



Steaming and vacuum drying preserve active components, sensory and antioxidant properties of Flos Sophorae

Xiulan GUO^{1#}, Feng ZHANG^{1#} , Yu LIU¹, Mingzhu XIE¹, Renyong TANG^{1*}

Abstract

Flos Sophorae (FS) is medicinal and edible, while fresh FS is highly perishable. Thus, this study investigated the effects of different drying methods (hot air drying, HAD; vacuum drying, VD; freeze drying, FD) after steaming on the bioactive components and sensory quality of FS. The results indicated steaming for 1 min maximally retained the contents of total flavonoids, rutin, and quercetin in thermal drying (HAD and VD), meanwhile, the freeze-dried FS without steaming also had higher total flavonoids and rutin contents. Moreover, FD samples had the highest protein content and organoleptic properties, with closer color parameters to fresh FS, followed by the VD group. Considering comprehensively all data with processing cost, the sample of VD after steaming for 1 min was chosen to measure the antioxidant activity, which showed that FS extract was superior to Vc in ABTS• and •OH scavenging rates, and its DPPH• scavenging rate was stronger than Vc in the low concentration. These results demonstrated that vacuum drying after steaming for 1 min was suitable postharvest processing for fresh FS, which was beneficial to maintaining higher flavonoids, rutin, and sensory characteristics of FS, and its flavonoid extract had better antioxidant activity than Vc.

Keywords: Flos Sophorae; drying; steaming; flavonoids; sensory properties.

Practical Application: Vacuum drying after steaming for 1 min was an appropriate treatment for fresh Flos Sophorae.

1 Introduction

Flos Sophorae (FS) is a medicinal and edible substance as well as an ornamental plant, which is widely grown in China, Japan, and Korea (Fan et al., 2020). It contains large amounts of flavonoids such as rutin, quercetin, and kaempferol (Pi et al., 2018), which have anti-inflammatory, antibacterial, anti-cancer, hemostatic, and antihypertensive properties (Gong et al., 2021). Furthermore, FS was used as a potential natural antioxidant in Chinese sausages and marketed as a dietary supplement in the United States due to its anti-oxidative properties (Fan et al., 2020; Tang et al., 2019; Madden et al., 2022).

The herbal medicine is usually fixed by scalding and bleaching, microwaving, steaming, and so on, to destroy the enzymes at high temperatures and maintain the product quality (Duan et al., 2011). The fixed *Lonicera japonica* Thunb. had a higher content of rutin and other bioactive components in comparison with unfixed samples, and steamed flowers contained more bioactive composition than microwaved ones (Shi et al., 2021). Several studies also reported that steaming could retain more flavonoid components in Flos Sophorae, Medlar leaves tea, and *Lithocarpus polystachyus* (Liu et al., 2019; Zhang et al., 2009; Teng et al., 2021).

Drying is an essential means of preserving edible flowers because it can prevent microbial growth and inhibit enzymatic degradation (Zhao et al., 2019). Hot air drying is commonly used for food dehydration due to its simplicity (Ozay-Arancioglu et al.,

2022), and it retained more flavonoids in Flos Sophorae, such as rutin, quercetin, and kaempferol, than sun drying (Liu et al., 2019). Hot air drying at 55-60 °C was beneficial to the retention of the active constituents of flowers compared to other temperatures (Lu et al., 2020; Wang et al., 2021), and air-dried pea seeds at 55 °C also did not have off-flavors (Espinosa et al., 2022). In addition, hot-air-dried Sophora flowers had a higher rutin content, while freeze-dried flowers had closer sensory properties to fresh flowers, and a higher flavonoid composition compared with hot-air-dried and shade-dried ones (Wang et al., 2020). Juhari et al. (2021) also found that freeze-dried Roselle calyx resembled the cell structure of fresh ones, and vacuum drying had no impact on the color of the dry calyx. Therefore, freeze-drying could better preserve the quality of plants during processing, but the drying cost is higher (Ozay-Arancioglu et al., 2022), and vacuum drying could deserve to be studied for drying fresh Sophora flowers.

Fresh Sophora flowers are extraordinarily susceptible to spoilage and are usually fried to form dark yellow or black medicinal products (Gong et al., 2021), with poor visual perception. Therefore, a suitable method needs to be explored to minimize the changes in sensory properties and quality of FS as food and simultaneously extend its storage time. This study investigated the effects of steaming in combination with different drying methods on the flavonoid components and

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¹ School of Food and Biological Engineering, Chengdu University, Chengdu, Sichuan, China

*Corresponding author: tangrenyong@cdu.edu.cn

[#]These authors contributed equally to this work

quality characteristics of Flos Sophorae, and chose the best treatment to evaluate its antioxidant capacity.

