were removed immediately, weighed and frozen in liquid nitrogen, prior to storage at −80°C. All the experimental procedures were carried out in accordance with the guidelines of the Animal Care and Use Committee of the National Institute of Livestock and Grassland Science.

| Ingredients (%) | Control | Low lysine |
|----------------|---------|------------|
| Wheat gluten   | 26.00   | 26.00      |
| Skim milk      | 4.10    | 4.10       |
| Sucrose        | 20.00   | 20.00      |
| Cornstarch     | 36.54   | 36.54      |
| Soya bean oil  | 6.80    | 6.80       |
| AIN-93G vitamin mix | 1.00   | 1.00      |
| AIN-93 mineral mix | 3.50   | 3.50      |
| HCl-L-lysine   | 1.08    | 0          |
| DL-Methionine  | 0.11    | 0.11       |
| L-Threonine    | 0.10    | 0.10       |
| L-Tryptophan   | 0.01    | 0.01       |
| L-Valine       | 0.13    | 0.13       |
| L-Glutamic acid| 0       | 0.54       |
| Glycine        | 0       | 0.54       |
| Tyrosine       | 0.16    | 0.16       |
| Aspartic acid  | 0.42    | 0.42       |
| L-Isoleucine   | 0.05    | 0.05       |
| Calculated chemical composition |         |            |
| Crude protein (%) | 21.90  | 21.80      |
| Digestible energy (Mcal/kg) | 4.00  | 3.98       |
| Lysine (%)     | 1.30    | 0.46       |

1Containing the following (g/kg vitamin mix): retinyl palmitate (500,000 U/g), 0.80; cholecalciferol (400,000 U/g), 0.25; phytomenadione, 0.075; biotin, 0.02; cyanocobalamin (0.1%), 2.5; folic acid, 0.20; nicotinic acid, 3.0; calcium pantothenate, 1.6; pyridoxine-HCl, 0.70; riboflavin, 0.60; thiamin-HCl, 0.60; all-rac-o-tocopheryl acetate (50%), 15; choline bitartrate, 250; sucrose, 724.655.

2Containing the following (g/kg mineral mix): calcium carbonate, 357; monopotassium phosphate, 196; potassium citrate, 70.78; sodium chloride, 74; potassium sulfate, 46.6; magnesium oxide, 24; ferric citrate, 6.06; zinc carbonate, 6.15; manganese carbonate, 0.63; cupric carbonate, 0.324; potassium iodate, 0.010; sodium selenite, 0.0103; ammonium molybdate-4H2O, 0.080; sodium metasilicate·9H2O, 1.45; chromium KSO4·12H2O, 0.275; lithium chloride, 0.017; boric acid, 0.082; sodium fluoride, 0.064; nickel carbonate-4H2O, 0.031; ammonium metavanadate, 0.0066; sucrose, 221.
Calculation of fractional protein degradation and synthesis rates of skeletal muscle. Urine 3-methylhistidine concentrations were measured by a HPLC method after derivatization of fluorescamine with a treatment of perchloric acid and heating (12). The concentration of 3-methylhistidine in skeletal muscle was estimated to be 4.185 mg per 100 g body weight as described by Nishizawa et al. (12, 13). The fractional protein degradation rates (%/d) of skeletal muscle were calculated by dividing the amount of 3-methylhistidine excreted in the urine daily by the amount of 3-methylhistidine bound in the skeletal muscle pool. Fractional growth rates (%/d) of skeletal muscle were calculated as the rates of skeletal muscle protein gain divided by the average body weight measured before and after the urinary sample collection. The fractional protein synthesis rates (%/d) of the mixed muscle protein pool were calculated as the sum of fractional degradation and growth rates. The fractional degradation and synthesis rates on 0 d (Fig. 2) were calculated as the average of those values on each day from day –3 to 0 in order to compare those of rats fed the LC and the control diets. The fractional degradation and synthesis rates on day 7 (Fig. 2) were calculated as the average of those values on each day from day 4 to 7.

