

ORIGINAL ARTICLE

Valorization of lychee fruit peels waste for the sustainable production of value-added ingredient

Valorização do resíduo casca da fruta lichia para a produção sustentável de um ingrediente de valor agregado

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Abstract

Lychee (*Litchi chinensis* Sonn.) is an exotic fruit from Asia, recently introduced in Europe. With the increase in the world production of this fruit, many by-products are wasted during industrial processing, including their peels and seeds. Considering the utilization of fruit peels as edible material as a new functional ingredient, this work aimed to assess the nutritional and phytochemical characterization, as well as the antioxidant activity of two lychee peel extracts (alcoholic and hydroalcoholic). Also, it was evaluated the scavenging capacity against reactive species ($O_2^{\cdot-}$, H_2O_2 , NO^{\cdot}) of the two extracts. The peels, at a nutritional level, showed high levels of carbohydrates and total energy (76.8 ± 1.0 g/100 g and 331.4 kcal/100 g, respectively) and low protein and lipid content. Total phenolic and flavonoid contents were higher in alcoholic extraction (1578 mg GAE/g and 55.1 mg CE/g, respectively). The antioxidant activity evaluated *in vitro* by DPPH and FRAP assays was also higher in the ethanolic extract, verifying a positive correlation with the extractive yield of the bioactive compounds. In general, the ethanolic extracts of lychee peels showed higher antioxidant capacity and the maximum scavenging activity against reactive oxygen ($O_2^{\cdot-}$) and nitrogen species (NO^{\cdot}). The hydrogen peroxide scavenging activity observed in ethanolic extract (64 μ g/mL) was like the values obtained in the positive controls (quercetin and ascorbic acid, 62 μ g/mL, and 46 μ g/mL, respectively). These preliminary results suggest this undervalued ingredient is a promising source of bioactive compounds with high biological potential for the development of new products as functional ingredient, always focusing on sustainability.

Keywords: *Litchi chinensis*, peels; bioactive compounds; antioxidant activity; free radicals; sustainability; functional ingredient.



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Resumo

Lichia (*Litchi chinensis*) é uma fruta exótica originária da Ásia, recentemente introduzida na Europa. Com o aumento da produção mundial desta fruta, muitos subprodutos são desperdiçados durante o processamento industrial, incluindo-se cascas e sementes. Considerando a utilização de cascas de frutas como matéria comestível e possível ingrediente funcional, este trabalho teve como objetivo avaliar a caracterização nutricional e fitoquímica, bem como a atividade antioxidante de dois extratos (alcoólico e hidroalcoólico) de cascas de lichia. Também foi avaliada a capacidade sequestradora de espécies reativas ($O_2^{\cdot-}$, H_2O_2 , NO^{\cdot}) dos dois extratos. As cascas, em nível nutricional, apresentaram teores elevados de carboidratos e valor energético total ($76,8 \pm 1,0$ g/100 g e 331,4 kcal/100 g, respectivamente) e baixo teor de proteínas e lipídeos. Os teores de compostos fenólicos e flavonoides totais foram maiores na extração alcoólica (1.578 mg GAE/g e 55,1 mg CE/g, respectivamente). A atividade antioxidante avaliada *in vitro* pelos ensaios DPPH e FRAP foi, igualmente, superior no extrato etanólico, verificando-se uma correlação positiva com o rendimento extrator dos compostos bioativos. Em geral, os extratos etanólicos das cascas das lichias apresentaram maior capacidade antioxidante e máxima atividade sequestradora de espécies reativas de oxigênio ($O_2^{\cdot-}$) e nitrogênio (NO^{\cdot}). A atividade sequestradora de peróxido de hidrogênio observada no extrato etanólico (64 μ g/mL) foi semelhante aos valores obtido nos controles positivos (quercetina e ácido ascórbico, 62 μ g/mL e 46 μ g/mL, respectivamente). Estes resultados preliminares apontam este resíduo pouco valorizado como uma fonte promissora de compostos bioativos com alto potencial biológico para o desenvolvimento de novos produtos, como ingrediente funcional, sempre com foco na sustentabilidade.

