Analysis of the biological function and application status of apple polyphenols

Yujing Chen*
School of Shanghai University of Medicine & Health Sciences, Shanghai, China
*Corresponding author: chenyujing@stu.sumhs.edu.cn

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Abstract: Polyphenols are a class of secondary metabolites of plants with biological activity, and are the most abundant biologically active components in fruits and vegetables. Apple polyphenols are the general term for polyphenols contained in apples. The content of apple polyphenols is affected by fruit maturity, cultivation methods, environmental conditions and storage conditions. Apple polyphenols are particularly rich in immature apples, and the polyphenol content of immature fruits is 10 times that of mature fruits. A large number of efficacy studies have shown that apple polyphenols have multiple biological functions such as antioxidant, antibacterial, and elimination of odor, anti-cancer, lowering blood pressure, and lowering blood sugar. At present, apple polyphenols have been widely used in medicine, food, daily chemicals and other fields. This article analysed and comprehensively summarized the composition, physical properties, preparation technology, biological function and application status of apple polyphenols, and provides ideas for the future development and utilization of apple polyphenol resources.

1. Introduction

Apple polyphenols are the general term for polyphenols in apples, and they are a kind of important biologically active substances that occur naturally. The content of apple polyphenols is affected by a series of factors such as maturity planting method, geographical environment, storage method, etc., showing a large range of variability. Apple polyphenols are mainly extracted from immature apples and pomace. The content of polyphenols in immature apples is rich, which is about 10 times that in mature apples. The physiological activity research of apple polyphenols has been initially verified on multiple levels, including cell culture analysis, pathological model animal research, clinical intervention, population experiment and epidemiological investigation. A large number of efficacy studies have confirmed that apple polyphenols have a variety of biological functions such as anti-oxidation, antibacterial, elimination of peculiar smell, anti-cancer, lowering blood pressure, and lowering blood sugar. In recent years, with more and more researches on apple polyphenols, the physiological activity of apple polyphenols has attracted more and more attention. Apple polyphenols are the main material basis for the functional activity of apples, and are closely related to the formation of sensory qualities such as flavor, color and texture in apple products and processed foods. In this paper, by combing the research progress related to apple polyphenols, elucidating the composition, characteristics and preparation process of apple polyphenols, analyzing the physiological activities of apple polyphenols and the main applications on the market at present, so as to promote the comprehensive development and utilization of apple resources.

2. Introduction of apple polyphenols

2.1 Composition of apple polyphenols

Apple polyphenols are secondary metabolites of apples. They are the general term for polyphenols contained in apples. There are mainly 5 categories, including phenolic acids, flavanols, dihydrochalcones and anthocyanins [1]. Phenolic acid is mainly composed of chlorogenic acid and caffeic acid. Flavonoids mainly include catechins, epicatechins, proanthocyanidins, and quercetin.
Compared with mature apples, immature apples are similar in composition, but there are significant differences in content. The polyphenols of mature apples are mainly chlorogenic acid, catechins and proanthocyanidins, while immature apples are mainly compounds such as dihydrochalcone and quercetin.

2.2 The physical properties of apple polyphenols

Apple polyphenols are brown-red powder, 20% aqueous solution is reddish-brown, and 100% powder is yellow-brown. Both liquid and powdered apple polyphenols have a slight apple flavor and a bit bitter taste. Apple polyphenols are easily soluble in water, methanol, ethanol and other solvents. Apple polyphenols have good stability. They can be stored in a cool and dry environment for one year in powder form, and their properties and physiological functions are almost unchanged. The 0.1% to 1% aqueous solution of apple polyphenols was heated for 30 minutes (100°C) within pH 2 to 10, and the preservation rates were all above 80%.

2.3 Factors affecting the content of apple polyphenols

The content of polyphenols in apples varies greatly due to different planting conditions and growth environments, varieties, maturity, processing techniques and storage conditions. In the process of apple growth and maturation, the composition of polyphenols is not static. The apples of the same kind use starch-iodine staining method to determine the ripeness of the apples and the standard curve is prepared by gallic acid to determine the content of apple polyphenols of different ripeness. As the fruit matures, the total phenol content will decrease. Planting conditions and growth environment have a certain impact on apple polyphenols, that is, different varieties of apples have different polyphenol content. Studies have shown that there are big differences in the content of apple polyphenols between different varieties, whether it is the peel group or the peel and pulp group, and the peel group is significantly higher than the peel and pulp group [2]. Processing technology also affects the composition of polyphenols. This is mainly determined by the storage time of the apple and the processing methods such as sterilization and enzymes. The total phenol content in pressed apple juice was significantly lower than that in extracted apple juice. After the apple is harvested, before processing and storage, the content of main phenols will drop sharply to a level and then basically remain unchanged. The total phenol content of apples is relatively stable during cold storage. Some researchers have found that the content of flavonoids and chlorogenic acid in apples stored under ultra-low oxygen concentrations at 1°C is not significantly different from that under normal oxygen concentrations. During storage, only catechins, quercetin and chlorogenic acid have slight changes [3].

