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Bioavailability of synthetic and biosynthetic deuterated lycopene in humans☆

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Abstract

Current knowledge of the bioavailability of lycopene in humans is limited due to the inability to distinguish newly administered lycopene from the body reserves of lycopene. A quantitative method to assess the absorption and relative bioavailability of newly absorbed synthetic or natural lycopene was developed using two deuterated lycopene sources, in conjunction with an advanced LC/APCI-MS (liquid chromatography/atmospheric pressure chemical ionization-mass spectrometry) to analyze newly absorbed lycopene in blood samples of study subjects. Two subjects (1 male and 1 female) consumed hydroponically grown tomatoes containing deuterium-enriched lycopene (80–84 g wet weight tomato containing 16.3 and 17.4 μmol lycopene, respectively) and two subjects (1 male, and 1 female) consumed 11 μmol synthetic $^2H_{10}$ lycopene in 6 g of corn oil. Tomatoes were steamed and pureed. The doses were given together with a liquid formulated drink with 25% energy from fat. Our results showed that up to 34 days after taking an oral $^2H_{10}$ lycopene dose (synthetic or from tomato) with a liquid formula drink, the area under the curve of the average serum percent enrichment response of synthetic lycopene reached 33.9 (±1.7) nmol-day/μmol lycopene in the dose, whereas that of lycopene from the tomato dose was 11.8 (±0.3) nmol-day/μmol lycopene in the dose. Our study provides evidence that the absorption of physiological levels of lycopene in intrinsically labeled tomatoes can be studied in humans. From these preliminary investigations, we find that the bioavailability of synthetic lycopene in oil appears to be about three times higher than that of lycopene from steamed and pureed tomatoes.

Keywords: Lycopene; Mass spectrometry; Stable isotope; Intrinsic labeling; Blood response

1. Introduction

Remarkable inverse relationships have been reported between lycopene intake or serum lycopene values and the risk of cancer of the prostate, pancreas, lung and stomach [1]. Since lycopene can be detected in high concentrations in the prostate, testes and adrenal glands, the biological function(s) of lycopene in these organs are of great interest. In the US diet, lycopene is derived primarily from tomatoes and tomato products, and may account for up to 60% of blood carotenoid content in the US population [2]. However, our current knowledge of the bioavailability of lycopene in humans is limited due to our inability to distinguish between newly administered lycopene, or its metabolites, and the body reserves of lycopene and lycopene metabolites.

Labeling foods with stable isotopes such as deuterium (^2H) can provide a safe, reliable and efficient method to study the bioavailability of carotenoids in foods [3,4]. We have developed liquid chromatographic/atmospheric pressure chemical ionization–mass spectrometry methods (LC/

Abbreviations: HPLC, high-performance liquid chromatography; LC/APCI-MS, liquid chromatography/atmospheric pressure chemical ionization-mass spectrometry; THF, tetrahydrofuran.

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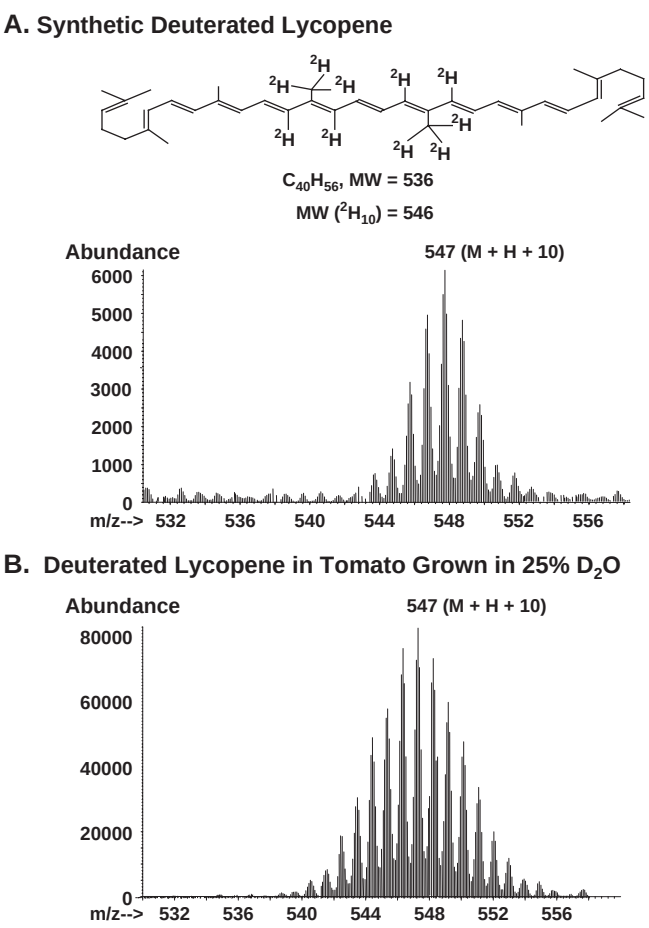

