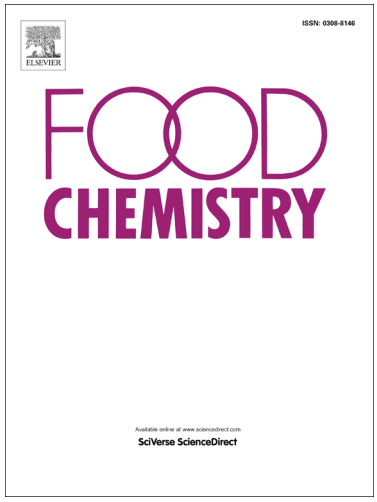
The influence of carrier material on some physical and structural properties of carrot juice microcapsules



The influence of carrier material on some physical and structural properties of carrot juice microcapsules

Emilia Janiszewska-Turak, Nicolò Dellarosa, Urszula Tylewicz, Luca Laghi, Santina Romani, Marco Dalla Rosa, Dorota Witrowa-Rajchert

PII: S0308-8146(17)30520-4
DOI: http://dx.doi.org/10.1016/j.foodchem.2017.03.134
Reference: FOCH 20838

To appear in: *Food Chemistry*

Received Date: 20 December 2016
Revised Date: 20 March 2017
Accepted Date: 22 March 2017

Please cite this article as: Janiszewska-Turak, E., Dellarosa, N., Tylewicz, U., Laghi, L., Romani, S., Dalla Rosa, M., Witrowa-Rajchert, D., The influence of carrier material on some physical and structural properties of carrot juice microcapsules, *Food Chemistry* (2017), doi: http://dx.doi.org/10.1016/j.foodchem.2017.03.134

The influence of carrier material on some physical and structural properties of carrot juice microcapsules

Emilia Janiszewska-Turak[1*], Nicolò Dellarosa[2], Urszula Tylewicz[2], Luca Laghi[2,3*], Santina Romani[2,3], Marco Dalla Rosa[2,3] and Dorota Witrowa-Rajchert[1]

[1] *Department of Food Engineering and Process Management, Faculty of Food Sciences, Warsaw University of Life Sciences (SGGW), Warsaw, ul. Nowoursynowska 159c, 02-776 Warsaw, Poland*

[2] *Department of Agricultural and Food Sciences, University of Bologna, Piazza Goidanich 60, 47521 Cesena, Italy*

[3] *Interdepartmental Centre for Agri-Food Industrial Research, University of Bologna, Via Quinto Bucci 336, 47521 Cesena, Italy*

Emilia Janiszewska-Turak - emilia_janiszewska_turak@sggw.pl, Nicolò Dellarosa - nicolo.dellarosa@unibo.it, Urszula Tylewicz - urszula.tylewicz@unibo.it, Luca Laghi – l.laghi@unibo.it, Santina Romani - santina.romani2@unibo.it, Marco Dalla Rosa - marco.dallarosa@unibo.it, Dorota Witrowa-Rajchert – dorota_witrowa_rajchert@sggw.pl

*Corresponding authors:

Dr. Luca Laghi – Tel: +39 0547 338105, Fax:+39 0547 382348

Ph. D. eng. Emilia Janiszewska-Turak, tel.: +48 22 5937566, fax.: +48 22 5937576

Abstract

This study investigates the effect of carrier type on the physical and structural properties of microcapsules of pure carrot juice. Low-crystallised maltodextrin (MD), gum arabic (GA), mixtures of MD and GA (1:1; 2:1; 3:1) and whey protein isolate (WPI) were used as carriers. Microencapsulation was carried out in a spray-drier at inlet air temperature of 160°C. Powders were investigated for dry matter, water activity, diameter, carotene content and hygroscopicity. In addition, differential scanning calorimetry (DSC) and time domain nuclear magnetic resonance (TD-NMR) were applied to analyse microcapsules glass transition temperature (Tg).

Carrot powders with GA used as a carrier material resulted in better carotenoids retention and higher stability of powders in terms of higher Tg, lower a_w and good hygroscopic properties. However, all powders showed a low a_w (below 0.26) and high dry matter content (98-99%) indicating a good potential for protection of microencapsulated carotenoids during the storage.

Key words: viscosity, DSC, TD-NMR, carrot juice, hygroscopicity

Abbreviations

a_w. – water activity

d.m. – dry matter

DSC – Differential Scanning Calorimetry

FID - free induction decay

GA – gum arabic

MD – Maltodextrin

MD:AG=1:1; 2:1; 3:1 – mixture of Maltodextrin and gum arabic expressed as weight ratio

WPI. – Whey Protein Isolate

$T_{melting}$ – temperature of melting point for a reference material used in DSC tests (^oC)

TD-NMR. – Time Domain Nuclear Magnetic Resonance

Tg – glass transition temperature (expressed in ^{o}C)

1. Introduction

Nowadays, there is an increased consumer demand for natural colorants, which could be obtained from vegetable juices. Carrot juice is particularly rich in carotenoids, in particular: β-carotene (8 mg/100g, which is about 50-60% of total carotene content), α-carotene (3.5 mg/100g – representing the 20% of total carotene content) and γ-carotene and ζ-carotene, responsible for the typical orange color of the carrot root (Ma et al., 2015). Natural colorants have been widely used for coloring different food products such as ice cream, yoghurt, cake mixes, gelatins, desserts, meat substitutes, sauces, salad dressings, processed cheese, snacks, confectionery, margarine, salad dressings, pastas, baked goods and mustard (Francis, 2002) . However, the limitation in using natural colorants at industrial level is their low resistance to pH changes, heating, light and oxidizing agents (Kandansamy & Somasundaram, 2012; Özkan & Ersus, 2014).

Microencapsulation is one of the methods used to protect these substances. The most common use of microencapsulation technique is spray drying, which consists in a division of the feed flux into small droplets by a spray disc, followed by their fast evaporation. Prior to spry-drying, a material, often defined as carrier, is added to create, along solidification, a protection for the active ingredient in the form of shell or matrix (Chranioti, Nikoloudaki, & Tzia, 2015; Domian, Brynda-Kopytowska, Cenkier, & Świrydow, 2015). The chemical and physical properties of the carrier material are a key factor for the success of spray-drying process. In general, the juices that undergo spray drying may get sticky due to the presence of low molecular weight sugars (such as glucose or fructose) and may therefore adhere to the walls of the spray drier chamber. Stickiness of a fluid is linked to its glass transition temperature

(Tg), being high when the fluid is at a temperature higher than its Tg. Carrier materials are generally added to the juices prior to drying in order to increase Tg, thus decreasing their stickiness, allowing the proper drying of the juice-carrier solution (Fazaeli, Emam-Djomeh, Ashtari, & Omid, 2012; Samborska, Langa, Kamińska□Dwórznicka, & Witrowa□Rajchert, 2015). The ideal carrier material i) has good rheological properties when used in high concentration; ii) creates stable emulsions; iii) does not react with the core substance during and after spray drying process; iv) is able to protect the core substance along shelf life; v) is soluble in aqueous or ethanol solution, so that it can be added to food (Chranioti et al., 2015; Domian et al., 2015; Gharsallaoui, Roudaut, Chambin, Voilley, & Saurel, 2007).

