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Research Progress on Traditional Medicine, Chemical Composition and Pharmacological Activity of Pine Pollen

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Abstract

In traditional Chinese medicine, "pine yellow" and "pine flower" are the pollen of Pinaceae, commonly Pinus Massoniana Lamb or Pinus tabulaeformis Carr. The pollen of Pinus Massoniana Lamb or Pinus tabulaeformis Carr is common. Pine flower is a unique species of medicinal and edible pollen in the Chinese pharmacopoeia, and is commonly found in ancient Chinese medical and herbal texts, such as "Derivatives of the Materia Medica", "Classical Materia Medica", and "Compendium of the Materia Medica". In this paper, we review the source, history of traditional medicine, and progress of research on pharmacological activity of Chinese pine pollen in order to provide reference for better research on its value.

Keywords: pine pollen, growth distribution, traditional medicine, chemical composition, pharmacological effects, research progress

1. Introduction

In traditional Chinese medicine, "pine yellow" and "pine flower" are the pollen of Pinaceae, commonly Pinus Massoniana Lamb or Pinus tabulaeformis Carr. The pollen of Pinus Massoniana Lamb or Pinus tabulaeformis Carr is common. Pine flower is a unique species of medicinal and edible pollen in the Chinese pharmacopeia, and it is commonly found in ancient Chinese medical and herbal texts, such as the "Derivatives of the Materia Medica", the "Classical Materia Medica", and the "Compendium of the Materia Medica". In this paper, we review the source, history of traditional medicine, and progress of research on the pharmacological activity of Chinese pine pollen to provide a reference for better research on its value.

Pine pollen has a long history of homologation in traditional Chinese medicine. In the first Chinese pharmacopoeia *Shennong Ben Cao Jing*, it is recorded that pine yellow is sweet, mild and non-toxic, and is used to treat cold and heat in the heart and abdomen, to facilitate urination, to eliminate blood stasis, and to lighten the body and improve vitality and prolong life (Deng Qian, Tong Shanshan, Ding Lixia, et al., 2012). In the Compendium of Materia Medica, it is written that pine flower, sweet, warm, non-toxic, can be used as medicine, which can moisten the heart and lung, warm and benefit the Qi, can be used to remove wind and stop bleeding, and can also be used to make wine. It is also recorded in *Ben Cao Feng Yuan* and *Ben Cao Zong Xin* (Dai Chengen, He Xiaoping, Zheng Fenfen, et al., 2019). With the improvement of modern extraction technology and modern medical research, the components of pine pollen are gradually separated and extracted, and pine pollen contains more than 200 kinds of sugars, lipids, proteins and amino acids, nucleic acids, peptides, vitamins, minerals and other nutrients required by the human body (He Xiaoyan, Sun Xueyuan & Yu Zhiyang, 2007). Pine pollen is known as "complete nutrition" or "micro nutrient bank". In terms of medicinal value, it has attracted the attention of researchers and scholars from all walks of life for its high medicinal value, minimal toxic side effects, and its versatility as a medicine.

This paper reviews the sources, growing regions and clinical medicinal values of pine pollen. In this paper, we review the sources, growth areas and clinical medicinal values of pine pollen to make a more comprehensive review and outlook on pine pollen, so that we can better study and apply this traditional Chinese medicine for the scholars.

2. The Source of Pine Pollen

Pine pollen is the pollen of the pine family, in China's pine trees have a wide variety of existing species, such as a wide distribution, among which oil pine, black pine, camphor pine, Yunnan pine and other species are more common.