Fig. 1. Deuterated lycopene from (A) synthetic lycopene and (B) from steamed-pureed tomatoes grown hydroponically with 25atom% D_2O and analyzed by LC-APCI-MS.

APCI-MS) to study carotenoids in biological samples [5]. We used synthetic lycopene (Fig. 1A) with a peak deuterium enrichment at M+10 ($^2H_{10}$) (provided by BASF, Ludwigshaven, Germany) to study the absorption of labeled synthetic lycopene in oil dose.

In addition, to better understand the absorption and relative bioavailability of lycopene from tomatoes, we used intrinsically labeled tomatoes (Fig. 1B) as an alternative source of deuterated lycopene in human subjects and an advanced isotope tracer LC/APCI-MS method to analyze the blood samples from those subjects.

2. Methods and materials

2.1. Subjects

We recruited two male (M) and two female (F) subjects from the general public, ages of 51.3±6.7 (46–60) years and BMI of 29.8±6.7 (23.7–37.0) kg/m^2. Informed consent had been obtained from all subjects under the guidelines established by the Institutional Review Board of Tufts University and the New England Medical Center.

Participants were housed in the Metabolic Research Unit of the Boston USDA Human Nutrition Research Center on Aging at Tufts University for a 10-day resident stay, and then were free living from day 10 to day 34 of the study.

2.2. Preparation of the deuterated tomato fruit

Tomato plants (cultivar Microtom) were grown hydroponically at the USDA/ARS Children's Nutrition Research Center in Houston, TX, USA. Plants were maintained in an environmental growth chamber (Conviron Model PGW36; Winnipeg, Manitoba, Canada) using a 16-h, 20 °C/8-h, 15°C day/night regime and 70% relative humidity. A combination of incandescent and fluorescent lamps provided photosynthetically active radiation at an intensity of 500 μmol of photons per square meter per second at the top of the plants. Tomato seeds were germinated on filter paper; seedlings were planted in polyethylene cups placed over the aerated hydroponic solution as previously described [6]. Four plants were grown hydroponically in 4.5 L of nutrient solution containing the following macronutrients in millimolars: KNO_3, 5; $NH_4H_2PO_4$, 0.3; Ca $(NO_3)_2$, 1; $MgSO_4$, 0.5; and the following micronutrients in micromolars: $CaCl_2$, 25; H_3BO_3, 25; $MnSO_4$, 2; $ZnSO_4$, 2; $CuSO_4$, 0.5; H_2MoO_4, 0.5; $NiSO_4$, 0.1. Iron was added in chelated form as Fe(III)EDDHA (*N*,*N*′ -ethylenebis[2-(2-hydroxyphenyl)-glycine]) at 10\ μM. MES buffer (adjusted with KOH) was added at 2 mM to maintain the nutrient solution pH between 5.4 and 5.8. Nutrient solutions were changed twice weekly, until plants reached reproductive maturity (i.e., flowering).

After plants had initiated flowering and indications of fruit formation were evident, the nutrient solution was replaced with a deuterium-enriched nutrient solution containing mineral salts at the concentrations described above. Labeled nutrient solution was made by combining stock salt solutions (in H_2O), deionized water (H_2O) and an appropriate amount of D_2O to achieve a final deuterium enrichment of 25atom%. All deuterium-enriched nutrient solutions were constantly aerated with a water vapor-free air supply to provide oxygen to the plant roots. Additional nutrient solution was added to the growth container, as required, in response to plant utilization of water and minerals. During the enrichment period, the container of plants was placed in a plastic tub fitted with a glass top. This arrangement maintained an enriched water vapor environment (≤25 atom-% D_2O) in the gas atmosphere surrounding the plants.