Palavras-chave: Cascas de *Litchi chinensis*; compostos bioativos; atividade antioxidante; radicais livres; sustentabilidade; ingrediente funcional.

Highlights

- Lychee peels waste as a renewable source of bioactive compounds
- Valorization of lychee peels as a functional ingredient
- Green-extraction technique for the analysis of antioxidant activity
- Enhancement as a functional ingredient against free radical damage

1 Introduction

Litchi chinensis Sonn. (Sapindaceae) is a subtropical to tropical tree originating from China the largest producing country, followed by Thailand, India, and Vietnam (Pareek, 2016). However, nowadays it has spread to more than 20 countries all over the world, including Bangladesh, Indonesia, Philippines, Nepal, and South Africa, as well as Australia and the United States of America (USA) (Sun et al., 2021). As far as we know, in Brazil, this plant was introduced in 1810 and only started to be commercialized in 1970. Currently, the commercialization and consumption of this fruit has increased considerably, being São Paulo the largest producing state. According to Hajare et al. (2010), the trends of global trade into agricultural products that had not previously been considered in the world food schemes, promote new marketing opportunities for exotic crops like lychee fruit. Due to its attractive appearance, floral fragrance, and delicious taste, fresh lychee has been accepted by the entire world population, registering an increase in its consumption. Thus, consumption of fresh fruit in Europe is developing towards a more sustainable approach to production and industrial-scale technological processes.

Over the years, whole lychee fruits have been used not only as a food source but also for medicinal purposes. Nowadays, medicine has started to pay great attention to functional ingredients for food production, which displays an additional function related to health promotion or disease prevention. In general, the number of phytochemicals present in the peel and fruit seeds is significantly higher than in the

pulp, which enhances the isolation of bioactive compounds or nutraceutical ingredients from these residues for later application in food products (Moreira-Araújo et al., 2019; Emanuele et al., 2017). Previous studies reported several health benefits of lychee fruit that might be related to polysaccharides, and polyphenols as the main bioactive compounds present in edible and non-edible parts of the fruit (Contreras-Castro et al., 2022; Dike et al., 2021; Zeng et al., 2019). As is general knowledge, polysaccharides play crucial roles in food, pharmaceutical, and cosmetic industries as thickeners, stabilizers/gelling agents, emulsifiers, and texture modifiers (Souza et al., 2022). Hence, they can be formulated as nanoparticles, hydrogels, patches, lenses, and filaments, among others. Thus, polysaccharides are often combined with other natural (e.g., proteins) or synthetic polymers to improve their properties, notably their lower mechanical properties. Among them, bioactive polysaccharides derived from natural resources (mainly from food by-products and edible fungus) have attracted much attention owing to their noncytotoxic properties and beneficial pharmacological effects such as antioxidant capacity, immunomodulation, and antitumor activities (Liu et al., 2023; Shan et al., 2022; Sun et al., 2021). Also, polyphenols are gaining more attention due to their therapeutic effects and their potential technological applications. The supplement of these compounds in suitable concentrations can present promising effects in the prevention of several diseases such as diabetes (Addepalli & Suryavanshi, 2018), obesity (Bhandarkar et al., 2019), Parkinson's, Alzheimer's (Ali et al., 2019). Several activities like antiviral, antioxidant, anticoagulant, hepato-, and cardioprotective have been assigned to lychee pulp and its by-products (Carvalho et al., 2020; Mutha et al., 2018). Lychee peels are recognized to possess high contents of ascorbic acid, minerals, and phenolic compounds, including gallic acid, flavonoids (procyanidin B4, procyanidin B2, and epicatechin), and anthocyanins (cyanidin 3-*O*-rutinoside, cyanidin 3-*O*-glucoside, quercetin-3-*O*-rutinoside, and quercetin-3-*O*-glucoside) (Chukwuma et al., 2021; Jiang et al., 2021). These compounds can also be used to improve the physicochemical properties of starch, in the preservation of foods, such as natural dyes, prebiotic ingredients, hydrogels, and polyphenol nanocomplexes (Araújo et al., 2021). According to Queiroz et al. (2015), dried lychee peels possess higher antioxidant activity compared to fresh fruit pulp, which allows their use as a functional ingredient, like flour for example. In a subsequent study, the same authors reported that intake of lychee peel flour may attenuate weight gain, reduce body mass index, glucose, and the levels of triacylglycerols, total cholesterol, low-density lipoprotein, hepatic enzymes, and leptin, besides the percentage of hepatic lipids, liver lipid peroxidation and frequency of severe steatosis (Queiroz et al., 2018). Considering the studies mentioned above, this work aimed to highlight the lychee peels in terms of their nutritional and phytochemical composition. In addition, green-extraction solvents were used to evaluate antioxidant activity with DPPH and FRAP techniques. The scavenging capacity against reactive species ($O_2^{\cdot-}$, H_2O_2 , NO^{\cdot}) of fruit peel extracts was also evaluated, being a pioneering study, with no record of any study published to date.

