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Plant Extracts from Mexican Native Species: An Alternative for Control of Plant Pathogens

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1. Introduction

Currently, control of plant pathogens requires employment of alternative techniques because traditional handling with synthetic chemicals has been caused various problems such as toxicity to users (Whalen et al., 2003) and impairment of beneficial organisms (Anderson et al., 2003). Another important aspect is that pathogenic organisms have generated resistance to the active ingredient of some synthetic fungicides in response to selection pressure due to high dose and continuous applications, causing great economic losses (Cooke et al., 2003; Leroux, 2003). An economical and efficient alternative for disease control is the use of natural products derived from plants (secondary metabolites) (Wilson et al., 1999), since it does not affect environment and their residues are easy to degrade. On the other hand, vegetal biodiversity in Mexico is available to be exploited, especially by their natural non-toxic, biodegradable compounds (Hernandez et al., 2007). The potential use of plant extracts to control plant pathogens has been reported in different laboratory (Hernández et al., 2010; Castillo et al., 2010; Jasso de Rodríguez et al., 2007; Lira et al., 2003; Osorio et al., 2010), greenhouse (Bergeron et al., 1995) and field studies (Hernandez et al., 2006, 2008). Based on the popular uses of plants in the Coahuila Southern region, botanical resources can be identifying by their antimicrobial potential against plant pathogens. However, still there is a lack of research in this field. As previously mentioned, there is a need for new plant disease management options with lower environmental and economic impact, which expresses a similar or greater effect in controlling pathogens. Mexico is one of the five countries with major biodiversity. It owns 10% of the world diversity with numerous endemic plant species. The arid and semi-arid areas cover 1,028,055 km², distributed in 19 states with less than 350 mm of rainfall per year, or 350-600 mm of annual rainfall, respectively. In Mexico, the Chihuahuan Desert is one of the most biologically rich deserts of the world. It covers an approximate area of 630,000 km², spanning the states of Chihuahua, Coahuila, Nuevo Leon, Durango, Zacatecas and San Luis Potosí, to until the Southwestern United States, corresponding to Arizona, New Mexico and Texas States (Fig. 1). In this desert are predominantly brushwood and grassland, among species highlighting the creosote bush (Larrea tridentata) (Jasso & Rodriguez, 2007).



Fig. 1. Location of the Chihuahuan Desert.

2. Control of plant pathogens using plant extracts

2.1 Bio-fungicide potential of chemical groups present in plant extracts

The botanical bio pesticides represent an alternative for pest control with low environmental impact and high food safety. Several products derived from plants have shown an antimicrobial effect. Among the main compounds present in these extracts are: flavonoids, phenols, terpenes, essential oils, alkaloids, lectins and polypeptides. Some plant extracts containing these metabolites has been extracted in water or other solvents, depending on its polarity, and in powder form (Bautista et al., 2003). The enormous diversity of secondary metabolites and biological properties present in plants, are still subject of study. The limited knowledge that currently exist about plant extracts, is an interesting point to begin studies with plants of almost any kind. However, there are some chemical derived from the knowledge base of the plants (Montes, 1996). Some families of plants may be more feasible for study, such as: Solanaceae for its high alkaloid content, or Mimosaceae that's present species rich in tannins, or *Lamiaceae* and *Meliaceae* because their wide diversity of terpenoids. For production of active ingredients, there are factors that determine variability in quality and quantity of metabolites. A plant may have different concentrations of a chemical in different vegetal parts: roots, leaves, flowers and fruit and may even be absent in one or more parts, so it is convenient to collect integral samples (Montes, 1996) and also, knowing thee chemical content of plants used in a given region, either as an insecticide, fungicide, nematicide, among others.

2.2 Effectiveness of phytochemicals of native species against plant pathogens

In the context of this problem, one of our research projects has as objective to study the potential of phytochemicals from the Chihuahuan semidesert plant biodiversity as an alternative to control plant diseases caused by fungus and bacteria. The results have shown the added value of the Mexican botanical diversity because the wide range of potential applications of their resources that is geared mainly towards the collection of wild plants for the extraction and marketing of raw material; good examples are candelilla wax and hard fibers of yucca. In some cases, these raw materials are exported to countries where they are purified and transformed into finished products. A new use of these plants promises to change the structure and concept of these sources to become active materials for disease

control. In the Table 1 is shown a listing of various plant species that we have studied against different species of plant pathogens with different habits of attack and producing different plant symptoms as root rots, leaf blights, anthracnose, fruit rot, rot grains and seeds, food molds, etc. as well as solvents used in the extraction of phytochemicals.

