

Quantifying the Influence of Thermal Process Parameters on in Vitro β -Carotene Bioaccessibility: A Case Study on Carrots

Lien Lemmens, Ines J. P. Colle, Sandy Van Buggenhout, Ann M. Van Loey, and Marc E. Hendrickx*

Laboratory of Food Technology and Leuven Food Science and Nutrition Research Centre (LForCe), Department of Microbial and Molecular Systems (M²S), Katholieke Universiteit Leuven, Kasteelpark Arenberg 22, 3001 Leuven, Belgium

ABSTRACT: This study describes a detailed and systematic investigation on the effect of thermal processing in terms of temperature and time (kinetic study) on β -carotene in vitro bioaccessibility in carrots. β -Carotene in vitro bioaccessibility increased with increasing processing temperature and time until steady-state conditions were reached after prolonged heating. The bioaccessibility values in steady-state conditions were temperature dependent. The experimental bioaccessibility data could adequately be modeled with a fractional conversion model. For the first time, modeling of processing-induced bioaccessibility changes is reported in literature. The results of the present kinetic study were used to estimate the impact of industrially relevant thermal processes on β -carotene bioaccessibility in carrots by simulation. It was shown that, to achieve a high β -carotene bioaccessibility, processing of carrots is essential (i.e., on the one hand, intense thermal processing or, on the other hand, mild thermal processing combined with intense mechanical processing).

KEYWORDS: β -carotene in vitro bioaccessibility, carrots, kinetics, thermal processing

INTRODUCTION

Thermal processing is an important food preservation unit operation.¹ Besides the desired inactivation of pathogenic and spoilage micro-organisms and deteriorative enzymes, several studies reported desired and undesired effects of thermal processing on other food quality aspects, such as nutrients, color, and texture.² Kinetic studies describing the effect of thermal processing on particular food quality aspects can be found in the literature. The identified kinetic models allow estimation of the impact of thermal processes on particular quality aspects and optimization of thermal processing conditions to maximize food product quality.³ In the context of the nutritional quality of food products, increased attention is given to the concepts of nutrient bioaccessibility and bioavailability. Nutrient bioaccessibility can be defined as the fraction of the ingested nutrients that is released from the food matrix⁴ and is available for intestinal absorption from the lumen,^{4,5} whereas nutrient bioavailability includes additionally nutrient absorption, tissue distribution, and metabolism.^{4,5} Typically, nutrient bioaccessibility is determined by in vitro methods, whereas nutrient bioavailability can be assessed in vitro and in vivo.⁴ Applying these concepts to the specific case of carrots, it would be worthwhile to study β -carotene bioaccessibility (and bioavailability), because carrots are the major source of β -carotene intake in most European countries⁶ and this micronutrient is water-insoluble and encapsulated in a structured system (tissue, cell, cell organelles).⁴ Moreover, important health benefits have been ascribed to its consumption, such as a reduced risk for cancer and cardiovascular disease (due to the antioxidative properties)^{7,8} and roles in vision, reproduction, and immune function (due to the provitamin A capacity).^{9,10} The typical chemical structure of β -carotene is depicted in Figure 1.

Carrots can be consumed in different ways: raw as such or, in most cases, mechanically and/or thermally processed (e.g., cooked, as part of a soup/sauce). It is relevant to study the effect

of these processing techniques on β -carotene bioaccessibility, as it reflects the effective nutritional value of the processed food products. Studies examining the influence of thermal processing on β -carotene bioaccessibility and bioavailability in different food matrices are available, each describing the effect of a specific thermal process on β -carotene bioaccessibility or bioavailability. A positive, enhancing effect of thermal processing on the amount of β -carotene released from the carrot matrix compared to raw carrots was observed in the studies of Hedrén et al.¹¹ (thermal processing = cooking), Lemmens et al.¹² (thermal processing = cooking and thermal treatments at 90, 100, and 110 °C), and Knockaert et al.¹³ (thermal processing = pasteurization and sterilization processes), whereas Hornero-Mendez and Minguez-Mosquera¹⁴ and Ryan et al.¹⁵ observed a decreased β -carotene release during digestion when, respectively, carrots or courgettes, red peppers, and tomatoes were cooked. In most cases, thermal processing was reported to increase β -carotene micellarization in carrots^{14,16,17} or in other vegetables.^{15,16,18} However, O'Sullivan et al.¹⁹ showed that the effect of cooking on β -carotene micellarization depended on the vegetable type that was considered. Recently, Tydeman et al.²⁰ concluded that heat had no positive effect on β -carotene bioaccessibility (measured as β -carotene partitioning into an emulsified oil phase) in carrots, and particle size reduction was identified to be more important than heating for bioaccessibility. Considering β -carotene bioavailability, Rock et al.²¹ and Edwards et al.²² observed increased bioavailability in cooked carrots compared to raw carrots, whereas Bugianesi et al.²³ detected no significant differences in β -carotene bioavailability between raw and domestically cooked cherry tomatoes.

