**Oxidative stability of vegetable purees enriched with n-3-LC-PUFA microalgal biomass:
impact of type of vegetable**

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Abstract

Vegetable purees can be interesting food products to enrich with omega-3 long chain poly-unsaturated fatty acids (n-3-LC-PUFA), since they are important carriers of nutrients and fibers and essential in a healthy diet. Due to their large number of double bonds, n-3-LC-PUFA are highly susceptible to lipid oxidation. Different vegetable purees, containing different types and amounts of endogenous antioxidants, may possibly show differences in oxidative stability. This study contributes to obtain more insight in the impact of the type of vegetable (broccoli, carrot or tomato) on the oxidative stability of purees enriched with heterotrophic (*Schizochytrium*) n-3-LC-PUFA rich microalgal biomass. Therefore, different vegetable purees enriched with microalgae were compared with each other and analyzed for their amount lipid oxidation products and antioxidants. Broccoli and tomato puree enriched with *Schizochytrium* showed a higher oxidative stability compared to enriched carrot puree. However, all of these *Schizochytrium* enriched purees were prone to oxidation.

Keywords

Microalgae, antioxidant, processing, omega-3, vegetable puree, oxidation

Abbreviations

α TEAC	α -tocopherol equivalent antioxidant capacity
DHA	docosahexaenoic acid
EPA	eicosapentaenoic acid
FRAP	ferric reducing antioxidant power
LPSC	luminol-chemiluminescence based peroxyl radical scavenging assay
n-3-LC-PUFA	omega-3 long chain poly unsaturated fatty acids

1. Introduction

Omega-3 long chain poly-unsaturated fatty acids (n-3-LC-PUFA), mainly eicosapentaenoic acid (EPA, 20:5 n-3) and docosahexaenoic acid (DHA, 22:6 n-3), are associated with beneficial effects in the prevention of cardiovascular disorders and the development of a normal brain function (Calder, 2014). The Food and Agriculture Organization of the United Nations recommends a daily intake of 250 mg n-3-LC-PUFA (FAO, 2010), although this amount is often not reached in a Western diet. In this respect, there is an increased interest to enrich food products with n-3-LC-PUFA as no change in eating habits is needed to enhance the intake.

Fish is the most well-known source of n-3-LC-PUFA, although it is under pressure due to a reducing stock and the accumulation of toxic compounds (PCB's, dioxins, lead,...) (Walker *et al.*, 2015). Heterotrophic microalgae have already shown their potential as an alternative source of n-3-LC-PUFA (Chacón-Lee and González-Mariño, 2010). Moreover, vegetarian diets often lack n-3-LC-PUFA which makes heterotrophic microalgae a unique vegetarian (and even vegan) source of n-3-LC-PUFA (Ganesan *et al.*, 2014).

n-3-LC-PUFA are susceptible to lipid oxidation due to their large number of allylic hydrogens. Oxidation of n-3-LC-PUFA results in loss of nutritional value, undesired flavor and the presence of toxic compounds and should thus be avoided (Frankel, 2005). The presence of (natural) antioxidants in the food matrix can help to protect the fatty acids from oxidation (Jacobsen, 2010). In this respect, fruit and vegetable-based products (for example soups, sauces, smoothies and juices) are an interesting matrix given their relatively high endogenous antioxidant capacity. Different vegetable purees, differing in type and amount of endogenous antioxidants, may however show a different oxidative stability when enriched with n-3-LC-PUFA, using heterotrophic microalgal biomass. Moreover, fruit and vegetable-based products are essential in a healthy diet and also contain other high-value compounds, like micronutrients (Darmon *et al.*, 2005).

Carotenoids, phenolic compounds and vitamin C are the main endogenous antioxidants in vegetables (Kaur and Kapoor, 2001). A combined effect of all these compounds determines the total antioxidant capacity (Wang *et al.*, 2011). A few studies determined the total antioxidant capacity of vegetables by measuring, amongst others, oxygen radical absorbance capacity (ORAC), ferric reducing antioxidant power (FRAP) and trolox equivalent antioxidant capacity (TEAC) of different vegetables. It should be mentioned that these methods only give an indication of the antioxidant capacity and the value is highly influenced by the used method, vegetable cultivar and growth conditions. Based on these studies it could be concluded that more commonly used vegetables greatly vary in antioxidant capacity (Ou *et al.*, 2002; Proteggente *et al.*, 2002; Szeto *et al.*, 2002; Bahorun *et al.*, 2004; Zhou and Yu, 2006; Miglio *et al.*, 2008).

