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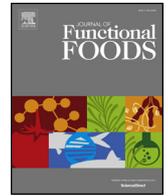
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Gastroprotective effect of soluble dietary fibres from yellow passion fruit (*Passiflora edulis* f. *flavicarpa*) peel against ethanol-induced ulcer in rats

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ABSTRACT

Yellow passion fruit peel (PFP) is considered an important source of dietary fibre (DF) and its consumption has been associated with health benefits in metabolic and gut disturbs. This work aimed to characterize the chemical structure of soluble DF (SDF) from PFP and evaluate its gastroprotective activity in acute gastric-ulcer model induced by ethanol in rats. SDF was composed of 92% of GalA and presented high methyl esterified homogalacturonan (DE = 70%), with relative M_w of 53 kDa. Oral pre-treatment of animals with SDF (0.1, 1 and 10 mg/kg) significantly reduced gastric ulcer lesions by 72%, 79% and 87% respectively. The gastroprotective effect was maintained when SDF was administered by the intraperitoneal route. SDF also prevented the depletion of GSH levels and gastric wall mucus when administered by both routes. These results demonstrated that ingestion of SDF from PFP exerts significant gastroprotective effects in vivo on experimentally induced gastric ulcers.

1. Introduction

Yellow passion fruit (*Passiflora edulis* f. *flavicarpa*) is a native plant of Brazil, where it is widely cultivated mainly for the industrial production of juice and soft drinks. Its processing generates large amounts of peel and seed waste since it can account for more than 50% of the fruit weight (López-Vargas, Fernández-López, Pérez-Álvarez, & Viuda-Martos, 2013; Seixas et al., 2014). Passion fruit is also well known for its therapeutic and health properties. Traditionally, its leaves have been used for the treatment of anxiety and insomnia (Barbosa et al., 2008). Likewise, surveys based on passion fruit peel (PFP) supplementation in both human and experimental models, demonstrated metabolic and health improvements which has been associated to its dietary fibre (DF) content (de Souza, Jonathan, Saad, Schols, & Venema, 2018; Macagnan et al., 2015).

In this context, DF is a nutritional concept that describes a wide variety of carbohydrates, including cellulose, hemicelluloses and pectins, that cannot be digested by endogenous enzymes of the human gastrointestinal tract thus, they can't be hydrolyzed nor absorbed by the small intestine (Brownlee, 2014; Englyst, Liu, & Englyst, 2007;

Poultanen, Fiszman, Marsaux, Steinert, & Mela, 2018). The main food sources of DF are plant origin foods, such as whole grains, vegetables, fruits, as well as their by-products such as seeds, leafs, pomace and peels (Albuquerque et al., 2019; Martins, Pinho, & Ferreira, 2017). The daily consumption of these foods, their by-products, and hence DF, is well recognized as an essential part of a healthy diet and thus benefits in the gastrointestinal tract function, cardiovascular diseases and diabetes (Albuquerque et al., 2019; Verspreet et al., 2016; Brownlee, 2014).

In this sense, PFP supplementation, which is considered an important source of DF (de Souza et al., 2018), have been investigated. Studies in humans demonstrated that diet supplemented with PFP flour was able to improve metabolic parameters, such as reducing fasting blood glucose and glycated haemoglobin in type 2 diabetes individuals (de Queiroz et al., 2012); as well as reduce fasting blood glucose and triglycerides levels in hypercholesterolaemic women (Ramos et al., 2007). In animal models, supplementation with PFP in isocaloric diet, protected against weight gain (Corrêa et al., 2014), decreased serum levels of triglycerides, improved glucose parameters (Corrêa et al., 2014; Macagnan et al., 2015), and increased faecal excretion of lipids.

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In high fat diet animal models, PFP improved insulin sensitivity, glucose metabolism and serum levels of incretins and leptin (Lima et al., 2016). More recently, PFP was tested in *in vitro* model of the upper gastrointestinal tract (TIM-1) and it was shown that PFP was able to slow down the absorption of glucose compared to other tested fruit by-products (de Souza et al., 2018).

Moreover, in animal models of induced colitis, PFP flour supplementation exerted an intestinal anti-inflammatory effect and attenuated the colonic damage caused by the dextran sodium sulphate. It demonstrated protective effects against symptoms and clinical observations, which included less diarrhoea, rectal bleeding and bloody faeces, weight loss and decreased food intake (Cazarin, Silva, Colomeu, Batista et al., 2014; Cazarin et al., 2016; Silva, Cazarin, Batista, & Maróstica, 2014).

In this sense, several studies trends towards DF and how it can affect metabolism, chronic diseases such as diabetes, as well as the intestine and its microbiota, although few studies analysed DF effects on the upper gastrointestinal tract (Brownlee, 2014). Thus, this work aimed to characterize the chemical structure of an isolated dietary fibre from PFP and evaluate its gastroprotective activity in acute gastric-ulcer model induced by ethanol in rats.