The most commonly used carrier materials for encapsulation colourants from juices by spray drying method are maltodextrin, gum arabic, whey protein isolate, and mixtures of them (Chranioti et al., 2015; Domian et al., 2015; Gharsallaoui et al., 2007). Maltodextrin (MD) is a polysaccharide, which molecular weight and properties depend on the hydrolysis process employed to obtain it from starch. Maltodextrin is classified by dextrose equivalent (DE) which measures the amount of reducing sugars present in a sugar product, relative to dextrose. High DE values lead to high levels of sweetness, solubility and hygroscopicity, but to low viscosity and Tg values (from 180 to 120^{o}C) (Roos & Karel, 1991). The biggest disadvantage of maltodextrin is the lack of emulsifying properties (Kandansamy & Somasundaram, 2012; Roos & Karel, 1991). Gum arabic (GA), also known as acacia gum, is particularly advantageous due to the low viscosity of its aqueous solutions and its ability to give rise to very stable emulsions, which facilitates the encapsulation of substances with a variety of characteristics (Kandansamy & Somasundaram, 2012; Özkan & Ersus, 2014). Moreover GA resulted to have a good encapsulation efficiency for other natural colorants from juices (Mahdavi, Jafari, Ghorbani, & Assadpoor, 2014). Whey protein isolate (WPI) is characterized by globular proteins, insoluble in milk at isoelectric point (around 5), but soluble at any other

pH value. Protein denaturation occurs at high temperature and moisture content, so that it is difficult to predict whey protein isolate stability in spray drying process (Kandansamy & Somasundaram, 2012).

As each pure carrier material is in some respect far from ideal, using mixtures of them is the most common choice in practical applications. For example, the combination of gum arabic with maltodextrin or other carriers results in better encapsulation efficiency (Fazaeli et al., 2012).

A key aspect of any carrier material used is its concentration in solution used to spray drying, with an optimum carrier material/ active substance dry matter ratio of 1:1 or 1:4 (Bayram, Bayram, & Tekin, 2008; Janiszewska, 2014).

The glass transition temperature (Tg) is a critical point also in dried powders along storage, modified by water uptake from the environment. In general, powders characterized by high water content and activity have low Tg (da Silva Carvalho et al., 2016; Fazaeli et al., 2012) and they are therefore prone to stickiness and caking phenomena, with negative consequences on textural, chemical and sensorial quality (Beristain, Azuara, Tamayo, & Vernon‐Carter, 2003).

Differential scanning calorimetry (DSC) is the most widely used technique to determine the Tg in pharmaceutical and food materials. Such technique is based on the measurement of the change in heat capacity, which occurs in samples around glass transition temperature. The analysis of Tg values by DSC has been performed in different spray dried powders, such as lemon juice powder (Paterson & Bröckel, 2015) and milk and grape/peach juice blend powders (Afifi, Abu Shelaibi, Laleye, & Ismail, 2009).

Recently, time domain nuclear magnetic resonance (TD-NMR) has been demonstrated to give information complimentary to DSC about water and polymers interactions around Tg of dried powders, because it is able to detect mobile water even when it has no translational mobility

according to DSC analysis (Rocculi et al., 2011). Indeed several works have appeared in the literature applying TD-NMR to foodstuff containing some of the molecules object of the present investigation, like malto-oligomers (Van Den Dries, Van Dusschoten, Hemminga, & Van Der Linden, 2000).

The aim of the study was to examine the effect of different carrier materials, namely maltodextrin, gum arabic, mixtures of them in different proportions and whey protein isolate, on the chemical and physico-chemical properties of carrot juice microcapsules. In particular, the powders were characterised for dry matter, water activity, particles diameter, hygroscopicity and glass transition temperature. Tg was investigated by two different but complementary methods differential scanning calorimetry (DSC) and Time domain nuclear magnetic resonance (TD-NMR). Moreover, the carotenoids content of microcapsules has been also investigated.

2. Materials and methods

2.1. Materials

Carrot juice employed in the present investigation was prepared from cultivar "Kazan", obtained from Agricultural Experimental Station (SGGW-WULS) in Żelazna (Poland). Low-crystallised maltodextrin with DE=11 (PPS "PEEPES" S.A., Łomża, Poland) with 9±2% of moisture content, gum arabic (Hortimex Sp. z o.o., Konin, Poland) with moisture content 5±1% and Whey Protein Isolate (WPI) (Hortimex Sp. z o.o., Konin, Poland) with moisture content 6±1% were used as the carriers.

2.2. Composition of juices and solutions for spray drying

To obtain the juice, the carrots were peeled and sliced with an approximate thickness of 0.5±0.1 cm. The slices were subjected to juice extraction by means of a kitchen extractor

(Kenwood, model JE600, UK). Raw juice was characterized by 10±0.2°Bx of solid soluble content (measured by Pocket Refractometer PAL-3 of ATAGO – Tokyo, Japan), 1040±0.2 kg/m^3 of density and 69.8±0.2 mPa·s of viscosity.

To compare microcapsules from different carrier materials, solid soluble content (ssc) concentration in the solutions was brought to 20% by adding the appropriate amount of the various carrier materials. The value of 20 °Bx of solid soluble content in solution was chosen on the basis of literature, in which the range 1:1 juice solid soluble content :carrier is suggested as the most suitable for spray drying (Janiszewska, 2014; Righetto & Netto, 2006). All solutions with GA and WPI were stored for 24 h prior to spray drying, to grant the complete hydration of the carrier and its homogeneity across the juice (Chranioti et al., 2015). At the opposite, solutions with MD was prepared immediately before processing. Each experiment described in the next sections was conducted on seven groups of samples: one constituted by juice alone, 3 constituted by juice and MD, GA or WPI and 3 where mixtures of MD and GA used in proportions 3:1, 2:1 and 1:1. Each group was constituted by 3 samples. Each of the following analysis was conducted in triplicate per sample, unless differently stated.

