

## Nutritional, Antioxidant, and Antidiabetic Potential of Wild Edible *Spondias lakonensis* Pierre Fruits

Vu Thi Huyen<sup>1</sup>, Lai Thi Ngoc Ha<sup>1</sup>, Bui Thi Hong Hue<sup>1</sup>, Le Thi Mai Linh<sup>2</sup>, Dang Cong Anh<sup>1</sup>, Nguyen Thi Hien<sup>2</sup> & Doan Thi Thuy Ai<sup>2\*</sup>

<sup>1</sup>Faculty of Food Science and Technology, Vietnam National University of Agriculture, Hanoi 12400, Vietnam

<sup>2</sup>Faculty of Natural Resources and Environment, Vietnam National University of Agriculture, Hanoi 12400, Vietnam

### Abstract

*Spondias lakonensis* Pierre is a wild edible fruit used in traditional medicine for the treatment of many diseases. In this study, the nutritional quality and biological activity of *S. lakonensis* fruits were investigated. The results of the nutritional components analyses were recorded for the vitamin C content ( $142.80 \pm 0.74$  mg/100 g) in the pulp, and total ash ( $2.88 \pm 0.07\%$  and  $7.25 \pm 0.19\%$ ), crude lipid ( $5.80 \pm 0.21\%$  and  $11.54 \pm 0.20\%$ ), and total protein ( $6.70 \pm 0.77\%$  and  $12.74 \pm 1.89\%$ ) in the pulp and seeds, respectively. At the suitable conditions for extracting polyphenol compounds (70% acetone at 30°C for 10min with a material/solvent ratio of 1/20 g mL<sup>-1</sup>), the value of the total polyphenol content (TPC) was  $12.32 \pm 0.07$  mg GAE g<sup>-1</sup> in the pulp and  $24.63 \pm 0.07$  mg GAE g<sup>-1</sup> in the seeds. The results of the DPPH tests indicated that the acetone extracts were good sources of antioxidants with IC<sub>50</sub> values of  $10.44 \pm 0.62$  µg mL<sup>-1</sup> (pulp) and  $29.20 \pm 1.76$  µg mL<sup>-1</sup> (seeds). The extracts also demonstrated strong  $\alpha$ -glucosidase inhibitory activities with IC<sub>50</sub> values of  $0.77 \pm 0.02$  µg mL<sup>-1</sup> in the pulp and  $10.24 \pm 0.15$  µg mL<sup>-1</sup> in the seeds. *S. lakonensis* fruits possess promising nutritional and pharmaceutical potential.

Received: August 18, 2024

Accepted: June 23, 2025

### Correspondence to

Doan Thi Thuy Ai  
dttai@vnua.edu.vn

### ORCID

Nguyen Thi Hien  
<https://orcid.org/0009-0004-6460-0563>

Doan Thi Thuy Ai  
<https://orcid.org/0009-0005-0300-0860>

Lai Thi Ngoc Ha  
<https://orcid.org/0000-0003-1020-3752>

Vu Thi Huyen  
<https://orcid.org/0000-0003-2174-9451>

Le Thi Mai Linh  
<https://orcid.org/0009-0003-1498-002X>

### Keywords

Anacardiaceae, *Spondias lakonensis*,  $\alpha$ -glucosidase, antioxidant, antidiabetic

### Introduction

*Spondias* is a genus belonging to the Anacardiaceae family. This genus is comprised of 18 species distributed widely in tropical regions around the world (Mitchell & Douglas, 2015). Previous studies on phytochemicals have shown the presence of various types of secondary compounds such as phenolics, flavonoids, sterols,

triterpenes, saponins, volatile compounds, amino acids, and polysaccharides in members of the *Spondias* genus (Tandon & Rastogi, 1976; Corthout *et al.*, 1991; Engels *et al.*, 2012; Olugbuyiro *et al.*, 2013; Lai *et al.*, 2014; Chaudhuri *et al.*, 2015; Pereira *et al.*, 2015; De Lima *et al.*, 2016). These plants have revealed useful bioactivities including cytotoxic, antimicrobial (Arif *et al.*, 2008; Muhammad *et al.*, 2011), antioxidant (Hazra *et al.*, 2008; Chalise *et al.*, 2010), anti-inflammatory (Da Silva Siqueira *et al.*, 2016), analgesic (Panda *et al.*, 2009), and  $\alpha$ -glucosidase inhibitory activities (Hoang *et al.*, 2015; Ojo *et al.*, 2018).

*Spondias lakonensis* Pierre is originally from Southern China, Cambodia, Laos, Vietnam, and Thailand, and can grow up to about 8-15m in height (Shaw & Forman, 1967). Ripe fruits of *S. lakonensis* are sweet and delicious, and stimulate digestion. A root decoction of *S. lakonensis* was also shown to improve weakness after childbirth (Hoang *et al.*, 2008). Despite *S. lakonensis* being used in traditional medicine, no chemical or bioactivity studies on *S. lakonensis* fruits have been reported. In this paper, we focused on extracting phenolic compounds, and evaluating the nutritional composition, antioxidant abilities, and  $\alpha$ -glucosidase inhibitory activities of *S. lakonensis* fruit extracts.

Plant phenolic compounds are commonly recognized as bioactive components associated with antioxidant properties and health benefits (Pierson *et al.*, 2012). It is well known that polyphenols are vulnerable to oxidative degradation, especially with prolonged exposure to heat, light, and oxygen. Therefore, in addition to factors such as agroclimatic conditions, organ and plant developmental stage, and their interaction with the genotype, the extraction conditions and methods can significantly impact the efficiency, stability, and yield of polyphenols (Rymbai *et al.*, 2023). However, there are currently no published data detailing the effectiveness of various extraction conditions for polyphenols from *S. lakonensis* fruit. This study examined the effects of various experimental parameters, namely extraction solvent, solvent/water ratio, material/solvent ratio,

extraction temperature, and extraction time, on the yield of polyphenols from *S. lakonensis* fruit. The findings will provide insights on extraction procedures and proper utilization of this wild edible fruit in value-added, processing, and pharmaceutical industries.