2 Materials and methods

2.1 Flos Sophorae treatment

Fresh Sophora flowers were purchased from Henan Province, China, and randomly divided into three parts, which were steamed for 0 min, 1 min, and 1.5 min, respectively. After steaming, each part was dried with hot air drying (HAD), vacuum drying (VD), and freeze drying (FD).

HAD was performed in a constant temperature drying oven ZFD-7600 at 55 °C (Zhengzhou Nanbei Instrument Equipment Co., Ltd., China), and VD was subjected to a vacuum dryer DZF-6020 at 50 °C and 0.08 Mpa (Shanghai Langgan Experimental Equipment Co., Ltd., China). For FD, the samples were pre-frozen at -20 °C for 24 h and then placed in a freeze dryer FD-1A-80 at -60 °C and 50 Pa (Beijing Boyikang Experimental Instrument Co., Ltd., China). The drying experiments were completed when the final moisture content was less than 11%. After drying, a part was chosen for sensory evaluation, and the rest was crushed and passed through a 60-mesh sieve and stored at -20 °C until use.

2.2 Physicochemical analysis

The moisture content in both fresh and dried materials was determined by the oven method at 105 °C, and protein content was detected according to the Kjeldahl method (Abifarín et al., 2021).

2.3 Determination of total flavonoids, rutin and quercetin

The total flavonoids content of FS was measured according to Jakovljević et al. (2015) with some modifications. Dried FS powder (0.2 g) was treated with a 20-fold volume of an ethanol solution (70%). The mixture was ultrasonically shaken for 30 min at 50 °C, after which the filtrate was collected, and the process was repeated twice. Both portions of ethanolic extracts were combined. The extract (1 mL) was aspirated into a volumetric flask, and 2 mL of 0.1 M aluminum trichloride and 3 mL of 1 M potassium acetate solution were added and diluted with 30% ethanol to a volume of 10 mL. The mixture was incubated for 30 min at ambient temperature and centrifuged at 8,000 rpm for 5 min (Changsha Xiangyi Centrifuge Instrument Co., Ltd., China). The absorbance of the supernatant was measured by spectrophotometer UV755B at 420 nm (Shanghai Youke Instrument Co., Ltd., China). Rutin was selected as the standard, and the results were calculated from the calibration curve and expressed as mg/g DM.

The determination of rutin and quercetin was based on the method of Tang et al. (2021) with slight modification. FS powder (0.02 g) was mixed with 60% ethanol (10 mL), and the suspension was ultrasonicated for 90 min and centrifuged at 8,000 rpm for 5 min. The supernatant was filtered through a 0.45 µm filter membrane and injected into LC-20A high-performance liquid chromatography (Shimadzu Co., Ltd., Japan) for analysis. Rutin ($\geq 98\%$) and quercetin ($\geq 97\%$) were purchased from Ruifensi Biotechnology Co., Ltd. (Chengdu, China).

2.4 Color measurement

The color of fresh and dried FS was examined according to the color system of the International Commission on illumination by using a chroma meter CR-400 (Konica-Minolta, Osaka, Japan). The color parameters were expressed in terms of lightness, redness, and yellowness values (Barani et al., 2020).

2.5 Sensory evaluation

The sensory evaluation of FS was conducted by 15 trained panelists, and they were acquainted with the sensory characteristics of fresh flowers in advance. The dried flower samples tested were prepared in white porcelain trays and placed under standard lighting while the panelists rated the color, shape, and aroma of the Sophora flowers using a 9-point hedonic scale (1 = dislike extremely, 9 = like extremely).

2.6 Antioxidant capacity determination

FS powder (1 g) was extracted twice by ultrasonication with a 20 mL 70% ethanol solution and filtered. Then the two portions of extracts were mixed and diluted to 1.81, 20.05, 48.52, 73.39, and 101.47 µg/mL, with 70% ethanol solution. Vc solutions with 10, 20, 50, and 80 µg/mL were prepared as controls.