2.4 Preparation process of apple polyphenols

For the extraction method of apple polyphenols, by reviewing and summarizing scientific and technological literature and technical data, the extraction method of apple polyphenols is currently mainly used organic solvent direct extraction method, ultrasonic assisted extraction method, microwave assisted extraction method, pressurized solvent extraction method, supercritical fluid extraction and so on. At present, the direct extraction of apple polyphenols by organic solvents is the most widely used extraction method in industrial production. There are two more mature extraction methods, as shown in Figure 1. Since the hydroxyl group contained in the structure of apple polyphenols has a certain polarity, water, ethyl acetate, acetone, etc. are usually used as the solvent. The method utilizes the principle of compatibility and has the advantages of simple production technology and high extraction rate of polyphenols. However, problems such as high solvent consumption and high production cost in the production process have not yet been solved.

The ultrasonic-assisted extraction method uses the strong cavitation effect, disturbance effect, mechanical vibration, high acceleration, crushing and stirring produced by ultrasonic radiation to increase the frequency and speed of the molecular motion of the substance and the penetration force of the solvent and accelerate the entry of the target component into the solvent. Compared with the organic solvent direct extraction method, trasonic-assisted extraction method greatly improves the extraction efficiency, saves the solvent and avoids the influence of high temperature on the extracted...
components. Microwave-assisted extraction is a new technology that uses microwave energy to improve extraction efficiency. The extraction solvent is heated by microwave energy to separate the target compound from the sample matrix into the solvent. With the continuous innovation of technological research, the research and application of new technologies such as pressurized solvent extraction, supercritical fluid extraction and high-pressure assisted extraction have opened up more and better ways for the research of apple polyphenol extraction technology.

Figure 1. Flow chart of two more mature direct extraction methods with organic solvents.

3. The biological function of apple polyphenols

Apple polyphenols have many biological functions such as anti-oxidation, antibacterial, elimination of peculiar smell, anti-cancer, lowering blood pressure, and lowering blood sugar.

3.1 Antioxidant

The antioxidant effect of apple polyphenols is accomplished by directly and indirectly scavenging free radicals. A large number of studies have shown that excessive production of free radicals and decreased scavenging ability are the biochemical mechanisms that trigger many diseases, and they are also the main cause of body aging or aging. It is concluded that apple polyphenols have a good scavenging effect on O$_2$·, ·OH and R·. Therefore, apples polyphenols have anti-oxidation, anti-aging effects and keep food fresh [4].

3.2 Antibacterial

Apple polyphenols have antibacterial effects. It can destroy the lipid layer of the bacterial cell membrane and because the morphological changes of the bacteria; inhibit the activity of harmful
bacteria to secrete toxins; inhibit the infection of harmful bacteria. The minimum inhibitory concentration (MIC) of apple polyphenols against bacillus, escherichia coli, pseudomonas and other tested bacteria is 0.1%. The antibacterial activity of apple polyphenols has good thermal stability, and its antibacterial effect is best under the environmental conditions of pH 5-6 and less than 0.3mol/L inorganic salt [5]. Streptococcus mutans is considered to be the main pathogenic bacteria causing dental caries. The glucosyltransferase (GTase) secreted by it can convert the soluble sucrose in the oral cavity into adhesive insoluble polysaccharides, thereby forming dental plaque. Dental plaque is a hotbed for the survival of cariogenic bacteria, more microorganisms will accumulate and multiply in it, produce acid to dissolve enamel and destroy teeth. Studies have shown that apple polyphenols have a significant inhibitory effect on GTase and have anti-caries effects. Apple Condensed Tannin (ACT) is the most effective inhibitor. Its inhibitory effect on GTase is 100 times higher than the epigallocatechin gallate (GECG) in green tea [6].

3.3 Eliminate odor

The fishy smell of fish is mainly caused by amines, and the main component is trimethylamine. In aquatic processing, adding apple polyphenols at 250 μg/mL when washing the fish section can reduce the volatile trimethylamine by about 70%. By adding apple polyphenols to dozens of salt-soaked water products, the fishy smell of all products has been suppressed.

