Caco-2 cell transport of purple sweet potato anthocyanins-phospholipids complex

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Revised: 28 September 2017 / Accepted: 19 October 2017 / Published online: 28 October 2017
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Abstract In this study, the role of phospholipids in transepithelial transport and the impact on the antioxidant activity of purple sweet potato anthocyanins (PSPAs) was evaluated. PSPAs were purified by column chromatography, and then PSPAs-phospholipids complex (PSPAs-PC) was prepared. In antioxidant assay in vitro, PSPAs-PC exhibited potential antioxidant activity; meanwhile, it exhibited relatively higher stability in mimic gastrointestinal digestion conditions. The inhibitory effect of PSPAs-PC on the oxidation of soybean oil was significantly higher after 15 days storage. The presence of phospholipids increased the transepithelial transport of PSPAs; its apparent permeability coefficient (*P_{app}*) was higher, while its efflux ratio was lower than PSPAs. Based on the above results, it clearly displays the potential of phospholipids in the promotion of intestinal transport of PSPAs, and further studies are needed to explore the in-depth mechanism of the bioavailability promotion effect of phospholipids.

Keywords Anthocyanins · Purple sweet potato · Phospholipids · Transepithelial transport

Introduction

Anthocyanins are closely associated with the wide range of colors in plants (Ravanfar et al. 2015). They possess various pharmacological properties and play important roles in food industry (Lim et al. 2013), in addition to fruits and berries, some vegetables can be major sources of anthocyanins in human diet. Nowadays, purple sweet potato (*Ipomoea batatas* (L.) Lam) (PSP) has attracted more attentions for its unique nutrition. PSP contains high anthocyanins compared with other colors, and they differ depending on varieties (Zhang et al. 2016). The bioavailability of anthocyanins is a fundamental factor for their physiological functions. In previous studies, urinary excretion and the recovery of anthocyanins in feces were low, which indicated that intact anthocyanins are poorly absorbed and an extensive biotransformation of anthocyanins after oral ingestion (Keppler and Humpf 2005).

Phospholipids are amphipathic molecules with considerable solubility in oil and aqueous mediums, which can promote the absorption of compounds into the blood stream. Recently, phyto-phospholipid complexation technique has emerged for improving the bioavailability of polyphenols with poor competency of solubilizing and crossing biological membranes (Khan et al. 2013). It has been reported the mechanisms proposed for the application of phospholipids include the formation of micelles or liposomes (Taylor et al. 2005), additionally, numerous advantages have been shown in the solubilizing property while considering them for a carrier system, therefore, it is expected that the combination with phospholipids might improve the biological effect of anthocyanins.

Because of high bioactivity, anthocyanins have been discussed as potential active ingredients in plants (De Pascual-Teresa et al. 2010). At present, most studies on

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anthocyanins have focused on their antioxidant and other activities *in vitro*; information about the bioavailability is limited. In our previously study, the results suggested that purple sweet potato anthocyanins (PSPAs) may have prebiotic-like activity by modulating intestinal microbiota, contributing to human health improvements (Zhang et al. 2016). Since phospholipids have the ability to improve the bioavailability of polyphenols *in vivo*, we therefore investigated the efficacy of phospholipids to aid in the transepithelial transport of PSPAs through a Caco-2 cell monolayer model, a widely used model for human intestinal drug absorption, and to evaluate the antioxidant activity and stability of PSPAs-phospholipids complex (PSPAs-PC).

Materials and methods

Reagents

Cyanidin 3- β -*O*-glucoside (C3G), 2,2-diphenyl-1-picrylhydrazyl (DPPH), 5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT), L-Ascorbic acid, 2,2'-azobis-2-methylpropanimidamide dihydrochloride (AAPH), 2,2'-azino-bis-(3-ethyl-benzothiazolin-6-sulfonate) diammonium salts (ABTS), 3-(4, pepsin, pancreatin and bile salt were obtained from Sigma (St. Louis, USA). Phospholipids (hydrogenated soy phosphatidyl choline) were purchased from Sihai Chemical Co., Ltd. (Zhengzhou, China). Butylated hydroxytoluene (BHT) and Tertiary butylhydroquinone (TBHQ) were purchased from Jingchun (Shanghai, China). Folin-Ciocalteu reagent and 2,4,6-tri(2-pyridyl)-s-triazine (TPTZ) was obtained from Fluka (Buchs, Switzerland). Dubelcco's Modified Eagle's Medium (DMEM) was purchased from Gibco/Invitrogen (Grand Island, NY, USA). The AB-8 resin was purchased from Sanxing Chemical Co. (Bengbu, China). Soybean oils without synthetic antioxidant added were provided by Jinyong Vegetable Oil Co. (Laiyang, China). All other chemicals and reagents were analytical grade.