Received: December 20, 2010

Revised: March 2, 2011

Accepted: March 5, 2011

Published: March 05, 2011

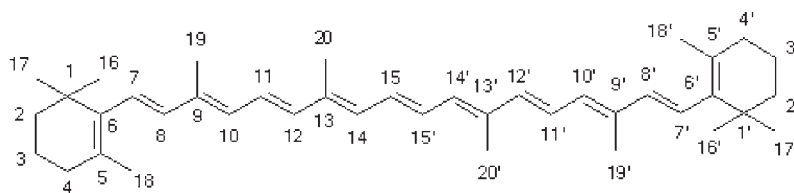


Figure 1. Chemical structure of β -carotene. The typical numbering of the carbon atoms is indicated.

From these studies, it is clear that no unambiguous effect of thermal processing on β -carotene bioaccessibility or bioavailability can be distinguished. Attention was drawn to this complex effect of thermal processing on carotenoid bioaccessibility and bioavailability in the review of Van Buggenhout et al.²⁴ as well. The conflicting information can be due to different factors, such as differences in methodologies to determine the bioaccessibility or bioavailability and (slightly) different starting material (e.g., particle size, (no) oil added during processing, (no) oil added during digestion). As indicated, all previous investigations in this context studied the effect of a single specific thermal process (e.g., a typical cooking or steaming process) on β -carotene bioaccessibility. Systematic studies on the effect of thermal process parameters (temperature and time) on β -carotene bioaccessibility are completely lacking.

Therefore, the aim of the present work was to examine in a detailed and systematic way the effect of temperature and time on β -carotene bioaccessibility in carrots. It was evaluated whether a kinetic model could be identified to adequately describe the evolution of β -carotene bioaccessibility as a function of treatment temperature and time. Such a kinetic model would allow prediction of β -carotene bioaccessibility values of thermally processed carrot products, which can be of particular relevance for the food industry. Special attention was given to the carrot tissue particle size in the present study, as it was shown previously that this parameter is essential in determining the effect of thermal processing on β -carotene bioaccessibility.²⁵

MATERIALS AND METHODS

Generation of Various Sized Carrot Tissue Particles. On the basis of a previous study indicating the importance of carrot particle size for β -carotene bioaccessibility,²⁵ it was decided to split the carrot tissue particles into two groups for this experiment: carrot tissue particles smaller than 125 μm , further referred to as “small particle fraction”, and carrot tissue particles with dimensions between 500 and 4000 μm , further referred to as “large particle fraction”. Hereby, it was assumed that the small particle fraction mainly contains broken cells and cell fragments (i.e., particles smaller than a carrot cell), whereas the large particle fraction mainly contains cell clusters.²⁵ For the preparation of the small carrot tissue particle fraction, carrots were peeled, cut into pieces, and blended with a kitchen blender for 2 min (ratio carrots/water = 1:1). Afterward, the blended carrots were high-pressure homogenized at 1000 bar (Panda2K, Gea Niro Soavi, Mechelen, Belgium), and this was repeated four times. Analysis of the particle size distribution by laser diffraction (Malvern Instrument Ltd., Worcestershire, U.K.) revealed that processing the carrots mechanically through this protocol resulted in carrot samples mainly containing carrot tissue particles smaller than 125 μm (95% of the carrot tissue particles was smaller than 125 μm). For the preparation of the large carrot tissue particle fraction, carrots were peeled, cut into pieces, and blended using a Grindomix (5 s at 2500 rpm for 300 g of carrot material, Grindomix GM200, Retsch, Haan, Germany). The technique of wet sieving (sieve shaker

equipped with sieves of 500, 1000, 2000, and 4000 μm (Retsch, Aartselaar, Belgium)) was used to isolate the carrot tissue particles in the range of 500–4000 μm .

Throughout the study, a single batch of carrots (*Daucus carota* cv. Nerac) was used (stored at 4 °C).

Thermal Treatments of Carrot Tissue Particles. For all thermal treatments, the carrot tissue particles (small or large carrot tissue particle fraction) were enclosed in stainless steel tubes (13 mm internal diameter, 16 mm external diameter, 150 mm length) and were preheated to 40 °C to shorten the come-up time required to reach the actual treatment temperature. The small carrot tissue particle fraction could be treated as such (water was already added during the preparation), whereas for the large carrot tissue particle fraction, deionized water was added to the reactor tubes as a heating medium.

To determine the relevant temperature/time range for the actual kinetic experiment, a screening study was performed: the carrot tissue particles (small or large particle fraction), enclosed in the reactor tubes, were thermally treated in an oil bath at temperatures ranging from 80 to 130 °C for a fixed time interval of 15 min. Afterward, the reactor tubes containing the carrot tissue particles were immediately cooled in an ice bath, and the samples were analyzed for β -carotene bioaccessibility. In this way, processing conditions under which changes in β -carotene bioaccessibility were expected could be identified.