Table 1. Growth and distribution of major tree species of pine pollen

Tree species	Distribution	Fl.	Maturity
Yaupon	Northeast, Central, Northwest and Southwest China	May	Early-mid October of the following year
Alpine Pine	Western alpine region	May	October of the following year
Sphagnum pine	Daxinganling, west of Hailar and southwest dune area of Heilongjiang Province	May ~ June	September to October of the following year
Cedar	Southern Tibet	October~ November	-
Five Needle Pine	Middle and lower reaches of Yangtze River	May	-
Money Pine	Middle and lower reaches of Yangtze River	April	October
Whitebark Pine	North and Central China	April ~ May	10~11
Huashan Pine	Central to southwestern China	April ~ May	September to October

	alpine region		of the following year
Lohan pine	South of Yangtze River	April ~ May	August~ September
Red Pine	Northeast Region	May	October~ November
Black Pine	Western Coastal Region	April ~ May	October of the following year
Horsetail Pine	Central and South China	April ~ May	October to December of the following year
Yunnan Pine	Southwest Region	April ~ May	October of the following year

3. Materia Medica and Canonical Records

Pine pollen in China has a thousand-year history of both medicinal and food use, and generations of medical literature, herbal pharmacopoeia, medical food and medicine, such as the pine pollen medicinal value and peculiar efficacy are very high evaluation (Ding Lingying, 2006; Chen YF, Xi Rong, Tang Shuang, et al., 2018).

Table 2. Application of pine pollen in traditional medicine

Traditional Medicine Ancient Books	Antiquarian Application Details
The Derivation of the Materia Medica, the Classical Materia Medica, the Compendium of Materia Medica, the Introduction to Medicine, and the Secret Jane Ji Yin	For postpartum hyperthermia, dizziness and chest fatigue. How to use: Pine Flower, Angelica Sinensis, Gypsum, Phellodendron, etc. are crushed into powder, add two coins of water, two coins of safflower, two twists of decoction into seven minutes, remove the residue and drink.
Puji Fang	Cure dizziness, dizziness and brain swelling are not applicable. How to use: Pine tree draws two liters of flowering heart, wraps it in gauze, soak it in wine for 5 days and drink it on an empty stomach.
The Infant Medical Mirror	Treating bleeding from traumatic injury. Usage: Pine pollen is applied directly to wounds externally.
The Authentic Fang and Vein Book	Cure prolonged dysentery, lingering and unclean. How to use: Take three coins of pine flower powder mixed with rice soup.
The Handbook of Wei Gu Xi, New Compilation of Experimental Prescriptions	(1) For the treatment of headache and dizziness, stuffy throat, watery diarrhea, fatigue and atrophy. How to use: 1 tael of pine flower, 5 taels of Chen Pi and Chuan Huang Lian plus 2 taels of licorice, fried and ground into powder, mix evenly, take 2 taels each morning and evening. (2) For vomiting blood, congestion in the chest, vomiting blood stasis, etc. Pine Flower, Radix Rehmanniae Praeparata, Peach kernel, Rhubarb and Citrus aurantium each one penny taken together. (3) How to use: Pine Flower, Ginseng, Atractylodes Macrocephala, Radix Paeoniae Alba, Rhizoma Macrocephala, Radix et Rhizoma Dioscoreae, 1 coin each. (4) Pine Flower, Radix Rehmanniae Praeparata, Radix Scutellariae, Radix Platycodon, Radix Glycyrrhiza, Radix Ginseng, Radix Rehmanniae Praeparata, Radix Mulberry Peel, one penny each, and one or two roots taken together. (5) For coughing up blood out of the lungs. It is used to treat blood in the stool, bleeding from the large intestine and damp heat in the internal organs. Usage: Pine