The oxidative stability of n-3-LC-PUFA enriched vegetable purees has never been described before. The objective of this study was therefore to obtain more insight into the oxidative stability of different vegetable purees enriched with n-3-LC-PUFA rich heterotrophic microalgal biomass (*Schizochytrium*). The vegetables used in this study (broccoli, carrot and tomato purees) were selected based on their commercial importance (FruitLogistica, 2018) as well as on their differences in endogenous antioxidants (Duthie *et al.*, 2013). The vegetable purees were enriched to reach a concentration of 80 mg n-3-LC-PUFA/100 g enriched puree and characterized for their oxidative stability during 12 weeks of storage at 37°C and amount of carotenoids, phenolic compounds and ascorbic acid. Furthermore, the impact of processing (high-pressure-homogenization, pasteurization or sterilization) was also investigated.

2. Materials and methods

2.1. Microalgal species

Schizochytrium sp. (heterotrophic) was obtained as freeze dried biomass from Bunge (St Just Desvern, Spain) and was stored at -80°C until further use. The composition of *Schizochytrium* biomass in terms of n-3-LC-PUFA (section 2.5.2), free fatty acids (section 2.5.3) and antioxidants (section 2.5.6-8) is shown in Table 1.

2.2. Preparation of vegetable purees

Broccoli, cultivar *Monrello*, and carrots, cultivar *Neria*, were obtained from REO Veiling Roeselare (Roeselare, Belgium). Tomatoes, cultivar *Bonaparte*, were obtained from Proefcentrum Hoogstraten (Hoogstraten, Belgium). Table 2 represents the composition of the vegetable purees in terms of dry matter, pH and antioxidants. The vegetables were cut into slices and blanched (8 min at 95°C) to inactivate endogenous enzymes. The blanched vegetables were diluted with water according to the ratio mentioned in Table 2 and then blended (Büchi mixer B-400, Flawil, Switzerland) and sieved (pore size 1 mm) to remove the larger parts, seeds and fibers. The resulting vegetable puree was mixed into a single batch and stored at -40°C until further use.

2.3. Experimental set-up

The general experimental set-up of this study is shown in Figure 1, which also shows the different steps to prepare the enriched vegetable purees. After each processing step (mixing, high-pressure-homogenization, pasteurization or sterilization), the purees were analyzed for their amount of n-3-LC-PUFA, carotenoids, phenolic compounds and ascorbic acid. Moreover, the (high-pressure-homogenized, pasteurized or sterilized) enriched vegetable purees were stored for 12 weeks at 37°C and analyzed for their amount of primary and secondary lipid oxidation products at 4 predefined time points (week 0, 4, 8 and 12). Based on statistical optimization, a 2*2 experimental set-up was selected, meaning that each enriched vegetable puree was prepared in duplicate and treated independently from its duplicate. Furthermore, each of these duplicates was analyzed twice according to the analyses

described in 2.5. This experimental set-up therefore takes into account the error on the production of the puree as well as the error on the chemical analysis.

2.4. Preparation and processing of enriched vegetable purees

2.4.1. Preparation of enriched vegetable puree

The *Schizochytrium* biomass was added to the thawed vegetable puree in order to reach a concentration of 80 mg n-3-LC-PUFA/100 g puree (0.24 g *Schizochytrium* biomass/100 g puree). At this concentration, the product can be labelled as ‘a product rich in n-3-LC-PUFA’ (European Commission, 2010). This claim can be mentioned if 80 mg EPA+DHA is present in the product per 100 g or per 100 kcal, whichever amount is limiting. The batch was mixed 2 times for 5 minutes at 7500 rpm with a labmixer Silverson L5M-A (East Longmeadow, MA, USA). The batch was then split up in two parts: one part was labelled as ‘mixed’ and the remaining part was further processed (2.4.2.).

2.4.2. Processing and storage

The remaining part of the batch obtained in 2.4.1. was high-pressure-homogenized (1 pass at 100 MPa) at 4°C. This high-pressure-homogenization step was applied to obtain a stable homogenous suspension and not necessarily to disrupt the microalgal cell (wall). The puree was collected in an ice bath. Again, the batch was split up in two parts: one part was labelled as ‘high-pressure-homogenized’ and the remaining part underwent further thermal processing.

