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ORIGINAL CONTRIBUTION

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Comparative multiple dose plasma kinetics of lycopene administered in tomato juice, tomato soup or lycopene tablets

Summary *Background* Lycopene is mainly provided in tomato and tomato products in Western diet. Among other factors the systemic availability of lycopene from natural sources is dependent on release from the cell matrix as achieved by food processing. *Aims of the study* The purpose of this study was to compare plasma concentration responses of total lycopene and its major isomers to dosing of the carotenoid as tomato juice, tomato soup or tablets containing synthetic lycopene. *Methods* Intake of lycopene rich food products was restricted throughout this randomized, parallel group study, including 6 volunteers per group. Following a 14 day lycopene depletion phase subjects ingested 20 mg of lycopene daily for 8 days as tomato juice, soup prepared from tomato paste or lycopene tablets. Lycopene plasma concentrations were monitored throughout the depletion and dosing phases and for 22 days post-dosing and kinetics were evaluated using both empirical and compartmental modelling. *Results* Irrespective of the lycopene treatment all-E lycopene was the predominant lycopene isomer, whereas 5-Z lycopene was the most abundant Z isomer. Plasma concentration response of total and all-E lycopene to dosing of the carotenoid in tablets and tomato soup was comparable but exceeded that of intake in tomato juice. No differences were noted in dose normalized 5-Z lycopene concentrations between groups. The estimates of efficient half-life were approximately 5 and 9 days for all-E and 5-Z lycopene, respectively. *Conclusions* The systemic availability of synthetic lycopene from a tablet formulation is comparable to that observed from processed tomatoes (soup from tomato paste) and superior to that from tomato juice. No differences were observed in disposition kinetics of natural and synthetic lycopene. The synthetic lycopene tablet formulation used in this investigation may be of value for future clinical investigations.

Key words lycopene – isomers – processing – availability – half-life

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Introduction

The health benefits of diets rich in fruits and vegetables have long been recognized. Lycopene is one of the most abundant carotenoids in human plasma and approximately 80 % of dietary lycopene is provided by tomatoes and tomato products [1, 2]. In his review of 35 studies, Giovannucci et al. [3] reported a statistically significant inverse relationship between intake of tomatoes or blood lycopene concentration and the risk of developing cancer. Evidence was strongest for a beneficial effect with regard to prevention of prostate cancer.

The systemic availability of lycopene from tomatoes is improved by processing [4–9]. Porrini et al. [5] found higher total lycopene plasma concentrations after ingestion of a tomato purée as opposed to raw tomatoes. In another study, tomato paste produced a 2.5-fold increase

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in peak concentrations of total and all-E lycopene and a 3.8-fold increase in AUC when compared with raw tomatoes [6]. Heating and homogenization of tomatoes appears to disrupt the cell wall structure of the tomatoes, thereby enhancing the availability of lycopene [4–9]. Similarly, the intestinal absorption of other carotenoids such as β-carotene is also improved by processing [10].

Several investigators have studied food-derived ("natural") lycopene supplements and found their systemic availability comparable to more highly processed tomato products [9, 11, 12]. The bioavailability of a water-dispersible tablet formulation of synthetic lycopene has not been investigated yet, although such products are sold on the market and widely advertised for risk reduction of cancer of the prostate. Furthermore, there is inconsistent information on plasma half-life of lycopene, ranging from 2 to 3 days to as long as 33 days [7, 13–15]. Half-life estimates of lycopene isomers have not yet been published. The purpose of this investigation was to compare the plasma kinetics, including the relative systemic availability, of lycopene and its major isomers (all-E, 5-Z) administered in different forms to healthy volunteers in a multiple dosing design. Lycopene was ingested in the form of unprocessed tomatoes in juice, as a processed tomato product (prepared as soup containing tomato paste) and as a tablet containing synthetic lycopene. The trial was carried out in male volunteers in view of the potential benefits of lycopene in preventing prostate cancer. Intake of dietary lycopene was monitored throughout the study.

Subjects and methods

Subjects

After approval of the study protocol by the local ethics committee, 18 healthy male subjects aged between 18 and 45 years gave their written informed consent to participate in the study. At screening, a physical examination including ECG, clinical chemistry, hematology and serology testing for hepatitis A and B as well as HIV-1/–2, revealed no abnormal findings. Three smokers participated in the study, which were allocated to treatment groups B (1 subject) and C (2 subjects) (see below for definition of treatment groups). Cigarette smoking was restricted to 10 cigarettes per day. None of the subjects was enrolled in any study at least 3 months prior to study beginning and none of the subjects took any supplements before the study. Demographic characteristics of the subjects are summarized in Table 1.