Plants were grown on the deuterium-enriched nutrient solution until all fruit had matured (i.e., full-color development). Fruits were harvested and shipped overnight on ice to the USDA/ARS HNRC at Tufts University in Boston, MA. Upon arrival, they were sliced and steamed for 10 min. Afterwards, they were pureed in a blender for 3 min, portioned and kept at −70°C in plastic containers until being analyzed or used for the feeding studies.

2.3. Diets

For the 2 weeks before the resident period, subjects were instructed to eat their normal diet, but without vitamin supplements or foods containing large amounts of carotenoids and tomatoes (from lists provided by the dietician). Compliance was checked by food diaries taken over the last 3 days of the 2-week run-in period. The volunteers were randomly assigned to receive lycopene either from gelatin capsules (1 M, 1 F) or steamed-pureed tomato (1 M, 1 F). The gelatin capsules contained 6.0 mg synthetic $^{2}H_{10}$ lycopene in 6 g corn oil. The 80- or 84-g steamed-pureed tomato doses contained 8.9 or 9.5 mg deuterated lycopene, respectively, with $^{2}H_{10}$ as the predominant lycopene isotopomer in the tomatoes.

On day 1, after an overnight fast, the volunteers consumed one gelatin capsule (with 6 g of corn oil) with a formulated liquid breakfast (14 g fat, 18 g protein, 480 kcal), or the steamed-pureed tomatoes with a formulated liquid breakfast (20 g fat, 18 g protein, 530 kcal). The fat in each formulated liquid meal was derived from coconut milk and cream. The percentage of energy from saturated, monosaturated and polysaturated fatty acids was estimated as 20%, 1.4% and 1.6%, respectively. Five hours after breakfast, the volunteers consumed a lunch consisting of a formulated liquid diet (14 g fat, 18 g protein, 480 kcal). In the evening, 10 h after the breakfast, the volunteers received a dinner containing 31 g fat and 35 g protein with a total energy content of 880 kcal (containing 7 μg, or ~0.013 μmol of carotenoids). For the first 9 days of the study, the volunteers consumed a 2-day rotation diet containing 100 μg vitamin A and 25 μg (0.05 μmol) carotenoids per day at the Nutrition Center in Boston, and from day 10 to day 34 they were free living and ate their normal diets.

2.4. Blood sample collections

Blood samples were collected at 0, 3, 5, 7, 9, 11, 13 h of the first day after consuming the deuterated lycopene; 12-h fasting blood samples were subsequently collected on various days while subjects stayed in the Boston Nutrition Center (until day 10), or while free living (until day 34). Blood samples were kept in the dark and at room temperature for half an hour after drawing, and then were centrifuged with Sure-sep II (Organon Teknika, Durham, NC) at 4 °C and 800×*g* for 15 min. Collected serum was stored at −70°C until subsequent processing and analysis.

2.5. Tomato extraction

To determine the lycopene content in tomato doses, samples of steamed-pureed tomato (0.5 g) were vortexed for 30 s with 10 mL methanol and incubated for 90 min at room temperature in a shaking incubator at 120 RPM (Thermolyne). After homogenization (Polytron) at 8000 RPM 5–10 s in an ice bath, samples were centrifuged at 3500 RPM for 10 min, and the methanol layer was transferred to a 50-mL volumetric flask. The residue was re-extracted with a total of 35 mL of tetrahydrofuran (THF) by vortex followed by centrifugation at 3500 RPM for 10 min. The methanol and THF extracts were combined and brought up to 50 mL with THF. One milliliter of the combined extract was evaporated to dryness under N_2, and the residue was re-dissolved in 1 mL ethanol, sonicated and vortexed for 30 s. A 50-μL aliquot of the ethanol extract was injected onto an HPLC.

2.6. Serum extraction

Three milliliters of chloroform/methanol (2:1 in volume) was added to a 100-μL serum sample to dissociate the protein and fat-soluble nutrients, and to extract the fat-soluble nutrients into the chloroform layer. The mixture was vortexed and centrifuged for 10 min at 4°C and at 800×*g*. The chloroform layer was collected. Hexane (2 mL) was added to the aqueous layer to re-extract fat-soluble nutrients and nonpolar straight chain fat-soluble carotenes to increase the extraction efficiency. The hexane layer was combined with the chloroform layer and evaporated under N_2 on an N-EVAP (Organomation Associates, South Berlin, MA). The residue was dissolved in 100 μL of ethanol, and 50 μL was injected onto an HPLC.

