Effective Timing of Curcumin Ingestion to Attenuate Eccentric Exercise–Induced Muscle Soreness in Men

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Summary Curcumin is known to have potent anti-inflammatory effects. We have reported that acute curcumin ingestion attenuates eccentric exercise–induced muscle damage. This study aimed to examine the effect of curcumin ingestion timing (before or after exercise) on the changes in muscle damage markers after eccentric exercise. In this randomized, single-blind, parallel design study, 24 healthy young men performed 30 maximal isokinetic (120˚/s) eccentric contractions of the elbow flexors using an isokinetic dynamometer. Subjects were randomly assigned to ingest 180 mg/d of oral curcumin either 7 d before (PRE) or 4 d after exercise (POST) or 180 mg/d of oral placebo 4 d after exercise (CON). The maximal voluntary contraction (MVC) torque of the elbow flexors, elbow joint range of motion (ROM), muscle soreness, and serum creatine kinase (CK) activity were measured before, immediately after, and 1–4 d after exercise. Changes in these variables were compared over time. In the POST group, ROM were higher at 3–4 d and muscle soreness was lower at 3 d after exercise compared with the CON group (p < 0.05). However, in the PRE group, there were no significant differences compared with the CON group in changes in ROM and muscle soreness. Meanwhile, there were no significant differences among the groups in terms of changes in MVC torque and serum CK activity. Our results suggest that curcumin ingestion after exercise had a more beneficial effect in attenuating muscle soreness.

Key Words eccentric contractions, elbow flexors, maximal voluntary contraction, delayed-onset muscle soreness, supplementation

Unfamiliar exercises and/or muscle contractions during high-intensity exercise, especially eccentric exercise, can induce muscle damage (1). This mechanical stress triggers an inflammatory response and the production of reactive oxygen species by promoting the activation of transcription factors, such as nuclear factor κB (NF-κB), which leads to secondary muscle damage (1–3). As a result, it is suggested that this secondary muscle damage leads to prolonged decreases in muscle strength and range of motion (ROM), delayed onset muscle soreness (DOMS), and increases in creatine kinase (CK) activity in the blood (4). Muscle function impairment caused by damage and its subsequent inflammatory responses may reduce the ability to perform daily training and reduce athletic performance. Therefore, it is important to prevent or minimize muscle damage and promote recovery after exercise.

Curcumin is a natural polyphenolic substance extracted from turmeric. It has various physiological effects, such as membrane-protective effects and anti-inflammatory and antioxidant responses (5, 6). These mechanisms have been reported to suppress the activity of NF-κB, which is a modulating factor for cytokines and cyclooxygenase (7). Therefore, these actions are considered to have a positive effect on exercise-induced muscle damage. We previously reported that curcumin (150 mg) ingestion 1 h before and 12 h after eccentric exercise (total: 300 mg curcumin ingestion) attenuates the loss of maximal voluntary contraction (MVC) torque and increases in CK activity after exercise (8). In addition, previous studies have reported that curcumin ingestion before and after exercise improves DOMS (9, 10) and decreases CK activity (11). Thus, it is possible that curcumin ingestion before and after exercise can effectively attenuate eccentric exercise–induced muscle damage in humans. However, little is known about the effective timing (before or after exercise) of ingestion of curcumin for muscle damage.

Therefore, the purpose of this study was to compare the effect of curcumin ingestion before or after exercise on changes in muscle damage markers after eccentric exercise in a randomized, single-blind parallel study.

MATERIALS AND METHODS

Subjects. Twenty-four healthy men were recruited in the present study. They were randomly assigned to one of three groups with different supplement inges-
Curcumin Ingestion Timing for Muscle Damage

Supplementation & frequency
1: Placebo after breakfast
2: Placebo after dinner
3: Curcumin after breakfast
4: Curcumin after dinner

Measurements (MVC, ROM, soreness, CK)

Fig. 1. Schematic illustration of the experimental protocol. All subjects ingested a 90-mg curcumin supplement (closed circles) twice daily (total: 180 mg/d) after breakfast and dinner or an equivalent oral placebo (open circles). On the exercise day, subjects performed 30 maximal eccentric contractions of the elbow flexors. The MVC torque of the elbow flexors, ROM of the elbow joint, muscle soreness, and serum CK activity were measured before (Pre), immediately after (Post0), and 1–4 d (1 d, 2 d, 3 d, and 4 d, respectively) after exercise. Ex, exercise; MVC, maximal voluntary contraction; ROM, range of motion; CK, creatine kinase; CON, group that ingested placebo; PRE, group that ingested curcumin pre-exercise; POST, group that ingested curcumin post-exercise.

