

## A water extraction preparation method for the preparation of black soldier fly oil by CNPs@Cu immobilization catalyst

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The black soldier fly (*Hermetia illucens L.*) is a typical insect with a high fat content. It can convert large organic waste into high value-added biological matter, which can effectively solve the world health problem that is difficult to manage waste such as human and animal waste, and at the same time obtain nutritious black soldier fly biological products (Tao,2023). Because of its appropriate ratio of unsaturated fatty acids and saturated fatty acids in body fat, lauric acid and other medium-chain fatty acids are abundant, and have broad prospects for the development of feed oil, edible oil and industrial oil, compared with fishmeal, soybean and other insects, the content of black soldier fly larvae and pre-pupa oil is particularly rich (15%~49%), and the content of medium-chain saturated fatty acids dominated by lauric acid is relatively stable and rich. (Miebach,2023). Due to the distinctive fatty acid composition of black soldier fly oil, it has the potential to be used as a feed or edible oil, as well as as industrial oil such as cosmetics and biodiesel.

The larval biomass of the black soldier fly is rich in protein, lipid and various trace elements, which is one of the products of the biomass conversion and utilization industry, and its biomass contains 38.76% crude protein and 43.25% crude fat, which has high processing value. The resource utilization rate and added value of the black soldier fly are very low. How to develop the black soldier fly into high value-added edible and industrial products is one of the important contents of the diversified utilization and development of black soldier fly resources.

The main methods for extracting oils and fats in traditional ways include pressing, extraction, heating extraction, and supercritical CO<sub>2</sub> fluid extraction. In view of the close binding of oil and protein in black soldier fly, the current oil extraction process does not take into account the subsequent utilization of protein. Compared with the above methods, the water enzymatic method does not use organic solvents, has low operating temperature and low energy consumption, and with the help of the principle of immiscible water and oil, the hydrolyzed protein is catalyzed by protease to release oil, so as to achieve the purpose of simultaneously obtaining oil and protein hydrolysate, which is suitable for materials where oil and protein coexist, and can ensure the quality and activity of oil and protein hydrolysate. (Phongpradist,2023) In addition, the water enzymatic method, supplemented by physical enhancement such as ultrasound, has also been shown to achieve a high oil extraction rate in a short time. However, the traditional water enzymatic method and its physical enhancement method inevitably use a large number of free proteases for catalytic reactions, which are not recyclable, which will inevitably increase the cost of oil extraction. Therefore, the development of immobilized enzyme technology suitable for ultrasonic-assisted water enzymatic extraction is expected to construct a new synchronous separation process of oil and protein that comprehensively utilizes black soldier fly resources (Getachew,2023).

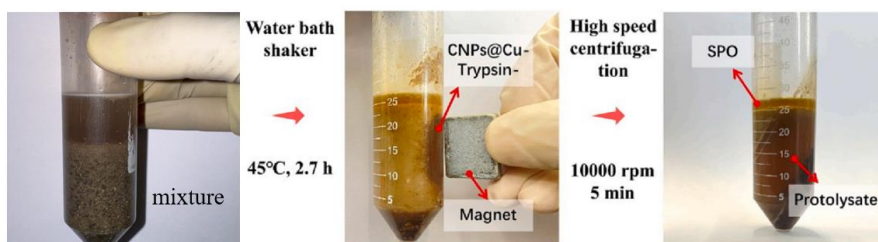


Figure 1 The application process of CNPs@Cu-Trypsin on the construction of CNPs@Cu-Trypsin-assisted aqueous extraction technique started

Table 1 provides a detailed list of ultrasound assisted organic solvent extraction, enzyme assisted water extraction, and CNPs@Cu-Trypsin-assisted oil extraction conditions of water extraction technology. The results show that the application of CNPs@Cu-Trypsin technology can achieve higher extraction rates of black soldier fly oil, with lower time costs and lower temperature requirements. The water extraction method has the highest oil yield ( $33.53 \pm 3.5\%$ ) and extraction rate ( $85.04 \pm 5.1\%$ ) for black soldier flies. Compared with ultrasound assisted organic solvent extraction and enzyme assisted water extraction technologies, the extraction time of water extraction technology was shortened by 32.5% and 55% respectively (from 10 and 6 hours to 4 hours), and the temperature was reduced by 10% and 43.75% respectively (from 80 and 50 °C to 45 °C), which may be beneficial for maintaining the nutritional value of the collected lipids and peptides.

Table 1 Extraction efficiency and oil properties corresponding to different extraction processes

Techniques	Temperature (°C)	Extraction time (h)	Yield Oil (%)	Acid value (mg/g)	Iodine value (g/100g)	Saponification value (mg/g)
Soxhlet extraction	ND	10	42.91±2.6	1.40±0.08	57.51±6.61	207.01±9.54
Enzyme-assisted aqueous extraction	50	10	29.91±5.3	2.48±0.32	66.14±4.25	222.15±6.44
Solvent extraction	60	4	28.45±4.1	1.95±0.24	61.95±3.25	204.15±6.27
CNPs@Cu-Trypsin-assisted	45	4	33.53±3.5	1.29±0.01	50.84±9.86	190.13±6.73

The aqueous extraction and preparation method of oil from black soldier fly using CNPs@Cu immobilization catalyst was mainly studied, and a new method for preparing oil from black soldier fly larvae and dried insects was studied. Compared with the prior art, the preparation method of the present invention is simple to operate, the enzymatic hydrolysis stability is high, the specificity is strong, and the oil yield is high (Ganesan,2024). Therefore, improving the effect of enzyme-assisted water extraction by trypsin immobilization and divalent copper ion chelation technology is a feasible strategy for green and sustainable extraction of edible insect oils and proteins without the use of organic solvents.

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