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Theme

Antifungal activity of *Peganum Harmala L.* essential oil against Tomato phytopatogenecity

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Dedication

I dedicate this modest work as a sign of respect and rebirth to my dear parents my beloved **Father** Bouderssa and my sweet **Mother** Sahra whose affection, love, encouragements and prays day and night makes me able to get this far

Also to all my dear beautiful sisters Halima ,Meriem, Soumia, Nour el-houda and Imen whom without them nothing will be complete

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To all my friends Sekour Samia, Boultif Soufounizia and Hadad Yousra

And to my colleague Habanda aya aicha

Sihem

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List of abbreviations

- LD : Lethal dose
- LT : Lethal time
- MIC : Minium ihibitory concentration
- MBC : Minium Bactericidal concentration
- Ppm : Part per million
- PDA : Potato dextrose agar

Introduction

Introduction

Tomato production and consumption are constantly increasing. It is noteworthy that tomatoes are not only sold fresh, but also processed as soups, sauces, juices or powder concentrates. The tomato ranks 7th in worldwide production after maize, rice, wheat, potatoes, soybeans and cassava, reaching a worldwide production of around 160 million tons on a cultivated area of almost 4.8 million hectares in 2011 (FAOSTAT, 2011).

Peganum harmala L. (P. harmala) commonly known as Syrian Rue is a widely used medicinal plant from the family Nitrariaceae. It is also known as Wild Rue orSyrian Rue because of its resemblance to plants of therue family. It is a perennial plant which can grow toabout 0.8 m tall.*Peganumharmala*is used as an analgesic and anti-inflammatory agent. In Yemen it was used to treatdepression, and it has been established in the laboratorythat harmaline, an active ingredient in *Peganumharmala*,is a central nervous system stimulant (Pathan *et al*., 2012)

Essential oils have been important substances since early times. Essential oils are valuable plant products, generally of complex composition comprising the volatile principles contained in the plant and the more or less modified during the preparation process (Hamid *et al*., 2011).

Since ancient times, essential oils are recognized for their medicinal value and they are very interesting and powerful natural plant products. They continue to be of paramount importance until the present day. Essential oils have been used as perfumes, flavors forfoods and beverages, or to heal both body and mind for thousands of years (Abdelouaheb, 2012).

Plants are the most important source of chemical compounds. Primary plant metabolism synthesizes essential compounds, which are present in all plant species. There is growing evidence that these compounds when applied on other plants, they can protect the plant from the pathogens and pests (Manonmani *et al.,* 2009).

Fungi are an extremely versatile class of organisms comprised mostly of saprophytes, thriving on dead organic material. The soil-borne fungus, Fusarium oxysporum is the causal

agent of vascular wilt, a disease that affects a large variety of economically important crops worldwide (Ortoneda *et al.*, 2004).

Numerous fungicide applications are often needed to control fungal plant pathogens and consequently there is increasing demand for alternative means to control them .Natural plant products are one of the most important alternatives which do not have indiscriminate hazardous effects like synthetic products (A. Sarpeleh *et al .,* 2009).

The present study consists a synthesis evaluation a on the antifungal activity in vitro of the plant on the growth of phytopathogen resepansable on rot and wilt diseases of tomato plant then research active chemical compounds that it contains and which make them valuable plant.

this work is written as follows:

- The first chapter we will cite different agents that kill tomatoes and focus on tomato Fusarium wilt .
- The second chapter we will present an bibliographic etudes about the genus *Peganum* specifying on the sepecie *Peganum harmala* as an anti-phytopathogenic agent , then we will stat an analyse of the biologic activities of P.harmala essential oil and extracts.

Chapter 1: Tomato Phytopathogenicity

Chapter 1 : Tomato Phytopathogenicity

I. Generality

The cultivated tomato, Solanum lycopersicum L., is the world's most highly consumed vegetable due to its status as a basic ingredient in a large variety of raw, cooked or processed foods. It belongs to the family Solanaceae, which includes several other commercially important species. Tomato is grown worldwide for local use or as an export crop.

Tomato is one of the best studied cultivated dicotyledonous plants at the molecular level and has been used as a model species for research into gene mapping, gene characterisation (e.g. plant pathogen resistance genes) and gene transfer approaches. It is also useful to study other plant traits such as fruit ripening, hormone function and vitamin biosynthesis.

Ⅰ.1 Tomato importance

Originating from the Andes, tomatoes (Solanum lycopersicum L.) were imported to Europe in the 16th century. At present, this plant is common around the world, and has become an economically important crop. Furthermore, this plant is a model species for introducing agronomically important genes into dicotyledonous crop plants (Paduchuri *et al.,* 2010).

The tomato is considered a protective food because of its particular nutritive value, as it provides important nutrients such as lycopene, beta-carotene, flavonoids, vitamin C and hydroxycinnamic acid derivatives.

Furthermore, this crop has achieved tremendous popularity especially in recent years with the discovery of lycopene's anti oxidative activities and anti-cancer functions *(*Wu *et al.,* 2011).

Thus, tomato production and consumption are constantly increasing. It is noteworthy that tomatoes are not only sold fresh, but also processed as soups, sauces, juices or powder concentrates. The tomato ranks 7th in worldwide production after maize, rice, wheat, potatoes, soybeans and cassava, reaching a worldwide production of around 160 million tons on a cultivated area of almost 4.8 million hectares in 2011 (FAOSTAT, 2011).

Overall the value of exported tomatoes increased by an average 5.8% for all exporting countries since 2015 when tomatoes shipments were valued at $8.5 billion. Year over year, exported tomatoes depreciated by -6.2% from 2018 to 2019. At the continent level, European countries sold the highest value for exported tomatoes during 2019 with shipments amounting to $4.3 billion or 47.6% of worldwide exports. North American exporters were responsible for 31.6% worth, while Asian suppliers supplied a 10.7% share trailed by Africa at 9.4%. Smaller percentages originated from Latin America (0.6%) excluding Mexico but including the Caribbean, then Oceania (0.1%) led by New Zealand and Australia. (exports, 2019) .

I.2 Botanical description

S. lycopersicum is a perennial herbaceous plant, although in temperate climates it is grown as an annual. (Pavan *et al.,* 2009) :

- The growth habit can be erect or prostrate. The root system, fibrous and fasciculate, can reach a depth of 1.5 m, although most roots explore the soil in the first 0.6–0.7 m.
- Leaves (20–30 cm long) are generally compound and pinnate, with 7–11 leaflets (each one up to 8 cm). Some cultivars are characterized by simple leaves (also referred to as potato leaves).
- Both leaves and stem are densely covered by hairs excreting the characteristic tomato smell.
- The flowers, yellow, are 1–3 cm across in full bloom and are brought by an inflorescence which can have different branching patterns. The flowers have individual stamens fused together to form a yellow flask-shaped cone that surrounds the carpels .The flower morphology favours self-pollination.
- The fruit is botanically a berry consisting of seeds within a fleshy pericarp developed from the ovary. The number of carpels in the flowers corresponds to the number of locules in the fruits. Tomato fruitscontain 93–97% of water. The dry matter is composed by sugars (40–60%), proteins and amino acids (15–20%), organic acids (4–10%), minerals (in particular potassium), vitamins and pigments (vitamin A, C, lycopene and b-carotene), insoluble matter (cellulose, emicellulose and pectins).
- Seeds are rough, flattened and discoidal, with 1000 seeds weighing 2.5–3.5 g.

