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A Review on Plant Metabolites and Ayurvedic Medicine

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ABSTRACT

From ancient times, Ayurveda has its own relevance and is known for its mystical effect. In Hindu mythology, during Satyug when Lord Hanuman went all the way to Himalayas to find a magical herb which had the power to cure any malady known as Sanjeevani booti (Selaginella bryopteris) to save the life of Lakshmana. Ayurveda marks its presence even today and has achieved different level of excellence. As it is said that Sanjeevani glowed in dark, so did the Ayurveda or science of life. It is an ancient system of natural and holistic medicine which broadly utilizes the secondary metabolites released from certain plants. Like in Ashwagandha various secondary metabolites are present like Withaferin A which has anti-cancer properties. It also has great effect on anxiety, diabetes, high cholesterol, arthritis, tumours, tuberculosis and a number of other diseases. Formation of memories can also be improved using Ashwagandha, so it is used to treat Alzheimer's disease. In this presentation we will discuss the process for obtaining the secondary metabolites from various medicinally important plants. For obtaining these vital secondary metabolites a suitable extraction technique is also mandatory. Extraction is an important step for the isolation of phytochemicals from plant materials. A specific technique and optimization of various parameters can be utilized for the isolation of a peculiar type of secondary metabolites. In this paper we have discussed some of the main ingredients found in some of the most powerful medicinal plants which includes Withaferin A extracted from Withania somnifera, digoxin from Digitalis purpurea and Morphine from Papaver somniferum. We have also discussed some of the most suitable processes which are used for their extraction depending on their properties. Like in Withaferin A, we used solvent extraction and cell suspension culture for its isolation. We have also discussed their role in the treatment of present diseases. A newer approach is also introduced of derivatization of active ingredient which leads to its enhancing effect which can be highly useful to produce or synthesize more effective drugs.

Keywords: *Primary Metabolite, Secondary Metabolite, Digoxin, Morphine, Withaferin A, Ayurveda.*

1. INTRODUCTION

The most well renowned work –Atharva Veda, which was developed by the creation of the great rishis or the seers of ancient India, reveals that Ayurveda has primarily originated from India ages back.

The Sanskrit word Ayurveda is derived from the root words Ayuh meaning "life" or "longevity" and Veda meaning "science" or "sacred knowledge". Therefore Ayurveda translates as, "the sacred knowledge of life". It works to maintain the health in the healthy, to prevent disease in order to promote quality of life and to heal the sick. Ayurveda is the most ancient science of healing which enhances longevity. It has influenced many of the older tradition methods of healing including Tibetan, Chinese and Greek medicine. Hence, Ayurveda is considered by many as the "Mother of Healing". The tradition of Ayurveda was dimmed due to the natural and human calamites and also by the invasion of foreign cultures into India. The sacred texts were either destroyed or stolen. However, divine plants that sustain long life and good health have now been rediscovered. Today there is a revival of traditions and ancient cultures inherent to Ayurveda, which is a true gift of the ancient civilization to the modern world.

Exploitation of nature particularly plants has been carried out since ancient times in search of new drugs and medicines. As a result, a large number of medicinal plants have been emerged for the treatment of various diseases. Almost 95% of the drug prescriptions

in India are plant based. But rather than using the whole plant, studies are carried out to use only the secondary metabolites. Researchers have proved that primary and secondary metabolites are responsible for medicinal activity of plant. Vaguely, Ayurveda uses different plant extract or bioactive components for producing various drugs and medicines, but new researches and development in this field aims at utilizing these plant metabolites from the plant directly which ameliorate their effect.

Plant Metabolites

For growth and development and also to perform different special functions, plants synthesize various organic compounds which are known as Metabolites. These are prepared using various enzyme-mediated chemical reaction also known as metabolic pathways. These plants metabolites are broadly categorized into two types depending on the utilization and role played by them in the plant body.

a. Primary Metabolites: These metabolites have the functions that are essentially required for the growth and development and are therefore synthesized in all plants. It includes different types of carbohydrates, lipids, proteins and nucleic acids. Since they are omnipresent in all plants, they act as fundamental component or sometimes product of essential metabolic pathways such as glycolysis, the Calvin cycle and the Krebs cycle. So they also enable a plant to synthesize, assimilate and degrade organic compounds.

b. Secondary Metabolites: These are the metabolites that are used for the extremely diverse functions apart from growth and development. They assist plants for performing important functions such as competition, protection and species interaction. These secondary metabolites are highly specific in nature and are used by humans in medicines, flavorings and recreation drugs. Unlike primary metabolites, the absence of secondary metabolites will not lead to the death of the plant.