Two different thermal treatments were performed. Therefore, the remaining part obtained was divided in a part for **pasteurization** ($10^{\circ}\text{C P}_{90^{\circ}\text{C}} = 3 \text{ min}$) and a part for **sterilization** ($10^{\circ}\text{C F}_{121^{\circ}\text{C}} = 5 \text{ min}$). The mechanical and thermal treatments were performed according to Gheysen *et al.* (2019).

The samples were stored according to Gheysen *et al.* (2019).

2.5. Analyses

2.5.1. Lipid extraction

The lipids were extracted with chloroform/methanol (1/1) as described in Gheysen *et al.* (2018).

2.5.2. Determination of n-3-LC-PUFA content

The n-3-LC-PUFA content was determined by chromatographic separation and FID detection after methylation of the extracted lipids according to the method described in Gheysen *et al.* (2018) and expressed as mg/100 g dry biomass or as mg/100 g (enriched) puree (wet).

2.5.3. Determination of free fatty acid content

The total free fatty acid content was determined by chromatographic separation after the formation of diethylamide derivatives as described in Gheysen *et al.* (2018) and expressed as mg/100 g dry biomass.

2.5.4. Determination of primary oxidation products

The determination of the primary oxidation products was performed according to the method described in Gheysen *et al.* (2019). All data points were normalized (to a reference point of zero at zero weeks) and expressed as mg/100 g enriched puree (wet). This allows to compare the impact of oxidation during storage.

2.5.5. Determination of secondary oxidation products

Volatile secondary oxidation products were analyzed according as described in Gheysen *et al.* (2018). The semi-quantitative concentrations of different selected indicator compounds originating from lipid oxidation were calculated as the area of the volatile indicator component divided by the response factor of the external standard hexanal. All data points were normalized (to a reference point of zero at zero weeks) and expressed as mg/100 g enriched puree (wet). This allows to compare the impact of oxidation during storage.

2.5.6. Determination of carotenoids

The carotenoid content was determined by high-pressure-liquid-chromatography (HPLC) according to the method described in Gheysen *et al.* (2019) and expressed as mg/100 g dry biomass or as mg/100 g (enriched) puree (wet).

2.5.7. Determination of total phenolic content

The determination of the total phenolic content was done with the Folin-Ciocalteu reagent according to the method described in Gheysen *et al.* (2019) and expressed as mg/100 g dry biomass or as mg/100 g (enriched) puree (wet).

2.5.8. Determination of ascorbic acid

The ascorbic acid content was determined by HPLC coupled with fluorescence detection after a derivatization reaction as described in Gheysen *et al.* (2019) and expressed as mg/100 g dry biomass or as mg/100 g (enriched) puree (wet).

2.6. Correction physical losses

The values of all parameters were corrected for physical losses according to Gheysen *et al.* (2019). By this correction, physical losses that occurred during processing, stickiness to glassware, mixer, etc., taken into account.

2.7. Statistical analysis

Results were statistically evaluated by a two way analysis of variance (ANOVA) to consider the impact of vegetable puree and processing. The results of primary and secondary oxidation and the degradation of carotenoids (for each individual carotenoid) were evaluated by one way ANOVA to consider the impact of time for each enriched vegetable puree. The total amount of phenolic compounds and the amount of ascorbic acid was evaluated by a two way ANOVA to consider the impact of treatment and type of vegetable puree. The differences were determined by a *post hoc* Tukey test. All statistical tests were performed with $\alpha = 0.05$ (JMP Pro 14, SAS Institute Inc.).

3. Results and discussion

3.1. Lipid oxidation

The formation of primary and secondary oxidation products during 12 weeks of storage at 37°C is shown in **Table 3**. No significant increase in the peroxide value (primary oxidation products) could be observed in any of the enriched purees, irrespective of processing, except for pasteurized enriched tomato puree at week 4 and pasteurized enriched broccoli puree at week 12. The absence of an increase in the peroxide value can imply that (i) the enriched puree is stable during storage or (ii) the puree is completely oxidized during storage and the peak maximum is located between two measuring points. Despite the fact that in most purees no significant change in peroxide value could be observed, a visual analysis of the graphs of enriched tomato purees showed an increasing trend at week 4 followed by a decrease upon further storage, which indicates highly oxidized purees. A similar trend could be observed in high-pressure-homogenized enriched broccoli puree. Moreover, sterilized enriched broccoli puree contained large error bars, which is more often seen for oxidized purees due to the evolution of a lipid oxidation process. Small time shifts of the oxidation process may lead to larger differences in the peroxide value once the oxidation is started compared to samples at the initiation phase (Frankel, 2005).