## Materials and Methods

### Plant materials

Fresh fruits of *S. lakonensis* (**Figure 1**) were collected in Gia Lam district, Hanoi, in 2019 at the coordinates of North at 21°1'15'' and East at 105°56'18''. The fruit samples were washed and freeze-dried at -47°C using a ModulyoD Freeze Dryer 230, and then stored at -18°C at the Department of Chemistry, Faculty of Environment, Vietnam National University of Agriculture.

### Chemicals

Folin-Ciocalteu reagent, 2,2-diphenyl-1-picryl-hydrazyl (DPPH),  $\alpha$ -glucosidase enzyme (CAS No 9001-42-7), *p*-nitrophenyl- $\alpha$ -D-glucopyranoside, 4-nitrophenol, and dimethyl sulfoxide were purchased from Sigma-Aldrich (USA). All solvents used in the extractions and other chemicals (gallic acid, ascorbic acid, sodium carbonate, sulfuric acid, and hydrochloric acid) were of analytical grade and purchased from China.

### Methods

#### Evaluation of the pulp

From the harvested fruits, a 200-g sub-batch of the entire fruit sample was weighed before shelling, after shelling, and after removing the pulp. The whole fruits, pulp, and seeds of *S. lakonensis* wild fruits are shown in **Figure 2**. The different parts were then freeze-dried for 48 hours and their masses were determined. The contents of the seeds and pulp were determined by the following equations:

$$\% \text{ pulp} = \frac{\text{weight of pulp}}{\text{weight of entire fruits}} \times 100 (\%)$$

$$\% \text{ seeds} = \frac{\text{weight of seeds}}{\text{weight of entire fruits}} \times 100 (\%)$$



**Figure 1.** Fruits of *S. lakonensis* on its peduncle



**Figure 2.** Fruits, dry pulp, and dry seeds

#### *Determination of the total ash content*

The total ash content was determined using the method described by TCVN 1-2017. The sample was washed with distilled water and dried in an air oven. An empty crucible was weighed, 2.0g of the dry sample was added, and the crucible with the sample was weighed again. The crucible was transferred to a muffle furnace and its temperature was maintained at 500°C for 5 hours. The process was completed when there was no black speck in the ash. The percentage of ash content on the dried sample was evaluated as:

$$\text{Ash content} = \frac{\text{weight of ash}}{\text{weight of dried sample}} \times 100 (\%)$$

#### *Major macronutrients determination*

##### *(i) Determination of the crude lipid content by the Soxhlet method*

The crude lipid content was determined by the Soxhlet method using *n*-hexane as the solvent (Carpenter & Ward, 2017). In short, the solvent was heated and volatilized, and then condensed above the sample. The solvent dropped onto the sample and extracted the fat. At 15-20min intervals, the solvent flowed through a siphon to

the heating flask to start the process again. Crude lipid content on the dried sample was measured as:

$$\begin{aligned} \text{Crude lipid content} \\ &= \frac{\text{weight of extract}}{\text{weight of dried sample}} \\ &\times 100 (\%) \end{aligned}$$

##### *(ii) Determination of the total protein content by the Kjeldahl method*

The total protein content was determined by the Kjeldahl method (International, 2009). A dried sample (0.2g) was placed in a Kjeldahl flask. Twenty milliliters of concentrated H<sub>2</sub>SO<sub>4</sub> was added along with 1.0g of a catalytic mixture containing potassium sulfate, cupric sulfate, and titanium dioxide. The flask was placed on a heater to start digestion until the mixture became clear. The digest was cooled and 100mL distilled water was added. Twenty milliliters of the digest was added to 10mL of 30% NaOH in a distillation tube. The distillate was then titrated by a standard 0.01 N HCl solution. The total protein content was measured as:

$$\begin{aligned} \text{Total protein content} \\ &= \frac{0.00014 \times (V_2 - V_1) \times 100 \times 6.25}{V \times m} (\%) \end{aligned}$$

where  $V_1$  and  $V_2$  are the volumes of 0.01 N HCl consumed for the blank and sample titration (mL),  $V$  is the volume taken for distillation (mL), and  $m$  is the weight of the dry sample (g).

### (iii) Determination of the ascorbic acid content

The ascorbic acid (Vitamin C) content was determined by iodometric titration (Silva *et al.*, 1999). A sample (5.0g) was weighed and placed into a conical flask, and 100mL of 1% HCl was added to dissolve the solid. The mixture was kept in the dark for 2 hours and then was filtered to get the extracted solution. A starch indicator (3-5 drops) was added to the extracted solution. This mixture was titrated with a 0.01 N iodine solution. The ascorbic acid content was calculated as:

$$\text{Vitamin C} = \frac{V_{I_2} \times 0.01 \times 176}{m} \times 100 \text{ (mg/100 g DW)}$$

where  $V_{I_2}$  is the volume of 0.01 N  $I_2$  consumed for sample titration (mL), and  $m$  is the weight of the sample (g).