	Flower, Radix et Rhizoma, Phellodendron (fried), Sophora japonica, Elm, Aconite, Radix Angelicae Sinensis, Radix et Rhizoma each 2 yuan, Phellodendron, Paeonia lactiflora, Atractylodes macrocephala, Gardenia jasminoides, Rhizoma Chuanxiong each 1 yuan.
The "Good Prescriptions of Wonderful Effect", "The Standard of Treatment", "The Complete Book of Ancient and Modern Medicine"	Cure heart meridian heat, tongue blood out. How to use: Stir-fried pine yellow and cypress leaves with glutinous rice, pounded and sieved, each serving of medicine for three coins, add one calanders of water, decoction to seven minutes, remove the dregs, warm.
The Drinking and Dining Guide	Tonic for the middle, strengthen the muscles and bones. How to use: boil mutton, herb nuts, beans, together into soup, strain, cooked sheep's chest a, pine yellow juice two, ginger juice half, together with fried, onion, salt, vinegar, coriander leaves, mix well, eat.
The Precious Family Recipe of Mr. Ding Ganren	Treatment of bedsores. How to use: Mung bean powder one or two, cypress two or two, pine flower powder, talcum powder five money each, research into a fine powder, apply sesame oil to the affected area.
Life-Saving Collection, External Treatment of Longevity Formula	Cure pox sores scratching. Usage: Pine pollen is applied externally.
The Evidence and Treatment of Female Medicine in Zhulin	Cure hunchback caused by early sitting in childhood. How to use: One tael each of Pine Pollen, Citrus Aurantium (bran fried), Fructus Anemarrhenae, and Dulcimer, five coins each of Ephedra Officinalis, Rhubarb, Qian Hu, and Gui Xin.
The Complete Ulcer Doctor	To treat wounded sores. The first thing to do is to wash and dry the sore with cold rice slop water, then apply the medicine to the surface and bandage it well, changing it daily.
Dictionary of Chinese Medicine	For the treatment of newborn red buttocks, diaperitis and summer sweat rash in children. How to use: Sprinkle pine pollen on the affected area. Or carry 200g of Pan Gui, 30g of Centella asiatica, decoct in water and wash the affected area, then sprinkle pine pollen.
New Compilation of Experimental Formulae, External Treatment for Longevity Formulae	1) To treat cerebral leakage. How to use: Pine pollen, sniff into the nose. 2) How to use: Pine pollen applied or pine pollen two money, Huang Dan one money, research even grits.

4. Pharmacological Activity of Pine Pollen

4.1 Enhance the Immunity of the Body

Liu, M. et al. (2014) observed the effects of horsetail pine pollen polysaccharide (PPM60-D) and its sulfate ester (SPPM60-D) on mouse spleen B lymphocytes, intracellular free calcium ion concentration ([Ca2+]i) and antibodies, and introduced that SPPM60-D could bind to B lymphocyte Toll-like receptor 4 (TLR4), stimulate TLR4-PI3K-PLC-. The IP3R signaling pathway opened calcium channels activated by calcium pool release (CRAC), thus elevating [Ca2+]i to activate B lymphocytes, which in turn increased B lymphocyte proliferation and antibody production in vitro. Thus, it can be shown that pine pollen enhances human immunity and promotes the development of immune organs in the spleen, bone marrow and lymph nodes.

4.2 Inhibition of Prostate Hyperplasia

Pine pollen sterol has a good inhibitory effect on prostate enlargement, and its mechanism inhibits prostate enlargement by inhibiting 5α a reductase to reduce the level of dihydrotestosterone in rats. Baoan Zhu (2016) after treating mice with different doses of pine

pollen sterols, the prostate tissue and testicular tissue were taken and weighed, and the prostate tissue was stained with HE in paraffin sections to observe the histomorphology, and the levels of prostate-specific antigen, testosterone and dihydrotestosterone in the serum of rats were measured by ELISA, and the 5α- reductase activity was determined by high performance liquid chromatography. It was shown that the sterols in pine pollen can inhibit dihydrotestosterone, thus regulating bladder and urethral smooth muscle and inhibiting prostate hyperplasia (Huang Aizhao, Li Yicong, Liang Chaofeng, et al., 2020).