Table 1 Mean (SD) characteristics of the groups

	Group		
	Juice	Soup	Tablet
Subjects			
n	6	6	6
Age (years)	29.7 (5.8)	29.0 (4.6)	29.3 (7.3)
Height (cm)	179.7 (6.9)	180.7 (8.7)	180.2 (10.5)
Weight (kg)	77.3 (8.2)	80.0 (10.0)	80.4 (7.8)
BMI (kg/m^2)	23.9 (1.7)	24.5 (2.6)	24.8 (1.6)
Daily Fat Intake (g)	105.2 (29.6)	106.4 (16.3)	113.8 (24.6)
Baseline Conc. (μmol/L) (day 0)			
Total Lycopene	0.37 (0.19)	0.34 (0.12)	0.40 (0.09)
all-E	0.15 (0.07)	0.13 (0.06)	0.15 (0.03)
5-Z	0.12 (0.06)	0.10 (0.04)	0.13 (0.03)
Dose			
Total Lycopene (mg/d)	18.98	23.28	22.8
all-E (%)	92	86	73
5-Z (%)	4	6	19

Study Design

This was an open label, randomized, parallel group study with three treatment groups. The study consisted of three phases beginning with a 14 day lycopene depletion phase during which subjects were asked to avoid foods containing lycopene such as tomatoes, tomato sauce, tomato paste, ketchup, watermelon, guava and rose hip puree. Subjects continued to maintain this diet with minimal lycopene content for the duration of the whole study. Dietary intake of lycopene as well as other foods was documented in a nutritional diary throughout the study.

Following the lycopene depletion phase, subjects were randomly assigned to one of the three treatment groups. Each group (n = 6 subjects per group) ingested approximately 20 mg of lycopene daily for 8 consecutive days according to one of the following treatments:

Treatment A: 4 lycopene tablets (5 mg, lycopene 5 % TG, Roche Vitamins Ltd.) with 240 mL minestrone soup with dinner. The presence of lycopene in the minestrone soup was avoided by careful selection of the soup ingredients.

Treatment B: Tomato soup (240 mL) prepared by adding tomato paste (Thomy, Switzerland) to minestrone soup with dinner.

Treatment C: Tomato juice (170 mL) (Del Monte, Migros, Switzerland) with dinner.

A defined amount of tomato paste was added to the freshly prepared minestrone soup to attain the intended daily lycopene dose of approximately 20 mg. After preparation of this tomato soup, 10 samples of the soup were drawn and analyzed for their lycopene content. Aliquots of 240 mL of minestrone with/without tomato paste were kept frozen until the day of consumption. The

mean value of the final lycopene concentration per mL soup x 240 mL was the basis for the daily dose, as given in Table 1. The actual lycopene doses from tablets and juice were also defined analytically. The fat content per 240 mL serving of soup was 18.3 g for the minestrone and 18.2 g for the tomato soup.

The lycopene dosing period was followed by a 22 day post-dosing phase.

Blood samples were obtained in the morning after an overnight fast 14, 11, 7, 3 and 0 days prior to lycopene dosing as well as 2, 5, 7, 8, 9, 10, 12, 14, 17, 21, 25 and 29 days after the beginning of the lycopene dosing phase. Blood (7.5 mL) was collected into pre-cooled monovettes containing EDTA, centrifuged at 4 °C and 2,000 g. The plasma was separated under appropriate light shielding and stored for less than 3 months at –35 °C until analysis.

Analytical methods

Lycopene in tablets, food and plasma was measured using previously published procedures [16, 17]. Tomato juice and soup were homogenized and mixed with acetone and magnesium sulfate using a dispersion instrument [16]. An aqueous suspension was prepared from the tablets followed by extraction into dichloromethane/ethanol [17]. Extracts from food and tablets were purified by aluminum oxide in an open column. The eluate was evaporated, the residue dissolved in n-hexane/acetone and analyzed by normal phase HPLC [17]. Plasma samples were treated with ethanol to precipitate plasma proteins followed by extraction with n-hexane. All-E and 5-Z lycopene were quantified at a wavelength of 470 nm. The lower limit of quantification was 20 μg/L. The method is specific and selective for all-E lycopene and related cis-isomers versus other carotenoids commonly present in plasma. The average inter assay precision (CV) was < 5 %. Total lycopene represents the sum of all-E, 5-Z, 9-Z, 13-Z lycopene and three additional non-identified Z-isomers.

Cholesterol and triacylglycerol concentrations in plasma were determined using enzymatic methods [18, 19].

Data analysis

Lycopene isomers contributing to more than 10 % of total plasma lycopene concentration on the average were evaluated. Accordingly, plasma concentrations of total lycopene, all-E lycopene and 5-Z lycopene were plotted versus time for the entire study period (Day –14 to 29). To allow for valid comparisons between groups during the dosing phase, concentrations were dose-normalized to compensate for the small dose deviations between groups and to account for the relative abundance of the 5-Z isomer in the synthetic tablet (Table 1). A standard dose of 20 mg total lycopene, 16.73 mg all-E lycopene and 1.93 mg 5-Z lycopene was used for dose normalization of concentrations. The increase above pre-dose plasma concentrations associated with lycopene ingestion in each of the three treatment groups was assessed by subtracting the baseline concentration at the end of the lycopene depletion phase (Day 0) from the concentration measured during the lycopene dosing phase. The dose-normalized maximal increase in lycopene concentration (ΔC_{max}) was calculated for each treatment. In addition, the incremental increase in plasma concentration was plotted versus time and the trapezoidal rule used to calculate the area under the incremental increase in concentration-time curve (AUΔC) from Day 0 until Day 9.