2.3. Viscosity of the solutions for spray drying

The viscosity measurements were carried out using a Haake MARS 40 (Thermo Electron GmbH, Karlsruhe, Germany) controlled stress rheometer, equipped with a temperature controller unit type TM-PE-P (Thermo Electron GmbH, Karlsruhe, Germany). A cylinder-rotor CC25 DIN/Ti (diameter 25.08 mm, length 37.60 mm, cone angle 120°) attached to rheometer was used for rheological measurements. Volume of the sample shear rate range and temperature during test were 17 cm^3, 10–100 s^{-1} and 20°C, respectively. All measurements were made in 5 replicates. After measurement, correlation to the appropriate model was made in RheoWin program version 4.62.0003 (Thermo Fisher Scientific). The following models

have been chosen on the basis of literature surveys for carrot juices viscosity (Van Hecke, Nguyen, Clausse, & Lanoisellé, 2012).

Ostwald –de Waele: $\tau = K \cdot \gamma^n$ (1)

Bingham: $\tau - \tau_0 = \eta \cdot \gamma \quad \tau > \tau_0$ (2)

where:

τ – shear stress (Pa); τ_0 – yield stress (Pa); η – viscosity (Pa s), γ – shear rate (s^{-1}); K - consistency coefficient (Pa s^n); n - flow behaviour index (-).

2.4. Spray drying

Drying was carried out in the semi-industrial spray drier LAB S1 (drying tower with diameter of 1000 mm, height of the cylindrical portion of 820 mm and height of conical portion of 1020 mm) (Anhydro, Copenhagen, Denmark). Based on previous experiments carried out on beetroot (Janiszewska, 2014), the following parameters for the drying process were chosen: spray disk speed of 39,000 rpm and raw material flux rate of $0.4 \cdot 10^{-6}$ m^3/s. Drying was carried out with the co-current method and the inlet air temperature 160°C at a constant air flow of 0.055 m^3/s. Outlet air temperature was 90±4°C. Powder was collected in the cyclone, then transferred to glass "twist-off" jars. Dried powders were stored in jars in a dark place.

2.5. Dry matter and water activity of microcapsules

The dry matter content in powders was measured by gravimetrical method. A powder sample of 1 gram was placed in a dryer at 95°C until constant weight. Dry matter content was calculated as ratio of mass after and before drying. Water activity was measured in Rotronic Hygroscop DT (Switzerland) at room temperature.

2.6. Size of microcapsules

The size of the particles was measured by laser diffraction method on Cilas Particle Size Analyzer 1190 (Orléans, France). Measurements were made in liquid dispersion created by using isopropyl alcohol (in which powders do not dissolved). Each time the obscuration was about 19-20%. This test was performed in 20 replicates. The diameter of particles was expressed as d50, the value dividing the particles in two groups equally populated. Pictures of microcapsules presented in supplementary material were made on a desktop scanning electron microscope, Hitachi TM3000 (Hitachi High-Technologies Corporation, Tokyo, Japan) operated at an accelerating voltage of 10 kV.

2.7. Hygroscopic properties of microcapsules

In order to investigate the hygroscopic properties, batches of 1 g of powder were placed in a desiccator filled with H_2O at 20°C (a_w=1). Adsorption was analysed for 50 h, determining the mass of the sample after 1, 3, 4, 5, 7, 24 and 50 h.

According to Janiszewska *et al.* (Janiszewska, Witrowa-Rajchert, Kidoń, & Czapski, 2013), changes in hygroscopic properties of powders were described by the kinetic equation:

$$\frac{m_{H_2O}}{m_{d.m.}} = a + b\left(1 - \frac{1}{1 + bc\,\tau}\right) \qquad (3)$$

where a, b and c are the constant coefficients of the equation 3, τ is the time expressed in hours, m_{H2O} is the water increment in the sample (g) and $m_{d.m.}$ is the mass of dry substance (g).

2.8. Tg of microcapsules by means of DSC

The glass transition temperature of the powders was determined using a differential scanning calorimeter (model Pyris 6 DSC, Perkin Elmer Corporation, Wellesley, USA). The DSC was equipped with a low temperature cooling unit Intracooler II (Perkin-Elmer Corporation, Wellesley, USA). The equipment was calibrated with indium ($T_{melting}$ 156.6°C) and tin ($T_{melting}$

231.88°C). Approximately 8-10 mg of carrot powders or juice were placed into aluminium pans (50 mL), hermetically sealed and placed in DSC chamber. Dry nitrogen was used as the purge gas (20 mL/mim). Powder samples were stabilized at -10°C for 5 min, then heated at 10°C/min to 130°C and then cooled to 25°C at 50°C/min. Juice samples were stabilized at -10°C for 5 min, then heated at 10°C/min to 80°C and then cooled to 25°C at 50°C/min. An empty pan was used as a reference.

2.9. Tg of microcapsules by means of TD-NMR

About 1 g of each spray-dried sample was loaded in a 10-mm outer diameter NMR tube that was subsequently sealed. On each sample, a free induction decay (FID) was acquired using a one-pulse sequence implemented in a Bruker 'The Minispec' NMR spectrometer (Bruker Corporation, Germany), operating at 20 MHz. The acquisition parameters included 8 µs of initial dead time, the acquisition of 16 scans of 1 ms each, a dwell time of 0.4 µs and 1 s of recycle delay between the scans. The decay curves were registered at temperatures between 5 and 72 °C, monitored by means of a thermocouple (National Instruments, Italy). The obtained signals were fitted, by means of Levenberg-Marquardt algorithm implemented in 'R' software (R Foundation for Statistical Computing, Austria), to the following equation, which includes the so-called Abragamian function and two exponential contributions (Buitink, van den Dries, Hoekstra, Alberda, & Hemminga, 2000; Kumagai, MacNaughtan, Farhat, & Mitchell, 2002; Van Den Dries et al., 2000):

$$S_{(t)} = A_1 \exp\left(\frac{-a^2 t^2}{2}\right) \frac{\sin bt}{bt} + A_2 \exp\left(\frac{-t}{T_{2,s}}\right) + A_3 \exp\left(\frac{-t}{T_{2,m}}\right) \quad (4)$$

The two exponential decays appearing as second and third elements of this equation describe the relaxation of the solid-like (s) and mobile-like (m) protons, with T_2 being their spin-spin relaxation time. The gaussian curve appearing in the first addend describes the behavior of the

protons pertaining to the most rigid molecules of the system, with *a* and *b* being respectively the standard deviation of the line and its half-width, assumed to be rectangular. According to Abraham(Abraham, Fisher, & Loftus, 1988) from a and b it is possible to calculate the second moment of the rigid protons (M_2), which gives an insight into the strength of the dipolar interactions, according to the equation

$$M_2 = a^2 + \frac{b^2}{3} \quad (5)$$

Examples of raw and fitted FID data at two different temperatures, together with M2 shown for each temperature, are displayed in figure 1. The second moment M_2 appears as linearly and inversely proportional to the increase of temperature both above and below the Tg, but the slope of the linear relationship is markedly different between the two cases (Kumagai et al., 2002). In order to accurately calculate the Tg, two distinct linear models were therefore built, so that their intersection identified the glass transition temperature (van den Dries, van Dusschoten, & Hemminga, 1998).