2. Material and methods

2.1. Yellow passion fruit peel flour

Yellow passion fruits (*Passiflora edulis*) cultivated in Torre de Pedra/SP, Brazil, harvested in June 2010, were used in the present study. After sanitation, it was performed the separation between pulp and peel. The peels were cut and dried at 50 °C (Marconi, Piracicaba/SP – Brazil) until to reach 10% of moisture. Once dry, it was ground in a hammer mill (20 mesh) to obtain flour, which was stored in the dark plastic packaging at room temperature until analysis (24 °C).

2.2. Extraction of soluble dietary fibre

Passion fruit peel flour was defatted with hexane (1:7, w/v, 2x), air dried and submitted to the standard enzymatic-gravimetric method AOAC official (Method 991.43) to obtain soluble and insoluble dietary fibres. Briefly, samples undergo sequential enzymatic digestion by heat-stable α -amylase, protease, and amyloglucosidase to remove starch and protein. The enzymatic treatments were performed in a heated water bath with temperature adjusted for each enzyme, under constant stirring. Samples were suspended in water (1:20, w:v), and heated until 90 °C. α -Amylase was added (5 units/g of sample) and samples were treated for 3 h. The suspension was cooled to 60 °C, amyloglucosidase (1.3 units/g of sample) was added and remained for 1 h. At the end of this treatment, the Lugol's iodine test was performed to confirm the absence of starch. Then, pH was adjusted to 7.0 with 10% NaOH solution and protease (≤ 1 unit/g of sample) was added to the suspension and incubated for 1 h. Finally, samples were heated to 80 °C, for one hour to inactivate the enzymes.

The enzymatically-treated suspensions were centrifuged (5000 rpm/25 min.) and supernatant (containing soluble dietary fibre – SDF) was separated from the precipitated (containing insoluble dietary fibre – IDF). Samples were then submitted to dialysis against tap water at room temperature, for two days, in 12–14 kDa size exclusion membranes (Spectra Por®). Each material was freeze-dried, and yields were determined and expressed as a percentage based on the weight of dry passion fruit flour.

2.3. Monosaccharide composition

Neutral monosaccharide composition was determined by derivatization of SDF to alditol acetates after hydrolysis and reduction. Briefly, samples were submitted to hydrolysis with 2 M TFA for 8 h at 100 °C,

followed by NaBH₄ reduction and overnight acetylation with Ac₂O-pyridine (1:1, v/v). Analyses were conducted on GC-MS (Varian, model Saturn 4000), with He as a carrier gas. A capillary column (30 m \times 0.25 mm i.d.) of DB-225, kept up 50 °C during injection for 1 min, then adjusted at 40 °C/min to 220 °C and held at this constant temperature for 19.75 min, was used for the quantitative analysis.

Uronic acids were determined according to Filisetti-Cozzi and Carpita (1991) spectrophotometric method.

2.4. Determination of homogeneity and relative molecular weight

The homogeneity and relative molecular weight of SDF were determined by high-performance steric exclusion chromatography (HPSEC). Briefly, four columns were used in series (7×10^6 Da, 4×10^5 Da, 8×10^4 Da and 5×10^3 Da, Ultrahydrogel, Waters) and a Waters 2410 refractometer was used as detection equipment. The eluent was 0.1 M NaNO₂ containing 200 ppm NaN₃ at 0.6 mL/min. SDF sample, previously filtered through a membrane (0.22 μ m, Millipore), was injected at a concentration of 1 mg/mL. Standards dextran (487 kDa, 266 kDa, 124 kDa, 72.2 kDa, 40.2 kDa, 17.2 kDa and 9.4 kDa), from Sigma were used to obtain the calibration curve and molecular weight reference.

2.5. Nuclear magnetic resonance spectroscopy

SDF sample was analysed for its chemical structure by means of ¹H, ¹³C and 2D ¹H-¹³C HSQC correlation map. Analyses were performed at 70 °C on a Bruker AVANCE III 400 NMR spectrometer, operating at 9.5 T, observing ¹H at 400.13 MHz and ¹³C at 100.61 MHz, and equipped with a 5-mm multinuclear inverse detection probe with z-gradient. The chemical shifts are expressed in ppm relative to CH₃ signal from internal reference acetone (δ 30.2/2.22). Bruker supplied all pulse programs.