For the actual kinetic study, the carrot tissue particles, enclosed in the reactor tubes, were thermally treated in an oil bath at temperatures ranging from 90 to 120 °C for time intervals ranging from 0 to 60 min. Afterward, the reactor tubes containing the carrot tissue particles were immediately cooled in an ice bath, and the samples were analyzed for β -carotene bioaccessibility. During the thermal treatments, the complete time/temperature profile of the carrot tissue particles was registered.

Determination of the β -Carotene in Vitro Bioaccessibility.

To determine β -carotene bioaccessibility, the carrot tissue particles were digested in vitro on the basis of the method of Hedrén et al.¹¹ with minor modifications as described by Lemmens et al.¹² In the present study, digestion in the mouth (mastication) was not considered, because all carrot tissue particles were small enough to swallow as such.²⁵ The method comprises gastric and small intestinal digestion. For the gastric digestion, NaCl (0.9% in water)/ascorbic acid (1% in water) solution and stomach electrolyte solution (containing NaCl (0.30%), KCl (0.11%), $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ (0.15%), KH_2PO_4 (0.05%), and $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$ (0.07%) dissolved in water) were added to the carrot tissue particles. The pH of the samples was adjusted to 4, and gastric juice (0.52% porcine pepsin (Sigma-Aldrich) in electrolyte solution) was added, followed by an incubation for 30 min at 37 °C with end-over-end shaking. The further stay in the stomach was imitated by adjusting the pH to 2 and by incubating the samples for 30 min at 37 °C with end-over-end shaking. The small intestinal digestion started with increasing the pH to 6.9 and adding duodenal juice (containing 0.4% porcine pancreatin (Sigma-Aldrich), 2.5% porcine bile extract (Sigma-Aldrich), 0.5% pyrogallol (Sigma-Aldrich), and 1% tocopherol (Sigma-Aldrich) in water) before the incubation was continued for 2 h at 37 °C. Each time the samples were incubated, the headspace was flushed with nitrogen to minimize oxidation reactions. To finalize the in vitro digestion, the digested and undigested materials were separated using a

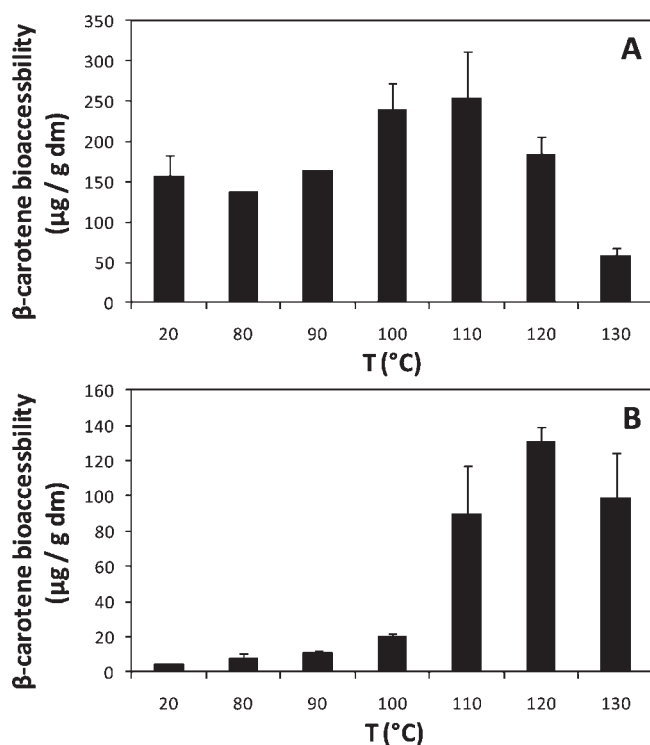


Figure 2. β -Carotene bioaccessibility ($\mu\text{g/g dm}$) (\pm standard deviation) as a function of treatment temperature for thermal treatments with a fixed time of 15 min: (A) treatments of the small carrot tissue particle fraction ($<125 \mu\text{m}$); (B) treatments of the large carrot tissue particle fraction ($500\text{--}4000 \mu\text{m}$). A temperature of 20°C refers to the untreated carrot tissue particles.

filter paper (Macherey-Nagel 615 1/4, folded filters, 185 mm diameter), and β -carotene could be extracted from the filtrate. A detailed description on the extraction and quantification procedure can be found in the study of Lemmens et al.²⁶ A RP-HPLC system (Agilent Technologies 1200 Series, Diegem, Belgium), equipped with a C_{30} column (25°C) ($5 \mu\text{m} \times 250 \text{ mm} \times 4.6 \text{ mm}$, YMC Europe, Dinslaken, Germany) and a diode array detector (DAD) (450 nm), was used. β -Carotene bioaccessibility was determined in duplicate. In the present study, the concentration of *cis*-isomers in the digested samples was too low to quantify (limit of detection and limit of quantification were, respectively, 0.0005 and $0.002 \mu\text{g}$ on column) and, hence, the results presented for β -carotene bioaccessibility always refer to *all-trans*- β -carotene bioaccessibility.

