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Interactions of allelopathy and competition affecting Ziziphus spina-christi and Prosopis juliflora seedlings

Thobayet S. Alshahrani
West Virginia University

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Thobayet S. Alshahrani

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Ray R. Hicks, Jr., Ph.D., Chair
Sven Verlinden, Ph.D.
Devinder K. Bhumbla, Ph.D.
Richard B. Thomas, Ph.D.
Jonathan R. Cumming, Ph.D.

Division of Forestry

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Abstract

Interactions of allelopathy and competition affecting growth of *Ziziphus spina-christi* and *Prosopis juliflora* seedlings.

Thobayet S. Alshahrani

This study consists of two parts, provides data on the interference between *Ziziphus spina-christi*, native to Saudi Arabia, and *Prosopis juliflora*, an introduced species. Dry leaves from both species were milled and used to prepare extracts using 0g/l, 5g/l, 20g/l, 40g/l, 60g/l, and 100 g/l of dried leaves. Leaf extract of *P. juliflora* was used to irrigate *Z. spina-christi* with 120 ml weekly over a 7- month growth period, and vice versa. Plant height, plant diameter, number of leaves, leaf area, chlorophyll content, total root length, number of root tips, root diameter, root surface area, root volume, shoot dry weight, root dry weight, and root to shoot ratio were measured monthly. *Ziziphus spina-christi* leaf extract produced a negative impact on all studied parameters of *P. juliflora* and the negative impact increased with increasing extract concentrations. Lower leaf concentration extracts of *P. juliflora* positively affected the studied parameters of *Z. spin-christi*. At 60 g/l the growth of *Z. spina-christ* was highly stimulated but at 100 g/l all the growth parameters were reduced. The study explored the negative effect of *Z. spina-christi* leaf extracts on the growth of other plant species where no such study has been conducted before.

The second part is a study of the effect of interaction between interspesific competition and nitrogen levels on growth of *Prosopis juliflora* and *Ziziphus spina-christi* seedlings for 7-months. Dry weight, root:shoot ratio, leaf area, plant height, plant diameter, total chlorophyll, relative yield, and aggressivity were measured and/or calculated. In mixed plantings with a high level of nitrogen, *Prosopis juliflora* exceeded the growth of the native species in most studied parameters but the effect of intraspesific competition was high. Under a low level of nitrogen the native species growth exceeded that of *P. juliflora*. In monoculture plantings, both species showed a reduction in growth parameters with increasing numbers of plants per pot and the reduction in low levels of nitrogen exceeded that in high levels of nitrogen. For total dry weight and leaf area, seedlings of *Z. spina-christi* grown in low levels of nitrogen exceeded that of seedlings grown in high levels of nitrogen.

DEDICATION

To my wife
To my children
Omar
Tamador
Norah
Mohaned

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Chapter 1:
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Abstract

This study explores the allelopathic interference between *Ziziphus spina-christi*, native to Saudi Arabia, and *Prosopis juliflora*, an introduced invasive species. Dried leaves from both species were milled and used to prepare leaf extracts with concentrations of 0 g/l, 5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l. The leaf extracts of *P. juliflora* were used to irrigate *Z. spina-christi* with 120 ml per week during a 7-month growth period, and vice versa. Growth parameters such as plant height, plant diameter, number of leaves, leaf area, chlorophyll content, total root length, number of root tips, root diameter, root surface area, root volume, shoot dry weight, root dry weight, and root to shoot ratio were measured monthly.

Ziziphus spina-christi leaf extract showed a negative impact on most studied parameters of *P. juliflora*. The negative impact increased with increasing extract concentrations, but no mortality was observed. Seedling heights for *P. juliflora* averaged 50.16 cm and 32.98 cm for 5 g/l and 100 g/l treatments, respectively. Chlorophyll *a* and total chlorophyll decreased with increasing extract concentrations and there were significant differences between lower and higher concentrations. Leaf area of *P. juliflora* averaged 100.89 cm^2 at 5 g/l but it diminished to 45.3 cm^2 with 100 g/l. Total root length, root surface area, number of root tips, and root volume decreased with increasing leaf extract concentration of *Z. spina-christi*. Average root diameters of *P. juliflora* did not vary with extract concentrations.

Leaf extract of *P. juliflora* had a positive effect on the studied parameters of *Z. spina-christi*. Growth was stimulated with increasing leaf extract concentration up to 60 g/l. However, at 100 g/l all the growth parameters declined. Average seedling height was the highest at 60 g/l averaging 40 cm, but it decreased to 25 cm at 100 g/l. Chlorophyll *a* average, and total chlorophyll showed no significant differences between 5, 20, 40, and 60 g/l but it declined at 100 g/l. Leaf area was the lowest at 100 g/l, 168.13 cm^2 , but it was the highest at 60 g/l with 308 cm^2. Average root length, average root

diameter, root surface area, and number of root tips increased with increasing concentration, again except for the 100 g/l concentrations where there were reductions in all measured root parameters.

This study emphasized that the allelopathic effect depends on the species, donor and recipent, and the concentration of the allelochemical. The study also revealed that *Z. spina-christi* leaf extract had a negative effect on the growth of another plant species, a finding that has never been reported previously.

Chapter 1:
The reciprocal effects of leaf extracts of *Ziziphus spina-christi* and *Prosopis juliflora* on seedling growth.

Introduction

There are numerous examples where introduction of exotic plant species have threatened native species. The alien species may become invasive and displace the native species because of its aggressive behavior outside its natural range (Chaloupka and Domm, 1986). Growth of the alien species can exceed the growth of local plants because of the absence of damaging agents, presence of special growth requirements, or rapid growth rate, resulting in the species' ability to out-compete natural plants for resources. Several mechanisms, such as competition for minerals, water, and light, and allelopathy may be involved in the interaction between these plants. It is most likely that plants compete with each other by more than one mechanism (Newman, 1988).

When two or more species occupy the same site they interact, positively or negatively, consequently affecting each other's fitness. Competition will occur when resources are limited and a negative interaction takes place because one of the species will be less successful in the presence of a competitor. The success of individual plants will therefore depend on their ability to compete with individuals of their own and other species under the given conditions. In the case of underground competition, the competitive ability of any species will depend strongly on its root architecture and its ability to secure resources in large quantities. Species with highly branched root systems consisting of a main axis and primarily laterals, also called herringbone architecture, are the most efficient in gathering nutrients. Invading success in some plant species may depend on the combination of root architecture and their ability to produce allelochemical compounds in roots or leaves to curtail the growth of competing species.

Allelopathy is a mechanism that enables one plant species to prevent the growth of another by producing growth-inhibiting substances. Allelopathy is defined as: any direct or indirect beneficial or harmful effect of one plant (donor) on another (recipient) through the production of chemical compounds that escape into the environment (Chon et al., 2000). In higher plants, allelopathy can influnce one or more phases of growth,

including seed germination and/or seedling growth. Generally, germination is less sensitive than is seedling growth, especially root growth (Miller, 1996; Peterson, 1956). Allelochemicals result in a negative interaction between species by affecting physiological processes (Patterson, 1981; Mersie and Singh, 1988). This inhibitory mechanism is important in ecological interactions between species and is an important determinant of the outcome of plant-plant interactions (Nilsson et al., 1998). On the other hand, some allelochemical substances have a stimulating effect on the growth of the recipient plant at low concentrations (Buta and Spaulding, 1989). Many plants produce allelochemical compounds but some have no negative effect because either biotic or abiotic triggering factors are lacking in the soil (An et al., 2001). Allelochemical compounds may be selective in their action, or plants may be selective in their response (Zeng et al., 2001). Undoubtedly, concentrations of secondary metabolites in the soil are an important factor to investigate affecting the allelopathic characteristics of particular species (Rice, 1979). Thus, allelopathy must be viewed as a complex phenomenon that is concentration-dependent (Hall et al., 1982; Hedge and Miller, 1992).

In Saudi Arabia many exotic tree species have been introduced in plantations where the native species did not meet requirements of the native populace. In addition, many local species are slow growing, have low productivity and a low rate of seed germination. For example, *Prosopis spp.*, *Eucalyptus spp.*, *Casuarina spp.*, and *Acacia spp.* are the most common species that have been used in plantations throughout the country. The most important exotic species is *Prosopis juliflora,* which was first introduced to the eastern region of Saudi Arabia 25 years ago and recently has become naturalized throughout the country (Shalabi and Alqarawi, 1997; Zoghet and Al-Asheikh, 1999). In addition, humans have contributed significantly in the spread of *P. juliflora*, an invasive species that threatens native species due to its ability to outcompete under extreme environmental conditions.

Prosopis juliflora is considered an invasive plant in Eastern and Central Sudan (Richardson, 1998), Pakistan (Noor et al., 1995), Ethiopia, and India. The success of the species to dominate is related to the allelopathic effect of its leaves which contain a growth inhibiting substance that apparently negatively affects the growth and fecundity of other species (Nakano et.al., 2002). *P. juliflora* produces heavy leaf litter which

increases the concentration of allelochemicals to amounts sufficient to affect the growth of native species. Leaves of *P. juliflora,* like many invasive species, contain high levels of inhibitory substances compared to other plant parts (Chon and Kim, 2002; Sen and Chawan, 1970; Tefera, 2002; Vandermast et al., 2002). The effect of allelopathic agents can include a reduction in mineral uptake, such as Ca, Mg and S (Walker et al., 1991), a reduction in photosynthesis, a lowering of the water content (Lodhi and Nickell, 1973), inhibition of specific enzymes, effects on stomatal activity (Spurr and Barnes, 1980), inhibition of respiration (McCahon et al., 1973), reduction of shoot and root growth (Baziramakenga et al., 1994), changes in the morphology of root architecture (Kim et al., 1995; Hedge and Miller, 1992a), and a reduction in the amount of chlorophyll in the leaf (Batish et al., 2002; Jayakumar et al., 1995; Viles and Reese, 1996). Allelopathy has been reported to enhance the capability of *P. juliflora* as an invasive species to prevent the growth of other species. Therefore, allelopathy promotes a change in community composition because native species have difficulty in surviving and regenerating and may become exterpated (Vivrette and Muller, 1977).

Ziziphus spina–christi (L.) is a very important native species in Saudi Arabia. The genus belongs to the family Rhamnaceae, which includes 40 species. *Z. spina–christi* is an indigenous tree of the Arabian Peninsula and although it grows wild throughout the country, it is concentrated in the Southern and south–western regions of Saudi Arabia (Said 1986). Natural distribution of the species in patches, in small or large numbers, is related to the availability of favorable sites for growth. The thick and woody seed coat of *Z. spina–christi*, which is about 1.38 mm (p. obs.), may play an important role in the lack of or delayed seed germination observed in this species. This may in turn reduce its chances to occupy sites and successfully compete for resources with other species. *Z. spina-christi* is very important to some local inhabitants of Saudi Arabia. Leaves of *Z. spina-christi* are a source for medicinal compounds used for several ailments by the indigenous population (Zoghet and Alsheikh, 1999; Weinges and Schick, 1995). It is used for its cleaning properties and is a food staple for animals. The flowers of the species produce a very highly prized honey that is widely marketed. Leaves of the species contain flavonoids (Nawwar, et al., 1984), saponin, alkaloids, and other potentially allelochemical substances that may play an important role in the interaction between *Z.*

spina-christi and other plant species such as *P. juliflora*. In Saudi Arabia it is important to understand the interaction in order to make informed decisions on the management of introduced species. Therefore, it is essential to asses the impact of *P. juliflora* on one of the region's most important native plant's seedling development since no such studies have been carried out before. Thus, a study of the allelopathic properties of the invasive species *P. juliflora* on this important native species is essential and will lead to better management decision in dealing with both species. A study of the negative interaction may help in stemming the spread of *P. juliflora.* The objectives of this study are;

1) to determine the reciprocal effect of *P. juliflora* leaf extracts as a potential source of allelopathic compounds on seed germination and seedling growth of Z. *spina-christi.*

2) To explore which growth stage for the two species is more susceptible to inhibitor compounds.

3) To investigate the effects of extract concentration on the seedling growth of the two species.

Literature review

Role of allelopathy in plants

Importance of allelopathy

Allelopathy is the production of allelochemical materials by one plant that can affect another plant. Whitman (1988) divided allelochemicals into 4 categories depending on whether the response of the receiver is adaptively favorable to the emitter but not the receiver (allomones), is favorable to the receiver but not the emitter (kairomones) or is favorable to both emitter and receiver (synomones) (Kohli et al., 1998).

Allelochemical compounds can influence plant community structure, where an individual species creates patches that alter the establishment and growth of other plant species in specific zones around the plant (Nilsson et al., 2000; Rai and Tripathi, 1984). In plant succession a dominant species may, by allelopathic suppression, speed its invasion into the succeeding community and delay its replacement by other species (Whittaker and Fenny, 1971). For example, in California, *Eucalyptus globulus*, native to Australia, displaced native vegetation as a result of the tree's allelopathic effects and its impact on nutrient cycling thus reducing the uptake of N, P, K, Fe, and Mo in turn reducing plant physiological processes (Watson, 2002; Alsaadawi et al., 1986; Walker et al., 1989). Another example is *Ailanthus altissima* where allelopathy is important in the establishment and persistence of *Ailanthus*. Both toxic exudates from roots and foliage extracts contribute to the aggressiveness of *Ailanthus altissima* (Heisey, 1990). The effect may occur when ground-cover toxins interfere with nutrient uptake by damaging or destroying root cells, root hairs, and mycorrhiza (Chick and Kielbaso, 1998).

In plants, all tissues contain potential allelochemicals and every interaction in allelopathy involves a group of compounds working together. The chemical structure of these compounds vary widely and include lactones, acids, coumarins, quinines, steroids, flavonoids, trepenoids, alkaloids and tannins (Whittaker and Fenny, 1971). All have potential inhibitory activity, and most of them produce different biological effects (Einhellig, 1995). Inhibiting substances such as phenolics and terpenes can be produced in leaves, roots and decaying tissues (Fuerst and Putnam, 1983). Many differences in allelopathy among extracts from different tissues of plants have been reported (Qasem

and Foy, 2001). Differences might be related to allelopathic compounds being produced in larger quantities in certain tissue, imparting a higher level of inhibition, compared to others (Chon and Kim, 2002). Chou and Leu (1992) found that aqueous extract of leaves, flower, and twigs of *Delonix regia* showed different patterns of toxicity and found that the highest inhibition was observed with floral extracts. The degree of inhibition increased with an increase of concentration of the extracts. However, the 5%, (W:V) flower extracts showed the highest inhibition toward bioassay species Lucerne, lettuces and Chinese cabbage comparing to 1%, 2%, 3%, and 4% extracts.

In plants allelochemicals can be present in seeds, leaves, stems, flowers, buds, bark, pollen grains, fruits, roots, and rhizomes (Qasem and Foy, 2001). Seed longevity in some species may be attributed to allelelopathic agents that protect seeds from attack and decay by microorganism. Presence of allelochemical compounds in seeds is an advantage for some species, helping them to invade and dominate a plant community. In South Africa *Sesbania punicea* (Cav) Benth., a noxious weed producing many seeds rich in the cytotoxic alkaloid sesbanimide, invades natural vegetation and forms dense stands. Such situations are typical of species that release allelopathic compounds from seeds which inhibit seedling growth of other species (Staden and Grobbelaar, 1995). The seed pericarp of *Prosopis juliflora* contains allelochemicals that inhibit seed germination and seedling growth of the same species called autotoxicity, (Warrag, 1994) as well as other species, termed phytotoxicity (Bennet and Bonner, 1953; Goel et al., 1989; Noor et al., 1995). Sen and Chawan (1970) found that leaf extracts had a more inhibiting effect than fruit extracts. A common situation, illustrated by *P. juliflora,* is the case where allelochemical activity is enhanced by the synergism of major and minor phenolic acids and nonpolar organic compounds (Goel et al., 1989).

Leaves often have allelochemical inhibitors that find their way to the environment by leaching or decay. Allelochemicals of leaves when compared to the allelochemicals of other parts in the plant are the most effective in inhibiting growth and development of other species (Vandermast et al., 2002; Tefera, 2002; Chon and Kim, 2002). Many substances are deposited in leaves. Abscission of the leaf enables the plant to distribute these substances in the immediate environment. For example, leaves of *Empetrum hermaphroditum* produce high levels of phenolics that inhibit the growth of other species

(Nilsson et al., 1998). In *Acacia confusa* leaves have inhibitory substances that curtail the growth of many different species (Chou et al., 1998). For *P. juliflora*, the inhibitor chemicals were isolated and identified as syringin, lariciresinol, and L-trypotophan (Nakano et al., 2002; Nakano et al., 2001). Dhawan (1995) found that the extracts of *P. juliflora* showed strong inhibitory properties compared to the extracts of *P. cineraria* toward *Parthenium hysterophru.* The seeds of *Parthenium hysterophrus* soaked in *P. juliflora* extracts showed poor germination, seedling growth, and produced weak seedlings that died shortly after initial establishment.

In nature, allelopathy is often responsible for interference between plants and its negative effects can be confused with competition. For example, a reduction in the growth of yellow nustsedge, *Cyperus esculentus*, was observed when grown with *Ipomoea batatas* in a greenhouse experiment where yellow nutsedge growth was reduced by 50% relative to the control (Harrison and Peterson, 1991a). Rai et al. (1998) studied allelopathic effects of 3 trees species, *Casuarina equisetifolia*, *Eucalyptus tereticornis*, and *Leucaena leucocephala* on five crops (sunflower, greengram, sesame, cowpea, and sorghum). In this study it was shown that the response of the recipent crop depends on the donor tree. The extracts of the all three trees depressed the germination of sunflower. However, the cowpea was not affected by any of them. Therefore, for different recipent species, the response to allelopathic inhibitors may vary. Recipent species not only vary in their response to allelopathic compounds but the impact could occur in above-or below-ground tissue growth. For instance, the variation in root/shoot dry mass ratio for *Pinus taeda* seedlings grown in *Myrica cerifera* leaf litter reflected a differential responses for above and below ground portions (Tolliver et al., 1995).

Allelochemical compounds can be distributed to the environment in different ways including volatilization, exudation from roots, leaching from plants by rain, or decomposition of above–ground or below-ground residues (Whittaker and Fenny, 1971; Rice, 1984). Toxic materials, if present, will find their way into the soil sooner or later, and have an effect, direct or indirect on the target plant. When the allelochemical substances are released to the environment, they are subject to various physiochemical and biological processes, and they may be detoxified or toxified by soil organisms and/or may serve as carbon skeletons for the production of new toxins by organism in the soil

(Blum et al., 1999). However, some species contain inhibitors in free form that are discharged to the environment without hydrolysis while other species release their phytotoxins after hydrolysis (Lodhi, 1978). It is clear that plant species vary in their inhibitory substance(s) and vary as to the way those chemicals are released into the environment.

Role of allelopathy in trees

Allelopathic species can be found in all classes of plants including gymnosperms and angiosperms. Allelopathic chemicals can also be found among algae, fungi, and mosses. Much research, however, has focused on tree species. Many tree species belonging to different plant families are known to exhibit allelopathic effects. Tree species often use a specific set of allelopathic compounds for reducing its competition for resources with other species.

In arid regions, several tree species are widely used as shelterbelts, windbreaks, or boundary plantations. Most of these species are considered to be allelopathic. For example, *Acacia arabica*, *Acacia tortilis*, *Leucanea leucocephala*, *P. juliflora, P. cineraria,* all belonging to the Mimosaceae family, are frequently used for purposes such as shelterbelts, windbreaks, fuelwood, canal-side plantations and sand dune stabilization. *Zizyphus rotundifolia* (Rhamanaceae), used in agroecosystems, has been found to affect crops through its allelopathic effects (Kohli et al., 1998). Also, *Pinus densiflora*, *P. thunbergii,* and *P. rigida,* used in plantations, have also been shown to exhibit allelopathy (Kil, 1989). In the Myrtaceae family there are many allelopathic species such as *Eucalyptus camaldulensis* (del Moral and Muller, 1970), *Eucalyptus treticornis* (Rai et al. 1998), and *Eucalyptus globulus* (Watson, 2000). Several other examples of allelopathic trees have been reported, including, *Quercus falcata* var. pagodaefolia (cherrybark oak) (Rice, 1979), *Pinus densiflora* (red pine) and, *Pinus thunbergii* (black pine) (Kil, 1989). Lodhi (1976) found that leaf water leachate of sycamore, hackberry, red oak, and white oak reduced the seed germination radical growth and seedling growth for some herbaceous species tested.

In desert plants, specifically, allelopathic substances play an important role in reducing the competition between species. For example, saponin in some desert plants

inhibits growth of other species. Askham and Cornelius (1970) found a negative effect of saltbush, *Atriplex canescens*, containing saponin, on seed germination of many subject plants at high concentrations, but interestingly, the growth was stimulated at low concentrations. Other desert shrubs such as *Rhazya stricta* contain alkaloides (Atta ur Rahman et al., 1991) that completely inhibited the germination of many other range plants in Saudi Arabia (Assaeed and Al-Doss, 1997). The importance of such toxic substances is greater in desert region because of restricted rainfall inhibiting leaching from soil allowing allelopathic compounds to residue for a long time (Sen and Chawan, 1970).

Role of allelopathy in plant invasion

Allelopathy has been suggested as the key strategy for the impressive success of many invasive plants that have become dominant in their invaded plant communities (Ridenour and Callaway, 2001). Expansion of invasive species depends on the ecology of the invaded communities and in many cases the density. For example, *Phalaris arundinacea* invades disturbed or low-density plant communities (Morrison and Molofsky, 1998). However, plant communities with high biomass production tend to be less conducive to invasion because litter accumulation inhibits seedling establishment of the invasive species (Burke and Grime, 1996). When invading species become dominant allelopathy is believed to play an important role in further invasion. The lack of co-evolved tolerance of resident species to new chemicals produced by the invader allows the invading species to dominate plant communities quickly (Hierro and Callaway, 2003). In addition, competitive strategies for resources and neighbor suppression allow invasive species' populations to aggressively take over a site or community by increasing the mortality of native species (Fischer et al., 1994; Tilman, 1988).

Allelopathy and plant growth

Allelopathic substances produced by many plant parts can negatively affect germination and seedling growth of many other annual or perennial plants. The response of particular species varies depending on the source plants from which allopathic substances are obtained. Allelopathic substances can have a strong, moderate, or slight

impact on the growth of other plants (Kil and Yun, 1992). Aqueous extract of leaves from *Lantana camara*, for example, reduced root and shoot biomass in soybean (Mersie and Singh, 1987) but leaf extracts of *Abutilon theophrasti* completely inhibited the growth of soybean seedlings (Colton and Einhelling, 1977). For perennial species, such as *Pinus sylvestris*, seedlings were negatively affected by the above-ground components of the shrubs *Empetrum hermaphroditum* and *Pleurozium schreberi* in indoor expermints; yet *E. hermaphroditum* had stronger effect on seed germination (Zackrisson et al., 1997). The efficacy of allelopathic compounds can also vary depending on the donor part of the source plant. For instance, in *P. juliflora,* fruit extracts were more inhibitory to shoot growth of different cultivated plants when compared to extracts of root, stem, leaf and flower. Interestingly, the root extracts of *P. juliflora* significantly promoted seedling growth of *Triticum aestivum* and *Zea mays* (Noor et al., 1995).

When plants are exposed to allelochemicals, their growth and development are affected. The readily visible effects include inhibited or retarded germination rate, seeds darkened and swollen, reduced root or radical and shoot or coleoptile extension, swelling or necrosis of root tips, curling of the root axis; discoloration, lack of root hairs, and increased number of seminal roots which anchors the young plant and absorbs minor amounts of water and nutrients for the first two to three weeks. In general, a reduction in dry weight accumulation and lowered reproductive capacity of recipent plants is observed. In the target plant, allelopathic compounds can affect resource allocation. El-Khatib et al. (1998) found that high concentration of extracts from *Zilla spinosa* affected the root length of several species and resulted in a significant increase in shoot/root ratio of those species. The reduction was accompanied by changes in root morphology. Roots of the target species had very few lateral roots and roots were generally more thickened. In another study, root to shoot biomass ratios increased in shrub species treated with aqueous leachates derived from the invasive species *Acacia cyclops* (Rutherford and Powrie, 1993). Springer (1996) found that tall fescue extracts (*Festuca arundinacea* Schreb) increased seedling shoot length of clover species whereas root lengths decreased.

Seedling dry weight may, however, be a better indicator for allelopathic effects (Ahn and Chung, 2000). Kil (1989) found that dry weight ratios for seedlings of 35 recipent species were the most severely inhibited out of all measurements taken. Root

length can also be used as a key parameter because root elongation responds rapidly and markedly to allelopathic compounds (Chon et al., 2002). The effect of allelopathic substances on roots is sometimes manifested by increasing rooting depth, increasing root volume or reducing root volume. For example, Roder et al. (1988) found that leachates from sandbur [(*Cenchrus longispinus* (Hack)] plants reduced initial root growth and increased shoot growth of switchgrass (*Panicum virgatum* L.) seedlings without affecting germination which indicated the selective susceptibility of certain seedling organs to allelopathic compounds. In another study by Hoque et al. (2003) the allelopathic effects of different concentrations of aqueous leaf extracts from *Eupatorium odoratum* [*Chromolaena odorata*] on the germination and growth behaviour of some agricultural crops such as *Cicer arietinum*, *Brassica juncea*, *Cucumis sativus*, *Phaseolus mungo* [*Vigna mungo*], *Raphanus sativus* and *Vigna unguiculata* were examined. *E. odoratum* leaf extracts caused a significant inhibitory effect on germination, root and shoot elongation and development of lateral roots of recipent crops. The study also revealed that the inhibitory effect was much more pronounced in primary root and lateral root development rather than in shoot development and germination.

Allelopathic compounds negatively affect root and shoot length (Hu and Jones, 1997; Inderjit and Dakshini, 1992; Xavier, 1990; Murthy et al., 1995; Qasem, 1995; Mughal, 2000). Moreover, a reduction in root volume limits nutrient and water supply to shoots, consequently decreasing shoot growth. Allelopathic compounds can change the morphology of the recipent plant. Kim et al. (1995) found a change in the structure of root tips of the species *Raphanus sativus* var. *hortensis* after treatment with various concentrations of the leaf extract of *Pinus rigida*. Also Chon et al. (2002) found that coumarin inhibited root elongation and cell division of alfalfa which resulted in the thickness of the seminal roots being abnormally enlarged due to inhibition of the longitudinal root growth. Also, Vandermast et al (2002) found that chestnut blight, *Cryphonectria parsitica*, extracts lower germination rates of lettuce seeds. The radicals of extract-treated lettuce were significantly shorter and thinner, more easily broken, and were less likely to develop secondary roots compared to water-treated plants. Jayakumar et al. (1995) found that the aqueous extract of fresh leaves of *Caesalpania coriaria* (Jacq.) inhibited the growth of *Parthenium hysterophorus*; the decrease in plant dry

weight, shoot height, and leaf area was proportional to the increase in concentration of leaf extract. In some species growth of early roots was more sensitive to water-soluble extracts from other species (Dias and Moreira, 2002). Moreover, direct contact of the allelochemicals with the young root increased the possibility of injury (Drost and Doll, 1980). However, allelopathy can affect plant growth in some growth stages and have no effect in others and the response may depend on the environmental growth conditions and the concentration of compounds.

Plant species can be classified into three types according to their allelopathic sensitivities as target plants. The first group are susceptible species that show low germination and seedling growth when exposed to donor extracts. The second group are non-susceptible species that show high germination and seedling growth when exposed to extracts, and the third group are intermediate species that show moderate reduction in germination and seedling growth in extracts (Kil and Yun, 1992). Furthermore, it has been observed that allelopathic compounds of an inhibitor or allelopathic plant do not curtail the growth of all plants, but affect sensitive species in a particular stage of growth. For instance, extracts from *Urochloa mosambicensis* (Hack.) had no adverse effect on *Stylosanthes scabra* cv. Seca germination, but reduced Seca seedling root length (Hu and Jones, 1997*)*. In another study, Chaves and Escudero (1997) found that a falvonoid compound from *Cistus ladanifer* had no direct effect on the germination of *Cyndon dactylon* and *Rumex crispus*, but caused a reduction in cotyldon and root size and subsequent seedling development. Also, Jose and Gillespie (1998) found that growth reduction were greater in soybean than in corn when exposed to juglone

Allelopathy as a concentration phenomenon

Stimulatory and inhibitory effects of plant extracts are a function of concentration. Like all other growth regulators in soil, concentrations of allelochemical substances must reach appropriate threshold levels before inhibition of germination and/or growth can occur. Thus, allelopathy is viewed as a concentration-dependent phenomenon. To exert allelopathic effects on target plants, allelochemicals, after release into the environment, often need to persist in the soil; sometimes they must undergo biotic and abiotic changes, and reach sufficient concentrations to exert their effect (An et al., 2001). In soil, the

effective amount of allelochemical present is the difference between the amount produced and the amount inactivated. Thus, the amount of allelochemicals in the soil at a given time varies depending on the relative rates of addition, decomposition or inactivation, the soil properties, and other physiochemical conditions. However, during movement of allelochemical substances, the abiotic (physical and chemical) and biotic (microbial) properties of soil can limit the phytotoxicity of chemicals in terms of quality and quantity required to cause injury (Inderjit, 2001).

At low concentration, stimulatory effects have sometimes been observed for aqueous plant extracts that have inhibitory effects at higher concentrations under laboratory conditions. Saxena et al. (1996) found that aqueous extracts of pearl millet (cv. MH 179) increased the germination, root length and total dry matter of other pearl millet (*Pennisetum glaucum* (L.) R. Br) cultivars at 20 g./l but the highest concentration (80 $g.l^{-1}$) resulted in a 60% decline in seed germination compared to the control. Also Kil and Yun (1992) found that the water extracts of leaf tissue of *Artemisia princeps* var. *orientalis* slightly increased the dry weight of many target plants, whereas it proportionally inhibited dry weight at higher concentrations. A study by Buta and Spaulding (1989) found that low concentration extracts of *Festuca arundinacea* often stimulated the growth of several other grass species, while high concentrations inhibited growth. Norby and Kozlowski (1980) found a variable response of red pine (*Pinus resinosa* Ait) seedlings as target species to the water-soluble extracts of many allelopathic ground cover species in different concentrations. However, *Lonicera tatarica* extracts had the highest negative effect on dry weight of red pine seedlings compared to the control. On the other hand, red pine height growth was inhibited by all extracts. In contrast, Tefera (2002) found a stimulating effect of stem extracts of *Parthenium hysterophorus* on shoot length of *Eragrostis tef* at all concentrations tested. On the other hand, leaf extract inhibited seed germination and had a deleterious effect on shoot length. Allelopathic substance(s) may affect seed germination and seedling early growth or have no affect at all. Mallik and Prescott (2001) found that leaf or litter leachate of salal (*Gualtheria shallon*), a species known to be allelopathic, in different concentration had no effect on seed germination and primary growth of western hemlock (*Tsuga heterophylla*).

In addition to concentration, many other factors increase or decrease the activity of allelochemical substances such as age of the recipent and donor plant, method of preparing the extract, and type of soil. In some allelopathic species, age has no effect on allelopathic potential. For example, Goel et al. (1989) found that the inhibitory activity in *P. juliflora* did not change with age. Also, Djanaguiraman et al. (2002) determined the allelopathic activity of aqueous leachates of *Eucalyptus globulus* leaves, with leaf age (juvenile, mature and senescent) on seed germination and growth of green gram, black gram and cowpea. The results indicated that the leaf leachates of *E. globulus* significantly decreased the germination of black gram, green gram and cowpea as compared to a control. Among the various ages of leaves used, senescent leaves were more inhibitory than the mature or juvenile ones.

In allelopathy studies, extraction methods are very important when acquiring allelochemical substance(s). However, there are two generally recognized methods for extracting allelopathic substances from plants. One method employs using organic solvents such as methanol to release the allellochemical substance(s), or alternatively use of hot or cool distilled water, and soaking plant tissues for a specific time. It is clear that the effect of the extracts depends on the efficiency of the extraction method. The distilled water method is the most commonly used in allelopathy studies and more nearly approximates the process that occurs in nature (Alam et al., 2001; Matizha and Dahl, 1991; Noor et al., 1995; Nilsson and Zackrisson, 1992; Butcko and Jensen, 2002; Chou et al. 1998; Conway and Smith, 2002; Escudero et al., 2000; Goel et al., 1989; AL-Humaid and Warrag, 1998; Hu and Jones, 1997; Jobidon and Thibualt, 1981). Studies have indicated that boiling during extraction had a variable effect on extract activity (Peters et al., 1986; Jonsen et al. 1984). Ahn and Chung (2000) found that warm water extracts were more phytotoxic than hot extracts. Hot water extraction is believed to result in allelochemical binding or degradation, consequently reducing its inhibitory effect on the tested species.