2.6. Animals

Female Wistar rats (180–200 g), obtained from the vivarium of Federal University of Paraná, were kept in plastic cages containing pine shavings (maximum of 5 rats per cage) and maintained at 22 \pm 2 °C under a 12/12 h light/dark cycle, with free access to food (Nuvi-Lab CR-1, Quimtia S/A, Colombo, PR, Brazil) and water. All animal protocols were approved by the Committee of Animal Experimentation of Federal University of Paraná (CEUA/BIO - UFPR: n° 1123) and conducted in agreement with the “Guide for the Care and Use of Laboratory Animals” (8th edition, National Research Council, 2011).

2.7. Acute gastric lesions induced by ethanol in rats

Gastric lesions were induced by oral administration (p.o.) of ethanol, as previously described (Robert, Nezamis, Lancaster, & Hanchar, 1979). Following 18 h of fasting with free access to water, animals (n = 6) were orally pre-treated with vehicle (V: water, 1 mL/kg), sucralfate (S: 100 mg/kg) or omeprazole (O: 40 mg/kg). SDF fraction was administered orally (at 0.1, 1 and 10 mg/kg) or intraperitoneally (at 1 mg/kg, i.p.). One hour after the pre-treatments, acute gastric lesion was induced by ethanol p.a. administration (1 mL/animal, p.o.), and then 1 h later, the animals were euthanized by thiopental overdose (100 mg/kg, i.p.) followed by cervical dislocation. The stomachs were excised, opened along the greater curvature, cleaned with saline, stretched flat and photographed to measure the haemorrhagic gastric lesions area (mm²) using computerized planimetry software (Image Tool® 3.0). Subsequently, the glandular portion of the stomach was divided in two parts and weighted for determination of gastric wall mucus and reduced glutathione (GSH) levels.

2.8. Measurement of wall mucus and reduced glutathione (GSH) levels in gastric tissue

The analysis of gastric mucus level was performed according to Corne, Morrissey, and Woods (1974). The gastric tissue, prior divided and weighted, was incubated in 0.1% Alcian Blue solution for 2 h, then washed twice with 0.25 M sucrose for 15 and 30 min, for subsequently extraction of wall mucus complexed with Alcian Blue with 0.5 M magnesium chloride solution. This extract was mixed in equal parts with ether and centrifuged for 10 min at 3600 rpm. Finally, the absorbance of the supernatant was spectrophotometrically measured at 598 nm, and the results were expressed in μg of Alcian Blue/g of tissue using a standard curve of Alcian Blue (6.25–100 μg).

The reduced glutathione (GSH) levels were evaluated in the other half of gastric tissue homogenized with 0.2 M potassium phosphate buffer (pH 6.5), according Sedlak and Lindsay (1968). The homogenate was mixed with 12.5% trichloroacetic acid, then centrifuged for 15 min at 3000 rpm (4 °C). The obtained supernatant was pipetted with 400 mM TRIS-HCl buffer (pH 8.5) and 10 mM 5,5'-dithiobis-(2-nitrobenzoic acid) (DTNB), which results in a colorimetric reaction. Then, GSH levels were quantified by the absorbance measured at 415 nm and interpolated into a standard curve of GSH (0.375–3 μg), and the results were expressed as μg of GSH/g of tissue.

2.9. Statistical analysis

Results were expressed as mean \pm standard error of mean (SEM) and the statistical significance ($P < 0.05$) was determined using one-way analysis of variance (ANOVA) followed by Bonferroni's test ($n = 5\text{--}6$ animals) using GraphPad Prism® software version 6.0 (San Diego, CA, USA).

3. Results and discussion

The defatted yellow passion fruit peel powder (50 g), submitted to the enzymatic-gravimetric method, centrifuged and dialysed against tap water, yielded approximately 78% of total dietary fibre (TDF, g/100 g of dried weight) in which, 20% (10 g) were soluble dietary fibres (SDF) and 58% (29 g) were insoluble dietary fibres (IDF). This agrees with previous studies that employed dietary fibre extraction methodologies, which reported 57.9–81.9% for TDF, 11.7–20.1% for SDF, and 40–62.4% for IDF (Canteri et al., 2010; Cazarin, Silva, Colomeu, Batista et al., 2014; Cazarin, Silva, Colomeu, Zollner et al., 2014; Cazarin et al., 2016; Hernández-Santos et al., 2014; Lima et al., 2016; López-Vargas et al., 2013; Macagnan et al., 2015; Silva et al., 2014; Yapó & Koffi, 2008a).

On monosaccharide composition, IDF presented a complex monosaccharide composition, typical of hemicellulosic polysaccharides, with high amounts of xylose, followed by galactose, uronic acids, glucose, mannose, arabinose and rhamnose (Table 1). SDF fraction was mainly

Table 1
Monosaccharide composition of dietary fibres obtained from passion fruit peel flour.