The dry matter content of the carrot tissue particles (small and large particle fractions) was determined, and β -carotene bioaccessibility values are expressed on a dry matter basis.

Data Analysis. In this study, a fractional conversion model was proposed to fit the experimental data points. Hence, it is assumed that when carrot tissue particles are heated at a specific temperature, β -carotene bioaccessibility reaches a plateau value after prolonged heating (steady-state conditions are attained). Generally, a fractional conversion model can be described by eq 1

$$B = B_f + (B_0 - B_f) \exp(-kt) \quad (1)$$

where B represents β -carotene bioaccessibility, B_f β -carotene bioaccessibility in steady-state conditions, B_0 initial β -carotene bioaccessibility ($\mu\text{g } \beta\text{-carotene/g dm}$), t reaction time (min), and k reaction rate constant (min^{-1}).

Data analysis was performed using nonlinear two-step regression analysis (SAS, v9.2, Cary, NC). In the first step, the kinetic parameters k

and B_f were estimated for each temperature, and in the second step, the temperature dependence of the kinetic parameters was investigated. The quality of fit was evaluated graphically by means of the parity plot.

Simulation Examples. From a more practical point of view, the results of this kinetic study can be applied to industrially relevant processing conditions, that is, a typical pasteurization process (resulting in a $10^\circ\text{C}P_{90^\circ\text{C}}$ value of 10 min), a typical sterilization process, and a typical UHT process (resulting in a $10^\circ\text{C}F_{121^\circ\text{C}}$ value of 6 min). By simulation, the impact of these processes on β -carotene bioaccessibility was estimated. The simulation was carried out for this specific batch of carrots and, thus, when the carrot variety is changed, the absolute values may differ but the general trends observed should be similar. As the carrot tissue particle size was shown to be an important factor determining β -carotene bioaccessibility, four carrot samples, each having a different particle size distribution, were included in this simulation example: blended carrots (sample A), blended carrots further high-pressure homogenized at 200 bar (sample B), blended carrots further high-pressure homogenized at 1000 bar (sample C), and blended carrots further high-pressure homogenized at 1000 bar for four times (sample D). The particle size distribution curves were experimentally determined by laser diffraction, whereas the β -carotene bioaccessibility values were estimated/predicted on the basis of the results of the present work (kinetic models).

RESULTS AND DISCUSSION

Screening Study. In Figure 2, the temperature screening of β -carotene bioaccessibility is shown for small (Figure 2A) and large carrot tissue particles (Figure 2B). For a fixed treatment time of 15 min, the effect of temperature on β -carotene bioaccessibility is different for small and large carrot tissue particles. For the small particle fraction, Figure 2A illustrates that β -carotene bioaccessibility was only minimally affected by the treatment temperature. For temperatures ranging from 80 to 110°C , a small increase of β -carotene bioaccessibility with increasing treatment temperature could be observed, although the differences were limited, especially compared to the effect of temperature on β -carotene bioaccessibility for the large particle fraction (Figure 2B). Also at the highest temperature applied in this study (130°C), a difference between the small and large carrot tissue particle fraction could be detected. For both particle fractions, isomerization might become important at this temperature, explaining the lower amount of β -carotene that is bioaccessible. However, it is clear that for the small particle fraction (Figure 2A), isomerization is more pronounced at 130°C compared to isomerization observed in the large particle fraction (Figure 2B). This implies that β -carotene isomerization is dependent on the carrot tissue particle size: for small carrot tissue particles, β -carotene is less protected and hence more vulnerable to degradation by isomerization compared to β -carotene in the large carrot tissue particles. Furthermore, Figure 2B clearly shows that for the large particle fraction, thermal treatments could be used to increase β -carotene bioaccessibility to a large extent. Especially at 110 and 120°C , high values for β -carotene bioaccessibility were detected compared to raw carrot tissue particles or carrot tissue particles that had been treated at lower temperatures. The following explanation is suggested to describe the effect of temperature on the β -carotene bioaccessibility of large carrot tissue particles. The release of β -carotene during digestion (β -carotene bioaccessibility) can be seen as a “multiple barrier” process: overcoming different “hurdles” is required (organelle membranes, cell membrane, cell wall) to liberate β -carotene from the cell.^{27,28} By increasing the

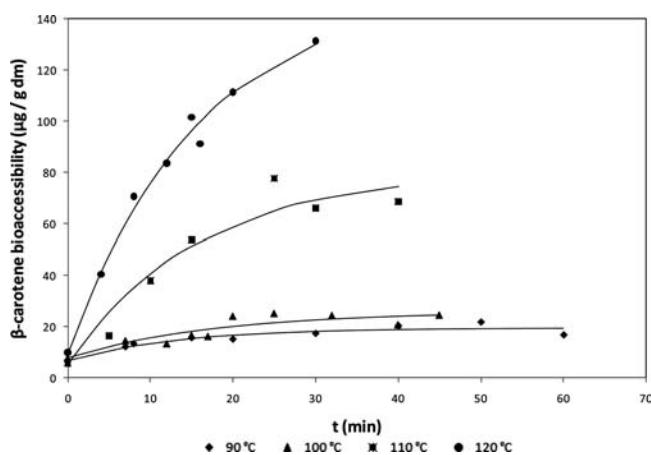


Figure 3. β -Carotene bioaccessibility ($\mu\text{g/g dm}$) as a function of treatment time for different temperatures modeled by a fractional conversion model. The full lines represent the bioaccessibility values predicted by the kinetic model, whereas the experimental data are represented by the symbols.

