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Potential of Tannnins: A Review

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Abstract: Traditional systems of medicine like ayurvedic system have major treatment across globe. Tannins play an important role and has wide applications. Tanninsare water-soluble polyphenols that are present in many plant foods. They have been reported to be responsible for decreases in feed intake, growth rate, feed efficiency, net metabolizable energy and protein digestibility in experimental animals. Therefore, foods rich in tannins are considered to be of low nutritional value. However, recent findings indicate that the major effect of tannins was not due to their inhibition on food consumption or digestion but rather the decreased efficiency in converting the absorbed nutrients to new body substances. The anticarcinogenic and antimutagenic potentials of tannins may be related to their antioxidative property, which is important in protecting cellular oxidative damage, including lipid peroxidaton. The antimicrobial activities of tannins are well documented. The growth of many fungi, yeasts, bacteria and viruses was inhibited by tannins. We have also found that tannic acid and propyl gallate, but not gallic acid, were inhibitory to foodborne bacteria, aquatic bacteria and off-flavor-producing microorganisms. Their antimicrobial properties seemed to be associated with the hydrolysis of ester linkage between gallic acid and polyols hydrolyzed after ripening of many edible fruits. Tannins in these fruits thus serve as a natural defense mechanism against microbial infections. Tannins have also been reported to exert other physiological effects, such as to accelerate blood clotting, reduce blood pressure, decrease the serum lipid level, produce liver necrosis and modulate immunoresponses.

Key words: Tannins, antioxidative, antimicrobial, physiological effects

INTRODUCTION

The term tannin was first applied by Seguin in 1796 to denote substances present in plant extracts, which were able to combine with protein of animal hides; prevent their putrefication and convert them into leather. Tannins are astringent, bitter-tasting plant polyphenols that bind and precipitate proteins. The term tannin refers to the source of tannins used in tanning animal hides into leather; however, the term is widely applied to any large polyphenolic compound containing sufficient hydroxyls and other suitable groups (such as carboxyls) to form strong complexes with proteins and other macromolecules. Tannins have molecular weights ranging from 500 to over 3,000 (Handa and Kapoor, 2003). Examples of gallotannins are the gallic acid esters of glucose in tannic acid ($C_{76}H_{52}O_{46}$), found in the leaves and bark of many plant species Tannins may be employed medicinally in antidiarrheal, hemostatic and antibacterial (Bele *et al*., 2009) compounds. Also, they produce different colors with ferric chloride (either blue, blue black, or green to greenish black) according to the type of tannin. Plant parts containing tannins include bark, wood, fruit, fruitpods, leaves, roots and plant galls. Examples of plant species used to obtain tannins for tanning purposes are wattle (*Acacia* sp.), oak (*Quercus* sp.), eucalyptus (*Eucalyptus* sp.), birch (*Betula* sp.), willow (*Salix caprea*), pine (*Pinus* sp.), quebracho (*Scinopsis balansae*). Tannins can complex with: Proteins, starch, cellulose, minerals.

OCCURANCE OF TANNIN

Tannin occur abundantly in following sources Barks, Seeds, Leaves, Roots and rhizomes:

- Barks of cinnamon, wild cherry, cinchona, willow, acacia mimosa and otherspecies, oak and hamamelis
- Seeds of cocoa, guarana, kola and areca
- Leaves of hamamelis and green tea
- Roots and rhizomes of kramaria (rhatany) and fern

Physicochemical properties: Tannins dissolve in water to form colloidal solutions but their solubility varies with their degree of polymerization. They are soluble in alcohol and acetone. The stability of aqueous solution varies with the structure and is generally moderate e.g., During the extraction with boiling water (i.e., in the condition of a decoction) tannin such as geranin decomposes in 30 min to gallic acid, ellagic acid and corilagin. Like all phenols tannin reacts with ferric chloride. Hydrolysable tannins

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are polyestsers of glucose and upon hydrolysis, they release the sugar and either gallic acid, hexahydrodiphenic acid or both (Edwin-Jarald, 2007).

Extraction: Tannins are generally extracted with water and acetone mixture (methanol is to be avoided because it causes the methanolysis of galloyl depside). The optimal yield is obtained from the fresh tissues or from the frozen or lyophilized tissues, because in the dried drugs, part of the tannin is irreversibly combined to other polymers. After eliminating the acetone by distillation, the pigments and lipids are removed from the aqueous solution y a solvent extraction (e.g., with dichloro methane). Next an ethyl acetate extraction of the aqueous solution separates the dimeric proanthocyanidins and most gallotannins.