Materials

Purple sweet potatoes were obtained from a local market in Ningbo, China. Briefly, 100.0 g of PSP powder was extracted with 3,200 mL of acid-ethanol (HCl, 1.5 mol/L) at 80 °C for 1 h. The supernatants were combined and concentrated, and then applied to AB-8 column chromatography. The effluents were collected, concentrated, and afford PSPAs extract (Zhang et al. 2016).

Characterization of PSPAs

PSPAs samples were analyzed according to our reported method by HPLC-ESI-MS/MS (Zhang et al. 2016). The separation was achieved on a Zorbax Stablebond Analytical SB-C18 column (4.6 mm $\times$ 250 mm, 5 μ m, Agilent, CA, USA). Elution was performed by mobile phase A (2% formic acid) and B (methanol), which was performed with a linear gradient as follows: 0–2 min, 10–20% B; 2–40 min, 20–55% B; 40–41 min, 55–80% B; 41–45 min, 80% B. The quantifications of anthocyanins were based on peak areas and calculated as equivalents of standards. The temperature of the column oven was 40 °C, flow rate was 0.6 mL/min, and the injection volume was 5 μ L.

Preparation of PSPAs-PC

PSPAs-PC was prepared according to the reported method (Maiti et al. 2007). The complex was prepared with phospholipids and PSPAs as a mass ratio of 2:1, they were placed in a 50 mL flask and 10 mL of ether was added as the reaction medium at 50 °C for 3 h. The resultant clear mixture was evaporated, filtered, dried under vacuum.

PSPAs content

The content of PSPAs in the phospholipids complex was determined as follows. Phospholipids complex of 5 mg were dissolved in 50 mL of solvent I (methanol:water = 40:60, v/v), a 20 μ L aliquot of the resulting solution was analyzed according to our reported Folin-Ciocalteu procedure (Zhang et al. 2012). A calibration curve of C3G was prepared, and PSPAs content was standardized against C3G and expressed as mg C3G equivalent per gram of sample on a dry-weight basis (DW).

Investigation of stability

In vitro gastrointestinal digestion of PSPAs and PSPAs-PC was performed according to previous reported method (Mills et al. 2008). In gastric conditions, samples including PSPAs and PSPAs-PC at the same final concentration of PSPAs were adjusted to pH 2 and then 1 mL pepsin (0.108 g) dissolved in HCl (0.1 M; 10 mL) was added and incubated at 37 °C. In small intestine conditions, the pH was increased to 7 and 2.5 mL pancreatin (80 mg) dissolved in NaHCO₃ (0.5 M; 10 mL) and 2.5 mL bile salt (500 mg) dissolved in NaHCO₃ (0.5 M; 10 mL) were added, and the incubation was continued at 37 °C. After 1, 3, 6, and 12 h, 50 μ L of each sample were taken and analyzed for the content of PSPAs. For the investigation of the stability of each anthocyanin in PSPAs and PSPAs-PC,

we have also measured their recoveries after incubation in mimic gastric and small intestine conditions for 12 h by HPLC–ESI–MS/MS.

Antioxidant activity

In this analysis, PSPAs and PSPAs-PC were at the same final concentration of PSPAs. The DPPH and ABTS free radical-scavenging activity were determined at 517 nm, 734 nm by 50%, respectively (Zhang et al. 2012; Stratil et al. 2006). The ability to reduce ferric ions of each sample was expressed as mmol FeSO₄ equivalents per gram of sample (DW) (Benzie and Strain 1996). In addition, ORAC is a typical hydrogen transfer-based assay with a competitive reaction between antioxidant compounds and fluorescein, which compete for the scavenging of peroxy radicals, generated in situ by thermal decomposition of azo compounds (Protti et al. 2017). Samples were suitably diluted with phosphate buffer (0.075 M, pH 7.4). 200 µL of diluted sample and 1500 µL of 9.6×10^{-8} M fluorescein solution (phosphate buffer) were incubated at 37 °C. 300 µL of AAPH (0.133 M, in phosphate buffer) were added in order to activate radical production. The emission intensity was recorded in the 495–700 nm range for 45 min ($\lambda_{\text{exc}} = 485$ nm). The calibration lines were constructed by plotting the net area under the curve (after 45 min) versus the concentration of the standard antioxidant. The results of this assay were expressed as µmol of vitamin C equivalents/g (VCE/g) by comparison with the calibration line obtained for the chosen standard.

