Ultrasound-assisted Aqueous Enzymatic Extraction of Corn Germ Oil: Analysis of Quality and Antioxidant Activity

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Abstract: An efficient method of ultrasound-assisted aqueous enzymatic extraction of corn germ oil was established, and its quality and antioxidant activity were studied. The optimum auxiliary extraction conditions with ultrasound were as follows: the ultrasonic treatment time was 20 minutes, and the ultrasonic temperature was 40°C. The corn germ oil extracted by the aqueous enzymatic method had better quality indexes in comparison with that extracted by the solvent. The fatty acid compositions of corn germ that was oil-extracted by two kinds of methods had almost no significant differences. The tea polyphenols (TP) exhibited remarkable antioxidant effects on corn germ oil during a storage stability test. Meanwhile, we focused on the antioxidant activity of corn germ oil, and the results showed that corn germ oil could effectively scavenge 1,1-diphenyl-2-picrylhydrazyl (DPPH), hydroxyl radical and superoxide anion free radicals. Thus, corn germ oil is a kind of functional oil with excellent antioxidant activity and could be used as a free radical scavenger in the food or other industries.

Key words: corn germ oil, ultrasound, aqueous enzymatic extraction, quality, antioxidant activity

1 Introduction

As we know, the main extraction methods of corn germ oil are traditionally pressing and solvent extraction1). The aqueous enzymatic method is a new kind of oil extraction technology, and research of this method begun in recent years both in China and abroad2). This method causes the oil release by means of mechanical fragmentation, and enzymes degrade the plant cell components3). We used cellulase and α-amylase as hydrolases in this experiment. Cellulase can efficiently degrade the plant cell wall4), while α-amylase can break down starch5), making oil precipitation easier and increasing the oil extraction rate. Most pretreatment methods of the aqueous enzymatic method were heat and steam treatments, and the ultrasonic treatment method is a new kind of oil extraction technology6). The strong vibration, the cavitation effect7) and other functions that result from ultrasound can increase the extraction efficiency, making the quality of the oil better, saving raw materials and improving the oil yield at the same time8).

The unsaturated fatty acids in the oil will oxidate and decompose due to various complex factors during the transport or storage process9). To inhibit lipid oxidation and keep the oil fresh, the most effective method that we can make use of is adding antioxidants to the edible oil. In this way, we can easily prolong the shelf life of the oil. Among those that are currently used frequently are BHT (butylated hydroxytoluene), BHA (butyl hydroxy anisid), TBHQ (tedinki hydroquinone) and other synthetic antioxidants; these antioxidants have certain effects of preventing the oil from rancidity. However, some animal experiments have shown that they have toxic10) and carcinogenic effects11). People doubt of the safety of synthetic food additives. These factors make use of these additives, which are controlled, and many countries have introduced regulations that strictly limit the amount added. Thus, increasing attention is being paid to the development12) of natural food antioxidants13) with no side effects, which have the advantages of high safety, strong antioxidant capacity, easy preservation and other characteristics.

In recent years, many scholars have researched the antioxidant activity of corn tassel14) and corn fiber15), but the antioxidant activity of corn germ oil has not been investigated. Reactive oxygen species (ROS) are the intermediate product of human metabolism, and they maintain a
dynamic balance in the human metabolism. However, when lesions occur, such as in cancer, aging, or cardiovascular and cerebrovascular diseases, ROS will accumulate. Screening plant seed oils with antioxidant properties is very beneficial for supplying it as dietary food components as specific preventative pharmaceuticals. The DPPH radical is one of the few stable radical sources, and it has been extensively used to determine antioxidant electron-donating and free radical-scavenging ability. In addition, the hydroxyl free radical and the superoxide free radical are also ROS.

In this experiment, we studied the quality of corn germ oil extracted by the ultrasound-assisted aqueous enzymatic method, including physical and chemical properties, fatty acid composition and storage stability, which was compared with that of traditional solvent (hexane) extraction. The relationship between the extraction methods of corn germ oil and its quality would be comprehensively evaluated through these indexes. In addition, the antioxidant activity of corn germ oil was also studied. We investigated the scavenging effect of corn germ oil on free radicals in vitro antioxidant systems to provide a theoretical basis for the development of comprehensive utilization of corn by-products and functional oils.