Soil, as a growing medium, is important in increasing or reducing the accumulation of allelochemical compounds. Accumulation can depend on soil structure and texture. Conditions for allelopathic interference with host species were most favorable in soils that were poorly drained, poorly aerated, shallow, and showed high

colloidal content (del Moral and Muller, 1970) because these factors permit toxin concentrations to reach physiologically significant levels. Interestingly, sandy soils are much more effective in enhancing allelopathic responses as a result of their low field capacity. Sandy soils, therefore, reduce the diluting effect that water normally has with regard to the toxic compounds (Goel et al., 1989). Patrick (1971) observed a greater inhibition of corn height and weight in sandy and light textured soils. Also, soils with high organic matter seemed to adsorb and inactivate coumarin more when compared to sandy soils with lower organic matter content (Takahashi et al., 1994).

In most ecosystems, there are differences in climatic patterns within and between years. These in turn are important in affecting temporal variability of secondary metabolite production. Temporal variation in the production of these metabolites could induce corresponding shifts in the biotic interaction among plants (Nilsson et al. 1998). Nilsson et al. (1998) found large differences in production of phenolic compounds between years and with shoot age. Richardson and Williamson (1988) found that the inhibitory effects of sand pine (*Pinus clausa*) varied based on the month that the samples were collected and was highly correlated with monthly precipitation. Lodhi (1978) found that the degree of germination and radical growth inhibition from decaying leaf litter of of sycamore and red oak showed a stronger inhibition of germination and radical growth of brome grass in January than in April and August.

Physiological mechanisms of allelopathy

Nutrient uptake

Allelochemical compounds can result in negative effects on growth and development by affecting one or more of the physiological functions of the recipent plants. One of the documented effects of allelopathic agents includes a reduction in mineral uptake, such as Ca, Mg and S (Walker et al., 1991). Inderjit and Dakshini (1992) found that the concentration of Mg, Zn and P were higher, but K was lower, in the shoot of plants that were grown in soil treated with water soluble compounds of the species *Pluchea lanceolata.* Gogoi et al. (2002) found a reduction in N, P, K content of rice shoots when treated with aqueous extracts of some allelopathic weeds.

An alteration of the mineral content of the plant has been observed in many studies with specific allelochemicals. Baziramakenga et al. (1994) found a reduction in the amount of P, K, Mg, Mn, Cl, SO, Zn and Fe on the roots of soybean (*Glycine max* L.) when treated with benzoic acid and trans-cinnamic acid but shoots showed greater accumulation of Ca, Mg, and Zn, while P and Fe content was reduced. For phenolic acids, Alsaadawi et al. (1986) found a reduction in the uptake of N, P, K, Fe, and Mo with increasing concentrations of phenolic acids.

The allelopathic effects of some species may be direct by affecting plant nutrition or indirect by, for example, affecting plant–microorganism interactions. Norby and Kozlowski (1980) found a reduction in phosphorus concentration in needles of red pine that were treated with water extracts of the foliage of *Lonicera tatarica*. The direct effects for some phenolic acids, are the result of complexes that form with plant nutrients in the soil (Kruse et al, 2000), interfering with the nutrient uptake. This in turn can cause lower concentrations of nutrients in plant tissues (Einhellig, 1986). On the other hand, ferulic acid has been found to increase the uptake of certain ions, but this effects is dependent on the age of the acceptor (Einhellig, 1986). Some plants exude certain compounds into their rhizosphere, which changes the availability of nutrients of the surrounding soil. These compounds are, among others, organic acids such as citric acid, fumaric acid amino acids, and phenolics or phytosiderophores. Most of these compounds work by changing the pH of the soil and/or function as chelating agents of nutrients (Marschner, 1995). This, of course, affects the donor plant as well as other plants with roots entering this rhizosphere. In most cases this effect will be positive, as the purpose of these chemicals is to increase the availability of nutrients most needed by the plant. However, the effect can be negative if leaching or depletion is the result, or if two succeeding plants, or two plants with overlapping roots are in need of different nutrients, as the compounds exuded to make one mineral more available can make another mineral less available. One example is *Pluchea lanceolata,* the presence of which has been noted to influence soil chemical characteristics such as pH, electrical conductivity and content of potassium and chloride of the soil in the vicinity of its roots (Kruse et al., 2000). The *Pluchea lanceolata*-infested soils had significant negative effects on seedling growth of various crop plants compared to non-infested soils. It is therefore possible that the effect of allelopathic

plants can be a result of the allelochemicals in the soil and/or to changed soil nutrients. Generally, phenolic acids and many other allelochemicals are considered to have an important influence on nutrient cycling in terrestrial ecosystems. Phenolic monomers and phenolic acids can form complexes with nutrients and, thus, influence the nutrient availability and nutrient turnover in soil (Appel, 1993; Kuiters, 1991).

Allelochemicals can decrease the absorbtion of mineral nutrients by changing the balance between absorbed forms of nutrients (Yang et al., 2002). For example, some plant roots exude allelochemicals, such as volatile terpenoids, which may inhibit the oxidation of ammonium to nitrate, by affecting the nitrifying microorganisms. This will of course lead to a change in N-availability, as most plants prefer to take up N as NO_3^-, although some plants can take it up in either form. In addition to their effect on nutrient form and balance, allelochemicals can change the water relations of target plants, as well as inhibit root hair formation which in turn leads to changes in the uptake and transport of mineral nutrients thus reducing the plant growth and development.

Photosynthesis and chlorophyll content

Allelochemical compounds can affect photosynthesis in a number of species by decreasing chlorophyll content (Lodhi and Nickell, 1973). In rice, *Oryza sativa*, Yang and co-workers (2002) found the production of chlorophyll and porphyrin in leaf tissue was increasingly inhibited as phenolic allelochemical concentrations increased. In this study it was suggested that Mg chelatase may be the major target of the phenolic allelochemicals. Zeng et al. (2001) found that secalonic acid F (SAF) reduced chlorophyll content at high concentrations resulting in yellowing of seedling leaves of *Sorghum vulgare*, hairy beggarticks (*Bidens pilosa* L.), and barnyardgrass (*Echinochloa crus-galli* (L.) Beauv.). Interestingly, chlorophyll content of sorghum increased when low concentrations of SAF were applied. Kumari and Kohli (1987) also found a reduction in chlorophyll content in Ragweed parthenium (*Parthenium hysteropborus*) as a result of autotoxicty of the species when it was treated with its own leaf leachates, a physiological response referred to as autotoxicity. Leachates were less effective in reducing chlorophyll content when added to the root system than when applied directly to the leaves. Studies such as Viles and Reese (1996) corroborate the above mentioned observations. They

found that the chlorophyll content of lettuce seedlings decreased as a result of adding aqueous extracts of *Echinacea angustifolia* D.C. Extracts from *E. angustifolia* shoots were less inhibitory to seed germination and growth of the tested species than root extracts. In contrast to most observations on allelochemical effects on chlorophyll content, Pandey (1994) found that the amounts of chlorophyll a, b, and total chlorophyll in the leaves of paddy rice seedlings growing in residues of the allelopathic species parthenium (*Parthenium hysterophorus* L.) were comparable in amounts to that of seedlings grown in distilled water. Einheling and Ramussen (1979**)** found that soybean plants treated with different phenolic acids had less chlorophyll than untreated plants. However, grain sorghum [*Sorghum bicolor* (L.)] seedlings exhibited no change in chlorophyll content even though growth was inhibited. Chlorophyll content, in some instances, has been shown to increase in some species when exposed to what would normally be considered allelochemical-containing species. El-Darier and Youssef (2000) observed an increase in the biosynthesis of photosynthetic pigments in *Lepidium sativum* L. at 50% strength of the alfalfa aqueous extracts used in this particular study. The chlorophyll a/b ratio attained its maximum at the highest concentrations used.

Materials and methods*:*

Plant Material

Seeds of *Ziziphus spina-christi* for this study were collected from trees grown at the Agriculture Research Station in Derab, Riyadh, Saudi Arabia (latitude 24º 34′, longitudinal 46º 43′). The research station is located at an elevation of 500 m and receives an average rainfall of 83 mm/y. Evaporation is measured at 2739.5 mm/y. The average daily maximum temperature is 48ºC in August and 31.8ºC in December. Seeds of *Prosopis juliflora* were obtained from Lawyer Nursery Inc, USA. Leaves of *Z. spina-christi* and *P. juliflora* were collected from the above described site in Saudi Arabia and then air shade dried at 48ºC for 72 hours. Leaves were ground to a powder stored in colored plastic containers and saved at room temperature until further use.

Z. spina-christi and *P. juliflora* were grown from seed in containers under greenhouse conditions (WVU plant sciences greenhouses). In order to establish the plants, seeds were sown in sterilized sand at a approximate depth of 3 mm. After germination two seedlings were transplanted per 10 × 10 × 32 cm plastic pot filled with 2840 g of sterilized sand. Nutrient poor sand was used to establish plants as recommended by Alsaadawi et al. (1986). A total of 336 pots were established, 168 pots with *Z. spina-christi* and 168 pots with *P. juliflora* seedlings. Seedlings were grown until the third true leaf was completely unfolded (approximately 8 weeks) before treatments were applied. A complete nutrient solution, Johnson's solution (Johnson et al. 1957), was used to fertilize the seedlings once a week with 200 ml per pot throughout the experiment. Seedlings were watered with tap water as needed.

Extract preparation

Dry powder of leaves of *Z. spina-christi* and *P. juliflora* was used to prepare extracts. Powdered leaves were soaked in distilled water for 24 h at 27 ºC at 5, 20, 40, 60, and 100 g./l (W:V). The extracts were filtered through eight layers of cheese cloth to remove solid materials. Next the extracts were filtered through No. 1 Whatman filter paper under suction. Leaf water extracts were stored in plastic containers in a refrigerator at 5 ºC for up to 2 weeks. New extracts were prepared as needed. Distilled water was used as the control treatment (0 g /l).

Treatments

Seedlings were irrigated weekly with 120 ml of the leaf extracts or the same amount of distilled water with twenty-eight pots representing each concentration (treatment). Four pots per treatment were harvested at thirty-day intervals for a period of seven months. The experiment was laid out in a randomized complete block design. Four blocks were used and every concentration was replicated 7 times within the block for a total of 84 pots per block. For every concentration combination four pots represented the four blocks that were harvested. In every block, pots were marked from 1 to 84. Extract's containers with a capacity of 120 ml were given corresponding numbers from 1 to 84 with the numbers on pots matching the numbers on the extract containers. For example, the container with title "B1, AP, 100 g/l, 56" corresponds to a pot in block 1, the recipent is *Prosopis juliflora,* the extract concentration is 100 g/l and the pot number is 56. Seedlings were grown in the greenhouse at 30ºC (D/N) and a 13 hour photoperiod was provided by HPS lights (400 watts).

Data Collection

Every month and before harvesting plants for further measurements, the following data were collected: number of leaves, plant height (cm) from the cotyledon scars to the stem apex, and stem diameter (mm) at the cotyledon scar using a digital caliper (± .04 mm). Leaf number was counted for every seedling then leaf area was measured for all leaves with a leaf area meter (L-COR 3100 LiCor Inc., Lincoln, Nebraska, USA),

Root measurements included root length and root dry weight. Prior to measurement of roots, sand was removed gently with water to reduce root loss. Roots of the pair of plants in each container were separated, and then transferred to a moist plastic bag for individual measurements. Fresh weight measurement was not taken to avoid root destruction. Roots for every seedling were spread gently over a computer scanner (UMAX 4000U with a resolution 1200 dpi by 2400 dpi and dimensions 21.59 cm by 35.56 cm) then scanned at 600 dpi using Adobe Photoshop 5.5 (Adobe, 2001). The images were saved in tiff format to be analyzed later with WinRhizo basic software (Regent Instruments Inc, 2002). Before analyzing root images with WinRhizo, and to eliminate any duplicate measurements of root length because of shadows, editing of the

images was performed by removing shadows with Adobe Photoshop 5.5. After the scanning process, roots were dried at 75ºC for 48 hr and dry weights were obtained.

Chlorophyll content for each seedling was measured by using the methanol extraction method of Porra et al. (1989). After choosing leaves randomly from the upper third of the plant, leaves were sliced into small pieces and weighed. For every seedling, about 0.0270 g, ±0.0005, of fresh leaf tissue was transferred to a test tube and placed on ice. Two milliliters of cold methanol was added to every test tube with tissue, sealed with Parafilm and extracted overnight in the refrigerator at 5ºC. After 24 hr, a 0.5 ml sample of the extract was used to determine chlorophyll content spectrophotometerically at two wavelengths, 665.2 nm and 652.0 nm. Chlorophyll a and b content and total chlorophyll a+b were calculated using the following equations (Porra et al. 1989).

Chlorophyll concentrations in nmol/ml=

$$\text{Chl } a = 18.22\, A^{665.2} - 9.95\, A^{652.0}$$

$$\text{Chl } b = 33.78\, A^{652} - 14.96\, A^{652.0}$$

$$\text{Chls } a + b = 24.93\, A^{652.0} - 9.95\, A^{665.2}$$

Where A is absorbance at the given wavelength.

Statistical analysis

Measurements were averaged for every month then the overall average for all months was computed. Later means and standard errors for the averaged data were obtained then data sets contrasted using analysis of covariance where the alpha-level for all statistical analyses was set at 0.05. Since the two tested species are inherently different, the observations of the control for each species were used as a covariate to adjust treatment means. Dead seedlings were ignored from the analysis except in shoot and root dry weight. After performing the analysis of covariance, a t-test was used to compare least squares means to determine the significant sources of variation between concentrations.

Results

The interactions between the sources of variance, recipent by concentration**,** are the focus of results throughout this chapter, in order to test the hypothesis that the seedlings of a species are unaffected by extracts of varying concentrations from the other species.

The response behavior of the two species was dissimilar in most properties studied. Growth characteristics of *Ziziphus spina-christi* were significantly promoted by extracts of *Prosopis juliflora* at all concentrations except the highest concentration. Conversly, *P. juliflora* was significantly inhibited in most properties studied by increasing extract concentrations from *Z. spina-christi*.

Seedlings height

The analysis of covariance shows that regarding seedling height the interaction between recipent and concentration (P=0.0001) is highly significant (Table 1).

Prosopis juliflora as recipient

Leaf extracts of the native species *Z. spina-christi* generally have an adverse effect on height of seedlings of the invasive plant *P. juliflora* at high concentrations. Average heights of seedlings decreased with increasing leaf extract concentrations. The highest concentration (100 g/l) had the most negative effective (Table 2). There were significant differences among treatment means of different extract concentrations (Table 4, shaded cells) and (Fig. 1). There were no significant differences in means for heights between 5 g/l and 20 g/l (P=0.5763) and the differences were not significant between 100 g/l and 60 g/l (P=0.8814). Analysis of covariance showed that the three-way interaction between recipent by month by concentration was not significant during the 7 months growth period demonstrating that the allelopathic effect was consistent over the course of the study (Table 1). Although the three-way interaction was not significant, *P. juliflora* seedling height demonstrated a significant effect of concentration of *Z. spina-christi* extract over the growth period since the two-way interaction of months by concentration was significant (Appendix A, Table 1).

***Ziziphus spina-christi* as recipient**

The response of *Z. spina-christi* seedling heights to aqueous leaf extracts of the invasive species was almost the reverse of *P. juliflora.* Seedlings heights of *Z. spina-christi* were increased with increasing leaf extract concentrations except for the concentration of 100 g/l which reduced seedling heights (Table 2). A t-test indicated highly significant differences between height means of 5, 20, 40, and 60 g/l and 100 g/l (Table 4, non-shaded cells) and (Fig.1). The height of *Z. spina-christi* during the 7-month growth showed a reduction at the highest concentration of 100 g/l (Appendix A, Table 2).

Seedlings diameter

Analysis of covariance indicated that the interaction was significant between recipent and the concentrations on seedling diameter (P=0.0001) (Table 3).

***Prosopis juliflora* as recipient**

P. juliflora average seedling diameters declined with increasing aqueous leaf extract concentrations of *Z spina-christi* where the minimum diameter was 3.21 mm with 100 g/l (Table 2). Diameter showed significant differences between the main effect of concentrations (Table 5, shaded cells, Fig. 2) but diameter means at the low concentrations, 5 g/l and 20 g/l, were not significantly different from each other. Stem diameter was significantly different between concentrations within months and between months. Extracts of *Z. spina-christi* stimulated the stem diameter at the low concentration (5 g/l and 20 g/l) but at the highest concentration (100 g/l) the diameter declined between treatments and among months (Appendix A, Table 1).

***Ziziphus spina-christi* as recipient**

Seedlings of *Z. spina-christi* responded to the aqueous leaf extracts of *P. juliflora*. Seedlings having increasing diameters with increasing concentrations of extract except at 100 g/l where diameter declined (Table 2). Concentration means had significant differences where 100 g/l was significantly different from 5, 20, 40, and 60 g/l (Table 5, Fig. 2). The diameter of *Z. spina-christi* seedlings showed a reduction over time at the concentration 100 g/l, but at the concentration of 60 g/l the diameter increased with time (Appendix A, Table 2)

Shoot dry weight

For shoot dry weight the recipent by concentration interaction was highly significant (P=0.0001) (Table 6). Thus different recipent species responded differently according to extract concentration and type of donor.

***Prosopis juliflora* as recipient**

The extracts of *Z. spina-christi* showed a negative impact on dry weight of *P. juliflora* seedling shoots (Table 7, Fig. 3). Shoot dry weight declined with increasing concentrations. Average shoot dry weight was the highest (4.02 g) at the lowest concentration of 5 g/l and lowest (1.88 g) at the highest concentration of 100 g/l. Overall the means of concentrations were significantly different (Table 8, shaded cells) but comparisons between 5 g/l and 20 g/l as well as 20 g/l and 40 g/l were not significantly different. Even though shoot dry weight increased with time, biomass allocation toward shoots in seedlings at the high concentration (100 g/l) was reduced compared to seedlings at the lower concentrations, 5 g/l and 20 g/l (Appendix A, Table 3).

***Ziziphus spina-christi* as recipient**

Shoot dry weight for *Z. spina-christi* increased with increasing aqueous leaf extract concentrations from *P. juliflora*, but at the highest concentration (100 g/l) shoot dry weight decreased (Table 7). Mean dry weights for seedlings treated with extract concentrations of 5 g/l, 20 g/l, and 40 g/l were not significantly different (Table 8), but there were significant differences between these concentrations and the highest extract concentration (100 g/l). The increase in shoot dry biomass was observed with increasing growth period and with increasing concentration of *P. juliflora*. At the 100 g/l concentration biomass was slightly reduced and less was allocated to shoots with time (Appendix A, Table 4).

Root dry weight

The interaction between concentration and recipent was significant for root dry weight (P=0.0001) (Table 9). This significant interaction indicated that the two species respond differently when treated with extracts of the other species. In the following paragraphs, the nature of these responses are explained for each species, as well as the responses to extract concentration and time of treatment.

***Prosopis juliflora* as recipient**

Root dry weight decreased with increasing concentration of leaf extracts of *Z. spina-christi* (Table 7, Fig. 4). There were significant differences in means between 5 g/l and 60 g/l (P=0.0154), and 5 g/l and 100 g/l (P=0.0006), and 20 g/l and 60 g/l (P=0.0540) (Table 10, shaded cells). There was no significant difference between the lowest two concentrations, 5 g/l and 20 g/l, for means of root dry weight. With time (months), root dry weight did not increase as rapidly at the highest two concentrations 60 g/l and 100 g/l compared to the other concentrations (Appendix A, Table 3).

***Zizphus spina-christi* as recipient**

Table (7) shows that *Z. spina-christi* increased its root dry weight with increasing concentration of aqueous leaf extracts of *P. juliflora*. However, at the highest concentration (100 g/l) there was an extreme reduction in root dry weight. Results of a t-test show that there are no significant differences between concentration 5, 20, and 40 g/l regarding the response of root dry weight of *Z. spina-christi* (Table 10, non-shaded cells). Root dry weight of *Z. spina-christi* generally increased with time and with increasing concentration. At the highest concentration (100 g/l) root dry weight was extremely reduced (Appendix A, Table 4).

Chlorophyll *a*

Analysis of covariance showed a very highly significant interaction between recipent and concentration (treatments) for chlorophyll *a* leaf content (P=0.0004) (Table 11). Chlorophyll *a* for both tested species tended to be reduced with increasing aqueous extract concentrations.

***Prosopis juliflora* as recipient**

Chlorophyll *a* content in *P. juliflora* was reduced with increasing concentration of aqueous leaf extracts of *Z. spina-christi* (Table 12, Fig. 5). There was a significant difference between concentrations (treatments) (Table 13, shaded cells). In *P. juliflora* increasing time and concentration of *Z. spina-christi* generally reduced leaf content of chlorophyll *a* (Appendix A, Table 5).

***Ziziphus spina-christi* as recipient**

Aqueous leaf extract of *P. juliflora* reduced chlorophyll *a* content in leaves of *Z. spina-christi* (Table 12, Fig. 5). A t-test for treatment means indicted no significant differences between 5 g/l, 20g/l, 40 g/l, and 60 g/l but there was a significant difference between all treatments and the 100 g/l treatment (Table 13, non-shaded cells). With time, leaf content of chlorophyll a in *Z. spina-christi* showed a reduction when treated with 100 g/l of *P. juliflora* extract (Appendix A, Table 6).

Chlorophyll *b*

Table 14 shows the analysis of covariance for the interaction between recipent and extract concentrations (treatments) on leaf chlorophyll *b* content, which was highly significant (P=0.0001).

***Prosopis juliflora* as recipient**

Leaf extracts of *Z. spina-christi* affect chlorophyll *b* content in *P. juliflora* leaves (Table 12, Fig. 6). A t-test showed significant differences between means of all concentration except between 100 g/l, and 60 g/l (Table 15, shaded cells). Chlorophyll *b* content in *P. juliflora* declined over time and this decline was greater with higher concentrations of extract (Appendix A, Table 5).

***Ziziphus spina-christi* as recipient**

With concentrations of 5 g/l and 20 g/l of *P. juliflora* extract, chlorophyll *b* increased in *Z. spina-christi*. At 100 g/l, leaf content of chlorophyll *b* declined (Table 12, Fig. 6). Differences between means of treatment 5 g/l, 20 g/l, 40 g/l, and 60 g/l were not significant but there were highly significant differences between these treatments and the 100 g/l concentration (Table 15, non-shaded cells). Leaf extracts of *P. juliflora* at 100 g/l caused a reduced leaf content of chlorophyll *b* in *Z. spina-christi* with time (Appendix A, Table 6).

Total chlorophyll *ab*

Analysis of covariance (Table 16) shows a highly significant interaction between recipent and concentration on the chlorophyll *ab* leaf content (P=0.0001).

***Prosopis juliflora* as recipient**

Total chlorophyll *ab* in *P. juliflora* leaves was lower as applied extract concentrations of *Z. spina-christi* increased (Table 12, Fig. 7). At the lowest concentration of 5 g/l the amount of chlorophyll *ab* was 11.31 nmol/ml but at the concentration of 60 g/l total leaf chlorophyll content was only 6.47 nmol/ml. The least squares analysis of means indicated where significant differences between concentrations on total chlorophyll *ab* content occurred (Table 17, shaded cells). Extract concentrations of 60 g/l and 100 g/l produced no significant difference between means for chlorophyll *ab* (P=0.7409). Total chlorophyll *ab* concentration was erratic during the growth period of 7 months for the 5 g/l, 20 g/l, and 40 g/l concentrations. At the highest extract concentrations of 60 g/l and 100 g/l there was a reduction with time in total chlorophyll *ab* content (Appendix A, Table 7).

***Ziziphus spina-christi* as recipient**

There was a slight decrease in leaf content of total chlorophyll *ab* with increasing aqueous leaf extract of *P. juliflora* through the 60 g/l treatment (Table 12, Fig. 7). At 100 g/l there was a sharp decline at total chlorophyll *ab* content. A t-test (table 17) indicated a highly significant difference between means of the highest concentration (100 g/l) and all other treatments (P=0.0001). However, there were no significant differences among the other treatments means for total chlorophyll *ab*. At the 100 g/l concentration there was also a reduction in total chlorophyll *ab* over time (Appendix A, Table 8). For all extract concentrations other than 100 g/l total chlorophyll *ab* concentrations were erratic over time (Appendix A, Table 8).

Leaf Area (LA)

There was a highly significant interaction between recipent and concentration for leaf area of seedlings (P=0.0001, Table 18).

***Prosopis juliflora* as recipient**

Leaf area of *P. juliflora* seedlings decreased with increasing leaf extract concentrations of *Z. spina-christi* (Table 19, Fig. 8). The 100 g/l concentration reduced the leaf area by 55% compared to the 5 g/l concentration. A t-test showed significant differences between treatment means (Table 21 shaded cells). Leaf area in *P. juliflora* seedlings generally increased over time but with increasing concentrations of *Z. spina-christi* extract, increases in leaf area was more erratic with time (Appendix A, Table 9).

***Ziziphus spina-christi* as recipient**

For *Z. spina-christi* the response to aqueous extract concentrations of *P. juliflora* is presented in Table 19 and Figure 8. *Z. spina-christi* seedlings treated with an extract of 5 g/l were not significantly different from those treated with 100 g/l (P=0.6020) but differences were significant among 100 g/l and 20 g/l, 40 g/l, and 60 g/l (Table 21). Leaf area in *Z. spina-christi* varied erratically with increasing concentration and generally increased over time (Appendix A, Table 10). Seedlings treated with 100 g/l had the highest leaf area size at the end of growth period as compared to other concentrations.

Number of leaves

There was a highly significant interaction between treatments and recipent for the number of leaves produced by seedlings (Table 20).

***Prosopis juliflora* as recipient**

Number of leaves in *P. juliflora* generally decreased with increasing extract concentration of *Z. spina-christi* (Table 19, Fig. 9). A t-test indicated that the only significant difference was between means of 5 g/l and 100 g/l (P=0.0307) (Table 22 shaded cells). Number of leaves in *P. juliflora* increased with time except for the last month where the number decreased, especially at concentrations of 40 g/l and higher (Appendix A, Table 9).

***Ziziphus spina-christi* as recipient**

Leaf extracts from *P. juliflora* of increasing concentrations increased the number of leaves in *Z. spina-christi* at all levels except the 100 g/l concentration (Table 19, Fig. 9). There were significant differences between treatments means (Table 22). Among months, the number of leaves generally increased with increasing concentration but was reduced at 100 g/l. Over time the number of leaves produced by *Z. spina-christi* did not follow a linear pattern (Appendix A, Table 10).

Total root length

Interaction between recipent and treatments for total root length was highly significant (P=0.0001, Table 23). However, the response was different for the two tested species.

***Prosopis juliflora* as recipient**

Average total root length of *P. juliflora* was negatively associated with concentration of extract from *Z. spina-christi*. Total root length decreased by about 30% as leaf extract concentrations increased from 5 g/l to 100 g/l (Table 24, Fig. 10). The mean for 5 g/l was significantly different from 60 g/l and 100 g/l. In addition, 40 g/l was significantly different from 100 g/l (Table 25, shaded cells). Total root length increased with time within the treatments but there appeared to be a lagged growth for roots when higher concentrations of extract were used. The total root length for seedling with higher concentration seemed to almost recover after 5-months. This could be due to development of a "resistance" to the extract or perhaps the pot volumes had begun to limit root development in the lower concentration treatments (Appendix A, Table 11)

***Ziziphus spina-christi* as recipient**

P. juliflora leaf extracts apparently caused a small increase in total root length for *Z. spina-christi* (Table 24, Fig. 10). However, at the highest concentration, 100 g/l, the total root length was reduced by about 35%. The comparison of means for total root length indicated significant differences between 5, 20, 40, and 60 g/l against the highest concentration of 100 g/l (Table 25). With time the total root length of *Z. spina-christi* increased at all concentration but for 100 g/l the increase was lower in contrast with the other treatments (Appendix A, table 12).

Root surface area

The analysis of covariance (Table 26) demonstrates a highly significant effect of the interaction between recipent and extract concentration on the root surface area (P=0.0001).

***Prosopis juliflora* as recipient**

Root surface area of *P. juliflora* decreased with application of increasing extract concentrations of *Z. spina-christi* (Table 24, Fig. 11). At the lowest concentration of 5 g/l the root surface area was 133.81 cm^2 but at the highest concentration of 100 g/l the root surface area was 99.93 cm^2 which is a reduction in percentage of root surface area between the two concentrations of about 25%. There were significant differences between most concentration means (Table 27, shaded cells), although means of 60 g/l and 100 g/l were not significantly different (P=0.4139). Root surface area increased with time except for the six-month period for 20 g/l and 100 g/l (Appendix A, Table 13).

***Ziziphus spina-christi* as recipient**

In contrast to *P. juliflora, Z. spina-christi* seedlings increased their root surface area with increasing concentration of leaf extracts of *P. juliflora* except at the 100 g/l concentration (Table 24, Fig. 11). At the 100 g/l concentration there was a reduction in root surface area of approximately 38% compared to the 60 g/l value. The comparisons between root surface area means indicated that significant differences occurred between the 5, 20, 40, and 60 g/l concentrations compared to the highest concentration of 100 g/l (Table 27). There also was a significant difference between means of 5 and 60 g/l (P=0.001). Root surface area in *Z. spina-christi* varied between months but it tended to increase with time at all concentrations except 100 g/l where the surface area peaked at month 5 and declined in months 6 and 7 (Appendix A, Table 14).

Number of root tips

The analysis of covariance (Table 28) indicated a significant interaction between recipent and the extract concentration on number of root tips (P=0.0029). As with other variables, the two species responded differently to treatment with aqueous extracts from the other species.

***Prosopis juliflora* as recipient**

Number of root tips in *P. juliflora* was affected by *Z. spina-christi* leaf extracts by producing a decreasing number of root tips with increasing concentrations of extract (Table 24, Fig. 12). The highest average of root tip numbers was 2547.75 with the treatment 5 g/l and the lowest number was 1843.39 with 100 g/l (Table 24). There were significant differences between treatments as to their effect on number of tips (Table 29). Significant differences for number of tips occurred between 5, 20 and 40 and 100 g/l. There was a consistent reduction in tips with increasing extract concentrations. The number of tips generally increased with time but, there was considerable variation with some increasing and others decreasing (Appendix A, Table 13)

***Ziziphus spina-christi* as recipient**

All concentrations except the 100 g/l concentration of *P. juliflora* leaf extract positively affected number of root tips in *Z. spina-christi* seedlings (Table 24). The 100 g/l concentration caused a reduction in the average number of tips. A t-test indicated that there were no significant differences between treatments (concentrations) 5, 20, 40, and 60 g/l regarding number of tips but there was a significant difference between these treatments and the highest concentration, 100 g/l (Table 29, Fig. 12). Numbers of root tips for *Z. spina-christi* increased with time (Appendix A, Table 14). For all months, seedlings treated with 100 g/l had the lowest number of tips compared to other treatments.

Root volume

The analysis of covariance indicated a highly significant interaction between recipent by treatments on root volume (P=0.0001, Table 29). However, the response of root volume for the two species tested was not consistent over the range of extract treatments.

***Prosopis juliflora* as recipient**

Increasing concentrations of *Z. spina-christi* leaf extract generally reduced root volume in *P. juliflora*. At the concentration of 5 g/l the volume was 1.38 cm^3 but the volume was about 24% less at 100 g/l (Table 24). There were significant differences between means of root volume among treatments. A significant difference occurred between 5 g/l and 60 g/l (P=0.0620), 5 g/l and 100 g/l (P=0.0182), and 20 g/l and 100 g/l (P=0.0603) (Table 31, shaded cells, Fig. 13). Root volume in *P. juliflora* increased with time but seedlings treated with 100 g/l of extract showed the lowest rate of volume increase (Appendix A, Table 15).

***Ziziphus spina-christi* as recipient**

Extract concentrations of *P. juliflora* significantly affected the root volume of *Z. spina-christi*. As extract concentrations increased the root volume increased except at the 100 g/l level, where the volume was reduced (Table 24, Fig. 13). The t-test revealed significant differences between means of most treatments. Only the comparison between extract concentrations of 20 and 40 and 20 and 60 g/l were not significantly different (Table 31, non-shaded cells). Seedlings of *Z. spina-christi* increased their root volumes over time. Seedlings treated with 100 g/l of extract showed a tendency for root volume to increase at a lower rate with time, resulting in a root volume for this treatment that was lowest during the growth period (Appendix A, Table 16).

Root:Shoot ratio

The interaction between recipent and concentration was significant for the variable root to shoot ratio expressed on a dry weight basis (P=0.0001, Table 32). The two species displayed a different response to the treatments.

***Prosopis. Juliflora* as recipient**

P. juliflora seedlings had an increased root to shoot ratio with increasing leaf extract concentrations of *Z. spina-christi*. At the highest concentrations, 100 g/l, the ratio was 0.75 but at the low concentration, 5 g/l, the ratio was 0.55 (Table 33). The mean of 5 g/l was significantly different from 60 g/l and 100 g/l (Table 34, Fig. 14). Means did not show significant differences between 60 g/l and 100 g/l (P=0.4127) or between 5 g/l and 20 g/l (P=0.5213) and between 40 g/l and 60 g/l (0.1686).