Measurement of serum hormones and lysine. Serum concentrations of insulin-like growth factor-I (IGF-I), insulin-like growth factor binding protein-3 (IGFBP-3), corticosterone and insulin were determined by commercial kits: Rat IGF-I ELISA (10-29200, DSL), Mouse/Rat IGFBP-3 EIA Kit (E-031, Mediagnost), rat corticosterone EIA kit (10-81100, DSL) and Rat Insulin ELISA KIT (AKRIN-030, Shibayagi), respectively. In order to determine serum amino acid concentrations, samples were deproteinized with one volume of 6% 5-sulfosalicylic acid. Supernatant fluids were filtered through a 0.45 μm syringe filter unit (Advantec Toyo) to remove the traces of protein. Free amino acids in the supernatants from serum samples were measured using an automated amino acid analyzer (L-8500, Hitachi).

RNA isolation and RT-PCR. Total RNA was extracted from the samples of gastrocnemius muscle by the guanidine thiocyanate/phenol-chloroform extraction method with TRIzol reagent (15596-018, Invitrogen) and the primers purchased from Qiagen (QuantiTect Primer Assays) for atrogin-1/MAFbx (QT00194698), MuRF1 (QT0182634), m-calpain large subunit (QT00182819), μ-calpain large subunit (QT00175259), proteasome C2 subunit (QT00178003), cathepsin L (QT00180054), caspase-3 (QT00186333) and glyceraldehyde-3-phosphate dehydrogenase (GAPDH, QT00423122). The quantification of mRNA was performed using a LightCycler ST300 (Roche Diagnostics) starting with reverse-transcribed total RNA. GAPDH expression was used as the internal control.

Statistical analysis. All results are expressed as least square means±SE. Data on body weight, feed intake, fractional synthesis rates and fractional degradation rates were analyzed using the mixed procedure for repeated measurements in the SAS package (SAS version 9.2, SAS Institute) with the treatment and time as fixed effects. We used two-way analysis of variance for serum hormones, amino acids and mRNA expression with the GLM procedure in the SAS statistical package (SAS version 9.2). When the effect of the treatments and/or the times were significant, statistical significances between control values at a time point: **p<0.01, *p<0.05.

RESULTS

Growth rates, feed intake, feed efficiency and gastrocnemius muscle weight

Despite equal total energy and protein intake, body weight (p<0.05, Fig. 1A) and daily growth weight gain during the first 2 wk (p<0.01, Fig. 1B) was lower in rats fed the low lysine diet compared with those of the rats...
fed the control diet. Feed intake was not different between the LC and the control groups during the first 2 wk, whereas after changing the diet, feed intake of the LC group was lower than that of the C group \((p<0.01\), Table 2). On day 1, 1 d after changing the diets, daily body weight gain of the LC group was already higher than that of the C group \((Fig. 1B, p<0.05)\). A higher daily body weight gain was also observed on day 3, 5 and 7 \((Fig. 1B, p<0.05)\). Due to the change of dietary lysine levels from day 0, the growth rate of the LC group during the 3rd week was 80% higher than that of the 2nd week \((p<0.05, Table 2)\). Feed efficiency of the LC group was lower than that of the C group \((p<0.05, Table 2)\) during the first 2 wk. Despite the reduction of feed intake, the LC rats showed compensatory growth with the lysine sufficiency. Therefore, after changing the diet, the feed efficiency of the LC group was higher than that of the C group \((p<0.05, Table 2)\). Due to the change of dietary lysine levels from day 0, the feed efficiency of the LC group during the 3rd week was 70% higher than that of the 2nd week \((p<0.05, Table 2)\).