2 Material and methods

2.1 Plant material

Fresh lychee fruits (~10 kg) were collected in October 2021 from a local market in Oporto, Portugal. Peels were manually separated, cut into small portions, and dried in an oven at 40°C for 48 h, and also grounded to get a fine powder using a Blender Moulinex type 320.2.00. Lychee peel powder was stored in plastic bottles hermetically sealed and protected from light.

2.2 Proximate analysis

Macronutrients (moisture, ash, fat, protein, and carbohydrates) were analysed following the Association of Official Analytical Chemists methods (Association of Official Analytical Chemists, 2012). The moisture content was instrumentally determined using an infrared moisture analyser (SMO 01, Scaltec

Instruments, Heiligenstadt, Germany). The ash content was determined by incineration. The crude fat was determined by using a Soxhlet apparatus. The protein content ($N \times 6.25$) was determined using the Kjeldahl procedure. To calculate the total carbohydrate content, the sum of the contents obtained for ash, crude fat, and protein was subtracted at 100 g. Energy value was calculated according to the general Atwater factors: Energy (kcal) = $4 \times (\text{g protein}) + 3.75 \times (\text{g carbohydrate}) + 9 \times (\text{g fat})$. Results were expressed as g per 100 g of dried mass.

2.3 Antioxidant profile

2.3.1 Extracts preparation

Powdered samples (~1 g) were extracted using two different solvents: 50 mL of ethanol, and ethanol-water (50:50 v/v) during 1 h, at 50 °C, on a heating plate (Mirak, Thermolyse, USA) under constant stirring (600 rpm) (Costa et al., 2014). Extracts were filtered, concentrated under vacuum, and then lyophilized. All extracts were stored at -20 °C for future analysis.

2.3.2 Total phenolic contents

Total phenolic contents were quantified, in triplicate, making small changes to the protocol described by Vinha et al. (2021). Briefly, 500 µL of each extract was mixed with 2.5 mL of Folin-Ciocalteu reagent (1:10) and 2.5 mL of sodium carbonate solution (7.5%, m/v). The mixture was incubated (15 min at 45 °C), followed by 30 min at room temperature. Absorbance was measured at 765 nm, using a Synergy HT microplate reader (BioTek Instruments, Synergy HT GEN5, USA). A calibration curve was prepared with gallic acid (0 - 100 mg/L, $R^2 = 0.9992$) and results were expressed as mg of gallic acid equivalents (GAE)/ g of dried weight.

2.3.3 Total flavonoid contents

Total flavonoid contents were determined in triplicate according to Vinha et al. (2016). 1 mL of each extract was mixed with 4 mL of distilled water and 300 µL of sodium nitrite (25%). After 5 min of incubation at room temperature, 300 µL of 10% AlCl_3 were added to the mixture. 2 mL of sodium hydroxide (1 M) and 2.4 mL of ultrapure water were also added. Absorbance measurements were performed at 510 nm, using a Synergy HT microplate reader (BioTek Instruments, Synergy HT GEN5, USA). A calibration curve was prepared with epicatechin (0-450 mg/L, $R^2 = 0.9986$), and results were expressed as mg of epicatechin equivalents (CE)/ g of dried weight

2.4 In vitro antioxidant activity

2.4.1 DPPH free radical scavenging assay

The DPPH assay was used to evaluate the free radical scavenging activity of lychees peel extracts. The reaction mixture was prepared directly on a 96 well plate between different sample concentrations (30 µL) and an ethanolic solution (270 µL) containing DPPH radicals (6×10^{-5} M) in each well. The reduction of the DPPH radical was observed at 517 nm at two-minute intervals, during 30 min. The concentration resulting in 50% inhibition (IC_{50}) of DPPH was compared with the standard (Vinha et al., 2016). All measurements were done in triplicate.