Source of plant extract	Plant pathogen	Solvents		
Flourensia cernua	Rhizoctonia solani	Methanol		
F. microphylla	Phytophthora infestans	Chloroform		
F. retinophylla	Alternaria sp	Hexane		
Origanum majorana	Fusarium oxysporum	Diethyl		
Bouvardia ternifolia	Colletotrichum coccodes	Ethanol		
Aloe vera	Colletotrichum gloeosporoides	Water		
Larrea tridenta	<i>Pythium</i> sp.	Lanolin		
Agave lechuguilla	Botritys cinerea	Cocoa butter		
Yucca filifera	Alternaria alternata			
Opuntia ficus-indica	Alternaria dauci			
Lippia graveolens	Penicillium digitatum			
Carya illinoensis	Phytophthora cinnamomi			
	Colletotrichum truncatum			
	Fusarium verticillioides			
	Fusarium solani			
	Fusarium sambucinum			
	Clavibacter michiganensis subsp. michiganensis			
	Clavibacter michiganensis subsp. nebraskensis			

Table 1. Chihuahua semi-desert native plant species studied to determine their effect against various plant pathogens and solvents used for phytochemicals extraction.

The dose-response analysis of extracts from semi-desert plant species indicate that these phytochemicals have anti fungal and anti bacterial properties because most of the extracts inhibited mycelium or bacterial growth of various pathogens studied. This effect varies according to plant species and solvent used for phytochemicals extraction (Gamboa *et al.*, 2003b; Hernández *et al.*, 2006, 2008, 2010; Castillo *et al.*, 2010).

2.3 Factors involved in phytochemicals recovery from Chihuahuan semi-desert native plant species

The diversity of compounds in plant tissues is extensive from the chemical point of view, which has resulted in the identification of thousands of these compounds, with unknown biological functions, which represents a challenge in the search for compounds that are highly efficient for crop plant diseases control. The main groups of secondary metabolites present in plant tissues are polyphenols, terpenes and nitrogen compounds. Tannins (polyphenols) are phytochemicals widely distributed in the plant kingdom, found in roots,

leaves, seeds and fruits. These compounds are secondary metabolites with a molecular weight range of 300-20 000 D (Gonzalez *et al.*, 2009). The tannins are non nitrogen compounds, flavor astringent, amorphous, mostly soluble in water and alcohol (Medina *et al* 2007). Its hydrophilic nature allows them associate with carbohydrates, alkaloids and proteins by hydrogen bonds, covalent bonds and hydrophobic interactions. The tannins are divided according to their sugar content, polymerization and esterification in hydrolysable tannins, condensed tannins and complex tannins (Fig. 2).

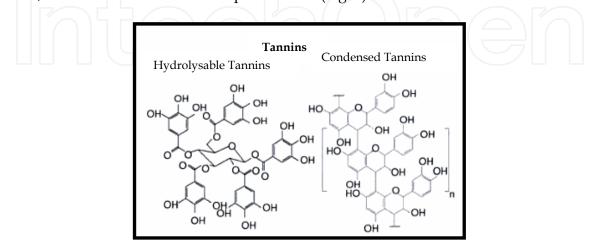


Fig. 2. Chemical structures of hydrolysable and condensed tannins

Hydrolysable tannins. These compounds are esters formed by a molecule of sugar (usually glucose) attached to a variable number of molecules of phenolic acids (gallic acid or its dimer, ellagic acid). By hydrolysis with acids, bases and hydrolytic enzymes may be broken the glycosidic bond to liberate the sugar and phenolic compounds. They have a central core consisting of a sugar or a shikimic acid analogue, which is esterified by gallic acid units or related compounds (C6-C1) (Mata *et al.*, 2008).