2.7. HPLC analysis

Concentrations of carotenoids were measured by an HPLC equipped with a YMC C30 column (Waters, Milford, MA) and a Waters 994 Programmable Photodiode Array Detector with the wavelength set at 450 nm for carotenoids and 340 nm for retinoids [7]. The concentrations of total lycopene together with the percentage isotopic enrichment of lycopene determined by LC/MS were used to calculate the molar enrichments of $^{2}H_{10}$-lycopene in the processed sera and tomato samples.

2.8. LC/APCI-MS analysis

To determine the percent enrichment of labeled lycopene, an APCI-MS method was developed based on a previous method used for determining the enrichment of β-carotene [5]. One to 2 mL serum samples (whole serum) were added to an NH_2 column (3 ml, 500 mg, J.T. Baker, Phillipsburg, NJ) preconditioned with hexane. Hexane was used as the eluent. The lycopene–hexane eluent was evaporated under N_2. The residue was resuspended in 70 μL of methyl *t*-butyl ether/methanol (2:1, v/v) and injected onto an HPLC system equipped with a C30 column (Bischoff Chromatography, Leonberg, Germany). The lycopene fraction from the HPLC separation was collected and dried under N_2, resuspended in 70 μL ethanol, and 60 μL injected into an LC/APCI-MS with a C18 Pecosphere column (Perkin-Elmer, Norwalk, CT) with 100% methanol. The data acquisition parameters were as follows: (1) mass range was set from *m*/*z* 530–560; (2) skimmer 1 was 30.7 V, capillary exit offset was at 71.6 V and trap drive was 47.4; (3) accumulation time was set at 10.0 μs and (4) the number of averages was set at 15 spectra. We used the Bruker Data Analysis and Esquire-LC MS

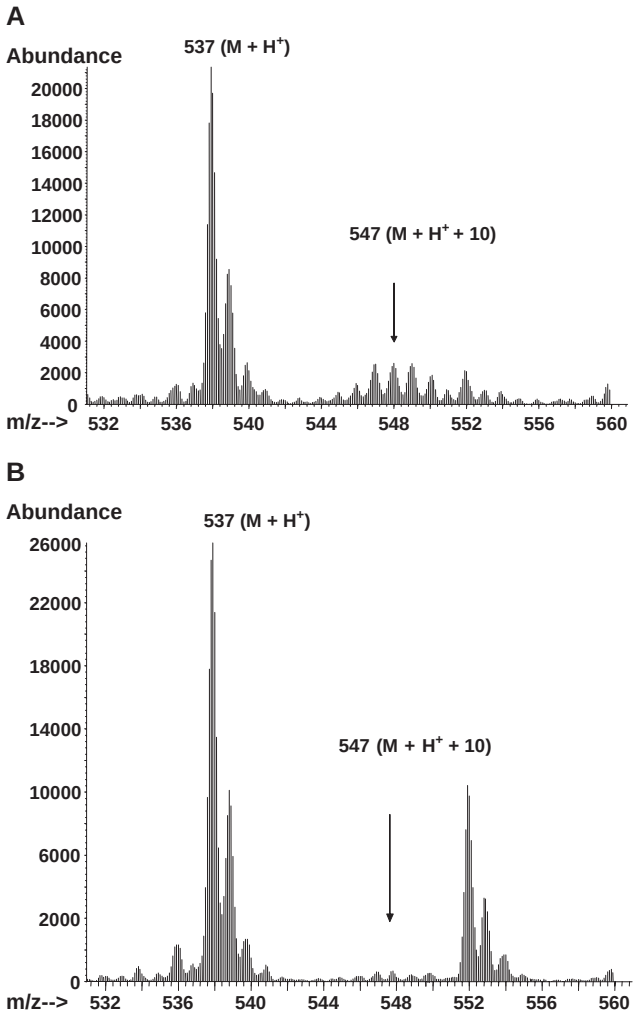

Fig. 2. Deuterated lycopene in serum 11 h after the ingestion of either synthetic lycopene (A) or steamed-pureed tomatoes grown hydroponically with 25atom% D_2O (B) and analyzed by LC-APCI-MS.

Processing version 1.6 m. The LC was an Agilent 1100 (Andover, MA), and the mass spectrometer was a Bruker Esquire LC (Billerica, MA). All experimental procedures were carried out under red light.