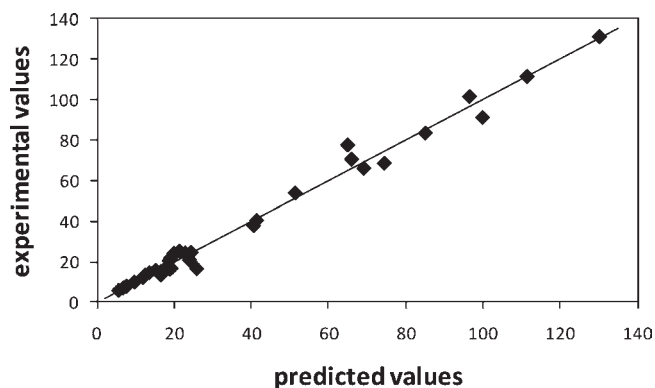


Figure 4. Parity plot (relationship between predicted and experimental bioaccessibility values ($\mu\text{g/g dm}$)) for the model describing the evolution of β -carotene bioaccessibility as a function of treatment time for different temperatures. The full line has a slope of 1 (perfect fit).

treatment temperature, additional barriers are removed, resulting in a more complete release of β -carotene. The large increase of β -carotene bioaccessibility for carrot tissue particles treated at high temperatures ($T > 100\text{ }^\circ\text{C}$) might be associated with additional processing-induced pectin changes at these temperatures,²⁹ affecting the carrot tissue structure and consequently facilitating the release of β -carotene during digestion. The possible link between pectin structural changes and β -carotene bioaccessibility was already highlighted by Lemmens et al.¹² and Epriliati et al.³⁰

The data obtained in this screening study allow identification of process and product parameters for the actual kinetic study: it is only relevant to perform a kinetic study on the large carrot tissue particle fraction, because the effect of temperature on the β -carotene bioaccessibility of the small particle fraction is minimal. The results are in agreement with previous results of Lemmens et al.,²⁵ who showed that the effect of thermal processing on β -carotene bioaccessibility is strongly dependent on the carrot tissue particle size. As processing conditions for the actual kinetic study, temperatures between 90 and 120 $^\circ\text{C}$ and treatment times between 0 and 60 min were selected.

Table 1. Overview of the Kinetic Parameters (\pm Standard Deviation; Based on 95% Confidence Interval) and the Correlation Coefficient of the Parity Plot for the Model Describing the Evolution of β -Carotene Bioaccessibility as a Function of Treatment Time during Thermal Treatments of Large Carrot Tissue Particles at Different Temperatures^a

T ($^\circ\text{C}$)	k ($\times 10^{-3}\text{ min}^{-1}$)	B_f ($\mu\text{g/g dm}$)
90	74.6 ± 21.1	19.5 ± 1.2
100	55.9 ± 24.4	26.0 ± 3.6
110	62.9 ± 23.1	80.5 ± 11.8
120	63.1 ± 9.1	151.3 ± 11.0

^a R^2 , parity plot = 0.989.

Actual Kinetic Study. In Figure 3, the experimental data and the model fit, assuming a fractional conversion model for the evolution of β -carotene bioaccessibility as a function of treatment time, are shown for the different temperatures. It can visually be detected that the kinetic model adequately describes the experimental data. The corresponding parity plot, plotting the experimental bioaccessibility values versus the predicted bioaccessibility values, has a high correlation coefficient, which is an indication for a good quality of fit (Figure 4). The estimated kinetic parameters (reaction rate constants (k) and bioaccessibility values in steady-state conditions (B_f)) at the different temperatures and the correlation coefficient for the parity plot are listed in Table 1.

