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Resistance exercise acutely increases MHC and mixed muscle protein synthesis rates in 78–84 and 23–32 yr olds

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Hasten, Debbie L., Jina Pak-Loduca, Kathleen A. Obert, and Kevin E. Yarasheski. Resistance exercise acutely increases MHC and mixed muscle protein synthesis rates in 78–84 and 23–32 yr olds. *Am J Physiol Endocrinol Metab* 278: E620–E626, 2000.—We determined whether short-term weight-lifting exercise increases the synthesis rate of the major contractile proteins, myosin heavy chain (MHC), actin, and mixed muscle proteins in nonfrail elders and younger women and men. Fractional synthesis rates of mixed, MHC, and actin proteins were determined in seven healthy sedentary 23- to 32-yr-old and seven healthy 78- to 84-yr-old participants in paired studies done before and at the end of a 2-wk weight-lifting program. The in vivo rate of incorporation of 1-[^{13}C]leucine into vastus lateralis MHC, actin, and mixed proteins was determined using a 14-h constant intravenous infusion of 1-[^{13}C]leucine. Before exercise, the mixed and MHC fractional synthetic rates were lower in the older than in the younger participants ($P \leq 0.04$). Baseline actin protein synthesis rates were similar in the two groups (P = not significant). Over a 2-wk period, participants completed ten 1- to 1.5-h weight-lifting exercise sessions: 2–3 sets per day of 9 exercises, 8–12 repetitions per set, at 60–90% of maximum voluntary muscle strength. At the end of exercise, MHC and mixed protein synthetic rates increased in the younger (88 and 121%) and older participants (105 and 182%; $P < 0.001$ vs. baseline). These findings indicate that MHC and mixed protein synthesis rates are reduced more than actin in advanced age. Similar to that of 23–32 yr olds, the vastus lateralis muscle in 78–84 yr olds retains the capacity to increase MHC and mixed protein synthesis rates in response to short-term resistance exercise.

sarcopenia; aging; muscle amino acid metabolism; weight lifting; mass spectrometry; myosin heavy chain

SKELETAL MUSCLE PROTEIN MASS and voluntary muscle strength deteriorate with advancing age. This contributes to the development of physical frailty, reduced physical function, impaired mobility, and loss of independence in the very old (9, 10, 15, 22, 23). Among several factors (e.g., chronic illness, undernutrition) that contribute to muscle wasting and weakness in old age, the lack of skeletal muscle contractile activity (sedentary lifestyle) exerts a major influence on sarcopenia that is potentially preventable with an appropriate exercise intervention (9, 10).

Two weeks of daily weight-lifting exercise training increased the fractional and absolute rates of vastus lateralis mixed muscle protein synthesis (~150%) in healthy sedentary 63- to 66-yr-old men and women (25). Initially, the rate of vastus lateralis mixed muscle protein synthesis was 38% lower in 63–66 yr olds than in 23–25 yr olds. After 2 wk of resistance exercise done at a similar relative intensity, the rate of mixed muscle protein synthesis increased to the same rate in both 23–25 and 63–66 yr olds. The reduced rate of mixed and myofibrillar protein synthesis in 60^{+}-yr-old adults and the stimulatory effect of resistance exercise on the rate of amino acid transport into muscle and the rate of muscle protein synthesis in younger adults have been reported by others (4, 7, 8, 22). However, it is not known whether weight-lifting exercise can acutely increase mixed and myofibrillar protein synthesis rates in 78^{+}-yr-old as well as younger women and men.

Several investigators have determined the in vivo fractional synthetic rate of mixed muscle proteins. This measurement reflects the average synthesis rate of several muscle proteins. It is desirable to determine the synthetic rate of specific contractile [myosin heavy chain (MHC), actin], mitochondrial, and sarcoplasmic proteins isolated from small muscle samples and to evaluate the effects of advancing age and exercise training on the synthetic rate of specific muscle proteins (2–4, 11, 19). Balagopal et al. (4) reported that the synthesis rate of MHC, but not sarcoplasmic, proteins decreased with advancing age. Rooyackers et al. (19) reported that the rate of subsarcolemmal mitochondrial protein synthesis was lower in 54- ± 1-yr-old men and women (middle age) than in 24- ± 1-yr-old men and women, but no further decrease was observed in 73- ± 2-yr-old men and women. These studies suggest that the synthetic rates of individual muscle proteins are regulated differently with age and demonstrate the importance of isolating specific muscle proteins when determining the effects of advancing age and various interventions on muscle protein metabolism.