For kinetic analysis, lycopene plasma concentrations were neither dose-normalized nor baseline corrected. Time-concentration profiles were fitted to non-compartmental and to compartmental models using nonlinear least squares regression analysis (SAAM II, version 1.2, SAAM Institute, Seattle, WA). Measurement errors were assumed to be independent and normally distributed with a mean of 0 and a fractional SD of 0.05. Weights were chosen equal to the inverse of the variance of the measurement error. The standard error of the parameter estimates was determined from the covariance matrix of the least squares fit and expressed as the coefficient of variation (CV).

Assessment of half-life of total, all-E and 5-Z lycopene was based on empirical modeling of plasma concentrations in the post-dosing phase (Day 9–28) of the study. To select the best empirical model, in addition to parameter precision, weighted residuals inspection and parsimony criteria were used, in particular by formally testing for non-randomness of errors as judged by run-tests, the F-ratio test (for nested models), the Akaike information criterion and the Schwarz criterion [20, 21]. The plasma concentration time profile in the post dosing phase was evaluated by the following equation: $C(t) = A^*e^{-\alpha t} + B$. Competing models were the mono-exponential and the bi-exponential model in addition to elimination according to a Michaelis-Menten mechanism.

Half-life estimates for all-E and 5-Z lycopene were further evaluated by compartmental modeling. The approach assumed that absorption and clearance remain constant from dose to dose and that the rate constant of lycopene absorption exceeds that of elimination at least by an order of magnitude. Under these assumptions the lycopene concentration time course is determined by plasma half-life over the entire duration of the trial from initial lycopene depletion, to the dosing and the post-dosing phases. As a consequence, modeling the complete concentration time course from the beginning (Day –14)

to the end of the experiment broadened the data basis for estimating plasma half-life. For the compartmental analysis, data sets were analyzed using a one-compartment model with first order absorption and disposition and corresponding rate constants k_a and k_e.

The following differential equation describes the model:

$$\frac{dC}{dt} = k_a * \sum_i q_i - k_e * (C - P_2)$$

where C represents lycopene concentrations at time t, with $q_i = 0$ for $t\text{-}T_i < 0$ and $q_i = P_1 * e^{-k_a * (t - T_i)}$ otherwise. Oral dosing occurred at time points T_i with $i = 1..8$.

The pharmacokinetic parameters estimated were the rate constant k_e, the apparent daily lycopene input into plasma due to dosing (P_1), the lycopene plasma contribution remaining unaffected due to lycopene dosing and depletion (P_2), and the initial plasma concentration at day –14 (P_3). The parameter P_3 was closely constrained to the actually measured concentration for Day –14. The model assumed rapid intestinal absorption and accordingly the absorption rate k_a constant was set to 50 x k_e ($days^{-1}$). P_1 was parameterized in terms of the absorbed all-E lycopene dose per distribution volume. P_2 is equivalent to the constant term B of the empirical model.

For one subject of the tablet group the fitting procedures for the total and lycopene isomers yielded parameters B and P_2, respectively, with 95 % confidence intervals including 0, and by inference, these parameters were not established. The results for this subject were not considered for further evaluation.

Statistical analysis

Descriptive statistics (mean, standard deviation or standard errors) were calculated for all measured parameters. Differences between the three treatments in the maximal incremental increase in plasma concentration (ΔC_{max}) and AUΔC response were assessed using one-way ANOVA with post-hoc analysis by the Tukey test. Treatment effects (treatment A including lycopene tablets versus treatment B including tomato soup) on parameters derived from kinetic modeling were analyzed by using Student's t-test. Matched samples, such as kinetic parameters resulting from the two modeling approaches (empirical versus compartmental modeling) were compared using Student's paired t-test.

All statistical analyses were conducted using S-PLUS 6.0 (Insightful Corp.). Statistical tests for model selection, comparison of treatment groups, and kinetic parameters were performed at the $\alpha = 5\,\%$ level.

Results

Table 1 lists the demographic characteristics of each group as well as the actual lycopene dose and the percentage of all-E and 5-Z isomers associated with the different products. Lycopene concentrations declined by approximately 50 % during the lycopene depletion phase (Fig. 1). Inspection of the nutritional records indicated an average dietary lycopene intake of less than 0.2 mg/day for all groups, an amount equivalent to less than 1 % of the treatment dose of approximately 20 mg/day. No statistically significant differences were observed between treatment groups in baseline concentrations of lycopene at the start of the dosing phase. Plasma concentrations exhibited considerable intersubject variability (CV = 25–40 %). Normalization for cholesterol (but not triacylglycerol) concentration reduced the coefficient of variation by approximately 25 % (data not shown). The changes in the dose-normalized plasma concentration-time profile of total, all-E and 5-Z lycopene during the dosing phase of the study are presented in Figs. 2–4.