Condensed tannins. Proanthocyanidins are polymers or oligomers formed by covalent flavan-3ol (group of catechins) and flavanes4-ol (group of leucoantocianidinas). The term proanthocyanidins (PAS), is derived from the oxidation reaction that produces anthocyanidins (ACS) red in acid-alcohol solutions. Two abundant types of proanthocyanidins, which are formed by the condensation of catechin and epicatechin units that relate to cyanidin and are known as procyanidins and those whose monomeric units are gallocatechin and epigallocatechin, delphydinin resemble and are known as prodelphynidins. They have a structure similar to that of flavonoids. These substances are not hydrolysable by acids or enzymes. Strong acids, heat or oxidizing agents make them red or dark substances, insoluble in most solvents (Ramirez *et al.*, 2008).

Exploration of phytochemicals from native species has allowed recovery of them in form of resin using various solvents as indicated in Table 2. In most cases, methanol, chloroform and acetone are the most commonly used solvents for phytochemicals extraction. However, the use of non-conventional solvents allowed recovery of polyphenols (PT), equivalent to gallic acid and catechin with highly variable recovery rates. There are several factors involved in production and recovery of metabolites, such as the solvent used in the extraction process, for example in Table 2, is showed the amount of resin recovered from the same plant specie in relation to the solvent used. The solvent efficiency for most metabolites recovery is associated with its polarity, so it is important to perform studies about the specific implications of the solvent for high phytochemicals recovery.

Solvent	Fresh Leaf weight (g)	Dry weight of resin (g)	Recovery (%)
Methanol : Chloroform	460	87.8	19.1
Hexane	460	26.6	5.8
Diethyl	460	44.2	9.6
Ethanol	460	37.8	8.2

Table 2. Recovery of resin from *Flourensia cernua* leaves using four different solvents. Guerrero *et al.*, 2007.

In addition to solvents, there are other factors involved in the recovery efficiency of phytochemicals, such as environmental conditions during the plant growing season, in Fig. 3 it is showed that there is a differential in phytochemicals production as a consequence of locality or place of plant growth. These results refer that the ecological conditions where shrubs grown have an effect on the phytochemical compound characteristics and on their antifungal action, one last factor is the selection of plant species. In Table 3 are presented differences on phytochemicals production by different plant species, which could be correlated with the content of secondary compounds in their tissues.

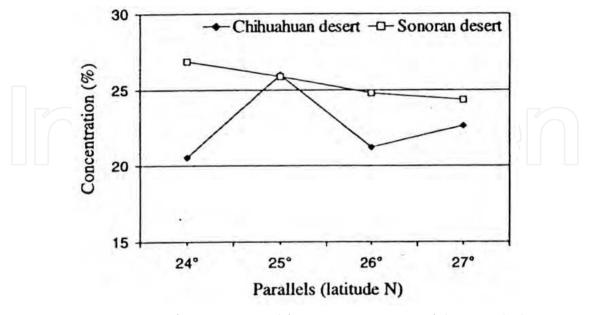


Fig. 3. Mean concentration of resin extracted from *Larrea tridentata* foliage with three different solvents. *Larrea tridentata* foliage was collected from four sampling locations in the Sonoran and Chihuahuan desert. Lira *et al.*, 2002.

Source of plant extract	Hydrolysable Tannins (mg/g)	Condensed Tannins (mg/g)	Total tannins
Larrea tridentata	17.3769 a	37.153 a	54.530 a
Carya illinoensis	1.0821 d	31.980 b	33.062 b
Opuntia ficus indica	1.1573 d	29.020 b	30.177 b
Agave lechuguilla	4.2952 c	22.629 c	26.924 с
Lippia graveolens	5.7996 b	19.090 d	24.889 c
Yucca filifera	0.7953 d	12.005 e	12.801 d
Flourensia cernua	4.7652 c	4.854 f	9.619 e

Table 3. Total tannins content in seven species from the Mexican plant semi-desert area. Means with the same letter are not statistically different according to the Tukey multiple range test (P < 0.05).