2.10. Quantification of carotenoids in the microcapsules

Determination of total carotenoids was made according to the methodology reported by (Scott, 2001). One gram of juice or 0.5 g of microencapsulated carrot powder were weighted into the centrifuge tubes. 20 mL of distilled water were added to the sample, followed by the addition of 1 mL of Carrez I and II, to deproteinize the sample. After centrifugation (2000g, 5 min.). 20 mL of acetone were added to the sediment and once more centrifuged. Decanted yellow acetone solution was placed into a funnel for liquid phase separation. The procedure was repeated until a transparent solution of acetone was obtained. 25 mL of petroleum ether were added to the funnel. The petroleum ether phase was centrifuged. The solution was then placed into a 50 mL flask. Absorbance was measured in a spectrophotometer Helios Gamma

(Thermo Spectronic, Cambridge, UK), with petroleum ether as a blank sample at 450 nm wavelength.

Carotenoid content (expressed as β-carotene) was calculated from the following formula:

$$\rho(C_{40}H_{56}) = \frac{A_{450} \cdot 4.00 \cdot \left(\frac{V}{m}\right)}{d.m.} \quad (6)$$

where:

$\rho(C_{40}H_{56})$ – total carotenoids content (mg/kg d.m.); A_{450} – absorbance; 4.00 – average conversion factor, based on a ring test taking into account the average rate of absorption of β-carotene in petroleum ether and dilution made during the analysis; m – mass of the sample (g); V - volume of the graduated flask (50 mL); d.m. – dry matter.

2.11. Encapsulation efficiency

Encapsulation efficiency was calculated from the equation:

$$EE = \frac{\rho(C_{40}H_{56})_p}{\rho(C_{40}H_{56})_j} \cdot 100 \quad (7)$$

where:

indexes p - powder, j - juice.

2.12. Statistical methodology

Data are presented as mean ± standard deviation. Significance of inter-group differences was determined by one-way analysis of variance (ANOVA), calculated with Statistica v12.5 software. Individual group differences were identified using the Tukey (HSD) multiple range test at a significance level of 0.05. Coefficients of determination (R^2) and root mean square errors (RMSE) were used in this study to determine the goodness of fit. Fitting viscosity and higroscopicity to equations was made by means of Table Curve 2D v 5.01 (SYSTAT Software Inc.).

3. Results and discussion

3.1. Apparent viscosity of carrot juice and its solutions employed for microcapsules formation

Figure 2 shows the relationship between apparent viscosity and shear rate for one sample per type observed in the present work. Both carrot juice alone and its mixtures with carriers appeared as pseudoplastic fluids, because characterized by a viscosity inversely related to shear rate (shear thinning behaviour).

Such behaviour was expected because typical of multiphase materials, as vegetable and fruit juices, which are constituted by a dispersion of insoluble components (materials of cellular walls) in a water solution (Augusto, Cristianini, & Ibarz, 2012). The resulting flow curves were therefore investigated in parallel by means of the power equation of Ostwald-de Waele and to Bingham model (Tab. 1). Hypothesis which can explain the behavior of the pseudoplastic liquid assumes that increasing shear rate causes a gradual ordering of asymmetric particles. In this way, the fluid's microstructure, created by macromolecule entanglement or particle–particle interaction, gets broken down under shear. Apparent viscosity begins to decrease with increasing shear rate until molecules start behaving like solids (Muthukumarappan, Tiwari, O'Donnell, & Cullen, 2016).

Although the two models highlighted similar trends, Ostwald-de Waele equation showed a better fit to the experimental data, as it can be deduced by r^2 values, reported in table 1. According to such equation, the viscosity of juice alone was the lowest (69.8 mPa·s at share rate 100 s^{-1}), while the addition of carriers caused a significant increase. The highest viscosity values of juice-carrier mixtures were observed for carrot juice with gum arabic (117.3 mPa·s at share rate 100 s^{-1}). This behaviour could be related to the GA molecular weight. Moreover, branched structure of gum arabic in solutions can determine a small hydrodynamic volume, what leads to creation of viscous GA solutions at high concentrations (Montenegro,

Borsarelli, Valle, & Boiero, 2012). Focusing on the samples where MD and GA were mixed in different proportions, viscosity was found to be proportional to gum arabic concentration, probably because of its emulsifying properties. Indeed, samples with the highest and other two with lower concentration demonstrated a significant difference in viscosity (Table. 1). Maltodextrin alone led to an increase of viscosity of juice, even if at a lower extent than gum arabic, probably because it is not characterized by a branched structure-and is less hydrophilic (Islam, Phillips, Sljivo, Snowden, & Williams, 1997).

Focusing on flow behaviour index *n* from Ostwald-de Waele equation, it can be seen that it increases with the addition of carriers and with the substitution of MD with GA (Tab. 1). However, consistency index could not be directly compared in that form, because it is expressed as Pa multiplied by s^n, with the coefficient n peculiar for each tested sample. An average value for *n* was therefore considered, equal to 0.3446, in order to obtain a new, and comparable, consistency indexes *K*. Juice alone was characterized by the lowest *K* value, in connection to its low content of soluble substances (10^oBx). Addition of carriers and also substitution of MD with GA as carrier material caused a statistically significant increase of K. This phenomenon can be linked to the suitability of GA for stable emulsions, which can be more viscous than solutions with other carrier materials (Kandansamy & Somasundaram, 2012; Muthukumarappan et al., 2016; Özkan & Ersus, 2014).