### Extraction of polyphenol compounds

#### (i) Selection of the extraction procedures

The study on the polyphenol extraction process of the fruits of *S. lakonesis* was conducted in single-factor experiments. The factors changed were the extraction solvent (ethanol, acetone, methanol, and water), solvent/water ratio (50-100%, v/v), material/solvent ratio (1/10; 1/20; 1/25; 1/30 g mL<sup>-1</sup>), extraction temperature (20-50°C), and extraction time (5-40min). The experiments were independent of each other. The mixture of materials and solvents was centrifuged (Hermle Z300, Germany) at a speed of 3000rpm for 10min at room temperature.

#### (ii) Determination of the total polyphenol content

The total polyphenol content (TPC) was determined by the Folin-Ciocalteu method (Singleton & Rossi, 1965). A mixture of 2.5mL of Folin-Ciocalteu reagent (1/10 diluted) and 0.5mL of plant extract was shaken for 5min. Then 2mL of 7.5%  $\text{Na}_2\text{CO}_3$  was added and the mixture was kept in the dark for 2 hours.

Absorbance was monitored at 760nm using a HACH DR 3900 UV-VIS spectrophotometer. Results were expressed as mg of gallic acid equivalents per gram of dry weight of the sample (mg GAE/g DW).

### Evaluation of the biological activities of the fruit extracts of *S. lakonesis*

#### (i) Antioxidant activity of the fruit extracts

The antioxidant activity was examined by the DPPH test (Tabart *et al.*, 2009) using ascorbic acid as a reference. An extract was diluted in methanol and then 3mL of this extract and 1 mL of DPPH (0.1mM) were mixed in one reaction tube with a cap. The mixtures were set aside in dark conditions for about 30min, and the absorbance was measured at 517nm. The percentage of inhibition of DPPH free radicals of the sample was calculated using the equation:

$$\% \text{ Inhibition} = \frac{A_0 - A_x}{A_0} \times 100 (\%)$$

where  $A_0$  is the absorbance of the control (mixture of 3mL methanol and 1mL of DPPH), and  $A_x$  is the absorbance of the sample.

#### (ii) Assay for $\alpha$ -glucosidase inhibitory activity

The  $\alpha$ -glucosidase enzyme inhibitory was evaluated using the previously described method of Moradi-Afrapoli *et al.* (2012). Samples were dissolved in dimethyl sulfoxide. Two mL of a sample and 40mL of 0.5 U mL<sup>-1</sup>  $\alpha$ -glucosidase were mixed in 120mL of 0.1M phosphate buffer (pH 7.0). After 5min, 40mL of 5mM *p*-nitrophenyl- $\alpha$ -D-glucopyranoside was added to the mixture and incubated at 37°C for 30min. The absorbance was measured at 405nm using a microplate reader (Molecular Devices, Sunnyvale, CA). Acarbose was used as a reference.

### Statistical analysis

The experiments were replicated three times and data were expressed as means  $\pm$  SD on a dry basis. The value of  $\text{IC}_{50}$  was calculated using the GradPad prism 8.0. Statistical analysis was confirmed by analysis of variance (ANOVA) using Turkey's Test via Minitab 16.0. A *P*-value of  $<0.05$  was considered significant.

## Results and Discussion

### Nutrition composition

The results showed that the pulp percentages were recorded as  $72.13 \pm 2.92\%$  and  $58.84 \pm 2.08\%$  for fresh and dry fruits, respectively. Similarly, the seed contents were noted as  $27.87 \pm 2.92\%$  in fresh and  $41.16 \pm 2.08\%$  in dry fruits. The nutrition composition of the *S. lakonensis* fruits is shown in **Table 1**. The total ash, crude lipid, and total protein of the pulp were found to be lower than those of the seeds. The total ash values were  $2.88 \pm 0.07\%$  and  $7.25 \pm 0.19\%$  in the pulp and seeds, respectively. This residue reflects the overall mineral composition present in the biomass, making it an important quality parameter. Determining the total ash is a key aspect of biomass analysis for nutritional or compositional assessment. It was found that *S. lakonensis* has insignificant amounts of crude lipids in its pulp ( $5.80 \pm 0.21\%$ ) and seeds ( $11.54 \pm 0.20\%$ ). The total protein contents in the pulp and seeds were recorded as  $6.70 \pm 0.77\%$  and  $12.74 \pm 1.89\%$ , respectively. The protein level in the pulp of *S. lakonensis* was found to be higher than that of *S. pinnata* (L.) Kurz (Khomdram *et al.*, 2014). The pulp of *S. lakonensis* contained  $142.8 \pm 0.74$  mg/100g of ascorbic acid. This value currently reported was higher than those values

reported for *S. dulcis* (42mg/100g) and *S. pinnata* (21mg/100g) (Das *et al.*, 2015). The high value of ascorbic acid in *S. lakonensis* pulp makes it useful in the prevention of scurvy, bleeding gums, limb pain, and blindness.

### Polyphenol extraction

#### Effect of the extraction solvent

To extract polyphenol compounds from *S. lakonensis* fruits, the effects of solvents with different polarities on the polyphenol yield was studied first. The variations in values of the total polyphenol contents are shown in **Table 2**. Acetone was recognized as the most compelling dissolvable for extricating polyphenol substances, with ethanol, methanol, and water yielding progressively lower amounts. These discoveries suggest an inverse relationship between solvent polarity and TPC extraction productivity from *S. lakonensis* fruits, where less polar solvents result in higher extraction rates compared to their more polar counterparts. Acetone-water (70%, v/v) was the best solvent for the extraction of polyphenols from *S. lakonensis* with the TPC values of  $14.23 \pm 0.85$  mg GAE g<sup>-1</sup> and  $24.43 \pm 1.53$  mg GAE g<sup>-1</sup> in the pulp and seeds, respectively. Water as extraction solvent occupied the lowest place in the phenolic

**Table 1.** Nutritional composition of *S. lakonensis* fruits

Parameters	Pulp	Seeds
%	$72.13 \pm 2.92$	$27.87 \pm 2.92$
Ash (%)	$2.88 \pm 0.07$	$7.25 \pm 0.19$
Crude lipid (%)	$5.80 \pm 0.21$	$11.54 \pm 0.20$
Total protein (%)	$6.70 \pm 0.77$	$12.74 \pm 1.89$
Ascorbic acid (mg/100 g fresh weight)	$142.80 \pm 0.74$	-

Note: Values are shown as mean  $\pm$  standard deviation of three extractions.