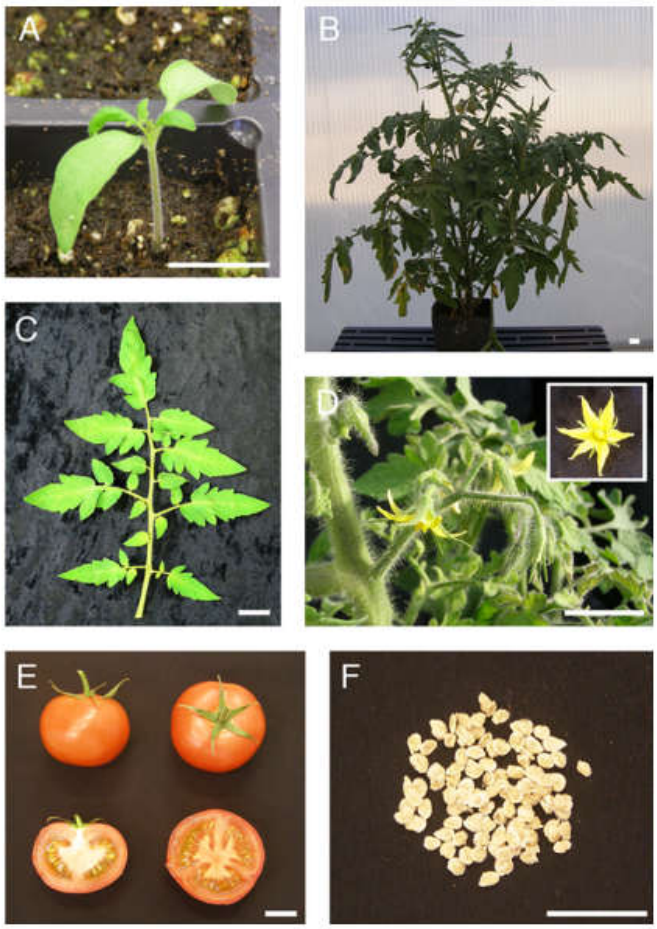

Figure 1: Tomato plants and parts: (A) seedling; (B) 40-d-old plant; (C) leaf; (D) flowers; (E) fruit; (F) seeds. Bar, 2cm *(Sinha et al., 2008)*.

Ⅰ.3 Classification

From botanical point of view, the tomato is a fruit. Nevertheless, it contains a much lower sugar content compared to other fruits. It is a diploid plant with 2n = 24 chromosomes. The tomato belongs to the Solanaceae family, which contains more than 3,000 species, including plants of economic importance such as potatoes, eggplants, tobacco, petunias and peppers (Bai & Lindhout, 2007).

In 1753, Linnaeus placed the tomato in the Solanum genus (alongside with potato) under the specific name S. lycopersicum. In 1754, Philip Miller moved it to its own genus, naming it Lycopersicum esculentum (MR, 2007).

Nevertheless, the designation of the tomato was for a long time a subject of consideration and discussion by many scientists.

- Kingdom : Plantae
- Division :Magnoliophyta
- Class : Magnoliopsida
- Order : Solonales
- Family : Solonaceae
- Genus : Solonum

Ⅱ. Diseases and their casual agent

Table 1: some of tomato diseases and their Casual agents (Ravi Shankar, 2014)

Disease	Casual agent
Buck eye fruit and root rot	Phytophthora capsici
Cercospora leaf mold	Pseudocercospora fuligena
Corcky root rot	Pyrenochaeta lycopersici
Didymella stem rot	Didymella lycopersici
Didymella stem rot	Didymella lycopersici
Bacterial canker	Clavibacter michiganensis pv.Michiganensis
Bacterial speck	Pseudomonas syringae pv. tomato
Bacterial wilt	Ralstonia solanacearum
Pith necrosis	Pseudomonas corrugata
Alternaria stem canker	Alternaria alternata f.sp. lycopersici
Black root rot	Thielaviopsis basicola

Ⅱ.1 Tomato Fusarium Wilt

Ⅱ.1.1 Fusarium genus (Pathogen agent)

The genus Fusarium is one of the most studied of the fungi because of its importance as a plant pathogen, toxin producer and, more recently, as a human pathogen. (Leslie *et al.,* 2005)

Fusarium is a filamentous fungi (Sordariomycetes: Hypocreales: Nectriaceae) containing phytopathogenic and toxigenic species. The genus Fusarium was first described by Link in 1809 as Fusisporium and is presently known as Fusarium (Anjul *et al.,* 2017).

Fusarium species have been important for many years as plant pathogens causing diseases such as crown rot , head blight, and scab on cereal grains; vascular wilts on a wide rangeof horticultural crops ; root rots; cankers; and other diseases such aspokkah-boengas crown rot and bakanae disease of rice ((Booth, 1971) as cited in (PAUL E. *et al.,* 1994)).

The genus Fusarium belongs to the Ascomycota phylum, Ascomycetes class, Hypocreales order, while the teleomorphs of Fusarium species are mostly classified in the genus Gibberella, and for a smaller number of species, Hemanectria and Albonectria genera (Moretti Antonio, 2009).

The genus is highly diverse with twenty monophyletic species complex and outgroups of nine species. Infestation of Fusarium coincides with that of the flowering plants nearly 91.3 million years ago (Anjul *et al.,* 2017)

Fusarium species are widely distributed in soil and on subterranean and aerial plant parts, plant debris, and otherorganic substrates .They are common intropical and temperate regions and are also found in desert,alpine, and arctic areas, where harsh climatic conditions prevail (PAUL E. NELSON, 1994).

Fusarium species are distributed on the plants, in soil and in water either as parasites, endophytes or saprophytes. (Anjul *et al.*, 2017)

Fusarium wilt is caused by a fungus, *Fusarium oxysporum* f. sp. *lycopersici,* that enters the plant through the roots and grows up through the vascular tissue. The fungus destroys cells of the vascular tissue, causing starvation in nearby branches of the plant.

Ⅱ.1.2 Disease development

Disease development is favored by warm temperatures, dry weather, acidic soil and root-knot nematodes. The Fusarium wilt fungus may be introduced to soils in several ways, such as through wind, water, wildlife or equipment. These fungi become established readily in most soils and can remain in the soil for years.

Ⅱ.1.3 Disease symptoms

Symptoms include drooping, yellowing, wilting, and dying of the lower leaves, often on one side of the plant**.**

These symptoms may appear on successively younger leaves with one or more branches being affected and others remaining healthy.

After a few weeks, browning of the vascular system (Figure2) may be observed by slicing the stem open lengthwise with a knife. This brown discoloration inside the stem can be found from the roots to the top of the plant. Plant growth is stunted and, under warm conditions, the plant may die. (WC Nesmith *et al.,* 2014)

Figure 2: Fusarium wilt symptoms include wilt, yellowing, and browning of foliage. Symptoms often occur only on one side of the plant. (Photo: Edward Sikora, Auburn University, Bugwood.org) (WC Nesmith *et al.,* 2014).

Ⅱ.1.4 Tomato Fusarium wilt control

There are three strains (races 1, 2 and 3) of the Fusarium wilt fungus, defined by the host varieties they are able to attack. Regardless of the variety planted, follow good cultural practices for reducing the level of the Fusarium wilt fungus and its effects on the plants.

- Crop rotation of four to five years effectively reduces the inoculum level.
- Clean all equipment, tools and stakes used in an infested field before using in a noninfested field.
- Soil pH between 6.5 and 7.0 and the use of nitrate, rather than ammoniacal, nitrogen fertilizer reduces severity.
- Soil fumigation is an effective control method for commercial growers, and soil solarization has shown benefits for gardeners.

Chapter 2: Medicinal Plants: Agent anti-phytopathogenic “Peganum”

Chapter 2: Medicinal Plants: Agent anti-phytopathogenic "Peganum"

I. Generality

I .1 Medicinal plants

The term of medicinal plants include a various types of plants used in herbalism and some of these plants have a medicinal activities. These medicinal plants consider as a rich resources of ingredients which can be used in drug development and synthesis. Besides that these plants play a critical role in the development of human cultures around the whole world.