3.2. Physical properties of carrot juice microcapsules

All obtained powders presented a dry matter content in the 98-99% (Tab. 2), a positive characteristic in sight of long storage times of this kind of products (da Silva Carvalho et al., 2016). The lowest values of dry matter were registered for microcapsules with MD, in connection to the high binding properties of this carrier (Fazaeli et al., 2012). Similar results have been found for spray dried blueberry extracts (Ferrari, Germer, & de Aguirre, 2012;

Ferrari, Marconi Germer, Alvim, & de Aguirre, 2013) and black mulberry powders (Fazaeli et al., 2012). Even if GA samples showed a significantly higher dry matter content than those with MD, their mixes showed an irregular behaviour. Such finding could be related by the active role played by the juice to which the carrier is added to, as the contrasting findings described in the literature allow deducing (da Silva Carvalho et al., 2016; Ferrari et al., 2013).

Water activity of a food system highlights the fraction of total moisture that is available for biochemical reactions and bacterial growth (da Silva Carvalho et al., 2016). In a low water activity environment (aw=0.00-0.25) limited lipid oxidation, low enzyme activity and beginning of browning reactions are expected. With water activity in the 0.3-0.5 region, powder material tends to collapse, giving rise to caking, while growth of moulds, yeast and bacteria are expected in a_w above 0.6 (Labuza & Altunakar, 2007).

The values of water activity in all tested powders were below 0.26, ensuring a high microbiological stability (Tab. 2). Change of carrier material form MD to GA or WPI caused significant decreases in water activity, in correspondence to the values of dry matter (Labuza & Altunakar, 2007; Tapia, Alzamora, & Chirife, 2008).

3.3. Tg of the microcapsules

A parameter related to water activity is the glass transition temperature (Tg), because powders with low a_w show generally high Tg values. In particular, Roos and Karel (Roos & Karel, 1991) observed a Tg value of 84°C in maltodextrin DE=10 samples kept in surroundings with $a_w = 0.23$, while they observed a Tg of 38°C when water activity raised to 0.52. Tg values depend also on the carrier material nature. Indeed for maltodextrin, gum arabic and their mixture MD:GA=2:1 the Tg was 60°C at $a_w = 0.2$ (Ramoneda, Ponce‐Cevallos, Buera, & Elizalde, 2011), while for WPI, at the same a_w of 0.2, Tg raised to 90°C (Schuck et al., 2005).

In high water activity conditions powder absorbs water, causing the decrease of Tg value, as Roos and Karel observed (Roos & Karel, 1991). In powders with high a_w and low Tg some undesirable processes can take place like caking, crystallization or stickiness of powders. This occurrence can result in structure changes, such as agglomeration into hard pieces or even solidification of all powder.

In the present investigation, the highest values of Tg were observed for GA powders (45.9°C/41.6°C obtained by DSC and NMR respectively), with Tg proportional to GA content in case of mixes with MD (Tab. 2).

The highest Tg value for carrot juice powders based on GA can be related to the carrier agent itself as its molecular weight, branching structure and also its possibility of mixing with all components from carrot juice before spray drying. However da Silva Carvalho *et al.* (2016) have stated that usually Tg of obtained capsules is similar to Tg of carrier material used and only small interactions between components take place.

The lowest values were obtained for carrot juice powders based on the WPI (34.9°C/28.3°C obtained by DSC and NMR respectively), independently from the method employed for testing (Tab. 2). This behavior is related to the water content of the powders and is in agreement with the results obtained by Ramoneda *et al.* (2011) and Schuck *et al.* (2005). It is of interest to notice that the same trend was observed for other matrices with similar water content (da Silva Carvalho et al., 2016; Ferrari et al., 2013), but these showed Tg values systematically higher than those reported in the present investigation, witnessing that the substance to which the carrier is added play an active role in this respect.

It is also worth remarking that Tg estimated by TD-NMR was systematically lower than the one estimated by DSC. In connection to TD-NMR ability to detect mobile water even when it has no translational mobility according to DSC analysis (Rocculi et al., 2011).

3.4. Size of microcapsules

Microcapsules with the widest diameter were obtained when WPI was used as a carrier material, in connection to the structure of the particles obtained after microencapsulation process (Fig. S2). Particles with the lowest diameter were obtained for powders based on mixture of carrot juice with carrier MD:GA=3:1. Replacing MD with GA as carrier material caused a statistically significant increase in particles diameter, with the exception of the mixture MD:GA=3:1 (Tab. 2). These results are in agreement with previous investigations (da Silva Carvalho et al., 2016; Fazaeli et al., 2012) on similar matrices. Interestingly, the results presented here seem more clear-cut than those from such works, possibly because the present experimental conditions avoided any cacking phenomena hindering clear observations.

3.5. Hygroscopicity of the microcapsules

Hygroscopicity was tested on the investigated carrot juice microparticles in order to gain insight about their physical stability in case of storage at very severe conditions, represented by water activity close to unity.

Kinetics of vapour adsorption are shown in Figure S1, by means of relative increment in water mass as a function of time. Carrot juice powders with WPI absorbed vapour at the highest rate, so that after 50 h they adsorbed 0.49 g H_2O/g d.m. The slowest rate was registered for carrot juice powders with maltodextrin, which absorbed in 50 h 0.33 g H_2O/g d.m.

According to kinetic equation (3), relative water mass increments at equilibrium were calculated (Tab. 3), on the basis that a long time for water uptake is positively related to a high stability of powders. The highest equilibrium values were observed for powders based on WPI, followed by those based on GA, with powders based on MD showing the lowest values. In the microcapsules where GA progressively substituted MD as a carrier, the time needed to

reach equilibrium appeared proportional to the GA employed, with the exception of mixture MD:GA=1:1 only. According to Bazaria and Kumar (Bazaria & Kumar, 2016), such order could be mainly determined by the particles moisture content, because high water concentration gradient between the product and the surrounding air favors a high hygroscopicity.

3.6. Quantification of carotenoids and microencapsulation process efficiency

In the carrots used to obtain the juice the carotene content was 1443±86 mg/(kg d.m.), while in the juice itself it was 860±11 mg/(kg d.m.).

Microcapsules based on WPI (Fig. 3a) showed the highest carotene concentration (573 mg/kg d.m.), statistically higher than those based on MD (423 mg/kg d.m.), characterized by the lowest concentration, with microcapsules based on GA showing intermediate values, proportional to the level of substitution of MD with GA. Such trend paralleled the one highlighted for microcapsules diameter, suggesting that the higher volume/surface ratio shown by particles with wider diameter played a key role. In this respect, it is worth noticing that viscosity of the fluid undergoing spray drying acts as a confounding factor, as it is known that viscosity modulates the formation of semi-permeable barriers across the fluid, hindering the internal circulations of active material within droplets (Jafari, Assadpoor, He, & Bhandari, 2008; Rajabi, Ghorbani, Jafari, Mahoonak, & Rajabzadeh, 2015).