Fraction	Neutral sugars ^a						Uronic acids ^b
	Ara	Rha	Gal	Glc	Man	Xyl	
SDF	3.0	tr ^c	2.3	1.8	tr ^c	tr ^c	92.0 ^c
IDF	7.9	1.8	18.2	14.0	10.7	47.4	17.0 ^d

^a % of peak area relative to total peak areas, determined by GC-MS.

^b Determined spectrophotometrically using the modified *m*-hydroxybiphenyl method (Filisetti-Cozzi & Carpita, 1991).

^c Galacturonic acid was employed in the calibration curve.

^d Glucuronic acid was employed in the calibration curve.

^e Trace amounts.

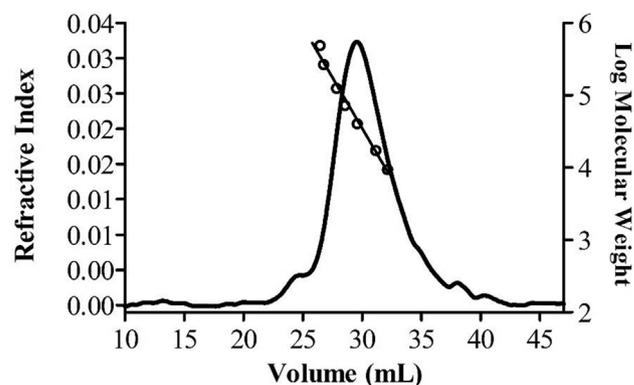


Fig. 1. HPLC elution profile of soluble dietary fibres (SDF) from passion fruit peel. Refractive index detector. Elution volume of dextran standards of molecular weight 487 kDa, 266 kDa, 124 kDa, 72.2 kDa, 40.2 kDa, 17.2 kDa and 9.4 kDa (left to right) were employed to construct the calibration curve.

composed of galacturonic acid (92%) suggesting the presence of pectin. The remaining eight percent presented relative amounts of arabinose, galactose and glucose, as shown in Table 1.

Size exclusion chromatography was used to analyse the SDF fraction, and one main peak with the relative molecular weight of 53 kDa was observed (Fig. 1).

The ¹³C and ¹H NMR spectra and 2D ¹H-¹³C HSQC correlation map of SDF (Fig. 2) were in agreement with monosaccharide composition and showed typical signals of α -D-GalpA units. Those at δ 99.3/5.14 and δ 100.1/4.96 are from C-1/H-1 of non-esterified and esterified α -D-GalpA units, while the signal at δ 52.8/3.81 was assigned to $-\text{COOCH}_3$. Also signals at δ 71.5/4.68 and δ 70.6/5.07 from non-esterified and esterified C-5/H-5 respectively, and at δ 78.5/4.46 from C-4/H-4 of the GalpA units. Finally, signals at δ 68.2/3.98 and δ 68.2/3.75 from C-2/H-2 and C-3/H-3. These assignments, in addition to the results of sugar composition, suggest the presence of a methyl esterified homogalacturonan in SDF portion of PFP. Thus, the degree of methyl-esterification (DM) was determined by ¹H NMR spectroscopy, showing a value of approximately 70%, which can characterize it as highly methoxylated pectin.

Pectin extraction from PFP, aiming food applications and utilization of a by-product of the processing industries of juice, has been studied by several authors. The most common methods used for the extraction of pectins from PFP include: calcium-ion chelators or diluted acids (nitric acid, sulfuric acid, acetic acid, tartaric acid or citric acid) under conventional heating (60–100 °C); and autoclave or microwave-induced heating (Canteri et al., 2010; Contreras-Esquivel et al., 2010; Kliemann et al., 2009; Pinheiro et al., 2008; Seixas et al., 2014; Yapó & Koffi, 2006, 2008b; Yapó, 2009). These studies showed that the extraction procedure (acid type and concentration) of pectin has a significant effect on its yield and macromolecular parameters (GalA content, degree of methyl esterification, molar mass and intrinsic viscosity). The GalA content in these studies varied from 65.5% to 88.2%. The pectin present in SDF fraction presented 92% of GalA and was obtained in very good yield (20%, g/100 g dry weight). Pectins with various degrees of methyl esterification have been extracted from passion fruit peels, also depending on the type of extraction. Low methoxy pectins (with DM = 5–45.94%) were observed by Yapó & Koffi (2006, 2008b) and Kliemann et al. (2009) while high methoxy pectins (with DM = 54.0–79.6%) have been extracted by others (Canteri et al., 2010; Contreras-Esquivel et al., 2010; Pinheiro et al., 2008; Seixas et al., 2014; Yapó, 2009). Pinheiro et al. (2008) observed that weak extraction conditions, such as low citric acid concentration, increased the degree of methyl esterification of the pectins. We found a DM of 70% for pectin in SDF fraction, showing that the applied DF extraction methodology can extract the pectin close to its native form. Regarding molar mass of