**Table 2.** Effect of the extraction solvents on the TPC (mg GAE g<sup>-1</sup> DW)

Solvent	Pulp	Seeds
70% Acetone	$14.23^a \pm 0.85$	$24.43^a \pm 1.53$
70% Methanol	$6.30^b \pm 0.15$	$16.75^b \pm 0.82$
70% Ethanol	$7.44^{bc} \pm 0.53$	$16.44^b \pm 0.51$
Water	$5.87^c \pm 0.54$	$6.76^c \pm 0.08$

Note: Values are shown as mean  $\pm$  standard deviation of three extractions, columns labeled with different letters (a, b, or c) indicate statistically significant differences ( $P < 0.05$ ).

extraction yields with the TPC values of  $5.87 \pm 0.54$  mg GAE g<sup>-1</sup> in the pulp and  $6.76 \pm 0.08$  mg GAE g<sup>-1</sup> in the seeds. The variations in the extract yields from *S. lakonensis* fruits using different solvents might be explained by the differences in polarity of the different compounds in the samples (Babbar *et al.*, 2014). As the extraction with acetone gave the highest TPC values from the fruits of *S. lakonensis*, acetone-water was chosen as the solvent for the following steps.

#### *Effect of the solvent/water ratio*

Water-acetone mixtures at different ratios (50-100%, v/v) were used as the extraction solvents in this study. The ratios had an observed effect on the TPC values (Table 3) ( $P < 0.05$ ). The TPC values increased as the acetone concentration increased, reaching the maximum TPC values of  $12.13 \pm 0.46$  mg GAE g<sup>-1</sup> and  $25.33 \pm 0.80$  mg GAE g<sup>-1</sup> in the pulp and seeds, respectively, with the 70% (v/v) mixture. Then, the TPC decreased when the acetone concentration increased to 100%. Many other studies have also used acetone to extract polyphenol compounds. For example, the TPC of eight cowpea genotypes were from 15.05 to 19.99 mg GAE g<sup>-1</sup> after being extracted with 70% acetone (Yusnawan *et al.*, 2021). Therefore, this result shows that the polyphenols in the fruits of *S. lakonensis* have a

polarity corresponding to 70% acetone, and this solvent was used for the evaluation of the other factors in the extraction process.

#### *Effect of the material/solvent ratio*

The impact of the material/solvent ratio on the extraction of polyphenol compounds from the fruits of *S. lakonensis* was evaluated with five ratios (1/10, 1/20, 1/25, 1/30 g mL<sup>-1</sup>) over a 10 min extraction period with a 70% acetone solution at 30°C. The amounts of TPC extracted are presented in Table 4. The results of the one-way analysis of variance showed that the material/solvent ratio showed significant differences among the ratios studied ( $P < 0.05$ ). The TP contents increased when the ratio increased from 1/10 to 1/20 g mL<sup>-1</sup> and then remained fairly constant at the 1/20, 1/25, and 1/30 g mL<sup>-1</sup> ratios. This phenomenon could be explained in that increases in the volume of the solvents led to easier extractions of the phenolic compounds. However, large amounts of the solvents also dissolved other components, and affected the dissolution of the polyphenol compounds components. According to our results, a material-to-solvent ratio of 1/20 g mL<sup>-1</sup> was chosen for evaluating the other parameters.

**Table 3.** Effect of the solvent/water ratio on the TPC (mg GAE g<sup>-1</sup> DW)

Solvent/water (v/v)	Pulp	Seed
50	$6.14^c \pm 0.83$	$20.97^{bc} \pm 0.91$
60	$10.44^b \pm 0.72$	$22.48^b \pm 0.70$
70	$12.13^a \pm 0.46$	$25.33^a \pm 0.80$
80	$9.47^b \pm 0.29$	$18.99^c \pm 0.75$
100	$4.14^d \pm 0.52$	$12.60^d \pm 0.57$

Note: Values are shown as mean  $\pm$  standard deviation of three extractions, columns labeled with different letters (a, b, c, or d) indicate statistically significant differences ( $P < 0.05$ ).

**Table 4.** Effect of the material/solvent ratio on the TPC (mg GAE g<sup>-1</sup> DW)

Material/solvent (g mL <sup>-1</sup> )	Pulp	Seeds
1/10	$8.99^b \pm 0.63$	$20.69^b \pm 0.45$
1/20	$12.94^a \pm 0.21$	$23.38^a \pm 0.66$
1/25	$12.73^a \pm 0.71$	$24.17^a \pm 0.88$
1/30	$12.29^a \pm 0.55$	$24.11^a \pm 0.65$

Note: Values are shown as mean  $\pm$  standard deviation of three extractions, columns labeled with different letters (a or b) indicate statistically significant differences ( $P < 0.05$ ).