The efficiency of encapsulation of carotenoids by different carrier types (Fig. 3b) showed the same trends of carotenoids content, with the lowest value (48%) for MD and the highest value for WPI (66%).

4. Conclusion

The results obtained in this study show that the powders with GA used as a carrier material resulted more adequate for microencapsulation of carrot juice, in terms of better carotenoids retention and higher stability of powders. In fact, concerning powder stability, a higher dry matter content and Tg values, lower water activity and better hygroscopic properties were observed for GA samples compared to the MD ones. Carrot powders with WPI presented very similar properties to those with GA, even if the Tg of these samples was the lowest, suggesting that these samples may undergo caking phenomena during storage. The mixture of MD and GA could also offer a good potential for protection of microencapsulated carotenoids when stored at room temperature, at water activity lower than 1.

5. Acknowledgments

The study has been funded by Scholarship of Warsaw University of Life Sciences. The work has been co-financed also by a statutory activity subsidy from the Polish Ministry of Science and Higher Education for the Faculty of Food Sciences of Warsaw University of Life Sciences.

6. References

Abraham, R. J., Fisher, J., & Loftus, P. (1988). *Introduction to NMR spectroscopy*. Chichester, UK: Wiley.

Afifi, H. S., Abu Shelaibi, A., Laleye, L., & Ismail, I. (2009). Measurement of thermal characteristics of spray-dried milk and juice blend. *International journal of food sciences and nutrition, 60*(sup7), 89-98.

Augusto, P. E., Cristianini, M., & Ibarz, A. (2012). Effect of temperature on dynamic and steady-state shear rheological properties of siriguela (Spondias purpurea L.) pulp. *Journal of Food Engineering, 108*(2), 283-289.

Bayram, O. A., Bayram, M., & Tekin, A. R. (2008). Whey powder as a carrier in spray drying of sumac concentrate. *Journal of food process engineering, 31*(1), 105-119.

Bazaria, B., & Kumar, P. (2016). Effect of whey protein concentrate as drying aid and drying parameters on physicochemical and functional properties of spray dried beetroot juice concentrate. *Food Bioscience, 14*, 21-27.

Beristain, C. I., Azuara, E., Tamayo, T. T., & Vernon□Carter, E. J. (2003). Effect of caking and stickiness on the retention of spray□dried encapsulated orange peel oil. *Journal of the Science of Food and Agriculture, 83*(15), 1613-1616.

Buitink, J., van den Dries, I. J., Hoekstra, F. A., Alberda, M., & Hemminga, M. A. (2000). High critical temperature above T g may contribute to the stability of biological systems. *Biophysical Journal, 79*(2), 1119-1128.

Chranioti, C., Nikoloudaki, A., & Tzia, C. (2015). Saffron and beetroot extracts encapsulated in maltodextrin, gum Arabic, modified starch and chitosan: Incorporation in a chewing gum system. *Carbohydrate polymers, 127*, 252-263.

da Silva Carvalho, A. G., da Costa Machado, M. T., da Silva, V. M., Sartoratto, A., Rodrigues, R. A. F., & Hubinger, M. D. (2016). Physical properties and morphology of spray dried microparticles containing anthocyanins of jussara (Euterpe edulis Martius) extract. *Powder Technology, 294*, 421-428.

Domian, E., Brynda-Kopytowska, A., Cenkier, J., & Świrydow, E. (2015). Selected properties of microencapsulated oil powders with commercial preparations of maize OSA starch and trehalose. *Journal of Food Engineering, 152*, 72-84.

Fazaeli, M., Emam-Djomeh, Z., Ashtari, A. K., & Omid, M. (2012). Effect of spray drying conditions and feed composition on the physical properties of black mulberry juice powder. *Food and bioproducts processing, 90*(4), 667-675.

Ferrari, C. C., Germer, S. P. M., & de Aguirre, J. M. (2012). Effects of spray-drying conditions on the physicochemical properties of blackberry powder. *Drying Technology, 30*(2), 154-163.

Ferrari, C. C., Marconi Germer, S. P., Alvim, I. D., & de Aguirre, J. M. (2013). Storage stability of spray-dried blackberry powder produced with maltodextrin or gum arabic. *Drying Technology, 31*(4), 470-478.

Francis, F. J. (2002). *Food colorings*: CRC Press, Boca Raton, FL.

Gharsallaoui, A., Roudaut, G., Chambin, O., Voilley, A., & Saurel, R. (2007). Applications of spray-drying in microencapsulation of food ingredients: An overview. *Food Research International, 40*(9), 1107-1121.

Islam, A., Phillips, G., Sljivo, A., Snowden, M., & Williams, P. (1997). A review of recent developments on the regulatory, structural and functional aspects of gum arabic. *Food Hydrocolloids, 11*(4), 493-505.

Jafari, S. M., Assadpoor, E., He, Y., & Bhandari, B. (2008). Encapsulation efficiency of food flavours and oils during spray drying. *Drying Technology, 26*(7), 816-835.

Janiszewska, E. (2014). Microencapsulated beetroot juice as a potential source of betalain. *Powder Technology, 264*, 190-196.

Janiszewska, E., Witrowa-Rajchert, D., Kidoń, M., & Czapski, J. (2013). Effect of the applied drying method on the physical properties of purple carrot pomace. *International Agrophysics, 27*(2), 143-149.

Kandansamy, K., & Somasundaram, P. D. (2012). Microencapsulation of colors by spray drying-a review. *International Journal of Food Engineering, 8*(2).

Kumagai, H., MacNaughtan, W., Farhat, I. A., & Mitchell, J. R. (2002). The influence of carrageenan on molecular mobility in low moisture amorphous sugars. *Carbohydrate Polymers, 48*(4), 341-349.

Labuza, T. P., & Altunakar, B. (2007). Water Activity Prediction and Moisture Sorption Isotherms. In *Water activity in foods: fundamentals and applications* (pp. 109-154). Oxford, UK: Blackwell Publishing Ltd.

Ma, T., Tian, C., Luo, J., Sun, X., Quan, M., Zheng, C., & Zhan, J. (2015). Influence of technical processing units on the α-carotene, β-carotene and lutein contents of carrot (Daucus carrot L.) juice. *Journal of Functional Foods, 16*, 104-113.

