

### Carotenoid transfer to oil upon high pressure homogenisation of tomato and carrot based matrices

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#### ABSTRACT

The transfer of carotenoids into the oil phase during digestion is limited by matrix related factors that hamper carotenoid release and the conditions of low gastric acidity that limit carotenoid solubility. Therefore, the aim of this study was to investigate the use of high pressure homogenisation (HPH) as a driving force to favour carotenoid transfer to oil before digestion. The level of bio-encapsulation and carotenoid hydrophobicity were investigated as factors that may influence carotenoid transfer from tomato and carrot based matrices to oil upon HPH. The results indicated that the selective transfer of a particular carotenoid depended on its hydrophobicity. The cell wall in tomato cell clusters represents a limiting factor for carotenoid transfer. Overall, the findings indicate that HPH is efficient in transferring carotenoids to the oil phase and this can be crucial to improve the nutritional quality of carrot and tomato-based products.

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#### 1. Introduction

There is evidence that dietary carotenoids from fruits and vegetables are beneficial to human health (Margalit et al., 2012; Mayne, 2013; Zaini, Brandt, Clench, & Le Maitre, 2012). Nevertheless, it is known that despite their prevalence in fruits and vegetables, the absorption of carotenoids during digestion can be low and highly variable depending on the diet and hostrelated factors (Faulks & Southon, 2005). This is because in order to confer their health effects, carotenoids must first be released from the matrix, solubilised in the lipid phase of chyme followed by transfer into mixed micelles in the small intestine before being taken up by the body and finally reach their site of action (Castenmiller, West, Linssen, van het Hof, & Voragen, 1999). As a result, the health benefits of carotenoids are strongly dependent on their bioaccessibility (i.e. fraction of an ingested nutrient that is released from the food matrix and made available for intestinal absorption). It is apparent that the different steps involved in the carotenoid absorption process are conditioned by different factors that can be controlled/ modified in order to improve carotenoid bioaccessibility (Fernández-García et al., 2012). In this context several studies in the past decades have focused on understanding the factors

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determining the bioaccessibility of carotenoids from fruit and vegetable materials.

It is well established that processing, thermal and/or mechanical, can be exploited to facilitate carotenoid release from the matrix and as a consequence, influence carotenoid bioaccessibility (Colle, Lemmens, Van Buggenhout, Van Loey, & Hendrickx, 2010a; Knockaert, Lemmens, Van Buggenhout, Hendrickx, & Van Loey, 2012a; Panozzo et al., 2013; Svelander, Lopez-Sanchez, Pudney, Schumm, & Alminger, 2011). Nevertheless, processing may lead also to carotenoid trans-cis isomerisation. The cis isomers possess different biological properties such as decreased pro-vitamin A activity and altered antioxidant capacity as compared to the respective trans isomers (Schieber & Carle, 2005). It is important to note that while processing disrupts cellular structures and organelle membranes which can facilitate carotenoid release (Van Buggenhout et al., 2010), some modifications of the matrix have been reported to actually entrap carotenoids limiting their release during digestion. In addition, carotenoid release depends on the structural barriers (i.e. chromoplast sub-structures and cell walls) naturally present in the matrix (Palmero et al., 2013). Moreover, some extrinsic factors, such as the co-ingestion of lipids, have been demonstrated to largely affect carotenoid bioaccessibility (Colle et al., 2013; Huo, Ferruzzi, Schwartz, & Failla, 2007; Salvia-Trujillo, Qian, Martín-Belloso, & McClements, 2013). In this context, carotenoid solubilisation into the oil phase has been highlighted as one of the critical steps in their absorption (Hedrén, Diaz, & Svanberg, 2002). Thus, the type of carotenoid and its solubility in the lipid phase play an important role in determining carotenoid bioaccessibility. For instance, a lower bioaccessibility of lycopene compared to  $\beta$ -carotene was reported and attributed to be related to the lower solubility of the former in dietary lipid compared to the latter (Svelander et al., 2011). Recently, a study by Palmero, Panozzo, Simatupang, Hendrickx, and Van Loey (2014) demonstrated the strong dependency of carotenoid transfer efficiency during digestion from the matrix into the oil phase on the level of bio-encapsulation as well as carotenoid hydrophobicity. This implies that the positive effects that can be realized by employing processing to enhance carotenoid release are diminished greatly by interplay of the factors that are controlling the movement of carotenoids from the matrix to the oil during digestion. These challenges are arising mainly from their low water solubility (due to their highly lipophilic nature), high melting point (due their existence in crystalline form) and poor chemical stability (due to the conjugated system of double bonds) (Boon, McClements, Weiss, & Decker, 2010).