Ziziphus spina-christi as recipient

A decreasing root to shoot ratio in *Z. spina-christi* accompanied increasing the extract concentration of *P. juliflora* (Table 33). However, the trend was somewhat erratic. For example, there were significant differences between the 5 g/l and 40 and 100 g/l treatments, but not between 5 g/l and 60 g/l. Conversely, the 100 g/l treatment was significantly different from 60 g/l, but not 40 g/l (Table 34, Fig. 14).

Discussion:

The species tested showed two different and relatively consistent patterns of sensitivity to extracts from the other species for most properties studied. *Z. spina-christi* seedling appeared to benefit from the lower extract concentrations of *P. juliflora*, but seemed to reach a threshold between 60 g/l and 100 g/l, above which a dramatic negative effect occurred. For *P. juliflora* seedlings, treated with *Z. spina-christi* extracts generally demonstrated an incremental negative effect with increasing extract concentrations.

Although both roots and shoots of *P. juliflora* were negatively affected by *Z. spina-christi* extracts, the shift in relative allocation of biomass to root with increasing extract concentrations indicated by an increasing root:shoot ratio, reflected a condition of environmental stress. A shift in relative allocation of biomass to roots has been observed under conditions of environmental stress by other workers (Rutherford and Powrie, 1993).

The reduction in growth for *P. juliflora* may due to allelopathy that may induce inhibition of nutrient uptake (Ismail and Chong, 2002). Nutrient uptake correlates with root characteristics such as root length and the increase in extract concentration reduced root length. There was a strong correlation between root length and root volume (r=0.97, Table 35) and root volume and number of tips (r=0.70). This may help to explain the reduced growth of *P. juliflora* under increasing extract concentrations. In a similar study, El-Khatib and Abd-Elaah (1998) found that with the highest concentration of extract, 7.5% of *Zilla spinosa* (recipent species) experienced a highly significant reduction in root length compared to lower concentrations.

A reduction in chlorophyll content in response to allelopathy has been reported in a number of plants (Batish et al. 2002, Alsaadawi et al. 1986; Viles and Reese 1996; Colton and Einhelling 1980). In this study, it is not clear whether the observed loss in chlorophyll was due to degradation of chlorophyll already present in the plant or due to direct inhibition of chlorophyll biosynthesis. However, the loss of chlorophyll is expected to reduce photosynthetic ability and so the growth and development of the plant. It is possible that the reduction in chlorophyll is due to the reduction in ion uptake since some of the ions are involved in the chlorophyll structure such as Mg-prophyrin and/or in the metabolic pathway of chlorophyll biosynthesis (Rice, 1984; Yang et al., 2002). Reduction

in leaf numbers and leaf area may in turn reduce photosynthesis (r=0.88, Table 35). In *P. juliflora* the species sheds its leaves heavily under normal condition (Goel et al., 1989) but shedding increases under environment stress thus reducing the amount of chlorophyll-bearing tissue, therefore the rate of photosynthesis (Appendix B, Fig. 1, 2). *P. juliflora* allocated above-ground biomass toward stem instead of leaf, in contrast to *Z. spina-christi*. The correlation between root surface area and shoot dry weight (r=0.87, Table 35) and root surface area and root to shoot ratio (r=0.50, Table 35) may indicate an ability of the plant to shift its growth toward above- or below-ground parts to avoid or reduce stress impacts on growth. However, increasing root to shoot ratio in *P. juliflora* with increasing extract concentrations of *Z. spina-christi* may also be, in part, due to the reduction of leaf number as a result of leaf shedding.

The increase in dry matter of plants is linked to carbon fixation and any loss in efficiency of photosynthesis might be detrimental to growth (Einhellig, 1986). Osmotic potential of aqueous leaf extracts is probably not the cause of growth reduction in *P. juliflora* or *Z. spina-christi*. Del Moral and Cates (1971) determined that the osmotic potential of a large number of plant extracts that possessed allelopathic qualities did not occur in osmotically inhibitory concentrations. They drew the conclusion that growth inhibitions caused by the extracts were due to allelochemical properties of organic materials in the extracts.

The response of *P. juliflora* to *Z. spina-christi* extracts tends to be incremental. Conversely, *Z. spina-christi* seedlings experienced growth stimulation at low concentrations of *P. juliflora* extracts, possibly due to the presence of inorganic nutrients in the extracts (Butcko and Jensen, 2002; Heisey, 1990) that compensate for the small amount of toxins at the lower concentrations. At high extract concentrations, inhibition may have resulted when toxins reached threshold concentrations. In *Z. spina-christi* chlorosis was observed in plants treated with 100 g/l of *P. juliflora* (Appendix B, Fig 3) compared to seedlings grown at lower concentrations (Appendix B, Fig. 4, 5, 6). The yellowish color in *Z. spina-christi*, demonstrated a reduction in photosynthesis and resulted in decline in biomass. Similarly, Jayakumar et al. (1995) found an incremental reduction in dry weight, shoot height, leaf area, and total chlorophyll in *Parthenium*

hysterophorus L. with increasing leaf extract concentrations of *Caesaplinia coriaria* (Jacq.).

In *Z. spina-christ* root to shoot ratio decreased with increasing extract concentrations of *P. juliflora* and that may help explain the tolerance of *Z. spina-christi* to the phytoxic compounds in *P. juliflora* extracts (Appendix B, Fig. 7). High concentration of *P. juliflora* extracts resulted, not only in growth retardation in *Z. spina-christi*, but also leaf dehydration (Appendix B, Fig 8), causing shrinkage and a decrease in leaf area.

It is interesting that the concentration 60 g/l of *P. juliflora* leaf extract was found to inhibit the growth of different plant species by several authors but it stimulated shoot and root growth of *Z. spina-christ* (Appendix B, figure 6 and 11). It is well documented that biological activity of allelochemicals is concentration dependent, often with a response threshold. However, plant growth may be stimulated below the threshold, but mild to severe growth reductions may be observed above the threshold concentration, depending upon the sensitivity of the receiving species, the plant process, and the environmental conditions (Einhellig, 1986). This is apparently the case of *Z. spina-christi* and its response to *P. juliflora* extracts. Furthermore, *Z. spina-christi* has a greater tolerance for *P. juliflora* extracts than some other species as illustrated by its higher threshold.

Growth stimulation may be induced in alternative ways. Several phenolic acids have been found to promote the growth regulator indole acitic acid (IAA) while several others suppress IAA destruction (Lee et al., 1982). The effective quantity of an allelopathic substance is the difference between the amount produced and the amount inactivated (An et al., 2001). Thus, retention and the effective quantity of the inhibitor in soil depends upon the relative rate of addition, decomposition or inactivation. However, *Z. spina-christi* may have a mechanism to avoid/reduce the negative impacts of allelochemicals by maintaining inhibitor concentrations below the growth-inhibition threshold. Orcutt and Nilsen (2000) suggested that plants can tolerate allelochemicals due to (i) an ability to reduce uptake of allelochemicals at the root surface, (ii) compartmentalization of allelochemicals away from molecular target sites and (iii) detoxification of allelochemicals.

Conclusion

Results from this study were in contrast with other studies that emphasized *P. juliflora* as an allelopathic species via leaf extracts (Dhawan, 1995; Goel et al. 1989; Goel and Nathawat, 1990; Al-Humaid and Warrag, 1998; Noor et al. 1995; Sen and Chawan, 1970). The stimulatory effects of *P. juliflora* were not observed previously but this is the first study involving *Z. spina-christi* and allelochemicals respond differently in different plant species (Oudhia, 2000; Chick and Kielbaso, 1998).

In the present study aqueous leaf extracts were used to determine if allelopathic effects could be detected between two species. The study indicates no negative effect of soluble water extract of the invasive species *P. juliflora* on the growth of the native species *Z. spina-christi* except at the highest concentration of 100 g/l, which may or may not be realistic in field conditions. The magnitude of allelopathic interaction has been shown to be dependent upon the concentration and chemical stability of the active compounds, as well as upon the tolerance of the species. Conversely, extracts from *Z. spina-christi* had a consistent and incremental negative effect on *P. juliflora* seedlings. These results suggest that allelopathy is probably not the mechanism used by *P. juliflora* to invade the habitat of *Z. spina-christi*. However, generalizations can not be made about the effect of *P. juliflora* on the native species *Z. spina-christi* because the experimental conditions may not represent natural field conditions. The consistent decrease in biomass of *P. juliflora* indicates that this species is susceptible to the negative influences of *Z. spina-christi* under the conditions tested. The specific toxins were not identified and their concentrations in the extracts were not quantified in this study. Since this is the first study of its kind, investigating the interaction between the two species, more studies need to be conducted to demonstrate the nature of the interaction. The following questions appear relevant:

- Do leaf water-soluble extracts stimulate the natural release of allelochemicals from *P. juliflora* through litter decomposition which affect the growth of other species?

- Are the extracts from other plant structures (bark, pods, fruit, and flower) of *P. juliflora* and *Z. spina-christi* more or less allelopathic than those from leaves on the growth of other species?

- Is their any effect of season on the content and impact of the inhibitor?

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Tables and Figures

Table 1. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on height**.**

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control height	1	0.85335	0.85335	0.01	0.9112
Blocks	3	486.70315	162.2343	2.37	0.0717
Concentration	4	6170.4085	1542.602	22.54	<.0001
Recipent	1	914.17152	914.1715	13.36	0.0003
Recipent *concentrations	4	3837.7881	959.4470	14.02	<.0001
Months	6	12520.469	2086.744	30.49	<.0001
Months* concentrations	24	3370.9528	140.4563	2.05	0.0040
Recipent *Month	6	295.68243	49.28040	0.72	0.6339
Recipent*Month*Concentration	24	1562.6242	65.10934	0.95	0.5324

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

Table 2. Means and standard errors for the influence of aqueous leaf extracts of *Prosopis juliflora* on seedlings height and diameter of *Ziziphus spina-christi* and vice versa over a 7- month growth period.

	Recipient			
	P. juliflora[1]		*Z. spina-christi*[2]	
Concentrations (g/l)	**Height (cm)**	**Stem diameter (mm)**	**Height (cm)**	**Stem diameter (mm)**
5	50.16 ±3.51	4.00 ±0.22	36.80 ±2.65	3.18 ±0.16
20	48.92 ±3.67	3.92 ±0.22	38.44 ±2.61	3.41 ±0.18
40	43.68 ±3.63	3.74 ±0.23	34.20 ±2.87	3.24 ±0.20
60	32.65 ±2.70	3.43 ±0.22	40.35 ±2.63	3.76 ±0.20
100	32.98 ±2.18	3.21 ±0.19	25.33 ±3.04	2.64 ±0.23

-*P. juliflora*[1] as a recipient of *Z. spina-christi* leaf extracts
-*Z. spina-christi*[2] as a recipient of *P. juliflora* leaf extracts

Table 3. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on diameter.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control diameter	1	1.0295492	1.02954917	3.76	0.0538
Blocks	3	5.6757582	1.89191941	6.91	0.0002
Concentrations	4	13.4056966	3.35142414	12.25	<.0001
Recipent	1	8.37905684	8.37905684	30.62	<.0001
Recipent * Concentrations	4	10.2098695	2.55246738	9.33	<.0001
Months	6	65.7302740	10.9550457	40.03	<.0001
Months* Concentrations	24	9.3584855	0.38993689	1.42	0.0986
Recipent*Month	6	3.86463673	0.64410612	2.35	0.0322
Recipent *Month* Concentration	24	4.74223551	0.19759315	0.72	0.8257

-Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
-Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

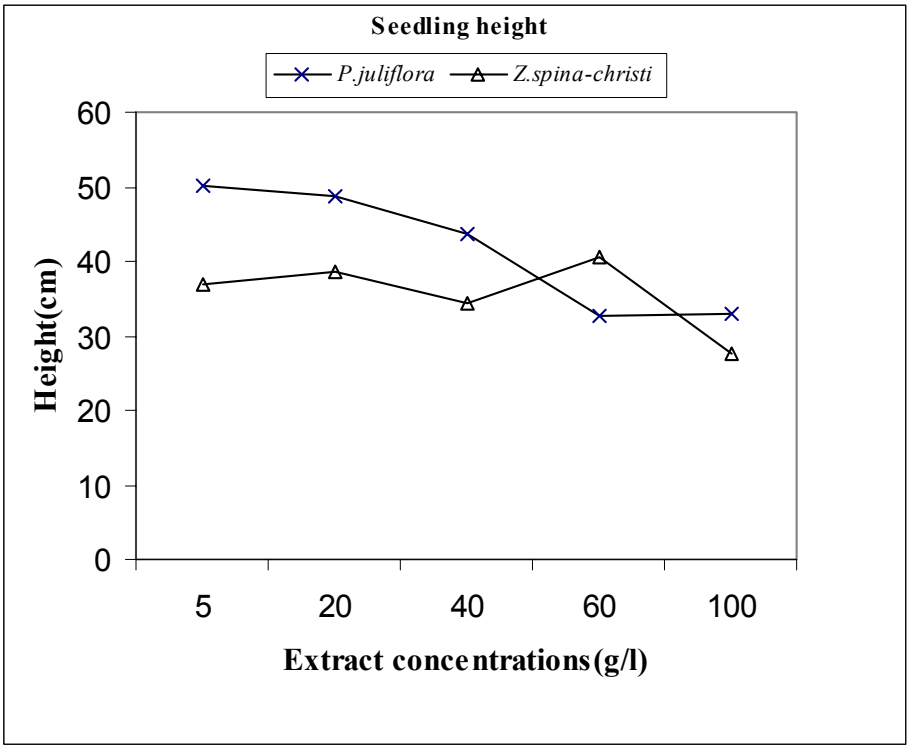

Figure 1. Effect of aqueous extract of *P. juliflora* (donor) on average height of *Z. spina-christi* (recipent) and effect of aqueous extract of *Z. spina-christi (*donor) on average height of *P. juliflora* (recipent).

Table 4. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent*concentration on height. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		-0.5597 0.5763	-2.9294 0.0038	-7.9207 <.0001	-7.7712 <.0001
20	-0.7414 0.4593		-2.3697 0.0188	-7.3609 <.0001	-7.2115 <.0001
40	1.1776 0.2404	1.9189 0.0564		-4.9913 <.0001	-4.8419 <.0001
60	-1.6064 0.1098	-0.865 0.3881	-2.7839 0.0059		0.1494 0.8814
100	3.7839 0.0002	4.4495 <.0001	2.7268 0.007	5.2260 <.0001	

-Shaded cells represent *P. juliflora.* - Non-shaded cells represent *Z. spina-christi.*

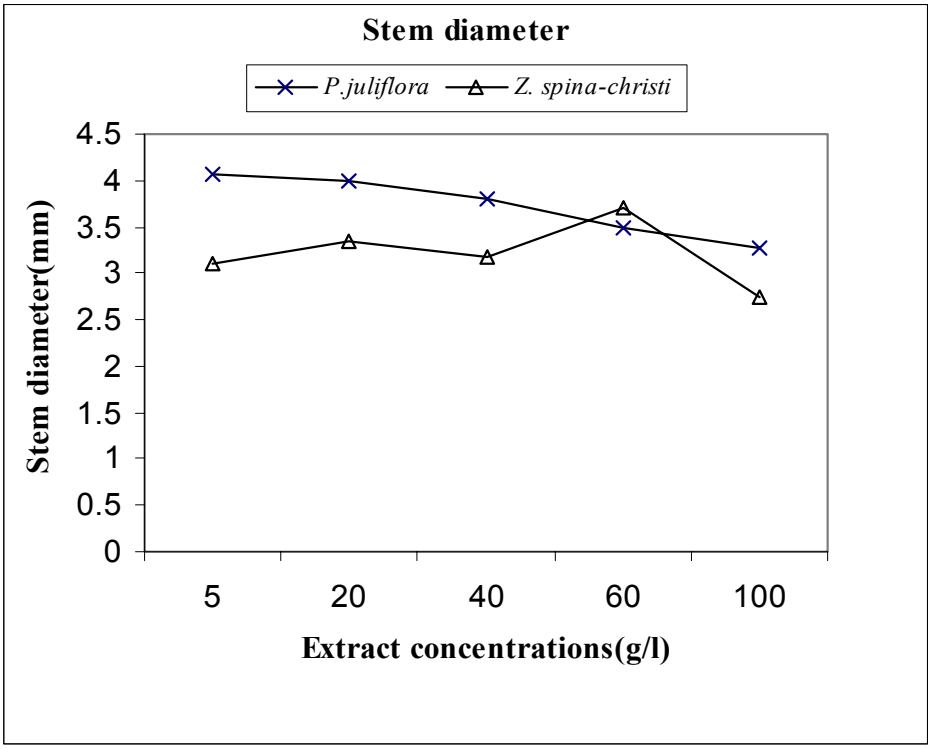

Figure 2. Effect of aqueous extract of *P. juliflora* (donor) on average diameterof *Z.spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi (*donor) on average diameter of *P. juliflora* (recipient).

Table 5. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent*concentration on diameter of *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		0.5748 0.5661	1.8393 0.0674	4.0489 <.0001	5.6711 <.0001
20	1.6988 0.0909		1.2645 0.2075	3.4742 0.0006	5.0963 <.0001
40	0.4598 0.6461	-1.2389 0.2168		2.2097 0.0283	3.8318 0.0002
60	4.1895 <.0001	2.4907 0.0136	3.7296 0.0003		1.6221 0.1064
100	-2.3524 0.0196	-3.8778 0.0001	-2.7653 0.0062	6.1143 <.0001	

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*

Table 6. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on shoot dry weight.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control shoot dry weight	1	0.0203144	0.0203144	0.02	0.8896
Block	3	11.728331	3.9094438	3.72	0.0123
Concentrations	4	144.83658	36.209146	34.46	<.0001
Recipent	1	0.1833599	0.1833599	0.17	0.6766
Recipent*concentrations	4	54.078032	13.519508	12.87	<.0001
Month	6	155.08759	25.847932	24.60	<.0001
Month*concentrations	24	65.156769	2.7148654	2.58	0.0002
Recipent*Month	6	2.2947382	0.3824564	0.36	0.9011
Recipent*Month*concentrations	24	27.236829	1.1348679	1.08	0.3692

-Concentrations : (5 g/l, 20/g/l, 40g/l, 60 g/l, and 100 g/l**)**
-Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

Table 7. Means and standard errors for the influence of aqueous leaf extracts of *Prosopis juliflora* on seedlings root and shoot dry weight *Ziziphus spina-christi* and vice versa over a 7-month growth period.

	Recipient			
	P. juliflora[1]		*Z. spina-christi*[2]	
Concentrations (g/l)	**Shoot dry weight (g)**	**Root dry weight (g)**	**Shoot dry weight (g)**	**Root dry weight (g)**
5	4.02 ±0.46	2.34 ±0.31	2.95 ±0.35	2.95 ±0.43
20	3.626 ±0.4	2.22 ±0.27	3.76 ±0.46	3.25 ±0.47
40	3.30 ±0.44	2.05 ±0.30	3.44 ±0.49	2.89 ±0.49
60	2.43 ±0.34	1.74 ±0.27	4.00 ±0.46	3.65 ±0.49
100	1.88 ±0.23	1.48 ±0.21	1.47 ±0.31	1.06 ±0.25

-*Z. spina-christi*[2] as recipent of *P. juliflora* leaf extracts.
-*P. juliflora*[1] as recipent of *Z. spina-christi* leaf extracts.

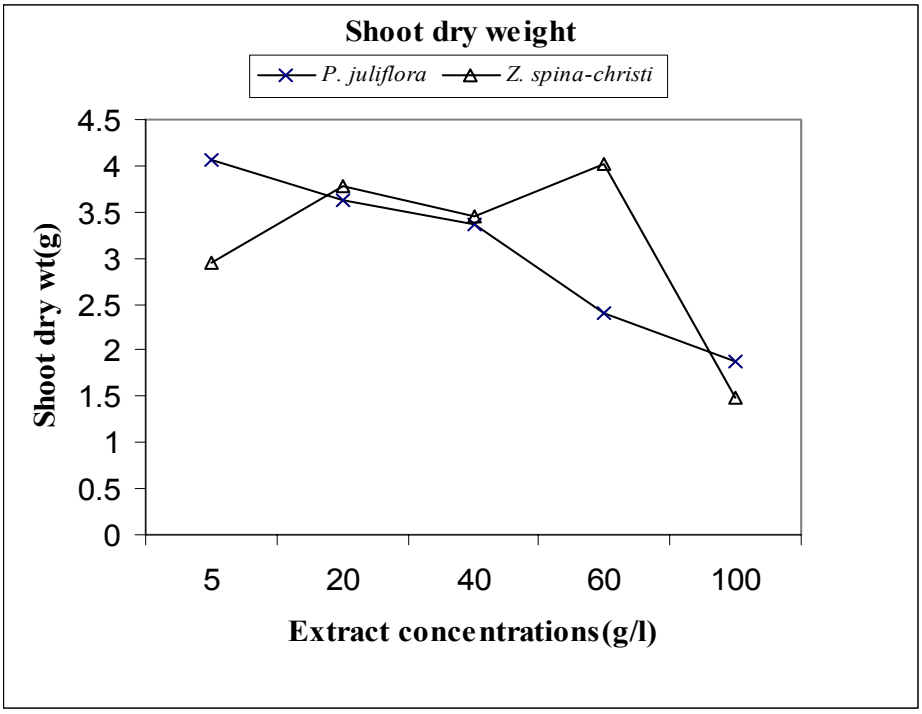

Figure 3. Effect of aqueous extract of *P. juliflora* (donor) on average shoot dry weight of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi (*donor) on average shoot dry weight of *P. juliflora* (recipient).

Table 8. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent *concentration on shoot dry weight of *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		1.5480 0.1232	2.5255 0.0123	5.8988 <.0001	7.7659 <.0001
20	2.9747 0.0033		0.9775 0.3295	4.3508 <.0001	6.2179 <.0001
40	1.8019 0.0731	-1.1728 0.2423		3.3734 0.0009	5.2405 <.0001
60	3.8326 0.0002	0.8579 0.3920	2.0307 0.0436		1.8671 0.0633
100	-5.4095 <.0001	-8.3843 <.0001	-7.2114 <.0001	-9.2421 <.0001	

- Shaded cells represent *P. juliflora*--- Non-shaded cells represent *Z. spina-christi.*

Table (9): Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on root dry weight**.**

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control root dry weight	1	2.4363948	2.4363948	2.88	0.0914
Blocks	3	10.0483408	3.3494469	3.96	0.0090
Concentrations	4	85.6508811	21.412720	25.28	<.0001
Recipent	1	35.8678069	35.867807	42.35	<.0001
Recipent *Concentrations	4	39.2884101	9.8221025	11.60	<.0001
Months	6	133.996839	22.332807	26.37	<.0001
Months*Concentrations	24	52.7689917	2.1987080	2.60	0.0002
Recipent *Months	6	23.0930917	3.8488486	4.54	0.0002
Recipent *Months*Concentrations	24	26.6469216	1.1102884	1.31	0.1593

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent : (*Ziziphus spina-christi* and *Prosopis juliflora***)**

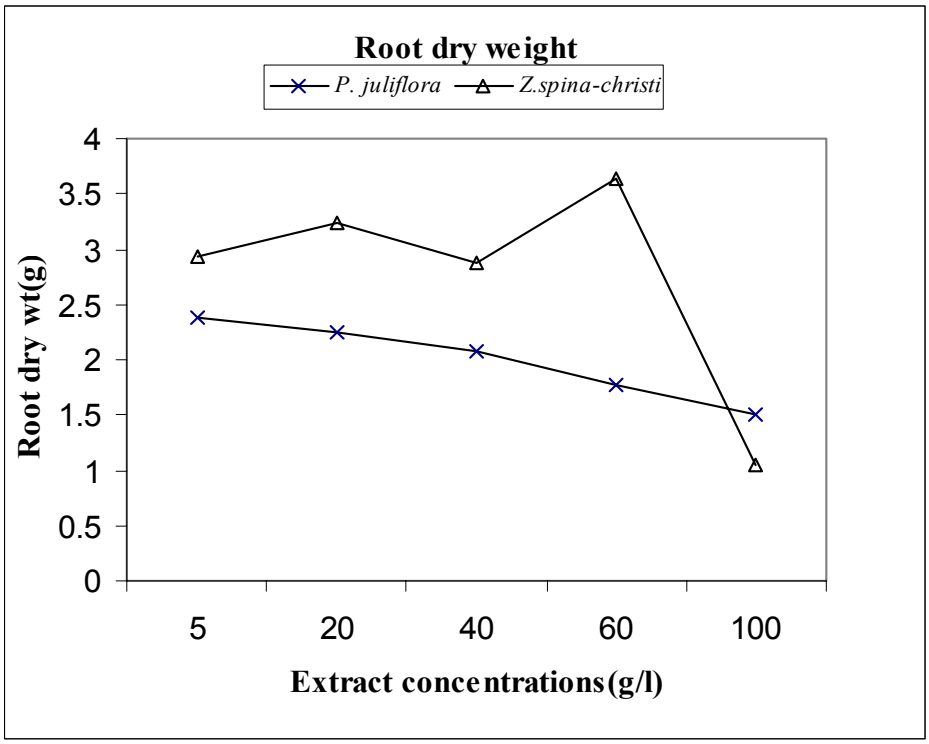

Figure 4. Effect of aqueous extract of *P. juliflora* (donor) on average root dry weight of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christ (*donor) on average root dry weight of *P. juliflora* (recipient).

Table 10. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent *concentration on root dry weight of *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		0.5054 0.6138	1.1856 0.2372	2.4436 0.0154	3.5012 0.0006
20	1.2476 0.2136		0.68016 0.4972	1.9382 0.0540	2.9958 0.0031
40	0.2141 0.8307	-1.4617 0.1453		1.2581 0.2098	2.3157 0.0216
60	2.8618 0.0046	1.61420 0.1080	3.0759 0.0024		1.0576 0.2915
100	-7.6528 <.0001	8.9004 <.0001	7.4387 <.0001	10.5146 <.0001	

- Shaded cells represent *P. juliflora* -Non-shaded cells represent *Z. spina-christi*.

Table 11. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on chlorophyll *a.*

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control Chlorophyll a	1	99.519437	99.519437	6.03	0.0149
Blocks	3	65.630411	21.876804	1.33	0.2671
Concentrations	4	957.15689	239.28923	14.51	<.0001
Recipent	1	2193.3519	2193.3519	132.99	<.0001
Recipent *Concentrations	4	358.82502	89.706255	5.44	0.0004
Months	6	385.67674	64.279457	3.90	0.0011
Months*Concentrations	24	581.76605	24.240252	1.47	0.0814
Recipent *Months	6	78.741290	13.123548	0.80	0.5743
Recipent *Months*Concentrations	24	335.56150	13.981729	0.85	0.6723

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent**: (** *Ziziphus spina-christi* and *Prosopis juliflora***)**

Table 12. Means and standard errors for the influence of aqueous leaf extracts of *Prosopis juliflora* on seedlings chlorophyll *a*, *b*, and total chlorophyll *ab* content of *Ziziphus spina-christi* and vice versa over a 7-month growth period.

	Recipient					
	P. juliflora[1]			*Z. spina-christi*[2]		
Concentrations (g/l)	**Chl *a*** *nmol/ml**	**Chl *b*** *nmol/ml**	**Chls *a+b***	**Chl *a*** *nmol/ml**	**Chl *b*** *nmol/ml**	**Chls *a+b***
5	8.79 ±0.62	2.23 ±0.22	11.31 ±0.77	17.41 ±0.72	4.58 ±0.45	22.00 ±0.87
20	8.53 ±0.68	2.149 ±0.35	10.99 ±0.93	17.34 ±0.89	5.45 ±0.37	22.80 ±1.14
40	6.16 ±0.78	1.60 ±0.22	7.80 ±0.94	16.40 ±1.00	4.76 ±0.30	21.16 ±1.26
60	5.05 ±0.58	1.06 ±0.24	6.47 ±0.72	16.88 ±0.94	5.00 ±0.32	21.89 ±1.14
100	5.45 ±0.96	1.31 ±0.30	7.152 ±1.21	9.61 ±1.49	2.38 ±0.45	12.00 ±1.89

- *P. juliflora*[1] as a recipient of *Z. spina-christi* leaf extracts
-*Z. spina-christi*[2] as a recipient of *P. juliflora* leaf extracts
-* chlorophyll per tissue in *nmol/g* = (*nmol/ml**2)/ (tissue weight (g))

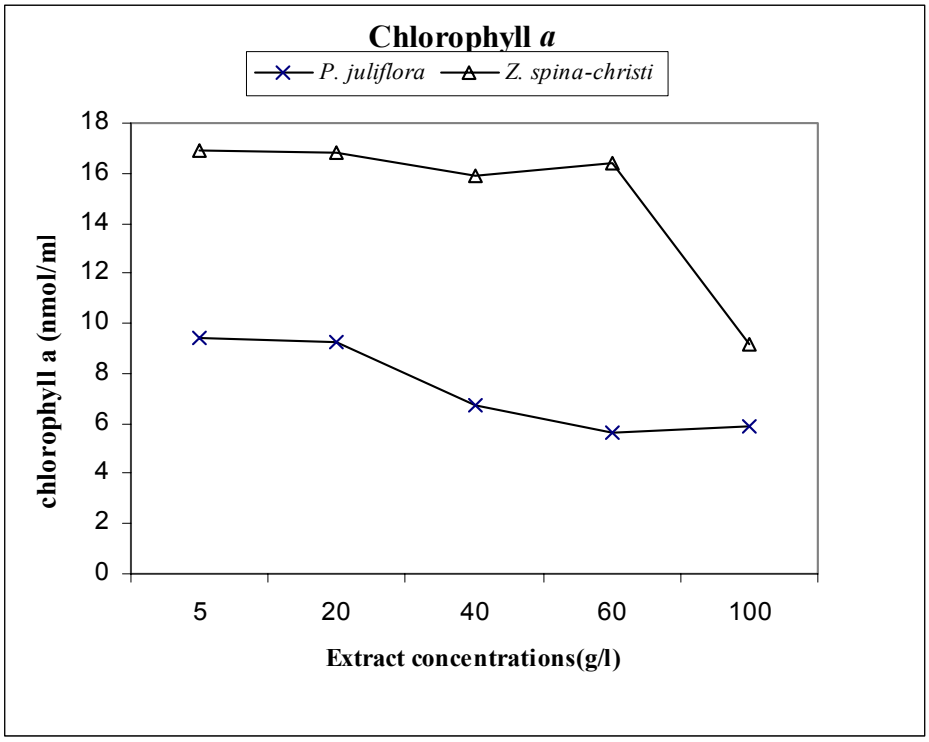

Figure 5. Effect of aqueous extract of *P. juliflora* (donor) chlorophyll *a* content of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on chlorophyll *a* content of *P. juliflora* (recipient).