Moreover, some plants consider as important source of nutrition and as a result of that these plants recommended for their therapeutic values. These plants include ginger, green tea, walnuts and some others plants. Other plants their derivatives consider as important source for active ingredients which are used in aspirin and toothpaste.

Medicinal plants have a promising future because there are about half million plants around the world, and most of them their medical activities have not investigate yet, and their medical activities could be decisive in the treatment of present or future studies. (Hassan, 2012).

Medicinal plants are traditionally used for a very long time to treat common illnesses and more serious. Their actions come from their chemical compounds: primary and secondary metabolites, and no doubt from the synergy between the various compounds present. For centuries, in Algeria as in all countries of the Maghreb, medicinal and aromatic plants are used mainly in rural areas by the elderly who are still experiencing some herbal tea recipes.

In Algeria, collection of medicinal and aromatic plants to extract, after distillation, essential oils for the manufacture of cosmetics, pharmaceuticals as well as flavors for food products, is a virgin field. The distillation of plants is sufficiently known, but remains largely untapped, despite the availability in Algeria of large tracts of forests and fields, whose territory covers important plant resources distributed on the coasts, plains, mountains, steppes, the Sahara and around water points (Reguieg, 2011).

I.2 Family Zygophyllaceae

Zygophyllaceae, or the bean caper family, is a loose-knit assemblage of 22 genera and 285 species that mainly grow in the desert or saline environments of temperate and tropical

regions. Most members are shrubs to small trees, often resinous, with opposite or spirally arranged leaves. The five-parted flowers typically have 10 anthers, each with a gland, and a well-developed nectary disk. Fruits are commonly capsules or schizocarps.

Ⅱ.Genus Peganum

Ⅱ.1 Botanical description

Peganum harmala commonly known as Syrian rue and Wild rue is a flowering plant and is widely distributed in the Central Asia, North Africa and Middle East. It has also been introduced in America and Australia. This plant is known as "Harmal" in North Africa and "African Rue", "Mexican Rue" or "Turkish Rue" in United States (Madadkar Sobhani A et al., 2002). P. harmalahas been known as "Espand" in Iran (Ramezanloo, 2012).

- *Peganum harmala L*. (2n = 24)is a perennial herbaceous, branched into 5-13 stems (figure 3)
- glabrous plant which grows upto 30- 100 cms in height with a thick rhizome

Figure 3: Peganum harmala plant

- The leaves are palmatisected into 3-5 linear lobes which are 3-6 cms long and 1.5-3.0 mm wide.

Figure 3 : Peganum harmala flower

- Flowers arise by 1-3 on apexes of branches (figure 3) ,rather large (25 to 30 cm) which bear whitish-yellow petals in color, are formed of :
    - Five green sepals, linear, persistent which exceed the corolla.
    - Five elliptical petals.
    - Ten to fifteen stamens with a very broad fillet in their lower part.
    - The ovary, globular, rests on a fleshy disc and results in a fruit which is a spherical capsule, with three cells, 6 to 8 mm depressed at the top, surrounded by persistent sepals and opening by 3 or 4 valves to release seeds.

Figure 4 : Peganum harmala Fruit

- The fruits (figure 4) are globular capsule with 3 chambers 0.9-1.3 cm in diameter and containing 35-45 angular blackish seeds (Nissar Ahmad Khan *et al.,* 2017) . The plant is not usually grazed by animals due to its bitter taste.
- The seeds (figure 5) : numerous, small, angular, subtriangular, dark brown in color, with a reticulate outer seed coat, have a bitter flavor; they are harvested in summer

Figure 5 *:* Peganum harmala seeds

Ⅱ.2 Classification of Peganum

It belongs *to Zygophyllaceae* family in the order of *Zygophyllales* that contains about 22 genera and more than 250 species. Although it belongs to the family *Zygophyllaceae* but its taxanomic position is still debatable and a separate family *Nitrariaceae* has been proposed for this genus (Sheahan, 1996)

According to cronquist 1981 Classification

- Kingdom : Plantae
- Division :Magnoliophyta
- Class : Magnoliopsida
- Order : Sapindales

- Family : Zygophylaceae
- Genus : Peganum

According to APG Ⅲ (2009) Classification

- Kingdom : Plantae
- Division : Angiospermae
- Class : Dicotyledoneae
- Order : Sapindales
- Family: Nitrariaceae
- Genus : Peganum

Ⅱ.3 Distribution

Peganum is a genus of five to six species distributed in the old world from the Mediterranean to Mongolia and in the New world from Texas to Mexico (table 2) (Decreane *et al.,* 1996).

Table 2: World distribution of different species of the genus Peganum (Nida *et al.,* 2014)

Species	Continent	Country
Peganum harmala	Asia	China, India, Afghanistan, Kazakhstan, Kyrgyzstan, Mongolia, Pakistan, Tajikistan, Turkmenistan, Uzbekistan, W. Asia, Iran, Iraq, Syria, Turkey, Jordan, Israel, Greece, Arabia.
	Europe	Russia, S. Europe
	N. America	USA, northern Mexico
	Africa	N. Africa
	Australia	Australia
Peganum multisectum	Asia	China
Peganum nigellestrum	Asia	China, Mongolia
	Europe	Russia
Peganum mexicanum	N. America	USA

Source: Flora of China and (Hooker, 1875) as cited in (Nida *et al.,* 2014)

Ⅱ.4 Pharmaceutical and traditional uses

Ⅱ.4.1 Pharmacological uses

Peganum harmala is a medicinal plant with antimicrobial (Arshad *et al.,* 2008), anti-inflammatory and analgesic properties (Monsef *et al.,* 2004). Carboline alkaloids obtained from various parts of the plant are used against number of diseases (Sobhani *et al.,* 2002).

The seeds of *Peganum harmala* are known to possess hypothermic and hallucinogenic properties and it is used as a medical remedy, incense, spice or condiment with abortifacient, narcotic, aphrodisiac, stimulant, sedative, emmenagogue, and emetic properties, and employed for the treatment of syphilis, fever, hysteria, malaria, neuralgia, parkinsonism, rheumatism, colic, asthma and eye complaint

Many studies have been conducted on the antibacterial, antifungal, antiviral and antitumour effects of *P. harmala* seeds. In Moroccan traditional medicine, seed powder is sometimes used on skin and subcutaneous tumours. (Lamchouri, 2014).

Peganum harmala is one of the most frequently used medicinal plants to treat hypertension and cardiac disease worldwide (Tahraoui *et al.,* 2007).

Ⅱ.4.2 Traditional uses

In traditional medicine, *P. harmala* has been used among societies to treat some nervous system disorders such as Parkinson`s disease (Leporatti ML, 2009), in psychiatric conditions such as nervosity (González *et al.,* 2010) , and to relieve rigorous pain.

Various studies have shown different antiparasidal, antifungal, antibacterial and insecticidal effects of the alkaloids derived from *P. harmala* seeds. It has also been used widely as an anti-fungal and antiparasidal agent in traditional medicine of some parts of the world. For instance, in Saudi Arabia it has been so common to use *P. harmala* against fungal infections. (Milad *et al. ,*. M., 2013).

Ⅱ.5 Toxicity

Overdose ingestion of *Peganum harmala* for medicinal use or as a recreational psychoactive product is toxic and several cases of toxicity have been already reported. It produces paralysis, euphoria, convulsions, hallucinations, digestive problems (nausea, vomiting), bronchodilator, hypothermia and bradycardia (Nida *et al.,* 2014).

I. Biological Activity

Ⅲ.1 Larvacidal activity

- **1st article :**

(Shengxiang *et al.,* 2020)Studied larvicidal activity of essential oils from *Peganum harmala*, and two other medicinal plants against The yellow fever mosquito (Aedesaegypti L., Culicidae).