From the above, processing fruit and vegetable based matrices in the presence of oil can be exploited to facilitate transfer of carotenoids to an oil phase as a means of introducing the lipid phase prior to ingestion. In this respect, high pressure homogenisation (HPH) can be a useful tool to facilitate the transfer of carotenoids from their natural location in the matrix to the oil phase. This transfer may for instance be affected by diffusion or by turbulent mixing or generally by the combined action of diffusion, turbulence and convection. In general, the mechanism of mass transfer depends on the dynamics of the system in which it occurs (Bravo, 2011). By recognizing that mass can be transferred by random molecular motion in quiescent fluids, aided by the dynamic characteristics of the flow (Bravo, 2011), the applied homogenisation pressure during HPH can be exploited as the driving force for carotenoid transfer into oil. This can be useful particularly for those fruit and vegetable based matrices such as tomato where the formation of a fibre network that entraps lycopene in the matrix following processing are carried over through the digestion process. In this way, matrix related factors that hamper carotenoid release (Castenmiller et al., 1999) and the conditions of low gastric acidity that limit carotenoid solubility into the oil phase during digestion (Rich, Fillery-Travis, & Parker, 1998) can be circumvented. Nonetheless, the potential of HPH to facilitate carotenoid transfer to oil has not been reported. This could potentially pave the way to a new and/or different applications in the context of process design for targeted bioactive compounds such as carotenoids in fruit and vegetable processing. In this regard, it is crucial to establish whether factors such as carotenoid hydrophobicity and level of encapsulation which have been highlighted above as influencing the transfer efficiency into oil during digestion are persisting during processing. Therefore, the present work evaluated the transfer of lycopene,  $\beta$ -carotene and  $\alpha$ -carotene from tomato and carrot based matrices to oil as influenced by HPH.

#### 2. Materials and methods

#### 2.1. Materials

All chemicals and reagents used were of analytical or HPLCgrade. L- $\alpha$ -phosphatidylcholine and carotenoid standards (alltrans lycopene, all-trans  $\beta$ -carotene and all-trans  $\alpha$ -carotene) were purchased from Sigma-Aldrich (Borne, Belgium). Olive oil (extra virgin) was kindly donated by Vandemoortele (Ghent, Belgium). Red ripe tomatoes (Lycopersicon esculentum cv Prunus) and orange carrots (Daucus carota cv Nerac) were obtained fresh from a local shop in Belgium and stored at 4 °C for 1 day prior to use.

#### 2.2. Experimental set up

The effect of high pressure homogenisation on carotenoid transfer from tomato and carrot matrices to oil was investigated. At first carrot and tomato matrices were decomposed into chromoplast-enriched and cell clusters fractions, as described further in the text. In this way, different physical barriers that hinder carotenoid release (chromoplast substructure and cell walls) were considered. The different fractions isolated from each matrix were mixed with an oil-in-water emulsion (5% oil) and high pressure homogenised at different pressure levels (10, 30, 50, 70, 100 MPa). Non-homogenised samples were considered as control samples. Treated and untreated samples were then ultra-centrifuged (Beckman Optima XPN-100 Ultracentrifuge, Brea, CA, USA) at 65,000 g for 1 hour at 4 °C in order to recover the oil. The recovered oil was analysed for carotenoid content. In particular, two types of carotenoids were quantified in the isolated fractions from each matrix, namely all-trans lycopene and all-trans  $\beta$ -carotene in tomato, and alltrans  $\beta$ -carotene and all-trans  $\alpha$ -carotene in carrot based system. Carotenoid content in the recovered oil from each fraction at each pressure level was compared with carotenoid content in

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