Broadly, secondary metabolites are classified into three main groups:

- Terpenes
- Alkaloids
- Phenolic compounds

Terpenes: Terpenes are a disparate group of more than 30,000 lipid soluble compounds. As a large group, they exhibit a wide range of toxicity from deadly to entirely edible and this embraces its antimicrobial properties and attracts symbionts for the purpose of pollination, secondary protective roles and seed dispersal. Terpenes are used as a relaxant as it soothes spasm and crams.

They show cytotoxic activities upon a wide range of organisms ranging from bacteria and fungi to higher insects and vertebrates and hence are widely employed in herbal medicines. The most commonly used terpene, Bilobalide and Ginkgolides A, B, C and J is extracted from leaves of Ginkgo biloba which is widely used to treat Dementia. Terpenes like Iridoid glucosides have been used to treat infections, rheumatism and inflammations which are extracted from Harpago phytumprocumbens and Plantago sp.

Alkaloids: Alkaloids are wide group of plant constituent containing a nitrogen bearing molecule. Most of the alkaloids are extremely toxic, especially to mammals and act as effective nerve poisons, membrane transport inhibitors or enzyme inhibitors as long as they are administered in carefully regulated doses. Many potentially toxic plant derived alkaloids can be used for medicinal purposes.

Alkaloids such as morphine and codeine from the Opium poppy and cocaine from coca plant are used for pharmaceutical purposes as they act on nervous system and hence are used as painkillers. Another alkaloid vincristine and vinblastine from Periwinkle plant act as inhibitors of cell division and are used as effective drug to treat cancers of the blood and lymphatic systems.

Phenolic compounds: Most of the phenolic compounds are synthesized from the amino acid phenylalanine. Phenolic compound can be marked by the presence of one or more aromatic rings containing functional hydroxyl group. Some simple phenolic compounds such as salicylic acid can be of utmost importance in defense against fungal pathogens.

During a fungal attack the concentration of salicylic acid is increased in leaves of certain plants that enable the plant to mount a complex defense response. Some phenolic compounds like isoflavones have strong antimicrobial activity. Interestingly, aspirin, a derivative of salicylic acid is widely used in humans to reduce inflammation, pain and fever.

Table.1. Examples of some secondary metabolites, their use and bioactivity

Secondary Metabolites	Class	Plant species	Properties/Applications
Tetrahydrocan, Nabinol	Terpene	*Cannabis sativa*	Analgesic
Digitoxin	Terpene	*Digitalis lanata*	Heart Sufficiency
Ouabain	Terpene	*Strophantus gratus*	Heart Sufficiency
Aconitine	Alkaloid	*Aconitum napellus*	Analgesic
Colchicine	Alkaloid	*Colchicum autumnale*	Gout treatment
Umbelliferone	Phenolic	*Daucus carota*	Cross linked DNA, block cell division
Cocaine	Alkaloid	*Erythroxylum coca*	Analgesic, stimulant
Podophyllin urushiol	Phenolic	*Mayapple poison*	Cathartic
Anthocyanin	Phenolic	*Almost all plants*	Estrogenic

Major ingredients extracted from medicinal plants

WITHAFERIN A

Withaferin A is a steroidal lactone which is ono of the first withanaloide to be extracted from Winter Cherry (*Withania somnifera*), also known as Ashwagandha. It is now also derived from *Acnistus arborescens* and other member of Solanaceae family. It is used as an Ayurvedic medicine from a very long time. Naturally it is useful in plants and plays an important role in defense responses to pathogens. It also proves useful in adverse conditions like drought and low temperature.

