

Ultrasound-assisted Extraction of Plant-Based Proteins: A Novel Technique

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Received: 9.04.2021 | Revised: 13.05.2021 | Accepted: 18.05.2021

ABSTRACT

Ultrasound has been explored in recent times by researchers and food processing industries due to its capacity to increase extraction efficacy by enhancing mass transfer and rupturing cells due to acoustic cavitation. In protein extraction ultrasound treatment is generally used as a pretreatment in combination with the traditional method of extraction as it breaks the cell wall and improves the extractability. Ultrasound treatment not only improves the extraction rate but can also modify the functional properties of the protein product. The rate and yield of protein extraction depend on the operating conditions like power density of ultrasound, pulse, duration of ultrasonication, solid to solvent ratio, temperature, extraction solvent, pH, and so on. The current paper aims to present the impact of incorporating ultrasound technology with traditional technologies on the yield and functional properties of protein extraction from plant sources.

Keywords: *Ultrasound extraction; Plant proteins; Novel technique; Mechanism of ultrasound extraction.*

INTRODUCTION

The demand for protein ingredients has considerably increased in the last few years and is expected to grow in the coming years. Plant-based protein is one of the promising sources to fulfill the growing demand for protein ingredients, as they possess good nutritional and functional properties. There is a need to develop effective techniques to increase the yield and to improve the functional properties of the plant-based

proteins to make them cost-effective and industrially applicable.

Protein isolates contain protein more than 90% and concentrate contains more than 65% protein. Protein isolation is generally performed by solubilizing the protein at pH far away from its isoelectric point and then concentrating the protein through membrane filtration (ultrafiltration and diafiltration) or precipitating the protein at pH near to its isoelectric point (Rodrigues et al., 2012).

Cite this article: Ashfaq, A., & Younis, K. (2021). Ultrasound-assisted Extraction of Plant-Based Proteins: A Novel Technique, *Ind. J. Pure App. Biosci.* 9(3), 145-150. doi: <http://dx.doi.org/10.18782/2582-2845.8676>

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This is the conventional method of protein isolation but with the development of novel technologies, conventional methods are being modified to improve the yield and functional properties protein isolates. Ultrasound and microwave technology are novel techniques that are used on various protein-rich sources and showed positive results on the yield and protein functionality.

Ultrasound is a proven green technology that enhances the extraction efficiency when used as a pretreatment in combination with other classical extraction techniques (Rombaut et al., 2014). Ultrasound refers to sound waves having a frequency greater than the upper limit of hearing. Ultrasound waves travel in the medium through series of compression and rarefaction causing cavitation bubbles to generate. These bubbles grow in size by a process known as rectified diffusion and after attaining a critical size they collapse generating heat and damage to the surrounding medium like the disruption of biological cells. This property of ultrasound waves is used in extraction techniques where it causes disruption of cells and the release of intercellular material of interest. Sonication may cause some changes in the functional properties such as foaming, hydrophobicity, solubility, and emulsion of the isolated product due to the cavitation-induced shear (Rahman & Lamsal, 2021). This review aims to discuss the research work that has been done on the ultrasound-assisted protein extraction from plant sources and its impact on the functional protein of the extracted product, it will help to understand the principle and application of this technology in the field of protein extraction.

ULTRASOUND-ASSISTED PROTEIN EXTRACTION:

Ultrasound is an emerging technology in the field of extraction. Ultrasound-assisted

extraction has the potential to increase heat and mass transfer (Chemat et al., 2011). Ultrasound baths and ultrasound probes are used in the process of extraction. Ultrasound probes are found more efficient and powerful for the process of extraction because the ultrasonic intensity is delivered from a smaller surface as compared to an ultrasonic bath. High intensity of ultrasound probe causes increase in temperature and requires cooling for the process of extraction (Rombaut et al., 2014). In recent years ultrasound technique has become the source of attraction to researchers for the extraction of plant-based protein and modification of their functional properties (Rahman & Lamsal, 2021).

PRINCIPLE OF ULTRASONICATION:

Ultrasound waves are sound waves having a frequency in the range of 20 kHz to 10 MHz which is greater than the upper limit of human hearing. Ultrasound waves travel in the medium through series of compression and rarefaction. The phenomenon that aids in the process of extraction is cavitation (Rombaut et al., 2014). The alternating cycles of compression and rarefaction cause cavitation bubble Fig. 1 to grow and after attaining the critical size the bubbles collapse, creating microturbulence (Chemat et al., 2017). The collision of bubbles causes the generation of shear forces and temperature in the surrounding medium. at higher frequencies of ultrasound, the compression and rarefaction cycles are short, so they are less effective than the lower frequency of ultrasound (Rahman & Lamsal, 2021). When the bubble collapse, generates high-speed liquid jets which disrupts the cell wall and causes the release of intracellular material into the surrounding medium that as a result increases the extraction efficiency (Chemat et al., 2017).

