Effects of drying methods on the structure and emulsifying capacity of egg yolk lecithin

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Abstract: The purpose of this study was to investigate the effects of different drying methods, including ultrasonic vacuum drying, vacuum drying, vacuum freeze-drying, hot-air drying and spray drying, on the structure and emulsifying capacity of egg yolk lecithin based on Raman spectra. The results showed that ultrasonic vacuum drying and spray drying can induce the vibration of C–N bonds in the polar O–C–C–N^+ head skeleton of egg yolk lecithin. The shift of the peak attributed to the C–N bond from 717 cm^{-1} to 774 and 772 cm^{-1} indicated that the vibration of some C–N bonds in the O–C–C–N^+ skeleton had transformed from gauche to trans. Ultrasonic vacuum drying exerted the most intense effect on the C–C skeleton of egg yolk lecithin, with the greatest vibration peaks at 1062 cm^{-1}, 1128 cm^{-1}, and 1097 cm^{-1} in the Raman spectra of egg yolks. Specifically, it relieved gauche vibration and strengthened trans vibration in the C–C skeleton. Hence, the I_{gauche}/I_{trans} ratio of the egg yolk lecithin processed through ultrasonic vacuum drying decreased. Ultrasonic vacuum drying and spray drying decreased the I_{2850}/I_{2878} ratio of the vibration peak of C–H bonds in the lipid chains of egg yolk lecithin. The weakening of the symmetric stretching vibration of the C–H bond and the strengthening of antisymmetric stretching vibration indicated that orderliness among the molecular chains of lipid bilayer membranes had increased, whereas liquidity had decreased. The emulsifying capacities were highly significantly different among various egg yolk lecithin samples, in which the highest emulsifying capacity (49.58 m^2/g) was shown for the egg yolk lecithin prepared through vacuum freeze-drying, and ultrasonic vacuum drying produced the lowest emulsifying capacity (14.77 m^2/g). This study demonstrated that ultrasonic vacuum drying and spray drying drastically affected the structure of egg yolk lecithin. The appropriate drying method can be selected based on sample volume and production situation.

Keywords: drying, structure, emulsification capacity, egg yolk lecithin, Raman spectrum

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1 Introduction

The yolk accounts for 28%-29% of the total mass of an egg. Solids contribute 50% of the total mass of egg yolks, and 32% of the total mass of egg yolk solids is attributed to lipids. Phospholipids, such as lecithin, account for 30% of the total egg yolk lipids[1]. Egg yolk lecithin is a composite phospholipid, and its major functional constituent is phosphatidylcholine (PC), which accounts for 70%-80% of the total egg yolk lecithin[2]. Egg yolk lecithin possesses high nutritional value and has been called "the gold for brain strengthening and a great nutrient for health" by British scientists[3]. It releases choline after digestion by the human body. Choline reaches the brain through blood circulation. It can prevent intellectual deterioration and enhance memory. Moreover, it facilitates hepatocyte regeneration, increases the plasma protein content, promotes metabolism, and boosts immunity. In addition, it is a natural surfactant and has numerous critical physiological functions. Egg yolk lecithin is applied as a pharmaceutical accessory. For example, it is used as an emulsifier in fat emulsion injections. It has also been utilized to produce liposomes for targeted drug delivery[4]. Egg yolk lecithin is mainly extracted from egg yolk powder. Hence, drying methods for the production of egg yolk powder must ensure that the structure of egg yolk lecithin is maintained in the final product. The emulsifying capacity is one of the key functional properties for egg yolk lecithin and is closely related to its structure which is influenced by different processing methods[4]. Moreover, the suitable detection method should be applied to analyze the quality of dried egg yolk powder.

Infrared spectroscopy and Raman spectroscopy are common methods used to test the structures of materials. Although these methods function through different mechanisms, both acquire spectra on the basis of molecular vibration and can complement each other. Infrared spectroscopy is mainly employed to identify groups of molecules, whereas Raman spectroscopy is mainly applied to observe the skeletons of molecules[5,6]. Infrared spectrometry requires grinding solid samples with KBr and complicated sample preprocessing procedures. By contrast, Raman spectroscopy is a nondestructive detection method that does not require sample preprocessing. Herrero announced that the protein structure in muscle food could be precisely monitored by Raman spectroscopy[7]. Czaja et al. verified that gluten structure could be well characterized by Raman spectroscopy[8]. Therefore, Raman spectroscopy could be widely used as a convenient, rapid, and accurate method for the structural characterization of samples.