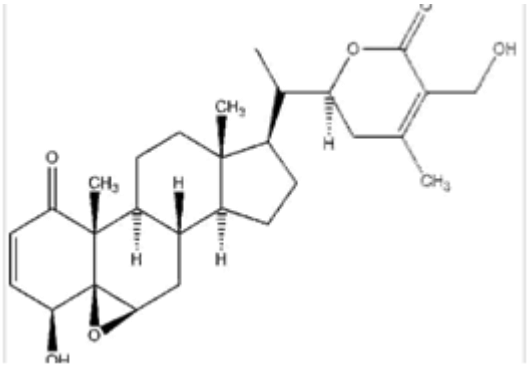

Figure.1.Chemical Structure of Withaferin A

They are highly reactive due to the presence of the ketone comprising an unsaturated A ring. This a ring acts as the main nucleophilic target site. Withaferin A is an important bioactive component in plants and has some peculiar properties due to which it is highly used in medicinal plants. These properties include:

- Withaferin A shows anti-inflammatory properties. It suppresses the manufacturing of many pro-inflammatory molecules. So it is highly useful in the treatment of arthritis and rheumatism.
- Anti-tumor and radio sensetising effect of Withaferin A has been discovered recently. It was first tested on human prostate cancer cell line, PC-3. It hinders tumour growth by ATP independent inhibition of heat shock protein 90(HSP 90).
- Many experiments have revealed immunosuppressive effects on human B and T lymphocytes and on mouse thymocytes.
- Withaferin A deactivates invasion capacity of cancer cell and cell motility. Various researches carried out in the field of cancer to decrease the growth and development of cancer cells.
- Withaferin A has a unique property to inhibit the formation new blood vessel from preexisting vessels which is also known as angiogenesis.
- This activity is a result of inhibition of chymotrypsin and activation of apoptosis is due to the inhibition of protein kinase C
- It shows anti-stress and anti-oxidant effect due to which Withaferin A is used against many diseases which are associated with free radical oxidative damage.
- Withaferin A is used as a tonic to prevent diseases and increase energy and thereby affecting the immune system.
- It has hemopoetic effect. Stimulation of stem cell proliferation and use in cancer chemotherapy are some of the major activities of Withaferin A.
- It affects the nervous system, endocrine system and cardio pulmonary system.

Extraction of Withaferin A:

METHOD I

According to the US Patent NO. 7,108,870 Withaferin A can be isolated from a plant material using a process comprising of the following steps.

- The plant material is first extracted in an extraction solvent having aqueous alcohol.
- The next step involves removal of fat also known as defatting.
- A withanolide preparation is obtained by subdividing with n-hexane followed by chromatographic separation.
- Withanolide aglycones have been portioned out during the defatting and are kept into chloroform followed by evaporation of chloroform to procure a chloroform extract.
- The chloroform extract is dissolved as done in step 4 in methanol.
- Further chromatographic separation is used to operate Withaferin A.

METHOD II

Cell culture:

1) Callus initiation and maintenance: The seeds of Withania somnifera are washed and allowed to germinate aseptically on sucrose agar medium foe three weeks. The hypocotyl portions from these 3 week old seedlings and are transferred on to MS medium aseptically. Finally, the callus induction is observed and sub cultured onto the half strength B5 medium at an interval of four weeks.

2) Cell suspension culture Initiation and maintenance: The callus (10% w/v) are transferred onto half strength B5 medium to initiate suspension culture and incubated. The culture is then filtered to get uniform suspension and 5% inoculums are taken from this filtered culture and are maintained at an interval of a week.

3) Induction of product synthesis: Filtered cell culture after washing with medium are transferred to a B5 medium having 1/4 of its concentration in 6 well plates and incubated for two weeks.

4) Elicitor Treatment: Elicitor solution is prepared by dissolving it in a suitable solvent and is added to the culture before incubation.

5) Extraction of Withaferin A: After obtaining cell fraction and cell free filtrate, cells are frozen and thawed and then with three volumes of di-chloro methane, they are extracted three times. This combination of di-chloro method and cell fraction after evaporation is dissolved in methanol and further saved for analysis.

6) Analysis: Analysis can be done either by TLC or HPLC technique.

MORPHINE

Morphine is the most ample anodyne opiate found in opium and is having great potential to relieve pain.