The effect of advancing age on mixed, MHC, mitochondrial, and sarcoplasmic protein synthesis rates has been reported (4, 15, 19, 22, 25). The acute effect of weight-lifting exercise on the synthesis rates of MHC and actin in younger and very old men and women has

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not been determined. The purpose of this study was to determine the fractional synthesis rates of mixed, MHC, and actin proteins in 23–32 and 78–84 yr olds. We also determined whether the muscle protein synthetic rates increase in response to short-term weight-lifting exercise similarly in the younger and elderly adults.

METHODS

Subjects

Seven healthy 23- to 32-yr-old men ($n = 4$) and women ($n = 3$) and seven healthy 78- to 84-yr-old men ($n = 3$) and women ($n = 4$) participated in this study. The procedures were approved by the Human Studies Review Board at Washington University School of Medicine. Informed consent was obtained after the purpose and procedures were described.

Before enrollment, the 78- to 84-yr-old participants were screened for cardiovascular, metabolic, and neuromuscular disorders that might interfere with their ability to exercise or confound the interpretation of the muscle amino acid metabolism studies. Participants received a physical examination, a medical history, blood chemistry profile, complete blood cell count, and a graded treadmill exercise test. The elderly participants also completed a modified physical performance test (PPT) to evaluate their level of physical frailty by objectively determining their ability to do normal activities of daily living (18). All subjects scored 36 out of 36 total possible points, indicating that these participants had a high level of function and were not physically frail.

Each participant's maximum voluntary muscle strength was determined by use of the one-repetition maximum (1-RM) technique. The 1-RM was determined as the heaviest weight that could be lifted once through the complete range of motion. The 1-RM was determined on seven weight-lifting exercises (Nautilus equipment) that included the chest press, inclined chest press, latissimus pull-down (wide and narrow grip), leg press, knee extension, knee flexion, and two free-weight-lifting exercises that included seated overhead press and overhead triceps extension. During the 2-wk exercise period, participants completed ten 1- to 1.5-h weight-lifting exercise sessions: 2–3 sets/day of the nine exercises listed above, 8–12 repetitions/set, 60–90% of maximum voluntary muscle strength.

Each participant's body fat and fat-free mass (FFM) were determined by dual-energy X-ray absorptiometry (Hologic QDR-1000/w system; Waltham, MA). The Hologic-enhanced whole body analysis software (v5.71) was used to process the images and determine body fat mass, FFM, and percentage of body fat.

These tests were followed by a 3-day meat-free controlled protein meal plan that was employed to normalize and stabilize the protein and energy intake of the participants, minimize fluctuations in body wt, and minimize creatinine and 3-methylhistidine intake. This latter requirement permitted the estimation of myofibrillar proteolysis and whole body muscle mass by determining 24-h urinary excretion of 3-methylhistidine and creatinine, respectively (5, 12). The weight maintenance meal plan consisted of 1.1–1.4 g protein$\cdot$kg$^{-1}\cdot$day^{-1}, 14–16% protein energy, 29–30% fat energy, and 54–57% carbohydrate energy. The younger subjects required 39 $\pm$ 1 kcal$\cdot$kg$^{-1}\cdot$day^{-1}, and the older subjects required 31 $\pm$ 1 kcal$\cdot$kg$^{-1}\cdot$day^{-1} to maintain body wt. A research dietician designed the meals, which were prepared in the Research Kitchen in the General Clinical Research Center (GCRC) and were served to the participants during outpatient and inpatient testing visits.

On the third day of the meal plan, each participant was admitted to the GCRC, where they received a 14-h 1-[^{13}C]leucine (MassTrace, Woburn, MA) intravenous infusion (prime = 7.58 μmol/kg; constant infusion = 7.58 μmol$\cdot$kg$^{-1}\cdot$h^{-1}) in the overnight fasted condition. The second [^{13}C]leucine infusion was started within 3–4 h of completion of the last supervised exercise session. Blood samples (5 ml) were collected before and at half-hour intervals during the last 2 h of the [^{13}C]leucine infusion. Plasma [^{13}C]leucine and α-[^{13}C]ketoisocaproic acid (KIC) enrichment were determined in these blood samples (13, 14, 20). Exhaled breath samples were collected into 20-ml evacuated tubes (Becton-Dickinson, Franklin Lakes, NJ) before and during the last hour of the tracer infusion. The ratio of exhaled $^{13}CO_2$ and $^{12}CO_2$ ($^{13}CO_2/^{12}CO_2$) was determined in these samples by means of an automated dual-inlet, triple collector gas isotope ratio mass spectrometer (IRMS). This measurement was used to calculate whole body leucine oxidation rates.