The trends observed in the secondary oxidation compounds, being a significant increase at week 4 followed by a stagnation phase, were similar in all enriched vegetable purees. The increase in volatile compounds was significantly more pronounced in the enriched carrot puree. Again, no significant impact of processing was seen in the evolution of the secondary oxidation compounds as a function of time.

Based on the secondary oxidation compounds, which are present due to the decomposition of primary oxidation products, it can thus be concluded that all enriched vegetable purees oxidized during storage. This implies that the peak maximum of the peroxide value was located between two measuring points, which led to the absence of significant differences in the primary oxidation. Despite

the generally recognized antioxidant capacity of vegetable purees, none of them was thus able to maintain the n-3-LC-PUFA derived from *Schizochytrium* oxidatively stable. N-3-LC-PUFA from *Schizochytrium* incorporated in carrot puree were less stable compared to broccoli and tomato puree given the significantly lower amount of volatile compounds in the latter two.

These results are in agreement with the results of Ranawana *et al.* (2016) and Duthie *et al.* (2013), who already showed the higher antioxidant capacity of broccoli and tomato compared to carrot in respectively bread and meat patties. Probably, the lower antioxidant capacity of the carrot puree is the main explanation for the observed differences. To confirm this hypothesis, the main endogenous antioxidants originating from the vegetable purees are discussed in **section 3.2**. Their link with the results of the oxidative stability is discussed in **section 3.3**.

3.2. Antioxidants

3.2.1. Carotenoids

Table 4 (week 0) represent the carotenoids present in the enriched vegetable purees. Since *Schizochytrium* did not contain carotenoids (**Table 1**), the enriched purees only contained carotenoids originating from the vegetable puree itself. Enriched broccoli puree contained lutein (0.5 mg/100 g enriched puree) and β -carotene (0.75 mg/100 g enriched puree), enriched carrot puree contained α -carotene (6.3 mg/100 g enriched puree) and β -carotene (9.5 mg/100 g enriched puree) and enriched tomato puree contained lycopene (5 mg/100 g enriched puree) and β -carotene (0.9 mg/100 g enriched puree). None of the carotenoids was significantly influenced by processing, being in accordance to literature (Hwang *et al.*, 2012; dos Reis *et al.*, 2015; Ma *et al.*, 2015).

3.2.2. Total phenolic compounds

The phenolic compounds present in the enriched purees are shown in **Table 5**. A significant impact of type of vegetable puree could be observed. Enriched broccoli puree contained the highest amount of phenolic compounds (17 mg GAE/100 g enriched puree) followed by enriched tomato puree (14 mg GAE/100 g enriched puree) and followed by enriched carrot puree (4 mg GAE/100 g enriched puree).

The lower concentration in enriched tomato and carrot puree can be attributed to the lower concentrations in the vegetable puree itself, since all purees were enriched with the same amount of *Schizochytrium* (0.24 g/100 g enriched puree) (**Table 1**). The results were in line with the results of Zhou & Yu (2006) who also showed that freeze dried broccoli has a tree times higher total phenolic content compared to freeze dried tomato and a six times higher content compared to freeze dried carrots. In none of the purees, a significant impact of processing was observed which was in accordance with the study of Gheysen *et al.* (2019) and Dewanto *et al.* (2002) where comparable processing steps were executed.

3.2.3. Ascorbic acid

Table 5 shows the amount of ascorbic acid in the enriched vegetable purees. A significant impact of processing and type of vegetable was observed. Thermal processing drastically reduced the amount of ascorbic acid in the enriched purees, irrespective of type of vegetable purees. In enriched carrot puree, no significant difference was observed between pasteurization and sterilization, while in enriched broccoli and tomato puree sterilization led to a supplementary reduction compared to pasteurization. In literature, ascorbic acid was already described as a thermolabile compound (Capanoglu *et al.*, 2010). The differences observed in the initial amount of ascorbic acid can be attributed to the different amounts in the vegetable purees as the ascorbic acid only originated from the vegetable puree itself and not from *Schizochytrium* (**Table 1 and 2**). Enriched broccoli puree contained a significantly higher amount (26 mg ascorbic acid/100 g mixed enriched puree) compared to enriched tomato puree (19 mg ascorbic acid/100 g mixed enriched puree). The lowest amount was observed in enriched carrot puree (4 mg ascorbic acid/100 g mixed enriched puree). These results are in line with the results obtained by Szeto *et al.* (2002).