2.4.2 Ferric reducing antioxidant power (FRAP) assay

The reductive potential (ferric reducing antioxidant power; FRAP) was determined, in triplicate, based on the chemical reduction of Fe^{3+} to Fe^{2+} (Vinha et al., 2021). To 35 μL of each extract, 265 μL of FRAP reagent (10 parts of 300 mM sodium acetate buffer at pH 3.6, 1 part of 10 mM TPTZ solution, and 1 part of 20 mM $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ solution) were added and the reaction mixture was incubated at 37 °C, during 30 min before reading at 595 nm. Solutions of known Fe(II) concentrations ($\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$) were used to perform the calibration curve (linearity range: 25 - 1000 μM , $R^2 = 0.9971$). The reducing power was expressed as an equivalent concentration (EC) to that of 1 mM FeSO_4 .

2.5 (ROS/RNS) Scavenging Assays

2.5.1 Superoxide radical scavenging assay

The reduction of molecular oxygen (O_2) produces superoxide (O_2^-), which is the main precursor to most other reactive oxygen species. In this experience, O_2^- was generated by the NADH/PMS system, and the antioxidant quercetin was used as positive control. The O_2^- scavenging activity was determined by monitoring the effect of the studied extracts (31.25 to 1000 $\mu\text{g}/\text{mL}$) and positive control (31.25 to 1000 $\mu\text{g}/\text{mL}$) on the O_2^- -induced reduction of NBT at 560 nm after 2 min (Fontana et al., 2001). The results were expressed as the inhibition (IC_{50}) of the NBT reduction to diformazan.

2.5.2 Hydrogen peroxide scavenging assay

The H_2O_2 scavenging activity was measured by monitoring the effect of the tested extracts (31.25 to 1000 $\mu\text{g}/\text{mL}$) and positive controls (31.25 to 1000 $\mu\text{g}/\text{mL}$) on the H_2O_2 induced oxidation of lucigenin. Ascorbic acid was used as positive control, due to being a recognized antioxidant. Results were expressed as the inhibition (lychee peel extracts in IC_{50}) of the H_2O_2 induced oxidation of lucigenin (Pedraza-Chaverri et al., 2004).

2.5.3 Nitric oxide scavenging assay

Nitric oxide (NO) is a signaling molecule that plays a significant role in the prolongation of inflammation and immunological response. The NO scavenging activity was measured by monitoring the effect of the tested extracts (7.81 – 500 $\mu\text{g}/\text{mL}$) and positive control (quercetin: 0.06-2 $\mu\text{g}/\text{mL}$) on the NO-induced oxidation of non-fluorescent DAF-2 to the fluorescent triazol fluorescein (DAF-2T) (Ebrahimzadeh et al., 2008). Quercetin was used as a positive control since it inhibits the induction of nitric oxide synthase and is a naturally occurring direct scavenger of nitric oxide. The results were expressed as the inhibition (IC_{50}) of NO-induced oxidation of DAF- 2.

2.6 Statistical analysis

The statistical analysis was evaluated by one-way Analysis of Variance (ANOVA) using spss® version 18. Posthoc LSD was used to analyse the differences between the means. A p -value of $p < 0.05$ was statistically significant and results are expressed as mean \pm standard deviation. All analytical techniques were done in triplicate. The IC_{50} values were calculated by the formula $Y = 100 \cdot A1 / (X + A1)$, where $A1 = \text{IC}_{50}$, $Y = \text{response}$ ($Y = 100\%$ when $X = 0$), $X = \text{inhibitory concentration}$. The IC_{50} values were compared by paired t-tests and the antioxidant activity was expressed in terms of IC_{50} ($\mu\text{g}/\text{mL}$ concentration required to inhibit the radical formation by 50%).