4.3 Protecting the Liver

The mice with acute liver injury were treated with total flavonoids from Pine Pollen, and the blank and model groups were treated with equal doses of saline by gavage after 10 d of continuous intervention. The liver pathological changes, peripheral blood glutamic aminotransferase (ALT) and glutamic oxalacetic aminotransferase (AST) activities, and changes of PI3K-AKT signaling pathway signature protein levels were compared among the three groups. The experimental results suggested that the liver in the model group showed obvious pathological changes, and the liver cells in the experimental group showed less vacuolation and inflammatory infiltration than those in the model group, and the liver cells in the experimental group showed slight necrosis. (Fan Berlin, 2005) In the model group, peripheral blood ALT, AST and Bax concentrations were significantly up-regulated and Bcl-2 expression was decreased. In contrast, in the experimental group, the above indexes were significantly improved, while PI3K and p-AKT levels in liver tissues of the model group were significantly decreased. The improvement suggests that total flavonoids of pine pollen have a protective effect on chemical liver injury, and its mechanism of action may be related to the mediation of PI3K-AKT signaling pathway.

4.4 Regulation of Blood Glucose and Lipid Metabolism

Fan Berlin et al. (2005) established a hyperlipidemic phenotype after feeding rats with a high-fat diet, and the rats were randomly divided into five groups: a negative control group, a high-fat model group and three dose groups of 1.0, 2.0 and 3.0 g/kg, and pine pollen was given prophylactically, and the TC, TG and HDL-C contents of the rats were measured before and after administration. The results of the study showed that TC decreased in both the middle and high dose groups, while TG decreased and HDL-C increased in the high dose group. This study suggests that pine pollen can regulate blood lipids in mice. Pan Xiaoling et al (2006) also randomly divided mice into negative control group and three dose groups of low, medium and high doses and gave pine pollen respectively. The results showed that the high dose group of pine pollen could significantly reduce the blood glucose level of tetraoxypyrimidine diabetic mice when compared with the negative control group in the experimental group. The above two experiments show that pine pollen has regulatory functions on blood lipids and blood glucose.

4.5 Improve Gastrointestinal Function

By studying the effects of chronic stress on the intestinal microecology of rats and the health effects of pine pollen, as well as the experiments of different doses of pine pollen affecting the movement of small intestine and the experiments of regular defecation, it is concluded that pine pollen can promote the peristalsis of gastrointestinal tract, improve the symptoms of constipation and have the function of laxative and laxative. (Peng L, Zhao P, Zhang JH, et al., 2016; Wu Jingjing, Huang Peihua, Zhu Liang, et al., 2010). From the above study, it can be shown that pine pollen can regulate the dysfunction of gastrointestinal tract, accelerate the peristalsis of gastrointestinal tract, improve constipation and laxative effect.

4.6 Anti-Fatigue Effect

The mice were given horsetail pine pollen total flavonoids and distilled water, and the indexes of swimming, pole climbing time, hypoxia tolerance time and blood lactate, urea nitrogen and liver glycogen after swimming were measured, indicating that horsetail pine pollen total flavonoids can prolong the swimming and pole climbing time of mice, and can reduce the serum BUN concentration after exercise, improve the reserve of liver glycogen, enhance the ability to clear lactate, and have anti-fatigue effects (Luo Yanhong, 2013).

4.7 Anti-Tumor

Some scholars used MTT method to detect the proliferation inhibitory effect of pine pollen on HepG2 cells at different concentrations and different action times, while flow cytometry was

used to detect apoptosis and LISA was used to detect the expression of Bcl-2 and PTEN protein. Or the contents of PIVKA-II, AFP and VEGF in the supernatant of cultured cells in the pine pollen treated group and HepG2 control group were detected by ELISA, and it was concluded that the contents of PIVKA-II and AFP were decreased and the contents of VEGF were extremely significantly decreased in the pine pollen treated group compared with the HepG2 control group. The above experimental results showed that pine pollen had significant proliferation inhibitory and pro-apoptotic effects on HepG2 cells (Yu, Wenjun, 2016; Dai Y. H., Zhang L. M., Yang S. H., et al., 2017).

4.8 Antioxidant Effect

By observing the survival rate of HepG2 cells when different mass concentrations of pine pollen and H2O2 were applied together, it was concluded that pine pollen has a protective effect against H2O2-induced oxidative damage in HepG2 cells. The correlation showed that pine pollen contains a variety of antioxidant vitamins as well as a variety of trace elements, and these components can scavenge free radicals in cells and inhibit the rate of lipid and protein peroxidation reactions in the body, thus achieving antioxidant and anti-aging effects (Hei Yurong, Peng Xiujuan & Yang XJ., 2019).