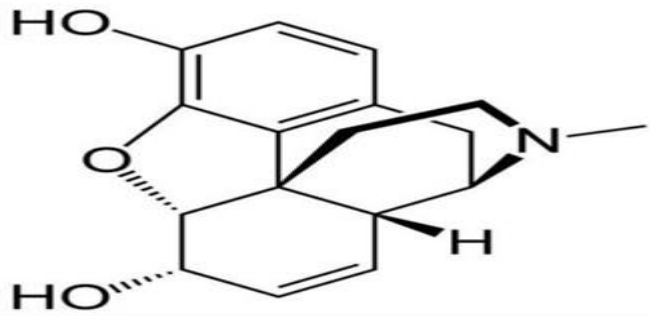

Figure2. Structure of Morphine

As a coin has two faces the drug has merits as well as demerits. Morphine is clinically used as a pain reliever but also has recreational purposes among drug users. And sometimes is known as a drug of abuse which has potential to down-regulate immune responsiveness. Albeit the exact mode of action how morphine induces immunosuppression has yet to be discovered. The seedpod extract or opium found in Papaver somniferum (poppy plant) is the most potent and widely used source of morphine. It is the first activated ingredient isolated from plant. The use of morphine came into existence 2000 years back but was first chemically isolated in 1806. The mode of action of morphine is through lymphocyte opiate receptors. Broadly there are two possible mode of action of morphine. Morphine act in both the central and peripheral nervous system. In the CNS, morphine has effects in many areas, comprehending the spinal cord. Within the peripheral nervous system, it acts in both the myenteric plexus and sub mucous plexus, present in the wall of the gut and therefore has constipating effect. In the peripheral tissues such as joints, morphine has anti-inflammatory effect.

Morphine has a wide area of application in the field:

As a pain reliever- It relieves the pain caused by myocardial infarction (heart attack), rheumatoid arthritis, cancer, sickle cell crisis, kidney stones, and labor pain or during any injury. It do not improve the overall outcome, in turn reduces the pain.

To treat shortness of breath- It treats the shortness of breath due to cancerous as well as non-cancerous causes. Regular, low dose, sustain release morphine decreases breathlessness safely, and maintain the benefit over time.

To treat opioid use disorder- Morphine is also available as slow release formulation for opiate substitution therapy (OST), for addicts who cannot survive on methadone or buprenorphine.

Extraction of Morphine

The extraction of morphine can be carried out from opium poppy by stirring, heating or preferably refluxing. The basic alcohol has a property to dissolve the morphine out of opium. This property of alcohol is exploited to extract morphine. This property of alcohol proves best at pH 9. Methanol, ethanol, isopropanol and mixtures of alcohols are preferred for solvent extraction of morphine. For extracting alkaloids from opium methanol proves to be an excellent solvent. The extraction is most efficient at a particular pH, which can be achieved during the separation by adding alkali or acid. More than 90% of morphine is extracted after refluxing in a basic solution of alcohol for about 1-2 hours. Hence, this process proves to be most efficient as well as takes less time.

Capillary zone electrophoresis (CZE) is a process used for the qualitative and quantitative determination of major alkaloids (morphine) in gum opium.

Major alkaloids are quantitatively extracted with 2.5% w/v aqueous acetic acid by using three extractions.

A 7:3 mixture of methanol and sodium acetate (100mM, ph. 3.1) at a potential of 15kV, with UV detection at 224nm is used to separate morphine by CZE.

Spiking of refined reference alkaloid standards in the opium extract is used for peak identification. The calibration plot is based on linear regression analysis. In the standard graph, the relative standard deviation (RSD) for migration time and peak area is in the range of 1.03-3.56% and 0.34-0.69%.

The detection limit of morphine dissolved in methanol is found to be 850 ng/mL.

DIGOXIN

Digoxin is a portent cardiac glycoside that is derived from Foxglove plant (Digitalis purpurea). Digitalis was earlier used to treat Dropsy which is an old term for Edema. Now the subsequent investigation has found that digitalis is most useful for edema that was caused by a weakened heart.