Conjugated dienes (CD) value

PSPAs and PSPAs-PC were added in soybean oils at the final concentration of PSPAs at 5 mg/mL, while TBHQ, BHT and phospholipids were added in at a final concentration of 5 mg/mL and mixed thoroughly by a magnetic stirrer (Selon Scientific Instrument, Co., Ltd, Shanghai, China), and the same volume of soybean oils were added as the control. Then they were placed in an oven at 60 °C for 3, 6, 9, 12 and 15 d, respectively. After the incubation, each oil sample was weighed into a volumetric flask and dissolved in isooctane (Wettasinghe and Shahidi 1999), the absorbance was read at 234 nm. The CD value was calculated using the following formula:

$$\text{CD value} = \frac{\text{absorbance at 234 nm}}{(\text{concentration of oil in g/100 mL} \times \text{length of the cell in cm})}$$

In vitro cytotoxicity assay

Caco-2 cells were cultured in DMEM supplemented with 10% FBS, 1% non-essential amino acids, 2 mM glutamine,

100 U/mL of penicillin and 100 µU/mL of streptomycin. Cultures were maintained in a humidified incubator at 37 °C with 5% CO₂. Caco-2 cells were pipetted into a 96-well flat-bottom plate in a density of 10^5 cells/mL. After 12 h, non-adherent cells were removed by washing with serum free culture medium. Then fresh medium (100 µL/well, control group) or test sample (100 µL/well, PSPAs and PSPAs-PC at a final concentration of PSPAs at 25, 50, 100 and 200 µg/mL) were added to each well, and they were incubated for 24, 48 and 72 h, respectively. The CTC₅₀ (50% cytotoxic concentration) was determined by estimating mitochondrial synthesis using the tetrazolium assay (Zhang et al. 2014). After the incubation, MTT solution (10 µL/well, 5 mg/mL) was added to each well, and the plate was incubated for an additional 4 h at 37 °C. Finally, 100 µL of 10% sodium dodecyl sulfate (SDS) in 0.01 N HCl was added to each well, and the plate was kept overnight for the dissolution of formazan crystals. The absorbance of each well was measured at 570 nm. The inhibition rate was calculated according to the following formula:

$$\text{inhibition rate (\%)} = 100 - \left(\frac{\text{mean OD of individual test group}}{\text{mean OD of control group}} \right) \times 100$$

A concentration–response curve was generated using inhibition rate (%) and sample concentration (µg/mL). The CTC₅₀ value is calculated from the concentration–response curve.

Permeability studies

For the transport experiment, Caco-2 cells were split at subconfluent densities by using trypsin/EDTA and seeded at a density of 10^5 cells/cm² on transwell polycarbonate insert filters (1.12 cm² of surface, 0.4 µm of pore size, 12 mm of diameter, Corning Costar Corporation) in 12-well plates (Willenberg et al. 2015). After seeding, the Caco-2 cells were fed with 0.5 mL and 1.5 mL of culture medium in the apical (AP) and basolateral sides (BL) and the medium was refreshed every 2 days. The monolayer cells with transepithelial electrical resistance (TEER), which exceeded 400 Ω cm², were used for the permeability experiment. The integrity of Caco-2 monolayers was confirmed by the paracellular flux of lucifer yellow, which was < 1% per hour. Firstly, PSPAs and PSPAs-PC samples at a final concentration of PSPAs at 50 µg/mL was added to AP and HBSS was added to BL. Then 20 µL of solution from BL were withdrawn at 30, 60, 90, 120 min. For the efflux study, both samples at the same concentration as mentioned above were added to BL, and the procedures were opposite with the absorption study.