Due to the isotopic profile and the detection limit, not all of the enrichment masses were measurable at very early or later time points. In addition, the interference masses (even after the HPLC purification), such as mass *m/z* 552 from the blood sample in Fig. 2A and B, made it unsuitable to measure the blood response for all the masses from *m/z* 540 to 556. Therefore, the actual enrichment of labeled lycopene in human serum was determined by the following steps. First, we used the LC-APCI-MS to measure *m/z* $(M+H)^+$=537 (^{1}H), 538 (^{13}C–^{1}H), 539 (^{13}C–^{13}C–^{1}H), 543 ($^2H_6$), 544 ($^2H_7$), 545 ($^2H_8$), 546 ($^2H_9$) and 547 ($^2H_{10}$), i.e., *m/z* 537–539 for unlabeled lycopene and *m/z* 543–547 for labeled lycopene in the circulation. Secondly, we determined the ratio of labeled peaks at *m/z* 543–547 (partial) to all labeled peaks at *m/z* 542–552 (complete) in the labeled lycopene doses. We found that for synthetic $^2H_{10}$ lycopene, the ratio of isotope masses *m/z* 543–547 (partial) to *m/z* 542–552 (complete) (Fig. 1A) was 0.43±0.01, and that for tomato $^2H_{10}$ lycopene, the ratio of isotope masses *m/z* 543–547 (partial) to *m/z* 540–556 (complete) (Fig. 1B) was 0.36±0.07. Finally, these abundances (partial/complete) were used to calculate the percent enrichment at every time point.

That is, in the case of the synthetic $^2H_{10}$ lycopene dose,

$$\%\ \text{Enrichment} = 100 \times [(m/z\ 543 - 547)/0.43] / [(m/z\ 543 - 547)/0.43 + (m/z\ 537 - 539)]$$

In the case of the biosynthetic tomato $^2H_{10}$ lycopene dose,

$$\%\ \text{Enrichment} = 100 \times [(m/z\ 543 - 547)/0.36] / [(m/z\ 543 - 547)/0.36 + (m/z\ 537 - 539)]$$

The concentration of deuterium-labeled lycopene in nanomoles per liter in the circulation was determined by multiplying the percent deuterium enrichment of serum lycopene by the concentration of total lycopene (all the cis and all-trans forms) determined by HPLC analysis. The enrichment for each time point at which blood samples were collected was plotted against time to calculate the areas under the curve.

2.9. Area under the curve of serum response for each micromole of lycopene in the labeled dose

Whole-body serum responses to the lycopene dose were determined by multiplying the total serum volume (0.0435 L/kg body weight [8]) by the concentration of labeled lycopene in the circulation. Areas under the curve for serum-labeled lycopene response (nmol-day/μmol labeled dose) were calculated by using the curve of total serum responses vs. time via Integral-Curve of Kaleidagraph (Synergy Software, Reading, PA). The response in area under the curve of labeled lycopene to each milligram of lycopene in the labeled dose was determined.

3. Results

3.1. Serum parameters

The volunteers' serum concentration of lycopene at the beginning of the study ranged from 0.088 to 0.243 μmol/L, as presented in Table 1. The serum concentrations of other major carotenoids are presented in Table 1, as well.

Table 1
Concentration of serum lycopene and other major carotenoids of each volunteer at the beginning of the experiment

Volunteer	Total lycopene	β-Carotene (μmol/L)	α-Carotene	Lutein	Cryptoxanthin
No. 1 (F)	0.243	0.099	0.022	0.096	0.048
No. 2 (M)	0.192	0.064	0.019	0.087	0.038
No. 3 (F)	0.176	0.447	0.242	0.141	0.077
No. 4 (M)	0.088	0.080	0.162	0.030	0.131

3.2. Serum response to labeled lycopene doses

After a single oral dose of either 80–84 g of pureed tomato dose (containing 16.3 or 17.4 μmol lycopene) or 11 μmol synthetic $^2H_{10}$ lycopene in 6 g of corn oil, we detected deuterium enrichment of lycopene in each subject up to 34 days. Two representative mass chromatograms of serum samples collected 11 h after the dose are shown in Fig. 2A and B for subject No. 2 and No. 4, respectively. The serum response curve in concentration (nmol/L) vs. time (day) of labeled lycopene from subjects receiving the synthetic $^2H_{10}$ lycopene dose over the 34-day period of the experiment is presented in Fig. 3. The insert shows both the labeled (left axis) and total (right axis) lycopene during the first 4 or 5 days. Both subjects showed a maximum serum response of $^2H_{10}$ lycopene at 11–13 h, with $^2H_{10}$ lycopene measurable in the serum for the entire 34-day course of the experiment. However, after a single oral dose of either 6 mg lycopene in oil or 9 mg of lycopene in the tomato preparation, we did not detect a consistent increase or decrease in serum concentration of lycopene over the first 24 h of the experimental period (see inserts of Figs. 3 and 4).