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Existing studies on egg yolk lecithin mainly focused on the extraction technologies and the functions of egg yolk lecithin. Liu et al.[9] found that a high voltage pulse electric field could improve PC extraction efficiency without drastically affecting the detection of the molecular structure of the PC through mid-infrared spectroscopy. Palacios et al.[10] demonstrated that the egg yolk lecithin extraction efficiencies of the non-deoiling and deoiling techniques with ethyl alcohol as a solvent were 23.9% and 13.5%, respectively. Asomaning et al.[11] optimized the enzymatic extraction of egg yolk lecithin by introducing transesterification. Akashi et al.[12] discovered that the addition of egg yolk lecithin to enteral formulations could protect emulsifiers from gastric acid damage, promote lipid adsorption, and ameliorate diarrhea. Nevertheless, the effects of drying methods on the structure of egg yolk lecithin and the effects of structural modifications on emulsifying capacity have not yet been reported.

In the lecithin industry, spray drying is the current commercial method for drying egg yolk lecithin, but egg yolk lecithin is easily oxidized or denatured during drying and the quality of the dried product is usually unsatisfactory. Thus, some alternative drying methods with low pressure and low temperature could be considered. In this study, egg yolk powder was processed from liquid egg yolk through five different drying methods including ultrasonic vacuum drying, vacuum drying, vacuum freeze-drying, hot-air drying and spray drying. Egg yolk lecithin was extracted from egg yolk powder samples prepared through different methods and the emulsifying capacities were quantified. The effects of drying methods on the polar O–C–C–N^+ head skeleton, C–C skeleton, and lipid bilayer membrane liquidity of egg yolk lecithin were investigated through laser Raman spectrometry. The relationship between the emulsifying capacity and the structure of egg yolk lecithin was discussed.

2 Materials and methods

2.1 Test materials

Fresh eggs were procured from a market in Luoyang, China. Standard egg yolk lecithin samples (purity>99.3%) were purchased from Shanghai Yuan Ye Biotechnique Co., Ltd. The analytical agent SDS was obtained from Luoyang AokeHuabo Instrument Distribution. Peanut oil (Lu Hua 5S pressing peanut oil) was acquired from a supermarket in Luoyang City.

2.2 Experimental methods

2.2.1 Processing of egg yolk powder

The processing of egg yolk powder is illustrated in Figure 1.

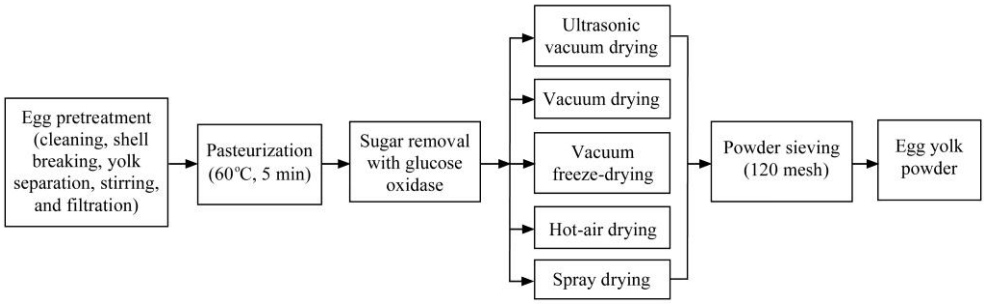
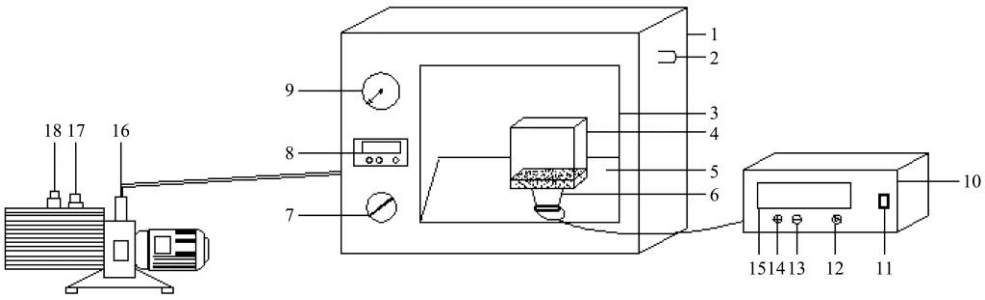


Figure 1 Processing of egg yolk powder

2.2.2 Drying methods

After the removal of sugar impurities, the liquid egg yolk samples were dried with five different methods, cooled, and ground. The powders were passed through a 120 mesh sieve to obtain the final egg yolk powder samples. The moisture content of dried egg yolk powder samples was less than 5%. The drying methods were performed under the following conditions:

Ultrasonic vacuum drying[13]: An ultrasonic system was installed into a vacuum dryer (DZF-6050, Yiheng Scientific Instrument Company, Shanghai, China) to achieve an ultrasonic vacuum drying device. The ultrasonic system was self-fabricated by Henan University of Science and Technology and mainly consisted of an ultrasound receiver, an ultrasonic transducer, and an ultrasonic generator. The schematic of the ultrasonic vacuum dryer is demonstrated in Figure 2, and detailed information and parameters of the ultrasonic system and the ultrasonic vacuum dryer could be seen in the literature[13]. Liquid egg yolk (500 g) was poured into the material tank of the ultrasonic system. The energy density of the ultrasonic wave, ultrasonic frequency, vacuum pressure, drying temperature, ultrasonication time, and drying time were set as 2 W/g, 28 kHz, 0.01 MPa, 50 ℃, 3 h, and 3 h, respectively.



1. Casebody 2. Air evacuation valve 3. Inner container 4. Ultrasonic receiver 5. Partition 6. Ultrasonic transducer 7. Vacuum valve 8. Temperature controller 9. Vacuum meter 10. Ultrasonic generator 11. Switch 12. Frequency sweep switch 13. Setting time 14. Setting power 15. Electronic display 16. Intake duct 17. Exhaust port 18. Gas ballast valve

Figure 2 Schematic of ultrasound enhanced vacuum dryer

Vacuum drying[14]: Liquid egg yolk was poured into a disk to form a layer with a thickness of 4 mm. The disk was then placed into a vacuum drying oven (DZF-6050, Yiheng Scientific Instrument Company, Shanghai, China) with the vacuum pressure, drying temperature, and drying time set as 0.01 MPa, 50 ℃, and 7 h, respectively.

Vacuum freeze-drying[15]: Liquid egg yolk was poured into a disk and then covered with a perforated piece of thin plastic film. The liquid was frozen for 1h under −30 ℃. The egg yolk was dried in a vacuum freeze-dryer (LGJ-10D, Sihuan Scientific Instrument Company, Beijing, China) after complete freezing. The thickness of the material layer, temperature, drying room pressure, and drying time were 4 mm, −60 ℃, 30 Pa, and 24 h, respectively.

Hot-air drying: Liquid egg yolk was poured into a disk to form a layer with a thickness of 4 mm and then dried for 24 h in a blast-drying oven (DHG-9420A, Yiheng Scientific Instrument Company, Shanghai, China) at the constant temperature of 60 ℃.

Spray drying[16]: Liquid egg yolk was poured into a constant-temperature water bath and preheated to 25 ℃ before drying in a spray-drier (SP-1500, Shunyi Experimental Apparatus Company, Shanghai, China). The spraying flow rate was 500 mL/h. Inlet temperature and outlet temperature were held at 120 ℃ and 90 ℃, respectively.

During the drying processes, the sample was taken out and weighed every 20 min, and the drying ended until the sequential two mass readings are approximately the same, with the corresponding moisture content of around 5% for the dried sample. All drying experiments were performed in triplicates.

2.2.3 Raman spectra of standard egg yolk lecithin samples and egg yolk powder

A confocal microscopic Raman spectrometer (XploRA ONE, HORIBA JOBIN YVON S.A.S, France) was applied to acquire the spectra of the egg yolk lecithin samples and egg yolk powder. The spectrometer was equipped with a 50×objective telephoto lens, 785 nm laser, and CCD detector. The spectrometer was operated with the laser power, spectral resolution, integral time, integration number, and spectral scanning range of 50%, 4 cm^{-1}, 20 s, 2 and 200-3500 cm^{-1}, respectively[17].

In each experiment, 200 points were selected randomly for scanning. The Raman spectra of standard egg yolk lecithin samples and egg yolk powder were recorded under the above conditions. Peaks attributable to egg yolk lecithin in the spectra of egg yolk powder dried through different methods and fresh liquid egg yolk were indexed to the characteristic peaks of standard egg yolk lecithin. The Raman spectra of fresh liquid egg yolk were used as the control group to analyze the effects of drying methods on the structure of egg yolk lecithin.

2.2.4 Extraction of egg yolk lecithin

Referring to previous studies[18], egg yolk lecithin was extracted by ethanol. Egg yolk powder of 30 g was weighed into a 250 mL beaker with the addition of 95% ethanol of 4 times volume, was stirred with a homogenizer (Angni Instrument Company, Shanghai, China) at a speed of 500 r/min for 15 min at 60 ℃ water bath, and was centrifuged at 3600 g for 8 min, and then the supernatant was poured out. The precipitate was extracted again using the same method described above. The supernatant was combined and placed in a refrigerator at 4 ℃ for 12 h to precipitate a neutral fat. After filtration, the filtrate was concentrated by a rotary evaporator (Yarong Biochemical Instrument Company, Shanghai, China) to obtain egg yolk lecithin.