4.9 Other Pharmacological Activities

In addition, studies have shown that pine pollen can also reduce the nitric oxide content in the renal tissue of acute renal failure and has a therapeutic effect on rats with acute renal failure (Zhu X.N. & Han L., 2015). The study of pine pollen extract on weighted swimming test, conventional stress hypoxia tolerance test and liver weight coefficient in mice also revealed that pine pollen extract has various effects such as analgesic, anti-inflammatory, anti-hypoxic and anti-fatigue (Shi-Bo Zhao, Suo-Yi Huang, Yu-Xin Wei, et al., 2020). Pine pollen contains a variety of vitamins and inorganic minerals, which can effectively prevent diseases caused by the lack of trace elements, such as rickets, etc. Pine pollen is rich in essential amino acids, which can benefit children's body and brain development.

5. Comparison of the Active Ingredient Content of Different Varieties of Pine Pollen in Common Use

Zhang Limei et al. (2008) employed various methods such as Soxhlet extraction, atomic absorption spectrophotometry and atomic fluorescence photometry, Hygroscopic ketone colorimetric method, and national standards for vitamin detection methods to determine and compare amino acids, minerals, fatty acids, and vitamins of horsetail pine pollen, Yunnan pine pollen, black pine pollen, and oleaginous pine pollen, respectively, and summarized in the following table:

Table 3. Nutritional composition analysis of pine pollen

Nutrients	Nutrients	Nutrient content comparison
Amino acids	Aspartic acid, Threonine, Proline, Tryptophan, Botrytisine, Serine, Glutamic acid, Glycine, Alanine, Fethionine, Methionine, Tyrosine, Phenylalanine, Lysine, Histidine, Arginine, Isoleucine, Leucine	Total amino acids: Pinus oleifera > Pinus nigra > Pinus equisetum > Pinus yunnanensis Total essential amino acids: Pinus radiata > Pinus nigra > Pinus equisetum > Pinus yunnanensis
Minerals	Calcium, potassium, magnesium, sodium, iron, copper, manganese, selenium, phosphorus	Pinus sylvestris > Pinus sylvestris > Pinus yunnanensis > Pinus nigra
Fatty acids	Myristic acid, palmitoleic acid, palmitoleic acid, stearic acid, oleic acid, linoleic acid, linolenic acid, arachidonic acid, eicosapentaenoic acid, eicosadioleic acid, lignocarboxylic acid	Saturated fatty acids: Pinus oleifera > Pinus sabdariffa > Pinus nigra > Pinus yunnanensis Unsaturated fatty acids: Pinus oleifera > Pinus sabdariffa > Pinus nigra > Pinus yunnanensis
Vitamins	Pantothenic acid, niacin, folic acid, vitamin A, E, C, vitamin B1, B2, B6, B12, etc.	Yaupon pine > black pine = Yunnan pine > horsetail pine

According to the above table, we can initially know that the content of active ingredients of different species of pine pollen varies. In the comparison of the four pollen species, we can assume that oleander has the highest content of amino acids, minerals, fatty acids, and vitamins.

6. Outlook

Pine pollen, as Chinese medicinal and food pollen, is rich in nutrients and active ingredients, and has a comprehensive effect on human physiological regulation, growth and development, and body repair, etc. A number of studies have shown that pine pollen can regulate immunity, anti-fatigue, antioxidant, anti-inflammatory, and also improve gastrointestinal function, regulate constipation, blood lipids and protect the liver, and in Chinese traditional Chinese medicine, pine pollen is considered to enter the heart meridian, lung meridian. It has the effect of "moistening the lungs and protecting the heart". Because of its many medicinal values, pine pollen should be promising for application in the food, drug and health care industries, either alone or as a formula.

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