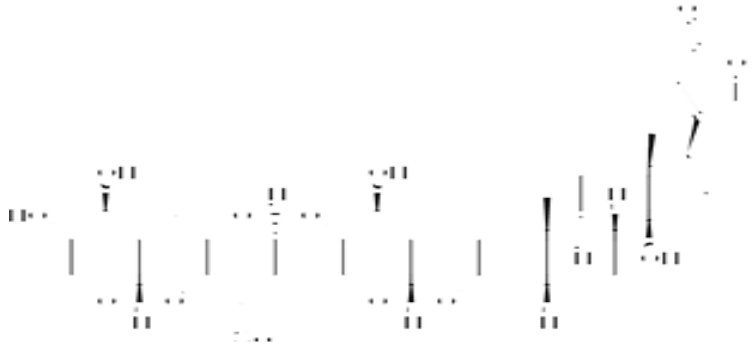

Figure.3.Chemical Structure of Digoxin

Digoxin is widely used in cardiac insufficiency and it causes positive isotropic, positive bathmotrophic, negative chronotropic and aromotropic effect. The most common indications for digoxin are atrial fibrillation and atrial flutter with rapid ventricular response. Digoxin is also used as a foeticidal agent or amniotically during abortion in the late second trimester and third trimester of pregnancy as it typically causes fetal demise.

Digoxins are inhibitors of cellular Na+ K+ ATPase. By inhibiting Sodium potassium ATPase, these cardiac glycosides cause intracellular sodium concentration to increase.

Also cardiac myosides and many other heart cells have a sodium calcium exchanger that is essential for maintaining sodium and calcium homeostasis. So inhibition of sodium potassium ATPase then leads to an accumulation of intracellular calcium via sodium calcium exchange system. In the heart increased intracellular calcium causes more calcium to be released there by making more calcium available to bind to Troponin-C which increases contractility. Inhibition of sodium potassium ATPase in vascular smooth muscles causes depolarization which causes smooth muscle contraction and vasoconstriction.

There are also some adverse effects of using digoxin which are concentration dependent. These are common in patient with low potassium level, since digoxin normally competes with potassium ions for the same binding site on sodium potassium ATPase pump. Common adverse effects include loss of appetite, nausea, vomiting, and diarrhea; blur vision, drowsiness and insomnia

Extraction of Digoxin

For the extraction of digoxin from Digitalis purpurea various methods can be employed. One of the method used is solvent extractions .In solvent extraction the plant parts (tubers, roots, stems, leaves etc.) are dried immediately in an artificial environment at low temperature (50-60°C) or dried preferably in shade so as to lower the initial large moisture content to maintain its prolonged storage life.

The dried plant materials are pulverized by mechanical grinders and the oil is removed by solvent extraction. The defatted material in the mixture is then extracted in a soxhlet apparatus or by soaking in water and alcohol mixture (95% v/v). The non-glucosidal impurities like vegetative residues which get extracted along with glycoside digoxin are removed by precipitating them with lead acetate solution. It is now passed through hydrogen supplied gas to remove excess of lead acetate. Lead gets precipitated as lead supplied, which is filtered out.

The filtrate contains the digoxin. The glycoside digoxin can be obtained by removal of the solvent under reduced pressure or any other suitable procedure. Further purification of the isolated digoxin is done by column chromatography.

Table2. Role of Plant Metabolites in present diseases

Metabolites	Disease	Mode of Action
Glucosinolates	Cancer	It has anti oxidative property and hence decreases the oxidative stress and induces cell apoptosis.
Saponins	Heart disease	Removes the cholesterol present in the cell or arterial membrane by binding to it. Since high LDL cholesterol intercalates cell membrane and decreases their flexibility.

Morphines	Pain and cough suppressant	Acts on the Central Nervous system and blocks pain signaling to the brain
Caffeine	Diabetes and heart disease	It contains polyphenol which fight against the disease.

Production of more effective drugs

Due to the presence of an active ingredient a plant is termed as a medicinal plant which may play a vital role in curing diseases. Ayurveda uses the same principle hence the crude extract of plant is generally used for medicinal purposes. Now -a-days scientists are trying to determine mode of action of these active ingredients which are then said to be a potent drug.