The aerial parts (leaves and stems) of *P. harmala* and *N. cataria* were collected from Alxa League of Inner Mongolia Autonomous Region, China .Leaves of P. amurense were collected during July 2014, from the suburbs of Changchun city, Jilin Province, China .

Aedesaegypti was originally obtained from the Chinese Center for Disease Control and Prevention. Anhydrous eggs of A. aegypti were hatched in glass trays filled with tap water. Larvae of A. aegypti were further cultivated in tubes on larval food (ground dog biscuits and

yeast tablets in a relationship 1:1, w/w) until the fourth instar stage. The larvicidal activity was evaluated according to the larval susceptibility assay suggested by the World Health Organization .

The fresh plant materials (400 g) of *P. harmala*, *N. cataria* and *P. amurensewere* subjected to hydrodistillation for 6 h using a Clevenger-type apparatus. The obtained essential oils were dried over sodium sulfate anhydrous and stored at 0 C after filtration. Total oil yields were expressed based on dry weight of the plant material.

Table 3: Concentrations required to kill 50% (LC50) and 95% (LC95) of the A. aegypti larvae obtained for the essential oils, thymol, eugenol and chlorpyrifos.

	LC50 (lg/ml)1	LC95 (lg/ml)1	Slope ± SD	Chi square (v2)
P. harmala oil	101.5 (92.4–109.4)	146.8 (134.5–152.9)	2.5 ± 0.26	10.99
N. cataria oil	47.3 (44.0–51.0)	86.8 (81.3–95.2)	1.83 ± 0.17	11.40
P. amurense oil	72.7 (68.0–78.7)	109.4 (103.0–121.4	2.37 ± 0.22	12.63
Thymol	37.1 (34.3–40.0)	54.1 (48.2–59.7)	1.91 ± 0.2	10.02
Eugenol	19.8 (19.4–20.9)	35.3 (33.3–39.8)	1.32 ± 0.13	9.64
Chlorpyrifos	2.1 (2.0–2.3)	6.05 (5.7–6.3)	0.91 ± 0.01	3.29

Table 3 shows the larvicidal activity of the essential oils, the oxygenated monoterpene thymol, the phenylpropanoid eugenol and the organophosphate insecticide chlorpyrifos on the early fourth instar larvae of A. aegypti. The LC50 values of thymol (37.1 mg/ml) and eugenol (19.8 mg/ml) were below those availablein the literature which were informed in the range 46.0–59.8 mg/ml and 60.9–93.3 mg/ml, respectively.

The oil of N. cataria had a strong larvicidal activity which was near to that of thymol and two folds lower than eugenol. P. amurense oil and P.harmala Oil were moderately active.

- **2nd Article**

(Kemassi Abdellah *et al.,* 2013) studied the biological activity of crude leaf essential oil of *Peganum harmala L*. on the larvae L5 and adult individuals of desert locust.

The biological material is made up of fifth stage larvae (L5) and imagos of locusts and leaves of *Peganum harmala L.,* harvested from Oued M'Zab (region of Ghardaïa, Algerian septentrional Sahara). The fifth stage larvae and imagos of locusts experienced result from a

mass breeding maintained in protection ecosystems in arid and semi-arid areas laboratory, University Kasdi Merbah Ouargla.

The leaves of P. harmala subjected to extraction are taken from seedling stage plants, harvested from their natural habitat of existence far places by man. With the help of a simple hydrodistillation assembly, the fresh leaves of *P. harmala* are brought to a boil for 6 hours, decanting is then performed. The resulting product was dried using anhydrous sodium sulphate to remove the little water that may have been retained in the organic phase. The resulting product is a pure essential oil, used for the treatment of insects.

Evaluation of lethal time 50 (LT_{50}) and 90(TL_{90}) for *P. harmala* essential oils on L_5larvae and adults of *S. gregaria*, has to confirm the speed of action of these extracts on L_5 larvae compared to adults.

The LT_{50} reported for the fifth stage larvae being shorter, it is of the order of 06 min 12', as for the assessment for adults is the19 min 21'.

As for the lethal time 90(TL_{90}) evaluated, they are 07 min. 56' and41min 43'for the fifth stage larvae and adults respectively.

The study of the toxicity of essential oils of *Peganum harmala* on the fifth stage larvae and adults of Schistocerca gregaria demonstrate their power to put insecticide on the desert locust.

The neurotoxicity symptoms were reported, then disorders and convulsive movements, inability to perchin the support is also noted in the larvae and adults exposed to essential oils of *P.harmala*, this reflect the neurotoxic effect of its crops extracts on desert locus.

From this perspective, the use of *P.harmala* essential oils against the locusts could be considered. These natural compounds could be a building block for the synthesis of new molecules with particular effectiveness of locusts and without risk of environmental poisoning.

- **3rd Article**

(Dahab, 2019) This experiments were conducted to check out the efficacy of two widely spread plants native to the deserts of the Arabian Peninsula specially Saudi Arabia where it is known locally as Arfag and Harmel against third instar larvae and emerged adults of T. granarium. The experiments were conducted using two different methods at diverse concentrations.

Samples of 50 g of Arfag and Harmel plants powder were extracted with organic solvent (Ethanol 80%) for about 72 hrs, using a soxhlet apparatus. After extraction, the

solvent removed by means of arotary evaporator, yielding the extracted compound. The non soluble portion of the extracted solid remaining in the soxhlet thimble was discarded. The extracts were stored in the refrigerato runder 4°C until needed for bioassay.

The prepared organic solvent (ethanol) were subjected to phytochemical screening to tentatively identify the different chemical groups present in each Ethanoic extract, using thin layer chromatography.

A sample of T. granarium was collected from different stores in the area. The insects were kept separately together with whole rice grains in earthen pots, covered with a muslin cloth and secured with rubber bands. The insects reared to breed under laboratory. To secure stock of larvae for the bioassay tests at room temperature.

The results of testing the insecticidal effects of all ethanolic extracts of the two plants, at each four dosage rates (5, 10 and 20, 40) µg/ml, showed insecticidal activity indicating that the rate of insect mortality increases with an increase in concentrations and exposure time, conversely the number of emerged adults decreased by an increase in concentration.

- **4th Article**

(AL-Hammoshi, 2011) this work aimed to examine the efficacy of *Peganum harmala Linn*. Alkaloids as potential antilieshmanial agents in vitro, and to determine their toxicity in mice.

The study included extraction and isolation of *Peganum harmala* crude alkaloids fromseeds. The isolated fraction that contains alkaloids was detected, using Myer`s and modified Dragendorff`s reagents. Then, the crude alkaloids were tested for their antileishmanialactivity against Leishmania tropica promastigotes in vitro including their effects on parasite growth and metabolism.

It was found that the studied alkaloids inhibited growth of the parasite remarkably. The inhibitory concentration of 50% of the promastigotes (IC50) at the log phase (96) hrs was 50 µg of the alkaloids/ ml of culture.

The extracted alkaloidal fraction from *Peganum harmala* seeds, resulted in decline of RNA, DNA, and protein content of the parasite and reduced specific activity of dihydrofolate reductase and thymidine phosphorelase enzymes. It also had obvious inhibitory effects on energy metabolism of the parasite.

Oral median lethal dose (oral LD50) of the extracted alkaloids was 1070 mg / kg bodyweight in Balb/c mice, using the up-and-down method.

On conclusion *Peganum harmala* alkaloids show promising antilieshmaneal activity and may have potential role in the search for novel antilieshmaneal drugs, as they affect metabolism of proteins, nucleic acids and energy of the parasite (in vitro) with aslight toxicity in mice (in vivo).