The serum response curve of labeled lycopene from subjects receiving the tomato $^2H_{10}$ lycopene dose is presented in Fig. 4. The inserts show the labeled (left axis) and total lycopene (right axis) during the first 4 days. These subjects showed a maximum $^2H_{10}$ response at 9–11 h, and again, the $^2H_{10}$ lycopene was measurable in the serum for the entire 34-day period of the experiment. All subjects responded to the lycopene dose, i.e., they were carotenoid absorbers. As seen in the insert, after taking the labeled tomato, the concentrations of lycopene in the circulation showed an increase followed by a decrease as compared to the baseline concentrations.

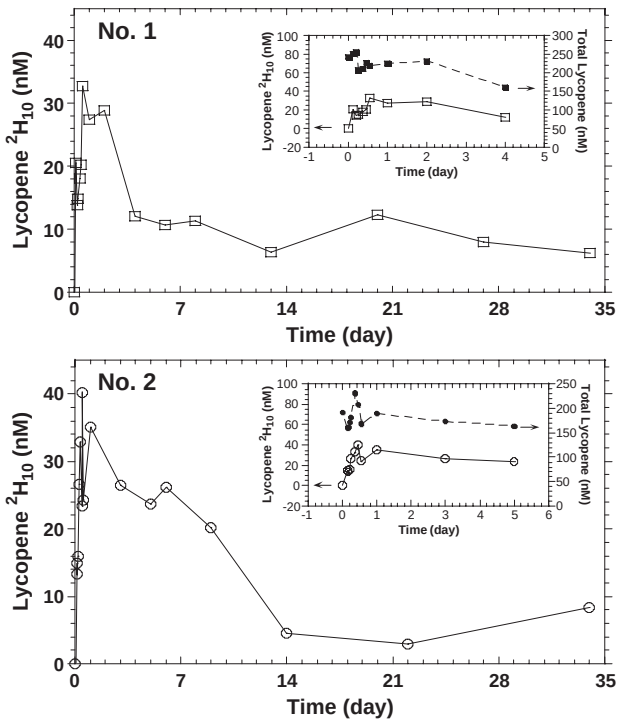

Fig. 3. Subjects' (No. 1, top panel and No. 2, bottom panel) serum responses of labeled lycopene after an 11-μmol (6 mg) dose of synthetic $^2H_{10}$ lycopene. Insert shows both labeled (left y-axis, solid symbol) and total lycopene (right y-axis) in serum measured during the first 4–5 days.

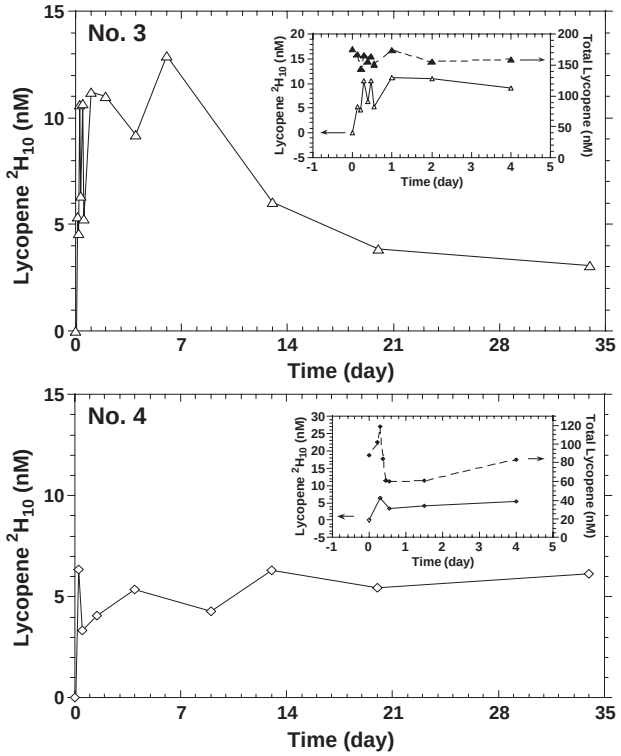

Fig. 4. Subjects' serum response of labeled lycopene after consuming steamed-pureed tomatoes containing 17.4 μmol (9.5 mg, subject No. 3) or 16.4 μmol (8.9 mg, subject No. 4) of $^2H_{10}$ lycopene. Insert shows both labeled (left y-axis, solid symbol) and total lycopene (right y-axis) in serum measured during the first 4 days.