The percutaneous needle muscle biopsy technique was used to remove a sample (100–120 mg) of muscle tissue from the vastus lateralis 1–1.5 h after starting the 1-[^{13}C]leucine infusion. A second muscle sample was removed from the contralateral vastus lateralis muscle 13–14 h after the infusion was started. The muscle samples were rinsed and blotted in sterile normal saline and cleared of any fat and connective tissue. They were immediately frozen in liquid nitrogen and stored at −80°C until analysis.

Sample Analyses and Calculations

Plasma α-KIC was isolated, prepared as the trimethylsilyl quinoxalinol derivative (14, 20), and analyzed for ^{13}C abundance using gas chromatography-electron impact-quadrupole-mass spectrometry (GC-MS; Hewlett-Packard 5890 Series II GC and 5970 Series mass selective detector, Hewlett-Packard, Avondale, PA). Muscle tissue fluid free amino acids were extracted by homogenizing 80–100 mg of tissue in 1–2 ml 10% TCA. The *n*-heptafluorobutyryl propyl esters of the muscle tissue fluid and plasma amino acids were formed (11), and their [^{13}C]leucine abundance was determined using GC-negative chemical ionization-electron capture-quadrupole-MS (Hewlett-Packard 5988A Series II GC and 5970 Series MS). The plasma α-[^{13}C]KIC enrichment and the tissue fluid [^{13}C]leucine enrichment [mole percent excess (MPE)] were used as surrogate measures for muscle [^{13}C]leucyl-tRNA enrichment, the intracellular precursor pool for leucine incorporation into protein. These measures were used to calculate the rates of mixed, MHC, and actin protein synthesis (2–4, 6, 11, 16, 19, 21). Protein synthesis rate (K_s, %/h) was calculated using the equation

$$K_s = \frac{\{[^{13}\text{C}]\text{leucine MPE increment in protein} \times 100\}}{[\text{precursor pool enrichment} \times (t_2 - t_1)]}$$

where ($t_2 - t_1$) was the time elapsed (h) between the two muscle samples.

Analysis of mixed muscle protein synthesis rate. To determine the [^{13}C]leucine abundance in mixed muscle proteins, 10–20 mg muscle samples were homogenized in 1 ml of 10% TCA (Tissumizer, Tekmar, Cincinnati, OH). The TCA extract was removed after centrifugation, and the protein pellet was hydrolyzed in 1 ml 6 N HCl at 110°C for 24 h. The *N*-acetyl-*N*-propyl (NAP) esters of the hydrolyzed amino acids were formed (1) and the [^{13}C]leucine abundance was determined using GC-combustion-IRMS, as previously described (24).

Isolation of MHC and actin for determination of synthesis rates. All procedures used to isolate MHC and actin from muscle samples were performed as previously described (11).

Crude myofibrillar proteins were isolated from 80- to 100-mg muscle samples using high-salt extraction and separated by means of SDS-PAGE. The MHC and actin protein bands were carefully cut from the gel and minced in hydrolysis tubes. The gel pieces were hydrolyzed in 3–4 ml 12 N HCl (110°C for 48 h). The NAP esters of the amino acids were prepared, and [^{13}C]leucine abundance was determined by GC-combustion-IRMS (24). In several muscle samples, it was difficult to recover adequate quantities of actin protein from the gel for [^{13}C]leucine enrichment analysis. Actin synthesis rates were determined in four out of seven older and seven out of seven younger subjects.

Calculation of muscle mass, myofibrillar proteolysis, and whole body protein synthesis. The whole body muscle mass was estimated from the average of three 24-h urinary creatinine excretion measures. Creatinine concentration was determined colorimetrically with an automated analyzer (Kodak Ektachem 700XR, Rochester, NY). Muscle mass was calculated on the basis of the assumption that 1.0 g/day of urinary creatinine is equivalent to 20 kg muscle (12). Total myofibrillar proteolysis was estimated from the average of three 24-h urinary 3-methylhistidine excretion measures. Urinary 3-methylhistidine concentration was determined with an automated amino acid analyzer (Beckman Instruments, Palo Alto, CA). An index of myofibrillar proteolysis was calculated by use of the ratio between the daily urinary 3-methylhistidine excretion and the daily creatinine excretion (μM 3-methylhistidine/mM creatinine).

The whole body leucine kinetic rates were calculated as previously described (13, 14, 16). The plasma leucine rate of appearance (R_a, leucine flux) is an estimate of the whole body rate of proteolysis and was determined using the average plasma α-[^{13}C]KIC enrichment measured during the last 2 h of the tracer infusion. The rate of whole body leucine oxidation was determined by use of the breath $^{13}CO_2$ abundance and the CO_2 production rate measured with a DeltaTrac open-circuit indirect calorimeter (Sensormedics, Yorba Linda, CA). The whole body nonoxidative leucine disposal rate is an estimate of the whole body rate of protein synthesis and was calculated as the difference between the whole body proteolytic rate and the leucine oxidation rate (13, 14, 16).