### Table 2. Effect of changing lysine levels from deficient to sufficient on feed intake, growth performance and gastrocnemius muscle weight of rats.

| Term | 1 wk | 2 wk | 3 wk | RMSE |
|------|------|------|------|------|
| **Term** | **1 wk** | **2 wk** | **3 wk** | **RMSE** |
| **Treat** | **C** | **LC** | **C** | **LC** | **C** | **LC** | **C** | **LC** |
| **Feed** | **C** | **LL** | **C** | **LL** | **C** | **C** | **C** | **C** |
| Body weight gain, g/d | 6.16b | 3.96d | 5.84b | 3.71d | 5.69bc | 6.67a | 0.44 |
| Feed intake, g/d | 12.0d | 12.5c | 13.9b | 13.6b | 14.8a | 12.9c | 0.75 |
| Feed efficiency rates | 0.51a | 0.32d | 0.46b | 0.29e | 0.38c | 0.52a | 0.02 |
| Muscle weight, g | — | — | 0.88c | 0.69d | 1.09a | 0.98b | 0.04 |

Data are means and RMSE, \(n=6\) per treatment group. Means without a common letter differ \((p<0.05)\).

1 Feed efficiency rates=body weight gain (g/d)/feed intake (g/d).
2 Muscle weight=gastrocnemius muscle weight (g).

### Fractional protein degradation and synthesis rates of skeletal muscle

The fractional protein degradation rates of skeletal muscle of the LC group fed the LL diet were not different from those of the C group. Therefore, after changing to the control diet, the fractional protein degradation rates of the LC group decreased and they were lower than that of the C group \((p<0.01, \text{Fig. 2A})\). From day 4 to 7, there was no difference between fractional protein degradation rates of skeletal muscle of the LC group and those of the C group. The fractional protein synthesis rates of skeletal muscle of the LC group rats fed the LL diet were lower than those of the C groups \((p<0.01, \text{Fig. 2B})\), while after changing to the control diet, the fractional protein synthesis rates of skeletal muscle of the LC group rats were higher than those of the C group on day 1, from 3 to 7 \((p<0.05)\).

### Expression of proteolytic-related genes

The expression of proteolytic related genes in gastrocnemius muscle of the LC rats fed LL diets was not different from that of the control group on day 0. On day 1 and 3, after changing the diet, the expression of atrogin-1/MAFbx, an E3 ligase of the ubiquitin-proteasome pathway in the muscle, of the LC rats given the control diet was lower than that of the control rats \((day 1, p<0.01; day 3, p<0.05, \text{Fig. 3A})\). Thereafter, the
Fig. 3. Effect of changing dietary lysine levels from deficient to sufficient on the mRNA expression of atrogin-1/MAFbx (A), MuRF1 (B) and caspase-3 (C) in gastrocnemius muscle of rats. Values are means ± SE, n = 6. Within LC group, means without a common letter differ (p < 0.05). Asterisk indicates significant difference versus control at a time point (** p < 0.01, * p < 0.05).

Fig. 4. Effect of changing dietary lysine levels from deficient to sufficient on serum IGF-I (A), IGFBP-3 (B), corticosterone (C) and lysine (D) in rats. Values are means ± SE, n = 6. Within LC group, means without a common letter differ (p < 0.05). Asterisk indicates significant difference versus control at a time point (** p < 0.01, * p < 0.05).
expression returned to the control level on day 7 (data not shown). The expression of MuRF1, an E3 ligase of the ubiquitin-proteasome pathway in the gastrocnemius muscle was not different between the LC group and the control group at any points of the sample collection (Fig. 3B). The expression of caspase-3, an initial and potentially rate-limiting protease cleaving actomyosin/myofibrils to produce substrates degraded by the ubiquitin-proteasome pathway, of gastrocnemius muscle was lower in the LC rats than in the control rats on day 1 ($p<0.05$, Fig. 3C). No difference was observed on day 3. The mRNA expression of other proteolytic-related genes (m-calpain large subunit, $\mu$-calpain large subunit, proteasome C2 subunit and cathepsin L) of gastrocnemius muscle was not different between the LC group and the control group at any points of the sample collection (data not shown).

