

# A Brief Review on *Trachyspermum Ammi.* and its Pharmacological Activity

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**Abstract:** *Ajwain seeds revealed to possess antiseptic, stimulant, carminative, diuretic, anaesthetic, antimicrobial, antiviral, nematocidal, antiulcer, antihypertensive, antitussive, bronchodilatory, antiplatelet and hepatoprotective as well as antihyperlipidemic effects, many of those were remarked by early Persian physicians. With reference to these pharmacological activities, Ajwain seeds can be a good candidate for to be applied in clinical practice. However, in spite of various experimental and animal studies, lack of comprehensive clinical trials aimed on regarded effects is still remained to reconfirm the traditional knowledge. Thymol in ajwain seeds is packed with anti-inflammatory, anti-bacterial and anti-fungal properties that help in providing instant relief from cold, fever and flu. It helps in clearing nasal blockage by discharging the mucus. Ajwain Works as an Expectorant: Ajwain has Thymol which has expectorant properties. Thus, it acts as a decongestant and helps in mucus removal to clear children's nasal passage and help them breathe easily. (1) *Trachyspermum ammi* (L.) Sprague or commonly Ajwain is a herbaceous herb belonging to the family Apiaceae and vastly grows in Egypt, Iran, Pakistan, Afghanistan, and India as well as European region. Known as Zenyan or Nankhah in medical and pharmaceutical manuscript of medieval Persia, seeds of Ajwain were highly administered by traditional healers and traditionally employed for different ailments. Due to its various chemical constituents, the herb seeds were also evaluated for its numerous pharmacological properties. Accordingly, current work was carried out to review the traditional and modern pharmacological properties of Ajwain regarding current and medieval reports. To this, respective databases were searched for the terms 'Trachyspermum ammi', 'Carum copticum', 'Ajwain' and 'Ajowan' without limitation up to early 2013. Information on the herb was gathered via electronic search using Pubmed, Scopus, Google scholar and SID (for articles in Persian language) as well as medical and pharmaceutical manuscripts of Persian medicine. (1).*

**Keywords:** Ajwain, *Trachyspermum ammi* (L.) Sprague, traditional medicine, herbal medicine, Apiaceae, nasal congestion

## I. INTRODUCTION

Nature has long served as a rich source of therapeutic floras that have been used to sustain human health. These floras have the power to heal numerous infectious diseases without causing any negative side effects (Shelef, 1983). Due to the existence of bioactive components, spices have been recognised for their therapeutic benefits in addition to their scent and flavor-giving abilities. Globally, an increasingly serious and horrifying issue has been the resistance of dangerous bacteria to various antifungal treatments. Globally, invasive fungal infections are an issue that cause 1.7 million fatalities annually. Chemotherapy, AIDS, and/or organ transplantation are all indicators of immunocompromised patients' high prevalence of these conditions. Over 300,000, 750,000, and 10,000 cases, respectively, of invasive aspergillosis, candidiasis, and mucormycosis have been reported annually most recently.(8)

The genus name *Trachyspermum* has originated from two Greek words, *Trachy* means rough and *spermum* means seeded, while species *ammi* is a Latin word. The plant belongs to the family Apiaceae (Umbelliferae). The Hindi name Ajwain is originated from two Sanskrit words Vavanaka or Ajomoda which are derived from the Greek word yavnaha. This plant has great medicinal importance and also used as a spice which is used in Indian dishes like in curry due to its aromatic smell and pungent taste. Each part of this plant exhibits many important pharmacological activities. The main utilizing parts of this plant are its fruits and seeds. Seeds are mainly used in snacks, pastries and as a spice. Decoction of

ajwain seeds is utilized as first line treatment in Ayurveda for abdominal discomfort, loose bowels, cough and stomach inconvenience. (6)