## 2 Materials and methods

### 2.1 Materials preparation

Corn germ was provided by the Xiangfang farm and stored at 4°C until it was ready for use. The cellulase (50,000 U/g, the optimum temperature: 40-50°C, pH: 4.8) was purchased from Shanghai Huishi Biochemical Reagent Co., Ltd. TBHQ, TP, Vitamin E, Vitamin C and 1,1-diphenyl-2-picrylhydrazyl (DPPH) were purchased from Shanghai Yuanye Biochemical Regent Co., Ltd. All other chemicals and reagents were of analytical grade.

### 2.2 Ultrasonic assisted aqueous enzymatic extraction procedure

The corn germ was sifted out by a 60 mesh after being crushed and then placed under a sieve until it was ready for the experiment. First, the raw material (50 g) was added to a certain amount of citric acid and sodium citrate buffer (0.05 M) prior to preparation. Next, the sample was treated by three different methods: water bath (20 min at 60°C), steam (20 min at 100°C) and an ultrasonic (20 min at 25°C). After the treatment and cooling to room temperature (23°C), we needed to adjust the optimum pH of cellulase and α-amylase using 0.5 M NaOH and 0.5 M HCl. Then, we placed the sample in a thermostatic mixer for an enzymolysis reaction (50°C for 2 h). After the reaction, enzyme deactivation was carried out at 95°C for 5 min. After enzyme deactivation, we removed the sample from the blender and cooled it to room temperature. Then, the sample was transferred into a centrifuge tube for centrifugation (8000 g for 20 min). After centrifugation, the supernatant oil layer was carefully removed by absorption using a micro-pipette. The lower layer of the oil emulsions was frozen and thawed. Finally, centrifugation was carried out again, resulting in more free oil separation. The free oil yield is the total of the two parts. The effect of the three pretreatment methods on the yield of free oil was compared. The oil yield was calculated as the weight of extracted oil divided by the total weight of oil present in corn germ sample (calculated based on a wet basis), shown as the following equation:

\[
\text{Oil yield} (\%) = \frac{\text{Weight of extracted oil (g)}}{\text{Total weight of oil in corn germ sample (g, wet basis)}} \times 100
\]

Meanwhile, we studied the effects of ultrasonic pretreatment conditions on the yield of free oil. The ultrasonic time was set from 0 to 40 min; temperature was set from 10 to 50°C.

### 2.3 Physicochemical properties of corn germ oil

#### 2.3.1 Determination of physicochemical properties

The physicochemical properties of corn germ oil included the fatty acid composition, refractive rate, acid value, iodine value, peroxide value and saponification value. Meanwhile, the control group, which was extracted with hexane, was analyzed.

Among these indexes, refractive rate, acid value, iodine value, peroxide value and saponification value were determined in accordance with Chinese standard methods (CSM). Fatty acid compositions of corn germ oil were determined by gas chromatography. The fatty acid composition analysis was performed according to Li et al.

#### 2.3.2 Storage stability test

The effects of three kinds of natural antioxidants on the storage stability of corn germ oil were tested by the Schaal oven method. The corn germ oil with antioxidants was placed in the oven at 65 ± 1°C without light for 20 days, and the peroxide value of the samples would be determined every four days. The storage stability of corn germ oil was studied from the effect of several aspects, including the antioxidant species, the added amount, the different antioxidant compounds and the synergistic agents on the antioxidant effect. Meanwhile, the control group and corn germ oil which had added synthetic antioxidant TBHQ were analyzed.

### 2.4 Determination of antioxidant activity

The experiment determined the scavenging abilities of corn germ oil on three different free radicals including the DPPH free radical, the hydroxyl free radical and the super-
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The antioxidant activity of corn germ oil is reported as an IC_{50} value (the concentration of the sample is required when 50% of the free radicals are scavenged). Simultaneously, the control group of antioxidant Vitamin C was analyzed.

2.4.1 DPPH scavenging assay

The reaction mixtures contained 2 mL of sample solution in ethanol and 2 mL of DPPH solution in ethanol (0.05 mM) in the test tube. The tubes were reacted for 30 min in darkness after shaking. The absorbance value of reactants was determined at 517 nm, which was denoted as A₀, while the absorbance value of other mixtures, including the 2-mL sample solution and the 2 mL of ethanol, were denoted as A₂. Then, we took 2 mL of ethanol and 2 mL of DPPH solution in a mixture and determined the absorbance value after the reaction, which was denoted as A₀. At the same time, the control group of antioxidant Vitamin C was determined. The calculation formula was as follows:

Scavenging rate (%) = \left[1 - \frac{A_1 - A_2}{A_0}\right] \times 100

2.4.2 Hydroxyl radical scavenging assay

The reaction mixtures of 2 mL of FeSO₄ solution (9 mM) in pure water and 2 mL of H₂O₂ solution (8.8 mM) in pure water were reacted at 37°C for 15 min. The absorbance value of the reactants was determined at 536 nm, which was denoted as A₀. Next, we added 2 mL of sample solution in pure water to the above mentioned mixtures and determined the absorbance value, again denoted as A₁. Meanwhile, the control group of antioxidant Vitamin C was determined. The calculation formula was as follows:

Scavenging rate (%) = \left[1 - \frac{A_0 - A_1}{A_0}\right] \times 100

2.4.3 Superoxide anion scavenging assay

The superoxide radical scavenging was investigated with the pyrogallol autoxidation method. First, 4.5 mL of Tris-HCl buffer (pH 8.2) and 3.2 mL of pure water were added to the test tube with 1 mL of sample solution. After reacting at 25°C for 10 min, 300 μL of pyrogallol solution (8 mM) in pure water was added to the mixtures immediately and reacted for 4 min. The addition of 100 μL of an HCl solution (6 M) terminated the reaction. Finally, we determined the absorption value, denoted as A₁, at 325 nm. The absorption value of pure water in the reaction instead of the sample solution was denoted as A₀. The control group of Vitamin C was also determined. The calculation formula was as follows:

Scavenging rate (%) = \left[1 - \frac{A_0 - A_1}{A_0}\right] \times 100

2.5 Statistical analysis

All data are presented as the mean ± standard deviation (SD) in at least three parallel measurements. The data were statistically analyzed using Origin 7.5 and SPSS 20.0. The differences between the means were determined by analysis of variance (ANOVA), and the statistical significance was established by letters in the figures and tables when p<0.05. The same letter indicated no significant differences, and the different letters indicated significant differences.

3 Results and discussion

3.1 Ultrasound-assisted aqueous enzymatic extraction of corn germ oil

Cellulase was selected to facilitate the destruction of the cell structure for the realization of oil from the lipid bodies. Three different pretreatment methods were adopted in this experiment. From the experimental results (Fig. 1), we know that compared with the auxiliary pretreatment methods of the water bath and steam heating ultrasound had a significant effect on improving the extraction rate of corn germ oil. That was to say, the combination of ultrasonic treatment and the aqueous enzymatic method had a significant effect on the bioprocess efficiency. The influence of different ultrasonic conditions on oil yield was studied. Figure 2 shows that the oil yield was found to increase (from 51.68% to 67.54%) with the increase in time from 0 to 20 min and then maintain constant when the time continuously increased. The research of Zhao, Kwok, and Liang showed that as the diffusion area reduced, the diffusion distance increased, and the diffusion rate would decrease when the diffusion front moved from the surface of perilla seed powder particles toward the interior. Thus, the continuous increase in ultrasonic time would not improve the oil yield. Similar results have also been shown

Fig. 1 Effect of three different pre-treatment methods on the yield of free oil water bath (20 min at 60°C); steam (20 min at 100°C); ultrasonic (20 min at 25°C) (p<0.05).
in the studies of ultrasound-assisted extraction of oils from flaxseed\textsuperscript{30} and perilla seeds\textsuperscript{25}. Thus, we choose 20 min as the best ultrasonic time. The effect of ultrasonic temperature on the oil yield was shown: the oil yield increased from 56.20\textdegree{} to 66.25\textdegree{} with an increase in temperature from 10 to 40\textdegree{} and then leveled out after 40\textdegree{}. Temperature affects the performance of the ultrasonic pretreatment by influencing the amount of vapor in ultrasonically generated bubbles. The vapor at high temperatures will contribute to forming vapor-filled bubbles. The explosion of those bubbles would generate an effect, termed the cushioning effect\textsuperscript{31}. As a result, the cavitational effect would be weakened; thus, the oil yield remained unimproved with continuously increasing the temperature. Therefore, 40\textdegree{} was selected as the optimum ultrasonic temperature. Subsequently, we began to extract the corn germ oil in the aqueous enzymatic method, which was assisted by the optimum ultrasonic conditions according to the references\textsuperscript{32, 33}.