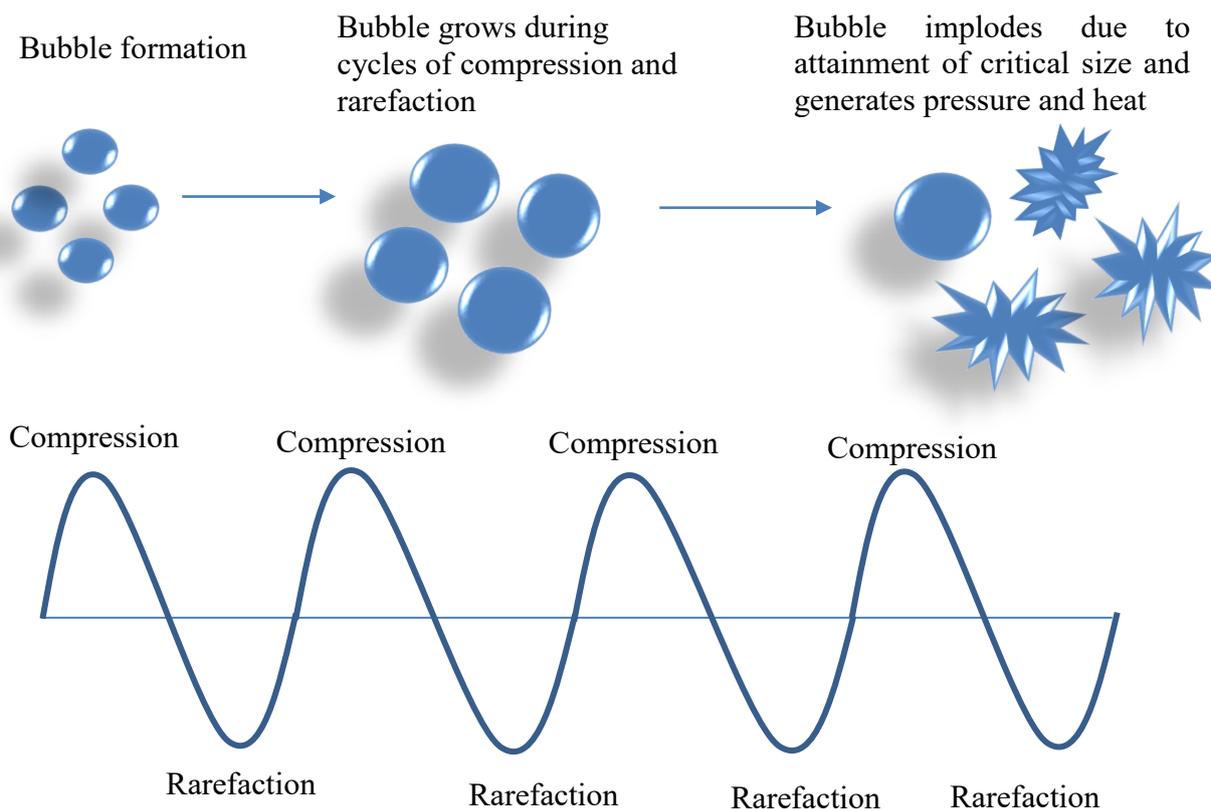


Figure 1: Generation of cavitation bubbles due to the propagation of sound waves via series of compression and expansion

APPLICATIONS OF ULTRASOUND TECHNOLOGY IN PLANT-BASED PROTEIN EXTRACTION:

Ultrasound can increase the extraction efficiency by increasing the rate of mass transfer between the plant material and surrounding solvent (Chemat et al., 2011). Ultrasound treatment increases the extraction yield, reduces the time of extraction, modifies the properties of extracted product, increases mass transfer and improves the solvent penetration.

The residue of wheat germ after the extraction of oil is known as defatted wheat germ and is a relatively higher source of protein containing essential amino acids methionine, lysine, and threonine. The most commonly used method for the extraction of protein from defatted wheat germ is alkaline extraction and isoelectric precipitation. (Zhu et al., 2009) used ultrasound-assisted technique for the extraction of protein from defatted wheat germ by reverse micelles and found that the extraction yield increases from 37% to

57% at ultrasonic output power 363 W, ultrasonic time 24 min and pulse mode 2.4s on and 2 s off (2.4 s:2 s). (Karki et al., 2009) studied the effect of ultrasound pretreatment on defatted soy flakes and concluded that the soy protein isolates yield increases at low and high-power ultrasound treatment for 120 s by 13% and 34% respectively. At high power treatment, soy flakes for 120 s few functional properties of soy protein isolate decreased and few increased like emulsification and foaming capacities decreased by 12% and 26% respectively, and solubility of the isolate improved by 34% when compared to control. (Dong et al., 2011) studied ultrasound-assisted, ultrafiltration, and isoelectric precipitation of rapeseed protein and obtained an increase in yield by 8.31%. (Li et al., 2017) reported that the countercurrent ultrasound-assisted extraction improves the extraction rate of rice dreg protein (RDP) and can be used industrially for the manufacture of the same as they found in the study that the extraction yield of RDP increased from 43.20% to

88.44% at NaOH concentration of 0.08 mol/L under ultrasonic concentration.