While the fruits of Ajwain is found to have antiseptic, anti-fungal/anti-bacterial and anthelmintic properties. The main active phytochemical constituents of the plant i.e. thymol and carvacrol are associated with many important therapeutic activities like antispasmodic, germicide, anti-fungal properties, antitussive, antiseptic and expectorant properties. The Ajwain seeds also have carminative, diuretic, anaesthetic, anti-microbial, antiviral, anti-ulcer, antiplatelet and hepatoprotective properties. (5)

#### **Plant Description:**

Ajwain is a small, highly branched annual herbaceous plant belongs to the Apiaceae (Umbelliferae) family. The stem of the plant is striated erect and including glabrous or minutely pubescent properties which may grow up to 90 cm tall. It consists of feathery leaves that are 2.5cm long with 16 umbellets which bears 6-16 flowers towards the head portion. The flowers of this plant are white and actinomorphic in nature. The leaves are pinnately shaped with a terminal and 7 pairs of leaflets. The fruits of the plant are small, egg shaped, aromatic and greyish-brown in colour. (2)

#### **Cultivation and collection:**

The crop is raised in cold climates both dry-land and irrigated. It thrives in loams or clayey loams but may grow in any type of soil. In the damp soil, seeds are dispersed from September to November. Depending on the climate, germination takes 5–15 days. The initial irrigation should be minimal. Two months or so pass before the blossoming begins. It's time to harvest in February or March. When the flower heads turn brown, the fruits are ready for picking. The plants were then dried after being pulled out by the roots. With care, rub the dried fruits apart with your hands or feet.

#### **Macroscopic Characters:**

It is widely grown in arid and semi-arid regions where soils contain the high levels of salts. Ajwain is a profusely branched annual herb, 60-90 cm tall. Ajwain is a voluminous 16 umbellets make up the compound umbel of the inflorescence. Including up to 16 actinomorphic, whitish, and male and bisexual; five bilobed petals on the corolla; five stamens. Fruit consists of two mericarps, grayish brown, ovoid, compressed, about 2 mm long and 1.7 mm wide, 5 ridges and 6 vittae in each mericarp, usually separate, 5 primary ridges. (2)

#### **Microscopic Characters:**

Transverse section of fruit shows two hexagonal structures attached with each other by a carpophore, epicarp consists of a single layer of tangentially elongated tabular cells. mesocarp consists of moderately thick-walled, rectangular to polygonal tangentially elongated cells having some vittae, carpophores and vascular bundles present as groups of thickwalled radially elongated cells, integument, barrel shaped of tangentially elongated cells. endosperm consists of thinwalled cells filled with embryo, oil globules, small and circular, composed of polygonal thin walled cells. The powder microscopic shows the presence of oil globules and groups of endosperm cells. (2)

#### **Chemical constituents:**

The ajwain essential oil contained thymol (87.75%) and carvacrol (11.17%) as major constituents and major nonphenolic components quantified were p-cymene (60.78%) and  $\gamma$ -terpinene (22.26%) Trachyspermum ammi seeds contain various chemical constituents such as fiber (11.9%), carbohydrates (38.6%), tannins, glycosides, moisture (8.9%), protein (15.4%), fat (18.1%), saponins, flavone and mineral matter (7.1%) like calcium, phosphorus, iron and nicotinic acid. Thymol and essential oil (2-4%) present in the fruits are considered as the major constituents (35%-60%) of the plant. The nonthymol constituents which are present in Ajwain are paracymene,  $\gamma$ -terpinene,  $\alpha$ - and  $\beta$ -pinenes, dipentene,  $\alpha$ -terpinene, and carvacrol. Camphene, myrcene, and  $\alpha$ -3-carene are present in small quantity in the plant. The alcoholic extract of plant consists of a highly hygroscopic saponin. A yellow coloured crystalline flavone extracted from the fruits which is a steroid like substance in nature contains glucopyranosyloxythymol, glucoside and 25% oleoresin. (1)

**Phytochemical Analysis:**

The phytochemical tests were carried out for the above mentioned plant extract using the standard procedures to identify the components Mentioned at the below. (12)

**Tests for alkaloids:**

**Dragendorff's test-**

To 0.5 ml of plant extracts the Dragendorff's reagent was added.(Potassium bismuth iodide solution). A reddish brown precipitate confirms that test as positive.