2.4 Plant extracts effect on microorganism's growth inhibition

Most plant pathogens have mycelia growth inhibition by effect of polyphenolic compounds and resins derived from Chihuahuan semi-desert plants. Inhibition ranging from 0 to 100% with a greater effect as concentration increase, i.e. as concentration of resin or polyphenols increases, the mycelia growth of plant pathogen is significantly reduced (Hernández et al., 2010; Castillo *et al.*, 2010). An example is shown in Fig. 4, where plant extracts obtained with three different solvents ffecting *R. solani* growth is showed, in general, phytopathogen growth is reduced as phytochemicals concentration increase, in some cases fungal radial growth was totally affected.

Inhibition of fungal mycelium growth by plant extract may be as effective that conferred by synthetic molecules. Table 4 shows the effect of synthetic molecules and phytochemicals on phytopathogens radial growth, both kinds of chemicals presented similar effects. This quality could be a plus for the use of plant extracts on disease control of crop fields, which could modifying the existing disease management systems while reducing the negative effect of some synthetic chemicals on the environment and develop a more friendly production system (Hernández *et al.*, 2006; Hernández *et al.*, 2008).

	Mycelia growth inhibition (%)			
Treatment	B. cinerea	C. coccodes	F. oxysporum	
Control (Destilled H ₂ O)	0 d	0 f	0 d	
Chemical control y	100 a	100 a	81.3 b	
L. tridentata 1000	96 c	80.2 b	63.2 c	
L. tridentata 2000	95.9 c	72.2 с	49.1 d	
L. tridentata 4000	99.5 a	64.4 d	41.8 d	

Table 4. Mycelia growth inhibition of *Botrytis cinerea, Fusarium oxysporum* and *Colletotrichum coccodes* after the incubation period containing different concentrations of *Larrea tridentata*. Lira *et al.*, 2006. y Chlorotalonil and prozycar were used at 2000 ul liter ⁻¹, z Numbers Followed by the same letter do not differ significantly according to Tukey test ($P \le 0.01$)

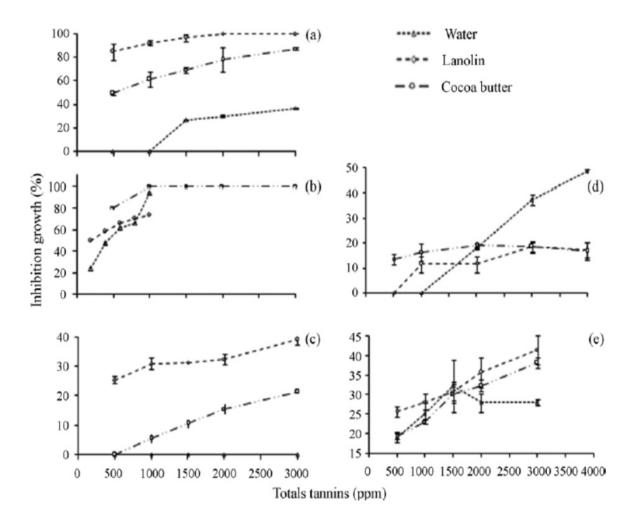


Fig. 4. Effect of plant extracts and total tannin concentration on inhibition of *R. solani* mycelia growth. (a) *Larrea tridentata* (b) *Flourensia cernua* (c) *Agave lechuguilla* (d) *Opuntia* sp. and (e) *Yucca* sp. Castillo, *et al.*, 2010.

In some extracts the overall effect on fungal mycelium growth inhibition can be maintained over time which gives fungicidal properties. Table 5 shows fungistatic and fungicidal action of some extracts on *P. infestans,* confirming the potential use of phytochemicals on plant disease control.

	Flourens	ia cernua	Origanur	n mejorana	Bouvardi	ia ternifolia
Dosis (ppm)	48 z	96	48	96	48	96
Metalaxil ^y	100.0 a	100.0 a	100.0 a	100.0 a	100.0 a	100.0 a
20000	88.44 ab	67.28 b	100.0 a	100.0 a	77.21 b	34.98 b
16000	81.63 b	48.56 c	100.0 a	100.0 a	57.14 c	1.23 c
12000	63.95 c	31.69 d	100.0 a	100.0 a	51.02 c	4.93 c
8000	49.66 c	9.15 e	100.0 a	100.0 a	22.45 d	0.0 c
4000	21.77 d	0.0 f	55.44 b	2.47 b	7.82 e	0.0 c
0	0.0 e	0.0 f	0.0 c	0.0 b	0.0 e	0.0 c

Table 5. Fungistatic and fungicidal effect of plant extracts obtained with methanol on *Phytophthora infestans* Mont (de Bary). ^z Hours, and ^y at 750 ppm. Gamboa *et al.*, 2003a.