Considering Figure 3 and Table 1, it can be observed that for all treatment temperatures, β -carotene bioaccessibility evolved to a steady state after prolonged heating and these B_f values were temperature dependent. The use of a fractional conversion model to fit the experimental data points was shown to be appropriate for this case study on β -carotene bioaccessibility in carrots, more specifically for the large carrot tissue particle fraction. For the lowest temperatures studied (90 and 100 $^\circ\text{C}$), β -carotene bioaccessibility increased only slightly upon heating. For temperatures above 100 $^\circ\text{C}$, thermal treatments clearly had a large positive (enhancing) effect on β -carotene bioaccessibility, even for short treatment times. These findings are in agreement with the trends observed in the screening study. Furthermore, in Table 1, it can be noted that the k values are not dependent on the treatment temperature (similar for all temperatures), whereas the B_f values are clearly increasing with increasing treatment temperature for the conditions studied in this investigation. This implies that, for all temperatures, the time required to reach a particular percentage of the B_f value is identical, whereas the value of the plateau in steady-state conditions is clearly influenced by the treatment temperature: by treating the carrot tissue particles at a higher temperature, more β -carotene could be released during digestion in steady-state conditions. The temperature dependence of the B_f values could accurately be described by a second-degree polynomial curve ($R^2 = 0.9956$) for the temperature range studied in this investigation. The equation of the polynomial curve ($y = 0.1608x^2 - 29.274x + 134.99$ with $y = B_f$ and $x = T$) can be used to estimate the bioaccessibility values of the plateaus when the thermal treatments are performed at other temperatures in the range of 90–120 $^\circ\text{C}$. From the polynomial curve, it can also be deduced that the effect of increasing the treatment temperature on the B_f values is more pronounced for thermal treatments performed at high temperatures ($T > 100\text{ }^\circ\text{C}$) than for thermal treatments

Table 2. β -Carotene Bioaccessibility Values for Different Processed Carrot Samples Estimated on the Basis of the Results of the Present Work (Kinetics Models) by Simulation^a

	estimated β -carotene bioaccessibility values ($\mu\text{g} / \text{g dm}$)			
	mechanically processed	mechanically processed + pasteurized	mechanically processed + sterilized	mechanically processed + UHT processed
sample A 9 % small particles 91 % large particles	18.4	28.9	118.8	30.1
sample B 12 % small particles 88 % large particles	23.0	33.2	120.1	34.3
sample C 40 % small particles 60 % large particles	65.9	72.8	132.1	73.6
sample D 87 % small particles 13 % large particles	137.9	139.4	152.2	139.6

^a Different intensities of mechanical as well as thermal processing were included. Samples A, B, C, and D refer to, respectively, blended carrots, blended carrots further high-pressure homogenized at 200 bar, blended carrots further high-pressure homogenized at 1000 bar, and blended carrots further high-pressure homogenized at 1000 bar four times. The particle size distribution curves of the different samples were determined experimentally by laser diffraction.

performed at temperatures below 100 °C. This supports the earlier observations and an important conclusion of this kinetic study, that is, only for carrot tissue particles that have been treated at high temperatures (130 °C > T > 100 °C), a large increase of β -carotene bioaccessibility was achieved compared to untreated carrot tissue particles (observed in screening study as well as in actual kinetic study).

It should be noted that the dynamic temperature conditions during heating up were not taken into consideration, because the dynamic phase was only limited in time (around 5 min) due to the preheating of the carrot samples to 40 °C. Hence, the temperature of the samples that were taken to be analyzed for β -carotene bioaccessibility was in most cases equal to the desired treatment temperature.

Implications for Industrially Relevant Processing Conditions. In Table 2, the results of the simulation examples are given. The conditions of an industrially relevant pasteurization process (90 °C, 20 min), sterilization process (120 °C, 20 min), and ultrahigh-temperature (UHT) process (130 °C, 50 s) were simulated, and the impact on β -carotene bioaccessibility was evaluated. Some important factors were considered when β -carotene bioaccessibility was predicted: (i) the percentage of small particles, (ii) the percentage of large particles, (iii) the β -carotene bioaccessibility of the small particles (untreated and treated, results from screening study), and (iv) the β -carotene bioaccessibility of the large particles (untreated (B_0), results from screening study). In the table, the processes are ordered with increasing intensity of mechanical processing (vertically) and with increasing intensity of thermal processing (horizontally). Important conclusions could be drawn from the table, and results from our previous investigation²⁵ could be validated. First of all, it is clear that the importance of thermal processing is dependent on the carrot tissue particle size and hence on the extent to which the samples have been mechanically processed. For carrot samples with a high amount of small particles (e.g., sample D), thermal processing is far less essential to achieve a high β -carotene bioaccessibility. On the other hand, carrot samples

containing mostly large particles (e.g., sample A) require intense thermal processing (sterilization) to reach a high value for β -carotene bioaccessibility. If the data are considered from the perspective of thermal processing, it can be deduced that for short (e.g., UHT process) or mild (e.g., pasteurization process) thermal processes, it is important to reduce the carrot tissue particle size by mechanical processing to obtain high β -carotene bioaccessibility, whereas more intense thermal processes (e.g., sterilization process) give rise to high β -carotene bioaccessibility values in all cases. These simulation examples, using the data of the present work, support the hypothesis postulated in the study of Lemmens et al.²⁵ to obtain high β -carotene bioaccessibility values, intense thermal processing of carrots or severe particle size reduction by mechanical processing of carrots is crucial.