3 Results & discussion

The proximate composition is an important criterion for determining nutritional benefits and food or food ingredient quality. Table 1 shows the results for the proximate composition of lychee fruit peel.

Table 1. Proximate analysis of lychee fruit peels, expressed in g/100 g dry weight.

Proximate analysis of lychee fruit peels	
Ash	2.5 ± 0.9*
Fat	1.8 ± 0.6
Protein	2.0 ± 0.7
Carbohydrates	76.8 ± 1.0
Total energy value	331.4 kcal/ 100 g 1386.6 kJ

*The values are expressed as mean ± SD of three parallel determinations on dry weight (dw) basis.

Regarding Table 1, the contents of carbohydrates and TEV were high (76.8 ± 1.0 g/100 g and 331.4 kcal/100 g, respectively) and a low protein and fat content were observed.

The high carbohydrate content is desirable since carbohydrate is a key class of naturally occurring organic compounds that are required for plant and animal life maintenance and offer raw materials for numerous industries. The carbohydrate content in lychee peels is within the range of values reported by Romelle et al. (2016) in six selected fruit peels. The low protein content (2.0%) found in lychee fruit peels can be interesting for the development of gluten-free products. Queiroz et al. (2018) have stated that lychee peel flour altered serum lipid levels in rats fed a high-cholesterol diet, aiding in the treatment of dyslipidaemia and hepatic steatosis, reinforcing its favourable benefits in lowering the risk of cardiovascular diseases. Indeed, it becomes necessary to broaden the use of food waste raw materials to produce gluten-free products, as well as to diversify the product range and enrich the nutrient content of meals. Thus, lychee peels can be considered a new functional ingredient to be incorporated as flour in the gluten-free foods sector. The ash content found in lychee peels (2.5%) can be considered a source of minerals. Similar ash content was reported in socotran pomegranate (*Punica protopunica* Balf. f.) peels (Shan et al., 2022), as well as in avocado (*Persea americana* Mill.) peels, tropical fruits grown in Brazil (Morais et al., 2017). Moreover, our results are within the values described by other authors in raw materials used as flours, specifically 0.6% (rice and maize flours), 2.25% of quinoa, 2.28% of tiger nut, 2.31% of buckwheat, and 0.34% of plantain flours (Culetu et al., 2021). Shukla et al. (2012) showed that lychee peels are an excellent source of carbohydrate (~81.1%), protein (~6.1%), and crude fiber (~4.327%), with low content in total fat (0.9%), presenting a similar nutritive value 356.917 kcal/100 g. Considering the importance of food for health maintenance and the few results on the benefits of the intake of lychee peel flour, a similar study described lower content of carbohydrates (59.7%) and higher content of protein (10.1%), total fat (7.1%) and similar total energy value (343.0 kcal/100 g) in the chemical composition of lychee peel flour (Queiroz et al., 2018). These findings are relevant to the use of these fruit peels as functional components because there is a growing exploitation possibility for fruit by-products, as well as increasing demand for natural ingredients with high nutritional content. In folk medicine and pharmacological studies, lychee and its by-products are related to some biological properties, including anticancer, anti-inflammatory, antifungal, antiviral, antioxidant, anticoagulant, antidiabetic, antihyperlipidemic, antihyperglycemic, hepato- and cardioprotective activities (Mir & Perveen, 2022; Huang et al., 2014; Jiang et al., 2013; Xu et al., 2011). Mutha et al. (2018) reported antioxidant, anti-inflammatory, and anti-microbial effects in the hydroalcoholic extract of lychee fruit peel. Bioactive compounds content and *in vitro* antioxidant activity were studied using two solvents free of toxicity, being the most suitable for maintaining green chemistry and optimal extraction in edible raw materials. Results are reported in Table 2.

Table 2. Phytochemicals content (total phenolics and total flavonoids) and antioxidant activity (DPPH and FRAP assays) obtained in ethanolic and hydroalcoholic lychee peel extracts.