3.3. Relation between oxidative stability and antioxidants

Large differences in oxidative stability were observed between the enriched broccoli and tomato puree on the one hand and enriched carrot puree on the other hand (**section 3.1**). Incorporation of

heterotrophic microalgae implies the absence of antioxidants originating from the microalga itself. Therefore, the antioxidants derived from the food product strongly affect the oxidative stability of enriched products. In parallel, differences in antioxidants, known for their impact on the oxidative stability, were seen (**section 3.2**).

At first sight, the results of the carotenoids present in the enriched purees seemed contradictory to the observed oxidative stability during storage. However, not only the total amount of carotenoids is crucial in the determination of the oxidative stability. It can be hypothesized that the lower total carotenoid concentration in enriched broccoli and tomato purees was (partially) compensated by the type of carotenoid (Miller *et al.*, 1996). In literature, different studies showed varying results concerning the antioxidant capacity of different carotenoids. All of them relied on fast methods of antioxidative capacity and none of them really monitored lipid oxidation like in this study. Woodall *et al.* (1997) showed a higher antioxidant capacity of lycopene compared to β -carotene based on the Fenton reaction. Müller *et al.* (2011) screened the capacity of carotenoids, among which lutein, β -carotene, α -carotene and lycopene, with different assays, TEAC, FRAP and luminol-chemiluminescence based peroxy radical scavenging assay (LPSC). The used assays, based on a variety of principles, gave different results. Measured by TEAC, lycopene showed a higher potential in scavenging synthetic radicals compared to α -carotene and β -carotene. Lutein showed the lowest potential. Based on the FRAP method, lycopene and lutein showed a higher ferric reducing potential. The LPSC assay in its turn showed a higher peroxy radical scavenging capacity for lutein, followed by α -carotene and β -carotene and followed by lycopene. As the different methods often gave different results, a weighed value, based on the different assays used, was calculated. Based on the latter, a higher antioxidant potential was obtained for lycopene and lutein compared to α -carotene and β -carotene. This supports that not only the total amount of carotenoids should be taken into consideration to make the link with the results of oxidative stability. Moreover, synergistic effects between different carotenoids might have occurred (Choe and Min, 2009).

280 Additionally, the amount of phenolic compounds and ascorbic acid was significantly lower in the
281 enriched carrot puree, which also showed the lowest oxidative stability during storage. This indicates
282 a possible role of these antioxidants in the prevention of lipid oxidation. However, a crucial role of
283 ascorbic acid seems doubtful as almost no significant impact of processing on the oxidative stability
284 was observed while the amount of ascorbic acid was highly influenced by processing.

285 Moreover, pro-oxidative effects of α - and β -carotene at higher concentrations can also possibly explain
286 the lower oxidative stability of enriched carrot puree (Ribeiro *et al.*, 2018). The conditions at which
287 these effects occur are hard to determine as the effect is influenced by environmental factors and
288 sample composition (Shahidi and Ambigaipalan, 2015).

289 Three hypotheses could thus be formulated to explain the higher oxidative stability of broccoli and
290 tomato compared to carrot: (i) the higher antioxidant capacity of lycopene and lutein compared to
291 α - and β -carotene, (ii) the higher concentration of phenolic compounds or (iii) the high concentrations
292 of α - and β -carotene resulting in pro-oxidative effects. A synergistic effect of different antioxidants
293 (Choe and Min, 2009) may be a fourth explanation.

294 The evolution of the carotenoids during storage was investigated (**Tables 4a–c**) in order to obtain more
295 insight in the possibility of hypothesis (i).

