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Optimization of an organic yogurt based on sensorial, nutritional, and functional perspectives

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ABSTRACT

The effects of purple grape juice (PGJ), grape skin flour (GSF), and oligofructose (OLI) on proximate composition, total phenolic content (TPC), antioxidant activity (AA), sensory, physicochemical, and textural properties of yogurts were analyzed using response surface methodology. Multiple regression models were proposed and results showed that PGJ increased the viscosity, AA, and TPC, while GSF increased the ash and total fiber contents of yogurts. GSF and OLI increased the hardness and consistency. A simultaneous optimization was performed to maximize TPC, ash and fibers contents, and sensory acceptance: a yogurt containing 1.7% GSF and 8.0% PGJ had a high fiber (5.60 ± 0.13%) and ash (0.76 ± 0.02%) contents, TPC (28.32 ± 2.10 mg GAE/100 g), AA toward DPPH (57.85 ± 1.36 mg AAE/100 g), and total reducing capacity (28.86 ± 5.19 mg QE/100 g). The optimized yogurt had 79% acceptability index, indicating the use of PGJ and GSF is a feasible alternative to increase the functional properties of yogurts.

1. Introduction

The search for foods that contain substances capable of improving the quality of life and that are easily incorporated into the conventional diet has increased in the past decade (Granato, Nunes, & Barba, 2017). Yogurt is the most popular fermented dairy product and has a high nutritional value, especially by containing a significant content of proteins and essential minerals, such as calcium. However, it is not considered a source of dietary fibers. Therefore, it is interesting to continuously investigate value-added ingredients, such as prebiotic fibers, to attract the consumers to consumption of dairy foods (Allgeyer, Miller, & Lee, 2010). Another demand of consumers is related to the development of organic foods. In this aspect, according to Nahed-Toral et al. (2013), factors such as the competition in the commercialization of healthy foods promote the diffusion of organic foods. Overall, consumers tend to define organic products as presenting a higher quality because they do not contain chemical residues and/or pesticides (Lee & Yun, 2015).

In Brazil, organic fruits are widely produced because of their high-added value and demand, but the addition of organic fruits in dairy foods is not a reality whatsoever. Fruit juices are widely consumed because of their freshness, sensory properties, and nutritional value (Granato, Karnopp, & van Ruth, 2015; Nowshehri, Bhat, & Shah, 2015). Grapes are mainly produced in southern states because of the weather conditions and Brazilians consume about 3 L of grape juice/person every year. From the grape industry standpoint, about 20% of grape pomace are generated when juice and/or wine is/are manufactured (Karnopp et al., 2015). Grape skin and seeds may be separated and dried using physical methods and may be used for the manufacture of flours and grape seed oil, respectively. Grape skin contains a relevant amount of anthocyanins and phenolic acids, such as gallic, ferulic, caffeic, syringic and *p*-coumaric acids, that have been linked to multiple *in vitro* and *in vivo* functionalities (Trošt et al., 2016; Jiao, Wei, Chen, Chen, & Zhang, 2017).

In many countries, grape pomaces do not have an adequate destination or useful utilization by the food industry. As grape pomaces constitute rich sources of bioactive compounds, their application as functional ingredients has gained increasing interest worldwide in the last decade (García-Lomillo & González-SanJosé, 2017; Granato, Carrapeiro, Fogliano, & van Ruth, 2016; Luo et al.,

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2017). For instance, Gil-Sánchez et al. (2017) studied the *in vitro* colonic fermentation of four grape pomaces extracts (skin and seeds) and verified that the phenolic compounds present in the extracts are metabolized into different active compounds by microbiota, confirming the bioactivity of grape pomaces. In this aspect, the development of dairy foods containing polyphenols and fibers from grape pomaces is a technological trend that can improve the nutritional and functional value of foods (dos Santos et al., 2017; Lachman et al., 2013).

In the scenario that consumers have become more aware about the consumption of high-fiber foods and those from alternative farming systems, such as the organic system, the development of organic foods containing grape pomace – a rich source of fibers and antioxidants – is an interesting application for technological purposes. In the same sense, although organic yogurts are perceived as healthier and more environmentally friendly compared to conventional yogurts (van Loo, Diem, Pieniak, & Verbeke, 2013), few options are available in the marketplace in many countries in South America and European countries. Additionally, the addition of oligofructose and other fructooligosaccharides in food products is well established (Crispín-Izidro, Lobato-Calleros, Espinosa-Andrews, Alvarez-Ramirez, & Vemon-Carter, 2015; Morais, Cruz, Faria, & Bolini, 2014; Röble, Ktenioudaki, & Gallagher, 2011).

Although the utilization of either grape pomace (Marchiani et al., 2016), grape juice (Silva et al., 2017), or both ingredients (dos Santos et al., 2017) in yogurts and other fermented milks was already the target of investigations, there is no information about the use of response surface methodology (RSM) to optimize a yogurt formulation containing organic PGJ and purple GSF. Taking advantage of limited research on the utilization of purple grape juice (PGJ) and grape skin flour (pomace) - GSF - originated from the organic grape juice industry in Brazil, we evaluated the combined effect of PGJ, GSF, and oligofructose on some quality traits of organic yogurts and optimized a yogurt formulation based on sensorial, nutritional, and functional perspectives using a multi-response statistical technique.