Table 2 lists the average maximal increases in dose-normalized plasma concentrations from baseline as well as the AUΔC response to dosing for each of the treatments. Plasma concentrations of total and all-E lycopene were more than doubled from baseline by administration of tablets containing synthetic lycopene. The maximal incremental increase in concentration and incremental area (AUΔC) response of total and all-E lycopene with tablets were both significantly greater than that produced by tomato juice (95 % confidence interval (CI) for the difference of means of the Juice-Tablet groups were –2.48 to –0.77 μmol*d/L and –0.48 to –0.11 μmol/L for AUΔC and ΔC_{max} of total lycopene and –1.46 to –0.41

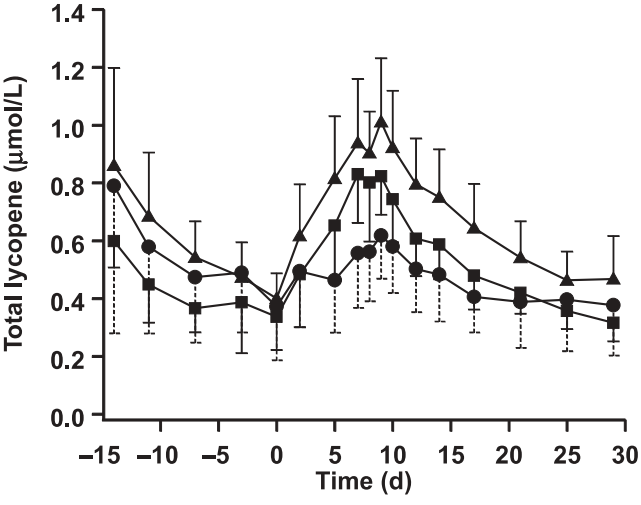

Fig. 1 Plasma concentrations mean (SD) of total lycopene in the three treatment groups over the entire study period. (●) Tomato juice group, (▲) Tomato soup group, (■) Lycopene tablet group

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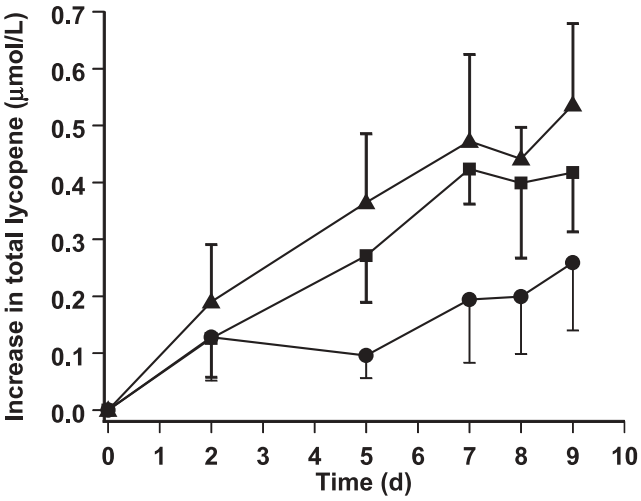

Fig. 2 Mean (SD) dose-normalized incremental increase in plasma concentration of total lycopene during the lycopene dosing phase of the study. (●) Tomato juice group, (▲) Tomato soup group, (■) Lycopene tablet group

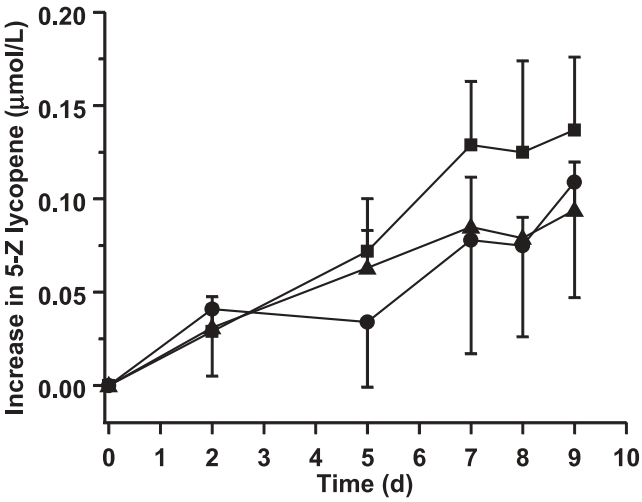

Fig. 4 Mean (SD) dose-normalized incremental increase in plasma concentration of 5-Z lycopene during the lycopene dosing phase of the study. (●) Tomato juice group, (▲) Tomato soup group, (■) Lycopene tablet group