Classification: Tannins are usually divided into hydrolyzable tannins and condensed tannins (proanthocyanidins). At the center of a hydrolyzable tannin molecule, there is a polyol carbohydrate (usually D-glucose). The hydroxyl groups of the carbohydrate are partially or totally esterified with phenolic groups such as gallic acid (in gallotannins) or ellagic acid (in ellagitannins). Hydrolyzable tannins are hydrolyzed by weak acids or weak bases to produce carbohydrate and phenolic acids. Condensed tannins, also known as proanthocyanidins, are polymers of 2 to 50 (or more) flavonoid units that are joined by carbon-carbon bonds, which are not susceptible to being cleaved by hydrolysis. While hydrolyzable tannins and most condensed tannins are water soluble, some very large condensed tannin is insoluble (Evans, 2005).

- **Chemical test:** Gold beaterskin test-A small piece of goldbeater's skin soaked in 2% HCl, rinsed with distilled water and placed in a solution of tannin for 5 min. The skin piece is washed with distilled water and kept in a solution of ferrous sulphate. A brown colour is produced on the skin due to the presence of tannins
- **Phenazone test:** A mixture of aqueous extract (5 mL) of a drug and sodium acid phosphate (0.5 g) is heated, cooled and filtered. A solution of phenazone (2%) is added to the filtrate. bulky coloured precipitate is formed
- **Gelatin test:** To a solution of tannin (0.5-1%) aqueous solution of gelatin (1%) and sodium chloride (10%) are added. A white buff-coloured precipitate is obtained (Harborne, 2007)

DRUGS CONTAINING TANNINS

Liquid orals-*Terminalia arjuna* (Myroballan)

- **Medicinal uses:** *T. arjuna's* traditional use as a cardiotonic has been confirmed by modern research. Although some results of these studies (performed since the 1930s) appear conflicting (e.g., increases and decreases in heart rate or blood pressure), the herb seems to work best when blood supply to the heart is compromised as in ischemic heart disease or angina Ayurvedic medicine employs *T. arjuna* to restore balance of the 3 humors. *T. arjuna* also has been used as an aphrodisiac, diuretic and for earaches. This species has reduced cholesterol levels, as well. Studies done on *T. arjuna* combinations find the herb to be the most potent hypolipidemic agent compared with *T. bellirica* and *T. chebula*. *T. arjuna* may also play a role as an anti-atherogenic
- **Chemical constituents:** *T. arjuna* contains tannins, flavonoids and sterols. Five oleane derivatives, namely, arjunic acid, arjunolic acid, arjungenin, arjunetin and arjunglucoside I from stem bark extract of *Terminalia arjuna*
- **Analysis:** Extraction and TLC Desitometric Determination of Triterpenoid Acids (Arjungenin, Arjunolic Acid) from *Terminalia arjuna* Stem Bark Without Interference of Tannin). The stem bark of *Terminalia arjuna* Linn. (fam: Combretaceae), commonly known as Arjuna in Indian systems of medicine, is a reputed drug used for various cardiac disorders. *T. arjuna* stem bark is reported to contain different groups of chemical constituents including phenolics, tannins, saponins and triterpenoid acids. From our earlier experience with tannin containing herbal drugs, we are aware that tannins interfere in the extraction of certain compounds and hence in their quantification. In the present experiment, we report a sample preparation method to overcome the interference of the tannins by adsorbing them with carboxy methyl cellulose, which facilitates the efficient extraction of the triterpenoid acids. Further we established TLC densitometric methods for the quantification of two of the triterpenoid acids of *T. arjuna* stem bark viz., arjungenin and arjunolic acid using HPTLC. The methods were validated in terms of accuracy, precision and repeatability. The calibration curve showed linearity in the range of 160-480 ng spot^{-1} and 160-560 ng spot^{-1} for arjungenin and arjunolic acid, respectively. The percentage of arjungenin and arjunolic acid were found to be 0.324 and 0.524% w/w in the stem bark by this modified method of extraction, which was many times higher than when compared to that using the extraction method without CMC (0.018 and 0.049%, respectively). The study reiterates the

importance of sample preparation in the quantification of non polar phytochemicals from herbal raw materials, such that the compounds of interest are extracted efficiently, overcoming the interference of other compounds like tannins in the matrix of plant material (Kalola and Rajani, 2006)