Statistics

The means and (the standard errors of the means) SE are reported. A 2-group repeated-measures ANOVA was used for between-group comparisons of mixed, MHC, and actin fractional synthesis rates and to compare baseline (initial) measures with the end of exercise (final) between the two age groups. A Student-Newman-Keuls post hoc analysis was used when the 2-group ANOVA indicated a significant ($P \leq 0.05$) main interaction. Change within a group was calculated as final measure minus initial and compared between age groups by means of a *t*-test.

RESULTS

In comparison with the older participants, the younger participants were taller ($P = 0.014$), had a lower percentage of body fat ($P = 0.005$), and had a greater whole body muscle mass ($P = 0.013$; Table 1). The 23–32 yr olds had greater maximum voluntary muscle strength (1-RM) than the older participants on all nine exercises tested ($P < 0.05$; Table 2). With only 3–4 men and women in each group of younger and elderly, no attempt was made to examine gender differences for any of the parameters; instead, kinetic parameters are expressed per unit FFM or muscle mass to correct for differences in leanness between younger and older men and women.

Table 1. *Descriptive characteristics*

Age of Group, yr	Age, yr	Weight, kg	Height, cm	Body Fat, %	FFM, kg	Muscle Mass, kg
23–32	27 ± 1	69.3 ± 4.1	176 ± 4*	17 ± 3†	57 ± 4	31 ± 3*
78–84	80 ± 1	68.4 ± 4.3	162 ± 3	31 ± 3	47 ± 4	17 ± 3

Values are mean ± SE; FFM, fat-free mass; *$P < 0.05$ and †$P < 0.01$ vs. older group.

The plasma α-[^{13}C]KIC enrichments determined during the final 2 h of the tracer infusion done at baseline and at the end of exercise were significantly greater than the tissue fluid [^{13}C]leucine enrichments for both younger and older participants ($P < 0.01$; Table 3). Initial and final plasma α-[^{13}C]KIC values were greater for the older participants than for the younger ($P < 0.01$). Initial and final tissue fluid [^{13}C]leucine was not significantly different between younger and older groups ($P > 0.05$). In the older participants, the muscle tissue fluid [^{13}C]leucine enrichment was equivalent to 70% of their plasma α-[^{13}C]KIC enrichment. In the younger participants, tissue fluid [^{13}C]leucine enrichment was 84% of their plasma α-[^{13}C]KIC enrichment.

By use of plasma α-[^{13}C]KIC to represent the ^{13}C enrichment in the precursor pool for protein synthesis (Fig. 1), the younger subjects were determined to have greater initial mixed and MHC protein synthesis rates than the older participants (mixed $P = 0.041$; MHC $P < 0.001$). At the end of 2 wk of weight-lifting exercise, MHC and mixed protein synthesis rates increased in both younger and older participants ($P < 0.001$; Fig. 1). The exercise-induced increases in MHC and mixed protein synthesis rates were similar (in absolute terms) in the younger and older groups ($P \geq 0.23$). In the younger group, the MHC protein synthesis rate increased (88%) from 0.038 ± 0.003%/h to 0.072 ± 0.002%/h, and the mixed muscle protein synthesis rate increased (121%) from 0.048 ± 0.003%/h to 0.100 ± 0.006%/h after 2 wk of exercise. In the older group, MHC protein synthesis increased (105%) from 0.024 ±

Table 2. *Baseline maximum voluntary muscle strength 1-RM in kg in younger and older participants*

Exercise	23–32 Yr Old	78–84 Yr Old
Chest press	48 ± 7	21 ± 3
Incline chest press	38 ± 6	20 ± 3
Overhead press	13 ± 2	6 ± 1
Triceps extension	6 ± 1	4 ± 1
Wide-grip lat. pulldowns	43 ± 5	22 ± 2
Narrow-grip lat. pulldowns	30 ± 3	20 ± 2
Leg press	53 ± 6	41 ± 4
Knee flexion	37 ± 3	16 ± 2
Knee extension	59 ± 6	33 ± 3
Average	36 ± 6	20 ± 4

Values are means ± SE. Younger subjects were stronger than older subjects on all exercises ($P < 0.05$). 1-RM, one-repetition maximum; lat., latissimus.