This potential is effective also by low concentrations of some plant extracts which inhibited the fungal mycelium growth. Besides, availability of raw material for polyphenols extraction is high. Table 6 and Fig. 5 are presented the inhibitory concentrations IC₅₀ of different plant extracts on *R. solani* growth.

		Fidu	Fiducially Limits 95%			
Source of plant extract	Solvents	IC ₅₀ (ppm)	Inferior	Superior		
Larrea trindetata	Water	3.87X10 ³	3.07X10 ³	5.21X10 ³		
Larrea trindetata	Lanolin	1.85X10 ²	6.86X10 ¹	2.93X10 ²		
Larrea trindetata	Cocoa	5.71X10 ²	4.77X10 ²	6.56X10 ²		
Flourensia cernua	Water	4.20X10 ²	1.73X10 ²	6.49X10 ²		
Flourensia cernua	Lanolin	2.12X10 ²	7.96X101	3.11X10 ²		
Flourensia cernua	Cocoa	4.54X10 ²	*	*		
<i>Opuntia</i> sp	Water	3.83X10 ³	3.24X10 ³	5.19X10 ³		
Opuntia sp	Lanolin	2.08X104	$1.06X10^{4}$	9.44X10 ⁴		
Opuntia sp	Cocoa	4.9X10 ⁸	*	*		
Agave lechuguilla	Water	NI				
Agave lechuguilla	Lanolin	$1.70X10^{4}$	7.08X10 ³	2.21X10 ⁵		
Agave lechuguilla	Cocoa	6.72X10 ³	4.14X10 ³	2.79X10 ⁴		
Yucca sp	Water	5.74X10 ⁴	$1.42X10^{4}$	1.00 X107		
Yucca sp	Lanolin	8.96X10 ³	5.14X10 ³	2.99X104		
Yucca sp	Cocoa	8.14X10 ³	5.25X10 ³	$1.81X10^{4}$		

Table 6. IC_{50} values for inhibition of mycelia growth *R. solani*, with different plant extracts. NI = Not inhibited to doses evaluated and * = Not permitted to identify fiducially limits.



Fig. 5. Inhibition of *R. solani* mycelia using different concentration of *F. cernua* extract.

The mechanism of action of these phytochemicals is variable, for example, toxicity of phenols for microorganisms is attributed to enzyme inhibition by oxidation of compounds. The mode of action of terpenes and essential oils has not been fully elucidated, but is postulated to cause cell membrane disruption by the lipophilic compounds. The postulated effect of alkaloids is that these compounds may alternate with DNA, while lectins and polypeptides are known that can form ion channels in the microbial membrane or cause competitive inhibition of adhesion of microbial proteins to host polysaccharide receptors (Cowan, 1999).

It has also been reported an excellent effect of plant extracts on growth inhibition of plant and food pathogenic bacteria. Table 7 shows the effect of plant extracts on growth of bacterial colonies, these plant extracts also reduce significantly the growth of food bacteria Figure 6.

	_	Bacteria Inhibition (%)		
		C. m. subsp. michiganesis		subsp. skensis
Source of plant extract	Concentration (PPM)	8 Days	4 Days	6 Days
Control (Distilled H ₂ O)	0	0	0	0
L. tridentata	500	100	100	100
L. tridentata	1000	100	100	100
L. tridentata	1500	100	100	100
L. tridentata	2000	100	100	100
F. cernua	50	100	100	100
F. cernua	150	100	100	100
F. cernua	300	100	100	100
F. cernua	450	100	100	100
A. lechuguilla	50	100	100	98.99
A. lechuguilla	150	100	100	100
A. lechuguilla	300	100	100	100
A. lechuguilla	450	100	100	100

Table 7. Effect of extracts from *Larrea tridentata, Flourensia cernua* and *Agave lechuguilla* on inhibition the growth of bacterial (*Clavibacter michiganensis* subsp. *Michiganensis* and *Clavibacter michiganensis* subsp. *nebraskensis*) colonies.