2.2.5 Emulsifying capacity test of egg yolk lecithin

This test was performed according to the references of Marefati et al.[19] and Ishii[20]. In this test, 30 mL of distilled water was combined with 10 mL of peanut oil, into which 300 mg of egg yolk lecithin was added. The mixture was then homogenized for 1 min at 16000 r/min. Subsequently, 0.2 mL of the homogenate was collected from the bottom of the container by using a transfer pipette and mixed with 40 mL of 0.1% SDS. The optical density of the mixture was quantified at 500 nm by using a UV-visible spectrometer (Shimadzu, Japan) with a 0.1% SDS solution as the blank control and three parallel samples. Emulsifying capacity was calculated as follows:

$$\mathrm{EAI}/(\mathrm{m}^2/\mathrm{g}) = \frac{2\times 2.303\times A_0\times n}{\rho\times\varphi\times 10000} \quad (1)$$

where, EAI is the emulsifying capacity; n is the dilution ratio (200 in this experiment); ρ is the mass concentration of egg yolk lecithin (g/mL); φ is the volume fraction of oil (1/4 in this experiment), and A_0 is the optical density of homogenized samples at 500 nm.

2.2.6 Spectral preprocessing and data analysis

All Raman spectra of 200 points were recorded and presented as the mean value. Data analysis was carried out with Origin 8.5 software. The statistical analysis including analysis of variance and multiple comparisons was carried out using SAS 6.0 software.

3 Results and analysis

3.1 Raman spectra of standard egg yolk lecithin samples

The Raman spectra of standard egg yolk lecithin samples are shown in Figure 3.

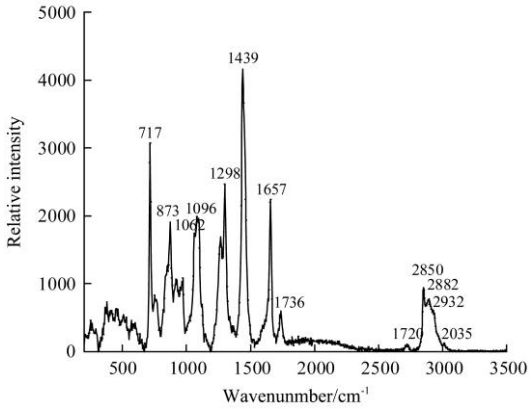


Figure 3 Raman spectra of standard egg yolk lecithin samples

Table 1 provides the affiliations of peaks in the Raman spectra of the standard egg yolk lecithin samples shown in Figure 3.

Table 1 Affiliations of peaks in the Raman spectra of standard egg yolk lecithin samples

Peaks /cm^{-1}	Reference peaks /cm^{-1}	Affiliations	References
717	717	ν_{sym}(C–N)$_{gauche}$	Sailer (1997) [21]
873	874	ν_{sym}(C–N)$_{gauche}$ and δ_d(CH_2)	Lhert (2000) [22]
1062	1062	ν(C–C)$_{trans}$	Wu(1995) [23]
1096	1096	ν(C–C)$_{gauche}$ and ν_{asym}(PO_2)	Wu (1995) [23]
1128	1128	ν(C–C)$_{trans}$	Wu (1995) [23]
1298	1296	ρ_t(CH_2)	Kint(1992) [24]
1439	1436	δ_d(CH_2)	Tantipolphan(2006) [25]
1657	1671	ν(C=C)	Tantipolphan (2006) [25]
1736	1735	ν(C=O)	Kint (1992) [24]
2720	2725	δ_S(CH_2) + ρ_W(CH_2)	Capelle(2000) [25]
2850	2845	ν_{sym}(CH_2)	Tantipolphan (2006) [15]
2882	2880	ν_{asym}(CH_2)	Capelle (2000) [25]
2932	2932	ν_{sym}(CH_3)	Sailer (1997) [21]
3035	3039	ν_{asym}N(CH_3)choline	Tantipolphan (2006) [15]

Notes: ν is stretching vibration; ν_{sym} is symmetric stretching vibration; ν_{asym} is asymmetric stretching vibration; δ_d is deformation vibration; ρ_t is torsional vibration; δ_S is shear vibration; ρ_W is rocking vibration.