The apparent permeability coefficient (P_{app}) was calculated according to:

$$P_{app} = (\Delta C \cdot V / \Delta t) \times (1 / AC_0)$$

where ΔC is the concentration ($\mu\text{g/mL}$) of the receiver chamber, V is the volume of the receiver chamber (mL), Δt is the duration of the transport experiment (s), C_0 is the initial concentration of the donor chamber ($\mu\text{g/mL}$), and A is 4.67 cm^2 in our study. The efflux ratio (ER) was calculated according to:

$$ER = P_{app} (\text{BL} \rightarrow \text{AP}) / P_{app} (\text{AP} \rightarrow \text{BL})$$

Statistical analysis

Measurements were performed at least in triplicate. The results were analyzed by SPSS version 16.0 (SPSS Inc., Chicago, IL, USA). Any significant difference was determined by one-way analysis of variance (ANOVA) followed by the Tukey test for multiple comparisons considering difference statistically at $P < 0.05$.

Results and discussion

Identification and quantitative analyses of PSPAs

By analyzing PSPAs with HPLC–ESI–MS/MS, 10 anthocyanins were separated (Fig. 1). To verify the identity of

different PSPAs, fragmentation patterns of MS/MS (m/z) were used to compare with radical groups as we have reported (Zhang et al. 2016). As showed in Table 1, cyanidin was characterized as the major anthocyanidin, while acylated cyanidin of anthocyanidins was the most common modification of anthocyanins in tested samples.

Stability of PSPAs-PC

As shown in Table 2, the recovery of PSPAs-PC decreased with the increase in incubation time in mimic gastric and small intestine conditions in vitro. For PSPAs in small intestine conditions, significant differences existed during the incubation ($P < 0.05$), while no significant differences ($P > 0.05$) were shown after 3 h in gastric conditions. For PSPAs-PC, it showed similar trends, while its recovery was relatively higher at the same incubation time. The activity of polyphenols depends on many factors; e.g. reductive potential, metals chelating ability, pH of medium and solubility (Decker 1997). The relatively low recoveries of PSPAs in small intestine conditions may be due to the instability at neutral conditions (Kosińska et al. 2012). Anthocyanins are planar molecules, and their aglycone forms are structurally based on 2-phenylbenzopyrilium and with hydroxyl and methoxyl groups in different positions (Kay et al. 2017; Selma et al. 2009). Although anthocyanins are stable under acidic conditions, they are converting to colorless derivatives and then insoluble brown pigments during the storage. Their degradation

Fig. 1 Representative HPLC elution profiles of anthocyanins from PSP

Table 1 Anthocyanins identified by HPLC–ESI–MS/MS in PSP

Peak no.	t_R (min)	$[M]^+$ (m/z)	MS/MS (m/z)	Anthocyanin
1	20.48	463	287	Cyanidin-3-glucuronide
2	21.42	611	449/287	Cyanidin-3,5-diglucoside
3	23.26	1081	919/449	Cyanidin-3-(caffeoyl)(<i>p</i> -coumaroyl)diglucoside-5-glucoside
4	25.39	949	787/707/503/287	Cyanidin-3-feruloylsophoroside-5-glucoside
5	28.18	949	817/419/287	Cyanidin 3-(sinapoyl)diglucoside-5-xyloside
6	30.96	907	743/433/271	Pelargonidin 3-(acyl)diglucoside-5-glucoside
7	32.48	965	803/449/287	Cyanidin3-(<i>p</i> -hydroxybenzoyl)(oxaloyl)diglucoside-5-glucoside
8	33.87	773	611/449/287	Cyanidin-3-diglucoside-5-glucoside
9	35.18	1125	963/449/287	Cyanidin-3-diferuloylsophoroside-5-glucoside
10	36.11	1111	949/449/287	Cyanidin-3-caffeoylferuloylsophoroside-5-glucoside

mechanisms involve hydrolysis, oxidation and condensation with other polyphenols (Dai et al. 2009). Therefore, it is important to control the effect of assay conditions, mainly buffers and pH value.

Phyto-constituents have excellent water solubility; nevertheless, they are poorly absorbed. For the relative large molecular size, the ability of anthocyanins to cross the lipid-rich outer membrane of small intestine enterocytes is limited, which may be an important reason account for their low bioavailability. Therefore, the development of novel drug delivery system is necessary. The improvement of bioavailability could be achieved by delivery systems, which can enhance the solubility of polyphenols in aqueous intestinal fluids (Cuomo et al. 2011). Phospholipids have the ability to unify themselves into several assemblies where drugs could be effectively entangled (Fricker et al. 2010). In our previous studies, DSC curves of PSPAs-PC showed the endothermal peaks of anthocyanins and phospholipids were disappeared, meanwhile, the phase transition temperature was lower than that of phospholipids, which indicated they combined and formed hydrogen bonds (Pathan and Bhandari 2011). In the present experiment, PSPAs-PC was prepared by a simple and reproducible method successfully, and it showed relatively higher stability in both acidic and near neutral conditions.