In conclusion, it can be stated that the present work studied for the first time systematically the effect of temperature and time on β -carotene bioaccessibility in a detailed way. The experimental data could adequately be modeled by a fractional conversion model. This investigation emphasized the importance of processing to increase the nutritional value (i.e., β -carotene bioaccessibility) of carrot-based food products: intense thermal processing (e.g., sterilization) or (intense) mechanical processing (e.g., high-pressure homogenization) of carrots is required to reach a high value for β -carotene bioaccessibility. It should always be kept in mind that during intense processing, degradation of β -carotene can occur. However, considering the results of a previous kinetic study on β -carotene isomerization in carrots,²⁶ it can be concluded that the impact of thermal processes on the β -carotene content was limited compared to the impact on β -carotene bioaccessibility: the relative β -carotene bioaccessibility increase that was achieved due to thermal processing was much larger than the accompanying relative decrease in β -carotene concentration.

The results of the present study are of importance for product and process design in the food industry (e.g., production of soups and sauces containing carrot particles). Predictions of the β -carotene bioaccessibility of processed carrot products can be

made. It is obvious that similar studies in other fruit and vegetable matrices or on other nutrients would enlarge the possibilities for intelligent product and process design with regard to the nutritional value of the food products.

AUTHOR INFORMATION

Corresponding Author

*Postal address: Laboratory of Food Technology and Leuven Food Science and Nutrition Research Centre (LForCe), Department of Microbial and Molecular Systems (M²S), Katholieke Universiteit Leuven, Kasteelpark Arenberg 22, Postbox 2457, 3001 Leuven, Belgium. Phone: +32 16 32 15 85. Fax: +32 16 32 19 60. E-mail: marc.hendrickx@biw.kuleuven.be.

Funding Sources

This research was financially supported by the Commission of the European Communities, Framework 6, Priority 5 'Food Quality and Safety', STREP Project Healthy Structuring (2006–023115). Sandy Van Buggenhout is a postdoctoral researcher funded by the Fund for Scientific Research Flanders (FWO). Ines Colle is financially supported by the Institute for the Promotion of Innovation through Science and Technology in Flanders (IWT).

ACKNOWLEDGMENT

The technical assistance of Margot De Haes and Heidi Roba during the experiments is greatly appreciated.