Egg yolk lecithin is composed of glycerin, choline, phosphoric acid, and saturated and unsaturated fatty acids. The molecular structure of egg yolk lecithin can be divided into three regions on the basis of spectral features: the hydrophobic chain consisting of C–H stretching modes, CH_2 deformation, CH_2 twisting, and C–C stretching modes, the interfacial region containing C=O stretching, and the polar headgroup region comprising of a band of PO_2^- asymmetric stretching and C–N stretching [15,26].

As shown in Figure 3 and Table 1, C–N stretching vibration occurred at 717 cm^{-1}.

The vibration of the carbonyl group in the interfacial region gave a weak band at 1736 cm^{-1}.

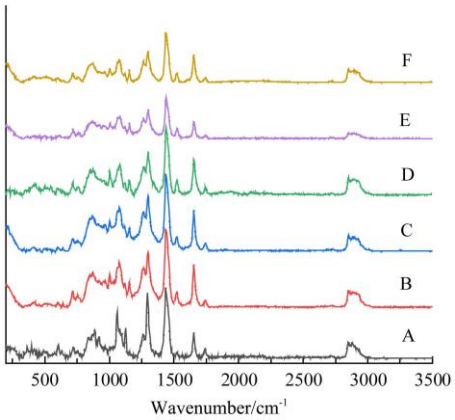
The stretching vibration region of the C–C skeleton was observed in the 1000-1200 cm^{-1} region. In this region, peaks at 1062 cm^{-1} and 1128 cm^{-1} were ascribed to the vibration peaks of the trans conformation of the bilayer C–C skeleton, whereas the peak at 1096 cm^{-1} was attributed to the stretching vibration peak of the gauche conformation of the C–C skeleton and PO_2^-[23].

The deformation peak of CH2 was located at 1439 cm^{-1}, and the peak at 1298 cm^{-1} belonged to the twisting mode of CH_2.

The stretching vibration mode of C–H was located in the 2800-3000 cm^{-1} region. The vibration peaks at 2850 and 2882 cm^{-1} were contributed by the symmetric and asymmetric stretching vibrations of methylenes, respectively. The peak at 2932 cm^{-1} was ascribed to the symmetrical stretching vibration of terminal methyls. The peak at 3035 cm^{-1} was attributed to the asymmetric stretching vibration of choline methyl[27].

3.2 Raman spectra of egg yolk powder and fresh liquid egg yolk

The Raman spectra of egg yolk powder and fresh liquid egg yolk samples are shown in Figure 4.



Note: (A) Ultrasonic vacuum drying; (B) Vacuum drying; (C) Vacuum freeze-drying; (D) Hot-air drying; (E) Spray drying; (F) Fresh liquid egg yolk.

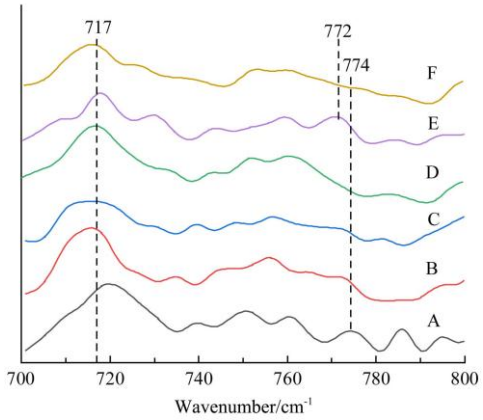
Figure 4 Raman spectra of egg yolk powder and fresh liquid egg yolk samples

Figure 4 illustrates that the spectra of egg yolk powder and fresh liquid egg yolk samples presented the characteristic peaks of standard egg yolk lecithin samples. In addition, the peak intensity and positions in the Raman spectra of egg yolk powder prepared through various drying methods were different from those in the Raman spectra of fresh liquid egg yolk samples.

3.3 Effects of drying methods on the polar O–C–C–N^+ head skeleton of egg yolk lecithin

The symmetric stretching vibration of the choline head base C–N is sensitive to the conformation of the O–C–C–N^+ skeleton. When O–C–C–N^+ is in its gauche conformation, the vibration peak of C–N appears at 717 cm^{-1}. When O–C–C–N^+ is in its trans conformation, the vibration peak of C–N moves to 770 cm^{-1}[28,29]. The orientation of the polar head changed from the parallel membrane plane to the vertical membrane plane when the conformation of O–C–C–N^+ transformed from gauche to trans. Therefore, the effects of the drying method on the polar headgroup region of egg yolk lecithin can be determined based on the vibration peak position of C–N.