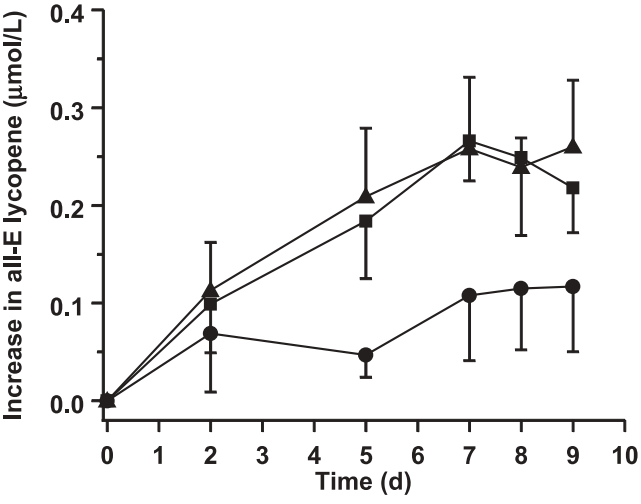

Fig. 3 Mean (SD) dose-normalized incremental increase in plasma concentration of all-E lycopene during the lycopene dosing phase of the study. (●) Tomato juice group, (▲) Tomato soup group, (■) Lycopene tablet group

µmol*d/L and –0.25 to –0.06 µmol/L for AUΔC and ΔC_{max} of all-E lycopene). Lycopene intake as tomato soup also resulted in a significantly higher AUΔC and ΔC_{max} in total lycopene and all-E lycopene compared to tomato juice (95 % CI for the mean differences in AUΔC and ΔC_{max} were –1.91 to –0.20 µmol*d/L and –0.40 to –0.03 µmol/L for total lycopene, those for all-E lycopene were –1.36 to –0.31 µmol*d/L and –0.254 to –0.06 µmol/L). No significant differences in AUΔC and ΔC_{max} were observed between tomato soup and tablets in total or all-E lycopene concentration (95 % CI for Soup-Tablet mean differences in AUΔC and ΔC_{max} were –1.42 to 0.29 µmol*d/L and –0.26 to 0.11 µmol/L for total lycopene and –0.62 to 0.43 µmol*d/L and –0.10 to 0.10 µmol/L for all-E lycopene, respectively). The dose-normalized increase in 5-Z lycopene concentration following administration of the tablets and soup was much smaller than the corresponding increases in all-E and total lycopene concentrations. However, the dose-normalized increase

Table 2 Mean (SD) dose-normalized increase in concentration (ΔC_{max}) and area (AUΔC) of lycopene following ingestion of tomato juice, tomato soup or lycopene tablets

		Treatment Group		
Compound	Parameter	Juice	Soup	Tablet
Total Lycopene	ΔC_{max} (µmol/L)	0.26 (0.12)	0.48 (0.08)[1]	0.56 (0.16)[1]
	AUΔC (µmol·d/L)	1.81 (0.52)	2.24 (0.40)[1]	2.80 (0.74)[1]
all-E Lycopene	ΔC_{max} (µmol/L)	0.13 (0.07)	0.29 (0.04)[1]	0.29 (0.08)[1]
	AUΔC (µmol·d/L)	0.63 (0.34	1.47 (0.32)[1]	1.56 (0.38)[1]
5-Z Lycopene	ΔC_{max} (µmol/L)	0.11 (0.06)	0.15 (0.04)	0.10 (0.03)
	AUΔC (µmol·d/L)	0.44 (0.30)	0.64 (0.17)	0.49 (0.12)

[1] Significantly different from Juice ($p < 0.05$) by Tukey's method

for 5-Z lycopene was similar for all three groups (95 % CI for Juice-Tablet group mean differences in AUΔC and ΔC_{max} were –0.36 to 0.27 μmol*d/L and –0.05 to 0.08 μmol/L, respectively, the corresponding differences for Juice-Soup groups were –0.50 to 0.12 μmol*d/L and –0.10 to 0.031 μmol/L and the intervals for the mean differences between Soup-Tablet groups were –0.164 to 0.46 μmol*d/L and –0.0145 to 0.121 μmol/L).