Table 13. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent *concentration on chlorophyll *a* in *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		0.1969 0.8441	2.4039 0.0172	3.4088 0.0008	3.1541 0.0019
20	-0.0626 0.9501		2.2093 0.0283	3.2119 0.0015	2.9594 0.0035
40	-0.9356 0.3507	-0.8730 0.3838		0.9659 0.3353	0.7416 0.4592
60	-0.4853 0.6280	-0.4227 0.6730	0.4503 0.6530		-0.2159 0.8292
100	-6.3095 <.0001	-6.2538 <.0001	-5.4778 <.0001	-5.8780 <.0001	

-Shaded cells represent *P. juliflora*- Non-shaded cells represent *Z. spina-christi*

Table 14. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on chlorophyll *b*.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control chlorophyll b	1	0.1535675	0.1535675	0.08	0.7823
Blocks	3	0.1760844	0.0586948	0.03	0.9932
Concentrations	4	107.24449	26.811122	13.37	<.0001
Recipent	1	463.84669	463.84669	231.35	<.0001
Recipent *Concentrations	4	60.906196	15.226549	7.59	<.0001
Months	6	45.799032	7.6331719	3.81	0.0013
Months*Concentrations	24	87.980153	3.6658397	1.83	0.0139
Recipent *Months	6	81.669897	13.611649	6.79	<.0001
Recipent *Months*Concentrations	24	68.456349	2.8523479	1.42	0.1004

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60gl, and 100 g/l**)**
- Recipent s (*Ziziphus spina-christi* and *Prosopis juliflora***)**

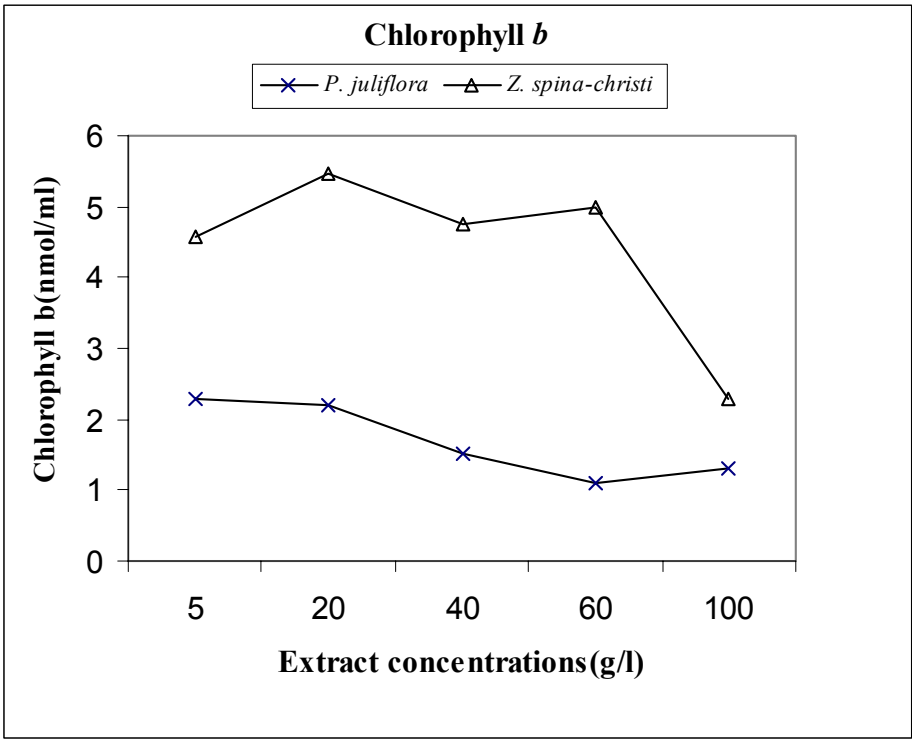

Figure 6. Effect of aqueous extract of *P. juliflora* (donor) chlorophyll *b* content of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on chlorophyll *b* content of *P. juliflora* (recipient).

Table 15. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent *concentration on chlorophyll *b* in *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		0.2172 0.8283	1.9671 0.0506	3.0528 0.0026	2.4811 0.0140
20	2.2882 0.0232		1.7524 0.0813	2.8357 0.0051	2.2663 0.0246
40	0.4591 0.6467	-1.8290 0.0690		1.0511 0.2946	0.5082 0.6119
60	1.0915 0.2764	-1.1966 0.2329	0.6324 0.5279		-0.5371 0.5918
100	-5.4032 <.0001	-7.4371 <.0001	-5.8114 <.0001	-6.3734 <.0001	

-Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*

Table 16. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on total chlorophyll *ab*.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control Chlorophyll *a+b*	1	99.275413	99.275413	3.80	0.0528
Blocks	3	66.117950	22.039317	0.84	0.4718
Concentrations	4	1677.9639	419.49099	16.05	<.0001
Recipent	1	3881.0427	3881.0427	148.48	<.0001
Recipent *Concentrations	4	724.39713	181.09928	6.93	<.0001
Months	6	512.32473	85.387455	3.27	0.0044
Months*Concentrations	24	876.87006	36.536252	1.40	0.1117
Recipent *Months	6	103.21913	17.203189	0.66	0.6835
Recipent *Months*Concentrations	24	502.80934	20.950389	0.80	0.7323

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

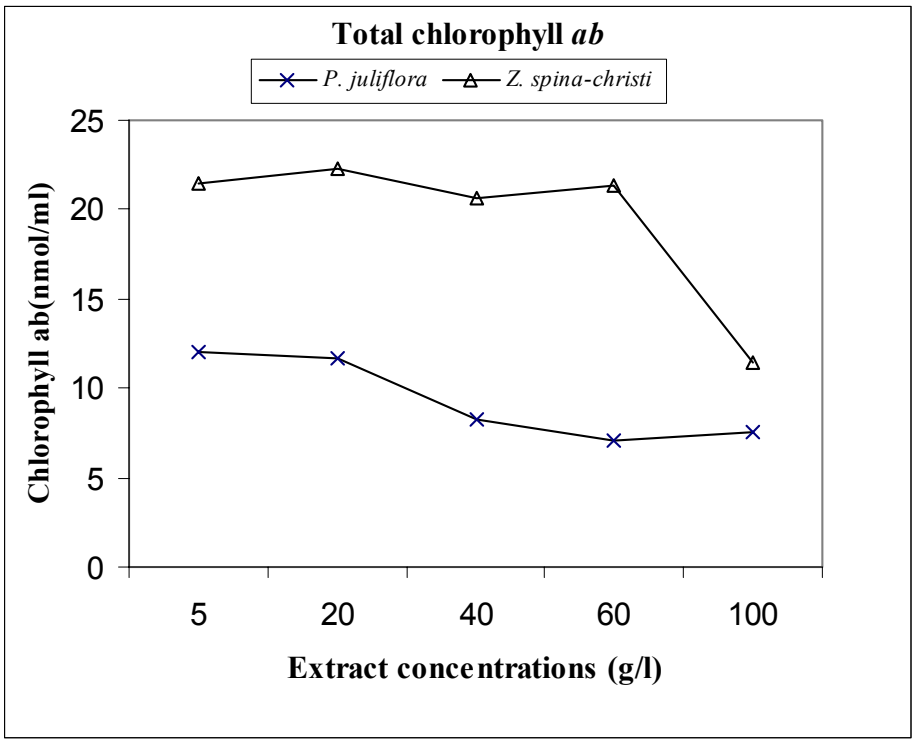

Figure 7. Effect of aqueous extract of *P. juliflora* (donor) on total chlorophyll *ab* content of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on total chlorophyll *ab* content of *P. juliflora* (recipient).

Table 17. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent *concentration on total chlorophyll *ab* of *P. juliflora* and *Z.spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		0.1956 0.8452	2.6924 0.0077	3.5022 0.0006	3.1312 0.0020
20	0.5839 0.5599		2.4969 0.0134	3.3066 0.0011	2.9379 0.0037
40	-0.6160 0.5386	1.2000 0.2316		0.8097 0.4191	0.4694 0.6393
60	-0.0832 0.9338	-0.6672 0.5054	0.5328 0.5948		-0.3312 0.7409
100	-6.5246 <.0001	-7.0438 <.0001	-5.9769 <.0001	-6.4506 <.0001	

-Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*

Table 18. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on leaf area.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control leaf area	1	7455.33600	7455.336	1.61	0.2058
Block	3	10999.6120	3666.537	0.79	0.4994
Concentrations	4	153345.550	38336.387	8.29	<. 0001
Recipent	1	1083588.07	1083588.074	234.22	<. 0001
Recipent *Concentrations	4	165552.106	41388.027	8.95	<.0001
Month	6	697757.850	116292.975	25.14	<.0001
Months*Concentrations	24	223963.967	9331.832	2.02	0.0049
Recipent *Months	6	705354.229	117559.038	25.41	<.0001
Recipent*Months*Concentrations	24	232658.752	9694.115	2.10	0.0032

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

Table 19. Means and standard errors for the influence of aqueous leaf extracts of *Prosopis juliflora* on leaf area and leaf number of *Ziziphus spina-christi* and vice versa over a 7-month growth period.

	Recipient			
	P. juliflora[1]		*Z. spina-christi*[2]	
Concentrations (g/l)	**Leaf number**	**Leaf area (cm^2)**	**Leaf number**	**Leaf area (cm^2)**
5	41.89 ±3.77	100.78 ±8.65	49.00 ±5.48	211.43 ±20.95
20	38.25 ±3.46	89.97 ±7.52	69.46 ±8.04	303.14 ±32.52
40	35.91 ±3.31	78.96 ±9.09	63.64 ±8.99	260.51 ±32.13
60	33.51 ±3.28	54.29 ±6.68	70.69 ±7.91	308.06 ±30.99
100	29.51 ±3.21	45.30 ±5.08	41.20 ±9.90	168.13 ±46.13

-*P. juliflora*[1] as recipient of *Z. spina-christi* leaf extracts
-*Z. spina-christi*[2] as recipient of *P. juliflora* leaf extracts

Table 20. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on number of leaves.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control number of leaves	1	449.19247	449.19247	1.20	0.2744
Block	3	2058.9090	686.30300	1.84	0.1421
Concentrations	4	7899.4879	1974.8720	5.28	0.0005
Recipent	1	39119.451	39119.452	104.61	<. 0001
Recipent *Concentrations	4	7170.2304	1792.5576	4.79	0.0010
Month	6	68767.789	11461.298	30.65	<. 0001
Month*Concentration	24	12348.798	514.53326	1.38	0.1224
Recipent *Month	6	28479.148	4746.5247	12.69	<. 0001
Recipent *Month*Concentrations	24	15343.233	639.30141	1.71	0.0255

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

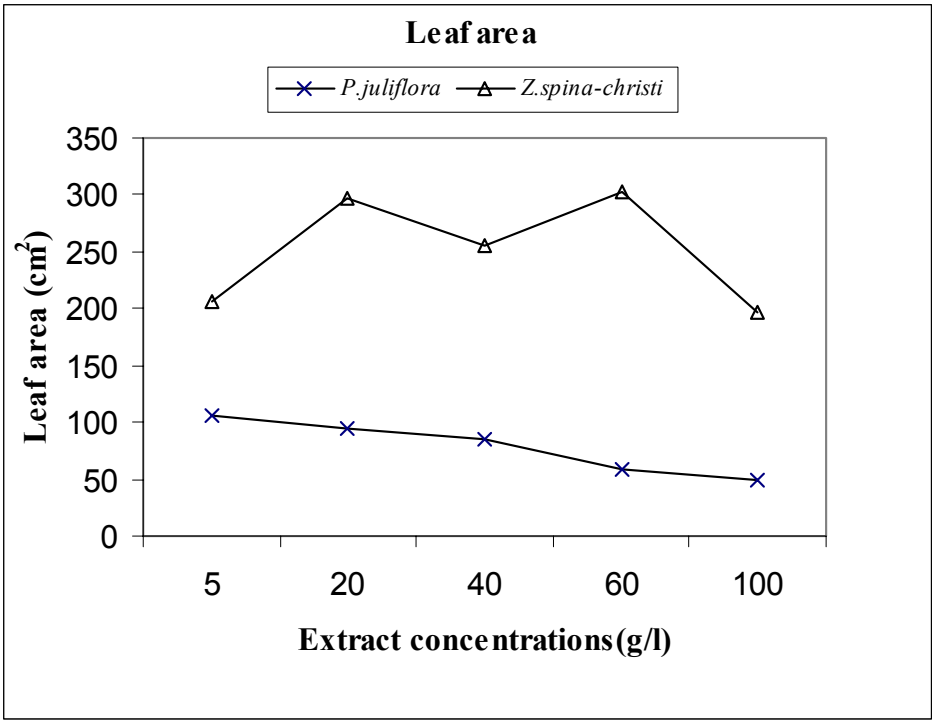

Figure 8. Effect of aqueous extract of *P. juliflora* (donor) on average leaf area of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on average leaf area of *P. juliflora* (recipient).

Table 21. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent *concentration on leaf area of *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		0.6541 0.5138	1.1913 0.2350	2.6284 0.0093	3.0815 0.0024
20	5.0454 <.0001		0.5372 0.5918	1.9743 0.0498	2.4274 0.0161
40	2.7000 0.0075	-2.3454 0.0200		1.4371 0.1523	1.8903 0.0602
60	5.3155 <.0001	0.2701 0.7873	2.6155 0.0096		0.4531 0.6510
100	-0.522 0.6020	-5.1331 <.0001	-2.9898 0.0032	-5.3799 <.0001	

- Shaded cells represent *P. juliflora*- Non-shaded cells represent *Z. spina-christi*

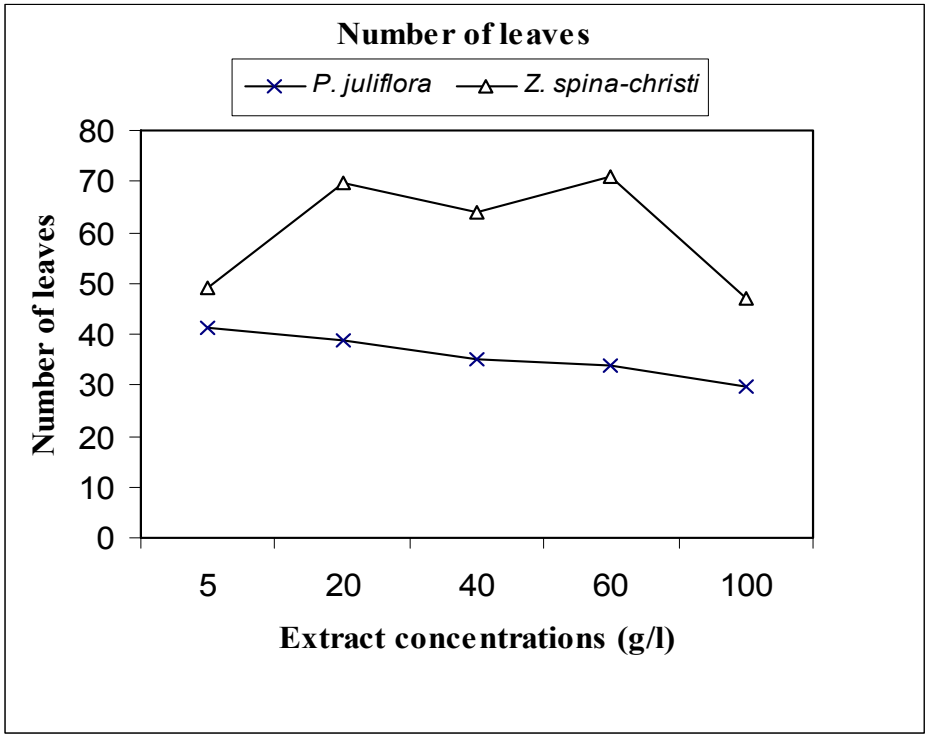

Figure 9. Effect of aqueous extract of *P. juliflora* (donor) on the number of leaves of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on number of leaves of *P. juliflora* (recipient).

Table 22. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent *concentration on number of leaves of *P.juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		0.4254 0.6711	1.1692 0.2438	1.3739 0.171	2.1774 0.0307
20	3.9597 0.0001		0.7438 0.4539	0.9486 0.3440	1.7520 0.0813
40	2.8333 0.0051	-1.1264 0.2614		0.2048 0.8379	1.0082 0.3146
60	4.1980 <.0001	0.2384 0.8118	1.3648 0.1739		0.8034 0.4227
100	-0.3331 0.7394	-3.9516 0.0001	-2.9222 0.0039	-4.1694 <0.0001	

- Shaded cells represent *P. juliflora*- Non-shaded cells represent *Z. spina-christi*

Table 23. Overall analysis of covariance for the effects of leaf extract concentrations and interactions with other factors on total root length.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control total root length	1	233366.06	233366.06	3.38	0.0673
Block	3	2068781.59	689593.86	10.00	<.0001
Concentrations	4	4039025.87	1009756.47	14.64	<.0001
Recipent	1	481750.80	481750.80	6.98	0.0089
Recipent *Concentration	4	1857312.90	464328.22	6.73	<.0001
Month	6	19222895.77	3203815.96	46.44	<.0001
Month*Concentration	24	2518680.12	104945.00	1.52	0.0634
Recipent *Month	6	1567803.24	261300.54	3.79	0.0013
Recipent *Month*Concentrations	24	1854922.55	77288.44	1.12	0.3239

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

Table 24. Means and standard errors for the influence of aqueous leaf extracts of *Prosopis juliflora* on root length, root diameter, root surface area, number of tips, and root volume of *Ziziphus spina-christi* seedlings and vice versa over a 7-month growth period.

	Recipient									
	P. juliflora[1]					*Z. spina-christi*[2]				
Concentrations (g/l)	**Total Root length (cm)**	**Root Surface Area (cm^2)**	**Number of tips**	**Volume (cm^3)**	**Root average diameter (mm)**	**Total Root length (cm)**	**Root Surface Area (cm^2)**	**Number of tips**	**Volume (cm^3)**	**Root average diameter (mm)**
5	1047.27 ±89.7	133.81 ±12.6	2547.75 ±165.4	1.38 ±0.13	0.37 ±0.016	1007.87 ±117.2	145.67 ±16.4	2175.89 ±226.89	1.71 ±0.19	0.43 ±0.01
20	949.14 ±81.7	123.679 ±11.7	2392.75 ±154.4	1.31 ±0.13	0.38 ±0.017	1147.10 ±121.4	169.20 ±17.9	2383.89 ±245.34	2.09 ±0.24	0.43 ±0.01
40	935.78 ±91.5	119.879 ±12.6	2240.73 ±154.2	1.24 ±0.14	0.37 ±0.01	1064.14 ±131	152.22 ±19.1	2323.50 ±257.83	1.78 ±0.22	0.41 ±0.02
60	847.68 ±98.7	108.22 ±12.9	2019.55 ±180.5	1.12 ±0.138	0.37 ±0.016	1191.95 ±119.7	179.52 ±18.6	2402.84 ±213.17	2.19 ±0.24	0.44 ±0.01
100	772.32 ±86.9	99.93 ±11.7	1843.39 ±152.6	1.05 ±0.12	0.37 ±0.017	624.51 ±115.7	89.11 ±18.1	1326.54 ±254.95	1.06 ±0.24	0.38 ±0.02

-*P. juliflora*[1] as a recipient of *Z. spina-christi* leaf extracts
-*Z. spina-christi*[2] as a recipient of *P. juliflora* leaf extracts

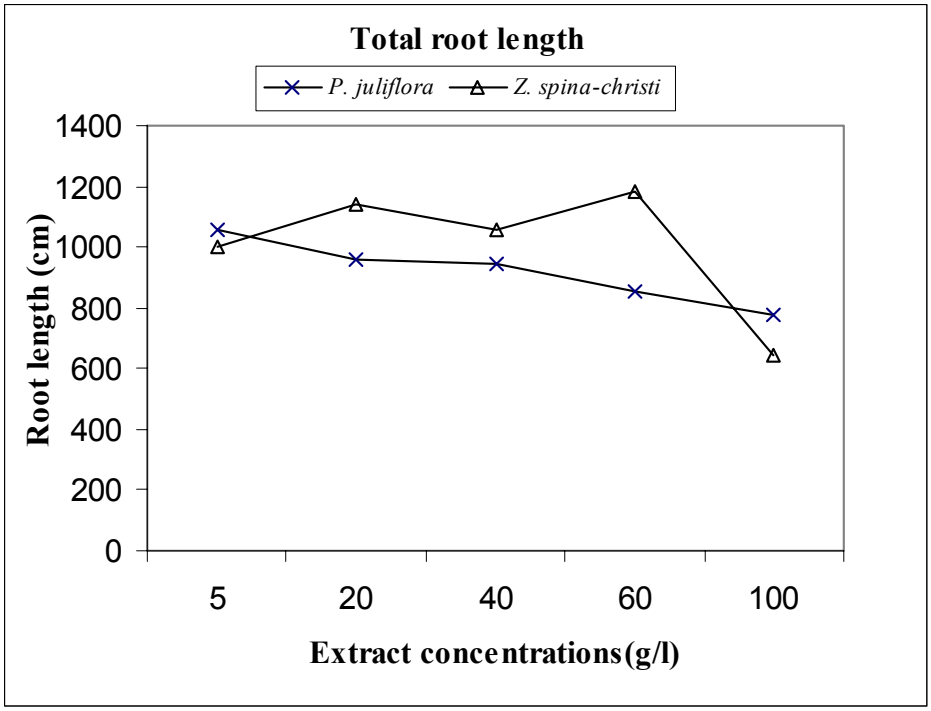

Figure 10. Effect of aqueous extract of *P. juliflora* (donor) on average: total root length of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on average total root length of *P. juliflora* (recipient).

Table 25. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent*concentration on total root length of *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		1.3979 0.1637	1.5883 0.1138	2.8433 0.0049	3.9169 0.0001
20	1.9835 0.0487		0.1905 0.8491	1.4455 0.1499	2.5191 0.0125
40	0.8016 0.4237	-1.1819 0.2386		1.2550 0.2109	2.3286 0.0209
60	2.6225 0.0094	0.6390 0.5235	1.8209 0.0701		1.0736 0.2843
100	-4.7921 <.0001	-6.6656 <.0001	-5.5493 <.0001	-7.2692 <.0001	

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi.*

Table 26. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on root surface area.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control root surface	1	1651.7961	1651.7961	1.15	0.2846
Blocks	3	32667.4712	10889.1571	7.59	<.0001
Concentration	4	86506.0704	21626.5176	15.07	<.0001
Recipent	1	64463.2126	64463.2126	44.91	<.0001
Recipent *Concentration	4	48810.3687	12202.5922	8.50	<.0001
Month	6	376814.2343	62802.3724	43.76	<.0001
Month*Concentration	24	54118.2559	2254.9273	1.57	0.0501
Recipent *Month	6	40443.3805	6740.5634	4.70	0.0002
Recipent*Months*Concentration	24	32790.4075	1366.2670	0.95	0.5316

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

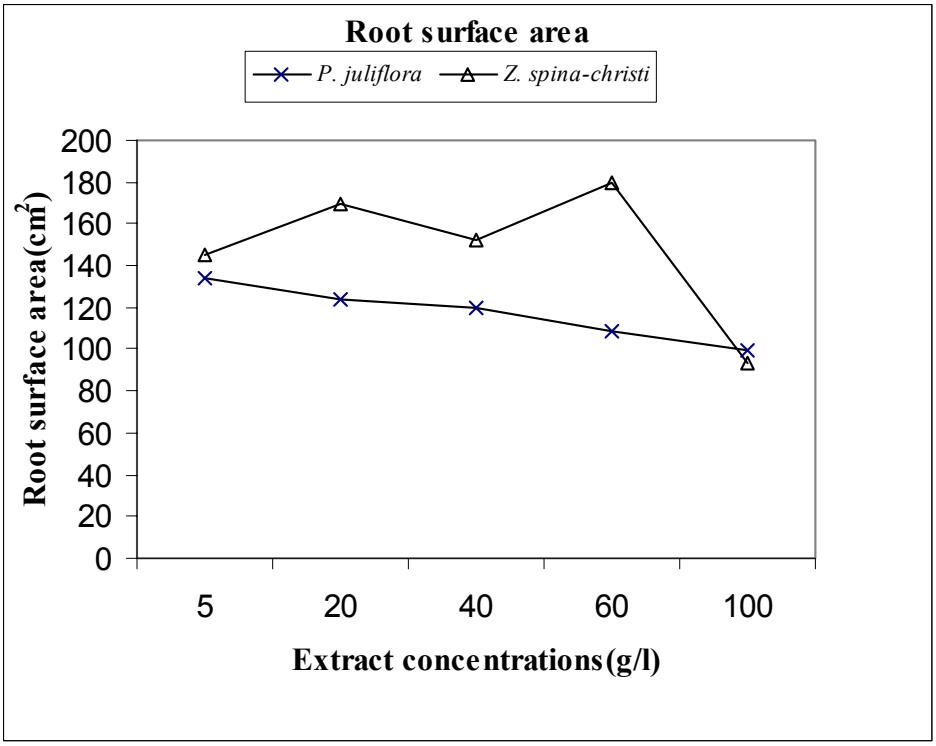

Figure 11. Effect of aqueous extract of *P. juliflora* (donor) on root surface area of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on root surface area of *P. juliflora* (recipient).

Table 27. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent *concentration on root surface area of *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		1.0003 0.3184	1.3756 0.1705	2.5267 0.0123	3.3454 0.0010
20	2.3243 0.0211		0.3753 0.7078	1.5264 0.1285	2.3452 0.0200
40	0.6474 0.5181	-1.6768 0.0951		1.1511 0.2510	1.9699 0.0502
60	3.3433 0.0010	1.0190 0.3094	2.6959 0.0076		0.8187 0.4139
100	-4.9160 <.0001	-7.1113 <.0001	-5.5275 <.0001	-8.0739 <.0001	

- Shaded cells represent *P. juliflora.* - Non-shaded cells represent *Z. spina-christi.*

Table 28. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on number of root tips.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control number of tips	1	61335.37	61335.37	0.15	0.6961
Blocks	3	10919063.30	3639687.77	9.08	<.0001
Concentration	4	21632019.83	5408004.96	13.49	<.0001
Recipent	1	289693.46	289693.46	0.72	0.3963
Recipent *Concentration	4	6667297.29	1666824.32	4.16	0.0029
Month	6	78151465.52	13025244.25	32.49	<.0001
Month*Concentration	24	10507440.03	437810.00	1.09	0.3553
Recipent*Month	6	7617487.82	1269581.30	3.17	0.0054
Recipent *Month*Concentration	24	8230075.40	342919.81	0.86	0.6624

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**

- Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

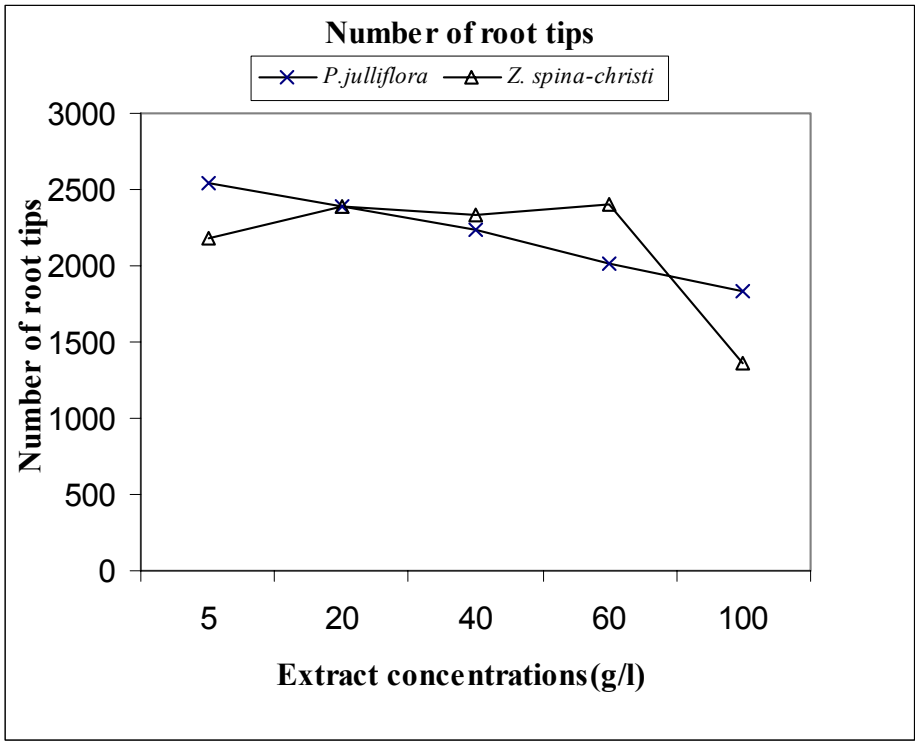

Figure 12. Effect of aqueous extract of *P. juliflora* (donor) on number of root tips of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on number of root tips of *P. juliflora* (recipient).

Table 29. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent *concentration on number of root tips in *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		0.9159 0.3608	1.8143 0.0711	3.1213 0.0021	4.1623 <.0001
20	1.2293 0.2204		0.8983 0.3701	2.2054 0.0286	3.2464 0.0014
40	0.8723 0.3841	-0.3569 0.7216		1.3070 0.1927	2.3480 0.0198
60	1.3411 0.1814	0.1119 0.9110	0.4688 0.6397		1.0410 0.2991
100	-4.5548 <.0001	-5.7158 <.0001	5.3787 <.0001	-5.8216 <.0001	

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi.*

Table 30. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on root volume.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control root volume	1	0.00033626	0.00033626	0.00	0.9724
Block	3	4.36728899	1.45576300	5.21	0.0017
Concentration	4	12.81202023	3.20300506	11.46	<.0001
Recipent	1	19.92762655	19.92762655	71.31	<.0001
Recipent *Concentration	4	8.32265111	2.08066278	7.45	<.0001
Month	6	51.59043728	8.59840621	30.77	<.0001
Month*Concentration	24	7.64204538	0.31841856	1.14	0.3038
Recipent*Month	6	8.81259896	1.46876649	5.26	<.0001

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent s: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

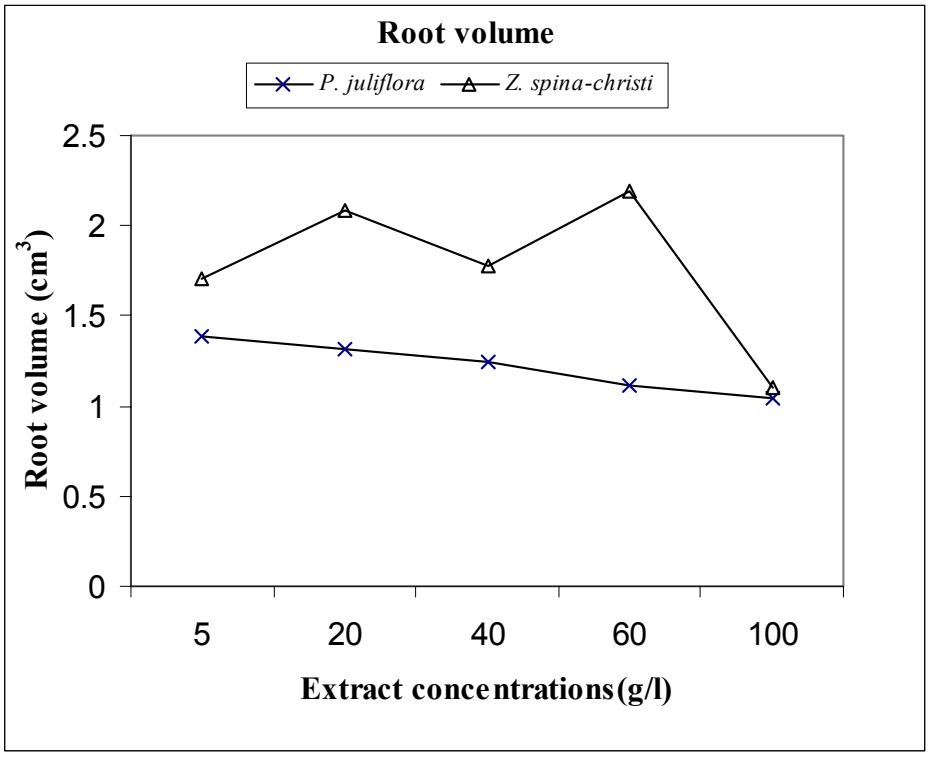

Figure 13. Effect of aqueous extract of *P. juliflora* (donor) on root volume of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on root volume of *P. juliflora* (recipient).

Table 31. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent*concentration on root volume in *P. juliflora* and *Z. spina-christi*. Pr > |t|.

Concentrations (g/l)	5	20	40	60	100
5		0.4904 0.6244	1.0010 0.3180	1.8769 0.0620	2.3799 0.0182
20	2.7009 0.0075		0.51062 0.6102	1.3865 0.1671	1.8895 0.0603
40	0.4979 0.6190	-2.203 0.0287		0.8759 0.3821	1.3789 0.1694
60	3.4176 0.0008	0.7166 0.4744	2.9196 0.0039		0.5030 0.6155
100	-4.0637 <.0001	-6.6149 <.0001	-4.5341 <.0001	-7.2918 <.0001	

-Shaded cells represent *P. juliflora*. - Non-shaded cells represent *Z. spina-christi*.

Table 32. Overall analysis of covariance for the effects of leaf extract concentrations and their interactions with other factors on root to shoot ratio.

Source	DF	Type III SS	Mean Square	F Value	Pr > F
Control Root: Shoot ratio	1	0.00000271	0.00000271	0.00	0.9932
Block	3	0.07430702	0.02476901	0.66	0.5793
Concentration	4	0.25803106	0.06450776	1.71	0.1488
Recipent	1	0.64952484	0.64952484	17.23	<.0001
Recipent*Concentration	4	1.21071506	0.30267876	8.03	<.0001
Month	6	2.27852839	0.37975473	10.08	<.0001
Month*Concentration	24	0.88715477	0.03696478	0.98	0.4931
Recipent*Month	6	0.52188298	0.08698050	2.31	0.0354
Recipent*Month*Concentration	24	0.70520501	0.02938354	0.78	0.7598

- Concentrations : (5 g/l, 20 g/l, 40 g/l, 60 g/l, and 100 g/l**)**
- Recipent: (*Ziziphus spina-christi* and *Prosopis juliflora***)**

Table 33. Means and standard errors for the influence of aqueous leaf extracts of *Prosopis juliflora* on root to shoot ratio of *Ziziphus spina-christi* and vice versa over a 7-month growth period.