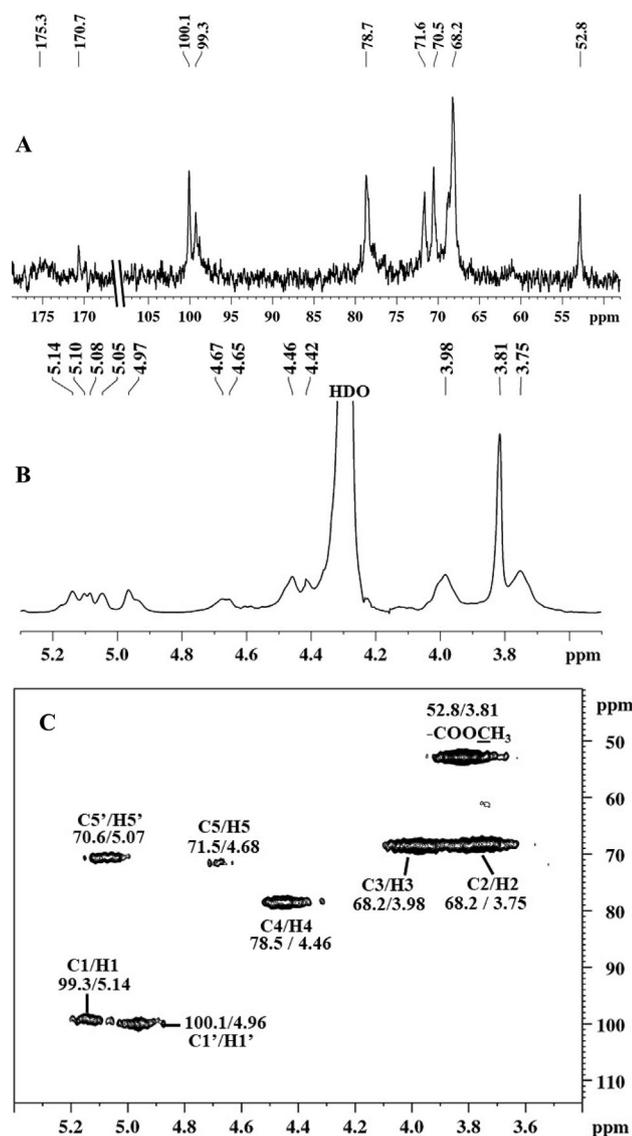


Fig. 2. NMR spectra of soluble dietary fibres (SDF) from passion fruit peel, in D_2O at $70^\circ C$. (A) ^{13}C NMR spectrum; (B) 1H NMR spectrum and (C) 2D 1H - ^{13}C HSQC correlation map. C1'/H1' and C5'/H5' are from C/H correlations of methyl esterified α -D-GalpA units.

pectin extracted from passion fruit peels, it ranges from 5.1×10^4 to 4.9×10^5 g/mol (Seixas et al., 2014; Yapo & Koffi, 2006, 2008b; Yapo, 2009). It has been observed that pectin may suffer degradation when extracted under severe conditions, presenting lower molar mass and exhibited a polydisperse and multimodal elution pattern in HPSEC (Yapo, 2009), while a homogeneous elution profile, with only one peak and high molar mass was observed by Canteri et al. (2010) and Contreras-Esquivel et al. (2010).

Once our objective was to evaluate *in vivo* gastroprotective activity of the soluble dietary fibre from passion fruit peels, despite all the optimized methodologies of pectin extraction available in the literature, the enzymatic-gravimetric dietary fibre method proved to be the method of choice. It extracted a pectin with high content of GalA, and it may mimic the physiological process of digestion in the gastrointestinal tract.

Several species of the genus *Passiflora* have long been used in folk medicine, especially as anxiolytic and sedative. The consumption of PFP as flour had been recommended for its bioactive compounds, but also for its high DF content. In this sense, a significant number of observational and meta-analysis research, showed that regular intake of