3.2 Physicochemical properties of corn germ oil

In this experiment, the effect of the aqueous enzymatic extraction method on the quality of corn germ oil was under analysis in comparison with the solvent (hexane) extraction method. As shown in Table \ref{table1}, the refractive rate obtained by the two extraction methods of corn germ oil had few differences. Our research results agree with those of Latif, Diosady, and Anwar\textsuperscript{34}. In their study, the effect of different enzymes on the aqueous enzymatic extraction of canola oil was investigated by comparing the quality of extracted oils, and no significant differences were observed in refractive index. Similar results were reported by Yang Li \textit{et al.}\textsuperscript{35}. They found the iodine value, refractive index, unsaponifiable matter, saponification value, and peroxide value of the perilla seeds oils were not significantly affected by different extraction means and enzymes. The acid value of corn germ oil extracted using the aqueous enzymatic method was slightly higher than that of the solvent extraction. This was mainly because the aqueous enzymatic extraction method was carried out in an acidic environment. In the ultrasonic treatment process, the pH of the buffer had a great influence on the acid value of the corn germ oil. Except for the pH, the physical and chemical indicators of the corn germ oil in the aqueous enzymatic extraction were better than those in the solvent extraction. In general, the corn germ oil had better qualities in the aqueous enzymatic extraction method.

\begin{table}
\caption{Analysis of physicochemical properties of corn germ oil by different extraction methods.}
\begin{tabular}{llll}
\hline
Physicochemical indicators & Aqueous enzymatic method & Solvent extraction \\
\hline
Refractive rate (n\textsubscript{D}20) & 1.4737 ± 0.04\textsuperscript{a} & 1.4739 ± 0.05\textsuperscript{a} \\
Acid value (mg/g) & 3.36 ± 0.13\textsuperscript{a} & 2.76 ± 0.10\textsuperscript{a} \\
Iodine value (g/100 g) & 102.72 ± 4.01\textsuperscript{a} & 105.67 ± 3.96\textsuperscript{a} \\
Peroxide value (meq/kg) & 0.86 ± 0.02\textsuperscript{b} & 1.82 ± 0.07\textsuperscript{a} \\
Saponification value (mg/g) & 182.02 ± 3.46\textsuperscript{a} & 193.28 ± 3.87\textsuperscript{a} \\
\hline
\end{tabular}
\end{table}

Values are given as the means ± SD from triplicate determinations; in the same line with different letters indicates significant differences (p < 0.05).
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The fatty acid composition of corn germ oil was studied by gas chromatography. We concluded that the aqueous enzymatic extraction method and solvent extraction method had few effects on the fatty acid contents. The corn germ oil produced from the two extraction methods was basically the same in terms of fatty acid compositions and contents, and the following results showed that corn germ oil of the two extraction methods had more than 70% unsaturated fatty acid contents. The main fatty acid compositions in corn germ oil were oleic acid, linoleic acid, palmitic acid, stearic acid, linolenic acid, and a small amount of arachidonic acid, palmitic acid and peanut acid (Fig. 3). The contents of each fatty acid composition are shown in Table 2. Abdulkarim et al. compared the content of fatty acids in the oil samples from enzymatic and hexane extractions, and no significant difference was found. These results concur with Winkler-Moser and Vaughn’s reports, which showed that the fatty acid compositions of all five oils extracted from distiller’s dried grains with solubles (DDGS) and the thin stillage were typical of corn oil.

### 3.3 Stability of corn germ oil

The effect of three kinds of natural antioxidants on the oxidation stability of corn germ oil was researched by studying TP, Vitamin C and Vitamin E.

#### 3.3.1 Effects of single antioxidants on the stability of corn germ oil

Figure 4(a) showed that peroxide values of all tested groups increased with the extension of storage time. The peroxide value of blank group increased fastest. TBHQ had the strongest antioxidant capacity, while the antioxidant effect of TP was the best in three kinds of natural antioxidants. The antioxidant capacities of Vitamin C and Vitamin E were the closest and weakest. Because natural antioxidants are more suitable for people in pursuit of green and healthy food resources, we studied the effect of adding the amount of TP on the stability of corn germ oil; see Fig. 4(b). We found that with TP contents increasing, the oxidation time of the oil could be significantly delayed. In the dosage range of allowed additives, with increased amounts of additive, we obtained better antioxidant effects. Some studies reported that CG oil showed the