The antioxidant capacity of FS flavonoid extracts included the ABTS, DPPH, and hydroxyl radical scavenging ability, as well as the reducing power. The ABTS assay was performed as described by Dudonné et al. (2009) with some modifications. Potassium persulfate solution (2.6 mM) was mixed with 7.4 mM ABTS solution (1:1, v/v) to react in the dark for 12-16 h to obtain an ABTS radical working solution. When the absorbance value of the working solution was 0.70 ± 0.02 at 734 nm, after adding 0.8 mL of sample liquid to 3.2 mL of ABTS working solution for 30 min in the dark, the absorbance was measured at 734 nm. The hydroxyl radical scavenging rate was determined by the method of Chen et al. (2019a), and the DPPH radical scavenging rate and the reducing power were measured according to Liu et al. (2008).

2.7 Statistical analysis

The experiments in our work were carried out in triplicates. The research results were expressed as mean value \pm standard deviation. One-way ANOVA was performed by SPSS 25 (Chicago, IL, USA), and then Duncan's test was analyzed for comparing groups at the 0.05 level of significance.

3 Results and discussion

3.1 Moisture content

The average initial moisture content of fresh Sophora flowers was $89.72 \pm 0.13\%$ in this study. The moisture content of FS was gradually reduced with the extension of drying time (Figure 1) and lower than 11% (Chinese Pharmacopoeia Commission, 2020) when dried with hot air at 55 °C for 4 h (Figure 1A), vacuum at 50 °C for 6 h (Figure 1B), and freeze for 12 h (Figure 1C). However, the steaming time had little effect on the drying rate.

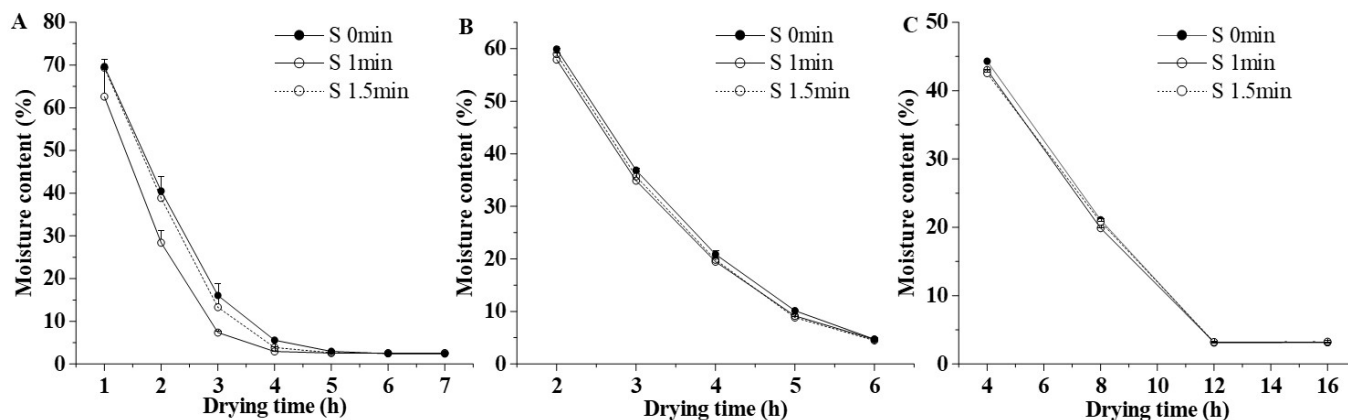


Figure 1. Effects of different treatments on the moisture content of Flos Sophorae: (A) hot air drying; (B) vacuum drying; (C) freeze drying. S 0min, steaming for 0 min; S 1min, steaming for 1 min; S 1.5min, steaming for 1.5 min.

Therefore, the FS treated with HAD for 4 h, VD for 6 h, and FD for 12 h was selected for subsequent tests.

3.2 Protein content

Flos Sophorae protein is abundant in essential amino acids, and the proportion of most essential amino acids conforms to the FAO/WHO model (Gong et al., 2021), which indicates that FS is a good source of plant protein. Freeze drying and vacuum drying protected FS proteins, which had higher processing performance and antioxidant properties than those by hot air drying (Ma et al., 2014). Similarly, this study found that the FD group had the highest protein content at the same steaming time, followed by the VD group. Meanwhile, the protein was remarkably lower after steaming for 1.5 min compared with other steaming times (Figure 2). This phenomenon may be due to the heating for a long time, which resulted in protein oxidation and degradation (Duque-Estrada et al., 2019; Jiang et al., 2022). In addition, the protein content ranged from 24.23 to 28.18 g/100 g in this research, and these were higher than other freeze-dried edible flowers, such as *Magnolia × soulangeana*, *Sambucus nigra* L., and *Robinia pseudoacacia* (Jakubczyk et al., 2022).