2. Material and methods

2.1. Raw materials

The organic bovine milk (total lipids: 4.91 ± 0.06 g/100 g; proteins: 3.47 ± 0.02 g/100 g; nonfat solids: 9.35 ± 0.03 g/100 g; lactose: 5.24 ± 0.01 g/100 g; pH 6.28 ± 0.04) was kindly provided by the Paraná Reference Center on Agroecology (Curitiba, PR, Brazil), and organic GSF and PGJ were supplied by Uva'Só (Bordeaux variety; Garibaldi, RS, Brazil), oligofructose (95% purity) was kindly provided by Clariant (São Paulo, Brazil), and organic sucrose was obtained at Native Alimentos (São Paulo, Brazil). It is important to note that the GSF and PGJ were produced using the same grape berries and harvested in 2013. The GSF was standardized to 28 Tyler mesh (~0.60 mm) and characterized by presenting 2.39 ± 0.57 g/100 g moisture, 3.85 ± 0.08 g/100 g ash, 9.83 ± 1.36 g/100 g proteins, 8.54 ± 0.30 g/100 g lipids, 54.81 ± 0.77 g/100 g total fibers, in which 51.02 ± 1.12 g/100 g were insoluble fibers and 3.79 ± 0.46 g/100 g were soluble fibers (Karnopp, Margraf, Maciel, Santos, & Granato, 2017). PGJ had the following characteristics: 17.15°Brix total soluble solids, 14.16 g/100 mL total reducing sugars (expressed as glucose), 1.578 g/100 mL titratable acidity (expressed as tartaric acid), 0.025 g/100 mL volatile acidity (expressed as acetic acid), optical density (λ = 420 + 520 + 620 nm) = 1.560, and pH = 3.13.

2.2. PGJ and GSF: Total phenolics and anthocyanins

The characterization of phenolic compounds in PGJ and GSF was carried out using both UV–Vis spectrophotometry and HPLC. The total phenolic content (TPC) and total monomeric anthocyanins were determined according to the procedures and experimental conditions described elsewhere (Granato, Koot, Schnitzler, & van Ruth, 2015).

GSF was extracted with ethyl alcohol 70% at 60 °C for 30 min under magnetic agitation using a proportion of GSF:solvent of 1:100 w/v, and filtered using a qualitative paper (Whatmann nº 1) prior to the analysis. Grape skin flour extract was filtered through a 0.45 μm Nylon membrane prior to injection. The HPLC system (HP, model 1100) was equipped with a Lichrospher RP_{18} (5 μm) column and the UV detector was set at 210 nm. The mobile phase consisted of solvent A (ultrapure water acidified with 1% phosphoric acid) and solvent B (acetonitrile). The gradient elution conditions started with 90% solvent A, and adjusted for 60% solvent A in 5 min, and then 90% solvent A in 45 min. The injection volume was 10 μL and the flow rate of 0.50 mL/min.

For purple grape juice analysis, the separation and quantification of phenolic compounds were carried out using a HPLC (Agilent Technologies, model 1100 series) equipped with a Phenomenex Gemini C_{18} column and guard column at 40 °C with 150 × 4.6 mm × 3 μm. An aliquot of 450 μL of the grape juice was diluted to 900 μL with a 0.85% phosphoric acid solution and filtered through a 0.45 μm polyvinylidene difluoride membrane prior to injection. The mobile phase was consisted of solvent A (0.85% phosphoric acid solution) and solvent B (acetonitrile). The gradient elution conditions started with 100% of solvent A and adjusted for 7% of solvent B in 10 min; 10% of solvent B in 20 min; 12% of solvent B in 30 min; 23% of solvent B in 40 min; 35% of solvent B in 45 min; and 100% of solvent B in 55 min. The flow rate was 0.50 mL/min and the injection volume was 10 μL (Granato, Koot et al., 2015).

Phenolic compounds [rutin, (+)-catechin, (−)-epicatechin, *trans*-resveratrol, quercetin, chlorogenic acid, gallic acid, and myricetin)] were quantified in triplicate by external validation using HPLC-grade phenolics. For that purpose, analytical curves were plotted using five concentrations of the standards and the determination coefficients were $R^2 > 0.99$ for all phenolics (data not shown).

2.3. Experimental design: Yogurt formulations

An augmented simplex-centroid mixture design (Table 1) containing ten formulations was used to evaluate the effects of OLI, PGJ, and GSF on the proximate composition, physicochemical properties, syneresis, sensorial traits, total phenolic content, and antioxidant activity of the yogurts. The total ingredients added in each formulation was established at 25 g/100 g. Therefore, even if

Table 1
Coded and real values of the organic yogurt formulations with addition of grape skin flour, oligofructose, and purple grape juice.

Sample	Grape skin flour (g/100 g)	Oligofructose (g/100 g)	Purple grape juice (g/100 g)
A	1.00 (2.5)	0.00 (0)	0.00 (0)
B	0.00 (0)	1.00 (5.0)	0.00 (0)
C	0.00 (0)	0.00 (0)	1.00 (25)
D	0.50 (1.25)	0.50 (2.5)	0.00 (0)
E	0.50 (1.25)	0.00 (0)	0.50 (12.5)
F	0.00 (0)	0.50 (2.5)	0.50 (12.5)
G	0.333 (0.83)	0.333 (1.67)	0.333 (8.33)
H	0.667 (1.66)	0.167 (0.84)	0.167 (4.17)
I	0.167 (0.42)	0.667 (3.34)	0.167 (4.17)
J	0.167 (0.42)	0.167 (0.84)	0.667 (16.7)