DF can prevent and support the treatment of metabolic, gastrointestinal or diet-related diseases (Guiné et al., 2016; Verspreet et al., 2016), in addition, surveys regarding DF supplementation, tended to focus on their effects on the gastrointestinal tract and its associated cancers, heart and bowel diseases, diabetes, hypercholesterolemia, and more recently the gut microbiota (Brownlee, 2014). Thus, assuming that PFP is a source of DF, some studies already evaluated its physiological effects and evidence shows that PFP presented health benefits, such as, improve insulin resistance in type 2 diabetics individuals (de Queiroz et al., 2012), anti-inflammatory effect and decreased colonic damage against induced dextran sodium sulphate model of mouse colitis (Cazarin et al., 2016), improved antioxidant status in animal models (Silva et al., 2014), as well as decreased lipodystrophy syndrome in HIV patients (Marques et al., 2016). Even though, few studies evaluated the effects of DF on the stomach or the upper gastrointestinal tract (Brownlee, 2014). To the best of our knowledge, no published research evaluated the effect of SDF from PFP on gastric ulcer models. Thus, the gastroprotective effect of SDF isolated from PFP was investigated in the ethanol-induced gastric ulcer. Oral administration of ethanol is a well-accepted model for induction of gastric ulcer and widely used to evaluate the gastroprotective effects of new agents against mucosa damage (Robert et al., 1979). It is well known that oral administration of ethanol abrogates the gastric mucosal defence, favouring the development of haemorrhagic lesions, due to the reduction of gastric mucus and GSH levels, which are important protective factors against stomach damage (Oates & Hakkinen, 1988).

Our results showed that the oral pre-treatment with SDF at 0.1, 1 and 10 mg/kg significantly decreased the ethanol-induced gastric lesions in 72.25%, 79.23% and 87.17% respectively, when compared to the vehicle group (V: 198.80 ± 33.87 mm 2) (Fig. 3A). It is also clear that the gastroprotective effect promoted by SDF did not display a dose-response relationship. Moreover, ethanol-induced gastric lesions were prevented in 93.06% by sucralfate, a drug that creates a physical barrier protecting the stomach against the gastric acidity, when compared to vehicle group (Fig. 3A).

Considering the gastric mucosal defensive factors, the oral treatment with SDF 0.1, 1 and 10 mg/kg was able to prevent the depletion of GSH levels in 52.20%, 41.91% and 50.33%, respectively, when compared to the vehicle group (V: 1659.29 ± 97.26 μ g of GSH/g of tissue) (Fig. 3B). Additionally, the depletion of gastric wall mucus was prevented by SDF 0.1, 1 and 10 mg/kg in 26.32%, 25.03% and 31.00%, respectively, when compared with vehicle group (3460.57 ± 72.66 μ g of Alcian Blue/g of tissue) (Fig. 3C). The treatment of animals with sucralfate was able to prevent only GSH levels (37.32%), without the preservation of gastric wall mucus (Fig. 3B and C).

Interestingly, when animals were treated with SDF (1 mg/kg) by intraperitoneal route, the gastroprotective effect was maintained, decreasing gastric lesion area in 72.56%, when compared to vehicle group (V: 168.32 ± 20.61 mm 2). This result is extremely valuable, since it is possible to discard that SDF promotes gastroprotection only by a physical barrier in the stomach (Fig. 4A). Furthermore, as observed with oral treatment, when SDF was administered by intraperitoneal route, the pectin from passion fruit peel preserved the gastric protective factors. SDF at 1 mg/kg (i.p.) was able to prevent the depletion of GSH levels in 40.81% and gastric wall mucus in 21.24%, when compared to the vehicle group (V: GSH: 1659.29 ± 97.25 μ g of GSH/g of tissue and Mucus: 3445.69 ± 72.34 μ g of Alcian Blue/g of tissue) (Fig. 4B and C). Since the mechanism of gastroprotection of sucralfate is attributable to the formation of a physical protective barrier, to compare the effect of SDF administered by intraperitoneal route, omeprazole was chosen as positive control. Omeprazole belongs to the proton pump inhibitors class, an antisecretory drug that inhibit irreversibly the gastric H^+ , K^+ -ATPase. Omeprazole decreased the gastric lesion in 91.40% and prevented the depletion of GSH and mucus levels in 37.37% and 26.61%, respectively (Fig. 4A–C).