As illustrated in Figure 5, ultrasonic vacuum drying and spray drying drastically affected the conformation of the polar headgroup region of egg yolk lecithin. A vibration peak was developed at 774 cm^{-1} in the spectra of the sample prepared through ultrasonic vacuum drying. An intense vibration peak was observed at 772 cm^{-1} in the spectra of the spray-dried sample. This result confirms that these drying methods modified the conformation of the O–C–C–N^+ skeleton in egg yolk lecithin from gauche to trans. By contrast, no vibration peaks were observed near 770 cm^{-1} in the spectra of the samples prepared through vacuum freeze-drying, vacuum drying, and hot-air drying. These results indicate that these methods negligibly affected the conformation of the O–C–C–N^+ skeleton.



Note: (A) Ultrasonic vacuum drying; (B) Vacuum drying; (C) Vacuum freeze-drying; (D) Hot-air drying; (E) Spray drying; (F) Fresh liquid egg yolk.

Figure 5 Raman spectra of the stretching vibration of C–N

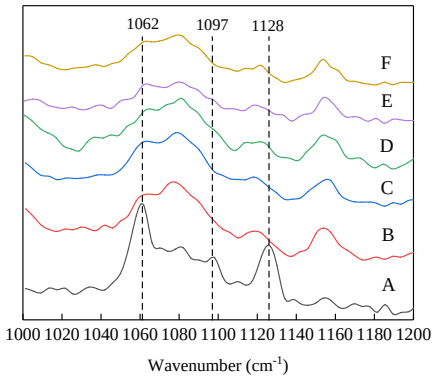
The massive energy exerted by ultrasonic propagation during ultrasonic vacuum drying reduced the orderliness of lecithin molecules and the liquidity of the bilayer membrane[30]. Molecules in the substance vibrated under the action of ultrasonic waves. Vibration frequency is equal to ultrasonic frequency. Molecular structures are intensively damaged by high frequency[31,32]. Therefore, ultrasonic vacuum drying influenced the structure of egg yolk lecithin.

Similar to ultrasonic vacuum drying, spray drying drastically influenced the structure of egg yolk lecithin. Under the high inlet temperature (120 ℃) of spray drying, materials acquired sufficiently high energy that stimulated and accelerated molecular vibration[33]. By contrast, the structure of molecules is hardly changed under the low temperatures of vacuum freeze-drying, vacuum drying, and hot-air drying due to their inadequate energy.

3.4 Effects of drying methods on the conformation of the C–C skeleton

The stretching vibration of the C–C skeleton was located in the1000-1200 cm^{-1} region (Figure 6). In this region, the vibration of C–C was highly sensitive to the conformation of chain molecules. The hydrocarbon chain can exhibit various conformations because of the rotation of C atoms in the chain.

System energy is minimized when the chain is in a trans configuration and the skeleton of the hydrocarbon chain is on a zigzag plane. A certain amount of energy is required to form various gauche conformations that then result in the different torsions of the hydrocarbon chain[34]. In the stretching vibration region of the C–C skeleton, the vibration peaks at 1062 cm^{-1} and 1128 cm^{-1} were contributed by the bilayer C–C skeleton of egg yolk lecithin, whereas the peak at 1097 cm^{-1} was ascribed to the vibration peak of the C–C skeleton in gauche conformation[26]. The orderliness of the C–C chain can be acquired by comparing the strengths of the trans and gauche spectra. High I_{gauche}/I_{trans} ratios indicate low lecithin molecule order and high liquidity. Otherwise, the orderliness of lecithin molecules had increased and liquidity had decreased[35]. The I_{gauche}/I_{trans} ratios of the samples are listed in Table 2.



Note: (A) Ultrasonic vacuum drying; (B) Vacuum drying; (C) Vacuum freeze-drying; (D) Hot-air drying; (E) Spray drying; (F) Fresh liquid egg yolk.

Figure 6 Raman spectra of stretching vibration of C–C

Table 2 I_{gauche}/I_{trans} ratios of egg yolk samples with different drying methods