Decay of plasma concentrations in the post-dosing phase were best fit according to a mono-exponential model with a constant in the form of $C(t) = A^*e^{-\alpha t} + B$. The precision of the estimated parameters for individual subjects was acceptable, as the CVs (%) were in general < 7 % for A and < 18 % for α and B. Averaged half-lives (0.693/α) of total and all-E lycopene ranged between 5 to 6.4 days in the soup and tablet groups respectively (Table 3). The half-life of 5-Z lycopene in subjects of the tablet group was significantly longer at 7.4 days in comparison to that of all-E lycopene, as revealed by a paired t-test (95 % CI for the difference of means –3.05 to –1.05 days). The half-life of total and all-E lycopene could not be accurately estimated after dosing with tomato juice, because the increase in plasma concentrations above baseline was too small. For the same reason, the half-life of 5-Z lycopene could be assessed only after tablet administration. No statistically significant differences were observed between corresponding half-lives resulting from intake of natural lycopene in the tomato soup and synthetic lycopene in tablets (Table 3) for total or all-lycopene, respectively. The 95 % CI for the difference of means (tomato soup group – tablet group) was –1.36 to 2.80 days for the half-life of total lycopene; the 95 % CI for the difference of corresponding means for the half-life of the all-E isomer was –1.69 to 0.93 days. Similarly, for corresponding lycopene isomers, ingestion of lycopene in tablets or tomato soup yielded comparable parameter estimates A (the dosing-dependent pre-exponential coefficient) and B (dosing-independent constant), respectively. For parameters derived from total lycopene data the 95 % CI of the mean differences were –0.32 to 0.10 and –0.27 to 0.01 μmol/L for A and B, respectively. For all-E lycopene related parameters the 95 % CI of the differences were –0.06 to 0.13 and –0.10 to 0.02 μmol/L for parameters A and B. The closeness of corresponding parameter values A and B is again consistent with a similar bioavailability of the two lycopene treatments. Because kinetic parameters of plasma disappearance of natural and synthetic lycopene were statistically indistinguishable for either total or all-E lycopene, the data basis could be extended to encompass the initial depletion phase. In turn, based on a compartmental modeling approach the total concentration-time profile, including depletion-, dosing-, and post-dosing phases, was fitted for all-E and 5-Z lycopene (Fig. 5). The precision of all estimated parameters for each subject was acceptable, as the CVs (%) were in general < 10 % for P_1 and P_3, and < 30 % for P_2 and k_e. The compartmental modeling approach confirmed the half-life determined for all-E lycopene of approximately 5 days based on experimental modeling (Table 4) (paired t-test, 95 % CI for mean differences in the tomato soup group was –1.96 to 2.12 days, 95 % CI for those of the tablet group –1.93 to 2.55 days). For the all-E isomer the parameter P_1, which reflects the systemic availability of dosed lycopene, was not significantly different for tomato-soup and tablet groups (95 % CI for the mean difference of the P1 parameters was –0.21 to 0.74 μmol/L). Furthermore, the constant B (Table 3) and the parameter P_2 (Table 4), both expressing the invariant portion of plasma lycopene concentration, were found to be comparable for corresponding groups (paired t-test, 95 % CI for parameters difference was –0.02 to 0.03 μmol/L). For subjects of the tablet group again the half-life of 5-Z lycopene was longer than that of the all-E isomer (95 % CI of the difference between isomers was –5.79 to –1.79 days); however, half-life estimates using the compartmental modeling approach were significantly longer than those obtained by empirical modeling of the post-dosing phase as assessed by paired t-test (95 % CI of the mean difference was 0.82 to 3.28). Consistent with the lower 5-Z lycopene dose administered with the tomato soup, the plasma-concentration re-

Table 3 Kinetic parameters of plasma lycopene disappearance

Lycopene	Group	N	A[a] (μmol/L)	B (μmol/L)	α (d^{-1})	$t_{1/2}$ (d)
Total	Soup	6	0.639 (0.114)	0.252 (0.048)	0.117 (0.038)	6.361 (1.696)
	Tablet	5	0.750 (0.191)	0.385 (0.135)	0.128 (0.030)	5.638 (1.261)
all-E	Soup	6	0.364 (0.064)	0.089 (0.016)	0.144 (0.030)	4.991 (1.010)
	Tablet	5	0.332 (0.076)	0.131 (0.062)	0.132 (0.023)	5.372 (0.892)
5-Z	Tablet	5	0.277 (0.077)[1]	0.096 (0.038)	0.094 (0.011)[1]	7.421 (0.838)[1]

Lycopene concentration-time profiles (Days 9 to 26) were fitted according to $C(t) = A^*e^{-\alpha t} + B$. Half-life ($t_{1/2}$) was estimated as 0.693/α.

Parameters are means ± 1 SE, no significant differences between Soup and Tablet group comparing corresponding parameters of either total or all-E lycopene, as based on two-sample t-tests

[1] Significantly different from rate constant α and half-life $t_{1/2}$ of all-E lycopene, as based on paired t-tests.