REFERENCES

- Richardson, P., Ed. *Thermal Technologies in Food Processing*; CRC Press: Boca Raton, FL, 2001.
- Holdsworth, S. D. Optimising the safety and quality of thermally processed packaged foods. In *Improving the Thermal Processing of Foods*; Richardson, P., Ed.; CRC Press: Boca Raton, FL, 2004; pp 3–31.
- Van Boekel, M. A. J. S. Statistical aspects of kinetic modeling for food science problems. *J. Food Sci.* **1996**, *61*, 477–489.
- Parada, J.; Aguilera, J. M. Food microstructure affects the bioavailability of several nutrients. *J. Food Sci.* **2007**, *72*, R21–R32.
- Holst, B.; Williamson, G. Nutrients and phytochemicals: from bioavailability to bioefficacy beyond antioxidants. *Curr. Opin. Biotechnol.* **2008**, *19*, 73–82.
- O'Neill, M. E.; Carroll, Y.; Corridan, B.; Olmedilla, B.; Granado, F.; Blanco, I.; Van den Berg, H.; Hininger, I.; Rousell, A.-M.; Chopra, M.; Southon, S.; Thurnham, D. I. A European carotenoid database to assess carotenoid intakes and its use in a five-country comparative study. *Br. J. Nutr.* **2001**, *85*, 499–507.
- Fraser, P. D.; Bramley, P. M. The biosynthesis and nutritional uses of carotenoids. *Prog. Lipid Res.* **2004**, *43*, 228–265.
- Rao, A. V.; Rao, L. G. Carotenoids and human health. *Pharmacol. Res.* **2007**, *55*, 207–216.
- Burri, B. J. β -Carotene and human health: a review of current research. *Nutr. Res. (N.Y.)* **1997**, *17*, 547–580.
- Dutta, D.; Chaudhuri, U. R.; Chakraborty, R. Structure, health benefits, antioxidant property and processing and storage of carotenoids. *Afr. J. Biotechnol.* **2005**, *13*, 1510–1520.
- Hedrn, E.; Diaz, V.; Svanberg, U. Estimation of carotenoid accessibility from carrots determined by an *in vitro* digestion method. *Eur. J. Clin. Nutr.* **2002**, *56*, 425–430.
- Lemmens, L.; Van Buggenhout, S.; Oey, I.; Van Loey, A.; Hendrickx, M. Towards a better understanding of the relationship between the β -carotene *in vitro* bio-accessibility and pectin structural changes: a case study on carrots. *Food Res. Int.* **2009**, *42*, 1323–1330.
- Knockaert, G.; De Roeck, A.; Lemmens, L.; Van Buggenhout, S.; Hendrickx, M.; Van Loey, A. Effect of thermal and high pressure processes on structural and health-related properties of carrots (*Daucus carota*). *Food Chem.* **2011**, *125*, 903–912.
- Hornero-Mendez, D.; Mínguez-Mosquera, M. I. Bioaccessibility of carotenes from carrots: effect of cooking and addition of oil. *Innovative Food Sci. Emerging Technol.* **2007**, *8*, 407–412.
- Ryan, L.; O'Connell, O.; O'Sullivan, L.; Aherne, A.; O'Brien, N. M. Micellarisation of carotenoids from raw and cooked vegetables. *Plant Foods Hum. Nutr.* **2008**, *63*, 127–133.
- Veda, S.; Kamath, A.; Platel, K.; Begum, K.; Srinivasan, K. Determination of bioaccessibility of β -carotene in vegetables by *in vitro* methods. *Mol. Nutr. Food Res.* **2006**, *50*, 1047–1052.
- Aherne, S. A.; Daly, T.; Jiwan, M. A.; O'Sullivan, L.; O'Brien, N. M. Bioavailability of β -carotene isomers from raw and cooked carrots using an *in vitro* digestion model coupled with a human intestinal Caco-2 cell model. *Food Res. Int.* **2010**, *43*, 1449–1454.
- Tumuhimbise, G. A.; Namutebi, A.; Muyonga, J. H. Microstructure and *in vitro* β -carotene bioaccessibility of heat processed orange fleshed sweet potato. *Plant Foods Hum. Nutr.* **2009**, *64*, 312–318.
- O'Sullivan, L.; Galvin, K.; Aherne, S. A.; O'Brien, N. M. Effects of cooking on the profile and micellarization of 9-*cis*-, 13-*cis*- and all-*trans*- β -carotene in green vegetables. *Food Res. Int.* **2010**, *43*, 1130–1135.
- Tydemann, E. A.; Parker, M. L.; Wickham, M. S. J.; Rich, G. T.; Faulks, R. M.; Gidley, M. J.; Fillery-Travis, A.; Waldron, K. Effect of carrot (*Daucus carota*) microstructure on carotene bioaccessibility in the upper gastrointestinal tract. I. *In vitro* simulations of carrot digestion. *J. Agric. Food Chem.* **2010**, *58*, 9847–9854.
- Rock, C. L.; Lovalvo, J. L.; Emenhiser, C.; Ruffin, M. T.; Flatt, S. W.; Schwartz, S. J. Bioavailability of β -carotene is lower in raw than in processed carrots and spinach in women. *J. Nutr.* **1998**, *128*, 913–916.
- Edwards, A. J.; Nguyen, C. H.; You, C.; Swanson, J. E.; Emenhiser, C.; Parker, R. S. α - and β -Carotene from a commercial carrot puree are more bioavailable to humans than from boiled-mashed carrots, as determined using an extrinsic stable isotope reference method. *J. Nutr.* **2002**, *132*, 159–167.
- Bugianesi, R.; Salucci, M.; Leonardi, C.; Ferracane, R.; Catasta, G.; Azzini, E.; Maiaini, G. Effect of domestic cooking on human bioavailability of naringenin, chlorogenic acid, lycopene and β -carotene in cherry tomatoes. *Eur. J. Nutr.* **2004**, *43*, 360–366.
- Van Buggenhout, S.; Alminger, M.; Lemmens, L.; Colle, I.; Knockaert, G.; Moelants, K.; Van Loey, A.; Hendrickx, M. *In vitro* approaches to estimate the effect of food processing on carotenoid bioavailability need thorough understanding of process induced microstructural changes. *Trends Food Sci. Technol.* **2010**, *21*, 607–618.
- Lemmens, L.; Van Buggenhout, S.; Van Loey, A.; Hendrickx, M. Particle size reduction leading to cell wall rupture is more important for the β -carotene bioaccessibility of raw compared to thermally processed carrots. *J. Agric. Food Chem.* **2010**, *58*, 12769–12776.
- Lemmens, L.; De Vleeschouwer, K.; Moelants, K. R. N.; Colle, I. J. P.; Van Loey, A. M.; Hendrickx, M. E. β -Carotene isomerization kinetics during thermal treatments of carrot puree. *J. Agric. Food Chem.* **2010**, *58*, 6816–6824.
- Rich, G. T.; Bailey, A. L.; Faulks, R. M.; Parker, M. L.; Wickham, M. S. J.; Fillery-Travis, A. Solubilization of carotenoids from carrot juice and spinach in lipid phases: I. Modeling the gastric lumen. *Lipids* **2003**, *38*, 933–945.
- Rich, G. T.; Faulks, R. M.; Wickham, M. S. J.; Fillery-Travis, A. Solubilization of carotenoids from carrot juice and spinach in lipid phases: II. Modeling the duodenal environment. *Lipids* **2003**, *38*, 947–956.
- De Roeck, A.; Duvetter, T.; Fraeye, I.; Van der Plancken, I.; Sila, D. N.; Van Loey, A.; Hendrickx, M. Effect of high-pressure/high-temperature processing on chemical pectin conversions in relation to fruit and vegetable texture. *Food Chem.* **2009**, *115*, 207–213.
- Epriliani, I.; D'Arcy, B.; Gidley, M. Nutritional analysis of fresh and processed fruit products. I. During *in vitro* digestions. *J. Agric. Food Chem.* **2009**, *57*, 3363–3376.