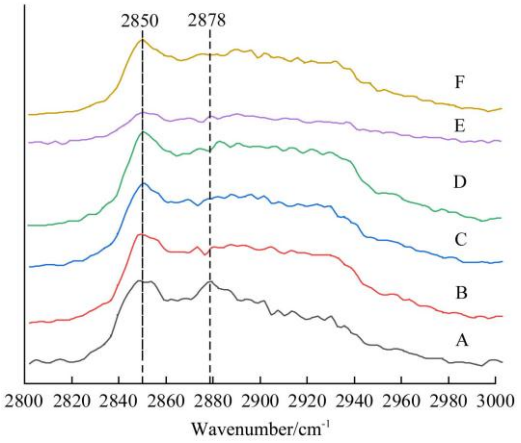
	Ultrasonic vacuum drying	Vacuum drying	Vacuum freeze-drying	Hot-air drying	Spray drying	Fresh liquid egg yolk
I_{1096}/I_{1062}	$(0.52\pm0.008)^b$	$(0.70\pm0.020)^a$	$(0.67\pm0.015)^a$	$(0.70\pm0.013)^a$	$(0.69\pm0.018)^a$	$(0.69\pm0.012)^a$
I_{1096}/I_{1128}	$(0.82\pm0.021)^b$	$(1.44\pm0.025)^a$	$(1.41\pm0.020)^a$	$(1.45\pm0.006)^a$	$(1.42\pm0.011)^a$	$(1.43\pm0.014)^a$

Note: Different letters represent significant differences (p<0.05).

As shown in Table 2, the I_{1096}/I_{1062} and I_{1096}/I_{1128} ratios of powdered egg yolk samples prepared through ultrasonic vacuum drying were lower than those of fresh liquid egg yolk and samples prepared through other drying methods. Moreover, as presented in Figure 6, the vibration peaks at 1062, 1128, and 1097 cm^{-1} in the Raman spectra of egg yolks prepared through ultrasonic vacuum drying were stronger than those in the spectra of fresh liquid egg yolk and samples dried through other methods, indicating that ultrasonic vacuum drying showed the most intense effect on the C–C skeleton. These results indicate that the conformation of the C–C skeleton was drastically affected by ultrasonic energy to change the order of the internal structure of a molecule while it was negligibly influenced by other drying methods.

3.5 Effects of drying methods on the bilayer membrane liquidity of egg yolk lecithin

Peaks within the 2800-3000 cm^{-1} region in the Raman spectra of egg yolk lecithin were attributed to the stretching vibration of some C–H bonds in the lipid chain (Figure 7). The symmetric stretching and antisymmetric stretching vibrations of the C–H bond manifested as two peaks at 2850 cm^{-1} and 2878 cm^{-1}, respectively. The vibration modes of these two peaks drastically influenced the lattice accumulation of the lipid bilayers and conformation between chains. The degree of order among the molecular chains of egg yolk lecithin could be inferred based on the ratio of the intensities of the two peaks at 2850 cm^{-1} and 2878 cm^{-1}. The I_{2850}/I_{2878} ratio is negatively correlated with the degree of order among the molecular chains of lecithin but is positively correlated with the liquidity of the lipid bilayer membrane[36,37]. The I_{2850}/I_{2878} ratios are shown in Table 3.



Note: (A) Ultrasonic vacuum drying; (B) Vacuum drying; (C) Vacuum freeze-drying; (D) Hot-air drying; (E) Spray drying; (F) Fresh liquid egg yolk.

Figure 7 Stretching vibration region of C–H

Table 3 I_{2850}/I_{2878} ratios of egg yolk samples with different drying methods

	Ultrasonic vacuum drying	Vacuum drying	Vacuum freeze-drying	Hot-air drying	Spray drying	Fresh liquid egg yolk
I_{2850}/I_{2878}	$(1.00\pm0.020)^b$	$(1.20\pm0.015)^a$	$(1.21\pm0.006)^a$	$(1.21\pm0.014)^a$	$(1.02\pm0.018)^b$	$(1.22\pm0.011)^a$

Note: Different letters represent significant differences (p<0.05).

As presented in Table 3, that I_{2850}/I_{2878} ratios of egg yolk samples processed through ultrasonic vacuum drying and spray drying were 1.00 and 1.02, respectively, and were lower than those of fresh liquid egg yolk and samples dried through other methods. The liquidity of the lipid bilayer membrane was considerably influenced by ultrasonic vacuum drying and spray drying but was slightly affected by other drying methods.

3.6 Effects of drying methods on the emulsifying capacity of egg yolk lecithin

Egg yolk lecithin was extracted with 95% ethanol from egg yolk powder samples prepared through different methods and the emulsifying capacities were quantified using the method described above. The results are shown in Figure 8, and different letters in the figure indicate that their differences are significant at the level of α=0.05.

The effects of different drying methods on the emulsifying capacity of egg yolk lecithin were analyzed by variance analysis and multiple comparisons, as shown in Tables 4 and 5.

It has been suggested in Table 4 and Table 5 that the effects of different drying methods on the emulsifying capacity of egg yolk

lecithin were highly significant.