Mahdavi, S. A., Jafari, S. M., Ghorbani, M., & Assadpoor, E. (2014). Spray-drying microencapsulation of anthocyanins by natural biopolymers: A review. *Drying technology, 32*(5), 509-518.

Montenegro, M. A., Borsarelli, C. D., Valle, L., & Boiero, M. L. (2012). *Gum Arabic: more than an edible emulsifier*: INTECH Open Access Publisher.

Muthukumarappan, K., Tiwari, B., O'Donnell, C. P., & Cullen, P. (2016). Ultrasound Processing: Rheological and Functional Properties of Food. In J. Ahmed, H. Ramaswamy, S. Kasapis, & J. & Boye (Eds.), *Novel Food Processing: Effects on Rheological and Functional Properties* (pp. 85-102): CRC Press.

Paterson, A. H., & Bröckel, U. (2015). Caking Development in Lemon Juice Powder. *Procedia Engineering, 102*, 142-149.

Rajabi, H., Ghorbani, M., Jafari, S. M., Mahoonak, A. S., & Rajabzadeh, G. (2015). Retention of saffron bioactive components by spray drying encapsulation using maltodextrin, gum Arabic and gelatin as wall materials. *Food Hydrocolloids, 51*, 327-337.

Ramoneda, X. A., Ponce□Cevallos, P. A., Buera, M. d. P., & Elizalde, B. E. (2011). Degradation of β□carotene in amorphous polymer matrices. Effect of water sorption properties and physical state. *Journal of the Science of Food and Agriculture, 91*(14), 2587-2593.

Righetto, A. M., & Netto, F. M. (2006). Vitamin C stability in encapsulated green West Indian cherry juice and in encapsulated synthetic ascorbic acid. *Journal of the Science of Food and Agriculture, 86*(8), 1202-1208.

Rocculi, P., Sacchetti, G., Venturi, L., Cremonini, M., Dalla Rosa, M., & Pittia, P. (2011). Role of water state and mobility on the antiplasticization of green and roasted coffee beans. *Journal of agricultural and food chemistry, 59*(15), 8265-8271.

Roos, Y., & Karel, M. (1991). Phase transitions of mixtures of amorphous polysaccharides and sugars. *Biotechnology Progress, 7*(1), 49-53.

Samborska, K., Langa, E., Kamińska□Dwórznicka, A., & Witrowa□Rajchert, D. (2015). The influence of sodium caseinate on the physical properties of spray□dried honey. *International Journal of Food Science & Technology, 50*(1), 256-262.

Schuck, P., Blanchard, E., Dolivet, A., Méjean, S., Onillon, E., & Jeantet, R. (2005). Water activity and glass transition in dairy ingredients. *Le Lait, 85*(4-5), 295-304.

Scott, K. J. (2001). Detection and measurement of carotenoids by UV/VIS spectrophotometry. *Current protocols in food analytical chemistry*.

Tapia, M. S., Alzamora, S. M., & Chirife, J. (2008). Effects of Water Activity (aw) on Microbial Stability: As a Hurdle in Food Preservation. In A. Fontana Jr, S. Schmidt, & T. Labuza (Eds.), *Water Activity in Foods: Fundamentals and Applications* (Vol. 13, pp. 239-271). Oxford, UK: Blackwell Publishing Ltd.

van den Dries, I. J., van Dusschoten, D., & Hemminga, M. A. (1998). Mobility in maltose-water glasses studied with 1H NMR. *The Journal of Physical Chemistry B, 102*(51), 10483-10489.

Van Den Dries, I. J., Van Dusschoten, D., Hemminga, M. A., & Van Der Linden, E. (2000). Effects of water content and molecular weight on spin probe and water mobility in malto-oligomer glasses. *The Journal of Physical Chemistry B, 104*(44), 10126-10132.

Van Hecke, E., Nguyen, P. U., Clausse, D., & Lanoisellé, J. L. (2012). Flow behaviour of carrot puree: modelling the influence of time, temperature and potato flakes addition. *International Journal of Food Science & Technology, 47*(1), 177-185.

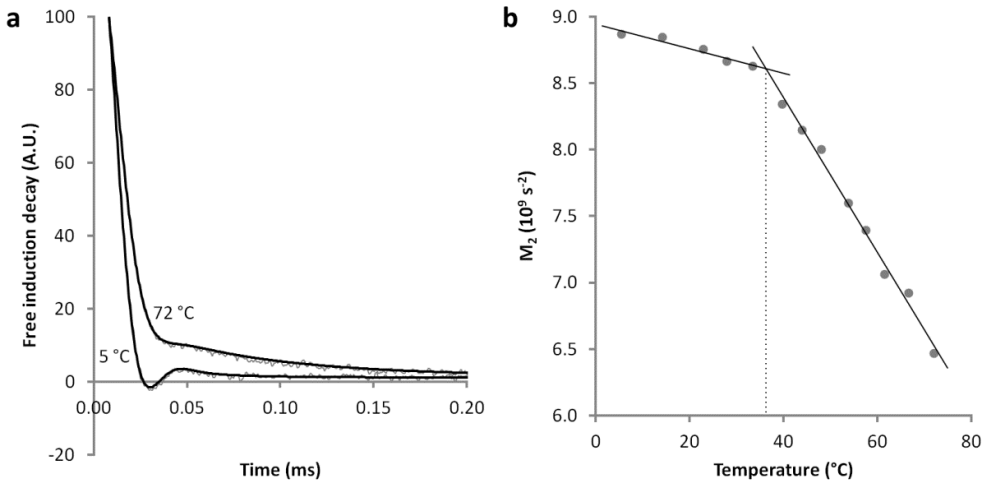
Özkan, G., & Ersus, S. (2014). Microencapsulation of natural food colorants. *International Journal of Nutrition and Food Science, 3*(3), 145-156.