3.3 The content of total flavonoids, rutin and quercetin

Flos Sophorae is enriched with flavonoids, which have antioxidant and hypoglycemic properties (Liu et al., 2019; Wang et al., 2017b). Rutin is one of the main flavonoids and commonly used as a marker to determine the medicinal quality of Sophora flower materials (Jin et al., 2015). In this study, the proper steaming (1 min) maximally retained total flavonoids, rutin, and quercetin contents of FS by thermal drying in HAD and VD (Figure 3), while steaming fixation reduced the total flavonoids and rutin content in the FD group (Figure 3A-3B). This result was supported by previous studies, Liu et al. (2019) reported that fixation process was beneficial for preserving rutin in hot-air-dried *Sophora japonica*, and steaming fixation was good for protecting the luteolin content of hot-air-dried *Callicarpa nudiflora* (Song et al., 2022). However, freeze-dried Sophora flowers had a higher flavonoid composition compared with hot-air-dried and shade-dried ones (Wang et al., 2020).

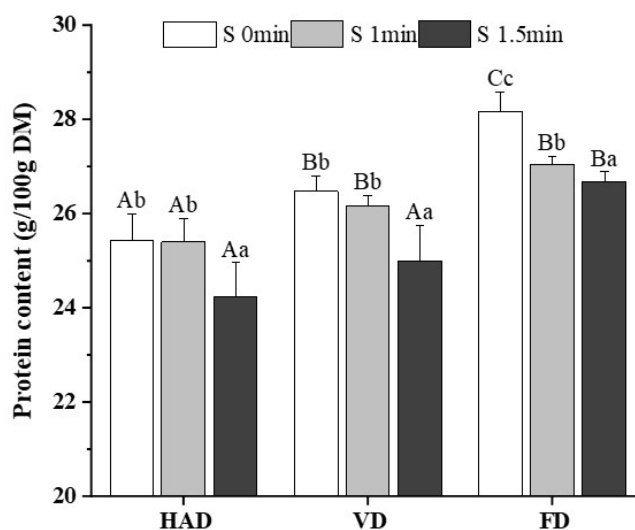


Figure 2. Effects of different treatments on the protein content of Flos Sophorae. HAD: hot air drying; VD: vacuum drying; FD: freeze drying; S 0min, steaming for 0 min; S 1min, steaming for 1 min; S 1.5min, steaming for 1.5 min. Different lowercase letters indicated significant differences between different steaming times ($P < 0.05$), and different capital letters indicated significant differences between different drying methods ($P < 0.05$).

This discrepancy could be due to the different temperatures during drying: proper steaming could deactivate the flavonoid-degrading enzymes (Duan et al., 2011) and thus prevent flavonoids by inhibiting the subsequent physiochemical response due to high temperature, while steaming could be unnecessary when less physiochemical response happens during freeze-drying.

After steaming for 1 min, the VD group had higher total flavonoids and rutin contents than the FD and HAD groups (Figure 3A-3B, $P < 0.05$). Similarly, Wei et al. (2019) reported that vacuum-dried *Eucommia ulmoides* Oliver male flowers had a higher content of total flavonoids than hot-air-dried and freeze-dried ones. There was the highest rutin content in lemon pomace after vacuum drying at 70 °C (Papoutsis et al., 2017).

Moreover, the FD group had higher total flavonoids and rutin content than the VD and HAD groups when without steaming (Figure 3A-3B, $P < 0.05$). This suggested that freeze drying itself could protect the active components of products due to its low processing temperature and lack of oxygen (Duan et al., 2010). Therefore, vacuum drying after steaming for 1 min or freeze drying has proven to be effective in preserving flavonoids and rutin in FS compared to conventional drying methods.