[a] Parameter evaluated at end of dosing at Day 8

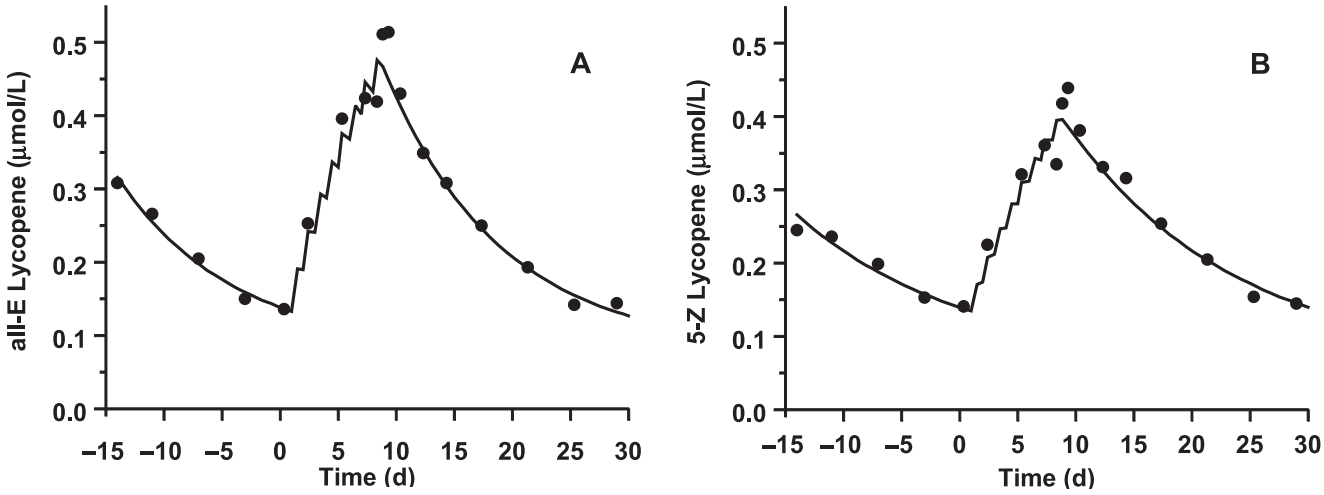

Fig. 5 Measured all-E lycopene (**A**) and 5-Z lycopene concentrations (**B**) and best fit lines based on the compartmental modeling approach as explained in the methods. The plot shows representative time-concentration profiles for a subject of the lycopene tablet group

Table 4 Kinetic parameters of compartmental analysis for all-E and 5-Z lycopene

Lycopene	Group	N	P_1 (nmol/L)	P_2 (nmol/L)	P_3 (nmol/L)	k_e (d^{-1})	$t_{1/2}$ (d)
all-E	Soup	6	71.4 (19.6)	90.0 (43.0)	265 (95)	0.155 (0.065)	5.07 (1.81)
	Tablet	5	57.9 (13.1)	121.3 (64.9)	372 (16)	0.136 (0.052)	5.68 (1.90)
5-Z	Tablet	5	38.6 (9.3)[1]	71.8 (23.7)	224 (67)	0.073 (0.003)[1]	9.47 (0.39)[1]

Compartmental modeling of all-E and 5-Z lycopene plasma concentrations time profiles using non-linear regression as described in Subjects and Methods. Data are means ± 1 SE of parameters assessed from individual fits for each subject, no significant differences between Soup and Tablet group for all parameters estimated for all-E lycopene as based on two-sample t-tests. [1] Significantly different from rate constant k_e and half-life $t_{1/2}$ of all-E lycopene, as based on paired t-tests; P_1 absorbed all-E lycopene dose per distribution volume and per day, during dosing phase; P_2 Basal plasma concentration unaffected by depletion and dosing; P_3 Initial plasma all-E lycopene concentration at day –14; k_e Disposition rate constant; the absorption rate constant was set to 50 x k_e (d^{-1}), Half-life ($t_{1/2}$) was estimated as 0.693/k_e

sponse was small and did not allow for modelling of the 5-Z lycopene profiles.

Discussion

Lycopene availability from natural sources is influenced by a number of factors including the degree of food processing and the lipid content co-ingested with a meal [4–9]. Processing not only disrupts the plant matrix releasing lycopene for absorption but also results in partial conversion of all-E lycopene, the most stable natural configuration, to Z isomers that are reported to be more highly bioavailable [4, 7, 22]. For the products studied in this investigation, the mean percentage of all-E lycopene was decreasingly lower from 92 % in tomato juice to 86 % in soup made from tomato paste to 73 % in tablets containing synthetic lycopene (Table 1). The absorption of lycopene is increased when tomato products are ingested with oil [4, 7]. In order to prevent this from confounding the assessment of lycopene availability in this study, the tablet was taken with a bowl of minestrone soup containing the same amount of fat as the tomato soup. In addition, subjects assigned to drink tomato juice ate a dinner with a similar fat content as the other groups. Dietary records maintained by the subjects confirmed that the daily fat intake was similar for all groups throughout the study (Table 1).

The results of this investigation indicate that the absorption of synthetic lycopene from a tablet formulation is comparable to that of a processed tomato product (soup prepared from commercial tomato paste) and superior to that of tomato juice. Administration of tablets for 8 days more than doubled the average total lycopene concentration above the average baseline concentration prior to dosing (Day 0) (Table 2). In contrast, the ΔC_{max} and incremental area (AUΔC) increase in total and all-E concentration with tomato juice was about half that observed following administration of tablets. No statistically significant differences were noted between tablets and soup in the increase in lycopene concentration. As with the tablets, the increase in total and all-E lycopene

concentration with soup was approximately twice that associated with tomato juice (Table 2, Figs. 2–3). This confirms previous reports [5–9] that processing and heating of tomatoes improves lycopene bioavailability.