Table 3. *Plasma α-[13C]KIC and tissue fluid [13C]leucine enrichments*

Group	Plasma α-[13C]KIC	Tissue Fluid [13C]leucine
Younger		
Initial	6.88 ± 0.18	5.80 ± 0.29
Final	6.56 ± 0.22	5.54 ± 0.26
Older		
Initial	8.19 ± 0.26	5.74 ± 0.21
Final	7.82 ± 0.12	5.45 ± 0.42

Mean ± SE. Plasma α-[13C]ketoisocaproic acid (α-[13C]KIC) was derived from average enrichment in 5 blood samples collected during last 2 h of infusion. Initial and final plasma α-[13C]KIC enrichment were significantly greater than tissue fluid [13C]leucine enrichment in both groups ($P < 0.01$). Initial and final tissue fluid [13C]leucine values were not significantly different between young and old groups ($P > 0.05$).

0.002%/h to 0.050 ± 0.007%/h, and mixed muscle protein synthesis increased (182%) from 0.037 ± 0.003%/h to 0.102 ± 0.009%/h after 2 wk of exercise. In the younger group, the baseline MHC protein synthesis rate was equivalent to 80% of the corresponding mixed muscle protein synthesis rate. In the older group, baseline MHC was equivalent to 65% of the mixed muscle protein synthesis rate. The initial rates of actin protein synthesis were not different between the younger (0.055 ± 0.010%/h) and older groups (0.063 ± 0.015%/h; $P = 0.59$), although this finding was limited to four out of seven older subjects. The actin protein synthesis rate tended to increase after 2 wk of exercise in both age groups [P = not significant (NS)]. In the younger group, the baseline actin protein synthesis rate was 14% faster than the mixed muscle protein synthesis rate. In the older group, the actin protein synthesis rate was 68% faster than the mixed muscle protein synthesis rate. The interindividual variability in the magnitude of the exercise-induced increments in protein synthetic rates was large: mixed (27–309%), MHC (6–175%), and actin (41–240%). As a result, the increments in mixed muscle protein synthesis rates were not greater than the increments in MHC or actin synthesis rates ($P > 0.05$) in either age group.

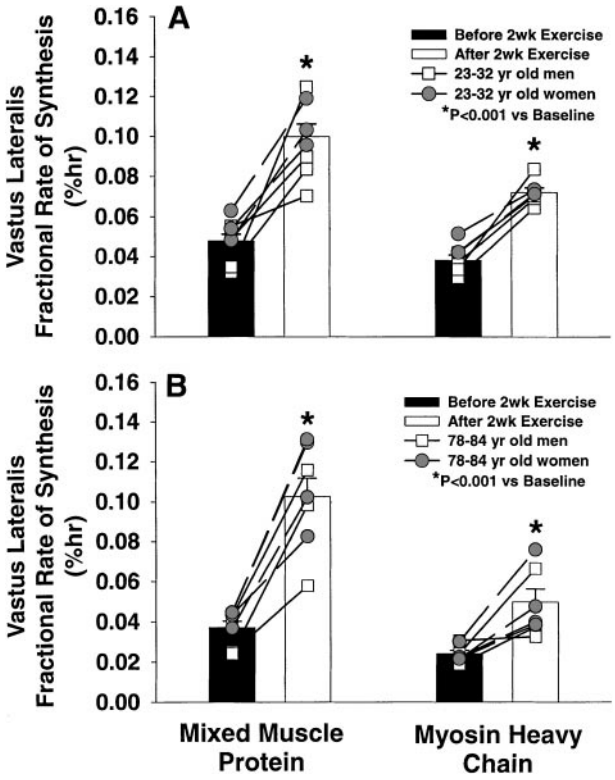

Fig. 1. Mixed and myosin heavy-chain (MHC) protein synthesis rates determined before and after short-term resistance exercise in 23- to 32-yr-old (*A*) and 78- to 84-yr-old (*B*) women and men. Plasma α-[13C]ketoisocaproic acid (α-[13C]KIC) was used to reflect enrichment in precursor pool for protein synthesis. Baseline MHC and mixed muscle protein synthesis rates were lower in 78–84 yr olds than in 23–32 yr olds ($P \leq 0.04$). See text for actin protein synthesis rates.