*Effect of the extraction temperature*

Temperature has a strong influence on the extraction of secondary compounds in general and polyphenol compounds in particular. As the temperature increases, the diffusion rate increases, and therefore the TPC extracted from the material increases. However, if the temperature rises too high, some phenolics may be degraded during the extraction process (Chirinos *et al.*, 2007). In this experiment, the polyphenol compounds were extracted with 70% acetone and the material/solvent ratio of 1/20 g mL<sup>-1</sup> for 10min at different temperatures from 20 to 50°C. **Table 5** presents the TPC values obtained at the different extraction temperatures. The statistical analysis of the TPC showed that the values increased when the extraction temperature increased from 20°C to 30°C. There were no statistically significant differences among the TPC at extraction temperatures ranging from 30 to 50°C ( $P < 0.05$ ). This result may be explained by the small particles of the material that facilitate the extraction of polyphenol compounds (Pinelo *et al.*, 2007; Kossah *et al.*, 2010). The temperature of 30°C was chosen for continuous experiments.

*Effect of the extraction time*

The effect of time on the extraction of polyphenols from the fruits of *S. lakonensis* is

shown in **Table 6**. The results showed that the extraction of polyphenols increased with the increase of time from 5min to 10min. However, no significant differences ( $P < 0.05$ ) were observed among the TPC with extraction times from 10 to 40min. This finding is consistent with previous research on the extraction of phenolic compounds from plants. For example, polyphenols were extracted for 30min from the leaves of *Gynura procumbens* (Lour.) Merr. (Pham *et al.*, 2016) and *Annona muricata* Linn. (Nguyen *et al.*, 2020), and for 40min from the aerial parts of *Vernonia amygdalina* (La *et al.*, 2017). Furthermore, the extraction kinetics of phenolics from *Inga edulis* leaves was shown to be characterized by two distinct phases: an initial rapid phase during the first 10min, followed by a slower phase that accounted for the remainder of the extraction time (Silva *et al.*, 2007). Therefore, 10min was suitable for the extraction of polyphenols from the fruits of *S. lakonensis*.

*Selected conditions*

The suitable extraction conditions for carrying out the determination of TPC of the fruits of *S. lakonensis* found from single-factor experiments were 70% acetone at 30°C for 10min with a solvent/material ratio of 20 mL g<sup>-1</sup>. Under these conditions, the TPC values were determined to be  $12.32 \pm 0.07$  mg GAE g<sup>-1</sup> DW

**Table 5.** Effect of the extraction temperature on the TPC (mg GAE g<sup>-1</sup> DW)

Temperature (°C)	Pulp	Seeds
20	8.65 <sup>b</sup> ± 0.27	13.87 <sup>b</sup> ± 0.62
30	12.59 <sup>a</sup> ± 0.94	23.71 <sup>a</sup> ± 0.87
40	12.63 <sup>a</sup> ± 0.51	23.77 <sup>a</sup> ± 0.63
50	12.86 <sup>a</sup> ± 0.08	24.51 <sup>a</sup> ± 0.59

Note: Values are shown as mean ± standard deviation of three extractions, columns labeled with different letters (a or b) indicate statistically significant differences ( $P < 0.05$ ).

**Table 6.** Effect of the extraction time on the TPC (mg GAE g<sup>-1</sup> DW)

Extraction time (min)	Pulp	Seeds
5	9.00 <sup>b</sup> ± 0.39	14.67 <sup>b</sup> ± 0.84
10	12.00 <sup>a</sup> ± 0.43	23.76 <sup>a</sup> ± 0.92
20	11.67 <sup>a</sup> ± 0.91	24.50 <sup>a</sup> ± 0.80
30	11.88 <sup>a</sup> ± 0.49	24.69 <sup>a</sup> ± 0.60
40	12.11 <sup>a</sup> ± 0.58	24.31 <sup>a</sup> ± 0.89

Note: Values are shown as mean ± standard deviation of three extractions, columns labeled with different letters (a or b) indicate statistically significant differences ( $P < 0.05$ ).



and  $24.63 \pm 0.07$  mg GAE g<sup>-1</sup> DW in the pulp and seeds, respectively. When compared with other fruits, the pulp of *S. lakonensis* fruits had a higher total phenolic content than *Rhodomyrtus tomentosa* fruits (Lai *et al.*, 2015), but had a total phenolic content comparable to that of berry fruits, which are known as good sources of phenolic compounds (Wu *et al.*, 2004). The polyphenol-rich extracts of the fruits of *S. lakonensis* were used to evaluate their antioxidant and alpha-glucosidase inhibitory activities.

### Biological activities of the fruit extracts of *S. lakonensis*

#### Antioxidant activity

The acetone extracts of the *S. lakonensis* fruits were tested for their antioxidant activities using DPPH. The extracts contained antioxidant compounds, which donated a hydrogen proton to the lone pair electron of the DPPH radicals. The results showed that the extracts had potential activities with IC<sub>50</sub> values of  $10.44 \pm 0.62$  µg mL<sup>-1</sup> (pulp extract) and  $29.20 \pm 1.76$  µg mL<sup>-1</sup> (seed extract), which were compared to ascorbic acid in the control test ( $11.85 \pm 0.14$  µg mL<sup>-1</sup>). In comparison, 90 Vietnamese medicinal plant extracts were reported to having DPPH IC<sub>50</sub> values ranging from 4.9 to more than 100 µg mL<sup>-1</sup> (Phan *et al.*, 2012), and wild edible fruits from Eastern Himalaya showed antioxidant activities with IC<sub>50</sub> values ranging from 0.17 to 0.67 mg mL<sup>-1</sup> (Rymbai *et al.*, 2013; 2023), while the pulp extract of *S. lakonensis* exhibited significantly stronger antioxidant activity. Although the strong activity in the DPPH radical scavenging assay may be the result of the high level of TPC in the plant extracts, the TPC in the acetone extract from the seeds was higher than that of the acetone extract from the pulp, whilst its antioxidant activity was lower in this study. Thus, it could be explained that phenolic compounds are highly determinative in the antioxidant activity of this fruit, and at the same time, it is necessary to characterize the phenolic compounds in this fruit to understand exactly which polyphenol compound acts as a strong antioxidant. Furthermore, although the *S. lakonensis* fruits contained lower polyphenol values than many of