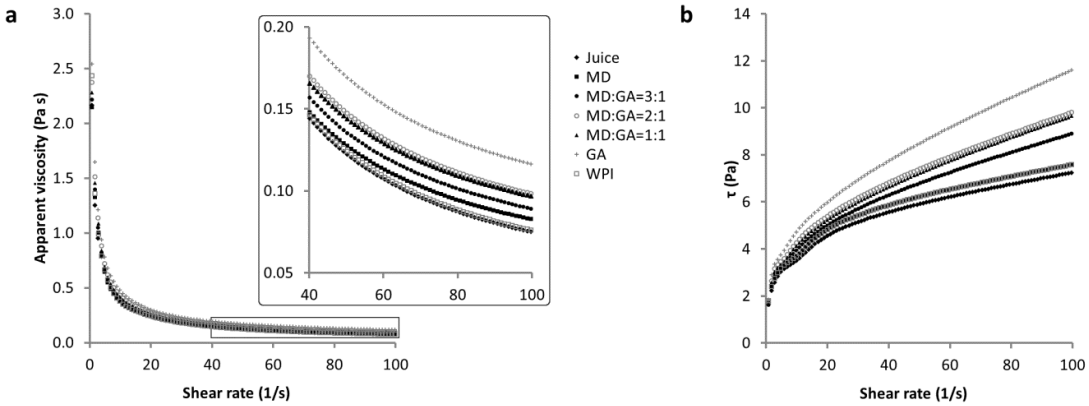
Figures captions

Figure 1. (a) FID data registered at two different temperatures (gray lines) by TD-NMR on one of the samples added with MD, fit by means of the model highlighted by equation 4 (black bold line); (b) for the same sample, M2 values, shown for each of the temperature values applied around Tg.

Figure 2. Effect of the carrot juice solutions type on its (a) apparent viscosity and (b) flow curve

Figure 3. Carotene percentage in carrot juice microcapsules. Mean values denoted with different letters differ statistically ($p<0.05$).





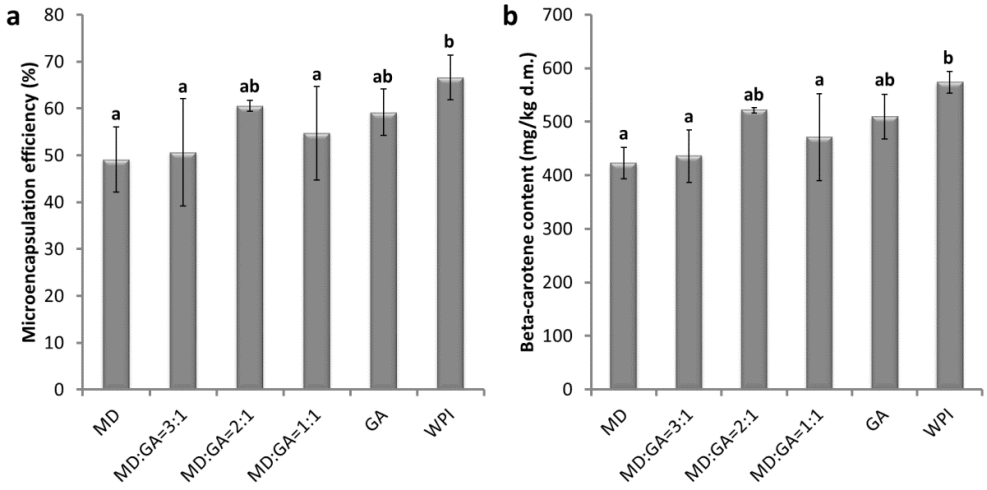


Table 1. Rheological properties of carrot juice and its solution

Tested juice or its solution	Tested apparent viscosity at share rate 100 s^{-1} (mPa s)	Bingham model			Ostwald de Waele Model				
		τ_0 yield stress (Pa);	η viscosity (mPa s),	r^2	Consistency coefficient K (Pa s^n)	Flow behaviour index n (-)	r^2	Calculated consistency coefficient K for average n=0.3446 (Pa s^n)	r^2 for calculation
juice	69.8±0.2a	3.51	45	0.923	2.0	0.288	0.999	1.46	0.978

juice with MD	82.1±0.7c	3.38	54	0.952	1.7	0.333	0.999	1.67	0.997
juice with MD:GA=3:1	89.2±1.1d	3.50	59	0.957	1.7	0.346	0.999	1.79	0.997
juice with MD:GA=2:1	98.3±0.5e	3.74	66	0.961	1.9	0.359	0.998	1.95	0.996
juice with MD:GA=1:1	99.1±2.0e	3.59	66	0.964	1.8	0.363	0.998	1.98	0.995
juice with GA	117.3±1.3f	4.02	82	0.970	1.9	0.392	0.998	2.25	0.987
juice with WPI	75.5±0.7b	3.55	46	0.924	1.9	0.298	0.999	1.58	0.986

Mean values in the same column denoted with different letters differ statistically at p=0.05

Table 2. Selected physical and thermal properties of powders

Tested powder	Dry matter (%) Xav±SD	Water activity (–) Xav±SD	d_{50} (μm) Xav±SD	Tg (DSC) [°C] Xav±SD	Tg (TD-NMR) (°C) Xav±SD
MD	98.23±0.09a	0.246±0.001c	28.26±0.21b	42.66±0.42ab	35.39±2.36abc
MD:GA=3:1	98.96±0.01b	0.252±0.003c	23.66±0.08a	42.78±2.96ab	31.42±1.42ab
MD:GA=2:1	98.50±0.06a	0.237±0.001b	30.87±0.19c	42.74±4.83ab	33.43±1.16ab
MD:GA=1:1	98.92±0.1b	0.236±0.001b	31.37±0.23d	43.31±2.12b	38.08±1.26bc
GA	99.03±0.1b	0.223±0.001a	32.89±0.17e	45.91±0.51b	41.58±1.09c
WPI	99.01±0.2b	0.232±0.003b	43.46±0.14f	34.92±4.36a	28.25±6.36a

Mean values in the same column denoted with different letters: a, b, c differ statistically at p=0.05.

Table 3. Coefficients of Eq. (3) describing kinetics of changes in relative mass increments during water sorption of carrot microcapsules

Tested powder	Coefficients of equation			r^2	RMSE	Equilibrium
	a	b	c		[%]	value
MD	0.0035	0.4102	0.1784	0.991	1.950	0.4137
MD:GA=3:1	0.0084	0.4391	0.1735	0.990	4.300	0.4475
MD:GA=2:1	0.0082	0.5843	0.1065	0.983	1.822	0.5925
MD:GA=1:1	0.0374	0.5208	0.1220	0.991	2.472	0.5582
GA	0.0041	0.5924	0.1251	0.988	1.852	0.5965
WPI	0.0146	0.6295	0.0983	0.975	4.528	0.6441

Microencapsulated carrot juices has low water activity

The carrier influences the amount of carotenoids microencapsulated

Better maintenance of carotenoids in GA and WPI powder samples was observed

Tg of microencapsulated carrot juice can be evaluated by DSC or TD-NMR