Muscle tissue [13C]leucine enrichment was lower than plasma α-[13C]KIC enrichment, so the mixed, MHC, and actin protein synthesis rates that were calculated by use of muscle tissue fluid [13C]leucine enrichment were greater than those calculated by use of plasma α-[13C]KIC (Table 4). Although it underestimates the actual rates of protein synthesis (6), plasma α-[13C]KIC is used because it allows us to compare the present findings with our previous studies, and it allows us to compare contractile protein synthesis rates with whole body protein synthesis rates. Using muscle tissue fluid [13C]leucine to represent the 13C enrichment in the precursor pool for protein synthesis (Table 4), the younger subjects had a greater initial MHC protein synthesis rate than the older participants ($P < 0.05$). At the end of 2 wk of weight-lifting exercise, MHC and mixed protein synthesis rates increased in both younger and older participants ($P < 0.001$; Table 4). The actin protein synthesis rates was increased after exercise in only the younger group ($P < 0.05$). The magnitude of the exercise-induced increase in MHC and mixed protein synthesis rates was similar in the younger and older groups ($P \geq 0.32$). In the younger group, the MHC protein synthesis rate increased (83%) from 0.047 ± 0.004%/h to 0.086 ± 0.004%/h, the mixed muscle protein synthesis rate increased (102%) from 0.057 ± 0.006%/h to 0.115 ± 0.008%/h, and the actin protein synthesis rate increased (78%) from 0.064 ± 0.014%/h to 0.114 ± 0.020%/h after 2 wk of exercise. In the older group, MHC protein synthesis increased (144%) from 0.032 ± 0.004%/h to 0.078 ± 0.012%/h, and mixed muscle protein synthesis increased (166%) from 0.056 ± 0.007%/h to 0.149 ± 0.017%/h after 2 wk of

Table 4. *Initial and final fractional synthesis rates (%/h) for mixed muscle, MHC, and actin proteins calculated with tissue fluid [13C]leucine as the precursor pool enrichment*

Group	Mixed	MHC	Actin
Younger			
Initial	0.057 ± 0.006	0.047 ± 0.004†	0.064 ± 0.014
Final	0.115 ± 0.008*	0.086 ± 0.004*	0.114 ± 0.020*
Older			
Initial	0.056 ± 0.007	0.032 ± 0.004	0.091 ± 0.016
Final	0.149 ± 0.017*	0.078 ± 0.012*	0.161 ± 0.044

Mean ± SE. MHC, myosin heavy chain. *$P < 0.05$ vs. corresponding initial measure. †$P = 0.027$ vs. initial for older group.

exercise. Based on a limited number of analyses, the initial rates of actin protein synthesis were not different between the younger and older groups ($P = 0.31$). In the younger group, the baseline actin protein synthesis rate was 13% faster than the mixed muscle protein synthesis rate ($P = 0.83$). In the older group, actin protein synthesis rate was 63% faster than the mixed muscle protein synthesis rate ($P = 0.003$), but this was based on a limited number of analyses.

Whole body leucine kinetic parameters were normalized to FFM (Table 5). As previously observed, the rates of plasma leucine flux (whole body proteolysis), leucine oxidation, and nonoxidative leucine disposal (whole body protein synthesis) were similar between younger and older participants when expressed per kg FFM. At the end of 2 wk of resistance training, these whole body kinetic rates were unchanged compared with baseline in both younger and older groups.

The urinary 3-methylhistidine excretion estimates of myofibrillar proteolysis were not affected at the end of 2 wk of resistance exercise in either the younger or older participants. At baseline, the daily urinary 3-methylhistidine-to-creatinine ratio in the younger participants was 13.0 ± 0.6 μM/mM, and at the end of exercise it was 13.7 ± 0.6 μM/mM. In the older participants, the 3-methylhistidine-to-creatinine ratio was 15.1 ± 0.6 μM/mM at baseline and 15.5 ± 0.6 μM/mM at the end of exercise training. At baseline, the urinary 3-methylhistidine-to-creatinine ratio in the older participants was greater than in the younger participants ($P = 0.03$).

There was a direct correlation ($P < 0.001$) between the rate of MHC protein synthesis and the rate of mixed muscle protein synthesis (Fig. 2) in both age groups when plasma α-[^{13}C]KIC was used to reflect the precursor pool enrichment, and baseline and final measures of mixed and MHC protein synthesis were included in the analysis (younger: $y = 0.52x + 0.02$, $r^2 = 0.68$; older: $y = 0.35x + 0.01$, $r^2 = 0.61$). Also, when tissue fluid [^{13}C]leucine was used as a surrogate measure of the precursor pool enrichment, mixed and MHC protein synthesis rates were directly correlated ($r = 0.60$, $P = 0.03$). Similar relationships for actin and mixed, or actin and MHC protein, synthesis rates were not observed.