Antioxidant activity of PSPAs-PC

DPPH scavenging activities of PSPAs and PSPAs-PC were shown in Fig. 2a. Both of them showed high scavenging activity on superoxide radical, and increased with the increasing of concentration. PSPAs-PC showed DPPH scavenging activity with an IC_{50} of 0.058 mg/mL. The value of IC_{50} (0.053 mg/mL) of PSPAs was less than PSPAs-PC, which exhibited high anti-oxidative property. The antioxidant abilities of samples determined by ABTS method were shown in Fig. 2b, and PSPAs exhibited considerably higher activity than PSPAs-PC. In addition,

their ABTS⁺ scavenging rates increased with the increasing of concentration. The ferric ion reducing activities of PSPAs and PSPAs-PC were shown in Fig. 2c. Similar to the results of DPPH and ABTS assay, the reducing activity for PSPAs-PC was relatively lower. The ORAC assay estimates the ability of removing peroxyl radicals through hydrogen transfer reactions. The antioxidant capacity values obtained by the ORAC assay ranged from 204.81 ± 15.36 to 252.66 ± 17.37 $\mu\text{mol/g VCE/g}$ for PSPAs-PC and PSPAs, respectively.

Although anthocyanins showed many biological activities, their absorptions in vivo are low. Studies have showed that anthocyanins are metabolized rapidly to different products after absorption, including glycosylation, sulfation and methylated derivatives (Ichianagi et al. 2008). To exert their biological properties, the health benefits of PSPAs may depend on their absorption in the intestine and the corresponding bioavailability. However, anthocyanin glucosides are the least stable compounds in the small intestine, likely due to the presence of β -glucosidase, the limited absorption of anthocyanins may also be associated with highly polar properties, due to hydroxyl groups as part of their structures (Steinert et al. 2008). Although PSPAs showed relatively higher antioxidant activity in vitro, the presence of hydroxyl groups in the structure led to poor stability and makes them difficult to dissolve in oil phase. In our study, PSPAs-PC was prepared by phospholipids and PSPAs, and showed satisfied antioxidant activity, which indicated the method of formulating such drug-phospholipids complex was effective and its potential application in manufacturing anthocyanins-rich food.

The effect of PSPAs on CD value

Edible fat-containing foods undergo oxidation during the processing and preservation, therefore it is desirable to control the processes of oxidation. The most effective means of preventing lipid oxidation is the addition of

Table 2 Recovery of PSPAs and PSPAs-PC after incubation in mimic gastric and small intestine conditions in vitro

	Recovery of PSPAs (%)				Recovery of PSPAs-PC (%)			
	1 h	3 h	6 h	12 h	1 h	3 h	6 h	12 h
Gastric	99.62 ± 0.29 ^a	98.12 ± 0.62 ^b	97.94 ± 0.78 ^b	97.88 ± 0.55 ^b	99.96 ± 0.21 ^a	98.75 ± 0.43 ^b	98.53 ± 0.36 ^b	98.51 ± 0.67 ^b
Small intestinal	64.31 ± 1.46 ^a	19.34 ± 0.85 ^b	7.43 ± 0.46 ^c	2.35 ± 0.12 ^d	77.23 ± 2.02 ^a	41.67 ± 2.42 ^b	16.29 ± 1.18 ^c	6.43 ± 0.32 ^d

Different lowercase alphabet letters indicate significant differences ($P < 0.05$) among different time points

antioxidants and food industry has been using synthetic antioxidants to inhibit lipid oxidation during the processing and storage. In recent years, with the awareness of the side effect of synthetic food additives, natural antioxidants have been popular for their safety, non-toxic and other advantages, while investigations on the application of natural antioxidants in oil protection has become a hot spot of oleo-chemicals.