(Ayim et al., 2018) obtained extraction yield of 63.5% under ultrasound-assisted alkali extraction as compared to the alkali extraction (56.4%), and alkali and enzyme assisted extraction (47.8%) from tea (*Camellia sinensis* L.) residue. In the isolation of protein from sunflower meal, ultrasound treatment was incorporated with the method of alkaline extraction and the highest yield (54.26%) was obtained at power density (220 W/L), temperature (45 °C) and extraction time (15 min) (Dabbour et al., 2018). (Ly et al., 2018) used ultrasound in the extraction of protein from defatted rice bran and found that the use of ultrasound enhances the yield in comparison with the traditional method. Ultrasound-assisted extraction modifies some of the functional properties of extracted protein like in ultrasonic extraction of protein from defatted rice bran, the water absorption capacity, oil absorption capacity, and emulsion stability remain unchanged while gelation capacity increased and foaming capacity, as

well as stability of protein concentrate, decreased (Ly et al., 2018).

(Nguyen & Le, 2019) observed the increase in protein yield by 10.6% under ultrasound-assisted alkaline extraction at pH 6.8 from defatted peanut meal. (Byanju et al., 2020) obtained that the protein extraction yield increased from 8.4% (unsonicated) to 33.45% for lower power density (2.5W/cm³) and up to 30.6% for higher power density (4.5W/cm³) respectively. (Gia Loi Tu, 2015) studied the ultrasound and enzyme-assisted albumin extraction from defatted pumpkin seed powder and reported that the ultrasonic extraction rate is two times faster than the enzyme-assisted extraction. They also found that the water and oil holding capacity of ultrasound-assisted pumpkin seed albumin concentrate was better than that of enzyme-assisted albumin concentrate. The foaming capacity and emulsifying stability of ultrasound-assisted pumpkin seed albumin concentrate were slightly lower in comparison to enzymatically assisted extraction (Gia Loi Tu, 2015).

Table 1: Conditions for ultrasound-assisted extraction of plant-based protein

Plant source	Solid to solvent ratio	The solvent used and pH	Power or power density	Sonication Time	Yield (%)	Reference
Defatted wheat germ	0.5g in 50ml	Reverse micelles (AOT, isoctane and KCl solution, pH 8	363W, pulse mode 2.4s on and 2s off (2.4s:2s)	24 min	extraction yield increases from 37% to 57%	(Zhu et al., 2009)
Defatted soy flakes	1:10	Water and NaOH; pH 8.5	0.30 and 2.56W/ml	2 min	Protein isolate yield increased by 13% and 34%	(Karki et al., 2009)
Defatted rapeseed meal	1:20	Phosphate buffer; pH 11.6	450 W	80 min	Protein yield increased by 8.31%	(Dong et al., 2011)
Rice dreg flour	24g in 480ml	NaOH 0.02 to 0.10 mol/L	448 W, pulse duration / pulse interval ratio 10/6	40 min	Extraction yield from 43.20% to 88.44%	(Li et al., 2017)
Tea residue	51.5g/L	NaOH concentration (0.05, 0.1, or 0.15 M)	377 W/L	13 min	Protein yield increase from 56.4% to 63.5%	(Ayim et al., 2018)
Sunflower meal	50g in 1000 mL	Water and NaOH; pH 8.0	220 W/L	15 min	Protein yield increased from 28.0% to 54.26%	(Dabbour et al., 2018)
Defatted peanut meal	1:20	Water and NaOH; pH 6.8	30W/g	15 min	Protein yield increased by 10.6%	(Nguyen & Le, 2019)
Defatted soy flakes	1:10	Water and alkali; pH 8.5	2.5 and 4.5W/cm ³	5 min	Protein yield increased by 33.45% and 30.6% at 2.5 and 4.5W/cm ³ respectively	(Byanju et al., 2020)

CONCLUSIONS

Ultrasound technology has been proven as a promising technology in plant-based protein extraction. It increases the yield and rate of protein extraction and also can modify the functional properties. Ultrasound treatment depends on the conditions of extraction (like sonication power, time of sonication, temperature, pH solid to solvent ratio) as well as on the sources of protein and for better yield

and functionality it requires optimization. However, on the lab scale, the results of ultrasound-assisted extraction are positive but on this scale, the ultrasound is a batch type and the results are found to vary on a commercial scale. Therefore, more study on the commercial level is required for the application of ultra sonication in the plant-based protein industry.

Acknowledgments

The authors are thankful to the Integral University for providing the basic facilities.

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