Supplementation: curcumin and placebo. All subjects ingested a 90-mg curcumin supplement (TheracurminCR-033P; Theravalues Corporation, Tokyo, Japan) twice daily (total: 180 mg/d) after breakfast and dinner or an equivalent oral placebo. This ingestion dose was determined based on curcumin’s acceptable daily intake (3 mg/kg/d), supported by the European Food Safety Authority database, assuming that the body weight of a healthy adult is 60 kg (12). Theracurmin is a highly bioavailable curcumin that was developed via using a microparticulation and surface-processing technique and has been shown to result in a much higher plasma concentration after intake compared with conventional curcumin powder (13, 14). TheracurminCR-033P consisted of 30.0% curcumin, 6.0% other curcuminoids, 38% maltose, 14.6% gum ghatti, 10.6% maltodextrin, and 0.8% citric acid. Placebo consisted of 11.0% Food Yellow No. 4 (tartrazine), 55.6% maltose, 20.3% gum ghatti, 12.1% maltodextrin, and 1.0% citric acid. The two supplements both contain the same additives except for curcumin. Placebo was ingested in capsule form identical to that of the curcumin to prevent the subjects from distinguishing between the capsules.

In the PRE group, curcumin was ingested 7 d before eccentric exercise; it was taken 24 h before the exercise to prevent the acute effect of Theracurmin. Meanwhile, in the POST group, curcumin was ingested 4 d after eccentric exercise. As a comparison condition, placebo was ingested 4 d after eccentric exercise.

Eccentric exercise. All subjects performed eccentric exercise of the elbow flexors on a BIODEX dynamometer (BIODEX System 4; Biodex Medical Systems, Inc., Shirley, NY, USA). Each subject was seated on the dynamometer chair with the arm supported by a padded armrest and secured at 45° shoulder flexion. The waist and chest were stabilized with straps. The forearm was supinated,
and the wrist was tightly fastened onto the lever arm. The exercise consisted of 30 maximal eccentric contractions of the elbow flexors at an angular velocity of 120°/s. The exercise motion range was from 130° to 10° of elbow flexion, where a fully extended elbow joint angle was considered to be 0°; thus, each contraction lasted 1 s (8, 15). The exercise limb was passively returned to the initial position at 10°/s, creating a 12-s rest between contractions. The subjects were instructed to contract the elbow flexors maximally to resist the elbow-extending action of the dynamometer for the whole motion range. The total work maintained at 120°/s was calculated using MATLAB software (MATLAB R2014a; The MathWorks, Inc., Natick, MA, USA).

Muscle damage markers.

MVC torque: The MVC torque of elbow flexion was measured using an isometric dynamometer (VTE-002; VINE, Tokyo, Japan). Subjects sat on a test chair with their arm positioned at 90° flexion of the shoulder and elbow joints and the wrist was fixed in a neutral position. The subjects performed three MVCs lasting about 5 s with a 30-s rest between trials. Torque signal was converted into a digital signal using an AD converter (PowerLab/16SP; ADInstruments, Castle Hill, Australia) and was recorded in a personal computer at 100 Hz. Peak active torque during the MVC was measured using LabChart version 7.2.5 (ADInstruments, Dunedin, New Zealand). The highest value of the three trials was used for analysis.

ROM: ROM was determined as the difference between the elbow joint angles. According to the protocol of a previous study (8, 16), each subject was asked to actively extend the elbow joint maximally (extended elbow joint angle) and to fully flex the elbow joint with the hand in supinated position (flexed elbow joint angle). Each angle was measured twice using a goniometer, and the mean of the two measures was used for the calculation of ROM.

Muscle soreness: Muscle soreness upon palpation of the upper arm and passive extension of the elbow joint was quantified using a visual analogue scale (VAS) with a 100-mm line with “no pain” at one end and “extremely sore” at the other end (8, 17). The subjects were instructed to sit with the arm relaxed, and the investigator palpated over the biceps brachii and extended the elbow joint maximally. The same investigator performed the tests in all participants to avoid inter-investigator measurement error.