Ⅲ.2 Antimicrobial activity

- **5th Article**

(Abdulkareem, 2020) The main objective of this research is to investigate the antimicrobial action of essential oils against grown microorganisms on the surface of acrylic resin denture base materials.

Four types of oils have been applied as Harmal (*Peganum harmala L*), Linseed or flax (*Linumusita tissimum L*), Radish (*Raphanus sativus*), and Black seed (*Nigella sativa*) purchased from Hemani International KEPZ Karachi Pakistan (KEPZ). These oils have been diluted with ethanol to prepare different concentrations of oils (0.25%, 0.5%, 0.75%, 1%, 5%, and 25%).

Test microorganisms are three bacterial types Streptococcus pyogenes, Staphylococcus aureus, and Pseudomonas aeruginosa plus one fungus Candida albicans obtained from the central public health laboratory of Kerbala. Test microorganisms have been cultured on various media, bacteria have been cultured on Muller Hinton Broth (MHB) and Sabouraud Dextrose Broth (SDB) for Candida albicans at 37°C for 24hr.

The results showed that all oils have shown a significant effect on bacteria (Streptococcuspyogenesa, Pseudomonas aeruginosa, and Staphylococcus aureus). Besides, radish oil has shown a significant effect on Candida albicans, contrary black seed, linseed and harmal oil didn't show an effect on Candida albicans.

Therefore, the value of MIC has shown 1%, 1%, 5%, and 0.25% for black seed oil, linseed oil, harmal oil, and radish oil, respectively. However, the MIC of radish oil on Candida albicans is 0.25%.

The result of biofilm counting of *Pseudomonas aeruginosa* and *Candida albicans* growing on denture surface has shown that radish oil is more effective than other oils in

reducing the biofilm formation of microorganism on denture base surface. Black seed oil, linseed oil, and harmal oil have also shown significant effectiveness against *Pseudomonas aeruginosa.*

On conclusion that *Peganum harmala* oil showed an antimicrobial effect on the biofilm which includes three types of bacteria .

- **6th Article**

(Saeide S *et al.,* 2016) The purpose of this study was to examine the evolution of antimicrobial activity of extracts of *Peganum harmala* and *Heracleum persicum* against *Acinetobacter baumannii*.

The seed of *Peganum harmala* and the *Heracleum persicum* leaf were collected in Iran and dried at room temperature. The specimens were ground and stored in a glass container and preserved until used.

Samples were justly dried and ground into fine particles and into a crude powder. In the first step, 10 g of each sample was drenched in 60 ml of 95% ethanol for one day (agitation sporadically with a shaker). Then supplies were strained (Whatman No. 1 filter paper). In the next step, filtrates were condensed with a rotary evaporator. Finally 0.97 g of prepared extracts were acquired and were stored at 4°C in an airtight screw-cap tube.

Table 4: Minimum Inhibitory Concentration of *Peganum harmala* Extract and Essential Oil Against *A. baumannii* (PPM).

Bacterial Code	MIC Extract/ Essential Oil	Bacterial Code	MIC EXTRACT/ Essential Oil
1	6.25/3.1	7	12.5/12.5
2	12.5/12.5	8	6.25/25
3	6.25/12.5	9	6.25/12.5
4	6.25/3.1	10	6.25/6.25
5	6.25/3.1	11	6.25/6.25
6	12.5/6.25	12	6.25/12.5

The results (table 4) showed the levels of MIC extract and essential oil of *Peganum harmala* were observed in ranges from 6.25 ppm to 12.5 ppm and 3.1 ppm to 25 ppm, respectively. The highest MIC value was observed at 12.5 ppm against *A. baumannii* .

Table 5: Minimum Bactericidal Concentration of *P. harmala L* and *H. persicum* Extract and Essential Oil against *A. baumannii* (PPM)

Bacterial Code	**MBC *P. harmala* Extract/Essential Oil**	**MBC *H. persicum* Extract/Essential Oil**
1	12.5/6.25	20/10
2	25/25	20/10
3	12.5/25	10/5
4	12.5/6.25	20/5
5	12.5/6.25	10/2.5
6	25/12.5	20/10
7	25/25	20/10
8	12.5/50	20/5
9	12.5/25	20/20
10	12.5/6.25	20/10
11	12.5/12.5	10/5
12	12.5/25	20/20

The levels of MBC extract and essential oil of *P. harmala* (table 5) were observed in ranges from 12.5 ppm to 25 ppm and 6.25 ppm to 50 ppm, respectively. The highest MBC values for extract and essential oil of *P. harmala* were observed at 25 ppm and 50 ppm respectively .

On onclusion of the results of this study we can suggest that the extract of *Peganum harmala* may be useful to treat bacterial infections.

- **7th Article**

(Ida Apostolico *et al.,* 2016) this study test the antimicrobial activity of the essential oil against some bacterial strains: *Staphylococcus aureus* (DSM 25693), *Bacillus cereus* (DSM 4313), *Bacillus cereus* (DSM4384), *Escherichia coli* (DMS 857) and *Pseudomonas aeruginosa* (ATCC 50071). All the oils showed different inhibitory activity. And were

evaluated for their possible in vitro phytotoxic activity against germination and initial radicle growth of *Raphanus sativus L., Lepidium sativum L., and Ruta graveolens L.*

The seeds of *P. harmala* were collected from Tunisia (region of Kasserine-Bouzguem), Algeria (South of Algeria-Bousaada), Libya (region of El Hisha-South Derna), Morocco (Marrakech-Haouz) and Egypt (region of Marsa-Mattrouh).

One hundred grams of dried seeds of each sample were ground in a Waring blender and then, subjected to hydrodistillation for 3 h according to the standard procedure described in the European Pharmacopoeia.

Using the agar diffusion test, we evaluated the potential antimicrobial activity of the EOs obtained from *P. harmala* grown in the five countries of the Mediterranean area. All samples were capable of inhibiting the growth of the bacteria used as tester strains

Escherichia coli is more sensitive to all oils, especially at a concentration of 15 µg/mL, with an inhibition area of 10.0 mm using the Egyptian oil. This value was higher than that shown by the control tetracycline against the same microorganism (6.0 ± 0.5 mm). The essential oil from Egypt showed quite similar effects against Bacillus cereus DSM 4384 at the same concentration. However, Staphylococcus aureus is more sensitive to essential oil from Morocco. Bacillus cereus DSM 4313 was the least sensitive to lower concentrations, but still shows a zone of inhibition of 9 ± 0 mm when the tests were effectuate with the essential oil from Libya at 15 µg/mL.

The five essential oils were evaluated for their activity against germination and radicle elongation of radish (*Raphanus sativus L.*)—a species frequently utilized in biological assays—of garden cress (*Lepidium sativum L.*), and rue (*Ruta graveolens L.).* The five oils seem to be effective against germination of all these species . In particular, treatment of seeds with concentrations of 100 µg/mL of the oil from Algeria is the most active against germination of radish. The concentration of 25 µg/mL of essential oils from Libya and concentrations of 50 µg/mL and of 25 µg/mL of essential oil from Tunisia inhibited significantly the germination of garden cress.

The essential oils from Algeria, Egypt and Libya, at all doses tested, significantly inhibited the radicle elongation of R.graveolens. However, concentrations of 25 µg/mL and 12.5 µg/mL of the oil from Tunisia are active against the germination of seeds oh the same

species. The five essential oils affected significantly, at all doses tested, the radicle elongation in R.graveolens.

As a conclusion the studied samples showed antimicrobial activity agains tboth Gram-positive and Gram-negative microorganisms, although it should be impossible to attribute such activity to a specific compound or group of biomolecules. However, the antimicrobial activity registered confirms some of the traditional uses of the plant. The essential oils studied showed different phytotoxicity, namely in the inhibition of radical growth of the three test species studied.