The gastroprotective effects of pectins in the ethanol-induced gastric

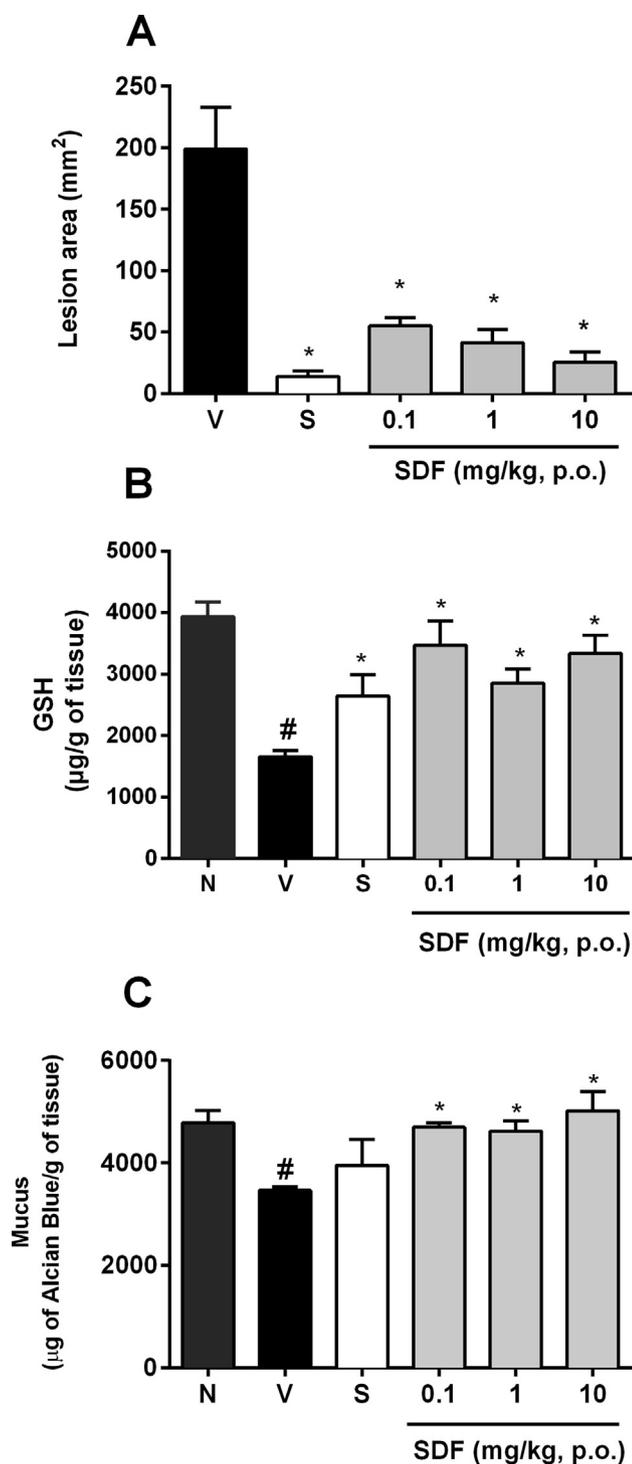


Fig. 3. Effect of oral pre-treatment with SDF on gastric ulcers induced by ethanol in rats. The animals (n = 5–6) were orally treated with vehicle (V: water 1 mL/kg), sucralfate (S: 100 mg/kg) or SDF (0.1, 1 and 10 mg/kg), 1 h before oral ethanol P.A. (1 mL/animal) administration. Panel A: lesion area; Panel B: gastric glutathione (GSH) levels and Panel C: gastric mucus. The results are expressed as mean ± S.E.M. *P < 0.05 when compared to the vehicle group (V) and # P < 0.05 when compared to the naive group (N).

ulcer model have already been reported in the literature. A pectic type II arabinogalactan from *Maytenus ilicifolia* (Cipriani et al., 2006), a rhamnogalacturonan from jambu (*Acmella oleracea*) (Nascimento et al., 2013), type I arabinogalactan from cactus (*Cereus peruvianus* Mill.) (Tanaka et al., 2010) and a pectin from wormwood (*Artemisia campestris*) (Corrêa-Ferreira et al., 2017), all identified as pectic