Bioactive compounds	Ethanolic extract	Hydroalcoholic extract
Phenolics (mg GAE/ g)	1578 ± 4 ^a	1061 ± 1 ^b
Flavonoids (mg CE/ g)	55.1 ± 0.9 ^a	41.2 ± 3 ^b
<i>In vitro</i> antioxidant activity		
	Ethanolic extract	Hydroalcoholic extract
DPPH (IC ₅₀ , µg/ml)	2.288 ± 0.063 ^a	8.980 ± 0.110 ^b
FRAP (EC)	491.503 ± 3.603 ^a	288.2 ± 2.446 ^b

Values are expressed as mean ± standard deviation (n=3). ^{a,b}Different letters in the same row indicate significant differences between mean values ($p \leq 0.05$).

Currently, there are several analytical methods used to determine the antioxidant activity. Furthermore, the phytochemicals found in fruits are chemically varied, making it impossible to quantify each antioxidant component independently. For this reason, the total content of phenolics and flavonoids, which are part of the most representative groups of secondary metabolites described in plant matrices, were determined. According to the examination of the results in Table 2, the phenolic content appears to be much higher than the overall flavonoid content, and ethanol has a stronger extractive capability. In fact, sustainable and environmentally friendly recovery of bioactive compounds is attracting ever-increasing attention (Chuo et al., 2022), and the recovery of these metabolites from agro-industrial wastes using aqueous/alcohol extraction has become more popular (Reungoat et al., 2020; Chen et al., 2019; Flourat et al., 2019). The total phenolic content of lychee peels ethanolic extract (1578 mg EAG/g) was substantially higher than some other authors' values. For instance, Shukla et al. (2012) described 336.57 mg GAE/g, while Silva et al. (2020) reported 328.41 mg GAE/g. Furthermore, total flavonoid content was consistently lower than total phenolic content, which corresponds with other research, including those conducted on different fruit peels (Begam et al., 2020; Suleria et al., 2020). As observed in total phenolics, many authors reported lower flavonoid levels in lychee fruit peels, including lower values than those reported in this work. Lal et al. (2018) reported total flavonoid contents between 0.75 and 96.37 mg EC/g in hydroalcoholic extracts of 30 lychee peels genotypes, which are in accordance with our results.

Despite their antioxidant characteristics, bioactive compounds need to undergo enzymatic hydrolysis in the digestive tract or be metabolized by the bowel microbiota to be absorbed (Ribeiro et al., 2020). Their bioavailability and assimilability could depend on the capacity of extraction methods to improve their recovery (Coelho et al., 2020). Also, the ever-growing demand to recover bioactive compounds from by-products encourages a constant search for accessible extraction methods. This work used solvents free of toxicity that support the concept of green chemistry in the environmental impact. On the other hand, extraction profitability depends on the chemical affinity between the compounds and the nature of the solvent. As food applications, these bioactive compounds, due to their multipurpose characteristics, could be used to establish novel and functional foods, which highlight antioxidant activities. Regarding Table 2, it is verified that the antioxidant activity measured by the DPPH and FRAP assays were significantly higher in lychee peel ethanolic extracts, knowing that a low IC₅₀ value indicates the strongest ability of the extracts to act as DPPH scavenger. Results proved a positive correlation between the content of bioactive compounds and antioxidant activity. The importance of this study emphasizes, once again, the potential applications of lychee peels, an underutilized by-product with biological potential. Many studies reported that the contents of bioactive compounds are always higher in the peels than in fruit pulps. In fact, Guo et al. (2003) reported 2 to 27-fold higher antioxidant activity in peels than in fruit pulps. Thus, and in a conclusive context, it can be mentioned that some fruit peel fractions possess strong antioxidant activity and may be rich sources of antioxidant compounds (Rakariyatham et al., 2020). More research into the powerful antioxidants found in fruit by-products, as well as the processes by which they protect against disease development, is urgently needed.

Oxidative stress is linked to the development of inflammatory processes, which stimulate the appearance/development of several metabolic changes, chronic disorders, or cancers. Although free radicals play an essential role in many biological processes, it is known that excessive free radical production determines the structural modification of cellular proteins and the alteration of their functions, leading to cellular dysfunction and disruption of vital cellular processes (Pelegrino et al., 2020). Moreover, high amounts of these reactive oxygen and nitrogen species can lead to cell damage and apoptosis, contributing to many diseases (Aranda-Rivera et al., 2022). Given the foregoing and the significant potential for lychee peel utilization, the scavenging capacity against reactive species ($\cdot\text{O}_2^-$, H_2O_2 , $\text{NO}\cdot$) of lychee peels ethanolic and hydroalcoholic extracts was evaluated. As far as we know, no study has been done with this matrix, making it even more valuable in the development of functional compounds based on lychee peels. The scavenging effect of the ethanolic and hydroalcoholic extracts against superoxide radical, nitric oxide, and hydrogen peroxide were evaluated and compared with positive controls (quercetin and ascorbic acid) (Table 3).