Table 5. *Whole body leucine flux, nonoxidative leucine disposal, and leucine oxidation rates determined before and after short-term resistance exercise in 23–32 yr olds and nonfrail 78–84 yr olds*

Group	Flux Rate	Nonoxidative Disposal Rate	Oxidation Rate
	μmol·kg FFM^{-1}·h^{-1}		
Younger			
Initial	128 ± 6	97 ± 4	31.5 ± 1.9
Final	134 ± 7	110 ± 6	24.4 ± 1.7
Older			
Initial	123 ± 4	88 ± 3	34.6 ± 2.0
Final	129 ± 6	102 ± 7	27.3 ± 1.4

Mean ± SE. No differences were found between younger and older subjects or before and after exercise.

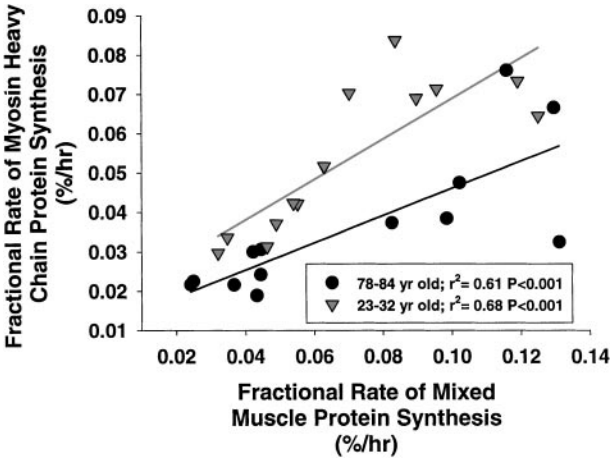

Fig. 2. Fractional rate of MHC protein synthesis was directly correlated with fractional rate of mixed muscle protein synthesis in both younger and older men and women. Measurements made before and after 2 wk of resistance exercise have been included.

DISCUSSION

The findings indicate that healthy 78- to 84-yr-old women and men with muscle weakness but who are not physically frail have lower MHC and mixed vastus lateralis muscle protein synthesis rates than 23–32 yr olds. This implies that muscle weakness and a reduction in the rate of muscle protein synthesis occur with advancing age, even in 78–84 yr olds who are not physically frail (as determined by PPT). Second, baseline actin protein synthesis rates were similar between the two age groups and faster than mixed and MHC protein synthesis rates. However, measurement variability was considerably greater for the actin protein synthesis rate than for the MHC or mixed muscle protein. Third, as a result of their lower whole body skeletal muscle mass, 78–84 yr olds had lower absolute rates (g protein synthesized/day) of MHC, mixed, and actin protein synthesis than 23–32 yr olds. Fourth, short-term resistance exercise increased mixed muscle and MHC protein synthesis rates in both age groups. This occurred regardless of whether plasma α-[^{13}C]KIC or muscle tissue fluid [^{13}C]leucine was chosen as the precursor pool for protein synthesis. Acute exercise tended to increase actin protein synthesis rates, but statistical significance was not achieved. This indicates that acute weight-lifting exercise induces an increase in the contractile protein synthesis rate in 78–84 yr olds similar to that observed in 23–32 yr olds. Thus the potent contractile protein anabolic actions of acute resistance exercise are not compromised by advanced age. Fifth, the direct correlation between mixed muscle and MHC protein synthesis rates (Fig. 2), combined with the observation that 2 wk of weight-lifting exercise increased mixed and MHC protein synthesis rates similarly in the younger and older subjects, suggests that measurements of the mixed muscle protein synthesis rate reflect resistance exercise-induced increments in the MHC protein synthesis rate. This might have been expected because MHC is the primary contractile pro-

tein present in skeletal muscle and constitutes ~25% of muscle protein (2, 3). However, the correlation found here cannot be applied generally to all experimental or pathological conditions where muscle protein wasting or anabolism occurs. For example, endurance exercise training would be predicted to have different and disparate effects on mitochondrial, enzymatic, sarcoplasmic, and contractile protein synthesis rates (4, 15, 19). Finally, the rates of whole body proteolysis, protein synthesis, and leucine oxidation were not affected by 2 wk of resistance exercise, and when expressed in kilograms of FFM were not different between younger and older subjects.