**Hager's test-**

To 0.5 ml of plant extracts, a few drops of Hager's reagent was added. Formation of yellow precipitates confirms the presence of alkaloids.

**Wagner's test-**

To 0.5 ml of plant extracts the Wagner's reagent was added. (Solution of Iodine in potassium Iodide). A reddish brown precipitate confirms that test as positive.

**Mayer's test-**

To 0.5 ml of plant extracts the Mayer's reagent was added. (Potassium mercuric iodide solution). A white creamy precipitate confirms that test as positive.

**Tests for carbohydrates:**

**Anthrone test-**

0.5 mg of plant extracts was shaken with 2.5ml of water, filtered and the filtrate was concentrated. To this 0.5ml of anthrone reagent solution was added. Formation of green or blue colour indicated the presence of carbohydrates.

**Benedict's test-**

0.5 mg of plant extracts was shaken with 2.5 ml of water, filtered and the filtrate was concentrated. To this 1.25 ml of Benedict's solution was added and boiled for 5 minutes. Brick red precipitate indicated the presence of carbohydrates.

**Fehling's test (free reducing sugars)-**

At the first step equal volume of Fehling's A (copper sulphate in distilled water) and Fehling's B (potassium tartarate and sodium hydroxide in distilled water) reagents are mixed carefully. Then few drops plant extracts was added and boiled. Brick red precipitate of cuprous oxide indicated the presence of free reducing sugars.

**Molisch's test-**

To 0.5 ml of plant extracts few drops of alcoholic  $\alpha$ -naphthol was added. Then 0.2 ml of concentrated sulphuric acid was added slowly along the sides of test tubes.Reddish-violet ring at the junction of the two layers indicated the presence of carbohydrates.

**Barfoed's test-**

About 0.5 mg of plant extracts was dissolved in distilled water and filtered. 1 ml of the filtrate was then mixed with 1 ml of Barfoed's reagent in a test tube and then heated on a water bath for a period of two minutes. A reddish precipitate of cuprous oxide confirms that test as a positive.

**Tests for flavonoids:**

**Shinoda's test-**

To 0.5 ml of plant extracts a piece of metallic magnesium was added, followed by addition of 2 drops of concentrated hydrochloric acid. Presence of deep red colouration indicated the presence of flavonoids in the extract.

**Ferric chloride test-**

To 0.5 ml of plant extracts a few drops ferric chloride solution was added. The presence of green colouration indicated the presence of flavonoids.

**Lead ethanoate test-**

To 0.5 ml of plant extracts 0.3 ml of lead ethanoate solution was added. A buff-coloured precipitate indicated the presence of flavonoids.

**Tests for glycosides:**

**Borntrager's test (Anthraquinone Glycosides)-**

1 ml of benzene and 0.5 ml of dilute ammonia solution were added to the plant extracts. A reddish pink colour indicated the presence of glycosides.

**Keller killiani's test (Cardiac glycosides)-**

0.4 ml of glacial acetic acid containing traces of ferric chloride and 0.5 ml of concentrated sulphuric acid were added to the plant extracts carefully. A reddish-brown colour formed at the junction of the two layers and the upper layer turned bluish green indicating the presence of glycosides.

**Test for resins:**

0.5 ml of plant extracts were treated with a few drops of acetic anhydride solution followed by one ml of concentrated sulphuric acid. Resins give colouration ranging from orange to yellow

**Tests for steroids and Triterpenoids:**

**Liebermann - Burchard Test-**

0.5 ml of plant extracts was treated with few drops of acetic anhydride, boil and cool, concentrated sulphuric acid is added along the sides of the test tube, shows brown ring at the junction of two layers and the upper layer turns green that shows the presence of sterols and formation of deep red colour indicated the presence of triterpenoids.