Marketed formulation-Arjuna rishta: Preparation of Arishtas-Arishtas are medicinal preparation with alcoholic content and are made by fermentation methods. The drugs either in the powder form or in the form of a decoction in a solution of sugar are soaked for a particular period of time during this period the solution with the drugs undergo fermentation generating alcohol and fascilitating extraction of active principles from the drugs. The alcohol generated in the preparation serve as a preservative. Drugs prescribed is prepared by boiling the coarse drug with water. This is then filtered and transferred to an earthen pot. sugar is dissolved by boiling. If honey is prescribed should not be boiled. Drugs to be smeared should be in fine powder form. Mouth of the pot is then covered with earthen lid and sealed by clay smeared cloth which is to be wound in 7 consecutive layers. The container should be kept in temperature control room to maintain constant temperature during fermentation, After the fermentation process the liquid in the plot is decanted strained and bottled.

Solid dosage form-triphala: Triphala is a combination of

- *Emblica officinalis* (Amla)
- *Terminalia chebula* (Hirda)
- *Terminalia belerica* (Behada)

Triphala is one of the longest-used herbal remedies in the world. Triphala, meaning three fruits, is made from the fruits of three trees that grow throughout India and the Middle East, including amalaki fruit (*Embelica officinalis*), bibhitaki fruit (*Terminalia belerica*) and haritaki fruit (*Terminalia chebula*). In preparing triphala, these fruits are dried, ground into powder and then blended together according to the precise directions of Ayrurvedic tradition.

- **Medicinal uses:** Triphala is taken as a general health tonic and useful for constipation. Triphala is a gentle laxative that can be used daily and is not habit-forming and has no adverse effects on the intestinal flora. Improves overall health by increasing the efficiency and absorption of digestion and reduces gas., triphala is used as a blood builder and purifier and may increase red blood cell count and hemoglobin levels. Some healers prescribe it for diabetes, for its balancing effect on blood sugar levels. It also has anti-cholesterol and anti-mucus properties in the body. Triphala is believed to strengthen the kidneys and liver and is prescribed for hepatitis sufferers. Triphala is a source of vitamin C and is believed to improve the function of the immune system. The herbs in triphala have anti-inflammatory properties. The remedy is prescribed for gout, a form of arthritis caused by excess uric acid in the body and other inflammatory conditions. Triphala is said to have a calming and tonic effect on the nervous system and is recommended for Alzheimer's disease and other degenerative disorders of the nervous system.

Another use for triphala is to strengthen the eyes, particularly in cases of cataracts, glaucoma and conjunctivitis. It can be used as eyewash and may reduce soreness and redness in the eyes. Triphala can also be applied topically to the skin to speed the healing of bruises and sunburn.

- **Chemical constituents:** The major chemical constituents of Amla are Phyllemblin, Ascorbic acid (vitamin C), Gallic acid, Tannins, Pectin etc. Myrobalam fruits are the important source of tannin. The tannin of myrobalam are pyrogallol type (hydrolysable tannin). Chebulagic acid, chebulinic acid and corilagin are the hydrolysable .Tannin while chebulic acid, ellagic acid and gallic acid are the other contents. It is also contain glucose, sorbital, gentiobiose and other Sugar in traces. The dried fruit *T. bellerica* contains about 20% tannins of both condensed and hydrolysable type. Other constituents identified in the fruit include lipids (-sitosterol, saponins, gallic and ellagic acids and their derivatives, glycosides and various carbohydrates
- **Analysis:** The fruits of *T. chebula*, *T. bellerica* and *E. officinalis* were powdered separately and 5 g of powder of each was extracted using 100 mL of 70% aqueous acetone by Soxhlet extraction. The concentrated extracts were evaporated to dryness under reduced pressure at 45° and the extractive values were calculated. These extracts were further used for the estimation of total polyphenols and antioxidant evaluation. The amounts of total polyphenols in the fruit extracts were determined according to the Prussian blue method using 1% gum acacia and 85% phosphoric acid as a color stabilizer. To 0.1 mL of sample solutions, 1 mL of 0.016 M $K_3Fe(CN)_6$ was added followed immediately by 1 mL of

0.02 M $FeCl_3$ in 0.1 N HCl. The contents were mixed well and kept at room temperature for 15 min. This was followed by addition of 5 mL of stabilizer containing water, 85% H_3PO_4 and 1% gum acacia in volume proportions of 3:1:1. The contents were vortexed and the colour density was measured at 700 nm against a reagent blank consisting of all of the reagents except the polyphenols using Shimadzu UV/Vis spectrophotometer-1601. The total polyphenol contents were calculated as% w/w of gallic acid equivalents (Palavy and Priscilla, 2006)