CD values reflect the number of conjugated double bonds formed in the initial oxidation of unsaturated fatty acid. It can be seen in Fig. 3, the formation rate of diene value significantly decreased in oil samples treated by PSPAs-PC and synthetic antioxidants. For PSPAs-PC, its ability to inhibit the formation of conjugated double bonds was lower than TBHQ and BHT, but relatively higher than PSPAs and the control, which indicated its potential antioxidant capacity in the oil phase. Recently, investigations suggested the limitation of synthetic antioxidant use, including BHT and TBHQ with regard to their toxicity, and consumers showed large interest in natural sources of anti-oxidative compounds (Ryo et al. 2009). In previous studies, we have developed a method for analyzing tea catechins in vegetable oils (Zhang et al. 2015), and detected the antioxidant activity of (–)-epigallocatechin gallate-phospholipid complex (EPC) in vitro, the results suggested that the prepared EPC has strong antioxidant activity and can effectively inhibit the oxidation of soybean oil, while its inhibitory effect on oil oxidation was dose-dependent (Chen et al. 2015). Within the scope of concentration used in this experiment, in addition to its anti-oxidation activity, PSPAs-PC does not affect the color and brightness of soybean oil tested; therefore, it may be used widely in edible oils and/or oil-based foods.

Transepithelial transport study

The cytotoxicity of PSPAs and PSPAs-PC was studied by in vitro MTT assay, and their CTC₅₀ values were 89, 117 and 178 µg/mL, and 96, 125 and 191 µg/mL for 24, 48 and 72 h, respectively, which showed the complex we prepared had no cytotoxicity for Caco-2 cell line in this study.

The apparent permeability coefficient (P_{app}) of Caco-2 cell monolayer was related to intestinal absorption. P_{app} (AP → BL) and P_{app} (BL → AP) values reflect the absorption and excretion of bioactivities in the intestine, respectively. As shown in Fig. 4, the absorption and excretion of PSPAs-PC increased dramatically compared with PSPAs. It is necessary to observe the efflux ratio variations of a drug for its absorption and excretion occurring simultaneously, and low efflux ratios result in high bioavailability, and our results revealed the bioavailability of PSPAs-PC was relatively higher.

Fig. 2 Antioxidant activities of PSPAs and PSPAs-PC determined by DPPH free radical-scavenging (a), ABTS (b), and FRAP assay (c)

Without phospholipids aided, polyphenols were poorly transported, which is consistent with previous reports (Cardona et al. 2015). It might be due to their instability at non-acidic conditions; moreover, anthocyanins undergo glucuronidation, sulfation or methylation by phase II enzymes during the absorption across intestinal epithelial cells (Lambert et al. 2007). To exert the health effects of plant polyphenols, this crucial issue encompasses dissolution, absorption, distribution and disposition in target tissues, and numerous factors may contribute to the variation of their metabolism (Bohn 2014; Duda-Chodak et al. 2015). For increased bioavailability, natural products are

Fig. 3 Comparison of PSPAs, PSPAs-PC, phospholipids (PP), BHT and TBHQ to conjugated diene value

Fig. 4 The bilateral P_{app} values of PSPAs and PSPAs-PC

demanded to have a good balance between hydrophilicity and lipophilicity (Semalty et al. 2010). Recently, particular attentions have been paid to the improvement of the bioavailability of plant polyphenols in vivo, and it has been demonstrated the increased intestinal transport has a closely relationship with the promoting bioavailability of anthocyanins.

Plant polyphenols, slowly transported via the paracellular pathway, could be converted to a lipid-compatible molecular complex known as phytosome. Phospholipids have shown to be incorporated in cell membrane to replace cellular phospholipids and affect membrane fluidity (Zhang et al. 2017). The increase of PSPAs-PC transport may also be attributed to the encapsulation of compounds due to aggregates formation; in addition, the presence of phospholipids may protect PSPAs from degradation potentially (Semalty et al. 2010). The potential of novel biomaterials in the promotion of intestinal transport of phytochemicals has been confirmed (Cardona et al. 2015), and these investigations may be fascinated in manufacturing

functional foods, containing high content of anthocyanins with increased absorption properties.

Conclusion

PSPAs-PC showed potential antioxidant activity and relatively higher stability in mimic gastrointestinal digestion conditions. Meanwhile, the inhibitory effect of PSPAs-PC on the oxidation of soybean oil was better than the control, and the presence of phospholipid increased the transepithelial transport of PSPAs in the Caco-2 cellular model. The results showed the potential of phospholipids in the promotion of intestinal transport of PSPAs, and provided theoretical basis for the addition of natural antioxidant components in vegetable oils and the manufacture of functional foods.

Acknowledgements This work was sponsored by National Natural Science Foundation of China (31501473), Zhejiang Provincial Natural Science Foundation of China (LQ15C200003), Key Research and Development Project of Zhejiang Province (2017C02039), People-benefit Project of Ningbo (2015C10061), and K.C. Wong Magna Fund in Ningbo University.

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