**Serum IGF-I, IGFBP-3, corticosterone, insulin and serum lysine**

Concentrations of serum IGF-I and IGFBP-3 in rats fed the LL diet were lower than those of the control rats ($p<0.01$, Fig. 4A and B). After the replacement of the diets, concentrations of serum IGF-I and IGFBP-3 already increased on day 1, and hence, there were no differences in concentrations of serum IGF-I and IGFBP-3 between the LC group and the C group on day 1 or day 3. The concentrations of serum corticosterone of the LC group rats fed the LL diet were tended to be higher than those of the rats given the control diet on day 0 ($p=0.06$). Concentrations of serum corticosterone decreased in the LC group rats due to the sufficiency of lysine, and hence, there were no differences in them between the two groups on day 3. The concentrations of corticosterone of the LC group on day 3 were lower than that on day 0 ($p<0.05$, Fig. 4C). The concentrations of serum free lysine of rats fed the LL diet were greatly lower than those of the rats given the control diet ($p<0.01$, Fig. 4D). On day 1, 1 d after changing the diets, the concentrations of serum free lysine of the LC rats fed the control diet were higher than that of the rats given the control diet ($p<0.01$, Fig. 4D). On day 3, there was no difference between the two groups. No effects due to dietary lysine levels or lysine sufficiency were observed in concentrations of serum insulin. The average concentration of serum insulin of all the rats was $5.97 \pm 0.48$ ng/mL.

**DISCUSSION**

Compensatory growth is an ability of animals to exhibit, after feed restriction, greater growth rates than in unaffected animals of the same chronological age (14). The present study demonstrates for the first time that changing dietary levels of a single essential amino acid from deficient to sufficient, in a diet containing adequate amounts of other nutrients and energy, induces compensatory growth in growing rats. In previous reports, although laboratory animals and livestock showed compensatory growth with sufficient intake of protein or amino acids, the tested diets used in those experiments were deficient in multiple amino acids, but not in a single amino acid (7, 8). Moreover, in these experiments, the terms of compensatory phase was long as 1 mo and the magnitudes of deficiency were severe. Hence, we investigated muscle protein metabolism with compensatory growth induced immediately after sufficiency of a single dietary amino acid, lysine. We focused on the protein metabolism of skeletal muscle because we found the magnitude of increase in gastrocnemius muscle weight of rats exhibiting compensatory growth was greater than that of the rats showing normal growth. This alteration of muscle protein metabolism induced by lysine sufficiency may be attributed to a change in the circulation levels of hormones relevant to muscle protein metabolism.

The suppression of proteolysis contributed to the growth increment during the first 3 d after the lysine sufficiency, whereas the increment of protein synthesis of skeletal muscle continuously contributed to it from day 1 to 7. Our results are in agreement with a previous study of ad libitum feeding after restricted feeding (15). These results from the present and the previous study suggest that suppression of proteolysis and increment of protein synthesis both contribute to the promoted growth rate, although there are differences in the timing of contribution.

We focused on skeletal muscle proteolysis during compensatory growth with lysine sufficiency following its deficiency. In a previous study, lysine directly inhibited activity of proteasome prepared from muscle of rats in vitro (16). Further, we have reported that oral administration of lysine suppressed muscle proteolysis in skeletal muscle of chicks (17). Therefore, a higher level of serum lysine might suppress proteolysis shortly after lysine sufficiency in the LC rats. To determine the molecular basis of the suppression of proteolysis after the lysine sufficiency, mRNA expression of proteolytic-related genes was examined. In the present study, we observed that the lysine sufficiency after its deficiency decreased the proteolysis and the expression of atrogin-1/MAFbx. Atrogin-1/MAFbx is a muscle specific ligase related to the ubiquitin-proteasome pathway, which is a major proteolytic pathway in the muscle cell. Thus, we infer that suppression of proteolysis with lysine sufficiency after lysine deficiency may result from suppressed components of the ubiquitin-proteasome pathway.