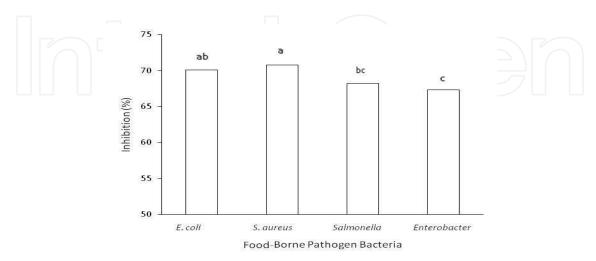


Fig. 6. Percentage of inhibition of food-borne bacterial growth because different plant extracts.

Most of the plant sources tested to date have demonstrated the antimicrobial potential of powders and extracts. Therefore, it is important to continue research for discover the active compounds responsible of the antifungal effect, and subsequently help to elucidate its biological effects and potential use. The available Mexican plant diversity should be used, especially because their biodegradable and nontoxic compounds.

2.5 Effect of solvent and extracts on spore production

Phytochemicals derived from semi-desert plants, shows not only action in arresting the growth of fungi and bacteria colony, but will also affect fungal sporulation, this action is presented in Table 8, showing the importance of the solvent used in phytochemical extraction and their effect on production of conidia for different fungal species.

Extracts	Alternaria alternata ^x	Penicillium digitatum ^x	Colletotrichum gloeosporoides ^y
Solvent			
Ethanol	0.93 b	0.24 a	5.50 c
Methanol: Chloroform	0.26 b	0.18 a	5.76 c
Hexane	4.40 a	0.19 a	7.77 b
Eter	3.33 a	0.24 a	9.29 a
Concentration			
0	3.13 a	0.61 a	8.07 a
500	2.78 ab	0.13 b	7.82 a
1000	2.66 ab	0.13 b	7.31 a
2000	1.64 ab	0.11 b	6.36 b
4000	0.94 b	0.08 b	5.80 b

Table 8. Comparison of four *Flourensia cernua* extracts and four concentrations on sporulation of three postharvest pathogens. \times 1.0 x 10⁵ conidia/ml, and $^{\rm Y}$ 1.0 x 10⁶ conidia/ml. Guerrero *et al.*, 2007

Likewise, one cans infer that the inhibition of sporulation is a function of the phytochemicals concentration used. Table 9, shows that conidia production is affected when phytochemicals concentration increase.

Sporulation inhibition x 10 ⁴ ml ⁻³				
Treatment	B. cinerea	C. coccodes	F. oxysporum	
Control (Destilled H ₂ O)	8.3 a ^z	6.7 a	47.7 bcd	
Chemical control	0.0 b	0.0 a	56.6 bc	
L. tridentata 1000	2.2 b	4.2 abc	25.0 bcde	
L. tridentata 2000	1.7 b	6.4 ab	20.3 cde	
L. tridentata 4000	1.7 b	2.8 ab	15.8 de	

Table 9. Effect of *Larrea tridentata* extracts on sporulation inhibition of *Botrytis cinerea, Colletotrichum coccodes* and *Fusarium oxysporum* after the incubation period (Lira *et al.,* 2006). Chemical controls used were chlorotalonil and prozycar at 2000 ul liter ⁻¹. ^z Number followed by the same letter do not differ significantly according Tukey test ($p \le 0.001$)

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2.6 General description of Chihuahuan semi-desert plant species sources of phytochemical compounds with anti-fungal properties

Larrea tridentata is a xerophytes evergreen plant that can survive hundreds or thousands of years through vegetative reproduction (asexual), because the roots produce new sprouts or shoots which are then converted into new plants (Brinker, 1993). The plant shows variation in height from 0.5 to 4 m. This height is a function of ploidy level (diploid 86 cm, 112 cm hexaploid and tetraploid 138 cm). There is one main stem, but the thick branches grow vertically or obliquely from the crown and root side is dichotomous. Its leaves are small and bifoliate, dark green to yellowish green with thick cuticles and a resinous coating, have short petioles and grow opposite on the branches (Fig. 7). The flowers are yellow usually appear in late winter or early spring, but can bloom at any time after a rain, grow near the ends of young shoots and buds solitary with five petals. The fruit are small between 4 to 7 mm in diameter, have a fuzzy cover and contains 5 seeds (Jasso de Rodriguez *et al.*, 2006; Lira, 2003).