3.3. Area under the curve of labeled lycopene serum response

The total area under the labeled lycopene response curve in serum for each micromole of lycopene ingested is

Table 2
Total area under the labeled lycopene response curve in serum up to 34 days per micromole of ingested lycopene

Volunteer	Lycopene dose	AUC of $^2H_{10}$ lycopene (nmol-day/μmol)	Average response
Synthetic lycopene			
No. 1	11 μmol (6 mg)	32.2	33.9
No. 2	11 μmol (6 mg)	35.5	
Steamed-pureed tomato lycopene			
No. 3	16.3 μmol (9.5 mg, 84 g tomato)	12.1	11.8
No. 4	17.4 μmol (8.9 mg, 80 g tomato)	11.4	

presented in Table 2. Synthetic lycopene in oil was absorbed three times as well as lycopene from tomatoes.

4. Discussion

4.1. Advantage of using labeled lycopene

After 2 weeks on a diet that avoided foods containing large amounts of carotenoids and tomatoes, the serum concentrations of lycopene and other major carotenoids in the study subjects (Table 1) were at the low end of the average human serum concentration of these carotenoids [7,9,10]. After taking a physiological dose of lycopene either by oil capsules or steamed-pureed tomato, the serum concentration of lycopene of these volunteers did not change consistently from 1 to 34 days. This shows that it is difficult to use serum concentration alone to study the blood response of lycopene to an acute dose of lycopene in humans. By labeling the lycopene dose, however, we can measure the enrichment of the labeled lycopene in the circulation up to 34 days after the dose. Our results confirmed that it is possible to use a physiological amount of either synthetic or biosynthetic deuterated lycopene to measure the serum response in human subjects.

4.2. Estimation of bioavailability of lycopene

Based on the highest concentration of labeled lycopene detected in the serum and the estimated serum volume of each subject [8], our calculation showed that about 1.2% of the synthetic lycopene dose appeared in the blood circulation. Although this may not represent the total lycopene absorbed or utilized by the human body, it is reasonable to assume that at least 1.2% of the lycopene dose got into the circulation as intact lycopene. This is in agreement with the study conducted by O'Neill and Thurnham [11] who found ~1.0 mg of lycopene was absorbed from a 38-mg dose contained in capsules (~2.6%).

Following the ingestion of a study dose of 23.6 mg total lycopene in tomato paste, Gärtner et al. [12] reported an increase of ~25 nmol/L lycopene in the chylomicron fraction of serum. Assuming the average body weight of an adult is 70 kg, this increase of lycopene in the chylomicron fraction represented a total of 75 nmol of lycopene in the circulation (70 kg×0.0435 L/kg=3.0 L blood). Thus, only 0.17% of the dose was detected in the circulation after the consumption of tomato paste (lycopene in fresh tomatoes was probably absorbed even more poorly than in tomato paste). In another report [13], three subjects took heated tomato juice containing lycopene (93.8 mg), equivalent to an amount of 2.5 μmol/kg body wt. An increase of 80–350 nmol/L in serum lycopene was observed. This represents an absorption of 0.14–0.57% of the 93.8-mg lycopene dose (again assuming 70 kg body wt. for the adults and 0.0435 L of serum per kg body wt.).

While only limited information is available regarding the absorption of an acute physiological dose of tomato lycopene in humans, multiple doses of 5 to 150 mg lycopene per day for 1 to 8 weeks given to human subjects have been reported [14–18]. For example, a recent report used 25 mg/day of lycopene for 8 weeks as either tomato paste or lactolycopene, a mixture of wheat whey and lycopene [19]. These investigators found that after 2 weeks of supplementation, serum lycopene reached a plateau, resulting in a plasma increase of ~0.5 μmol/L. The increase of lycopene in the blood circulation was 0.8 mg (assuming 70 kg body wt. and 0.0435 L/kg body wt.). Since a total of 350 mg lycopene dose was ingested, this means that 0.8/350=0.2% of the lycopene doses appeared in the circulation. These results are very close to our observation that after an acute dose of steamed (for 10 min) and pureed tomato (~9 mg lycopene), ~0.2–0.3% of the tomato lycopene was detected in the circulation based on the highest serum concentration of labeled lycopene.