	Recipient	
	P. juliflora[1]	*Z. spina-christi*[2]
Concentrations (g/l^{-1})	**Root: shoot ratio**	**Root: shoot ratio**
5	0.55 ±0.02	0.90 ±0.07
20	0.58 ±0.02	0.79 ±0.05
40	0.62 ±0.03	0.74 ±0.03
60	0.69 ±0.02	0.83 ±0.04
100	0.74 ±0.04	0.68 ±0.04

-*P. juliflora*[1] as recipient of *Z. spina-christi* leaf extracts.
-*Z. spina-christi*[2] as recipient of *P. juliflora* leaf extracts.

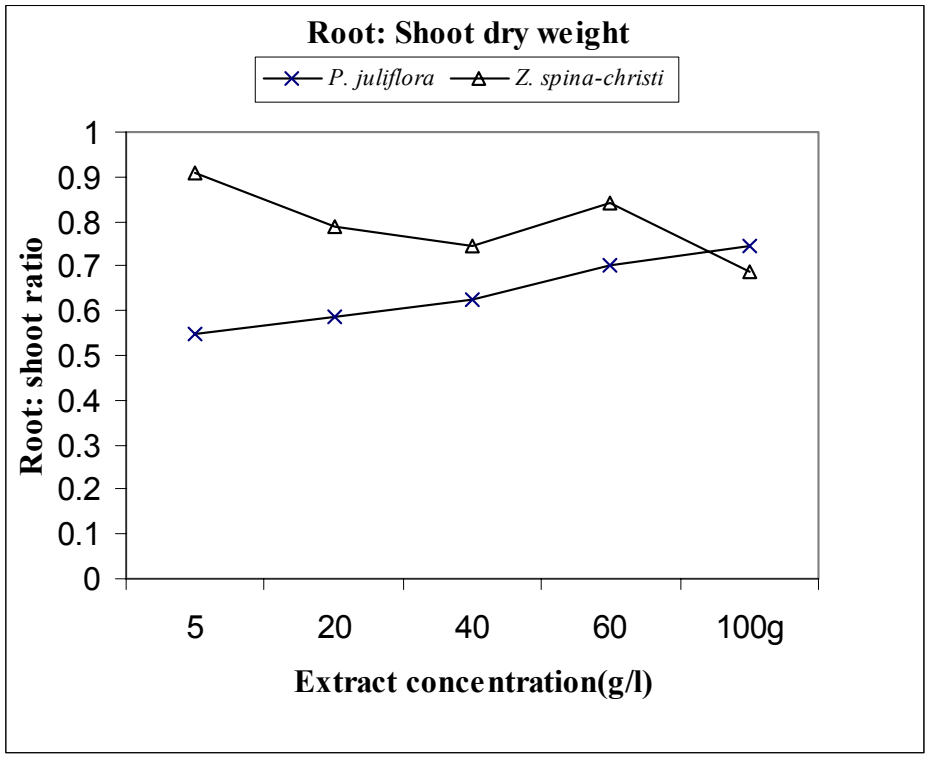

Figure 14. Effect of aqueous extract of *P. juliflora* (donor) on root to shoot ratio of *Z. spina-christi* (recipient) and effect of aqueous extract of *Z. spina-christi* (donor) on root to shoot ratio of *P. juliflora* (recipient).

Table 34. T- values (top) and p-values (lower) comparing least squares means for the interaction between recipent*concentration on root to shoot ratio of *P. juliflora* and *Z. spina-christi*. Pr > |t|

Concentrations (g/l)	**5**	**20**	**40**	**60**	**100**
5		-0.6425 0.5213	-1.4326 0.1535	-2.8143 0.0054	-3.6351 0.0004
20	-2.2509 0.0255		-0.7901 0.4304	-2.1718 0.0310	-2.9926 0.0031
40	-3.1012 0.0022	-0.8503 0.3962		-1.3816 0.1686	-2.2025 0.0288
60	-1.3062 0.1930	0.9448 0.3459	1.7950 0.0742		-0.8209 0.4127
100	-4.2007 <.0001	-1.9497 0.0526	-1.0995 0.2729	-2.8944 0.0042	

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi.*

Table 35. Correlation (*r*) among growth parameters measured.

	Root average diameter (RAD)	Chl *a*	Total Chls *ab*	Chl *b*	Stem Diameter (SD)	Root dry weight (RDW)	Shoot dry weight (SDW)	Height	Leaf area (LA)	Number of leaves (NL)	Number of root tips (NRT)	Root mass fraction (RMF)	Root: Shoot ratio (R:S)	Shoot leaf mass Fraction (SLMF)	Total leaf mass fraction (TLMF)	Root surface area (RSA)	Total root length (TRL)	Root volume (RV)
RAD		-0.019	-0.057	-0.15	0.53	0.49	0.48	0.50	0.47	0.47	0.26	0.34	0.32	0.013	-0.13	0.64	0.54	0.68
Chl a			0.98	0.74	-0.17	0.09	-0.021	-0.15	0.39	0.17	-0.005	0.14	0.16	0.67	0.59	0.036	-0.022	0.093
Tchl ab				0.85	-0.19	0.070	-0.039	-0.17	0.37	0.15	-0.008	0.11	0.13	0.66	0.60	0.009	-0.046	0.066
Chl b					-0.18	0.029	-0.051	-0.176	0.25	0.090	0.016	0.103	0.105	0.51	0.46	-0.033	-0.07	0.0091
SD						0.75	0.87	0.84	0.45	0.62	0.72	0.28	0.26	-0.41	-0.52	0.81	0.84	0.74
RDW							0.89	0.67	0.74	0.77	0.71	0.597	0.596	-0.044	-0.27	0.90	0.87	0.90
SDW								0.83	0.65	0.76	0.75	0.27	0.25	-0.21	-0.34	0.87	0.88	0.82
Height									0.39	0.52	0.68	0.13	0.14	-0.43	-0.50	0.73	0.76	0.67
LA										0.88	0.53	0.43	0.43	0.44	0.24	0.75	0.67	0.78
N L											0.65	0.38	0.36	0.17	0.001	0.81	0.78	0.80
NRT												0.322	0.320	-0.182	-0.314	0.80	0.87	0.70
R M F													0.98	0.087	-0.26	0.51	0.45	0.54
R:S														0.105	-0.24	0.50	0.44	0.53
SLMF															0.93	-0.065	-0.15	0.025
TLMF																-0.27	-0.33	-0.20
RSA																	0.97	0.97
TRL																		0.91

APPENDIX A

Tables of Means and Standard Errors for the Reciprocal Effects of Leaf Extracts for the *P. juliflora* and *Z. spina-christi* over a Seven-month Treatment Period.

Table 1. Effect of *Z. spina- christi* leaf extract concentrations on stem height and diameter of *P. juliflora* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)	***Height (cm)***						
5	19.43 ±1.97	42.12 ±2.46	45.25 ±4.29	49.75 ±4.00	58.81 ±6.19	66.77 ±6.90	69.00 ±8.24
20	16.18 ±1.84	39.15 ±1.89	41.81 ±2.42	42.37 ±3.98	69.06 ±5.72	67.83 ±2.82	66.06 ±3.23
40	18.37 ±3.30	26.56 ±7.33	41.68 ±2.97	38.18 ±7.10	58.75 ±6.25	65.75 ±6.47	56.50 3.25
60	14.68 ±1.35	19.37 ±3.72	29.18 ±5.71	43.06 ±4.52	36.12 ±4.98	40.93 ±7.58	45.18 ±4.95
100	18.08 ±3.54	30.28 ±3.26	29.06 ±4.95	25.50 ±4.10	41.93 ±3.66	46.25 ±2.74	39.75 ±3.60
Concentrations (g/l)	***Diameter (mm)***						
5	1.81 ±0.17	3.30 ±0.06	3.56 ±0.12	4.37 ±0.14	4.75 ±0.20	4.95 ±0.28	5.27 ±0.06
20	1.77 ±0.04	3.03 ±0.05	3.45 ±0.14	4.30 ±0.16	4.91 ±0.29	4.80 ±0.13	5.18 ±0.36
40	1.83 ±0.12	2.61 ±0.43	3.87 ±0.28	3.42 ±0.43	4.43 ±0.21	4.83 ±0.22	5.20 ±0.19
60	1.83 ±0.17	2.52 ±0.34	2.78 ±0.37	3.78 ±0.26	4.31 ±0.05	4.30 ±0.66	4.51 ±0.48
100	1.75 ±0.14	2.62 ±0.13	2.78 ±0.33	3.13 ±0.69	3.97 ±0.05	3.81 ±0.07	4.38 ±0.23

Table 2. Effect of *P. juliflora* leaf extract concentrations on height and stem diameter of *Z. spina-christi* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)			***Height (cm)***				
5	13.50	24.08	35.06	37.75	46.00	50.18	51.06
	±1.00	±2.39	±2.18	±3.54	±2.89	±3.26	±3.08
20	14.56	29.50	37.06	39.00	48.37	51.68	48.93
	±3.27	±4.26	±1.91	±3.58	±3.39	±5.24	±1.06
40	12.18	19.65	28.87	33.96	47.25	47.56	49.93
	±2.67	±3.46	±2.18	±1.35	±6.65	±2.39	±1.98
60	13.43	31.76	40.06	45.93	47.37	52.31	51.62
	±1.02	±3.85	±3.85	±2.34	±2.59	±2.98	±1.68
100	13.00	23.37	16.93	24.87	33.83	36.25	46.00
	±1.90	±2.87	±2.07	±8.06	±11.15	±1.25	±4.00
Concentrations (g/l)			***Diameter (mm)***				
5	1.72	2.42	2.90	3.37	3.86	3.76	4.21
	±0.09	±0.10	±0.07	±0.14	±0.32	±0.15	±0.06
20	1.73	2.86	3.22	3.55	4.28	4.06	4.20
	±0.17	±0.27	±0.21	±0.12	±0.30	±0.46	±0.29
40	1.78	2.15	2.86	3.20	4.20	4.27	4.23
	±0.14	±0.21	±0.14	±0.13	±0.59	±0.17	±0.29
60	1.77	3.06	3.32	4.27	4.80	4.71	4.41
	±0.12	±0.14	±0.11	±0.11	±0.12	±0.33	±0.19
100	1.76	2.45	1.83	2.45	3.63	4.02	3.72
	±0.12	±0.40	±0.18	±0.51	±0.63	±0.22	±0.57

Table 3. Effect of *Z. spina-christi* leaf extract concentrations on shoot and root dry weight of *P. juliflora* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)			***Shoot dry weight (g)***				
5	0.48 ±0.05	2.39 ±0.06	2.50 ±0.15	3.45 ±0.16	5.19 ±0.62	6.99 ±0.63	7.11 ±0.29
20	0.38 ±0.04	1.80 ±0.06	2.60 ±0.19	3.41 ±0.52	5.20 ±0.27	6.24 ±0.47	5.73 ±0.49
40	0.44 ±0.14	1.24 ±0.49	2.34 ±0.17	2.59 ±0.86	4.13 ±0.54	6.53 ±0.88	5.84 ±0.39
60	0.34 ±0.08	0.88 ±0.36	1.40 ±0.49	2.59 ±0.18	3.24 ±0.12	3.97 ±1.17	4.61 ±0.79
100	0.36 ±0.08	1.142 ±0.12	1.05 ±0.32	1.29 ±0.49	2.98 ±0.32	3.08 ±0.42	3.29 ±0.27
Concentrations (g/l)			***Root dry weight (g)***				
5	0.21 ±0.01	0.95 ±0.07	1.35 ±0.12	1.97 ±0.16	3.19 ±0.35	3.82 ±0.41	4.91 ±0.30
20	0.21 ±0.01	0.76 ±0.03	1.25 ±0.16	2.30 ±0.17	3.18 ±0.13	3.75 ±0.43	4.09 ±0.14
40	0.21 ±0.04	0.56 ±0.18	1.32 ±0.12	1.59 ±0.45	2.37 ±0.41	3.82 ±0.34	4.48 ±0.22
60	0.22 ±0.04	0.43 ±0.16	0.87 ±0.23	1.88 ±0.23	2.33 ±0.12	3.08 ±0.93	3.38 ±0.59
100	0.20 ±0.03	0.56 ±0.07	0.79 ±0.21	1.20 ±0.56	2.11 ±0.07	2.22 ±0.37	3.29 ±0.30

Table 4. Effect of *P. Juliflora* leaf extracts concentrations on shoot and root dry weight of *Z. spina-christi* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)	***Shoot dry weight (g)***						
5	0.49 ±0.08	1.13 ±0.17	1.83 ±0.25	3.14 ±0.20	3.77 ±0.68	5.50 ±0.29	4.77 ±0.41
20	0.49 ±0.15	1.76 ±0.38	2.48 ±0.33	3.65 ±0.31	5.50 ±0.53	6.31 ±1.50	±6.15 ±0.61
40	0.47 ±0.19	0.89 ±0.37	1.72 ±0.14	3.15 ±0.06	4.91 ±1.23	5.95 ±0.47	7.01 ±0.21
60	0.45 ±0.04	2.14 ±0.33	2.37 ±0.37	3.87 ±0.28	5.79 ±0.08	7.22 ±0.29	6.15 ±0.92
100	0.44 ±0.09	0.88 ±0.33	0.49 ±0.07	1.27 ±0.80	2.04 ±0.90	2.91 ±1.10	2.24 ±1.21
Concentrations (g/l)			***Root dry weight (g)***				
5	0.27 ±0.02	0.67 ±0.16	1.48 ±0.09	2.55 ±0.26	3.82 ±0.65	5.7316 ±0.4488	6.11 ±0.55
20	0.22 ±0.04	0.91 ±0.19	1.82 ±0.21	3.17 ±0.30	5.15 ±0.36	5.2071 ±1.4223	6.28 ±0.76
40	0.24 ±0.07	0.51 ±0.20	1.35 ±0.15	1.86 ±0.21	5.03 ±1.47	5.5946 ±0.7995	5.68 ±0.53
60	0.23 ±0.01	1.29 ±0.12	1.85 ±0.22	3.46 ±0.23	5.80 ±0.28	6.1876 ±0.9608	6.74 ±0.92
100	0.27 ±0.05	0.55 ±0.29	0.31 ±0.05	1.11 ±0.81	1.39 ±0.65	1.9239 ±0.8523	1.90 ±1.08

Table 5. Effect of *Z. spina-christi* leaf extract concentrations on chlorophyll *a* and chlorophyll *b* of *P. juliflora* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)				**Chl *a***			
5	10.24 ±2.40	7.66 ±1.59	8.89 ±2.00	9.09 ±1.84	10.82 ±1.42	7.23 ±1.04	7.56 ±1.38
20	10.50 ±3.24	6.81 ±0.90	7.79 ±1.52	9.32 ±1.64	10.97 ±2.39	6.60 ±0.79	7.69 ±0.99
40	8.47 ±2.96	8.51 ±3.19	3.49 ±0.12	7.33 ±1.31	8.31 ±2.38	5.02 ±1.30	2.58 ±0.72
60	6.65 ±1.47	3.13 ±0.79	3.12 ±1.08	4.94 ±1.78	8.72 ±0.97	5.95 ±1.40	2.85 ±1.26
100	14.67 ±3.00	4.13 ±0.83	3.00 ±1.25	5.13 ±2.37	4.25 ±1.70	4.33 ±1.09	2.54 ±1.37
Concentrations (g/l)				**Chl *b***			
5	2.49 ±0.58	0.78 ±0.64	2.12 ±0.95	2.52 ±0.46	2.86 ±0.44	2.42 ±0.15	2.45 ±0.32
20	3.42 ±1.29	-0.71 ±0.51	1.13 ±0.38	2.61 ±0.44	3.09 ±0.72	2.10 ±0.30	3.37 ±0.71
40	2.25 ±0.68	0.47 ±0.72	0.73 ±0.36	2.370 ±0.45	2.49 ±0.78	1.46 ±0.32	1.14 ±0.13
60	1.74 ±0.21	-0.74 ±0.25	-0.39 ±0.41	1.43 ±0.44	2.32 ±0.27	1.95 ±0.40	1.12 ±0.39
100	4.10 ±0.78	-0.48 ±0.31	0.31 ±0.35	1.62 ±0.43	1.04 ±0.36	1.46 ±0.32	1.20 ±0.29

Table 6. Effect of *P. Juliflora* leaf extract concentrations on chlorophyll *a* and chlorophyll *b* of *Z. spina-christi* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)				**Chl *a***			
5	21.03 ±1.27	14.74 ±1.70	17.99 ±1.93	14.04 ±1.22	20.14 ±2.16	15.22 ±1.43	18.72 ±0.97
20	20.81 ±3.09	13.36 ±1.34	19.61 ±2.64	18.87 ±1.77	20.64 ±1.48	12.54 ±1.29	15.58 ±0.93
40	17.62 ±3.65	12.75 ±3.47	15.48 ±1.93	17.44 ±0.95	21.77 ±3.79	13.89 ±0.69	15.81 ±1.22
60	17.15 ±2.05	17.19 ±4.72	19.58 ±1.61	15.89 ±2.72	19.64 ±2.49	14.06 ±0.28	14.69 ±2.02
100	15.20 ±2.48	7.20 ±2.47	2.70 ±0.58	11.91 ±4.75	10.89 ±6.46	11.18 ±1.88	7.77 ±1.09
Concentrations (g/l)				**Chl *b***			
5	6.05 ±0.58	5.34 ±0.65	4.77 ±0.43	4.86 ±1.65	6.48 ±0.90	4.28 ±0.61	0.32 ±0.39
20	6.91 ±1.80	5.47 ±0.47	5.43 ±0.97	4.60 ±0.64	6.30 ±0.74	3.45 ±0.25	5.99 ±0.78
40	5.71 ±1.00	4.59 ±0.75	3.65 ±0.60	4.58 ±0.29	5.66 ±1.40	4.51 ±0.87	4.61 ±0.25
60	6.29 ±0.77	6.85 ±1.46	4.76 ±0.51	4.73 ±0.54	4.58 ±0.55	3.51 ±0.15	4.26 ±0.47
100	4.23 ±0.67	2.43 ±0.32	0.67 ±0.19	3.60 ±1.23	2.32 ±1.46	2.85 ±0.21	-0.13 ±1.86

Table 7. Effect of *Ziziphus spina- christi* leaf extract concentrations on total chlorophyll *ab* of *P. juliflora* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)				**Chls *a+b***			
5	12.74 ±2.98	10.43 ±1.92	11.02 ±2.87	11.61 ±2.25	13.69 ±1.84	9.65 ±1.16	10.01 ±1.68
20	13.93 ±4.48	8.29 ±1.13	8.92 ±1.90	11.94 ±2.08	14.06 ±3.11	8.71 ±1.09	11.07 ±1.68
40	10.73 ±3.60	8.95 ±3.02	4.23 ±0.44	9.70 ±1.72	10.80 ±3.10	6.48 ±1.58	3.73 ±0.84
60	8.39 ±1.61	4.88 ±0.44	2.72 ±1.02	6.38 ±2.23	11.05 ±1.23	7.91 ±1.80	3.98 ±1.65
100	18.77 ±3.77	6.26 ±0.87	3.32 ±1.57	6.76 ±2.80	5.29 ±2.07	5.79 ±1.41	3.74 ±1.66

Table 8. Effect of *P. Juliflora* leaf extract concentrations on total chlorophyll *ab* of *Z. spina-christi* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)	**Chls *a+b***						
5	27.09	20.08	22.76	18.90	26.63	19.51	19.05
	±1.85	±1.36	±2.33	±1.85	±1.98	±2.00	±0.96
20	27.72	18.84	25.05	23.48	26.94	15.99	21.57
	±4.65	±1.16	±3.55	±2.32	±1.01	±1.55	±1.00
40	23.34	17.34	19.14	22.03	27.44	18.40	20.43
	±4.64	±4.17	±2.54	±1.21	±5.18	±1.12	±1.47
60	23.45	24.05	24.34	20.63	24.23	17.58	18.96
	±1.34	±5.89	±2.10	±3.24	±3.04	±0.41	±2.50
100	19.44	9.64	3.37	15.52	13.21	14.03	7.63
	±3.11	±2.80	±0.75	±5.97	±7.92	±2.10	±0.76

Table 9. Effect of *Z. spinac-christi* leaf extract concentrations on number of leaves and leaf area of *P. juliflora* over a 7-month growth period

Concentrations (g/l)	*Month*						
	1	*2*	*3*	*4*	*5*	*6*	*7*
	number of leaves						
5	12.62	32.37	30.50	38.62	56.87	60.62	61.62
	±0.55	±2.63	±1.42	±4.50	±10.31	±6.51	±5.85
20	10.75	34.12	29.25	44.75	44.62	47.25	57.00
	±0.85	±2.25	±3.19	±3.20	±5.75	±4.39	±14.90
40	10.50	28.62	27.87	32.37	52.62	54.62	44.75
	±2.09	±5.50	±1.50	±6.41	±8.40	±5.73	±3.81
60	10.12	22.12	27.25	35.50	55.12	40.62	43.87
	±1.23	±5.21	±2.52	±8.33	±7.07	±7.58	±4.58
100	10.12	28.12	20.25	24.87	58.37	30.37	34.50
	±1.24	±3.60	±5.03	±5.42	±9.88	±4.81	±3.66
Concentrations (g/l)			***Leaf area***				
5	31.30	93.29	81.83	77.79	136.42	170.33	114.47
	±2.85	±6.52	±5.21	±18.18	±13.67	±7.07	±6.90
20	22.54	85.62	73.59	79.94	110.78	135.30	121.98
	±5.11	±2.18	±3.97	±5.60	±8.79	±10.56	±22.06
40	29.66	41.35	73.18	55.67	130.16	138.09	84.61
	±11.88	±13.92	±9.59	±11.04	±14.21	±28.20	±11.82
60	19.46	26.72	30.74	50.34	96.62	89.24	66.90
	±8.24	±11.64	±6.40	±10.24	±8.38	±20.85	±7.72
100	20.33	48.93	21.96	30.59	85.39	68.99	40.90
	±5.59	±7.07	±3.34	±9.72	±5.99	±13.96	±4.18

Table 10. Effect of *P. Juliflora* leaf extract concentrations on number of leaves and leaf area of *Z. spina-christi* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)			***number of leaves***				
5	11.12	22.37	38.25	68.75	64.00	71.75	66.75
	±1.16	±4.33	±6.70	±14.66	±14.51	±5.08	±12.04
20	10.50	38.25	53.37	78.50	90.50	111.37	103.75
	±1.24	±12.85	±10.60	±16.73	±15.78	±21.33	±9.47
40	11.00	19.12	35.50	56.25	86.12	105.50	132.00
	±2.50	±9.53	±4.37	±4.11	±9.88	±10.72	±24.03
60	10.37	44.00	49.00	84.50	92.37	132.25	82.37
	±1.43	±4.01	±11.46	±10.18	±5.50	±21.40	±8.07
100	11.37	27.25	9.62	24.50	63.16	79.66	120.75
	±2.26	±16.75	±2.96	±11.55	±22.96	±35.46	±48.25
Concentrations (g/l)			***Leaf area***				
5	60.62	93.88	166.13	257.76	266.68	342.75	292.15
	±9.76	±17.53	±28.57	±15.64	±41.67	±22.41	±37.82
20	58.74	165.02	245.18	300.28	455.53	406.44	490.82
	±17.76	±45.94	±30.65	±27.51	±66.15	±72.30	±49.70
40	59.06	58.70	178.35	289.29	348.11	387.05	502.99
	±26.36	±24.19	±16.47	±15.66	±65.21	±29.78	±31.40
60	58.01	205.70	220.32	317.29	452.65	512.17	390.26
	±7.53	±38.94	±40.58	±29.87	±14.46	±44.99	±64.20
100	51.50	110.11	20.02	90.41	312.04	246.89	577.11
	±10.95	±53.62	±8.33	±65.13	±163.17	±108.88	±229.39

Table 11. Effect of *Z. spina-christi* leaf extract concentrations on total root length of *P. juliflora* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)	***Total root length***						
5	172.02 ±8.34	731.47 ±13.41	958.40 ±65.62	1156.89 ±42.23	1293.09 ±148.65	1434.08 ±96.21	1584.93 ±85.11
20	150.62± 3.65	620.86 ±18.55	888.08 ±82.69	1104.81 ±50.94	1267.35 ±88.19	1187.66 ±105.92	1424.63 ±72.15
40	157.21 ±32.53	500.99 ±96.74	859.38 ±35.62	933.41 ±150.78	1138.47 ±95.24	1450.43 ±34.12	1510.55 ±63.62
60	147.88± 35.17	384.36 ±80.28	643.85 ±105.93	1053.86 ±62.361	1044.83 ±57.76	1219.23 ±321.08	1439.75 ±213.42
100	118.43± 21.32	475.19 ±44.59	593.54 ±117.57	678.40 ±200.93	1180.70 ±144.52	1072.48 ±189.33	1287.46 ±59.21

Table 12. Effect of *P. Juliflora* leaf extract concentrations on total root length of *Z. spina-christi* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)			***Total root length***				
5	119.89 ±14.78	411.81 ±75.53	850.13 ±78.55	957.84 ±59.60	1346.86 ±228.08	1700.76 ±196.49	1667.79 ±160.49
20	122.95 ±21.34	571.01 ±103.38	927.45 ±80.19	1306.94 ±188.17	1531.60 ±56.98	1735.59 ±215.26	1834.16 ±138.79
40	125.07 ±16.18	399.51 ±102.03	781.77 ±60.03	1189.30 ±156.16	1526.66 ±384.35	1635.56 ±68.73	1791.10 ±247.83
60	118.48± 14.06	735.70 ±50.42	867.79 ±97.79	1261.87 ±38.02	1802.99 ±74.32	1863.00 ±183.75	1693.84 ±29.35
100	145.44 ±22.87	589.83 ±177.95	325.94 ±34.99	590.10 ±251.58	1113.94 ±450.43	1239.54 ±432.45	625.55 ±297.84

Table 13. Effect of *Z. spina-christi* leaf extract concentrations on root surface area and number of root tips of *P. juliflora* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)	***Root surface area***						
5	9.13 ±0.69	93.43 ±3.13	127.06 ±9.34	140.34 ±7.14	166.71 ±21.35	185.21 ±9.54	214.72 ±10.69
20	8.31 ±0.27	78.82 ±3.33	111.71 ±8.55	145.70 ±7.24	173.90 ±12.94	155.66 ±14.43	191.63 ±8.99
40	8.58 ±1.84	64.87 ±11.98	112.30 ±6.02	119.53 ±19.87	141.08 ±13.38	189.69 ±3.42	203.09 ±5.48
60	8.59 ±1.99	51.12 ±12.92	85.76 ±14.62	137.27 ±10.35	135.85 ±8.19	160.65 ±44.55	178.29 ±21.02
100	6.52 ±1.06	65.83 ±5.77	81.16 ±16.63	84.08 ±29.02	151.78 ±15.56	134.35 ±23.43	175.79 ±4.91
Concentrations (g/l)	***Number of root tips***						
5	1744.50 ±86.69	1462.63 ±60.60	2037.25 ±163.96	2950.38 ±227.94	3180.13 ±485.62	3231.00 ±279.53	3228.38 ±356.47
20	1443.88 ±86.80	1507.38 ±121.18	2316.50 ±547.59	2693.63 ±105.89	2815.38 ±267.15	2979.50 ±407.24	2993.00 ±208.12
40	1613.50 ±342.74	1263.63 ±317.40	1745.38 ±137.97	2264.25 ±271.98	3028.38 ±214.69	2951.88 ±280.46	2818.13 ±207.61
60	1233.63 ±153.75	976.25 ±217.13	1409.88 ±271.77	2359.63 ±248.68	2418.13 ±260.48	2895.13 ±478.65	2844.25 ±531.20
100	1089.38 ±257.50	1222.63 ±219.88	1489.38 ±205.77	1825.00 ±297.30	2606.13 ±576.50	2343.75 ±359.31	2327.50 ±212.293

Table 14. Effect of *P. Juliflora* leaf extract concentrations on root surface area and number of root tips of *Z. spina-christi* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)			**Root surface area**				
5	8.18 ±0.75	64.15 ±12.90	125.78 ±5.35	145.60 ±9.37	204.32 ±26.37	241.42 ±25.15	230.22 ±11.00
20	8.01 ±1.41	86.62 ±15.26	144.27 ±8.08	193.98 ±18.48	245.06 ±21.19	235.08 ±31.62	271.38 ±9.70
40	7.92 .04	54.12 ±14.85	125.15 ±7.44	159.61 ±21.05	214.73 ±52.04	235.30 ±10.59	268.73 ±31.46
60	8.02 ±0.99	113.96 ±10.05	134.38 ±11.97	186.02 ±9.25	276.90 ±6.86	261.39 ±30.60	275.96 ±5.42
100	9.75 ±1.34	85.88 ±30.26	41.33 ±5.21	87.15 ±46.98	163.85 ±75.67	162.29 ±53.81	108.92 ±56.82
Concentrations (g/l)			***Number of root tips***				
5	932.50 ±317.46	693.00 ±86.45	1662.50 ±127.28	2575.63 ±208.63	2783.88 ±510.77	3370.00 ±432.02	3213.75 ±529.90
20	669.50 ±235.26	943.13 ±137.73	1757.88 ±111.97	2983.00 ±416.09	3152.63 ±312.03	3787.63 ±301.69	3393.50 ±477.79
40	693.13 ±187.87	682.00 ±121.29	1801.88 ±150.54	3621.63 ±328.20	2872.38 ±713.34	3552.38 ±343.49	3041.13 ±411.09
60	952.13 ±214.21	1055.38 ±65.33	1907.88 ±38.46	3576.38 ±190.19	2817.25 ±380.10	3684.88 ±310.16	2826.00 ±149.06
100	663.38 ±220.83	855.75 ±193.25	744.13 ±77.25	1250.63 ±486.51	2007.17 ±700.30	3049.83 ±1487.09	1080.50 ±405.49

Table 15. Effect of *Z. spina- christi* leaf extract concentrations on root volume (cm^3) of *P. juliflora* over a 7-month growth period.

	Month						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)	***Volume (cm^3)***						
5	0.03 ±0.00	0.98 ±0.03	1.34 ±0.10	1.36 ±0.09	1.71 ±0.24	1.92 ±0.11	2.31 ±0.17
20	0.03 ±0.00	0.79 ±0.05	1.17 ±0.16	1.53 ±0.09	1.91 ±0.18	1.63 ±0.16	2.09 ±0.24
40	0.03 ±0.01	0.67 ±0.12	1.17 ±0.08	1.22 ±0.21	1.40 ±0.17	2.00 ±0.06	2.18 ±0.05
60	0.04 ±0.00	0.54 ±0.16	0.91 ±0.16	1.42 ±0.13	1.41 ±0.09	1.70 ±0.50	1.78 ±0.16
100	0.03 ±0.00	0.73 ±0.07	0.89 ±0.19	0.84 ±0.32	1.56 ±0.13	1.34 ±0.23	1.92 ±0.09

Table 16. Effect of *P. Juliflora* leaf extract concentrations on root volume (cm^3) of *Z. spina-christi* over a 7-month growth period.

	Months						
	1	***2***	***3***	***4***	***5***	***6***	***7***
Concentrations (g/l)			***Volume (cm^3)***				
5	0.04 ±0.00	0.80 ±0.17	1.48 ±0.07	1.77 ±0.13	2.50 ±0.30	2.74 ±0.26	2.57 ±0.21
20	0.04 ±0.00	1.04 ±0.18	1.81 ±0.08	2.32 ±0.14	3.16 ±0.49	2.96 ±0.77	3.25 ±0.17
40	0.04 ±0.00	0.59 ±0.17	1.60 ±0.08	1.82 ±0.23	2.42 ±0.58	2.70 ±0.14	3.23 ±0.33
60	0.04 ±0.01	1.41 ±0.16	1.66 ±0.11	2.19 ±0.19	3.42 ±0.23	2.98 ±0.55	3.59 ±0.16
100	0.05 ±0.01	1.01 ±0.41	0.42 ±0.05	1.05 ±0.67	1.93 ±0.99	1.69 ±0.53	1.57 ±0.85

APPENDIX B

Photographs of Experimental Conditions and Examples of Typical Plants from Various Treatments

Figure 1. *Prosopis juliflora* sheds its leaves.

Figure 2. The ability of *Prosopis juliflora* to recover its leaves in a short time.

Figure 3. The effect of aqueous leaf extract of *P. juliflora* at 100 g/l on *Z.spina-christi* chlorophyll content of yellowish leaves was low.

Figure 4. Stimulatory effect of *Prosopis juliflora* leaf extract (20 g/l) on the growth of *Z. spina-christi*.

Figure 5. *Ziziphus spina-christi* seedlings treated with *Prosopis juliflora* leaf extract at 40 g/l.

Figure 6. *Ziziphus spina-christi* seedlings treated with *Prosopis juliflora* leaf extract at 60 g/l.

Figure 7. *Ziziphus spina-christi* seedlings treated with *Prosopis juliflora* leaf extract at 100 g/l. Arrow indicates growing part of the root.