Fig. 7. Larrea tridentata

Secondary Metabolites: The main compounds reported in the literature are numerous. Distinguished by their higher content of dry weight basis of foliage phenolics, lignans, saponins, flavonoids, amino acids and minerals. The most important compound is nordihydroguaiaretic acid (NDGA) (Lira, 2003) found in the resin cell epidermal layers near the top and bottom of the leaves and stems.

Flourensia cernua D.C. Is a much-branched plant up to 2 m high, which exudes a resinous. It has slender branches, evergreen, light brown to gray with alternate leaves, composed of two leaflets, elliptic to oblong 17-25 mm long and 6.5-11.5 mm wide, acute at both ends, dark green, sometimes resinous paler underside and petiole of 1-2 mm (Hyder *et al.,* 2005). The flowers are in corymbs or panicles flower heads and has 12 to 20 flowers per capitulum. The fruit is a very hairy achene 6 mm long and 2 mm wide, laterally

compressed, 2 to 4 edges uneven hair 2-3 mm long, nearly obscured by the long hairs of the achene (Jasso *et al.*, 2006).

Secondary Metabolites: The chemical composition of *F. cernua* resin is sesquiterpenoids and triterpenoids (Kingston *et al.*, 1975), polyacetylenes, p-acetophenones, benzofurans and benzopirans (Bohlmann and Grenz, 1977) and flavonoids (Dillon *et al.*, 1976; Rao *et al.*, 1970). Advanced Studies on phytochemistry of leaves reveal the presence of resins, flavonoids (deoxy flavonoids), phytoalexins, coumarins, phenolic compounds, benzofurans, p-coumaric acid. Some particular compounds include dehydroflourensic acid and flourensadiol (Kingston *et al.*, 1975) that enable the use of this species with biological activity against pathogens.

Opuntia ficus-indica (L.) Mill. This plant tree 3-5 m tall, crown wide, glabrous, stems of 60-150 cm wide, obovate cladodes of 30-60 cm long, 20-40 cm wide and 19-28 cm thick, dark green covered with a layer of wax. Spines usually absent or up to 2 per areola, short: only 0.5-1 cm, weak, whitish. Flowers (6-) 7-9 (-10) cm long are orange to yellow. The fruit is sweet, juicy, edible, 5-10 cm long and 4-8 cm wide, pyriform, slightly sunken in the navel, pulpy and thin shell (Fig. 8). The seeds are obovate to disc-shaped 3-4 mm in diameter (FAO, 1999).



Fig. 8. Opuntia ficus-indica

Secondary Metabolites: According to the revised literature, has not been reported secondary metabolites present, although we did detect condensed polyphenols gallic acid equivalent, saponins and terpenes.

Lippia graveolens Kunth. Is an evergreen shrub, with life cycle of 3 to 10 years belonging to the family Verbenaceae, can reach up to 2.5 m tall with branched stems with many leaves 1-3 cm long and 0.5 to 1.5 cm wide, distributed in the opposite form, alternate and oval in shape with jagged edges, rough texture with light hairs. Presents individual inflorescences with small white flowers (Lahlou, 2004). The number of flowers per inflorescence is very variable most frequently found 10 flowers per inflorescence, its fruits are small capsules containing brown seeds, not more than 0.25 mm (Lahlou, 2004).