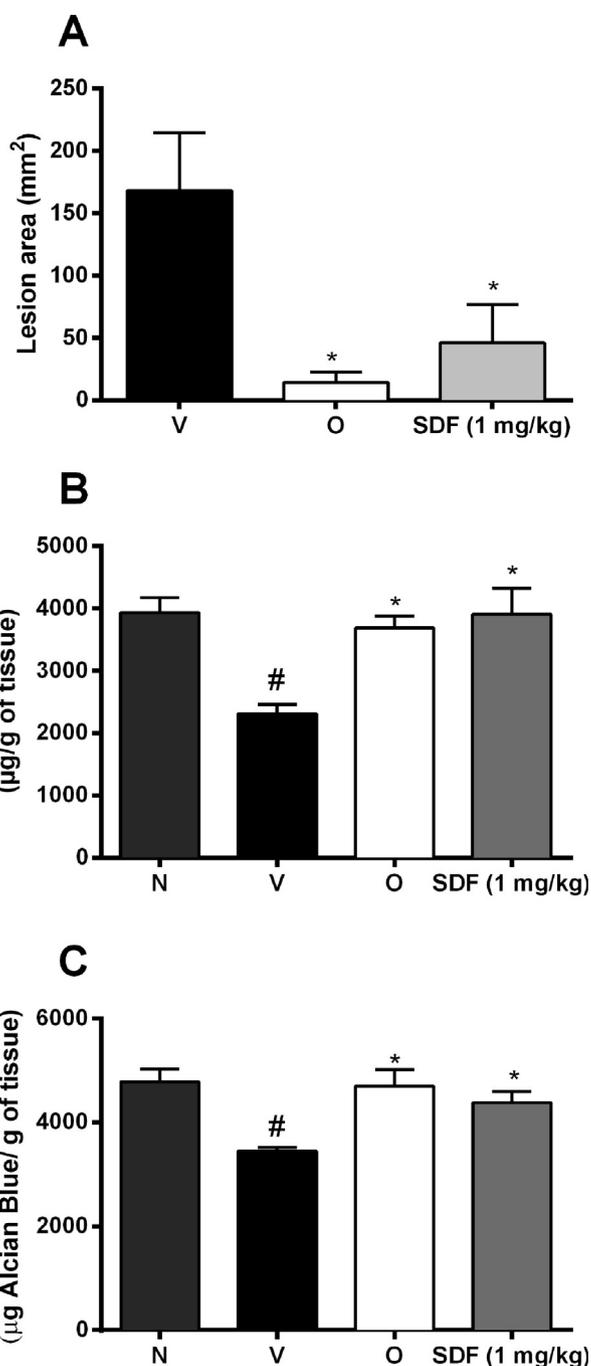


Fig. 4. Effect of intraperitoneal pre-treatment with SDF on gastric ulcers induced by ethanol in rats. The animals (n = 5) were orally treated with vehicle (V: water 1 mL/kg), omeprazole (O: 40 mg/kg) or intraperitoneally with SDF (1 mg/kg), 1 h before oral ethanol P.A. (1 mL/animal) administration. Panel A: lesion area; Panel B: gastric glutathione (GSH) levels and Panel C: gastric mucus. The results are expressed as mean ± S.E.M. *P < 0.05 when compared to the vehicle group (V) and # P < 0.05 when compared to the naive group (N).

polysaccharides and considered dietary fibres, demonstrated gastro-protective effects and the pectin from wormwood also maintained the GSH levels and the gastric mucus barrier. It is noteworthy that the polysaccharides cited in those publications were not considered as dietary fibres due to different extraction methods and, they were isolated from medicinal plants. From food sources, type I arabinogalactans from soybean meal (Cipriani et al., 2009) and from prunes (Cantu-Jungles et al., 2014) also promoted gastroprotection in the ethanol-induced gastric ulcer model. However, pectic polysaccharides from

prunes did not prevent the depletion of GSH and the amount of gastric mucus (Cantu-Jungles et al., 2014). It is notable that the gastroprotection promoted by SDF from passion fruit peel in the ethanol-induced gastric ulcer model was more effective than all the above cited pectic polysaccharides and the only one that has been tested and been active by the intraperitoneal route.

At this moment, some mechanisms suggest that gastroprotective effects could be displayed by the binding of the polysaccharides to the gastric mucosa surface, acting as coating agent; and/or to the reduction of gastric secretion (HCl and pepsin) as well as by protecting gastric mucosa due to increasing scavenging radicals and/or synthesis of mucus (Mellinger-Silva et al., 2011; Nergard et al., 2005). Thus, SDF from passion fruit peel presented its gastroprotective activity by preventing the depletion of GSH levels and gastric wall mucus and presented anti-ulcer effect even when administered by the intraperitoneal route. However, further studies are encouraged to investigate the mechanisms involved in the gastroprotective effects of SDF.

4. Conclusion

Yellow passion fruit peel is a very good source of dietary fibre (78%). Soluble dietary fibres (SDF) were isolated (yield of 20%) and composed of 92% of GalpA units and presented high methyl esterified homogalacturonan (DE = 70%), with relative M_w of 53 kDa. Oral and intraperitoneal pre-treatments of animals with SDF significantly reduced gastric ulcer lesions induced by ethanol and prevented the depletion of GSH levels and gastric wall mucus. These results demonstrated that ingestion of SDF from passion fruit peel exerts significant gastroprotective effects in vivo on experimentally induced gastric ulcers. Moreover, this study adds one more valuable application for yellow passion fruit peel, which is an unexploited by-product from the juice industry in many tropical and subtropical countries, like Brazil.

Declarations of interest

None.

Ethics statements

Female Wistar rats (180–200 g), obtained from the vivarium of Federal University of Paraná, were kept in plastic cages containing pine shavings (maximum of 5 rats per cage) and maintained at $22 \pm 2^\circ\text{C}$ under a 12/12 h light/dark cycle, with free access to food (Nuvi-Lab CR-1, Quimtia S/A, Colombo, PR, Brazil) and water. All animal protocols were approved by the Committee of Animal Experimentation of Federal University of Paraná (CEUA/BIO - UFPR: n° 1123) and conducted in agreement with the “Guide for the Care and Use of Laboratory Animals” (8th edition, National Research Council, 2011).

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