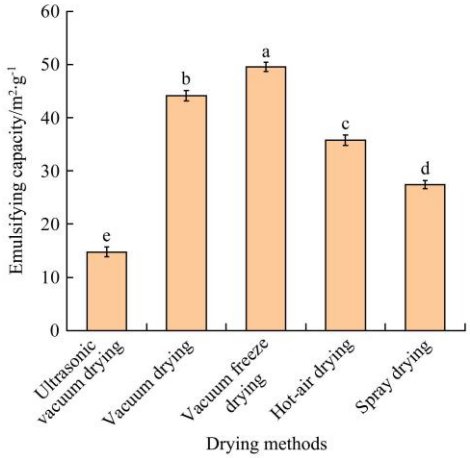


Figure 8 Effects of drying methods on the emulsifying capacity of egg yolk lecithin

Table 4 Variance analysis of the effects of different drying methods on the emulsifying capacity of egg yolk lecithin

Source	*SS*	*df*	*MS*	*F* value	Significance level
Drying methods	6855.57	4	1713.89	2079.19	α=0.01
Error	32.97	40	0.82		
Sum	6888.54	44			

Note: $F_{0.01}(4, 40) = 3.83$.

Table 5 Significance analysis of the effects of different drying methods on the emulsifying capacity of egg yolk lecithin

Drying methods	$\bar{x}_i$	Significance of difference	
		α=0.05	α=0.01
Vacuum freeze-drying	49.58	a	A
Vacuum drying	44.15	b	B
Hot-air drying	35.81	c	C
Spray drying	27.41	d	D
Ultrasonic vacuum drying	14.77	e	E

As shown in Figure 8, the highest emulsifying capacity of 49.58 m^2/g was exhibited by the egg yolk lecithin sample prepared through vacuum freeze-drying, followed by that of samples processed through vacuum drying, hot-air drying, spray drying, and ultrasonic vacuum drying. The lowest emulsifying capacity of 14.77 m^2/g was obtained by the egg yolk lecithin sample dried through ultrasonic vacuum drying. The structures of lecithin were altered by different drying methods and drying conditions, which led to the change of emulsifying capacity.

In this study, ultrasonic vacuum drying and spray drying altered the membrane orientation of the polar head region of egg yolk lecithin from parallel to vertical. This alteration exposed the hydrophobic groups of egg yolk lecithin and consequently weakened the hydrophilicity and emulsifying capacity of egg yolk lecithin[38,39]. Meanwhile, the combined effects of ultrasonic wave and high temperature decreased the liquidity of the lipid bilayer membrane[40,41]. This reduction was accompanied by increased lipophilicity and reduced emulsifying capacity[42].

Egg yolks begin to solidify at 65 ℃. The temperature of spray drying exceeded the solidification temperature of egg yolks and reached 120 ℃, under which emulsifying capacity drastically decreased[43,44]. In actual operation, egg yolk lecithin prepared through ultrasonic vacuum drying was partially insoluble in the oil-water mixture. The precipitation of particles and the rapid formation of different liquid layers after homogenization were indicative of extremely low emulsifying capacity[42,45]. The egg yolk lecithin prepared through vacuum freeze-drying exhibited the highest emulsifying capacity mainly because molecular vibration is insensitive to low temperature. The temperature of vacuum drying and hot-air drying was 50 ℃, which was lower than the temperature required for egg yolk solidification. Therefore, the egg yolk lecithin samples prepared through vacuum drying and hot-air drying presented moderate emulsifying capacity. Vacuum drying was quicker than hot-air drying and slightly degraded the structure of egg yolk lecithin. Therefore, the emulsifying capacity of samples processed through hot-air drying was slightly higher than that of samples prepared through vacuum drying.

4 Conclusions

Ultrasonic vacuum drying, vacuum drying, vacuum freeze-drying, hot-air drying, and spray drying influenced the structure of egg yolk lecithin at different degrees. Among others, the greatest influence was observed on the structure of egg yolk lecithin dried by ultrasonic vacuum, which can change its conformation, making the inter-chain order increased, and the fluidity of the membrane bilayer reduced. The influence of spray drying on the structure of egg yolk lecithin is less than that of the ultrasonic vacuum drying, with the little effect observed for other drying methods. The emulsifying capacity of egg yolk lecithin is closely related to its structure. The effects of drying methods on the emulsifying capacity of egg yolk lecithin are highly significant. The egg yolk lecithin sample prepared through vacuum freeze-drying presented the highest emulsifying capacity, followed by the samples processed through vacuum drying, hot-air drying, spray drying, and ultrasonic vacuum drying, respectively.

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