The main component that produces bad breath is methyl mercaptan produced by sulfur-containing amino acids, and apple polyphenols can inhibit the production of methyl mercaptan. In the deodorization test, sulfur-containing methionine was used as a substrate, and human saliva containing oral bacteria was added to produce methyl mercaptan at 37°C. When 300μg/ml apple polyphenol was added to the matrix, the production of methyl mercaptan was reduced by more than 50%. When the addition amount reached 1200μg/mL, the production of methyl mercaptan was completely suppressed. In human experiments, volunteers were asked to chew gums containing different amounts of apple polyphenols. When chewing gum without apple polyphenols, the content of methyl mercaptan in the mouth immediately began to increase after chewing; when chewing gum containing 0.25% of apple polyphenols, methyl mercaptan began to increase after 30 minutes; if the content of apple polyphenols in the chewing gum reaches 0.5%, the methyl mercaptan starts to increase 60 minutes after chewing. Experiments show that apple polyphenols have a good effect on inhibiting bad breath.

3.4 Anti-cancer

Epidemiological studies have shown that diet can prevent about 32% of cancer incidence. Studies have shown that apple polyphenols can prevent and delay the occurrence of cancer to a certain extent. The triterpenoids in apple peel have strong anti-proliferative activity, which may be part of the reason for the anti-cancer activity of apples [7]. Compared with apple flesh, apple peel has stronger antioxidant and proliferative activity [8]. The main part of the peel provided by the apple is the biologically active substance of the apple. The main reason may be that the apple peel contains a large amount of quercetin glycosides, but the flavonoids are not in the pulp. Most triterpenoids isolated from apple peel have a strong inhibitory effect on HepG2 human liver cancer cells [9], MCF-7 human breast cancer cells and CaCo-2 human colon cancer cells [7].

3.5 Lowering blood pressure and blood sugar

Apple polyphenols can inhibit angiotensin transferase (ACE), prevent vasoconstriction and increase in blood pressure, and are effective substances for preventing hypertension. The catechins and condensed tannins in apple polyphenols all have the effect of inhibiting the activity of ACE, while the condensed tannins have strong activity, so they are known as "natural anti-hypertensive drugs". Diabetes is characterized by high blood sugar. One of the protective mechanisms for diabetic patients against hyperglycemia is to increase the elimination of urine sugar. Glucose is continuously filtered and reabsorbed in the kidneys. Polar sugars need a certain carrier for active or facilitating transport through lipid-rich cell membranes. There are two types of sugar carriers, one is Sodium-linked glucose transporters (SGLTs) and the other is Glucose transporters (GLUTs) [10]. The degradation products
of phloridin have the best inhibitory effect on microbial activity. Phloridin specifically and competitively inhibits the transport of glucose molecules by SGLTs. Phloridin can promote the secretion of glucose, reduce fasting and postprandial blood sugar levels, without the side effects of hypoglycemia [11]. Treating diabetic rats with phloridin can restore normal insulin sensitivity and eliminate or reduce insulin resistance caused by glycotoxicity [12]. In a randomized, placebo-controlled trial, researchers found that long-term use of apple polyphenols can improve hyperglycemia in subjects with high normal and cut-off values [13]. Clinical transformation experiments also show that apple polyphenols have a positive regulatory effect on vascular oxidative stress uricemia and vascular endothelial function in overweight individuals. By inhibiting the individual's xanthine oxidase, it has a significant effect on reducing fasting blood glucose and uric acid levels [14].

4. Application of apple polyphenols

4.1 Application in medicine

The four active ingredients of chlorogenic acid, catechin, phloridin, and quercetin contained in apple polyphenols all have varying degrees of hypoglycemic effect [15], as shown in Table 1. Clinical studies have shown that phloridin has a very good effect in the treatment of diabetes. Apple polyphenol phloretin capsules are currently on the market.

Table 1. Four main bio-active components and functions in apple polyphenols.

<table>
<thead>
<tr>
<th>Name</th>
<th>Chemical formula</th>
<th>Effect</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlorogenic acid</td>
<td><img src="image" alt="Chemical formula" /></td>
<td>Lowering blood lipids, antibacterial, anti-inflammatory, anti-viral, protecting liver and choleretics, etc.</td>
</tr>
<tr>
<td>Quercetin</td>
<td><img src="image" alt="Chemical formula" /></td>
<td>Dilate blood vessels, lower blood pressure, lower blood lipids, reduce capillary permeability and fragility, prevent coronary heart disease, etc.</td>
</tr>
<tr>
<td>Catechin</td>
<td><img src="image" alt="Chemical formula" /></td>
<td>Lower serum total cholesterol (TG), triglycerides (TC), low-density lipoprotein (LDL-c) and increase high-density lipoprotein (HDL-c), etc.</td>
</tr>
<tr>
<td>Phloridin</td>
<td><img src="image" alt="Chemical formula" /></td>
<td>Effectively inhibit microbial activity, lower blood sugar, etc.</td>
</tr>
</tbody>
</table>