Figure 8. *Ziziphus spina-christi* seedling treated with *Prosopis juliflora* leaf extract at 100 g/l. Arrows indicates dehydrated leaves.

Figure 9. *Ziziphus spina-christi* seedling roots treated with *Prosopis juliflora* leaf extract at 5 g/l at the first month of treatment

Figure 10. *Ziziphus spina-christi* seedling roots treated with *Prosopis juliflora* leaf extract at 40 g/l.

Figure 11. *Ziziphus spina-christi* seedling roots treated with *Prosopis juliflora* leaf extract at 60 g/l.

Figure 12. *Prosopis juliflora* seedlings treated with *Ziziphus spina-christi* leaf extract at 100 g/l

Figure 13. *Prosopis juliflora* seedlings treated with *Ziziphus spina-christi* leaf extract at 100 g/l. Arrow indicates growing part of the root.

Figure 14. *Prosopis juliflora* seedlings treated with *Ziziphus spina-christi* leaf extract at 5 g/l.

Figure 15. *Prosopis juliflora* seedlings treated with *Ziziphus spina-christi* leaf extract at 100 g/l. Arrow indicates saponin bubbles in leaf extract.

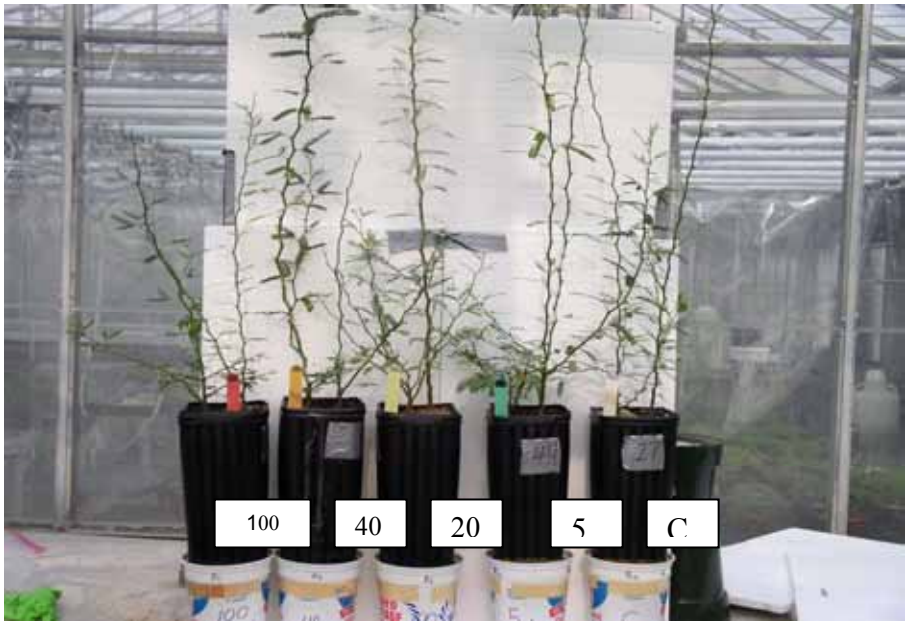

Figure 16. *Prosopis juliflora* seedlings treated with *Ziziphus spina-christi* at different extract concentrations at the end of growth period, 7 months.

Figure 17. *Prosopis juliflora* seedlings treated with *Ziziphus spina-christi* leaf extract at 40 g/l (#31), 60 g/l(#35), and 100 g/l(#62).

Figure 18. *Prosopis juliflora* seedlings treated with *Ziziphus spina-christi* at 5 g/l (#83), 20 g/l(#57) and control (#44).

Figure 19. *Ziziphus spina-christi* seedling treated with *Prosopis juliflora* leaf extract at 100 g/l.

Figure 20. *Prosopis juliflora* seedling roots treated with *Ziziphus spina-christi* leaf extract at 40 g/l.

Figure 21. *Prosopis juliflora* seedling roots treated with *Ziziphus spina-christi* leaf extract at 5 g/l.

Chapter 2

Interaction of interspecific competition and nitrogen levels on growth of *Prosopis juliflora* and *Ziziphus spina-christi* seedlings.

Abstract

This study was conducted under greenhouse conditions to study the effect of two nitrogen levels and plant density on the competition between *Z. spina-christi* and *P. juliflora* in a growth period of 7-months. The total density in mixed plantatings was 6 plants per pot (15.5 cm* 15 cm) and proportions of *Z. spina-christi* to *P. juliflora* were (0.67:0.33), (0.50: 0.50), and (0.33:0.67) under high and low levels of nitrogen. Plants were grown in monoculture planting in a density of 1, 2, 3, 4, and 6 plant per pot under high and low levels of nitrogen as well. Plant height, diameter, leaf area, total chlorophyll *ab*, total dry weight, root to shoot ratio, relative yield, relative yield total, and aggressivity were measured and/or calculated.

Results indicated that the two species responded differently to high and low levels of nitrogen. Under high nitrogen, in mixed plantings, most parameters of *P. juliflora* exceed these of *Z. spina-christi*. Height, leaf area, total dry weight, and total chlorophyll *ab* were reduced for *Z. spina-christi* seedlings with increasing proportions of *P. juliflora*. Under high nitrogen, *P. juliflora* tended to be more aggressive but the aggressivity and relative yield of *P. juliflora* wasreduced as its proportion increased in mixed culture.

Under the low nitrogen level, *Z. spina-christi* growth exceeded *P. juliflora* in most growth parameters, illustrating that, *Z. spina-christi* had a competitive advantage over *P. juliflora* under low nitrogen levels.

In monocultures, there was a reduction in most growth parameters with increasing numbers of plants per pot. The reduction was higher for plants grown in low nitrogen for both species, compared to plants grown in high levels of nitrogen. It appears that *P. juliflora* is more affected by intraspecific competition than interspecific competition, and under higher nitrogen levels, this invasive species is more aggressive than the native species that it is replacing. Since, *P. juliflora* is a nitrogen fixing

species, it is possible that under field conditions it gains a competitive advantage when nitrogen fixation occurs.

Chapter 2

Interaction of interspecific competition and nitrogen levels on growth of *Prosopis juliflora* and *Ziziphus spina-christi* seedlings.

Introduction

Historically, human introduction of plant species into new areas is designed to exploit their economic value as crop species, forage plants, or timber trees (Heywood, 1989). In arid and semi-arid areas introducing exotic plant species is often deemed necessary when the native species no longer meet the requirements of the local human population. Many local species have disadvantages such as slow growth, low productivity and low seed germination rates. In Saudi Arabia, in the Riyadh region, a numbers of exotic tree species have been introduced for public or private purposes, including 16 *Eucalyptus* species *(*Zoghot, 1997*)*, 19 *Acacia* species (Zoghet and Tag El- dien, 1996), 9 *Prosopis* species *(*Zoghot, 1997), and 2 *Casuarina* species. In all cases, the purpose of introduction was to provide needed benefits to humans. Among those species, however, several have became invasive in Saudi Arabia and neighboring countries. For example, *Prosopis juliflora* is considered an invasive species in Sudan (Richardson 1998), Pakistan (Noor et al., 1995), Ethiopia, India, United Arab Emirates (El-Kablawy, personal communication), and Oman. In most cases when a species has been observed as invasive in some regions on the world, the probability that it will be an invasive plant in other regions is high.

It is clear from these observations that introduced plant species can threaten terrestrial ecosystems by altering geomorphological processes, nutrient cycles, and displacing indigenous species such as *Ziziphus spina-christi* (Walck et al., 1999a). One process that leads toward exclusion of the native species is altered seed germination due to changes in soil conditions. Often this will lead to a decrease in the abundance of the native species and therefore a more homogenized ecosystem (Huenneke and Thomson, 1995). For many plant species, success in the new ecosystem can also be the result of freeing the plant from its natural enemies that hold them in check, therefore allowing them to utilize their full competitive potential and eradicate their new neighbors **(**Callaway and Aschehoug, 2000).

In plant populations or communities, competition for resources occurs between individuals of the same species or between members of different species. When a new plant is introduced into a community, one can expect that changing the environment of the native species will affect their growth (Harper, 1977). Interspecific competition is a significant factor in controlling the distribution and abundance of terrestrial plants in a variety of different habitats (Badger and Ungar, 1990) and therefore an important determinant of the structure and dynamics of plant communities (Aerts, 1999). For plants occupying stressful habitats, Tilman's (1988) model suggests that plants limited by the same resources compete strongly for that resource. For invasive species, this means that the invaders either are capable of gaining access to limited resources where the native species cannot or are able to use the resources more efficiently (Vitousek, 1990).

In nature, nutrient availability is one of the most important factors that can limit plant performance and therefore change the competitive relationships between plant species. Competition at the seedling stage is extremely important and will, to a great extent, determine later success. In nutrient poor-environments, e.g. deserts, plants must vigorously compete for water and nutrients and use both physiological and morphological traits to gain an edge over the competition. However in nutrient poor habitats the plant's physiological traits are less important than morphological characteristics in increasing mineral nutrient uptake (Aerts, 1999). For some species their low competitive ability in securing nutrients may become one of the most important factors contributing to the species' narrow distribution.

To study the interference, or competition between species, De Wit (1960) developed a mathematical model to describe the interference between two species using a replacement series experiment. In this substitutive design the overall density of the two species is held constant but the proportion of the two species is changed so that the outcome of competition between the two species can be predicted (Weiner, 1980). Greenhouse replacement series commonly place invasive plant species together with native plant species to show potential interference between the two. For example, Weiss and Noble (1984) found that, by using a replacement series experiment, the seedlings of the invasive species *Chrysanthemoids monilifera* (DC) had a competitive

advantage over the native species *Acacia longifolia* because of its rapid growth, large leaf area, and high use of water. Therefore, replacement experiments have been commonly used to compare the relative efficiency of two species to capture resources and convert them into biomass (Daugovish et al., 2002).

Initially, *Prosopis juliflora* was introduced to the Middle East as a result of intentional anthropogenic dispersal but it has started to rapidly expand its range. *P. juliflora* has aggressively expanded its range into non-native habitats because of its extensive seed production, high seed germination rate, and rapid root growth. A number of other attributes of *P. juliflora* may also provide a competitive advantage over native species such as *Ziziphus spina-christi*. Because of the observed invasiveness of *P. juliflora* it is likely that the species will continue to invade communities and displace local species. The analysis of this interaction should clarify the mechanism by which *P. juliflora* is able to displace *Z. spina-christi* from its habitat.

The goal of this research was to determine the nature of the competitive interaction between the invasive exotic species *P. juliflora* and the native species *Z. spina-christi* and to interpret this in terms of observed invasiveness. I specifically wanted to determine how competition for nitrogen influenced the relative competitiveness of the two species. The hypotheses tested were:

1) Interspecific competition of *P. juliflora* seedlings and *Z. spina-christi* seedlings differs from intraspecific competition within either species, based on gained dry biomass.

2) In mixtures, *P. juliflora* is more efficient than *Z. spina-christi* in both high and low nitrogen environments.

In order to test these hypotheses we used a substitutive experiment or replacement series design, growing plants of the two species in containers at the same overall density and changing the proportion of species in the mixture. The experiments were replicated under conditions of low nitrogen fertilization and high nitrogen. Total biomass and biomass allocation were used as measures of impact in this greenhouse experiment. Data collected in this experiment were used to analyze the interspesific

and intraspesific interference between the two species. Specific questions we tried to address were:

1) What species will gain more biomass during the experiment under monoculture conditions?

2) How does one species affect the performance of the other species when grown together in various mixtures?

3) What is the interactive effect of nitrogen fertilization on competition and growth of the two species?

Literature review:

Plant and competition

Competition among plants is a process that governs the structure and function of many ecosystems. The seedling stage is an important developmental stage in the plant life cycle. A species can dominate or be dominated in an ecosystem depending on the ability of seedlings to compete with seedlings of other species. Weiss and Noble (1984) using pot experiments, found that the displacement of *Acacia longifolia* by the invasive species *Chrysanthemoides monilifera* could be explained by the greater competitiveness of *C. monilifera* at the seedling stage. In another study, Witkowski (1991) demonstrated that seedlings of the invasive species *Acacia saligna* had a relatively high growth rate at all levels of nutrient availability. This was shown to be the key factor that enabled *A. saligna* to be a successful competitor with the native species *Protea repens*. The long-term outcome of species interactions, however, depends on the overall competitive aggressiveness of species toward each other.

Competition can be defined as a depression in growth of one plant induced by limitation in resources to that plant as a result of the presence of a neighbor. The degree of growth suppression a plant can induce on its neighbors depends on two factors. First, the responsiveness of each species to resource supply. Second, a species' competitiveness will depend on the effectiveness of each species in competing for limiting resources in a specific environment (Beneke et al. 1992). Success of individual plants in an ecosystem depends on their ability to compete with individuals of their own species and other species. When species compete intensely for limited resources, species with low competitive ability become rare (Walck et al., 1999a). Competition begins when one or more vital resources such as water, light and/or nutrients are limited for plant growth. Plant succession, in turn, is determined by the dynamic interaction between resource availability and the ability of different species to obtain these limited resources (Van Auken and Bush, 1987). Plants can differ quite significantly in their interaction with each other when they occupy the same site. Sometimes a positive interaction between species can be observed where one plant species may facilitate the growth of another plant species. This interaction depends on

the age, resource availability and the position of the plant relative to the resource. For instance, in a greenhouse experiment the growth of *Cynodon dactylon* was enhanced when the plants were grown with *Acacia smallii* but the best performance for Acacia was in a monoculture (Cohn et al., 1989). In another experiment acacia individuals had a negative effect on the growth of eucalyptus. In this case its intrespecific competition was stronger than its intraspecific competition with eucalyptus (Bauhus et al., 2000). It is well documented that plants behave differently in response to inter- and intraspecific competition when responding to resource constraint factors such as fertility levels.

Under unfavorable growth conditions superior competitors will supplant non-competitive species. Kooijman and Bakker (1995) found that *Scorpidium scorpioides* was replaced by *Sphagnum subnitens* as a result of changing environmental conditions. In another example, Austin et al. (1985) found that *Carthamus lanatus* L. had the highest yield when grown in a mixed plantation with *Carduus nutans*, *Carduus pycnocephalus*, *Cirsium vulgare*, *Onopordum aff. Illyricum*, and *Silybum marianum* at low nutrient concentrations. This was not the case for the total dry weight of the species at the medium and high level of nutrients where the species had low yield. On the other hand, Mynhardt et al (1994) found that interspecific competition lowered the number of lateral tillers per plant and tuft height in *Anthephra pubescens* in a mixed stand when compared to plants in pure stands. The investigators concluded that *Anthephra pubescens* was a poor competitor in these circumstances. Clearly, differences among the competitive abilities of plant species can be important factors controlling the distribution and composition of vegetation (Gerry and Wilson, 1995).

Under growth limiting conditions, competition strategies are extremely important to plants (Kenkel et al., 1991). The competition for a given mineral depends on the importance of that mineral to the plant. The shrub *Artimsia tridentate* absorbed much more phosphorus from the root space it shared with *Agropyron spicatum* than from the root space it shared with *Agropyron desertorum*, and *A. desortorum* took up more phosphorus than *A. spicatum* when competing with *A. tridentate*. In some species, mineral absorption increases in the presence other species. Allen (1982) found that *Bouteloua gracilis* exhibites increased uptake of P and K when grown with *Salsola kali* var. *tenuifolia.* Witkowski (1991) found that *Protea repens* showed reduced

growth at high levels of nutrient availability when grown with the invasive species *Acacia saligna*. The competitive edge of *A. saligna* over *P. repens* may be related to plant morphology. Hill and Shimamato (1973) indicated that the strongest competitor among the five genotypes of ryegrasss (*Lolium perenne*) had longer leaves and a more erect growth pattern, therefore enabling these genotypes to convert incoming light more efficiently than other genotypes in the study.

Regarding below-ground competition, plants depend on several root characteristics including relative growth rate, total biomass, fine root density, rooting depth, and total surface area to gain a competitive edge (Casper and Jackson, 1997). Casper and Jackson found that sagebrush, *Artemisia tridentate*, had eight- to ten- fold fewer roots than the non-native tussock grass *Agropyron desertorum* in fertilized nutrient patches one week after the patches were created. The *Agropyron desertorum* had apparently responded to the fertilization by producing additional roots. Fetene (2003) showed that *Hyparrenia hirta* produced high root yields compare to shoot when competing with *Acacia etabaica*. Increased root yields indicated that the competition strategy of Hyperenia was facilitated through the response of root growth. It should be clear from these observations that above- and below-ground competition often depends on plant morphology.

Intra- and inter-specific competition negatively affects individual plant biomass when plant density reaches a certain point. Yields of species in mixtures and monocultures can therefore give an indication of competitive performance of a species and the impact of plant-to-plant interactions under different environmental conditions. Allen (1982) found that *Salsola kali var. tenuifolia* negatively affected the harvested biomass of *Agropyron smithii* and *Bouteloua gracilis* when growing in a mixed culture compared to plants of *A. smithii* and *B. gracilis* grown in a monoculture.

Performance and yield of species vary even though they are growing under the same conditions. Superior competitors have a superior ability to occupy a patch of resource rich soil (Crick and Grime, 1987) and to convert soil resources to biomass. Characteristics of superior competitive species include the production of large root surface area per unit of total plant biomass (Jones et al, 1989) and efficient uptake of water and nutrients per unit root surface (Crick and Grime, 1987).

For plants, competition pressure depends on the availability of the limiting factor and the ability of the plant to change allocation of biomass to shoots or roots during the competitive interaction. In general plants that normally occupy nutrient-poor environments are able to compete for nutrients more successfully than those that normally inhabit nutrient-rich habitats (Shontz and Shontz, 1972).

Nitrogen and plant competition

Generally when nitrogen is a limiting factor to plant growth, the plant allocates growth to root volume or biomass to increase nitrogen uptake. Whigham (1984) found that the level of competition between plants of the same species was more severe for unfertilized plants than plants growing in nutrient-rich environments. Plants in unfertilized plots were small and produced proportionately more root biomass in response to limited nitrogen levels compared to those in fertilized plots.

Many plants can modify their growth allocations to shoot or root to assist in the capture of the most limited resources. It is clear that nutrient availability is often important in interspecific competition in natural plant communities (Aerts, 1999). Biomass allocation can therefore determine the success of a species in a given habitat (Gross et al. 1985).

Densities and proportions of plant species in a given ecosystem affect intraspecific and interspecific interference because there is a clear relationship between plant yield, denisity of individuals, and resources available (Santos et al, 1997). In support of this statement, Santos et al. (1997) found that tomato plants, when grown with either nutsedge or yellow nutsedge, under non-limiting conditions of water and nutrients, increased in dry weight whereas nutsedge dry weight decreased as their relative proportions decreased in mixture. Relative yield indicated tomato to be a stronger competitor than either nutsedge species. Both nutsedges appeared to be weak interspecific competitors with tomato but strong intraspecific competitors. On the other hand, Weiner (1980) found in a study on the effect of plant density and species proportion that at a given density *Trifolium incarnatum* individuals generally showed a greater yield when competing with *Lolium multiflorum* than when competing with plants of their own species. However, the monoculture of *Trifolium incarnatum*

produced a higher total yield than *Lolium multiflorum.* Walck et al. (1999 b), in a replacement series study, concluded that height and relative yield total of the endangered species *Solidago shortii* decreased in the presence of the aggressive species *Festuca arundinacea* indicating that the two species competed for the same resources. For *Festuca arundinacea*, intraspecific competition seemed to be more intense than interspecific competition.

Growth is dependent on the availability of resources such as light, nutrients, etc. A change in the environment may affect the availability of resources therefore altering the performance of a species and changing biomass allocation. Bi and Turvey (1994) addressed this idea by showing that *Acacia melanoxylon* was more aggressive than other species in their study, since *A*. *melanoxylon* in mixture showed a smaller decrease in shoot/root ratio compared to its corresponding monoculture than the other species. Qasem and Hill (1994) recognized that in a replacement series experiment with two weed species, *Chnopodium album* L. and *Senecio vulgaris* L. the two species behaved quite differently. *Senecio vulgaris* dry matter per plant increased as its proportion increased in the mixture. On the other hand, *Chenopodium album* dry weight per plant was reduced as its proportion in the mixture increased.

Weiner (1980) found that the proportion of *Trifolium incarnatum* and *Lolium multiflorum* that results in the greatest total yield and the greatest increase in yield relative to the monoculture yield depends on density and nutrition. The increase in relative yield in a mixture was investigated at high density and low potassium and phosphorous. The results indicated that nutrient level and density affect competition out-come. Similarly, Cohn et al. (1989) proposed that nutrient levels and species proportions had significant effects on dry mass per plant when *Acacia smalii* competed with *Cyndon dactylon*. In their experiments they showed that in soil supplemented with nutrients, relative yield of *Cyndon dactylon* was significantly greater in all mixtures, with a corresponding decrease in the growth and *Acacia smalli.*

Plant species from nutrient deficient ecosystems can be excluded from habitats that have high concentrations of nutrients. McGraw and Chapin (1989) found in a greenhouse experiment that a species often found on nutrient-rich sites, *Eriophorum scheuchzeri*, showed a greater growth response to nitrogen than did a species

associated with low nutrient sites, *Eriophorum vaginatum*, when grown in a mixed culture. Interestingly, nitrogen use efficiency was higher for *Eriophorum vaginatum*.

Aggressive species change the growth of competing species by depleting resources. Fetene (2003) found that *Hyparrenia hirta* (L.) was more aggressive than *Acacia etabaica* Schweinf. It gained advantage by reducing the growth of *Acacia* seedlings through resources depletion. In some invasive woody species, resources such as nutrients and/or water, are captured more rapidly and in relatively larger amounts compared to native species. In fact, their productivity is frequently the reason such species are introduced in the first place.

Plant root biomass is a good indicator of below-ground competitive ability. The differences in plant root systems or architecture may contribute to their competitive ability for water and nutrients. The ability of a species to occupy an ecosystem niche can depend on several root characteristics such as relative growth rate, biomass, fine root density, root length, root depth and total surface area. For example, the thinner roots in *Agropyron desertorum* have twice the root length of *Agropyron spicatum* and this gives *A. desertorum* a distinct competitive advantage in sites with limited resources (Eissenstat and Caldwell 1988). Another example of root architecture and its ability to determine plant competitiveness can be found in *Prosopis glandulosa* (Torr.) which has an extensive lateral root system that contributes to efficient plant water uptake (Ansley et al. 1998).

Competition between plant species can also affect overall plant development through reduced leaf production and associated suppressed growth. Daugovish et al. (2002) found, in a replacement design, that yellow mustard mixed with oats reduced wild oat leaf number when compared to wild oat plants in monoculture. A species' growth pattern can significantly vary between a monoculture or mixed stand. For example, Knee and Thomas (2002) found significant differences in leaf area ratios and the relative growth rates between monocultures and mixtures. In other experiments, Patterson and Highsmith (1989) found a reduction in leaf area in cotton, *Gossypium hirsutum*, when competiting with velvetleaf, *Abutilon theophrasti* in a water-limited environment. Furthermore, the reduction in leaf area reduced the relative yield of

cotton. The same pattern has been observed in several perennial species where a reduction of seed numbers and vigor accompanied factors that reduce leaf area.

Plant height is also a good indicator of plant competitiveness. Estorninos et al. (2002) found, in a replacement series experiment, that the height of rice was reduced when grown at 1:2 rice-red rice mixtures. Caldwell et al. (1995) found that *Pinus resinosa* (red pine) seedling shoot and root growth was suppressed by three grass species due to competition.

Nitrogen, a macronutrient, is often growth limiting and therefore affects the growth of plants. The degree of growth reduction generally increases with increased levels of nitrogen deficiency. Sindel and Michael (1992) demonstrated that *Senecio madagascariensis* increased its relative growth when grown in mixture with oats (*Avena strigosa* cv. Saia). The relative growth rate of *S. madagascariensis* in mixed stands was greater at higher N and P levels. A greater percentage of dry matter was also partitioned into stems and flowering capitula thereby raising its relative reproductive and invasive potential. In another study, two greenhouse experiments were used to investigate the influence of amount and form of added nitrogen on the growth of maize and redroot pigweed (*Amaranthus retroflexus*) (Teyker et al. 1991). In this study, maize and pigweed were grown together in replacement series experiments. Pigweed responded more to supplemental N than maize and accumulated 2.5 times as much N in shoots at the high N supply than in the low nitrogen supply treatments. In another study, Cralle et al. (2003), found that low phosphorus levels did not affect the relative competitiveness of Italian ryegrass as much as that of wheat in a mixed culture. The dominant species may therefore be the species that can reduce the concentration of the limiting resource to low levels and still maintain vigor (Wedin and Tilman, 1993). In support of this argument, Wedin and Tilman (1993) found that *Agrpyron repens* displaced *Agrostis scabra* in all added-N plots after five years.

Materials and methods.

Plant material

For this study seeds of *Ziziphus spina-christi* were collected from trees grown at the Agriculture Research Station in Derab, Riyadh, Saudi Arabia (latitude 24º 34′, longitudinal 46º 43′). The research station is located at an elevation of 500 m and receives an average rainfall of 83 mm/y. Evaporation is measured at 2739.5 mm/y. The maximum average temperature is 48ºC in August and the average minimum temperature is 31.8ºC in December. Seeds of *Prosopis juliflora* were obtained from Lawyer Nursery Inc, USA.

Experimental design

Seed coats were removed from both species using a small pincher then seeds of *P. juliflora*, were mechanically scarified. On 11/8/02, seeds of *Z. spina christi* were placed in a closed transparent container filled to a depth of 1.5 cm with sterilized mortar sand. Four days later, seeds of *P. juliflora* were sown in separate containers because of their rapid germination. Before germination, sand was sterilized in an autoclave then containers and mortar sand were treated with fungicide. Containers were placed in a mist bed with bottom heat at a temperature of 25ºC.

After germination seedlings were transplanted into plastic pots (15.5 cm diameter by 15 cm deep) containing 2275 g of sterile sand. Sand was chosen because it is nutrient poor and easy to wash during the harvest. Before transplanting, a circular template with multiple overlapping rings was used to mark the planting location for every individual seedling to ensure that they were equally spaced, ≈ 4 cm, from neighboring plants in all densities in monoculture and mixture. The exceptions to this was the density of one plant per pot. Greenhouse day:night temperatures varied between 35-25ºC with additional illumination (sodium light) providing a 13-h/day photoperiod.

This experiment utilized a De Wit (1960) replacement series where the density of plants was held constant at six plants per pot (Cohn et al. 1989, Van Auken and Bush 1987, Bi and Turvey 1994) while varying the proportion of the two species. The proportions of *Z. spina-christi: P. juliflora* (Z:P) in mixture were 6:0, 4:2, 3:3, 2:4 and

0:6 plants/ pot and had the same order as in Figure 1 (Appendix A). The monoculture series for every species consisted of 1, 2, 3, 4, and 6 plants per pot. The experimental design was a randomized complete block design with four replications where pots were set randomly within blocks on the greenhouse table. Two levels of nitrogen, 20 ppm and 100 ppm, were used in the form of NH_4NO_3. Each pot name included the following information: block number, treatment, density of *Z. spina-christi,* and density of *P. juliflora* (e.g., Z:P, 4:2, HN, B1, consists of 4 *Z. spina-christi* plants and 2 *P. juliflora* plants with high nitrogen and placed in block 1).

Two weeks after transplanting treatments were started by adding 300 ml of Johnson's nutrient solution (Johnson et al. 1957) where N was varied in concentration between 20 and 100 ppm but amounts of all other nutrients were held constant. The interval between nutrient additions was 14 days and pots were irrigated with 300 ml of tap water between nutrient additions. Nutrient solutions or water was added to the center of the pot. After 211 days, the plants were harvested.

Before harvest, two leaves from the upper third of the plant were taken from randomly chosen plants to measure chlorophyll content. For *Z. spina-christi* leaves were sliced into small pieces then weighted. For *P. juliflora*, leaflets were removed and chosen randomly for chlorophyll analysis. For each test, approximately, 0.02 g of fresh leaf tissues was transferred to a test tube then placed on ice. Two millileters of cold methanol were added to the tubes with tissue then sealed with Parafilm and extracted overnight in the refrigerator at 5 ºC. After 24 hr a 0.5 ml sample of extract was used to determine chlorophyll using a spectrophotometer at two wavelengths, 665.2 nm and 652.0 nm. Chlorophyll *a* and *b* concentrations were calculated using the following equations (Porra et al. 1989):

Chlorophyll concentrations in nmol/ml=

$$\text{Chl } a = 18.22\, A^{665.2} - 9.95\, A^{652.0}$$

$$\text{Chl } b = 33.78\, A^{652} - 14.96\, A^{652.0}$$

$$\text{Chls } ab = 24.93\, A^{652.0} - 9.95\, A^{665.2}$$

Where A is the absorbance at the given wavelength.

Prior to harvest, the following measurements were made for each individual seedling in pots: stem height from the ground to the stem tip (cm). Stem basal diameter

above the cotyledon scars (mm), number of leaves longer then 0.5 cm, total leaf area (cm^2), root dry weight (g), stem dry weight (g), and leaf dry weight (g). Stem basal diameter was measured with digital caliper (± .04 mm) and leaf area with a Li-3100 area meter (LiCor Inc., Lincoln, Nebraska, USA). The leaflets of *P. juliflora* tended to shed as plants grew, leading to a reduction leaf area while *Z. spina-christi* seedlings retained most of their leaves. From each pot, the upper part of plants was collected. For roots, sand was removed gently by water to avoid breaking roots. Intertwined roots of the two species in mixed culture were separated whereas roots of species monocultures within pots were kept intact because of the difficulty of separation. They were then transferred to a paper bag. Plant tissues (leaf, stem, and root) were dried separately in for 48 hr at 75 ºC. Stem, leaf, and root dry weights were measured to the nearest milligram.

Data analysis

Relative yield

Relative yield of the two species was calculated using the following formula, according to (Snyder et al. 1994):

$$\text{Relative Yield (RY) of } \textit{Z. spina-christi} = \frac{Y_{ZP}}{(pY_{ZZ})}$$

$$\text{Relative Yield (RY) of } \textit{P. juliflora} = \frac{Y_{PZ}}{(qY_{PP})}$$

Where Y_{ZP} is the yield of the *Z. spina-christi* grow in the mixed planting and *p* is the initial proportions (0.67, 0.50, and 0.33) of *Z. spina-christi,* multiplied by the Y_{ZZ} which is the mean yield of *Z. spina-christi* grown in monoculture (6 plant per pot). Similarly, Y_{PZ} is the yield of *P. juliflora* grow in mixture with *Z. spina-christi* and "q" is the proportions of the species (0.67, 0.50, and 0.33), multiplied by the mean yield of *P. juliflora* grown in monoculture (6 plant per pot). Relative yield of the two species was used to calculate the relative yield total per pot.

Relative yield total (RYT)

Relative yield total (RYT) was determined to indicate if the species were sharing resources or interfering with each other by comparing their intraspecific yield per pot in monoculture to their interspecific yield per pot in pure culture (De Wit 1960; Harper 1977). The following formula was used to calculate (RYT):

$$\textbf{RYT}= \quad = pRY_Z + qRY_P$$

Where p = initial proportion of *P. juliflora* in a mixture and q initial proportion of *Z. spina-christi* in a mixture such that p+q=1.

The results of replacement experiments can be analyzed graphically using a replacement diagrams. When the competition between the two species is equal each species will contribute to the total yield in direct ratio to its proportion in the mixture and there is a linear increase in the yield for each species with its proportion in the mixture. When the competition is not equal the more aggressive species gains more yield than would be expected. The aggressivity index for the two species will be determined by the following formulas:

Aggressivity of *Z. spina-christi* $= RY_Z - RY_P$

Aggressivity of *P. juliflora* $= RY_P - RY_Z$

Where p = initial proportion of *P. juliflora* in a mixture and q initial proportion of *Z. spina-christi* in a mixture so that p+q=1 in a mixture of two species.

Results

Height

Both species (P=0.0457) and species × nitrogen level (P=0.0004) were significantly associated with seven-month height. For mixed plantings with high nitrogen, the height of *Z. spina-christi* decreased with increasing proportion of *P. juliflora*. Conversely, *P. juliflora* showed a reduction in height with decreasing proportions of *Z. spina-christi* (Table 1). Under the low level of nitrogen, *Z. spina-christi* showed an increase in height with an increasing proportion of *P. juliflora* (Table 1). T-test results indicated significant differences in height between seedlings within and between nitrogen levels for *P. juliflora* (Table 2, shaded cells). Conversely, *Z. spina-christi* heights were not significantly different between plants grown in different proportions (Table 2, non-shaded cells).