**Salkowski Test-**

0.5 ml of each extract was treated in chloroform with few drops of concentrated sulphuric acid, shaken well and allow to stand for some time, red colour at the lower layer indicates the presence of sterols and formation of yellow coloured lower layer indicates the presence of triterpenoids.

**Test for saponins:**

**Froth test-**

A pinch of the dried powdered plant was added to 3 ml of distilled water. The mixture was shaken vigorously. Formation of a foam indicated the presence of saponin.

**Tests for Tannins:**

**Lead acetate Test-**

To 0.5 ml of plant extracts, a few drops of 10 % lead acetate was added. Precipitate was formed, indicated the presence of tannins.

**Ferric chloride Test-**

To 0.5 ml of plant extracts, few drops of 0.1% ferric chloride solution was added. Formation of brownish green or a blue black colouration indicated the presence of tannins.

**Tests for Amino acids:**

**Millons Test-**

To 0.5 ml of plant extracts 2 ml of Millon's reagent (Mercuric nitrate in nitric acid containing traces of nitrous acid) was added. White precipitate appears which turns red when gentle heating.

**Ninhydrin Test-**

To 0.5 ml of plant extracts few drops of 5% ninhydrin was added and then boiled. Appearance of violet colour indicated the presence of amino acids.

**Test for starch:**

To 0.5 ml of plant extracts Iodine as reagent was added. Appearance of dark blue colour which disappeared on heating and reappears on cooling indicated presence of starch.

**Tests for organic acids:**

**Malic acid Test-**

To 0.5 ml of plant extracts few drops of 40% FeCl<sub>3</sub> solution was added. Formation of yellowish colour indicated the presence of malic acid.

**Oxalic acid Test-**

To 0.5 ml of plant extracts, few drops of 1% KMnO<sub>4</sub> and dilute H<sub>2</sub>SO<sub>4</sub> were added. Disappearance of colour indicated the presence of oxalic acid.

**Tests for inorganic acids:**

**Sulphate Test-**

To 0.5 ml of plant extracts, the lead acetate reagent was added. White precipitate which is soluble in NaOH, indicated the presence of sulphate.

**Carbonate Test-**

To 0.5 ml of plant extracts, dilute HCl solution was added. Libration of CO<sub>2</sub> gas, indicated the presence of carbonates.

**Test for ascorbic acid-**

To 0.5 ml of plant extracts, 2 ml water, 0.1 gram sodium bicarbonate and about 20 mg ferrous sulphate were added, shaken and allowed to stand. A deep violet colour was produced. To this 5 ml of 1M sulphuric acid was added, the colour disappeared showing the presence of ascorbic acid.

**Tests for phenolic compounds:**

**Lead acetate Test-**

To 0.5 ml of plant extracts few drops of 10% lead acetate solution was added. White precipitate indicated the presence of phenolic compounds.

**Tests for protein:**

**Biuret Test-**

To 0.5 ml of plant extracts, 4% NaOH solution and few drops of 1% CuSO<sub>4</sub> solution were added. Violet colour appears, indicated the presence of protein.

**Millon's Test-**

To 0.5 ml of plant extracts 2 ml of Millon's reagent (Mercuric nitrate in nitric acid containing traces of nitrous acid) was added. White precipitate appears which turns red when gentle heating.

**Test for oils and fats:**

A small quantity of the dried plant was pressed between the two filter papers. Oil stain on the filter papers indicated the presence of oils and fats.

**Test for coumarins:**

To 0.5 ml of plant extracts, the solution of 10% NaOH was added. The appearance of yellow colour indicated the presence of coumarins.