4.2 Application in food

4.2.1 Health Food

Apple polyphenols have a variety of medical and health care functions, such as preventing high blood pressure, anti-aging, anti-tumor, preventing dental caries, weight loss, promoting development, enhancing memory, improving sleep and so on. Therefore, it can be used in the development of health
foods to produce safe, healthy foods and beverages with special functions. For example, it can be developed as a functional product that assists in inhibiting tumors, lowering blood pressure, delaying aging, functional food for weight loss, and functional food for improving growth and development, memory, and sleep. Apple polyphenol capsules are currently on the market. At present, FUJIYA Food Production Company in Japan has launched lollipops containing apple polyphenols. Because this product has the effect of protecting teeth, it has been welcomed by consumers.

4.2.2 Food preservative

Apple polyphenols are natural, safe, and low-cost preparations that extend shelf life. Spraying a low-concentration apple polyphenol solution on fresh fruits and vegetables can inhibit bacterial reproduction, maintain the original color of fruits and vegetables, achieve the purpose of preservation and preservation, and will not pollute fruits and vegetables. Apple polyphenols can be used for preservation of fresh-cut dragon fruit, which can significantly maintain the color, delay softening, reduce the loss of soluble sugar, titratable acid, and polyphenols, while maintaining its antioxidant properties and inhibiting the growth of microorganisms. So as to achieve the purpose of improving the safety of the product [16]. In the preservation of braised pork, the use of apple polyphenols in combination with ascorbic acid or niacin-amide can significantly improve the color stability of fresh pork packaged in oxygen permeable plastic wrap, and achieve a high and long-term color protection effect [17]. When investigating the effects of apple polyphenols on the quality of grass carp surimi during storage at 4 °C, the researchers found that apple polyphenols have strong antioxidant properties in vitro, inhibit the degradation of myofibril protein, and prolong the sensory organs of grass carp surimi. During the shelf life, it has a good protective effect on the quality of grass carp surimi during cold storage, and apple polyphenols also play a significant role in other functional properties of protein, which has broad prospects in the research field of antifreeze protective agents. After storing the herring meat treated with apple polyphenols at 4°C for 2 days, it was found that the color of the fish in the control group changed significantly and the meat became yellow. The fish added with apple polyphenols still maintained a high degree of freshness after 10 days [18].

4.3 Application in daily chemical products

Apple polyphenols can be used as active ingredients to be added to daily necessities such as hair restorers, toothpastes, and cosmetics. Apple polyphenols can stimulate the proliferation of keratinocytes and induce the production of keratin, inhibit protein kinase C (PKC) isoenzymes and regulate transforming growth factor β, etc., so it has proliferation and regeneration functions on hair follicle cells. It can be used to treat hair loss. In clinical trials, subjects who applied proanthocyanidin B-2 (from apple) not only increased their hair density significantly, but also thickened their hair. The average hair diameter of 78.9% of people increased by more than 40 microns [19]. Apple polyphenols can inhibit the effect of GTase and can be added to toothpaste to prevent the formation of tartar. Apple polyphenols have multiple functions such as anti-oxidation, anti-aging, anti-radiation, whitening and moisturizing, and have good astringency and adhesion. It can promote synthesis and protect collagen, inhibit elastase and improve skin elasticity, thereby avoiding or reducing wrinkles. In clinical trials, applying more than 2% extracts every day, after 28 days, can significantly improve the wrinkles and skin firmness in the eye contour area [20].

5. Conclusions

As a class of natural compounds with significant physiological activity, apple polyphenols have relatively high application value and play a decisive role in human health. Apple polyphenols have a certain effect on the color, aroma, taste and shape of products in food processing. The research mainly focuses on the improvement of product color and fresh-keeping material quality. Apple polyphenols have a wide range of physiological functions, focusing on their antioxidant properties, which are involved in lowering blood sugar, lowering blood lipids, preventing various cancers, and regulating immune levels. However, the current researches on the physiological and biochemical effects of apple
polyphenols functional foods are not in the description and explanation of the phenomenon, and there are few experiments on the effects of certain active monomers in apple polyphenols. Future research needs to focus on the relationship between monomer components and mixtures as well as research on functionality.

Apple is the top of the four major fruits in the world, and China is the world's largest producer and consumer of apples. Although China's apples are rich in resources, their comprehensive utilization is still lagging behind. In some harvest seasons, waste is very serious. Apple is the fruit crop with the widest planting area, the highest yield, and the greatest economic effect. The uniqueness and application potential of apple polyphenol resources still need to continue to be explored and in-depth researched, so as to develop products in medical, health care, cosmetics and other various fields. Products with practical application value in the field can better serve human health.

References