### Table 2  Fatty acid constituents and their contents in corn germ oil by different extraction methods.

| Fatty acids (%) | Aqueous enzymatic method | Solvent extraction |
|----------------|--------------------------|--------------------|
| Palmitic acid (C16:0) | 12.80 ± 0.27a | 13.01 ± 0.26a |
| Palmitic acid (C16:1) | 0.17 ± 0.01a | 0.16 ± 0.01a |
| Stearic acid (C18:0) | 1.56 ± 0.06a | 1.75 ± 0.05a |
| Oleic acid (C18:1) | 22.09 ± 0.66a | 23.28 ± 0.61a |
| Linoleic acid (C18:2) | 51.56 ± 0.52a | 51.92 ± 0.57a |
| Linolenic acid (C18:3) | 0.50 ± 0.02a | 0.49 ± 0.01a |
| Arachidic acid (C20:0) | 0.33 ± 0.01b | 0.44 ± 0.02a |
| Arachidonic acid (C20:1) | 0.34 ± 0.01a | 0.38 ± 0.01a |

Values are given as the means ± SD from triplicate determinations; in the same line with different letters indicates significant differences ($p < 0.05$).
highest rate of increase in peroxides when stored at 40°C compared with oil extracted from DDGS and centrifugally extracted from a thin stillage syrup. This is because the latter contains more functional fats, such as tocopherols, tocotrienols, and carotenoids. DDGS oil has the lowest peroxide value, which is probably due to the presence of Maillard reactants, resulting in the drying of the distiller’s grains.

3.3.2 Effect of complex antioxidants on the stability of corn germ oil

The complex antioxidants made with a mixture (1:1) of natural antioxidants were added to corn germ oil. With the storage time increasing, the peroxide value (POV) of each experimental group increased, among which the control group increased fastest and the TP + Vitamin C group increased the slowest. On the twentieth day, the POV of each group, TP + Vitamin C, TP + Vitamin C + Vitamin E, TP + Vitamin E, Vitamin C + Vitamin E and control group (CK), by the sequence from small to large reached 16.25, 21.73, 23.65, 37.52 and 42.30 mmol/kg, respectively. We found that the composite antioxidant effect was better than that of a single antioxidant. The combination of TP and Vitamin C had the best antioxidant effect, with the lowest POV at 16.25 mmol/kg. Dai studied the synergistic antioxidant effect of polyphenols extracted from green tea with α-tocopherol (vitamin E) and L-ascorbic acid (vitamin C) against the peroxidation of linoleic acid in sodium dodecyl sulfate (SDS) micelles. Kinetic and mechanistic studies on the antioxidation process revealed that this antioxidant synergism was due to the regeneration of vitamin E by the green tea polyphenol and the regeneration of the latter by vitamin C. Our experimental results are consistent with their conclusion. Their synergistic antioxidant effect may stem from vitamin C regeneration to tea polyphenols. In addition, Chen et al. reported that addition of vitamin C could increase the stability of green tea polyphenols. Lotito

Fig. 4 (a), Effect of single antioxidant on the stability of corn germ oil. (b), Effect of the TP adding amount on the stability of corn germ oil. The corn germ oil with antioxidants was placed in the oven at 65(±1) ºC without light for 20 days, and the peroxide value of the samples would be determined every four days. POV, peroxide value; CK, control group.

Fig. 5 Effect of complex antioxidants and synergists on the stability of corn germ oil: (a), complex antioxidants; (b), synergists. The corn germ oil with antioxidants was placed in the oven at 65(±1) ºC without light for 20 days, and the peroxide value of the samples would be determined every four days. POV, peroxide value; CK, control group.
research shows that adding vitamin C can completely inhibit the decay of green tea polyphenols during 2,2'-azobis(2-amidinopropane) hydrochloride (AAPH)-initiated lipid peroxidation in human plasma. We already knew that citric acid had a capacity for synergism; therefore, we also investigated its synergistic antioxidant effects with TBHQ and TP. At the same time, we used Vitamin C as a comparison. Figure 5 shows the collaborative groups have a significantly lower POV compared with the control group, indicating that the experimental groups showed significant antioxidant effects of coordination, among which the TBHQ + CA group has the highest antioxidant activity with the lowest POV (9.40 mmol/kg) in 20 days.