Blood sampling and analyses: Blood was collected from the antecubital vein using a standard venipuncture technique. To obtain serum for CK activity, a 5-mL blood sample was collected using commercially produced vacuum-sealed serum collection tubes. To obtain plasma for the curcumin concentration, a 5-mL blood sample was collected using a tube containing ethylene-diaminetetraacetic acid. These tubes were centrifuged at 3,000 rpm for 10 min at 4°C, and the serum and plasma samples were stored at −80°C until analysis. The preparations and methods for measuring plasma curcumin concentrations were described previously (13, 18). In brief, each plasma sample (0.02 mL) was incubated with 0.1 mL sodium acetate buffer (pH 5.0) containing 1,000 U β-glucuronidase (Wako) at 37°C for 1 h to hydrolyze the curcumin conjugates. After the addition of 0.01 mL of internal standard (IS) working solution (500 ng/mL), 0.5 mL of chloroform was added as an extraction solvent. After extraction with chloroform, the dried extracts were reconstituted in 0.1 mL of 50% methanol and injected into a chromatographic system. Plasma concentrations of curcumin were measured using the HPLC-MS/MS system comprising the Prominence micro-LC system (Shimadzu, Kyoto, Japan) and an API 3200 tandem mass spectrometer (Applied Biosystems, Foster City, CA, USA) with (+) ESI, as described previously (14). Serum CK activity was measured using a test kit (Iatro LQ CKJ II; LSI Medience Corporation, Tokyo, Japan) from a commercial laboratory.

Statistical analyses. The Kolmogorov-Smirnov test was used to check for normality of distribution of parameters. In normally distributed parameters (MVC, ROM and muscle soreness in the palpation of the upper arm), the one-way analysis of variance (ANOVA) was used among the groups. When it showed significant differences, Tukey’s post hoc test was used to determine the differences among the groups. The level of statistical significance was set at p<0.05. In the non-normally distributed parameters (plasma curcumin concentration, muscle soreness in the extension of the elbow joint and serum CK activity), the non-parametric Kruskal-Wallis test was used among the groups. When it showed significant differences, a Mann-Whitney U test was used to determine the differences among the groups. The level of statistical significance was adjusted by Bonferroni (p<0.017). The subject characteristics and total work were performed using one-way ANOVA. All data are shown as mean±standard deviation (SD). Statistical analyses were performed using SPSS 24.0 (IBM Corp., Armonk, NY, USA).

RESULTS

Plasma curcumin concentration

At baseline, the plasma curcumin concentration of the PRE group was significantly higher than that in the CON and POST groups. At 1–4 d after exercise, the plasma curcumin concentration of the POST group was significantly higher than that of the CON and PRE groups and that at 1–3 d after exercise, the plasma curcumin concentration of the PRE group was significantly higher than that of the CON group (Fig. 2).

Exercise

The total work during eccentric exercise did not differ among the CON, PRE, and POST groups (1.644±410, 1.561±393, and 1.662±190 J, respectively).

MVC torque

MVC torque before exercise was not significantly different among the CON, PRE, and POST groups. As shown in Fig. 3, there were no significant differences among the three groups at all time points.

ROM

At baseline, ROM was similar among the three
groups. After eccentric exercise, ROM was significantly better 3–4 d after exercise in the POST group than in the CON group (Fig. 4).

Muscle soreness

In the palpation of the upper arm, the VAS score was significantly lower at 3 d after exercise in the POST group than in the CON and PRE groups (Fig. 5a). In the extension of the elbow joint, the VAS score was significantly lower at 4 d after exercise in the POST group than in the CON group (Fig. 5b).

Serum CK activity

Serum CK activity was within the normal reference range before exercise, without a significant difference among the three groups. There were no significant differences in the changes among the groups at all time points (Fig. 6).
DISCUSSION

This study investigated the effect of curcumin ingestion before or after eccentric exercise on changes in muscle damage markers in healthy young men. Results showed that curcumin ingestion after exercise significantly improved ROM and muscle soreness as compared with the CON group who ingested placebo. However, curcumin ingestion before exercise did not affect the changes in muscle damage markers compared with those of the CON group. These results suggest that curcumin ingestion after exercise had a more beneficial effect in attenuating muscle damage.