3.4 Color analysis

Among organoleptic properties, color is an important trait in the decision-making process for purchasing products (Susilo et al., 2022). The fresh FS was white with a slight yellow (lightness = 60.82 ± 0.04 , redness = -9.29 ± 0.08 , yellowness = 20.98 ± 0.08). The freeze-dried FS showed similar color with a slight increase in lightness compared to fresh flowers, and it had higher lightness and lower redness values than HAD and VD groups (Figure 4A-4B). Similarly, the brightness of freeze-dried *Begonia cucullata* also had an increasing trend compared with

fresh ones (Marchioni et al., 2022). In addition, HAD treatments induced an increase in yellowness (Figure 4C, $P < 0.05$), and this could be related to Maillard reactions, deterioration of some pigments, and non-enzymatic browning (Binici et al., 2021; Vega-Gálvez et al., 2009), which resulted in noticeable color changes of FS. Among the drying methods, the freeze-dried FS had closer color parameters to fresh flowers, followed by the vacuum-dried group.

Figure 4A indicated that steaming for 1-1.5 min maintained the higher lightness of Sophora flowers when thermal drying in HAD and VD, which suggested that steaming might protect the color of plants in some degree. Chen et al. (2019b) supported this hypothesis and observed that steaming fixation retained a better yellow-green color of tea.

3.5 Sensory evaluation

The sensory characteristics of a product affected its acceptability and preference by consumers (Cais-Sokolińska et al., 2021;

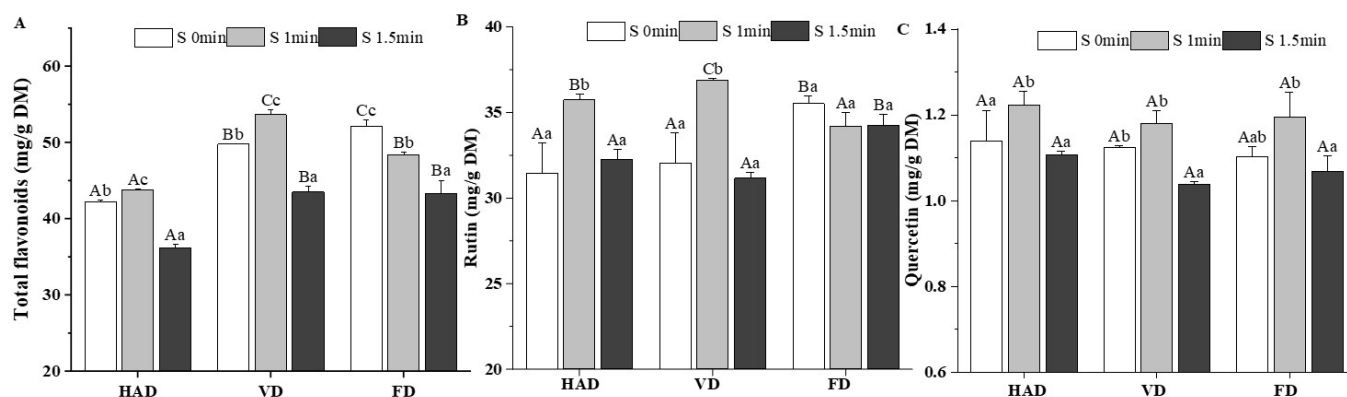


Figure 3. Effect of different treatments on the content of (A) total flavonoids; (B) rutin; (C) quercetin in Flos Sophorae. HAD: hot air drying; VD: vacuum drying; FD: freeze drying; S 0min, steaming for 0 min; S 1min, steaming for 1 min; S 1.5min, steaming for 1.5 min. Different lowercase letters indicated significant differences between different steaming times ($P < 0.05$), and different capital letters indicated significant differences between different drying methods ($P < 0.05$).