**Test for anthraquinones:**

To 0.5 ml of plant extracts few drops of 2% HCl was added. Red colour precipitate indicated the presence of anthraquinones. (12)

**Pharmacological Activities:**

Ajwain with its characteristics aromatic smell and pungent taste is widely used as spice in curries. In Indian system of medicine, Ajwain is administered for curing stomach disorder, a paste of crushed fruits is applied externally for relieving colic pains and a hot dry fermentation of the fruits is applied on chest for asthma. T. ammi has been shown to possess Antimicrobial, Hypolipidemic, digestive stimulant, Antihypertensive, hepatoprotective, antispasmodic, broncho-dilating, antilithiasis, diuretic, abortifacients, galactogogic, antiplatelet-, aggregatory, antiinflammation, gastroprotective, nematocidal, anthelmintic, detoxification of aflatoxins and ameliorative effects. Therapeutic uses of T. ammi fruits include stomachic, carminative and expectorant, antiseptic and amoebiasis antimicrobial seeds soaked in lemon juice with prunus amygdalus are given curing amenorrhoea and it is also used as antipyretic, febrifugal and in the treatment of typhoid fever. (2)

**Antibacterial and Antifungal Activities-**

Bacterial strains Gram positive Staphylococcus aureus, Streptococcus Pyogenes, Gram negative, Escherichia Coli, Pseudomonas aeruginosa and Fungal strains Candida albicans, and Aspergillus clavatus were chosen based on their clinical and pharmacological importance. To assay the antibacterial efficacy of Ajwain, acetone and aqueous extracts were tested against Enterococcus faecalis, Escherichia coli, Klebsiella pneumonia, Pseudomonas aeruginosa, Salmonella typhi, Salmonella typhimurium, Shigella flexneri, and Staphylococcus aureus using agar diffusion assay.

On the other hand, no activity was reported against Pseudomonas fluorescens and Micrococcus luteus. As Ajwain may have large amounts of Thymol or Carvacrol in its total essential oil, mentioned phenolic compounds are reported to be either bactericidal or bacteriostatic agents depending on the concentration. (2)



#### **Analgesic and Antinociceptive Effects-**

In order to evaluate the analgesic and antinociceptive activity of Ajwain, an In vivo investigation was carried out using a Tail-flick Analgesiometer Device. An experimental trial study has also been carried out to compare the antinociceptive effect of the hydroalcoholic extract of Ajwain with morphine sulphate using formalin test. Findings revealed that Ajwain extract exhibited antinociceptive effect on both early and late phases. (2)

#### **Antihypertensive, antispasmodic and broncho-dilating activity-**

The antihypertensive effect of *T. ammi* administered intravenously in vivo, and the antispasmodic and broncho-dilating action in vitro showed that calcium channel blockade that has been found to mediate spasmolytic effect of plant materials and it is being considered that this mechanism contributed to their observed result supported the traditional use of *T. ammi* in hyperactive disease states of the gut such as colic and diarrhoea as well as in hypertension. (2)

#### **Hepatoprotective activity-**

The Hepatoprotective actions in vivo showed that *Trachyspermum ammi* was 80% protective in mice against a normally lethal dose of paracetamol (1g/kg), it prevented the CCL4-induced prolongation of pentobarbital sleeping time in mice, and it tended to normalize the high serum levels of liver enzymes caused by CCL4-induced liver damage in rats. (2)

#### **Antilithiasis and diuretic activity-**

Antilithiasis and diuretic actions in vivo of *Trachyspermum ammi* on inhibiting oxalate urolithiasis induced in rats. In a further study of a possible diuretic effect the result found that *Trachyspermum ammi* was not effective in increasing the 24-h urine production. The results were reported satisfactory against pure calcium-oxalate stone. (2)

#### **Antiplatelet-aggregatory-**

Antiplatelet activity has been done on the dried ethereal extract of Ajwain. Antiplatelet-aggregatory experiments in vitro with blood from human volunteers, it showed that a dried the real extract of *Trachyspermum ammi* seeds, inhibited aggregation of platelets induced by arachidonic acid, collagen and epinephrine, Research study was intended to support the traditional use of *Trachyspermum ammi* in women post parturition. (2)

#### **Anti-inflammatory Effects-**

Ajwain was also evaluated for exhibiting anti-inflammatory effect. Accordingly, both total alcoholic extract and total aqueous extract possess in vivo significant anti-inflammatory effect. Anti-inflammatory potential of the total alcoholic extract (TAE) and Total aqueous extract (TAQ) of the Ajwain seeds was determined. (2)