the 90 plant extracts reported in the previous study, such as *Perilla ocymoides* L., *Piper lolot* C. DC., *Plantago asiatica* L., etc., its DPPH scavenging capacity was higher (Phan *et al.*, 2012). This could be explained by differences in the phenolic profile of this fruit. Our findings on the total phenolic content and antioxidant capacity of *S. lakonensis* fruits highlight its potential as an undiscovered source of phenolic antioxidants. This fruit warrants further investigation for its potential health benefits and its promising applications in the fields of food and pharmaceutical technology.

#### α-Glucosidase inhibitory activity

The α-glucosidase inhibitory activity of the fruit extracts was evaluated. The IC<sub>50</sub> values of the pulp and seed extracts were  $0.77 \pm 0.02$  µg mL<sup>-1</sup> and  $10.24 \pm 0.15$  µg mL<sup>-1</sup>, respectively, while that of acarbose was  $176.55 \pm 4.09$  µg mL<sup>-1</sup>. The IC<sub>50</sub> value of the acetone extract from the pulp of this fruit was lower than those reported for *S. cytherea* fruit ( $110.81 \pm 0.86$  µg mL<sup>-1</sup>) (Hoang *et al.*, 2015) and *S. mombin* leaves ( $12.05 \pm 0.02$  µg mL<sup>-1</sup>) (Ojo *et al.*, 2018). Previous studies have indicated that plant extracts could be potential agents to manage hyperglycemia via α-glucosidase inhibitory activity (Abesundara *et al.*, 2004; Iwai *et al.*, 2006; Qaisar *et al.*, 2014; Liu *et al.*, 2016; El-Manawaty & Gohar, 2018; Joycharat *et al.*, 2018). Numerous studies have indicated that phenolic compounds may be involved and contribute significantly to the α-glucosidase inhibitory activity of plants. However, in this study, the polyphenol content was lower than that of many other plant species. Thus, other bioactive compounds in *S. lakonensis* might also play a role in its α-glucosidase inhibitory effect, beyond just the phenolic compounds. The observed biological activity of these extracts is likely due to the presence of various secondary phytochemicals.

### Conclusions

Our present study suggests that *S. lakonensis* fruits possess promising nutritional and pharmaceutical potential. With high levels of TPC in both the pulp and seeds ( $12.32 \pm 0.07$  and  $24.63$



$\pm 0.07$  mg GAE  $g^{-1}$ ), the fruit exhibited strong antioxidant activity in the DPPH assays as well as a significant  $\alpha$ -glucosidase inhibitory effect. The considerable content of phenolic compounds of the fruit extracts may be linked to their bioactivities. Further chemical investigations and evaluations of the bioactivities of isolated compounds from this wild plant should be conducted to provide a new source of nutritious food with strong antioxidant and antidiabetic properties from wild edible fruits in Vietnam.

## Acknowledgements

The authors wish to thank Dr. Do Van Truong for his aid in botanical determinations.