Marketed formulation: Triphala tablet, triphala choorna:

- **Formulation of choornam:** This is a dry fine powder form of the drug choornam can be used both internally and externally
- **Preparation:** The drug selected is washed cleaned and dried. The fineness of the powder using a pulveriser. The fineness of the powder improve the therapeutic efficacy. In case of compound choornas each drug should be powdered separately and finally all individual drug powder are to be mixed. The choorna should be fine of atleast 80 mesh sieve

Semisolid dosage form: Rosemary leaf

- **Medicinal uses:** Rosemary is a stimulant of the circulatory system. Externally, it is used to treat bites, stings, sores, eczema, bruises and wounds. It is also used in lotions to ease rheumatism and arthritis Mixed with borax and used cold, it is said to make a nice-smelling hair wash that can prevent dandruff and stimulate hair growth. Rosemary is particularly effective at treating oily skin and oily hair, helping to restore proper balance and oil levels. Rosemary's powerful antimicrobial properties help to prevent infections and treat skin conditions such as athlete's foot, psoriasis, eczema, shingles and neuralgia

Internally, it's used to treat migraines, bad breath and to stimulate the sexual organs (but it can be an irritant to the stomach, intestines and kidneys, so use it sparingly). Rosemary is also used to treat nervous disorders, upset stomachs and to regulate the menstrual cycle and ease cramps. Mix the crushed leaves generously into meats, fish, potato salads, etc. at your next picnic to prevent food poisoning. The essential oil is used in aromatherapyas an inhalant and decongestant and to enhance memory and clear concentration. Rosemary is taken by mouth to treat indigestion, headache, stress, nervous tension, as well as to promote menstrual flow and to raise low blood pressure. It's put on the skin to stop redness and pain and to treat fibromyalgia and sciatica (pain in the muscles and nerves). Rosemary oil has been used to promote wound healing.

- **Chemical constituents:** Rosemary leaf contains phenolic acids (2-3% rosmarinic, chlorogenic, caffeic acids); phenolic diterpenoid bitter substances (up to 4.6% carnosol, rosmaridiphenol, rosmanol); triterpenoid acids (oleanolic acid, ursolic acid); flavonoids (apigenin, luteolin, nepetin, nepitrin); 1.2-2.5% volatile oil, of which 15-50% is 1,8-cineole, 15-25% a-pinene, 12-24% a-terpineol, 10-25% camphor, 5-10% camphene, 1-6% borneol, 1-5% bornyl acetate and tannins
- **Analysis:** *Rosmarinus officinalis* extracts were investigated by a combination of bioassays and biochemical analysis to identify bioactive compounds. The 2,2-diphenyl-2-picrylhydracyl hydrate (DPPH) radical scavenging method, Folin-Ciocaulteau method and HPLC chromatography were used to study the distribution and levels of antioxidants (AOXs). Antimicrobial activity analysis was carried out using the disk diffusion and broth dilution techniques. A good correlation between the AOX activities and total phenol content in the extracts was found. Although all rosemary extracts showed a high radical scavenging activity, a different efficacy as antimicrobial agent was observed. Methanol extract containing 30% of carnosic acid, 16% of carnosol and 5% of rosmarinic acid was the most effective antimicrobial against Gram-positive bacteria (minimal inhibition concentration, MIC, between 2 and 15 $\mu g\ mL^{-1}$), Gram negative bacteria (MIC between 2 and 60 $\mu g\ mL^{-1}$) and yeast (MIC of 4 $\mu g\ mL^{-1}$). By contrast, water extract containing only 15% of rosmarinic acid showed a narrow activity. MIC value of the methanol and water extracts is in a good correlation with the values obtained with pure carnosic acid and rosmarinic acid, respectively. Therefore, our results suggested that the antimicrobial rosemary extracts efficacy was associated with their specific phenolic composition. Carnosic acid and rosmarinic acid may be the main bioactive antimicrobial compounds present in rosemary extracts. From a practical point of view, rosemary extract may be a good candidate for functional foods as well as for pharmaceutical plant-based products (Moreno *et al*., 2006)

Marketed formulation-Lubrajel: Neutraceuticals-(I) Pomegranate:

Punica Granatum (Pomegranate)