We next measured concentrations of serum IGF-I, IGFBP-3 and corticosterone, which play pivotal roles in regulating protein metabolism. IGF-I is a major regulator of the anabolic drive of nutrients and when IGF-I binds to its priming binding protein, IGFBP-3, its half-life in blood circulation is extended (18). Rats fed the low lysine diet had lower concentrations of serum IGF-I and IGFBP-3. Similar results were already obtained in rats (19) and pigs (10). Our novel finding is that after the lysine sufficiency, serum concentrations of IGF-I and IGFBP-3 of the LC rats increased as quickly as 1 d after changing the diet. These results suggest that IGF-I bioavailability in the blood circulation was drastically increased after changing the diet in the LC rats. It was
reported that orally administrated lysine induced a release of growth hormone (GH) into the blood circulation (20). As GH induces the secretion of IGF-I from liver, stimulated release of GH to blood circulation may explain the increment of IGF-I level. However further investigations are needed in this respect. In contrast to IGF-I, the concentration of serum corticosterone levels were elevated in rats fed the low lysine diet, which is in agreement with a previous report (19). Although food restriction and caloric restriction increase concentration of blood corticosterone with activation of hypothalamic-pituitary-adrenal axis and increasing adrenocorticotropic hormone, there is no information on the eventual interactions between lysine deficiency and concentration of serum corticosterone.

Glucocorticoid promotes protein breakdown by enhancing the expression of atrogin-1/MAFbx in the ubiquitin-proteasome pathway (21). However, despite of high concentration of corticosterone of the LC rats fed the LL diet, its fractional protein degradation rates were not higher, which is in agreement with previous reports on prolonged experiments of low protein diet (22) and corticosterone treatment (23). Such cancelled up-regulation of protein degradation may be explained by glucocorticoid resistance (24). IGF-I is a major anabolic factor that regulates protein turnover in skeletal muscle. IGF-I suppressed protein degradation and prevented the dexamethasone-induced increase in proteolysis. Changes in overall proteolysis with dexamethasone, one of glucocorticoid, and IGF-I correlated tightly with changes in atrogin-1/MAFbx mRNA content (21). In the present study the rats exhibiting compensatory growth after the lysine deficiency showed a selective decrease in the mRNA expression of atrogin-1/MAFbx, but not MuRF1. In response to various atrophic stimuli, such as food-restriction, immobilization, diabetes and denervation, the up-regulation of mRNA expression of atrogin-1/MAFbx and MuRF1 was found to be skeletal muscle specific in rats (25). Previous studies showed that up-regulation or down-regulation of atrogin-1/MAFbx mRNA expression was more drastic than that of MuRF1 (21, 26). The magnitude of the increment of atrogin-1/MAFbx mRNA expression by dexamethasone was larger than that of MuRF1 (21, 27). Moreover administration of IGF-I and IGFBP-3 decreased the mRNA of atrogin-1/MAFbx but not MuRF1 (25). The degree of the hormonal change due to the lysine deficiency in the present study might not be large enough to down-regulate MuRF1 mRNA expression. Thus, lysine deficiency after its deficiency suppresses serum corticosterone and increases serum IGF-I and this hormonal transition may suppress expression of atrogin-1/MAFbx and proteolysis of skeletal muscle. In the present study, we also observed that lysine deficiency after its deficiency decreased the expression of caspase-3 in gastrocnemius muscle. Activation of caspase-3, an apoptotic protease, was proposed to trigger the initial step of muscle proteolysis (28). IGF-I may be involved in reduction of caspase-3 mRNA, because IGF-I also inhibited caspase-3 activation in cultured muscle cells (28) as well as proteolysis in skeletal muscle. However, there is no information about the effect of a single amino acid on caspase-3 mRNA expression. The role of caspase-3 in protein metabolism of rats exhibiting compensatory growth due to lysine sufficiency still remains to be elucidated.

In rats, the skeletal muscle ratio to body weight was high and protein accumulation in skeletal muscle was rapid in the growing phase (29). Furthermore, in this experiment, gastrocnemius weight per body weight of the LC group was higher than that of the control rats on day 7. Therefore, incrementation of skeletal muscle may largely contribute to compensatory growth. Fat accumulation or growth of internal organs may be involved in the compensatory growth. However, since there are no data about fat and internal organs, further investigation is needed in this respect.

In conclusion, the present study demonstrates that dietary lysine sufficiency after its deficiency induces compensatory growth. Increase in protein synthesis and reduction in protein degradation may be involved in the underlying mechanisms of this compensatory growth. Our data suggest that the compensatory growth with lysine sufficient is due to changing levels of hormones relevant to protein synthesis and degradation and to increment of serum lysine before and after changing diets.

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