In this study, the ROM of the elbow joint and muscle
soreness, as evaluated via VAS, were measured as markers of muscle damage. In the group who ingested curcumin before exercise, there was no significant difference in ROM or muscle soreness compared with that of the CON group. Meanwhile, in the group that ingested curcumin after the exercise, significant improvements in ROM and attenuation of muscle soreness were observed after 3–4 d of exercise. In the previous study, curcumin had a significant effect in suppressing the production of cyclooxygenase (19), prostaglandin E$_2$ (20), and histamine (21), which are algogenic substances. Furthermore, curcumin is associated with desensitization of nociceptor (reduction of nociceptive response) evaluated electrophysiologically with animal neurons in vitro and inhibition of transient receptor ion channels involved in the generation of painful stimuli such as TRPV1 and TRPA1 (22, 23). Thus, it is considered that muscle soreness may be suppressed by ingestion of curcumin after exercise due to a decrease in production of pain substance and pain sensation. Meanwhile, the days with significant improvement in ROM and muscle soreness were matched in the group that ingested curcumin after exercise. Therefore, there is a possibility that the restricted ROM was improved with the reduction of muscle soreness.

In this study, the plasma curcumin concentration of the PRE group remained about 40 ng/mL during exercise and almost 0.0 ng/mL for 2–4 d after the exercise. Meanwhile, in the POST group, the plasma curcumin concentrations were maintained at about 40–50 ng/mL for 4 d after the exercise, but there was no curcumin in the serum during exercise. In a previous study, continuous curcumin ingestion before and after exercise attenuates muscle soreness due to leg press (9) and downhill runs (10). However, when curcumin (150 mg) was ingested once before and after the exercise, the plasma curcumin concentration was less than 10 ng/mL 2 d after exercise, and there were no significant improvements in muscle soreness compared with those who ingested placebo (8). Thus, to improve muscle soreness, it may be necessary to ingest curcumin continuously after exercise to increase blood curcumin concentration.

MVC torque and serum CK activity measured in this study changed significantly after the exercise compared to before, but no significant difference was observed in the three groups. Curcumin is known to suppress the activity of NF-$\kappa$B and attenuate the decrease in muscle strength and has the effect of promoting muscle regeneration (24, 25). Moreover, regeneration from muscle damage due to eccentric exercise of the elbow flexors begins on day 3 after exercise (26). In this study, since the follow-up of the recovery period was short (4 d), the possibility that the period was not enough to determine the effect of curcumin is considered. Besides, serum CK activity is often used as a muscle damage marker in the blood (4, 8, 15), but it is known that the change after exercise is greatly different among individuals, and it is influenced by heredity (27, 28) and training history (29). In this study, we conducted experiments with three independent groups. Hence, individual variability was large (one subject in the PRE group had serum CK activity up to 50,000 IU/L), and the comparison with the same subject is preferable. Furthermore, in our previous study using a similar eccentric exercise protocol, the average serum CK activity in the control group was 7,600 IU/L and the decrease rate of MVC torque was $-40\%$ (8). In this study, it is considered that the degree of muscle damage was greater than that of the previous...
study because the serum CK activity and the decrease rate of MVC torque were much higher than those in the previous study. Therefore, it is possible that the effect of curcumin could be veiled by the considerably larger degree of muscle damage.

The limitation of this study was that the period of supplement ingestion could not be unified. In this study, subjects were unaware of the supplement they had ingested; thus, blindness to the beneficial effect of the supplement was maintained. However, the ingestion period was disjointed for each group. In the future, the supplement was maintained. However, the ingestion period was disjointed for each group. In the future, adopting an experiment design that standardizes the ingestion period of supplements before and after exercise among groups is needed.

In conclusion, the present study showed that ROM and muscle soreness were improved with curcumin ingestion 4 d after eccentric exercise of the elbow flexors. Meanwhile, curcumin ingestion before exercise had no effect on muscle damage markers in this study. These findings suggest that even if curcumin is ingested before exercise, the protective effect against muscle damage may be extremely small and that curcumin ingestion after eccentric exercise was more effective in attenuating muscle soreness and may contribute to improvement of performance after eccentric exercise–induced muscle damage.

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REFERENCES
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