When the mode of action is known, the metabolite can be further modified to synthesize more effective drug with enhanced effects. For example Azido thymidine (AZT) is an analog of thymidine having an azido (N3) group in the 3' position. AZT is used to prevent and treat HIV/AIDS. During DNA replication of HIV the reverse transcriptase is unable to bind the preceding nucleotide. The action of reverse transcriptase is inhibited due to the presence of azido group at 3' position in turn terminating nucleic acid chain.

Another example of how an active ingredient from a medicinal plant can produce more effective results is conversion of morphine to heroine by the process of acetylation using anhydride. Morphine extracted from opium plants is heated under reflux with ethanoic acid and concentrated sulphuric acid to synthesize heroine.

Heroine under the generic name diamorphine is used in severe physical trauma, post-surgical pains, and chronic pains including end stage cancer and other terminal illness. Diamorphine is more advantageous than morphine as it is more fat soluble and therefore low dose is required for the same effect in chronic pains.

After the modification has been done, the next step for the drug to be commercialized is to check for its efficacy. It can either be done using cell culture or plant tissue culture, and further clinical trials can also be done.

2. CONCLUSION

There are about 50000 plant species present on this earth, out of which 4444 of them are widely exploited for medicinal purposes. There is large number of plant species still left unexplored, which can be of great use to produce a wide range of drugs and medicines. The secondary metabolites from plant are exploited to a large extend for these purpose from a very long time. Secondary metabolites such as Withaferin A, digoxin and morphine have a prominent role in treatment of various diseases ranging from most common diseases like flu, cough and cold to some of the most deadly diseases like cancer and Alzheimer's disease. A lot of research has been carried out in the field of Ayurvedic medicine to use these active ingredients more effectively by knowing their mode of action and converting them to more potent drugs.

In this view, preserving our natural resources is crucial as new plant species and metabolites have yet to be discovered along with their role to escalate human health and development. As with the changing environment around us due to pollution, global warming and many other hazardous effects, the nature and intensity of the diseases is also increasing, therefore its need of the hour to protect these precious gifts of nature.

3. REFERENCES

[1] Charles L. Mehltretter, Francis B. Weakley, Inventors; Charles L. Mehltretter, Francis B. Weakley, assignee. 1955 .Solvent extraction of opium alkaloids. United States Patent 2,715,627
[2] Michael Wink (2015), Modes of action of herbal medicine and plant secondary metabolite. Medicines.2015, 2, 251-286
[3] JI Yubin , Yu Miao, Wang Bing, Zhang Yao, The extraction, separation and purification of alkaloids in natural medicine. Journal of Chemical and Scientific Research, 2014, 6,338-345
[4] In-Hyoung Yang, Lee-Han Kim, Ji-Ae Shin, Sung-Dae Cho, Chemotherapeutic Effect of Withaferin A in Human in Human Oral Cancer Cells. Journal of cancer therapy.2015, 6,735-742
[5] Cardiac Glycosides: Cardiovascular Pharmacology Concepts. c 2005-2015. Richard E. Klabunde [March 18, 2017]. http://www.cvpharmacology.com/cardiosti mulatory/digitalis
[6] Lakshmi- Chandra Mishra, Besty B. Singh, Simon Dagenais, Scientific Basis for the Therapeutic Use of Withania somnifera (Ashwagandha): A Review. Alternative Medicine Review.2000, 5,334-346
[7] Pallavi Thakur. Feb 12, 2015. Sanjeevani Booti: Is the divine herb from Ramayana for real? [March 19, 2017]. http://www.speakingtree.in/allslides/sanjeevni-booti-is-the-divine-herb-from-ramayana-for-real.
[8]Patient:Diamorphine for pain relief. Patient Platform Limited; [March 17, 2017]. http://www.patient.info/in/medicine/diamorphine-for-pain-relief
[9] The difference between morphine and heroin. c 2017. Sober Media Group [March18, 2017] http://heroin.net/types-of-heroin/morphine-and-heroin/
[10] Douglas D. Richman (1987), The Toxicity of Azidothymidine (AZT) in the treatment of patients with AIDS and AIDS - related complex. The New England Journal of Medicine.1987; 317:192-197
[11] Difference between primary and secondary metabolites. July 1,2013.http://www.differencebetween.com