When muscle tissue fluid [^{13}C]leucine was used to reflect ^{13}C abundance in the precursor pool for protein synthesis, the MHC protein synthesis rate was lower in older than in younger participants, but the mixed muscle protein synthesis rates were similar between younger and older participants. Balagopal et al. (4) reported similar findings when MHC and mixed muscle protein synthesis rates were determined in younger (23 ± 1 yr), middle-aged (52 ± 1 yr), and older (77 ± 2 yr) women and men. This may be attributed to the finding that tissue fluid [^{13}C]leucine enrichment was similar in younger and older subjects, whereas plasma α-[^{13}C]KIC enrichment was lower in the younger subjects. Balagopal et al. reported a ratio between tissue fluid [^{13}C]leucine and plasma α-[^{13}C]KIC to be 78% in younger, 63% in middle-aged, and 76% in older subjects. We found this ratio to be 84% in 23- to 32-yr-old and 70% in 78- to 84-yr-old women and men. Our findings suggest that advancing age only modestly alters the relationship between muscle tissue fluid [^{13}C]leucine and plasma α-[^{13}C]KIC. This may reflect differences in amino acid-ketoacid transport rates in muscles from older and younger subjects. It does not appear that acute resistance exercise altered the relationship between plasma α-[^{13}C]KIC and muscle tissue fluid [^{13}C]leucine in younger or older subjects.

We found that muscle proteins in 78- to 84-yr-old women and men respond to 2 wk of weight-lifting exercise by increasing mixed, MHC, and, to a lesser degree, actin protein synthesis rates. These exercise-induced increments in mixed and MHC were observed regardless of the choice of precursor pool enrichment. The exercise-induced increments in contractile protein synthesis rates observed in the 78–84 yr olds were similar to those observed in 23–32 yr olds after 2 wk of weight-lifting exercise at the same exercise intensity relative to baseline maximum voluntary muscle strength. This supports the notion that muscle contractile proteins respond acutely to weight-lifting exercise by increasing their rates of synthesis, even in 78–84 yr olds. This extends our previous observations in 63–66 yr olds who increased mixed muscle protein synthesis rates by 155 ± 24% after 2 wk of weight-lifting exercise. Our previous work (24, 25) and that of Phillips et al. (17) support the notion that the dramatic increments in mixed and MHC protein synthesis rates induced by acute (2 wk) resistance exercise are attenuated when the exercise program is continued for several more weeks and the participants' muscles become trained and accustomed to weight-lifting exercise.

With the use of plasma α-[^{13}C]KIC, the baseline MHC fractional synthesis rates for our younger (0.038 ± 0.003%/h) and older (0.024 ± 0.002%/h) subjects were similar to values previously reported by Balagopal et al. (4) for younger (0.036 ± 0.001%/h) and older (0.022 ± 0.002%/h) adults. In vivo human actin protein synthesis rates determined before and after resistance exercise have not been reported. The muscle actin content is substantially less than the MHC content. In several muscle samples, it was difficult to recover adequate quantities of actin protein for [^{13}C]leucine enrichment analysis; as a result, we had less statistical power to demonstrate that short-term weight-lifting exercise induces an increase in the actin protein synthesis rate. At baseline, it appears that human actin protein synthesis rates are faster than MHC or mixed muscle protein synthesis rates in both age groups.

The higher baseline urinary 3-methylhistidine-to-creatinine ratio in 78- to 84-yr-old men and women suggests that myofibrillar proteolysis was greater in the older than in the younger subjects. This, in combination with the lower rates of contractile protein synthesis observed in the older subjects, contributes to the muscle wasting that accompanies advanced age. Short-term weight-lifting exercise did not increase urinary 3-methylhistidine/creatinine excretion, suggesting that the exercise program did not increase the rate of myofibrillar proteolysis. Previous studies have not found a greater baseline urinary 3-methylhistidine-to-creatinine ratio in 63- to 66-yr-old women and men compared with 24-yr-old women and men, and 2 wk of resistance exercise did not increase 3-methylhistidine/creatinine excretion (22, 25). The current finding suggests that with very old age (78–84 yr) the possibility exists that both increased muscle proteolysis and reduced synthesis rates contribute to sarcopenia; however, we and others have questioned the appropriateness of using urinary 3-methylhistidine excretion as an indicator of whole body myofibrillar proteolysis in combination with measures of muscle protein synthesis localized to the vastus lateralis muscle (5, 22, 23, 25). It is possible that vastus lateralis muscle proteolysis was increased after 2 wk of exercise but that urinary 3-methylhistidine/creatinine was not adequately sensitive or specific to detect this increase.

Mixed and MHC protein synthesis rates were lower in 78–84 yr olds than in 23–32 yr olds. Two weeks of weight-lifting exercise rapidly stimulated MHC and mixed muscle protein synthesis rates in 23–32 yr olds and nonfrail 78–84 yr olds. Under these conditions, MHC and mixed muscle protein synthesis rates were directly related and increased similarly in the younger and older participants. Myofibrillar proteins retain the ability to increase their rates of synthesis in response to a short period of weight-lifting exercise, even in sarcopenic 78+ yr olds.

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