In the monoculture plantings, *P. juliflora* height was significantly affected by nitrogen (P=0.0001) and plant density (P=0.0001) but the interaction between nitrogen and density was not significant (P=0.1105). With high nitrogen, the height of plants decreased with increasing density per pot (Table 3). The reduction in height was 63% at the density of 6 plants/pot compared to a single plant. Height of plants grown in low nitrogen was also reduced as the number of plants per pot increased. The height for 6 plants per pot was 63% compared to the single plant per pot grown at the same level of nitrogen (Table 3). Results of a t-test indicated significant differences in height between seedlings of *P. juliflora* within and between levels of nitrogen (Table 4, shaded cells)

Height of *Z. spina-christi* monocultures was significantly affected by nitrogen (P=0.0001), plant density (P=0.0001), and their interaction (P=0.0992). The height decreased with increasing density under the low level of nitrogen (Table 3). However, the effect of intraspecific competition on height was greater in plants grown at low nitrogen levels where the reduction in height at 6 plants was 45% comparing to the height of individual plants grown alone. There were significant differences in height between seedlings within and between levels of nitrogen as determined by a t-test (Table 4, non-shaded cells).

Diameter

In the replacement series study, seedling diameter was significantly affected by nitrogen (P=0.0068), species (P=0.0219) the interaction between nitrogen and number of plants (P=0.0730), and the interaction between nitrogen and species (P=0.0001). With high levels of nitrogen, diameter of *Z. spina-christi* decreased with increasing proportion of *P. juliflora.* In contrast, diameter of *P. juliflora* decreased as its proportion increased in the pot (Table 5). With the low nitrogen treatment, the diameter of *Z. spina-christi* increased with increasing proportion of *P. juliflora*. There was a significant difference in diameter in *P. juliflora* within and between levels of nitrogen (Table 6, shaded cells).

In *P. julifora* monoculture pots, diameter was significantly affected by nitrogen (P=0.0001) and number of plants per pot (P=0.0001) but the interaction between nitrogen and number of plants was not significant (P=0.1918). The diameter per plant was reduced with increasing plant number in both high and low nitrogen treatments (Table 7). The average diameter for a single plant grown without competition was 6 mm but the average was 3.11 mm for plants grown in monoculture at a density of 6 plants per pot, which was a reduction of 48% (Table 7). As can be seen in Table 8, t-tests indicated significant differences in diameter between seedlings within and between levels of nitrogen for the monoculture tests.

Average diameter for *P. juliflora* at the low level of nitrogen was lower than high N and diameter was further reduced with increasing plant density per pot, where the average diameter for plants in pots with a density of 6 plants was 2.52 mm but at a density of one plant per pot the average diameter was 4.63 mm (Table 7).

Nitrogen had no significant effect on the diameter of *Z. spina-christ* in monoculture plantings (P=0.8985). Moreover, the interaction between nitrogen and number of *Z. spina-christi* plants per pot was not significant with respect to diameter (P=0.5752). Diameter of *Z. spina-christi* plants was, however, affected by density (P=0.0001), where diameter of plants was reduced by approximately 35% with increasing plant density (Table 7).

Under low levels of nitrogen *Z. spina-christi* plants had a slightly higher (not significant) average diameter in pots with one and two plants per pot compared to the

same densities grown in high nitrogen (Table 7). However there was a greater reduction in diameter with increasing plant density in the low N treatment compared to the high N treatment. The t-tests indicated significant differences in diameter from monoculture seedlings at different densities within and between levels of nitrogen (Table 8, non-shaded cells).

Leaf area

Leaf area in the replacement series study was significantly affected by species (P=0.0081), the interaction between nitrogen and proportions (P=0.0004), and the interaction between nitrogen and species (P=0.0001).

At the high level of nitrogen, leaf area in *Z. spina-christi* in mixture at the proportions of 0.67 (4Z:2P) and 0.50 (3Z:3P) did not differ significantly with increasing proportions of *P. juliflora* (Table 9). In contrast, leaf area of *P. juliflora* decreased with decreasing proportion of *Z. spina-christi* (Table 9). This seems to indicate that intraspecific competition among *P. juliflora* plants is more intense for individual plants of that species than competition with *Z. spina-christi*. Results of a t-test indicated significant differences in leaf area between seedlings of the same species within and between levels of nitrogen and within and between proportions of the two species (Table 10).

Under the low level of nitrogen and in mixed plantings, there was an increase in leaf area in *Z. spina-christi* with increasing proportions of *P. juliflora*, where the highest leaf area occurred in the proportion 0.33:0.67 (Z:P) (Table 9). Leaf area in *P. juliflora* was greatly reduced in the low level of nitrogen compared to the high level (Table 10).

Leaf area in *P. juliflora* in the monoculture was highly significantly affected by nitrogen (P=0.0001) and density of plants (0.0001). Under high levels of nitrogen, leaf area decreased with increasing numbers of plants. Leaf area for one plant per pot was 85.65 cm^2 compared to 19.13 cm^2 at 6 plants per pot (Table 11). Under low nitrogen, leaf area was dramatically reduced with increasing density. In the density of 6 plants per pot the average leaf area per plant was 5.55 cm^2, a reduction of almost 8- fold

compared to the single-plant pots. A t-test indicated significant differences in leaf area between seedlings and between levels of nitrogen (Table 12, shaded cells).

In monoculture, *Z. spina-christi*, density had a significant effect on leaf area (P=0.0001) whereas nitrogen and the interaction between plant density and nitrogen had no significant effect (P=0.4043) and (P=0.2929), respectively. Leaf area for single plants/pot of *Z. spina-christi* was approximately 160 cm^2. With increasing density of *Z. spina-christi*, average leaf area decreased between 33 cm^2 and 57 cm^2 for the 6 plants per pot treatment in low and high levels of nitrogen, respectively (Table 11). Results of t-tests indicated significant differences in leaf area between planting densities within and between levels of nitrogen (Table 12, non-shaded cells). In general *P. juliflora*, although it had lower leaf areas per seedling, was much more responsive to nitrogen fertilization while *Z. spina-christi* was more responsive to planting density.

Total dry weight

In mixed plantings, total dry weight per seedling was significantly affected by nitrogen (P=0.0116), species (P=0.0001), the interaction between nitrogen and species (P=0.0001), and the interaction between planting proportion and species (P=0.0001).

Average total dry weight per plant for *Z. spina-christi* in the 100 ppm nitrogen treatment decreased slightly with increasing proportion of *P. juliflora* (Figure 1). At the proportion (0.50:0.50) dry weights were 0.69 g and 2.51g for *Z. spina-christi* and *P. juliflora*, respectively. In *P. juliflora* the dry weight per plant decreased dramatically as its proportion in the pot was increased, which appeared to be more a function of intraspecific competition, as was verified by the monoculture experiment discussed later.

Under low nitrogen there was a reversal in response of total dry weight for the two species. *Z. spina-christi* gained biomass with increasing proportions of *P. juliflora*. In the proportion (0.33 Z:0.67 P) the average total dry weights were 2.03 g and 0.76 g for *Z. spina-christi* and *P. juliflora*, respectively (Figure 2). Conversely, *P. juliflora* plants tended to have slightly lower dry weights as their proportions increased in the pot, but not nearly so much so as in the high nitrogen treatments.

Total dry weight in monoculture of *P. juliflora* was significantly affected by nitrogen (P=0.0001), plant density (P=0.0001), and the interaction of nitrogen by density (P=0.0001). At the level of 100 ppm of nitrogen, dry biomass of *P. juliflora* decreased with increasing plant density. At the density of one plant per pot the total dry weight was 13.13 g but the dry weight per plant decreased at 6 plants per pot to 1.56 g **(**Figure 3**)**. Under low levels of nitrogen, per-plant dry weight of *P.juliflora* decreased with increasing plant density, but not as greatly as under the high nitrogen regime. The average dry weight per plant at 6 plants per pot was 0.67 g compare to 6.56 g for the single plant pots (Figure 4). It is obvious from this figure that total dry weight per plant for *P. juliflora* is highly responsive to nitrogen, which was not the case for *Z. spina-christi*. Results of t-tests showed significant differences in average total dry weight per plant for the two species within and between levels of nitrogen and at different pot densities (Table 13, shaded cells).

In *Z. spina-christi* the total dry weight in monoculture was significantly affected by nitrogen (P=0.0478) and plant density (P=0.0001), but the interaction between nitrogen and plant density was not significant (P=0.090). For both 100 and 20 ppm of nitrogen, with increasing density, plant dry weight decreased (Figure 3). It was surprising that at low N dry weight per plant in pots containing 1, 2, and 3 plants exceeded dry weights for plants grown in high levels of nitrogen at the same density (Figure 3). There were significant differences in total dry weight in *Z. spina-chrsti* between densities and between levels of nitrogen as indicated by t-tests (Table 13, non-shaded cells).

Root to shoot ratio

In mixed plantings root to shoot ratio was significantly affected by nitrogen (P=0.0002), the interaction between nitrogen and species (P=0.0002), and the interaction between nitrogen and proportion by species (P=0.0243). At the high level of nitrogen, *Z. spina-christi* root to shoot ratio showed an increase with increasing proportion of *P. juliflora* (Figure 5). In contrast, a reduction in root to shoot ratio for *P. juliflora* is accompanied by decreasing the proportion of *Z. spina-christi* (Figure 5). A t-test indicated significant differences in root to shoot ratio within and between

proportions and under high and low levels of nitrogen for *P. juliflora* and *Z. spina-christi* (Table 14).

With low nitrogen, *Z. spina-christi* tended to have a lower root to shoot ratio with increasing proportion of *P. juliflora* (Figure 6). *P. juliflora* responded dissimilarly in low nitrogen where there was little change in root to shoot ratio with increasing proportions of *Z. spina-christi*. In *Z. spina-christi* there were significant differences in root to shoot ratio in mixed plantings as the proportion of the species changed within and between nitrogen levels (Table 14, non-shaded cells).

Root to shoot ratio in monoculture plantings for *P. juliflora* was highly significant for nitrogen levels (P=0.0001). In 100 ppm of nitrogen, *P. juliflora* root to shoot ratio did not show a trend as its density increased from 1 to 6 plants per pot (Table 15). However, under low levels of nitrogen *P. juliflora* responded by increasing root to shoot ratio with increasing density. There were significant differences within and between levels of nitrogen in root to shoot ratio for *P. juliflora* in monoculture plantations as elucidated by t-tests (Table 16, shaded cells).

Root to shoot ratio in *Z. spina-christi* was significantly affected only by nitrogen (P=0.0001). In the 100 ppm nitrogen treatment, the species increased root to shoot ratio with increasing density per pot (Table 15). At a density of 6 plants per pot, however the species had a slightly lower ratio. In the low level of nitrogen the species showed an increased root to shoot ratio with increasing numbers of plants per pot. Results of t-tests indicated significant differences in root to shoot ratio for *Z. spina-christi* (Table 16, non-shaded cells). In general, *Z. spina-christi* maintained a higher root to shoot ratio than *P. juliflora* and *P. juliflora* responded more dramatically to lower N levels by increasing its root to shoot ratio. Higher planting density seemed to promote a higher root to shoot ratio, especially for *P. juliflora* at the lower N level.

Relative yield and relative yield total

Relative yield in the mixed plantings was highly significant for species (P=0.0001), the interaction between nitrogen and species (P=0.0001), and the interaction between nitrogen and species proportion (P=0.0049). The interaction

between nitrogen and proportion ratio did not significantly affect relative yield total (P=0.0866).

Relative yield in the high nitrogen level for *Z. spina-christi* decreased significantly with increasing proportion of *P. juliflora* (Table 17). For *P. juliflora*, the relative yield also decreased as its density proportion increased. However, at the proportion of (0.50:0.50), the relative yield of *P. juliflora* exceed that for *Z. spina-christi* (Table 17). This probably reflects a condition where resources other than N were limiting to the growth of *P. juliflora* and lower relative yields are a function of intense intraspecific competition. Under the low nitrogen level (20 ppm), the relative yield of *Z. spina-christi* increased with increasing the proportion of *P. juliflora*. Relative yield totals, except (0.33:0.67) in high level of nitrogen, were higher than 1 (Table 17).

Results of t-tests indicated significant differences in relative yield for both *P. juliflora* and *Z. spina-christi* within and between proportions and within and between levels of nitrogen (Table 18).

Aggressivity

Analysis of variance indicated that the effect of adding nitrogen was highly significant regarding the aggresivity of the two species (P=0.0001).

The aggeresivity index in the high nitrogen level shows that *P. juliflora* is more aggressive than *Z. spina-christi*, but *P. juliflora*'s aggresivity decreased as its proportion in mixture was increased (Table 19). At the low level of nitrogen *P. juliflora* and *Z. spina-christi* are almost equal in aggressivity except in the proportion (0.33 Z: 0.67 P) where *Z. spina-christ* is slightly more aggressive (Table 19). This may be related to greater intraspecific competition between plants of *P. juliflora* in a more resources limited environment.

Total chlorophyll *ab*

In mixed plantings, proportion of species in the pots is the only variable that was significantly associated with total chlorophyll *ab* (P=0.0419).

In *Z. spina-christi,* the total chlorophyll *ab* decreased with an increasing proportion of *P. juliflora* in the high level of nitrogen (Table 20), but the difference was only significant for the 4:2 (Z:P) versus the 2:4 (Z:P) ratio (Table 21). At the same level of nitrogen *P. julilfora* displayed the same trend, where chlorophyll *ab* decreased as its proportion increased, but these differences were not significant (Table 21). In fact, there were no significant differences among means for chlorophyll *ab* that were compared for the different proportion mixtures and nitrogen levels for *P. juliflora* (Table 21, shaded cells). In *Z. spina-christi* there were a few significant differences between means in proportion involving both high and low levels of nitrogen (Table 21).

In monoculture plantings, nitrogen significantly affected the total chlorophyll *ab* in both *Z. spina-christi* and *P. juliflora* (P=0.0001). Number of plants (density) and the interaction between number of plants and nitrogen had low significant effect on total chlorophyll *ab* for *P. juliflora* (P=0.0571) and *Z. spina-christi* (P=0.0495). In *Z. spina-christi*, total chlorophyll *ab* was reduced 50% in plants grown at the low level of nitrogen, compared to plants at the high level (Table 22). *P. juliflora* plants grown in the high level of nitrogen were also higher in total chlorophyll *ab* compared to plants grown in low nitrogen (Table 22). There were many significant differences between means in total chlorophyll *ab* for *P. juliflora* in different densities and under high and low levels of nitrogen (Table 23, shaded cells). Also, there were some significant differences in means for *Z. spina-christi* grown in different densities under high and low levels of nitrogen (Table 23, non-shaded cells). The species that was found to be less aggressive under high nitrogen conditions (*Z. spina-christi*) had the highest concentration of chlorophyll *ab*. However, as mentioned in a previous section, *P. juliflora* compensated by having higher leaf area than *Z. spina-christi* under high N levels (Table 9).

Discussion:

Interspecific competition is believed to be less intense between individuals under conditions where resources are severely limited (Grime, 1988). In terms of their performance in a controlled environment, *Z. spina-christi* and *P. juliflora* showed different responses to high and low nitrogen levels which indicates that the amount of nitrogen plays an important role in the interference between the two species. At high nitrogen levels the growth and total yield of *P. juliflora* in mixture was superior to that of *Z. spina-christi, P. juliflora* having greater dry weight indicating its ability to compete and dominate. Austian et al. (1985) found that *Carthamus lanatus* was dominant over *Silybum marianum* at low levels of nutrients but at high nitrogen levels *Silybum marianum* dominated. The aggresivty of *P. juliflora* under high nitrogen indicates its ability to dominate under such conditions. Although in mixed culture the presence of *Z. spina-christi* reduced the total dry weight per plant of *P. Juliflora*. The species was affected more by intraspecific competition as indicated by the dry weight production per plant of the species comparing the same number of plants in monoculture and polyculture. For example, at the high nitrogen level the total dry weight of 2 plants of *P. juliflora* in mixture with 4 *Z. spina-christi* was 3.71 g but the dry weight per plant for 6 plants *P. juliflora* in the monoculture was approximately 1.5 g.

In mixed cultures containing equal numbers of each species (3:3) the total dry weight of *P. juliflora* was greater than *Z. spina-christi* which reflects the greater ability of *P. juliflora* to convert resources to biomass. Competitive ability for soil resources (nutrients, water) is an important trait because competition for belowground resource can limit plant growth and plant dry weight. My results indicate a lower ability of *Z. spina-christi* to compete when nitrogen is abundant, which clearly emphasizes that changing resource availability alters the competitive relationship between the species. One of the more interesting outcomes of this research is the finding that total leaf area for seedlings of *Z. spina-christi* was independent of nitrogen fertilization levels, whereas *P. juliflora* responded to fertilization by more than doubling its leaf area. Furthermore, in mixed culture, under high N levels, *P. juliflora* produced a leaf area

about double the amount produced by *Z. spina-christi*. This situation was reversed under low N.

Root to shoot ratio can explain the ability of a species to allocate resource toward different structures. The root to shoot ratio in *P. juliflora* increased with increasing proportion of *Z. spina-christi* indicting that *P. juliflora* had aggressive root growth and a larger root system enables plants to rapidly obtain resources, an attributes for a species that is a strong competitor for resources (Grime 1977). Additionally, *P. juliflora* gained more biomass and out-competed *Z. spina-christi* at the high level of nitrogen which indicates that the native species has no competitive advantage over the exotic species in high nitrogen. Under low levels of nitrogen *Z. spina-christi* apperentely has an 'investment strategy' to occupy and survive in unfavorable environments (Mynhardt et al. 1994). The fact that *P. juliflora* is a legume capable of N fixation probably plays an important role in the competitive strategy of the two species under field conciliations where inoculation with nitrogen-fixing bacteria is possible.

Relative yield of *P. juliflora* exceeds that of *Z. spina-christi* and the interspecific competition is less intense than intraspecific competition in reducing RY for *P. juliflora* when the two species are grown together. Under the low level of nitrogen the native species had a competitive advantage over the exotic where *Z. spina-christi* gains more dry weight. Harper (1977) demonstrated that the competitive advantage could be shifted from a nitrogen fixer to a non-nitrogen fixer by manipulating soil nutrients (particularly nitrogen). Also, under low N relative yield (RY) of *Z. spina-christi* exceeded that of *P. juliflora* at the ratio of 0.33 Z: 0.67 P which may relate to the effect of intraspecific competition in a resource-limited environment on *P. juliflora*. Goldberg (1988) found that when nutrients and water supplies were low, the biennial *Melilotus alba* was dominant over *Solidago canadensis* after two years, but when nutrients and water were abundant, a competitive equilibrium was established between the two species.

In this study, relative yield total indicated that the two species were making demands on the same limited resources of the environment. Values of the relative yield total >1.0 suggest that the species may be competing for the same resources. However,

relative yield total for polycultures in the proportion 0.33 *Z*: 0.67 P growing in high nitrogen levels were <1.0 which indicates a mutual antagonism.

Our results were similar to those of Nicotra and Rodenhouse (1995) whose studies show that the intensity of competition significantly decreases with decreasing availability of resources. Nicotra and Rodenhouse (1995) found that reduction in nitrogen lowered the intensity of competition. However, intraspecific competition can affect the growth of individual plants which in turn affects their interactions with neighbors of other species.

Height of plants grown singly in pots exceeded heights of plants growing in monocultures of high density (6 per pot) at high levels of nitrogen and that may also indicate that the negative effect of competition is higher at higher nutrient levels. However, as plants grow larger in higher nitrogen levels, their resource demands are also increased (Lentz 1999).

Lentz (1999) found that root to shoot ratio increased with increasing density in intraspecific competition and with different nutrient concentrations. However, an increase in root:shoot is a common response of plants to lowered nutrient supply (Chapin 1980). The results of this study verifiy the importance of intraspecific competition on increasing root: shoot ratio under the lower level of nitrogen in *Z. spina-christi* and *P. juliflora*. In the monoculture experiment, *Z. spina-christi* did not experience a dramatic change in R:S with altered nitrogen (only 2 of 5 comparisons were significant). On the other hand, *P. juliflora* showed a significant increase in R:S with low nitrogen levels for all 5 comparisons. Species that are able to respond to the levels of resource supply by changing their growth allocation from shoot to root can facilitate their ability to capture the most needed resources. It has been reported that plants from nutrient-poor habitats are able to produce more organic matter per unit of mineral nutrient taken up (Vitousek 1982). *Z. spina-christi* may use nutrients more efficiently by producing relatively high amount of dry matter and high leaf area under low levels of nitrogen under field conditions. Nutrient-use efficiency (NUE) is considered to be an important plant characteristic, where NUE is measured as the ratio between total productivity and total nitrogen loss in litter (Vitousek 1982).

Nutrient use efficiency may explain why greater relative yield is characteristic of *Z. spina-christi* grown in mixed plantings at low levels of nitrogen. Nutrient balance of species grown in nutrient-poor ecosystems is determined by the balance between nutrient acquisition and nutrient loss. These plants have a large internal pool of nutrients giving them a high competitive ability based on nutrient balance (Aerts 1999).

In nutrient-poor environments, species produce relatively small amounts of litter due to the long lifespan of the various tissues. That may help explain the relatively high dry weight and leaf area in *Z. spina-christi* grown in low levels of nitrogen. The amount of litter produced under resource-limiting environments may play an important role in plant success, especially considering their high content of secondary compounds such as lignin and phenolics (Aerts 1997).

Based on my results, since the two species had different responses to nitrogen availability in mixtures, invasion is expected for *P. juliflora* where nitrogen is highly available but under low nitrogen availability the species may lose its competitive ability. However, under field conditions, the ability of *P. juliflora* to fix atmospheric nitrogen could confer an advantage to the species that would not appear in greenhouse studies like this one. The aggresivity index shows that *P. juliflora* is a superior competitor in high levels of nitrogen where the species gains more dry weight than *Z. spina-christi*. At the low level of nitrogen the aggresivity index shows that *Z. spina-christi* gains more dry weight as the proportion of *P. juliflora* increases. However, it is obvious that *P. juliflora* was also affected by intraspesific competition. Moreover, more resources are allocated toward roots in *Z. spina-christi* which enables it to acquire nutrients in nutrient-poor environments. In plants like *Z. spina-christ*, the physiological and morphological traits are very important to ensure survival in habitats with nitrogen deficiency.

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Tables and Figures

Table 1. Means and standard errors for the effect of species proportion and level of nitrogen on height per plant for *Ziziphus spina-christi* and *Prosopis juliflora* grown in mixtures at different plant proportions in high and low level of nitrogen

	Plant Height (cm)			
	Level of nitrogen			
***Z. spina: P. juliflora* proportions**	**High ***		**Low****	
	Z	**P**	**Z**	**P**
4:2 (0.67:0.33)	21.91 ±1.22	37.25 ±7.27	18.51 ±0.49	16.38 ±1.66
3:3 (0.50:0.50)	18.33 ±2.00	30.18 ±3.35	20.63 ±2.54	13.99 ±1.07
2:4 (0.33:0.67)	16.96 ±3.22	27.71 ±2.59	22.91 ±2.57	16.66 ±1.99

-* 100 ppm -** 20 ppm.

Table 2. t values (upper values) and p-values (lower values) comparing least squares means for the effect of interaction between nitrogen level and species proportion on height for *P. juliflora* and *Z. spina-christi* grown in various mixture in high and low levels of nitrogen. Pr > |t|.

Z. spina: P. juliflora	4:**2** HLN	3:**3** HLN	2:**4** HLN	4:**2** LLN	3:**3** LLN	2:**4** LLN
4:**2** HLN		-0.73 0.4777	-0.75 0.4624	3.33 0.0039	2.69 0.0154	3.42 0.0033
3:**3** HLN	-0.90 0.3794		1.01 0.1134	3.03 0.0076	2.55 0.0206	1.32 0.2050
2:**4** HLN	-1.73 0.1017	-0.85 0.4098		2.97 0.0086	2.65 0.0169	1.36 0.1918
4:**2** LLN	-0.01 0.9883	-0.87 0.3981	-1.56 0.1380		0.20 0.8420	1.27 0.2229
3:**3** LLN	-0.55 0.5874	-1.44 0.1671	-2.18 0.0438	0.54 0.598		1.29 0.2459
2:**4** LLN	-0.09 0.9316	-0.99 0.3373	-1.84 0.0839	0.07 0.9482	-0.46 0.6508	

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*
- Numbers in bold to make the comparison for height mean of *n* plants of *P. juliflora* to height mean of *n* plants of the same species (e.g. 4:**2** HLN to 2:**4** LLN where t= 3.42 and P= <0.0033)

Table 3. Means and standard errors for the effect of plant density and level of nitrogen on height per plant for *Z. spina-christi* and *P. juliflora* grown in monoculture plantings.

	Plant Height (cm)			
	Z. spina-christi		***P. juliflora***	
	Level of nitrogen			
Density (plant/pot)	**High***	**Low****	**High***	**Low****
1	34.63 ±5.67	37.88 ±2.62	74.88 ±3.03	44.88 ±7.95
2	35.94 ±1.34	32.94 ±1.61	44.44 ±7.48	31.56 ±3.77
3	31.37 ±0.60	27.66 ±0.81	35.46 ±2.80	24.31 ±2.25
4	31.44 ±1.65	23.65 ±1.92	30.78 ±1.5	19.41 ±2.19
6	29.09 ±1.40	20.57 ±0.81	27.62 ±1.12	16.59 ±0.88

.-* 100 ppm -** 20 ppm

Table 4. t- test (upper values) and p-values (lower values) comparing least squares means for the effect of interaction between nitrogen level and plant density on plant height for *P. juliflora* and *Z. spina-christi* grow in monoculture in high and low level of nitrogen. Pr > |t|.

Density And level of N	1 (HLN)	1 (LLN)	2 (HLN)	2 (LLN)	3 (HLN)	3 (LLN)	4 (HLN)	4 (LLN)	6 (HLN)	6 (LLN)
1 (HLN)		5.2181 <0.0001	5.2942 <0.0001	7.5336 <0.0001	6.8560 <0.0001	-8.7954 <0.0001	-7.6695 <0.0001	-9.6469 <0.0001	-8.2196 <0.0001	-10.1376 <0.0001
1 (LLN)-	1.0128 0.3193		0.0760 0.9398	2.3255 0.0276	1.6379 0.1119	-3.5773 0.0012	-2.4514 0.0203	-4.4289 0.0001	-3.0019 0.0054	-4.9195 <0.0001
2 (HLN)	0.4090 0.6854	0.6038 0.5505		2.2394 0.0327	1.5618 0.1288	-3.5012 0.0015	-2.3753 0.0241	-4.3528 0.0001	-2.9258 0.0065	4.8434 <0.0001
2 (LLN)	-0.5259 0.6029	-1.5386 0.1344	-0.9349 0.3573		-0.6776 0.5032	-1.2618 0.2168	-0.1359 0.8928	-2.1133 0.0430	-0.6863 0.4978	-2.6039 0.0142
3 (HLN)	1.0153 0.3180	2.0281 0.0515	-1.4244 0.1647	0.4895 0.6280		-1.9393 0.0619	-0.8135 0.4223	-2.7909 0.0091	-1.3639 0.1827	-3.2816 0.0026
3 (LLN)	-2.1814 0.0371	-3.1941 0.0033	-2.5903 0.0147	-1.6555 0.1083	-1.1699 0.2528		1.1259 0.2691	-0.8515 0.4012	0.5754 0.5693	1.3422 0.1896
4 (HLN)	-0.9915 0.3294	2.0041 0.0542	1.4004 0.1717	0.4654 0.6449	0.0240 0.9810	1.1900 0.2434		1.9774 0.0572	-0.5504 0.5861	-2.4680 0.0195
4 (LLN)	-3.4214 0.0018	-4.4341 0.0001	-3.8303 0.0006	-2.8955 0.0070	-2.4059 0.0225	-1.2399 0.2246	2.4300 0.0213		1.4270 0.1639	-0.4906 0.6272
6 (HLN)	-1.7230 0.0952	2.7357 0.0104	-2.1320 0.0413	1.1971 0.2406	-0.7076 0.4846	-0.4583 0.6500	-0.7316 0.4701	1.6983 0.0988		-1.9177 0.0647
6 (LLN)	-4.3791 0.0001	-5.3918 <0.0001	-4.7881 <0.0001	-3.8532 0.0006	-3.3637 0.0021	-2.1977 0.0358	-3.3877 0.0020	-0.9577 0.3459	-2.6561 0.0125	

-Shaded cells represent *P. juliflora* Non-shaded cells represent *Z. spina-christi*

Table 5. Means and standard errors for the effect of plant proportion and level of nitrogen on diameter per plant for *Z. spina-christi* and *P. juliflora* grow in mixture plantation under two levels of nitrogen.

	Plant Diameter (mm)			
	Level of Nitrogen			
***Z. spina: P. juliflora* proportions**	**High***		**Low****	
	Z	**P**	**Z**	**P**
4:2 (0.67:0.33)	2.13 ±0.12	3.89 ±0.16	2.16 ±0.10	2.38 ±0.04
3:3 (0.50:0.50)	2.03 ±0.09	3.78 ±0.22	2.22 ±0.29	2.33 ±0.12
2:4 (0.33:0.67)	1.57 ±0.31	3.29 ±0.14	2.61 ±0.12	2.50 ±0.19

-* 100 ppm -** 20 ppm

Table 6. t- test (upper values) and p-values (lower values) comparing least squares means for the interaction between nitrogen level and species proportion on diameter *P. juliflora* and *Z. spina-christi* grow in mixture in high and low level of nitrogen. Pr > |t|.

Z. spina: P. juliflora	4:**2** HLN	3:**3** HLN	2:**4** HLN	4:**2** LLN	3:**3** LLN	2:**4** LLN
4:**2** HLN		1.34 0.3405	-0.46 0.6514	5.29 <0.0001	3.08 0.0068	1.94 0.0690
3:**3** HLN	-1.05 0.3099		0.48 0.6392	6.71 <0.0001	4.29 0.0005	2.79 0.0125
2:**4** HLN	-1.29 0.2156	-0.38 0.7052		5.27 <0.0001	4.21 0.0006	2.57 0.0199
4:**2** LLN	-0.49 0.6320	-1.46 0.1613	-1.60 0.1282		1.07 0.3007	2.05 0.0556
3:**3** LLN	-0.92 0.3721	-2.00 0.0620	-2.23 0.0395	0.42 0.6833		1.43 0.1703
2:**4** LLN	-0.60 0.5576	-1.62 0.1231	-2.19 0.0426	0.18 0.8565	-0.17 0.8695	

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*
- Numbers in bold to make the comparison for diameter mean of *n* plants of *P. juliflora* to diameter mean of *n* plants of the same species (e.g. 4:**2** HLN to 2:**4** LLN where t= -1.94 and P= 0.0690)

Table 7. Means and standard errors for diameter per plant incorporating the effect of plant density and level of nitrogen for *Z. spina-christi* and *P. juliflora* grown in monoculture planting under two levels of nitrogen.

	Plant Diameter (mm)			
	Z. spina-christi		***P. juliflora***	
	Level of nitrogen		**Level of nitrogen**	
Density (Plant/pot)	**High***	**Low****	**High***	**Low****
1	3.65 ±0.27	3.88 ±0.24	6.0 ±0.24	4.63 ±0.36
2	3.21 ±0.24	3.39 ±0.02	4.69 ±0.06	3.95 ±0.16
3	2.82 ±0.15	2.74 ±0.15	4.15 ±0.16	3.21 ±0.16
4	2.56 ±0.10	2.38 ±0.11	3.52 ±0.5	2.93 ±0.07
6	2.43 ±0.08	2.21 ±0.15	3.11 ±0.06	2.52 ±0.13

-* 100 ppm -**20 ppm

Table 8. t- test (upper values) and p-values (lower values) comparing least squares means for of interaction between nitrogen level and plant density on plant diameter for *P. juliflora* and *Z. spina-christi* grown in monoculture in high and low levels of nitrogen. Pr > |t|.