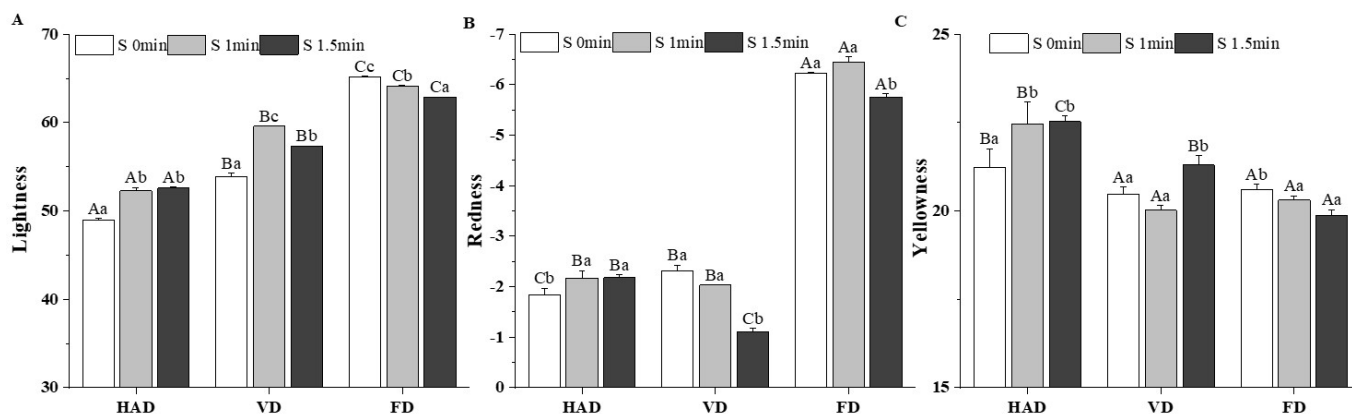


Figure 4. Effects of different treatment methods on the color parameters of Flos Sophorae: (A) Lightness; (B) Redness; (C) Yellowness. HAD: hot air drying; VD: vacuum drying; FD: freeze drying; S 0min, steaming for 0 min; S 1min, steaming for 1 min; S 1.5min, steaming for 1.5 min. Different lowercase letters indicated significant differences between different steaming times ($P < 0.05$), and different capital letters indicated significant differences between different drying methods ($P < 0.05$).

Los et al., 2021). Sophora flowers in the FD group were white and yellow, with complete appearance and light fragrance, and had the best color, shape, and aroma scores, followed by the VD samples (Table 1, $P < 0.05$). The sensory results were in agreement with the color results (Figure 4), and consistent with the results of Wang et al. (2020), who reported that freeze-dried Sophora flowers showed a minimal change in morphology, color, and smell. Additionally, proper steaming (1 min) elevated the color scores of Sophora flowers when thermal drying in HAD and VD compared to steaming for 0 min (Table 1, $P < 0.05$), and this was consistent with color results (Figure 4).

Table 1. Effects of different treatment methods on the senses of Flos Sophorae.

	Drying method	Steaming 0 min	Steaming 1 min	Steaming 1.5 min
Color	HAD	2.90 ± 0.25 ^{Aa}	4.20 ± 0.26 ^{Ab}	3.00 ± 0.22 ^{Aa}
	VD	4.30 ± 0.42 ^{Ba}	5.80 ± 0.31 ^{Bb}	4.80 ± 0.54 ^{Bab}
	FD	8.50 ± 0.32 ^{Ca}	8.40 ± 0.32 ^{Ca}	8.20 ± 0.40 ^{Ca}
Shape	HAD	4.20 ± 0.23 ^{Aa}	4.30 ± 0.36 ^{Aa}	3.80 ± 0.28 ^{Aa}
	VD	5.40 ± 0.32 ^{Ba}	4.90 ± 0.28 ^{Aa}	4.80 ± 0.41 ^{Ba}
	FD	8.50 ± 0.24 ^{Cb}	8.10 ± 0.33 ^{Bb}	6.80 ± 0.26 ^{Ca}
Aroma	HAD	4.00 ± 0.27 ^{Aa}	4.50 ± 0.33 ^{Aa}	4.20 ± 0.29 ^{Aa}
	VD	5.20 ± 0.49 ^{Ba}	6.00 ± 0.47 ^{Ba}	5.40 ± 0.36 ^{Ba}
	FD	8.40 ± 0.39 ^{Cb}	7.90 ± 0.48 ^{Cab}	7.30 ± 0.36 ^{Ca}

HAD: hot air drying; VD: vacuum drying; FD: freeze drying. Different lowercase letters indicated significant differences between different steaming times ($P < 0.05$), and different capital letters indicated significant differences between different drying methods ($P < 0.05$).

3.6 Antioxidant activity

The above experimental data showed that freeze drying could effectively retain protein components and sensory quality of Flos Sophorae, followed by the vacuum drying group, but vacuum drying after steaming for 1 min or freeze drying could maximally retain total flavonoids and rutin. Based on these results and processing cost, vacuum drying after steaming for 1 min was recommended to be an effective way to retain the quality of FS, and this group was chosen to measure the antioxidant activity.