## References

- Abesundara K. J., Matsui T. & Matsumoto K. (2004).  $\alpha$ -Glucosidase inhibitory activity of some Sri Lanka plant extracts, one of which, *Cassia auriculata*, exerts a strong antihyperglycemic effect in rats comparable to the therapeutic drug acarbose. *Journal of Agricultural and Food Chemistry*. 52(9): 2541-2545.
- Arif M., Zaman K., Fareed S. & Hussain M. (2008). Antibacterial, antidiarrhoeal and ulcer-protective activity of methanolic extract of *Spondias mangifera* bark. *International Journal of Health Research*. 1(4): 177-192.
- Babbar N., Oberoi H., Sandhu S., and Bhargav V. (2014). Influence of different solvents in extraction of phenolic compounds from vegetable residues and their evaluation as natural sources of antioxidants. *Journal of Food Science and Technology*. 51: 2568-2575.
- Carpenter C. & Ward R. (2017). *Food Analysis Laboratory Manual*. Springer: Luxembourg.
- Chalise J. P., Acharya K., Gurung N., Bhusal R. P., Gurung R., Skalko-Basnet N. & Basnet P. (2010). Antioxidant activity and polyphenol content in edible wild fruits from Nepal. *International Journal of Food Sciences and Nutrition*. 61(4): 425-432.
- Chaudhuri D., Ghate N. B., Singh S. S. & Mandal N. (2015). Methyl gallate isolated from *Spondias pinnata* exhibits anticancer activity against human glioblastoma by induction of apoptosis and sustained extracellular signal-regulated kinase 1/2 activation. *Pharmacognosy Magazine*. 11(42): 269-276.
- Chirinos R., Rogez H., Campos D., Pedreschi R. & Larondelle Y. (2007). Optimization of extraction conditions of antioxidant phenolic compounds from mashua (*Tropaeolum tuberosum* Ruiz & Pavón) tubers. *Separation and Purification Technology*. 55(2): 217-225.
- Corthout J., Pieters L., Claeys M., Berghe D. V. & Vlietinck A. (1991). Antiviral ellagitannins from *Spondias mombin*. *Phytochemistry*. 30(4): 1129-1130.
- Da Silva Siqueira E. M., Félix-Silva J., De Araújo L. M. L., Fernandes J. M., Cabral B., Gomes J. A. d. S., De Araújo Roque A., Tomaz J. C., Lopes N. P. & De Freitas Fernandes-Pedrosa M. (2016). *Spondias tuberosa* (Anacardiaceae) leaves: profiling phenolic compounds by HPLC-DAD and LC-MS/MS and in vivo anti-inflammatory activity. *Biomedical Chromatography*. 30(10): 1656-1665.
- Das K., Roy D., Nandi P., Kundu S. & Dutta P. (2015). *Spondias*-an underutilized potential fruit crop of West Bengal. III International Symposium on Underutilized Plant Species. 1241: 51-56.
- De Lima E. Q., De Oliveira E. & De Brito H. R. (2016). Extraction and characterization of the essential oils from *Spondias mombin* L. (Caj), *Spondias purpurea* L. (Ciriguela) and *Spondia* ssp (Cajarana do sertão). *African Journal of Agricultural Research*. 11(2): 105-116.
- El-manawaty M. & Gohar L. (2018). In vitro  $\alpha$ -glucosidase inhibitory activity of Egyptian plant extracts as an indication for their antidiabetic activity. *Asian Journal of Pharmaceutical and Clinical Research* 11(7): 360-367.
- Engels C., Gräter D., Esquivel P., Jiménez V. M., Gänzle M. G. & Schieber A. (2012). Characterization of phenolic compounds in jocote (*Spondias purpurea* L.) peels by ultra high-performance liquid chromatography/electrospray ionization mass spectrometry. *Food Research International*. 46(2): 557-562.
- Hazra B., Biswas S. & Mandal N. (2008). Antioxidant and free radical scavenging activity of *Spondias pinnata*. *BMC Complementary and Alternative Medicine*. 8(1): 1-10.
- Hoang L. S., Phạm H. N. & Trần V. M. (2015). Antioxidant and  $\alpha$ -glucosidase inhibitory activity of *Spondias cytherea* fruit extract. *Journal of Science*. 3(3): 54-59.
- Hoang V. S., Pieter B., & Kejler P. J. A. (2008). Traditional medicinal plants in Ben En national park, VietNam. *Blumea*. 53: 569-601.
- International A. 2009. AOAC Official Method 2001.11 Protein (crude) in animal feed, forage (plant tissue), grain and oil seeds. AOAC International Gaithersburg, MD.
- Iwai K., Kim M.-Y., Onodera A. & Matsue H. (2006).  $\alpha$ -Glucosidase Inhibitory and Antihyperglycemic Effects of Polyphenols in the Fruit of *Viburnum dilatatum* Thunb. *Journal of Agricultural and Food Chemistry*. 54(13): 4588-4592.
- Joycharat N., Issarachote P., Sontimuang C. & Voravuthikunchai S. P. (2018).  $\alpha$ -glucosidase inhibitory activity of ethanol extract, fractions and purified compounds from the wood of *Albizia myriophylla*. *Natural Product Research*. 32(11): 1291-1294.
- Khomdram S., Arambam S. & Devi G. S. (2014). Nutritional profiling of two underutilized wild edible fruits *Elaeagnus pyrifolia* and *Spondias pinnata*. *Annals of Agricultural Research*. 35(2): 129-135.
- Kossah R., Nsabimana C., Zhang H. & Chen W. (2010). Optimization of extraction of polyphenols from Syrian sumac (*Rhus coriaria* L.) and Chinese sumac (*Rhus typhina* L.) fruits. *Research Journal of Phytochemistry*. 4(3): 146-153.
- La T. H., Tran T. M. N., Nguyen T. T., & Do M. P. N., (2017). Effect of extraction on total polyphenol content and antioxidant activity of bitter leaf (*Vernonia amygdalina*). *Vanhien University of Science*. 5(4): 93-99.
- Lai T. K., Acharya K., Chatterjee S. & Sherpa N. L. (2014). Antipseudomonal Ergosteryl Triterpenes from the Paste