- **8th Article**

(Milad *et al.* ,. F., 2019) in this study smoke and extract of total alkaloids were investigated for antimicrobial activity against five different microorganisms (standards and hospital isolates). The antibacterial activity was evaluated using disc diffusion assay and minimum inhibitory concentration (MIC) was determined by serial dilution methods.

For the preparation of alchoholic extract of alkaloids the seeds of the plant were washed with petroleum ether, dried and ground into crude powder. In the first step, 100-150 g of dried powder was macerated in 1 L of 95% methanol for 12 hr at 50°C in water bath. After evaporating the solvent (i.e. methanol), the residue was dissolved in HCl (5%) until the pH of the final solution became 1, then the solution was filtered. In the next step, the filtrate was extracted twice by 30 ml petroleum ether to remove highly lipophilic compounds. The residue was basified by 10% NH4OH (pH 9-10) and extracted by chloroform (30 ml); finally, the chloroform was evaporated to dryness.

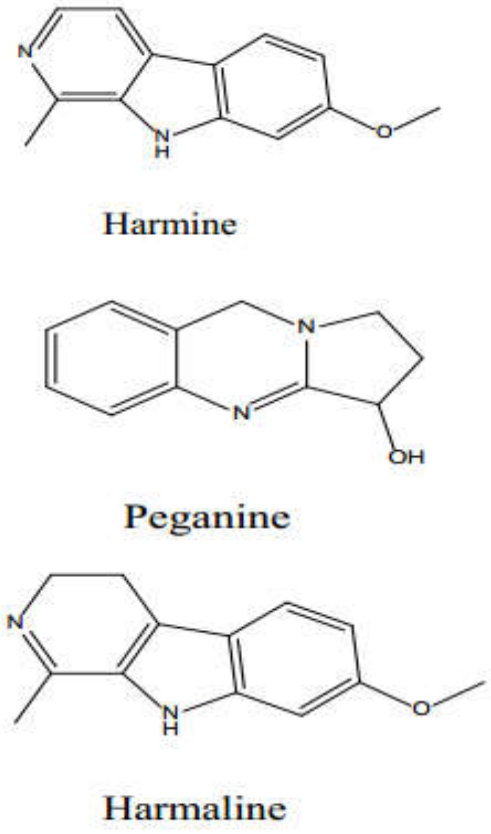

Figure 6: Chemical structures of alkaloids isolated from *P.harmala* .
(Milad *et al.* ,. F., 2019)

The smoke of burned seeds of *P.harmala* or formalin tablet in a watch glass was directed to a flask containing Mueller Hinton broth (150 ml) through a tube. One ml of each microbial suspension (equivalent to 0.5 ml McFarland standard, i.e. 108 CFU/ml) was added to the medium. Incubation of the flask was performed for 24 hr, at 37oC for bacteria and for 48 hr, 25oC for yeast and then, the growth of Microorganisms were observed. All the steps were performed under aseptic conditions.

Table 6: MICs (µg/ml) of total alkaloid extract, clotrimazole and amikacin against standard and pathogen microorganisms.

Microorganism	Standard (total alkaloid)	Pathogen (total alkaloid)	Standard and pathogen (clotrimazole)	Standard (Amikacin)	Pathogen (Amikacin)
Staphylococcus aureus	125±2.04	150±4.08	-	8±0.163	15±0.408
Escherichia coli	500±8.160	1500±16.32	-	4±0.408	10±0.816
Pseudomonas aeruginosa	1500±40.820	2000±40.82	-	4±0.408	50±1.632
Candida albicans	62.5±2.04	62.5±2.04	8±0.204	-	-
Micrococcus luteus	31.25±1.650	-	4±0.408	-	-

Table 7: Antimicrobial activity (equivalent of g seeds) of smoke against standard and pathogenic microorganisms.

Microorganism (standard)	Standard (smoke)	Pathogen (smoke)	Standard (Formalin tablets)	Pathogen (Formalin tablets)
Staphylococcus aureus	4±0.082	5±0.163	2±0.204	2±0.204
Escherichia coli	1±0.082	2±0.224	0.5±0.144	1.5±0.204
Pseudomonas aeruginosa	6±0.122	8±0.408	0.5±0.144	2±0.204
Candida albicans	1±0.04	1±0.04	1±0.163	1±0.163
Micrococcus luteus	2.25±0.04	0.5±0.144	-	-

As a final result of this study ,Our results showed that the alkaloids (table 6) and smoke (table 7) were specifically more effective on Candida albicans and Gram- positive bacteria (Micrococcus luteus and Staphylococcus aureus), while Gram- negative bacteria, especially Pseudomonas aeruginosa, were less sensitive.

- **9th Article**

(Jasim, 2019) this study test the Antibacterial, muscle relaxant, and hypnotic effects of seeds of *Peganum harmala* on mice.

Dry seeds of *P. harmala* were grounded in coffee machines for 2-3 min. The powder was mixed with sufficient amount of distilled water, and shaken overnight at room temperature. The mixture was filtered, and the solvent was removed by incubation at 37°C. Distilled water was used to dissolve the dried residue to give the required concentration.

The Gram positive bacterium was Staphylococcus aureus and Gram negative was Escherichia coli.

Three or four isolated colonies were inoculated in the 2 ml nutrient broth and incubated till the growth in the broth was equivalent to Mac-Farland standard (0.5%).

Kirby- Bouar method was performed by Muller Hinton agar (Oxoid) poured on disposable plates. Holes of 5 mm in diameter were made after solidification of the agar. E. coli and S. aureus were uniformly distributed on the surface of the agar. 0.4 ml of 5 and 10 mg/ml of *P. harmala* extract were placed in the holes. The plates were incubated at 37°C, and examined after 24 h for the presence of growth inhibition zones. Ampicillin of 10 mg was used as positive control, while distilled water was used as negative control.

Results revealed that the extract of *P. harmala* (10 mg /ml) exhibited border spectrum as well as greater activity against E. coli and S. aureus with inhibition zone varying between 2-15 mm . The water extract of *P. harmala* exerted antibacterial activity against E. coli at 10 mg/ml; the mean of the inhibitory zone is 3-12 mm at 20 mg/ml. It exerted also antibacterial activity against S. aureus.

The mean of the inhibitory zone is 2-9 mm.Table 2 shows the minimal inhibitory concentration of *P.harmala* against S. aureus.

All animals injected with the 100mg/kg.b.w of aqueous extract of seeds of *P. harmala* showed myorelaxation or incoordination; so all the animals dropped down from the wire 3 consecutive timesin 60 s.

P. harmala prolonged the sleeping effect of pentobarbitol sodium.Aqueous extract of P. harmala also induced muscle relaxation and prolonged the sleeping time induced by pentoparpitol.

These data suggest that *P.harmala* extract could inhibit the growth of S. aureus and E.coli strain in vitro and this activity may contribute to its chemopreventive effect.

Ⅲ.3 Antifungal activity

- **10th Article**

(N. Amezian *et al.,* 2007)This study shows Antifungal activity of Moroccan plants against citrus fruit pathogens , among the plnats *Peganum harmala* powder Essential oil and extract.

Twenty-one fresh plant samples were collected from the Souss valley (Agadir, Morocco) in 2003 .

Different parts of the plants were used (stem, leaves, flowers and seeds). Tests were carried out with powders and essential oils, as well as methanolic and chloroformic extracts.

The fungi used in this study, *Penicillium digitatum*, *Penicillium italicum* and *Geotrichum candidum*, were isolated from decayed citrus fruits. The fungi were maintained on Potato Dextrose Agar plates at 5 ◦C, with periodic transfers through citrus fruit to maintain the aggressiveness of the pathogen. A one-week-old culture of each fungus was used to inoculate the agar plates.