3.4 Antioxidant activity of corn germ oil

3.4.1 DPPH scavenging assay

As we can see from Fig. 6(a), with the increase of Vitamin C concentration in corn germ oil, its ability to scavenge free radicals is increased. Through analysis of the linear regression between concentrations and scavenging
rate, we could obtain the corresponding IC$_{50}$ value. The IC$_{50}$ values of corn germ oil and Vitamin C were 27.38 mg/mL and 0.19 mg/mL, respectively, and the IC$_{50}$ value of corn germ oil was approximately 144 times as much as Vitamin C. The corn germ oil’s ability to scavenge DPPH was weaker than that of Vitamin C, but the corn germ oil also had a remarkable DPPH radical scavenging effect. The scavenging ability of *Camellia oleifera* seed oil (COSO), extracted by an organic solvent and the salt effect-aided aqueous extraction process (AEP-SE), on DPPH was compared by Yu, X. et al.\(^\text{42}\), and the IC$_{50}$ values of oil obtained by organic solvent extraction and AEP-SE were 3.31 mg/mL and 2.27 mg/mL, respectively. This could be because *Camellia oleifera* seed oil contains more unsaturated fatty acids than corn germ oil.

3.4.2 Hydroxyl radical scavenging assay

The hydroxyl radical is the most active free radical, and it can attack all biological molecules by causing free radical chain reactions\(^\text{43}\). Thus, removing hydroxyl radicals is important for protecting living systems. As shown in Fig. 6 (b), corn germ oil was capable of scavenging hydroxyl radicals within a relatively low concentration range of 0.5-2.5 mg/mL. When the concentration was more than 1.5 mg/mL, the increasing trend of the scavenging capacity slowed down. The IC$_{50}$ values of Vitamin C and corn germ oil were 0.029 mg/mL and 1.574 mg/mL, respectively. Compared with the DPPH scavenging capacity, corn germ oil had a better capability to scavenged hydroxyl radicals. Jin-Won et al.\(^\text{44}\) investigated the antioxidative activity of methanol and ethanol extracts from five seed oils (*perilla seed oil, pine seed oil, Camellia seed oil, flaxseed oil, and olive oil*), and CSO-ME (*Camellia* seed oil extracts by methanol and ethanol) had the highest hydroxyl radical scavenging activity among all seed oil extracts, reaching 64.5%. This is because CSO-ME had more available antioxidant components. The free radical scavenging rate was positively correlated with the content of polyphenol compounds and their hydrogen donating capacity. The more polyphenol compounds, the more the reducing force accounted for the higher free radical scavenging rate\(^\text{45}\).

3.4.3 Superoxide anion scavenging assay

The Fenton reaction occurs with a superoxide anion of a metal ion catalyzing a reaction, which could produce a highly reactive hydroxyl radical; thus, the superoxide anion radical was commonly used for its scavenging ability. We can see from Fig. 6 (c) that with the increase of mass concentration, Vitamin C and corn germ oil had an increasing trend in superoxide anion radical scavenging capacity. When the concentration of corn germ oil ranged from 5-25 mg/mL, it had a significant scavenging effect on the superoxide anion. The IC$_{50}$ value of corn germ oil was 24.188 mg/mL, and that of Vitamin C was 0.022 mg/mL. Although the scavenging capacity of corn germ oil on superoxide anion free radical was weaker than that of Vitamin C, corn germ oil showed a higher capacity for scavenging the superoxide anion radical compared with DPPH.

4 Conclusions

The auxiliary method of ultrasound could exert a positive effect on the aqueous enzymatic method and significantly improved the free oil extraction rate. Because the ultrasonic waves break down cells and strengthen the transferred mass so that the buffer molecules can penetrate the tissue cells, a perfect contact with solute molecules is achieved. As a result, the cells of soluble components will easily be set free, and the rate of corn germ oil extraction is improved. In terms of physical and chemical indicators, the corn germ oil extracted by the aqueous enzymatic method had a better quality than that of solvent extraction, but the fatty acid compositions and contents had almost no differences between these two extraction methods. During the storage time, TP as a natural antioxidant had a better antioxidation effect on corn germ oil compared with Vitamin C and Vitamin E, but the capacity of TP was weaker than that of the synthetic antioxidant TBHQ. In contrast, the composite antioxidants had a better antioxidation effect relative to single antioxidant, resulting in a synergetic effect. Corn germ oil had certain scavenging capacities for the DPPH free radical and especially the hydroxyl free radical and the superoxide anion free radical. In summary, corn germ oil is a good antioxidant and free radical scavenger. If a certain amount of corn germ oil is added to edible oils and fats, not only could the storability be improved but also a positive effect on human health could occur.

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