Secondary metabolites. The biological components with higher capacity are in the essential oil consisting mainly of thymol and carvacrol, as well as some phenolic acids and flavonoids with antimicrobial properties (Lambet *et al.*, 2001)

Yucca filifera Chabaud. Plant tree to over 10 m high, much branched with leaves up to 55 cm long and 3.6 cm wide, linear-oblanceolate, constricted near the base, rigid, usually rough on both surfaces with numerous coiled filaments white, easily breakable, so are most noticeable in young leaves (Fig 9). The escape of the foliage stands; panicle more or less cylindrical, pendulous, up to 1.5 m long, multiflora, extended flowers, pedicellate, pedicels up to 2.7 cm long, 3.8-5.2 perianth segments cm long, 0.7-2.5 cm wide, inner segments slightly shorter and wider, filaments 1-1.5 cm long; pistil 2.3-2.5 cm long, ovary 1.8-2 cm long and 0.4 to 0.5 cm in diameter. Hanging fruit, oblong, 5-8.8 cm long, 2.7-3.3 cm in diameter, ending in a peak of 0.2 to 0.7 cm long. Seed 8 x 2 mm, somewhat rough.



Fig. 9. Yucca filifera.

Secondary Metabolites: The leaves and roots of the *Yucca* genus contains saponins, steroidal sapogenins and a high content of ascorbic acid.

Carya illinoensis. Cultivated species belonging to the family Juglandaceae is a tree that can reach 50 meters in height with a trunk up to 2 m in diameter, its bark is cracked and rough, greyish. Its leaves are deciduous, compound, odd-pinnate, lanceolate, large, oval, toothed, petiole short 6 to 12 cm wide. The flowers are very small, apetalous, monoecious and are grouped in catkins (earrings) cylindrical pendants, light green. The fruit or drupe, consisting of pericarp, mesocarp and seeds (almonds). The pericarp, as the mesocarp is a segmented structure into four parts that opens dehydrated freeing the endocarp and seed. A portion of the mesocarp and endocarp is known as husk. The nuts consist of the endocarp and the seed typically measure 2 to 6 cm long and weigh 4 to 12 g each. The seed has two cotyledons separated by a center wall, which come from flowers carpels (Fig 10).



Fig. 10. Carya illinoensis fruits

Secondary metabolites. Depending on the analyzed part of the plant one cans find different active principles in leaves, naphthoquinones (juglone, plumbagin, beta-hydroplumbagin) in the pericarp, organic acids, tannins and naphthoquinones, in the cotyledons, unsaturated fatty acids, in the integument, polyphenols and tannins, and walnut, Vitamin A, B, C and E, minerals and iodine. Meanwhile, Sasaki (1964) reports that the walnut husk contains azaleatin (Quercetin 5-methyl ether) and caryatin flavonol (quercetin 3.5-dimethyl ether).

3. Conclusion

The from Mexican semi-desert plant species have the ability to inhibit the development of mycelium and sporulation of fungi and stramenopiles and growth of bacterial pathogens. Under field conditions a decrease in disease incidence and severity have been reported. Phytochemicals derived from *Larrea tridentata* and *Flourensia cernua* show a wide spectrum of action towards different phytopathogens, this activity occurs even with non-conventional solvents such as water, lanolin and cocoa butter. The use of natural extracts in controlling plant diseases has low or no environmental impact, so they may become a viable option for development of organic and sustainable agriculture. However, it is necessary to develop research on molecular and biochemical changes that these compounds may have on pathogen and plant.

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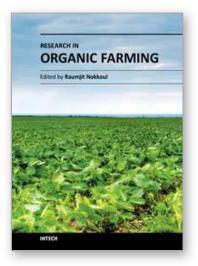
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Research in Organic Farming

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This book has emerged as a consequence of the difficulties we experienced in finding information when we first started researching. The goal was to produce a book where as many existing studies as possible could be presented in a single volume, making it easy for the reader to compare methods, results and conclusions. As a result, studies from countries such as Thailand, Spain, Sweden, Lithuania, Czech, Mexico, etc. have been brought together as individual chapters, and references to studies from other countries have been included in the overview chapters where possible. We believe that this opportunity to compare results from different countries will open a new perspective on the subject, allowing the typical characteristics of Organic Agriculture and Organic Food to be seen more clearly. Finally, we would like to thank the contributing authors and the staff at InTech for their efforts and cooperation during the course of publication. I sincerely hope that this book will help researchers and students all over the world to reach new results in the field of Organic Agriculture and Organic Food.

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