The powder of *Peganum harmala* completely inhibited the mycelial growth of the three pathogens.

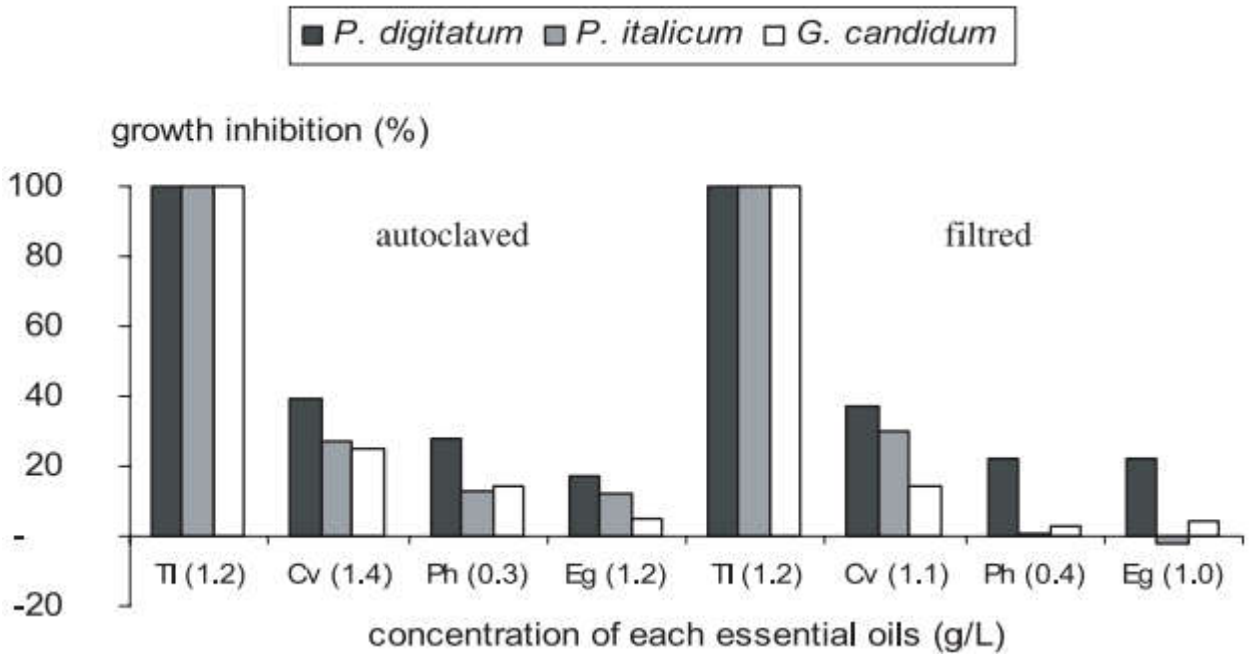

Figure 7 : Effect of essential oils from *Thymus leptobotrys* (Tl), *Cistus villosus* (Cv), *Peganum harmala* (Ph) and *Eucalayptus globulus*(Eg) on mycelial growth of three pathogens using two methods of sterilization.

Peganum harmala Essential oil showed (figure 7) a growth inhibition less than 40% on the three fungal pathogens.

Chloroformic and methanolic extracts of *P. harmala* tested at a concentration of 1% and 2% (w/v), respectively, exhibited a pronounced activity against the three pathogens.

- **11th Article**

(A. Sarpeleh *et al.,* 2009) this study demonstrated the antifungal activity of *P. harmala* extracts on phytopathogenic fungi which can be used as an alternative for chemical compounds.

The effect of water soluble extracts obtained from *P. harmala* on different fungi. There were highly significant differences (P d 0.01) among fungal species, extracts and interaction between mycelia growth of the fungi (A) and the extracts (B). Based on their mycelial growth, fungal species were grouped into 10 classes.

Maximum decrease occurred in the mycelial growth of V. Dahliae, while no inhibitory effect was detected for R. solani .There was a significant difference in the antifungal activity of the extracts taken from different parts of the *P. harmala* plants.

The maximum inhibitory effect was induced by seed extracts followed by the extracts taken from floral tissues and leaves, respectively .
The effect of individual extracts (leaves, floral tissues and seeds) on each of the fungal species (extract u fungi interaction) .

All extracts inhibited significantly the fungal growth (except for R. solani) with the maximum inhibitory effect being detected for seed extracts . Decreasein the mycelial growth of most of the isolates was obvious while P. drechsleri and V. dahliae did not show any growth in the culture media amended with seed extracts.

This study shows that the extracts of *P. harmala* plants have antifungal activity. Additionally,
Iranian traditional growers experienced that *P. harmala* has a great impact on yield as well as in the decrease of soil-borne diseases of melons when the *P. harmala* plants are buried 2–3 months prior to cultivation. The present study shows that aqueous extracts of *P. harmala* inhibit the growth of many agronomically important phytopatho genic fungi. This indicates the potential application of *P. harmala* extracts as an alternative or integrated method to chemical control of plant pathogenic fungi.

Ⅲ.4 Anti-inflammatory activity

- **12th Article**

(Pradeep Kumar *et al.,* 2015) The objective of this research work is to carry out the phytochemical screening and evaluate the analgesic, anti-inflammatory activities of *Peganum harmala Linn.* seeds.

In this study different extracts of *Peganum harmala (Linn)* seeds were evaluated for analgesic and anti-inflammatory activities using glacial acetic acid induced writhing and carrageen an induced rat paw edema models respectively.

Phytochemical screening revealed the presence of alkaloids, flavonoids in the alcoholic extract and alkaloids, flavonoids and steroids in the ethyl acetate extract.

Hence, the presence of alkaloids, flavonoids and steroids in the ethyl acetate extract could be attributed for observed significant analgesic and anti- inflammatory activities. However, research work is under progress to confirm the exact mechanism of action and to elucidate the structure of bioactive principle for the claimed analgesic and anti-inflammatory activities.

The present study provides an evidence for the analgesic and anti-inflammatory activities of *Peganum harmala (Linn)* seeds. Aspirin and diclofenac were used as standard drugs for screening the analgesic and anti-inflammatory activities respectively.

Aspirin used as standard drug for analgesic activity acts by obtunding of peripheral pain receptors and prevention of PG- mediated sensitization of nerve endings and the diclofenac used as standard drug for screening the anti-inflammatory activity act by inhibiting the prostaglandin synthesis and specially it is COX-2 selective .

Ⅲ.5 Antioxidant activity

- **13th Article**

(Abolhasani *et al.,* 2015) This study occurs around a test of antioxidant capacity and stability of Phenolic compounds from the seeds of *Peganum harmala*.

The antioxidant properties and total phenolic compounds extracted from *Peganum harmala* using water, ethanol and ethanol - water was evaluated. The highest amount of phenolic compounds from water extract, ethanol – water and then ethanol was obtained. Antioxidant activity of extracts tests to trap free radicals DPPH & β-carotene are investigated and compared with the synthetic TBHQ antioxidant.

Among the extracts, the highest amount of phenolic compounds extracted from the water extract and ethanol-water and ethanol extracts, respectively were the next steps.

DPPH radical scavenging test indicate that the water and alcoholic extracts of the highest DPPH radical scavenging power are and almost in a statistical range and are generally DPPH radical scavenging power when we compare these extracts with TBHQ, We see that the extracts in DPPH radical disable located at a much higher level.

The high antioxidant activity of beta-carotene antioxidants in model systems is the fact that phenolic compounds in the extracts can react with radicals generated as a result of lipid oxidation of lipids in cell membranes, resulting in injury to prevent oxidative degradation of lipids is inhibited.