Dose normalized concentrations of 5-Z lycopene increased by a similar extent in all groups (Table 2, Fig. 4), i. e. after adjustment for the higher content of the 5-Z isomer in the soup and tablet compared to juice. This is consistent with the findings of Porrini et al. [5], who reported that the AUC of total and all-E lycopene was significantly higher following administration of a tomato purée compared to raw tomatoes, while both increased concentrations of 5-Z isomers to a similar extent. Plasma concentrations of Z isomers account for about 50 % of total lycopene concentrations under equilibrium conditions [4]. Following lycopene dosing, the contribution of the 5-Z isomer to total lycopene plasma concentrations exceeded that of the administered lycopene products. Therefore, the increase in plasma concentrations of 5-Z lycopene following administration of products containing lycopene is not solely related to the content of dosed 5-Z lycopene. The elevated content of the 5-Z isomer in plasma may be accounted for by a longer residence time, which is in line with the observed longer half-life for 5-Z lycopene as compared to that of all-E lycopene. Alternative explanations are 5-Z lycopene isomerization of systemically available all-E lycopene, or preferential intestinal absorption.

Plasma concentrations of lycopene in the post-dosing phase appeared to level off after Day 21 rather than continuing to decline (Fig. 1). This applied in particular to the tomato juice group, for which lycopene concentrations were maintained over the four last sampling points. The mono-exponential plus constant term model was therefore compatible for modelling the post dosing-phase (Table 3). Several explanations might account for the preference of the mono-exponential plus constant term model, which does not represent the general integrated form of a corresponding compartmental model: i) in spite of dietary restrictions lycopene ingestion was not completely suppressed, ii) maintenance by a mechanism such as enterohepatic re-circulation fed by lycopene stores in the liver, and iii) the model could point to a bi-exponential model with a vanishing exponential term. Thus, a mono-exponential plus constant term model may be interpreted as the integrated form of a two-compartment model (two exponential terms) for the extreme case when the second rate constant approaches zero, i. e. the terminal half-life becomes very long and may not be evaluated from data obtained within the selected time window. Such interpretation implies that a very slow plasma disappearance will become indistinguishable from a plateau, in particular when the quality of the concentration data is corrupted by measurement errors. A half-life of 26 days for total lycopene has indeed been reported by Burri et al. [13].

Because lycopene administration in tomato juice elicited only a small increase in plasma levels, the corresponding concentration-time data were not subjected to kinetic analysis. For the remaining two groups, estimates of plasma half-lives for total and all-E were about 5 days (Table 3) and were comparable for natural and synthetic lycopene. It should be noted that we studied only 6 subjects in each group, small differences cannot be detected with this sample size. However, the half-life of all-E lycopene was confirmed by compartmental modelling of the time-concentration profiles from initial lycopene depletion in the pre-dosing phase to the end of the trial (Table 4). Because the time concentration profiles describe both plasma disappearance and accumulation of all-E lycopene, the emerging half-life represents an effective half-life [23]. Based on the effective half-life and under the assumption that systemic availability and clearance are constants from dose to dose, attainment of plasma steady state may be expected after 20 to 25 days of daily lycopene administration. The kinetic models predicted that continued administration of lycopene as tablets or soup at a dose of 20 mg could result in plasma steady state concentrations as high as 1.0–1.5 µmol/L. Richelle et al. [12] administered 25 mg of lycopene daily for 8 weeks in the form of a tomato oleoresin embedded in a whey protein matrix and attained plasma lycopene concentrations of 1 µmol/L. The comparatively high mean lycopene plasma concentrations (0.6–0.9 µmol/L) observed at the onset of the present study in our volunteers (Day –14) (Fig. 1) must therefore be attributed to a substantial average intake of the carotenoid with the diet. The pronounced variations in lycopene concentration at Day –14 (SD in Fig. 1) reflects the absence of adequate standardization of lycopene intake before subjects were entering the study, ingesting differing lycopene doses from food products exhibiting varying bioavailability.

Half-life for 5-Z lycopene was consistently longer than that found for the all-E isomer. However, empirical modelling yielded a somewhat shorter half-life (7 days) than the compartmental approach (9 days). This difference is probably related to the sampling period of the post-dosing phase, which is relatively short for determining a half-life of more than 7 days. In particular the data of Day 9 could still reflect a contribution of a more rapid distribution phase, thereby shortening the apparent half-life. Thus, compartmental modelling of the total concentration-time profile is more likely to provide the correct half-life.

The present study does not resolve any details on lycopene absorption, distribution and elimination. Such information could be gained from single dose studies using labelled carotenoids [24, 25]. Plasma lipoprotein carriers efficiently mediate carotenoid post-absorptive transport and distribution and these processes are rapid in comparison to elimination. In the present study, wide

sampling intervals ensured that the plasma concentration time profiles principally demonstrated accumulation and elimination processes, whereas faster processes (absorption and distribution) could not be resolved.

In conclusion, the study confirmed the importance of food processing for improving carotenoid availability. Appropriate food processing resulted in systemic lycopene availability comparable to that of synthetic lycopene formulated in tablets. Apparent half-life estimates came to about 5 and 9 days for all-E and 5-Z lycopene, respectively. The synthetic lycopene tablet formulation used in this investigation may be of value for future clinical investigations.

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