- of *Spondias pinnata* kruz. Bark, Pre-Treated with Curd-Brew. International Journal of Pharmaceutical Sciences Review and Research. 28(1): 143-146.
- Lai T.N.H., Andre C., Rogez H., Mignolet E., Nguyen T.B.T., Larondelle Y. (2015). Nutritional composition and antioxidant properties of the sim fruit (*Rhodomyrtus tomentosa*). Food Chemistry. 168: 410-416.
- Liu S., Yu Z., Zhu H., Zhang W. & Chen Y. (2016). *In vitro*  $\alpha$ -glucosidase inhibitory activity of isolated fractions from water extract of Qingzhuan dark tea. BMC Complementary and Alternative Medicine. 16(1): 378-385.
- Mitchell J. D. & Douglas C. (2015). A revision of *Spondias* L. (Anacardiaceae) in the Neotropics. PhytoKeys. 55: 1-92. DOI: 10.3897/phytokeys.55.8489.
- Moradi-Afrapoli F., Asghari B., Saeidnia S., Ajani Y., Mirjani M., Malmir M., Bazaz R. D., Hadjiakhoondi A., Salehi P. & Hamburger M. (2012). *In vitro*  $\alpha$ -glucosidase inhibitory activity of phenolic constituents from aerial parts of *Polygonum hyrcanicum*. DARU Journal of Pharmaceutical Sciences. 20(1): 37-42.
- Muhammad A., Rahman M., Kabir A., Kabir S. & Hossain M. (2011). Antibacterial and cytotoxic activities of *Spondias pinnata* (Linn. f.) Kurz fruit extract. Antibacterial and cytotoxic activities of *Spondias pinnata* (Linn. f.) Kurz fruit extract. 2(2): 265-267.
- Nguyen V. T., Nguyen N. Q., Pham N. T., Nguyen M. T., Nguyen T. N. Q., Le V. M., Nguyen T. X. L., Nguyen T. T. T., Nguyen T. T. N., & Huynh L. T., (2020). Effect of extraction conditions on total polyphenol and flavonoid content of Soursop leaves (*Annona muricata* Linn.). Nguyen Tat Thanh University Journal of Science and Technology. 9: 54-56.
- Ojo O. A., Afon A. A., Ojo A. B., Ajiboye B. O., Oyinloye B. E. & Kappo A. P. (2018). Inhibitory effects of solvent-partitioned fractions of two nigerian herbs (*Spondias mombin* Linn. and *Mangifera indica* L.) on  $\alpha$ -amylase and  $\alpha$ -glucosidase. Antioxidants. 7(6): 73-83.
- Olugbuyiro J., Moody J. & Hamann M. (2013). Phytosterols from *Spondias mombin* Linn with antimycobacterial activities. African Journal of Biomedical Research. 16(1): 19-24.
- Panda B., Patra V., Mishra U., Kar S., Panda B. & Hati M. (2009). Analgesic activities of the stem bark extract of *Spondias pinata* (Linn. f) Kurz. Journal of Pharmacy Research. 2(5): 825-827.
- Pereira C., Oliveira L. L. d., Gonçalves R., Amaral A. C. F., Kuster R. M. & Sakuragui C. M. (2015). Phytochemical and phylogenetic analysis of *Spondias* (Anacardiaceae). Química Nova. 38(6): 813-816.
- Pham T. K. Q., Nguyen V. M., & Nguyen T. H., (2016). Effect of extraction conditions on polyphenol content and antioxidant activity of the extract from *Gynura procumbens* (Lour) Merr. leaves. Vietnam J. Agri. Sci. 14(8):1248-1260.
- Phan, T. A. D., Nguyen, X. H., Nguyen, T. N., Tran, L. Q., & Nguyen, T. T. M. (2012). Study on DPPH free radical scavenging and lipid peroxidation inhibitory activities of Vietnamese medicinal plants. Natural Product Sciences. 18(1): 1-7.
- Pierson J. T., Dietzgen R. G., Shaw P. N., Roberts-Thomson S. J., Monteith G. R., and Gidley M. J. (2012). Major Australian tropical fruits biodiversity: Bioactive compounds and their bioactivities. Molecular Nutrition and Food Research. 56: 357-387.
- Pinelo M., Tress A. G., Pedersen M., Arnous A. & Meyer A. S. (2007). Effect of cellulases, solvent type and particle size distribution on the extraction of chlorogenic acid and other phenols from spent coffee grounds. American Journal of Food Technology. 2(7): 641-651.
- Qaisar M. N., Chaudhary B. A., Sajid M. U. & Hussain N. (2014). Evaluation of  $\alpha$ -glucosidase inhibitory activity of dichloromethane and methanol extracts of *Croton bonplandianum* Baill. Tropical Journal of Pharmaceutical Research. 13(11): 1833-1836.
- Rymbai H., Verma V. K., Talang H., Assumi S. R., Devi M. B., Vanlalruati, Sangma R.H.CH., Biam K. P., Chanu L. J., Makdoh B., Singh A. R., Mawleñ J., Hazarika S. & Mishra V. K. (2023). Biochemical and antioxidant activity of wild edible fruits of the eastern Himalaya, India. Frontiers in Nutrition. 10: 1039965. DOI: 10.3389/fnut.2023.1039965.
- Rymbai H., Srivastav M., Sharma R. R., Patel C. R. & Singh A. K. (2013). Bio-active compounds in mango (*Mangifera indica* L.) and their roles in human health and plant defence - a review. Journal of Horticultural Science and Biotechnology. 88(4): 369-379. DOI: org/10.1080/14620316.2013.11512978.
- Rymbai H., Ercisli S., Durul S. M., Kilicgun H., Bayyigit I. & Colak A.M. (2025). Fruit biochemical, bioactive and antioxidant characteristics of *Prunus nepalensis*. Applied Fruit Science. 67: 127. DOI: 10.1007/s10341-025-01371-1.
- Shaw H. A. & Forman L. (1967). The genus *Spondias* L. (anacardiaceae) in tropical Asia. Kew Bulletin. 1-19.
- Silva C. R., Simoni J. A., Collins C. H. & Volpe P. L. (1999). Ascorbic acid as a standard for iodometric titrations. An analytical experiment for general chemistry. Journal of Chemical Education. 76(10): 1421-1422.
- Silva E. M., Rogez H. & Larondelle Y. (2007). Optimization of extraction of phenolics from *Inga edulis* leaves using response surface methodology. Separation and Purification Technology. 55: 381-387.
- Singleton V. L. & Rossi J. A. (1965). Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. American journal of Enology and Viticulture. 16(3): 144-158.
- Tabart J., Kevers C., Pincemail J., Defraigne J.-O. & Dommes J. (2009). Comparative antioxidant capacities of phenolic compounds measured by various tests. Food chemistry. 113(4): 1226-1233.
- Tandon S. & Rastogi R. (1976). Studies on the chemical constituents of *Spondias pinnata*. Planta Medica. 29(02): 190-192.
- Yusnawan E., Inayati A. & Baliadi Y. (2021). Total phenolic content and antioxidant activity in eight cowpea (*Vigna unguiculata*) genotypes. IOP Conf. Series: Earth and Environmental Science. 924(2021): 012047.
- Wu X., Beecher G. R., Holden J. M., Haytowitz D. B., Gebhardt S. E., & Prior R. L. (2004). Lipophilic and hydrophilic antioxidant capacities of common foods in the United States. Journal of Agricultural and Food Chemistry. 52: 4026-4037.