Table 3. Superoxide radical ($\cdot\text{O}_2^-$), hydrogen peroxide (H_2O_2), and nitric oxide ($\cdot\text{NO}$) scavenging activities of ethanolic and hydroalcoholic extracts from lychee peels fruits and positive controls (quercetin and ascorbic acid).

Lychee peels extracts (IC_{50})	$\cdot\text{NO}$	H_2O_2	$\cdot\text{O}_2^-$
Alcoholic	54 ± 6	64 ± 0.3	205 ± 10
Hydroalcoholic	77 ± 5	155 ± 1	352 ± 5
Positive controls			
Quercetin	35 ± 2	62 ± 4	-
Ascorbic acid	-	46 ± 9	179 ± 3

IC_{50} = inhibitory concentration, *in vitro*, that decrease in 50% the number of reactive species in the tested media (mean ± standard error of the mean).

A comprehensive review of the results shows that the ethanolic extract clearly outperforms free radical scavenging. Regarding the $\text{O}_2^{\cdot-}$ -scavenging assay, ascorbic acid was the best scavenger (positive control). In this assay, quercetin presented no activity up to the highest concentration tested (1000 $\mu\text{g}/\text{mL}$). Regarding RNS-scavenging capacity, both extracts showed good scavengers to $\cdot\text{NO}$. Results showed that bioactivity is observed in both extracts (ethanolic and hydroalcoholic), being the alcoholic the one with the highest antiradical activity ($\text{IC}_{50} = 54 \mu\text{g}/\text{mL}$), similar to the control sample ($\text{IC}_{50} = 35 \mu\text{g}/\text{mL}$). To date, there are no identical studies that allow a comparison of results. So, further analysis with other radical species is suggested.

Although H_2O_2 is utilized as ROS in several *in vitro* experiments, it is unclear whether H_2O_2 really has a direct role in the experiments. Thus, hydrogen peroxide can be generated by the dismutation of $\text{O}_2^{\cdot-}$ or by the direct reduction of O_2 , and it is synthesized by enzymatic reactions (Nakai & Tsuruta, 2021). According to Kohen & Nyska (2002), the H_2O_2 is responsible for damage into heme proteins, with the release of iron, enzyme inactivation, and oxidation of DNA, lipids, -SH groups, and keto acids. In this determination, the results are promising. The hydrogen peroxide scavenging activity observed in lychee peel ethanolic extracts was like those obtained in controls (quercetin and ascorbic acid).

These scavenging activities are due to the high levels of polyphenols present in lychee fruit peels. According to Yang et al. (2018), lychee peels contain high contents of proanthocyanidins (known as tannins) which are acknowledged as natural agents to safely prevent acute damage and control chronic diseases at relatively low cost.

4 Conclusions

The results obtained in the present study indicate that lychee fruit peels exhibit free radical scavenging. The overall antioxidant activity of ethanolic and hydroalcoholic extracts might be attributed to lychee peels polyphenolic contents and phytochemical constituents. The findings of the present study suggest that this by-product might act as a potential source of natural antioxidant (functional ingredient) that could have great importance as therapeutic agents for biological systems susceptible to free radical mediated reactions. Thus,

this by-product can be used as an ingredient for the development of new products, not only in the food industry but also in pharmaceuticals and cosmetics areas, always focusing on the concept of sustainability. Despite this, by nutritional analysis, lychee peels stood out for their low energy value and low protein content, suggesting that this by-product may be useful as a value-add ingredient for specific foods (e.g., gluten-free flours).

Thus, further studies are suggested, namely in the incorporation of this by-product as an antioxidant and preservative agent in processed and packaged products.

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