- **Medicinal uses:** A decotion of seed is used to treat syphilis. Juice used to treat jaundice and diarrhoea. Juice of flower is used to treat nose bleeds. The fruit pulp and the seed are stomachic. Pomegranate juice enters into preparations for treating dyspepsia and is considered beneficial in leprosy. Because of their tannin content, extracts of the bark, leaves, immature fruit and fruit rind have been given as astringents to halt diarrhea, dysentery and hemorrhages
- **Chemical constituents:** One pomegranate delivers 40% of an adult's daily vitamin C requirement. It is also a rich source of folic acid and of antioxidants Pomegranates are high in polyphenols. The most abundant polyphenols in pomegranate are hydrolysable tannins, particularly punicalagins, which have been shown in many peer-reviewed research publications to be the antioxidant responsible for the free-radical scavenging ability of pomegranate juice
- **Analysis:** Quantitative determination of the polyphenolic content of pomegranate peel

The quantitative determination of total phenols, ellagic tannins and gallic and ellagic acids in the peel of the Tunisian pomegranate variety Chelfi, has been carried out. The ellagic tannin content is prominently less than the amount of total phenols, which led us to look for the presence of the condensed tannins. The determination of the content of catechic tannins in eight Tunisian varieties of the pomegranate was carried out using weekly samples over a period of 2 months.

Analysis of ellagic tannins. The absorbance was measured to its maximum at 590 nm by means of a spectrometer UV-Visible Shimadzu (UV. 260) (Ben-Nasr *et al*., 1996).

Marketed formulation: (i) Pomeratrol capsules 60-20 mg. (ii) t'Zerah - Pomegranate beauty and skin

Cosmetics: Hair dye: This product contains a natural tannin (from Neutral Henna a plant ingredient) known as hennotannic acid. Tannins are slightly astringent and their use will tighten the surface of the scalp and hair follicles, strengthening the follicles' grasp on each hair. Hennotannic acid coats the hair. It seals in oils and tightens the hair cuticle giving your hair a rich, healthy gentle shine.

- **Chemical constituents:** The constituents of Henna is found in it in a brown substance of a resinoid fracture, having the chemical properties which characterize the tannins and therefore named *hennotannic acid*. Dried, powdered leaves of henna contain about 0.5 to 1.5% lawsone, the chief constituent responsible for the dyeing properties of the plant. Henna also contains mannite, tannic acid, 2-hydroxy-1:4-naphthoquinone resin mucilage, gallic acid, glucose, mannitol, fat, resin and mucilage are also present. The colouring matter is the quinone and napthaquinone. Lawsone, the dye molecule in henna will bind with protein, such as the keratin in fingernails, hair and skin
- **Analysis:** A fixed wavelength of 248 nm; lawsone Rt = 6.3 min. The identification of lawsone was confirmed by UV. spectrum, Rt and by spike analysis using pure standard
- **Marketed formulation:** Herbal Heena- Herbal Henna is available in pure form and without any additives. It is obtained by mixing 13 different herbs. The product helps in removing dandruff and prevents hair loss. Herbal henna is popular in the international market because it increases the longevity of hair, is a natural conditioner and removes dryness. It is perfectly safe and has no ill effects

CONCLUSIONS

Tannins are astringent, bitter-tasting plant polyphenols that bind and precipitate proteins,applied to any large polyphenolic compound containing sufficient hydroxyls and other suitable groups (such as carboxyls) to form strong complexes with proteins and other macromolecules. Tannins have molecular weights ranging from 500 to over 3,000. Tannins have the property of binding proteins together, even precipitate them, so they affect the proteins in saliva making them stringy, hence the effect on the inside of the mouth. They also have the ability to react with bacterial cell walls, polysaccharides, carbohydrates and enzymes, all present in the mouth In the plant, tannins are defensive compounds that counteract bacteria and fungi by interfering with their surface proteins The sensation of astringency is caused by the tanning of the proteins in the saliva and mucous membranes of the mouth; lubrication is reduced and the surface tissues actually contract. Pharmaceutical, chemical, microbiological and analytical part also serves an important part to study drugs containing tannins. Therapeutic applications of tannin containing drugs also play an important role. They can have a large influence on the nutritive value of many foods eaten by humans and feedstuff eaten by animals. Tannins are common in fruits (grapes, persimmon, blueberry), in tea, in chocolate, in legume forages (trefoil), in legume trees (*Acacia* sp., *Sesbania* sp.), in grasses (sorghum, corn)

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