296 In general, all carotenoids showed a decrease over time in all enriched purees. This degradation, and
297 thus their antioxidant working, can be explained by the contribution of carotenoids in the reaction
298 with radicals or oxygen. Irrespective of the processing intensity, broccoli purees showed a reduction
299 up to 70% of β -carotene during 12 weeks of storage, while α - and β -carotene of carrot purees were
300 reduced up to 65 and 60%, respectively. Lycopene and β -carotene derived from tomato purees showed
301 a degradation up to 65 and 75% respectively. A significant reduction was mostly observed from week
302 4 of storage. This degradation was also visually observed by a change in color towards more colorless
303 products. No changes in texture could be observed visually, which means that the suspension is stable
304 most likely because of the high-pressure-homogenization treatment. As all carotenoids from the

vegetable purees degraded, their antioxidative effect is proven. Although, it can be doubted that hypothesis (i) is determinative on its own as comparable reductions for the different antioxidants were observed.

Moreover, it can be doubted whether or not a pro-oxidant effect of α -carotene is the primary explanation (hypothesis (iii)). Based on the comparison of this study with the study of Gheysen *et al.* (2018), carrot puree enriched with *Schizochytrium* showed a higher oxidative stability compared to aqueous suspensions enriched with *Schizochytrium*. It thus seems that the absence of strong antioxidants in carrot puree rather than pro-oxidant effects of α -carotene is the explanation for the lower oxidative stability in carrot puree compared to broccoli and tomato puree.

4. Conclusions

All vegetable purees enriched in n-3-LC-PUFA by the addition of heterotrophic microalgae were prone to oxidation. A higher oxidative stability of enriched broccoli and tomato puree was observed compared to enriched carrot puree, which could be attributed to (i) the higher antioxidant capacity of lycopene and lutein compared to α - and β -carotene, (ii) the higher concentration of phenolic compounds or (iii) synergistic effects between different antioxidants.

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325 **Author contributions**

326 Conceptualization, L. Gheysen and I. Foubert; Methodology, L. Gheysen, and T. Bernaerts; Formal
327 Analysis, L. Gheysen, N. Durnez and J. Devaere; Writing – Original Draft Preparation, L. Gheysen;
328 Writing – Review & Editing, N. Durnez, J. Devaere, T. Bernaerts, A. Van Loey, L. De Cooman and I.
329 Foubert; Supervision, I. Foubert and L. De Cooman; Project Administration, A. Van Loey. All authors
330 have approved this version of the manuscript.

331 **Conflict of interest**

332 The authors have declared no conflict of interest.

333 **Supporting information description**

334 /

335 **Data availability**

336 The data that support the findings of this study are available from the corresponding author upon
337 reasonable request.

338 **Ethics**

339 Ethics approval was not required for this research.

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Captions Figures

Figure 1 General overview of the experimental set-up.

Captions Tables

Table 1 n-3-LC-PUFA, free fatty acid and antioxidant composition of *Schizochytrium* biomass, expressed in mg/g dry biomass, mean ($\pm$ SD), n=3.

Table 2 Vegetable/H₂O ratio, pH, dry matter and antioxidant composition of unenriched vegetable purees, expressed in mg/100 g vegetable puree, mean ($\pm$ SD), n=3. Statistical differences per compound between the vegetable purees are indicated with a different letter, $\alpha=0.05$.

Table 3 Evolution of primary and secondary oxidation products during 12 weeks of storage at 37°C for broccoli, carrot, tomato purees enriched with *Schizochytrium* after high-pressure-homogenization (a), pasteurization (b) and sterilization (c) , mean ($\pm$ SD), n=2*2. The results are normalized and expressed as meq hydroperoxides/100 g enriched puree (for primary oxidation) or mg hexanal equivalents/100 g enriched puree (for secondary oxidation). Statistical differences in function of time (per enriched puree) are indicated with a different letter, $\alpha=0.05$.

Table 4 Evolution of the carotenoids of broccoli, carrot and tomato purees supplemented with *Schizochytrium* after high-pressure-homogenization (a), pasteurization (b) and sterilization (c) during 12 weeks of storage, mean ($\pm$ SD), n=2*2. The results are expressed as mg carotenoid/100 g enriched puree. Statistical differences in function of time (per enriched puree) are indicated with a different letter, $\alpha=0.05$.

Table 5 Total phenolic compounds and ascorbic acid concentration for vegetable purees enriched with *Schizochytrium* after mixing, high-pressure-homogenization, pasteurization, and sterilization, mean ($\pm$ SD), n=2*2. The results are expressed as mg gallic acid equivalent/100 g enriched puree (for total

451 phenolic compounds) or mg ascorbic acid/100 g enriched puree (for ascorbic acid). Statistical
452 differences (antioxidant) are indicated with a different letter, $\alpha=0.05$.

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