We used a ratio of partial to whole mass-ranges to determine the enrichment of labeled lycopene in the circulation. To determine the enrichment of labeled lycopene, the serum extract was first purified with a NH_2 cartridge column and then on an HPLC equipped with a C30 column. The peaks of lycopene collected from the HPLC chromatography was rechromatographed on a C18 column (total lycopene) before APCI ionization. After these chromatographic procedures, an interfering compound of m/z (M+H)=552 from serum was still detected in the MS analysis. This peak may correspond to the platelet-activating factor phospholipid, 1-octadecyl-2-acetyl-*sn*-glycero-3-phosphocholine, whose atomic mass is 551.7 [20]. Using the ratio of partial to whole mass-ranges to determine the enrichment of labeled lycopene in the circulation, this peak's interference can be eliminated.

Our results show that after adjustment for dose differences (11 to 17.4 μmol), the average AUC response was 33.9 nmol-day/μmol synthetic lycopene dispersed in oil and 11.8 nmol-day/μmol lycopene in steamed-pureed tomatoes. Thus, the absorption of synthetic lycopene by human volunteers was almost three times greater than that of lycopene from steamed-pureed tomatoes. It seems that the tomato matrix and the fact that the tomato was not heated in oil during the food preparation made the tomato lycopene less bioavailable. In addition, this difference could also be partially due to the difference in the amounts of doses used, 6 mg lycopene in the synthetic dose vs. 9 mg lycopene in the tomato dose, since the absorption efficiency is probably inversely related to the dose level (as is the case with β-C) [21].

An earlier report showed that tomato processing affected serum lycopene levels; that is, the absorption of lycopene from cooked tomato paste was 3.8 times greater than from a fresh raw tomato [12]. In addition, boiling tomato juice for 1 h in the presence of 1% corn oil significantly increased the bioavailability of lycopene compared with unheated tomato juice after a single dose of 2.5 μmol/kg body wt. [13]. Thus, many processing factors can affect lycopene availability. Although our preparation involved steaming

the tomatoes for 10 min and giving the dose with a liquid diet containing fat, the tomatoes were not heated with oil, and this might not have been optimal for absorption. Nevertheless, our method had adequate sensitivity to monitor labeled lycopene in serum for up to 34 days after feeding. By using this food-based, deuterated lycopene method, future experiments to compare the blood responses of labeled lycopene following different preparation conditions (e.g., length of heating period, amount of oil) would provide further useful information regarding the bioavailability of lycopene from tomatoes.

Our study investigated the blood response to a single acute dose of lycopene from different sources. If the doses are given repeatedly (chronic doses), the blood concentration of lycopene will eventually reach a plateau as described by Richelle et al. [19]. A recent publication [22] did not observe a difference in the increase of serum lycopene from baseline between synthetic and tomato-based lycopene. In their study, the lycopene doses from LycoVit or Lyc-O-Mato were in the form of beadlets or beads, respectively, with the lycopene derived synthetically (LycoVit) or from a natural source (Lyc-O-Mato; extracted from tomato). Although the Lyc-O-Mato dose also contained tomato-oleoresin, there were no tomato solids in the dose. Thus, there was no involvement of food matrix components in the blood response to either of these doses. In addition, we suspect that if doses of 15 mg/day were given for 28 days, the serum concentration of lycopene would plateau from either LycoVit or Lyc-O-Mato supplements.

4.3. Summary

In summary, our method can evaluate the bioavailability of dietary levels (9 mg) of tomato lycopene in humans. Our results show that deuterated lycopene can be detected in serum for up to 34 days after a single physiological dose of either a synthetic deuterated lycopene supplement or an intrinsically labeled tomato. The synthetic lycopene is almost three times more bioavailable than biosynthetic tomato lycopene from steamed-pureed tomato. Further studies are needed to investigate additional subjects and to study the factors affecting tomato lycopene bioavailability.

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