#### **Antitussive effects-**

Antitussive effect of Ajwain has been reported in traditional medical manuscripts. According to the results, both concentrations of Ajwain seeds revealed significant reduction of cough number which may be a result of its potent antitussive effect. Relative studies showed the inhibitory effect of both Ajwain extract and essential oil on Histamine (H1) receptors of isolated guinea-pig tracheal chains. In another study, in the field of respiratory, bronchodilatory effects of different fractions of Ajwain essential were examined. (2)

#### **Antifilarial activity-**

In vitro activity of a methanolic extract of the fruits of *Trachyspermum ammi* (Apiaceae) against *Setaria digitata* worms has been investigated. The crude extract and the active fraction showed significant activity against the adults *S. digitata* by both a worm motility and MTT [3-(4, 5-dimethylthiazol-2yl)-2, 5-diphenyltetrazolium bromide] reduction assay. The isolated active principles phenolic monoterpene screened for in vivo antifilarial activity against the human filarial worm *B. malayi*. *Trachyspermum ammi* crude extract exhibited macrofilaricidal activity. (2)

#### **Gastro protective Activity-**

*Trachyspermum ammi* fruit showed antiulcer activity by using different ulcer model. Animals pre-treated with ethanolic extract showed significant decrease in ulcer index and percentage ulcer protection in all models. The results suggest that the extract showed significant protection ( $p < 0.001$ ) by reducing ulcerative lesions when compared with control group of animals.

#### **Detoxification of aflatoxins-**

Aqueous extract of ajowan seeds was found to contain an aflatoxin inactivation factor. Thin layer chromatography analysis of the toxins after treatment with IF showed relative reduction of aflatoxin  $G1 > G2 > B1 > B2$ . Toxin

decontamination in spiked corn samples could be achieved using IF. This study emphasizes the potential of ajowan IF in aflatoxin removal from contaminated food commodities. (2)

#### **Anthelmintic activity-**

Anthelmintic activity of *Trachyspermum ammi*, shows its effect against specific helminths, e. g. *Ascaris lumbricoides* in humans and *Haemonchus contortus* in sheep. Anthelmintic Activity *Trachyspermum ammi* exert by interference with the energy metabolism of parasites through potentiation of ATPase activity and thus loss of energy reserves. The plant has also been reported to possess cholinergic activity with peristaltic movements of the gut, thus helping in expulsion of intestinal parasites which might also be a contributory factor to its anthelmintic activity. Antifilarial activity assessment of the Ajwain methanolic extract was done as an in vitro assay against adult bovine filarial *Setaria digitata* worms. In that investigation, a bioassay-guided fractionation was prepared by introducing the crude extract to flash chromatography. HPLC analysis was done for both crude extract and active fraction. Ajwain was also evaluated for its nematicidal activity. A survey was done on the total essential oil components of Ajwain that showed significant nematicidal activity against pinewood nematode, *Bursaphelenchus xylophilus*. Nematicidal activity of Ajwain essential oils LC50 values was measured as 0.431mg/ml [38] and it was mainly attributed to the activity of Thymol and Carvacrol. (2)

#### **Antiviral Effects-**

For the evaluation of Ajwain antiviral activity, an in vitro assay was carried out on the methanolic extract of the herb which showed significant inhibitory effects on Hepatitis C Virus (HCV) protease. (1)

## **II. CONCLUSION**

*Trachyspermum ammi* is an important medicinal plant, which has both nutritional as well as medicinal uses. This particular have a wide established and hidden therapeutic uses. This medicinal plant needs a scientific explore to the hidden curative and therapeutic potential. This review is an attempt to provide well assembled scientific data on the behalf of *Trachyspermum ammi*. It is expected that this review will attract attention towards medicinal potential, applications and commercialization of various Pharmacognostic, Phytochemical studies, Ethnopharmacology relevance, Pharmacological activities of *Trachyspermum ammi*.

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