Based on these results, we can say harmala seed due to a large amount of phenolic compounds with high antioxidant potential and hence can as a rich source of natural antioxidants in the manufacture of various products such as jam, marmalade, pickles and other food products in which the presence of lipid oxidation is likely to be used effectively.

Ⅲ.6 cardiovascular activity

- **14th Article**

(Rajkumar *et al.,* 2018)The present study was undertaken to find out the effects of the active principle extracted from the seeds of *Peganum harmala* on isolated frog heart muscle and frog rectus abdominus muscle preparations and the probable mechanism of action.

Peganum harmala seeds were crushed and treated with dilute 3% acetic acid and pressed after 48 hours. To this liquid, sodium chloride was added and kept in the refrigerator for a day, to isolate the harman alkaloids, which are precipitated as yellow material. This is dried in air to get yellow powder, which his used in experiments on isolated frog heart and frog rectus abdominus muscle.

Using the isolated frog heart, normal heart rate and cardiac output are noted and the responses are recorded on a rotating smoked drum on a kymograph.

Using the isolated frog rectus abdominus muscle, the responses to Harman alkaloids and their effect on frog rectus.

In this study, Harman alkaloids isolated from *Peganum harmala* seeds were shown to inhibit the normal contractions of isolated frog heart and the inotropic actions of Adrenaline, 5-Hydroxytryptamine and calcium. These inhibitions were concentration dependent and reversible. Harman alkaloids have not shown any effects on frog rectus abdominus muscle on their own and they have also not shown any effect on the contractions of frog rectus abdominus muscle induced by Acetyl-choline.

As a conclusion It seems that the Harman alkaloids isolated from *Peganum harmala* seeds affect the cardiac muscle contractions by blocking the calcium influx into the cell via voltage dependent calcium channels and receptor operated calcium channels. They do not affect the skeletal muscle contractions as it is dependent on sodium and potassium channels and calcium is released from sarcoplasmic reticulum inside the cells in response to depolarisation.

Ⅲ.7 Immune activity

- **15th Article**

(Toghyani *et al.,* 2015) The present study was carried out to evaluate the effect of different levels of dietary seed and extract of Harmal (*Peganum harmala L.*) on immunity of broiler chicks.

A total of 350 day-old Ross 308 broiler chicks were used in this experiment during a 42 d feeding trial including 1 to 14 d (starter period), 14 to 28 d (grower period) and 28 to 42 d (finisher period).

The chicks were randomly assigned into five different dietary treatments with four replicate pens. Dietary treatments consisted of control, 1 and 2 g/kg Harmal seed in diet, 100 and 200 mg/L Harmal seed extract (methalonic extract) in drinking water .

For the preparation of extract, one kg of *Peganum harmala* seeds were dipped in 3 liters of 80% aquous methanol for five days, filtered and then methanol was evaporated using rotary evaporator under low pressure. At d 42 of experiment, two birds from each replicate were selected randomly to evaluate the relative weights of Bursa of Fabricius and spleen as lymphoid organs were precisely removed and weighed and expressed as percentage of live body weight.

Antibody titers against Newcastle and influenza viruses and sheep red blood cell were measured at 30 d of age. Results showed that the relative weights of lymphoid organs were not affected by dietary treatments. Furthermore, antibody titer against Newcastle and influenza viruses as well as sheep red blood cell antigen were significantly ($P<0.05$) enhanced by feeding Harmal seed and extract.

In conclusion, the results indicated that dietary inclusion of Harmal seed and extract enhanced immunological responses in broiler chicks.

Conclusion

Conclusion

With the increasing concerns of pesticide residues in agricultural products and environment as well as the incidence of resistance in plant pathogens against chemical pesticides, the use of non-chemical methods including natural metabolite have assumed greater significance.

Natural plant products are one of the most important alternatives which do not have indiscriminate hazardous effects like synthetic products

This Work carried out as part of a reaserch on the natural products that can prevent or cure diseases through their Fungicidal, bactericidal and larvicidal effects

As our work was focused on many biological activities that the genus of Peganum showed with the precedent works that gives many results concerning Pharmaceutique and therapeutique effects on curing or preventing nemourous human diseases.

From several past decades the search for plant extracts and essential oils in an increasing scale. Plant extracts, such as essential oils (EOs), have been known for centuries for their ability to prevent and/or to cure diseases through their fungicidal and bactericidal effect.

And this goes as well with *Peganum harmala* different extracts, Essential oil, powder and smoke that showed a significant antibacterial ,anti-inflamatory, antifungus, antimicrobial activity , on which we can conclude that this species is a solution for many human diseases and different phytopathogens and expansion of our research in more domains in the future.

P.harmala showed an antifungal activity againt different fungus this allows to continue the research on the antifungal activity against Fusarium wilt in inhibiting the mycelia growth of Fusarium oxysporum.

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Annexes

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الملخص

تعتبر المنتجات النباتية الطبيعية من أهم البدائل التي ليس لها تأثيرات خطرة عشوائية مثل المنتجات الاصطناعية، مع الانتشار السريع لمسببات الأمراض النباتية في عالم النباتات ، تم التركيز على العوامل المسببة للأمراض النباتية وفي هذا التوليف ركزنا على تقييم الزيت الأساسي و مستخلصات نبتة الحرمل و تأثيرها البيولوجي في علاج العديد من الأمراض أو الحد منها ، حيث تم اختباره على النشاط المضاد للميكروبات ، النشاط المضاد للفطريات ، نشاط مبيدات اليرقات والنشاط المضاد للأكسدة. من هذه الأبحاث والنتائج يمكن أن يكون لدينا فرضية تعلن عن إجراء إعادة بحث في المستقبل حول النشاط المضاد للفطريات لزيت الحرمل الأساسي في السيطرة على مرض ذبول الطماطم بفطر الفوزاريوم. و استخدامه كمبيد للفطريات الحيوية.

الكلمات المفتاحية: الحرمل، النشاط البيولوجي، مسببات الامراض النباتية للطماطم ، النشاط المضاد للفطريات، مرض فطر الفوزاريوم.

Abstract

Natural plant products are one of the most important alternatives which do not have indiscriminate hazardous effects like synthetic products , with the fast spreading of phytopathogens in plants world, reaserches has been focused on phytopathogenic agents and in this synthesis we focused on the valuation of *Peganum harmala* Essential oil and extracts on Biological activity in treating many diseases or reducing it , as was tested on antimicrobial activity, antifungal activity, larvacidal activity and antioxidant activity. from these researchs and result we can have an hypothesis declaring to take a future reaserchs on the antifungul activity of *Peganum harmala* EO in controlling Tomato fusarium wilt disease. Using *P. harmala* EO as a biofungicide .

Key words : *Peganum harmala L.,* biological activity , Tomato phytopathogens , EO ,antifungal activity, fusarium wilt.

Résumé

Les produits végétaux naturels sont l'une des alternatives les plus importantes qui n'ont pas d'effets dangereux aveugles comme les produits synthétiques, avec la propagation rapide des phytopathogènes dans le monde des plantes, les recherches se sont concentrées sur les agents phytopathogènes et dans cette synthèse, nous nous sommes concentrés sur l'évaluation de Peganum harmala Huile essentielle et extraits sur l'activité biologique pour traiter de nombreuses maladies ou les réduire, comme cela a été testé sur l'activité antimicrobienne, l'activité antifongique, l'activité larvacide et l'activité antioxydante .A partir de ces recherches et de leurs résultats, nous pouvons avoir une hypothèse déclarant de prendre une future étude sur l'activité antifongique de Peganum harmala OE dans le contrôle de la maladie de la flétrissure fusarienne de la tomate. Utilisation de P. harmala EO comme biofongicide.

Mots clés : Peganum harmala , activity biologique , phytopathogenes de tomato , HE, activity antifangique, fusarium wilt.