In Figure 5A-5C, FS extract had higher scavenging rates of ABTS and hydroxyl radical than Vc at the same concentrations (1.81-73.39 $\mu\text{g/mL}$), but these results were inconsistent with the reports of Fan et al. (2020) and Wang et al. (2017a), who found Vc had higher ABTS and hydroxyl radical scavenging rates than FS extract in the concentrations of 0.2-0.8 mg/mL and 0.10-0.15 mg/mL, respectively. This difference may be caused by the varied sources and preparation methods of Flos Sophorae and test concentrations. Meanwhile, it had higher scavenging rates of DPPH radical than Vc in the low concentration range (Figure 5B), while Vc had higher total reducing power than FS in the high concentration (Figure 5D), which was similar to the result of Wang et al. (2017a). Overall, the antioxidant capacity of FS extract was positively correlated with the content of flavonoids, a similar correlation was observed by Fan et al. (2020). In addition, the IC_{50} value of FS extract for DPPH radical scavenging (32.880 $\mu\text{g/mL}$) was less than that of Vc solution (38.203 $\mu\text{g/mL}$) (Figure 5B). Therefore, the flavonoid extract of FS had better radical scavenging rates than Vc, which may

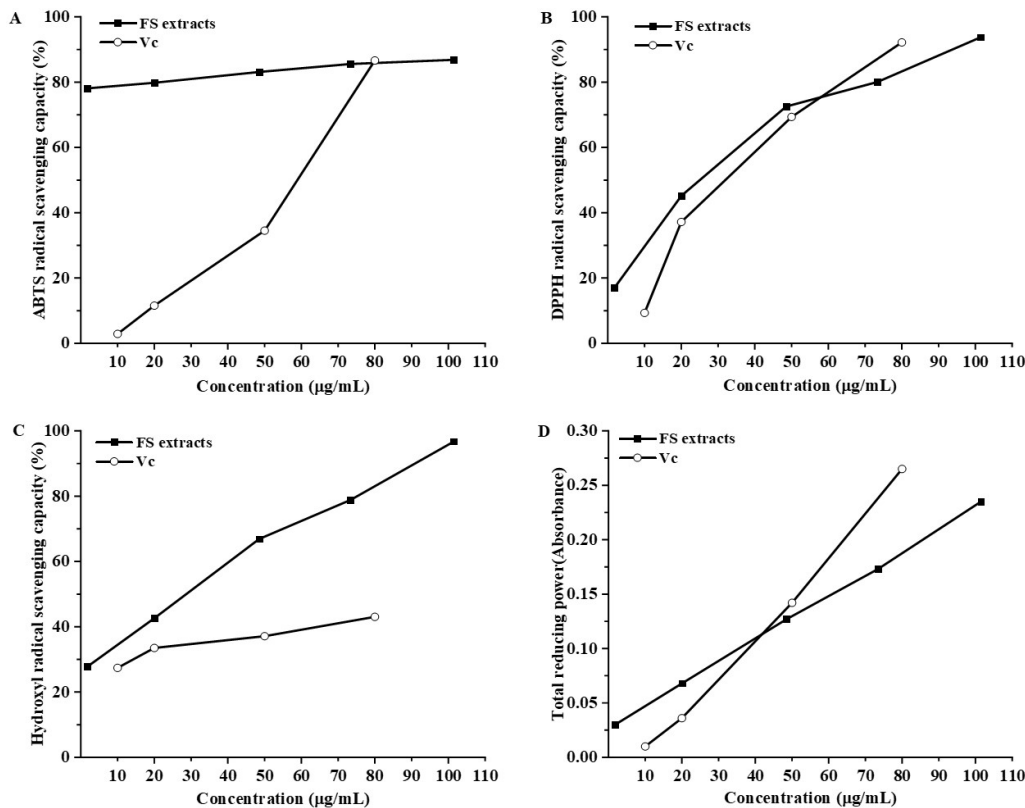


Figure 5. Effect of different concentrations of Flos Sophorae extracts in free radical scavenging rate: (A) ABTS•; (B) DPPH•; (C) •OH; (D) Total reducing power.

be associated with the higher content of rutin and quercetin in FS extract.

4 Conclusion

Based on the results obtained, vacuum drying after steaming for 1 min was an appropriate method for postharvest processing of fresh Flos Sophorae, with the highest contents of total flavonoids and rutin, better visual quality and protein content, and relatively low cost of processing. Furthermore, its flavonoid extract had better antioxidant capacity than Vc. Therefore, this research provided a good method for postharvest processing of Flos Sophorae as a food material, which could effectively protect its bioactive ingredients and sensory characteristics, and extend its shelf-life.

Acknowledgements

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