Density and level of N	1 (HLN)	1 (LLN)	2 (HLN)	2 (LLN)	3 (HLN)	3 (LLN)	4 (HLN)	4 (LLN)	6 (HLN)	6 (LLN)
1 (HLN)		5.3314 <0.0001	5.0891 <0.0001	7.9487 <0.0001	7.1732 <0.0001	-10.8245 <0.0001	-9.6209 <0.0001	-11.8989 <0.0001	-11.2123 <0.0001	-13.4903 <0.0001
1 (LLN)	-0.9531 0.3481		-0.2423 0.8102	2.6172 0.0138	1.8417 0.0754	-5.4930 <0.0001	-4.2894 0.0002	-6.6574 <0.0001	-5.8807 <0.0001	8.1587 <0.0001
2 (HLN)	1.8534 0.0737	2.8066 0.0087		2.8596 0.0076	2.0841 0.0854	5.7353 <0.0001	4.5317 <0.0001	6.8097 <0.0001	6.1231 <0.0001	8.4011 <0.0001
2 (LLN)	1.1120 0.2750	2.0652 0.0476	-0.7436 0.4642		-0.7754 0.4441	2.8757 0.0072	1.6721 0.1049	-3.9501 0.0004	-3.2635 0.0027	-5.5415 <0.0001
3 (HLN)	3.5302 0.0014	4.4834 <0.0001	1.6768 0.1040	2.4182 0.0219		3.6512 0.0010	-2.4476 0.0204	4.7256 <0.0001	-4.0390 0.0003	-6.3170 <0.0001
3 (LLN)	-3.5302 0.0014	-4.4834 <0.0001	-1.9946 0.0552	-2.7359 0.0103	-0.3177 0.7529		1.2036 0.2381	-1.0743 0.2912	0.3877 0.7009	2.6657 0.0123
4 (HLN)	-4.6334 <0.0001	-5.5866 <0.0001	-2.7800 0.0093	-3.5214 0.0014	-1.1032 0.2787	0.7855 0.4383		2.278 0.0300	1.5913 0.1220	3.8693 0.0005
4 (LLN)	5.3836 <0.0001	-6.3368 <0.0001	3.5302 0.0014	4.2716 0.0002	1.8534 0.0737	1.5356 0.1351	0.7502 0.4590		0.6866 0.4976	1.6240 1420
6 (HLN)	5.1718 <0.0001	-6.1250 <0.0001	3.3184 0.0024	4.0598 0.0003	1.6415 0.1111	1.3238 0.1956	0.5384 0.5943	0.2118 0.8337		2.2776 0.0260
6 (LLN)	6.0862 <0.0001	7.0393 <0.0001	-4.2328 0.0002	4.9742 <0.0001	2.5559 0.0159	2.2382 0.0328	1.4527 0.1567	0.7025 0.4878	-0.9143 0.3678	

-Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*

Table 9. Means and standard errors indicating the effect of plant proportion and level of nitrogen on leaf area (cm^2) per plant for *Z. spina-christi* and *P. juliflora* grown in different proportions in mixed plantings

	Leaf Area (cm^2)			
	Level of nitrogen			
***Z. spina: P. juliflora* proportions**	**High***		**Low****	
	Z	**P**	**Z**	**P**
4:2 (0.67:0.33)	22.66 ±3.79	47.60 ±3.30	35.58 ±3.57	4.57 ±0.75
3:3 (0.50:0.50)	23.99 ±5.66	34.18 ±7.42	46.88 ±11.08	4.09 ±0.79
2:4 (0.33:0.67)	13.77 ±4.46	20.02 ±1.60	51.04 ±5.55	6.63 ±1.33

-* 100 ppm -** 20 ppm

Table 10. t- test (upper values) and p-values (lower values) comparing least squares means for the effect of interaction between nitrogen level and species proportion on leaf area for *P. juliflora* and *Z. spina-christi* grow in mixture in high and low level of nitrogen. Pr > |t|.

Z. spina: P. juliflora	4:**2** HLN	3:**3** HLN	2:**4** HLN	4:**2** LLN	3:**3** LLN	2:**4** LLN
4:**2** HLN		-2.79 0.0125	-4.96 0.0001	6.61 <0.0001	6.05 <0.0001	5.66 <0.0001
3:**3** HLN	0.10 0.9204		2.31 0.0015	5.39 <0.0001	5.11 <0.0001	4.69 0.0002
2:**4** HLN	0.13 0.8971	0.09 0.9323		3.08 0.0068	3.21 0.0052	2.82 0.0119
4:**2** LLN	-0.94 0.3580	-0.66 0.5169	-0.37 0.7131		-0.49 0.6309	-0.13 0.9006
3:**3** LLN	-4.07 0.0008	-4.01 0.0009	-2.64 0.0171	2.83 0.011		0.37 0.7125
2:4 LLN	-3.55 0.0025	-4.59 0.0003	-6.09 <0.0001	2.65 0.0169	1.34 0.1971	

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*
- Numbers in bold to make the comparison for leaf area mean of *n* plants of *P. juliflora* to leaf area mean of *n* plants of the same species (e.g. 4:**2** HLN to 2:**4** LLN where t= -5.66 and P= <0.0001)

Table 11. Means and standard errors indicating the effects of plant proportion and level of nitrogen on leaf area per plant (cm^2) of *Z. spina-christi* and *P. juliflora* grown in different proportions in monoculture.

	Leaf Area (cm^2)			
	Z. spina-christi		***P. juliflora***	
	Level of nitrogen		**Level of nitrogen**	
Density (plant/pot)	**High***	**Low****	**High***	**Low****
1	160.78 ±40.97	161.19 ±13.81	85.65 ±7.40	40.58 ±9.85
2	103.49 ±7.56	103.99 ±9.49	48.70 ±5.09	18.63 ±3.04
3	75.14 ±8.40	67.07 ±5.57	35.37 ±4.13	9.47 ±1.01
4	57.31 ±6.68	48.56 ±5.50	28.34 ±8.09	7.73 ±1.82
6	56.63 ±5.09	32.72 ±1.57	19.13 ±2.45	5.55 ±0.73

-* 100 ppm -** 20 ppm

Table 12. t- test (upper values) and p-values (lower values) comparing least squares means for the interaction between nitrogen level and plant density on leaf area (cm^2) for *P. juliflora* and *Z. spina-christi* grow in monoculture in high (100 ppm) and low (20 ppm) levels of nitrogen. Pr > |t|.

Density and level of N	1 (HLN)	1 (LLN)	2 (HLN)	2 (LLN)	3 (HLN)	3 (LLN)	4 (HLN)	4 (LLN)	6 (HLN)	6 (LLN)
1 (HLN)		5.9865 <0.0001	4.9068 <0.0001	8.9008 <0.0001	6.6779 <0.0001	-6.6779 <0.0001	-7.6117 <0.0001	-10.3484 <0.0001	-8.8343 <0.0001	-10.639 <0.0001
1 (LLN)	-0.0195 0.9846		-1.0796 0.2889	2.9143 0.0067	0.6914 0.4946	-4.1320 0.0003	-1.6252 0.1146	-4.3619 0.0001	-2.8479 0.0079	-4.6525 <0.0001
2 (HLN)	2.7216 0.0107	2.7411 0.0102		3.9940 0.0004	1.7710 0.0867	-5.2116 <0.0001	-2.7048 0.0112	-5.4415 <0.0001	-3.9275 0.0005	-5.7321 <0.0001
2 (LLN)	2.6974 0.0114	2.7169 0.0108	-0.0242 0.9809		-2.2229 0.0339	-1.2176 0.2329	1.2892 0.2072	-1.4474 0.1581	0.0665 0.9474	-1.7381 0.0924
3 (HLN)	4.0684 0.0003	4.0878 0.0003	1.3467 0.1881	1.3709 0.1806		-3.4405 0.0017	-0.9338 0.3579	-3.6704 0.0009	-2.1564 0.0392	-3.9610 0.0004
3 (LLN)	-4.4519 0.0001	-4.4712 0.0001	-1.7303 0.0938	-1.7545 0.0896	-0.3836 0.7040		2.5068 0.0178	-0.2299 0.8198	1.2841 0.2089	-0.5205 0.6065
4 (HLN)	-4.9155 <0.0001	-4.9349 <0.0001	-2.1938 0.0361	-2.2180 0.0343	-0.8471 0.4037	-0.4635 0.6463		-2.7366 0.0103	-1.2226 0.2310	-3.0272 0.0050
4 (LLN)	-5.3310 <0.0001	-5.3505 <0.0001	-2.6094 0.0140	-2.6335 0.0135	-1.2626 0.2165	-0.8791 0.3864	-0.4156 0.6807		1.5140 0.1405	-0.2906 0.7733
6 (HLN)	-4.9478 <0.0001	-4.9672 <0.0001	-2.2261 0.0337	-2.2504 0.0319	-0.8794 0.3862	-0.4959 0.6236	-0.0324 0.9744	0.3832 0.7043		-1.8046 0.0812
6 (LLN)	-6.0840 <0.0001	-6.1035 <0.0001	-3.3624 0.0021	-3.3865 0.0020	-2.0156 0.0529	-1.6321 0.1131	-1.1685 0.2518	-0.7530 0.4573	-1.1362 0.2649	

- Shaded cells represent *P. juliflora* Non-shaded cells represent *Z. spina-christi.*

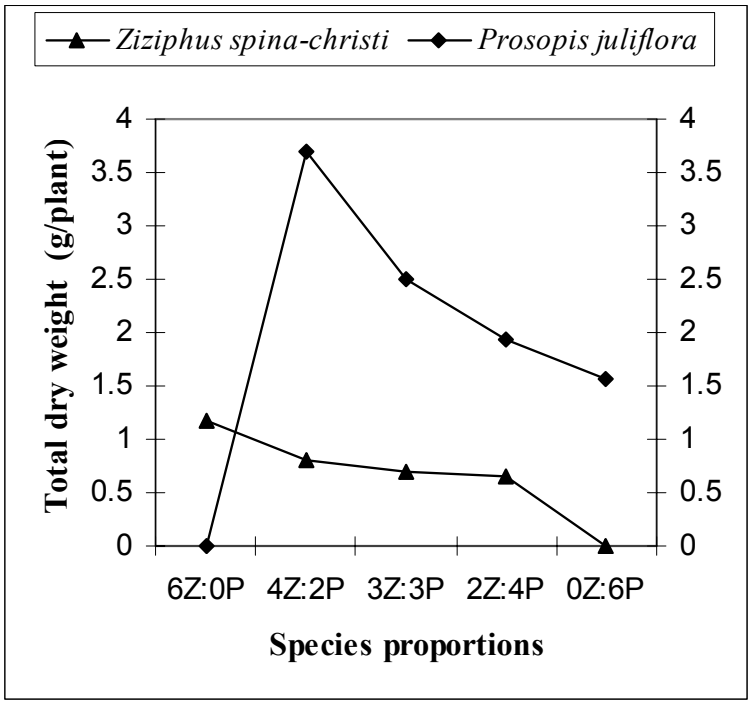

Figure 1. Replacement diagram showing dry weight for *Z. spina-christi* and *P. juliflora* in a replacement series with 100 ppm nitrogen.

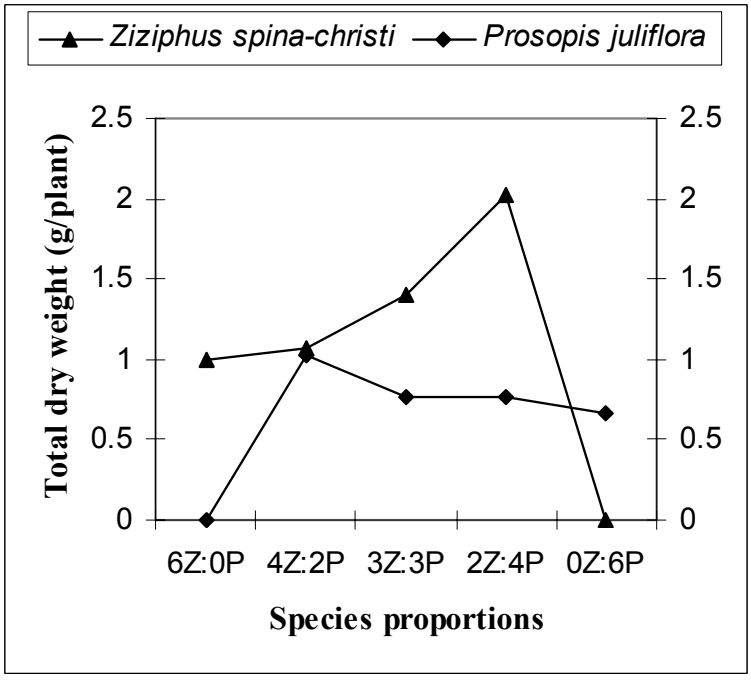

Figure 2. Replacement diagram shows dry weight for *Z. spina-christi* and *P. juliflora* in a replacement series with 20 ppm nitrogen.

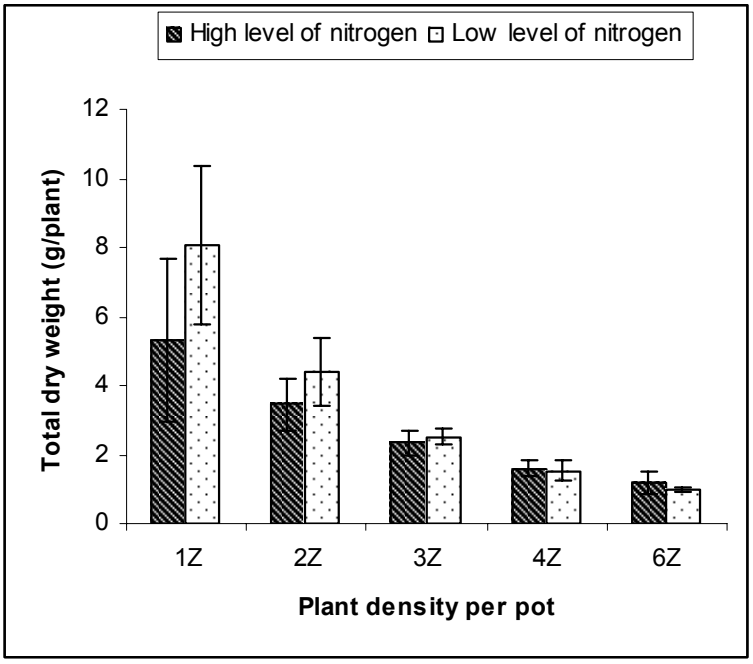

Figure 3. Total dry weight for *Z. spina-christi* grown in monoculture in different densities with high (100 ppm) and low (20 ppm) levels of nitrogen.

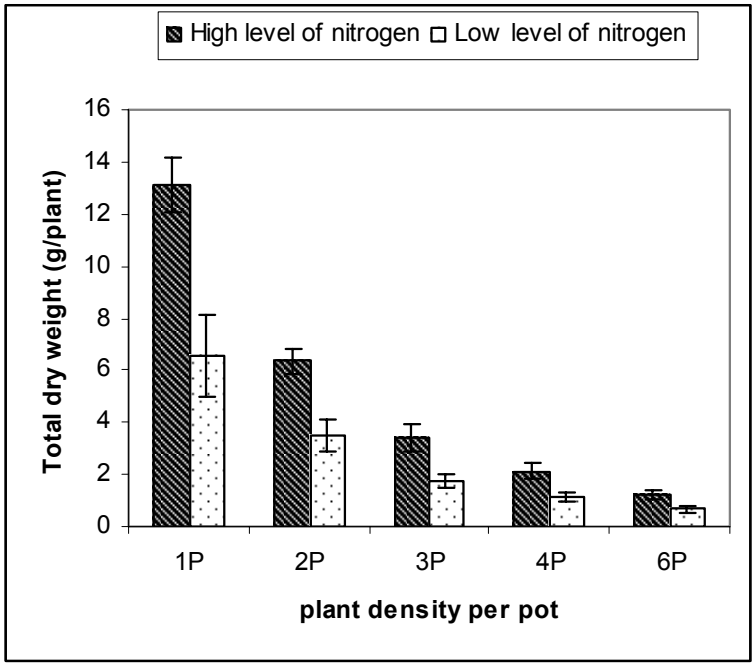

Figure 4. Total dry weight for *P. juliflora* grown in monoculture in different densities high (100 ppm) and low (20 ppm) levels of nitrogen.

Table 13. t- test (upper values) and p-values (lower values) comparing least squares means for the interaction between nitrogen level and plant density on total plant dry weight for *P. juliflora* and *Z. spina-christi* grown in monoculture in high (100 ppm) and low (20 ppm) level of nitrogen. Pr > |t|.

Density and level of N	1 (HLN)	1 (LLN)	2 (HLN)	2 (LLN)	3 (HLN)	3 (LLN)	4 (HLN)	4 (LLN)	6 (HLN)	6 (LLN)
1 (HLN)		-13.5711 <.0001	-14.003 <.0001	-19.8557 <.0001	-20.1039 <.0001	-23.5321 <.0001	-22.7153 <.0001	-24.7205 <.0001	-23.8935 <.0001	-25.739 <.0001
1 (LLN)	3.4113 0.0019		-0.4319 0.6689	-6.2846 <.0001	-6.5328 <.0001	-9.9609 <.0001	-9.1442 <.0001	-11.1494 <.0001	-10.3224 <.0001	-12.1679 <.0001
2 (HLN)	-2.2859 0.0295.	-5.6972 0.0001		-5.8527 <.0001	-6.1009 <.0001	-9.5291 <.0001	-8.7122 <.0001	-10.7175 <.0001	-9.8905 <.0001	-11.736 <.0001
2 (LLN)	-1.1308 0.2671	-4.5421 0.0001	1.1551 0.2572		-0.2482 0.8057	3.6763 0.0009	-2.8595 0.0076	-4.8648 <.0001	-4.0378 0.0003	-5.8832 <.0001
3 (HLN)	-3.6717 0.0009	-7.0830 0.0001	-1.3858 0.1760	-2.5409 0.0165		-3.4282 0.0018	-2.6113 0.0139	-4.6166 <.0001	-3.7896 0.0007	-5.6350 <.0001
3 (LLN)	-3.3564 0.0022	-6.7677 0.0001	-1.0705 0.2929	-2.2256 0.0337	0.3153 0.7547		-0.8168 0.4205	-1.1884 0.2440	-0.3614 0.7203	-2.2087 0.0351
4 (HLN)	-4.6024 0.0001	-8.0137 0.0001	-2.3165 0.0275	-3.4716 0.0016	-0.9307 0.3594	-1.2460 0.2224		-2.0052 0.0540	1.1782 0.2480	-3.0237 0.0051
4 (LLN)	-4.6678 0.0001	-8.0791 0.0001	-2.3818 0.0238	-3.5369 0.0013	-0.9960 0.3272	-1.3114 0.1997	-0.0653 0.9483		0.8270 0.4148	-1.0185 0.3166
6 (HLN)	-5.1097 0.0001	-8.5210 0.0001	-2.8238 0.0084	-3.9789 0.0004	-1.4379 0.1608	-1.7533 0.0898	-0.5073 0.6157	-0.4420 0.6617		-1.8455 0.0749
6 (LLN)	-5.3308 0.0001	-8.7421 0.0001	-3.0449 0.0048	-4.2000 0.0002	-1.6591 0.1075	-1.9744 0.0576	-0.7284 0.4720	-0.6631 0.5123	-0.2211 0.8265	

- Shaded cells represent *P. juliflora* -Non-shaded cells represent *Z. spina-christi*

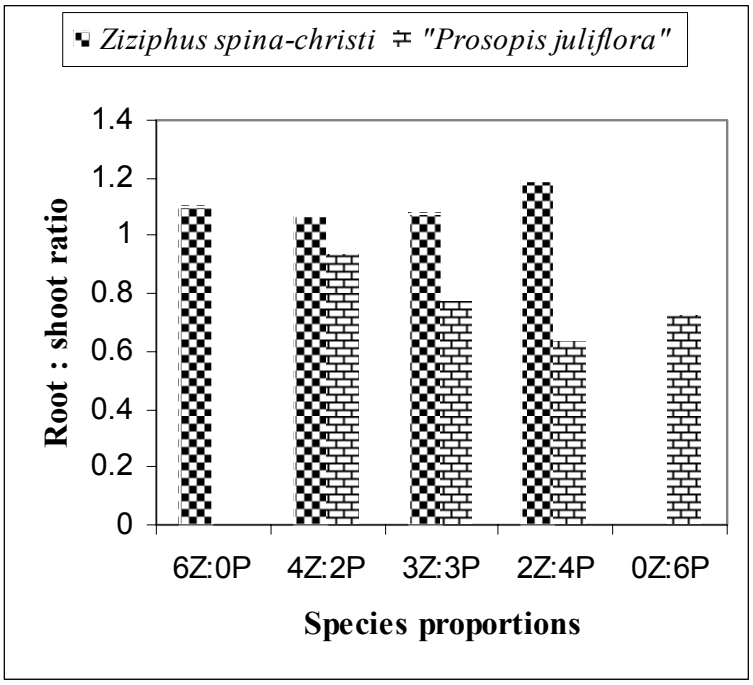

Figure 5. Diagram showing root to shoot ratio for *Ziziphus spina-christi* and *Prosopis juliflora* in mixed plantings with high levels of nitrogen (100 ppm).

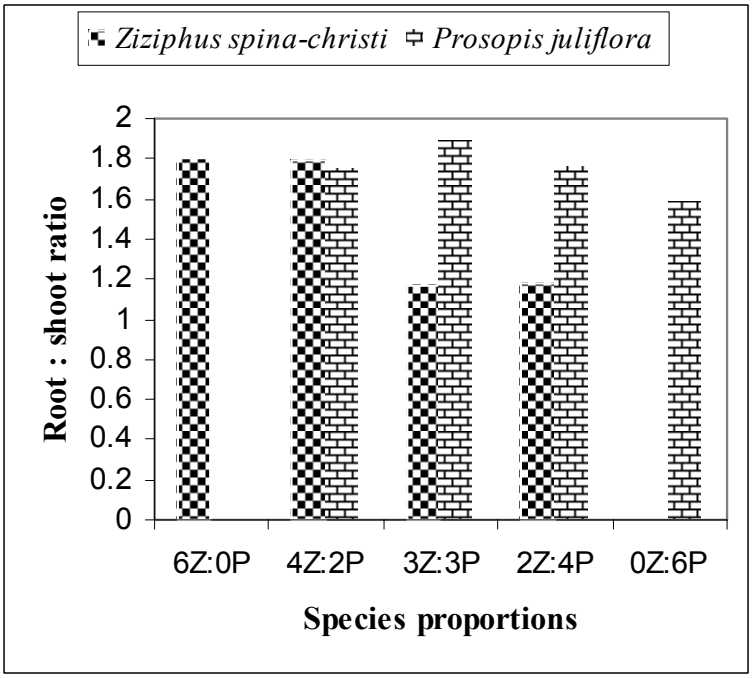

Figure 6. Diagram showing root to shoot ratio for *Ziziphus spina-christi* and *Prosopis juliflora* in mixed planting in low levels of nitrogen (20 ppm).

Table 14. t- test (upper values) and p-values (lower values) comparing least squares means for the interaction between nitrogen level and species proportion on root to shoot ratio for *P. juliflora* and *Z. spina-christi* grown in mixture in high (100 ppm) and low level (20 ppm) of nitrogen. Pr > |t|.

Z. spina: P. juliflora	4:**2** HLN	3:**3** HLN	2:**4** HLN	4:**2** LLN	3:**3** LLN	2:**4** LLN
4**:2** HLN	-	0.64 0.5331	1.34 0.1990	-4.44 0.0004	-4.95 0.0001	-4.69 0.0002
3:**3** HLN	-0.48 0.6352	-	0.69 0.4975	-3.74 0.0016	-4.23 0.0006	-3.97 0.0010
2:**4** HLN	-3.09 0.0066	-0.04 0.9878	-	-3.39 0.0035	-3.91 0.0011	-3.52 0.0026
4:**2** LLN	0.05 0.9595	-0.43 0.7620	-0.47 0.6427	-	0.55 0.5870	-0.08 0.9397
3:**3** LLN	0.08 0.9399	-0.39 0.6986	-0.44 0.6684	-0.03 0.9785	-	-0.62 0.5420
2:4 LLN	-2.52 0.0219	-3.03 0.0075	-0.52 0.6076	2.68 0.0159	2.78 0.0129	-

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*
- Numbers in bold to make the comparison for R:S mean of *n* plants of *P. juliflora* to R:S mean of *n* plants of the same species (e.g. 4:**2** HLN to 2:**4** LLN where t= -4.69 and P= <0.0002)

Table 15. Comparison of the effect of density per pot and level of nitrogen on root to shoot ratio of *Z. spina-christi* and *P. juliflora* grown in monoculture plantation under two levels of nitrogen.

	Root : Shoot Ratio			
	Ziziphus spina		***Prosopis juliflora***	
	Level of nitrogen			
Density (plant/pot)	**High***	**Low****	**High***	**Low****
1	1.18 ±0.20	1.66 ±0.13	0.64 ±0.09	1.06 ±0.16
2	1.25 ±0.12	1.43 ±0.12	0.91 ±0.16	1.26 ±0.19
3	1.33 ±0.13	1.64 ±0.15	0.67 ±0.009	1.49 ±0.10
4	1.36 ±0.14	1.77 ±0.22	0.78 ±0.04	1.51 ±0.09
6	1.10 ±0.09	1.80 ±0.14	0.72 ±0.01	1.58 ±0.14

-* 100 ppm -** 20 ppm

Table 16. t- test (upper values) and p-values (lower values) comparing least squares means for the interaction between nitrogen level and plant density on root to shoot ratio for *P. juliflora* and *Z. spina-christi* grown in monoculture in high (100 ppm) and low (20 ppm) levels of nitrogen. Pr > |t|.

Density and N level	1 (HLN)	1 (LLN)	2 (HLN)	2 (LLN)	3 (HLN)	3 (LLN)	4 (HLN)	4 (LLN)	6 (HLN)	6 (LLN)
1 (HLN)		-2.4402 0.0208	-1.5532 0.1309	-3.6064 0.0011	-0.1918 0.8492	4.9705 <0.0001	0.8328 0.4115	5.0518 <0.0001	0.4717 0.6405	5.4889 <0.0001
1 (LLN)	-1.4291 0.0213		0.8870 0.3821	-1.1662 0.2527	2.2484 0.0320	2.5303 0.0169	-1.6073 0.1184	2.6116 0.0139	-1.9684 0.0583	3.0488 0.0048
2 (HLN)	-0.5439 0.5905	1..8852 0.0691		-2.0532 0.0489	1.3614 0.1835	3.4173 0.0018	-0.7203 0.4769	3.4986 0.0015	-1.0814 0.2881	3.9358 0.0005
2 (LLN)	-1.3920 0.1742	1.0371 0.3080	-0.8480 0.4031		3.4146 0.0019	1.3641 0.1827	-2.7736 0.0094	1.4454 0.1587	-3.1347 0.0038	1.8826 0.0695
3 (HLN)	-0.9335 0.3580	1.4956 0.1452	-0.3896 0.6996	0.4584 0.6499		4.7788 <0.0001	0.6411 0.5263	4.8601 <0.0001	0.2799 0.7814	5.2972 <0.0001
3 (LLN)	2.3428 0.0260	-0.0863 0.9318	1.7988 0.0821	0.9508 0.3493	1.4093 0.1690		14.1377 0.0003	0.0813 0.9357	-4.4988 <0.0001	0.5184 0.6080
4 (HLN)	1.0671 0.2944	-1.3619 0.1834	0.5232 0.6047	-0.3248 0.7476	0.1336 0.8946	-1.2756 0.2119		4.2190 0.0002	-0.3611 0.7206	4.6561 <0.0001
4 (LLN)	2.9654 0.0059	0.5362 0.5958	2.4214 0.0217	1.5734 0.1261	2.0318 0.0511	0.6225 0.5383	1.8982 0.0673		-4.5801 <0.0001	0.4371 0.6652
6 (HLN)	-0.1104 0.9129	-2.5394 0.0165	-0.6543 0.5179	-1.5024 0.1435	-1.0438 0.3049	-2.4531 0.0202	-1.1775 0.2482	-3.0757 0.0045		5.0172 <0.0001
6 (LLN)	3.1092 0.0041	0.6800 0.5017	2.5653 0.0156	1.7172 0.0962	2.1757 0.0376	0.7663 0.4494	2.0420 0.0500	0.1438 0.8866	3.2195 0.0031	

- Shaded cells represent *P. juliflora* Non-shaded cells represent *Z. spina-christi.*

Table 17. Relative yields and standard errors per plant for *P. juliflora* and *Z. spina-christi* grown under two different N levels in different proportion per pot.

	High level of nitrogen*			**Low level of nitrogen****		
	RY		**RYT**	**RY**		**RYT**
***Z. spina-christi: P.Juliflora* proportion**	***Z. spina-christi***	***P. Juliflora***		***Z. spina-christi***	***P. Juliflora***	
4:2 (0.67:0.33)	0.63 ±0.09	1.79 ±0.21	1.144	0.959 ±0.13	0.957 ±0.09	1.082
3:3 (0.50:0.50)	0.49 ±0.09	1.38 ±0.1	1.066	0.995 ±0.24	0.90 ±0.08	1.073
2:4 (0.33:0.67)	0.38 ±0.09	1.11 ±0.12	0.993	1.19 ±0.19	1.01 ±0.07	1.198

-* 100 ppm -** 20 ppm

Table 18. t- value (upper values) and P- value (lower values) of least squares means for the interaction between level of nitrogen and proportion of species on relative yield in high (100 ppm) and low (20 ppm) levels of nitrogen. Pr > |t|.

Z. spina: P. juliflora	4:**2** HLN	3:**3** HLN	2:**4** HLN	4:**2**- LLN	3:**3** LLN	2:**4** LLN
4:**2** HLN	-	-2.3265 0.0263	-3.9147 0.0004	4.8052 <.0001	-5.1274 <.0001	-4.4979 <0.0001
3:**3** HLN	-0.8034 0.4275	-	-1.5882 0.1218	2.4787 0.0185	2.8008 0.0085	-2.1715 0.0372
2:**4** HLN	-1.4601 0.1537	-0.6567 0.5159	-	0.8905 0.3797	1.2126 0.2339	0.5832 0.5637
4:**2** LLN	-1.8653 0.0711	-2.6686 0.0117	-3.3253 0.0022	-	-0.3221 0.7494	0.3072 0.7606
3:**3** LLN	2.0776 0.0456	-2.881 0.0069	-3.5377 0.0012	0.2124 0.8331	-	0.6294 0.5334
2:4 LLN	3.2501 0.0027	4.0535 0.0003	-4.7102 <.0001	1.3849 0.1754	1.1725 0.2494	-

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*
- Numbers in bold to make the comparison for RY mean of *n* plants of *P. juliflora* to RY mean of *n* plants of the same species (e.g. 4:**2** HLN to 2:**4** LLN where t= -4.4979 and P= <0.0001)

Table 19. Aggressivety index for *P. juliflora* and *Z. spina-christi* grown in mixed plantings under two levels of nitrogen.

	Aggressivety Index*	
***Z. Spina: P. Juliflora* proportion**	**High level of nitrogen****	**Low level of nitrogen*****
4:2	-1.15	-0.0002
(0.67:0.33)	±0.25	±0.08
3:3	-0.895	0.093
(0.50:0.50)	±0.12	±0.28
2:4	-0.733	0.187
(0.33:0.67)	±0.15	±0.27

-*Aggressivty index was calculated as (Relative yield of *Z. spina* - Relative yield of *P. juliflora*).
-** 100 ppm
-***20 ppm
-The negative values indicate that RY of *Z. spina* are lower than RY of *P. juliflora*.
- Positive values indicate agressivity of *Z. spina-christi* and negative values indicate agressivity of *P. juliflora*.

Table 20. Means and standard errors for the effect of plant proportion on total chlorophyll *ab* of *Z. spina-christi* and *P. juliflora* grown in mixed plantings under two levels of nitrogen.

	Chlorophyll *ab* (nmol/ml)			
	Level of nitrogen			
***Z. spina: P. juliflora* proportions**	**High***		**Low****	
	Z	**P**	**Z**	**P**
4:2 (0.67:0.33)	14.22 ±0.77	5.35 ±1.20	6.32 ±1.16	3.95 ±1.91
3:3 (0.50:0.50)	11.11 ±1.11	4.79 ±0.93	6.05 ±1.22	3.00 ±0.38
2:4 (0.33:0.67)	8.32 ±1.92	4.59 ±0.85	5.17 ±1.24	2.92 ±1.15

-* 100 ppm -**20 ppm

Table 21. t- test (upper values) and p-values (lower values) comparing least squares means for nitrogen levels and plant density on total chlorophyll *ab* for *P. juliflora* and *Z. spina-christi* grow in monoculture in high (100 ppm) and low (20 ppm) level of nitrogen. Pr > |t|.

Z. spina: P. juliflora	4:**2** HLN	3:**3** HLN	2:**4** HLN	4:**2** LLN	3:**3** LLN	2:**4** LLN
4:**2** HLN	-	-0.63 0.5385	-0.62 0.5416	0.21 0.8389	-1.00 0.3323	-0.84 0.4137
3:**3** HLN	-1.54 0.1411	-	0.01 0.9885	-0.36 0.7260	0.35 0.7325	-0.21 0.8335
2:**4** HLN	-3.54 0.0025	-1.92 0.0712	-	-0.36 0.7250	0.37 0.7143	0.23 0.8172
4:**2** LLN	2.27 0.0363	1.25 0.2291	-0.26 0.7944	-	-0.79 0.4407	-0.67 0.5145
3:**3** LLN	-2.20 0.0418	1.22 0.2387	-0.23 0.8190	-0.03 0.9744	-	0.13 0.8951
2:4 LLN	-3.16 0.0057	-2.10 0.0507	0.38 0.7086	-0.89 0.3873	-0.84 0.4120	-

- Shaded cells represent *P. juliflora* - - Non-shaded cells represent *Z. spina-christi*

Table 22. Means and standard errors for the effect of plant density per pot and level of nitrogen on total chlorophyll *ab* for *Z. spina-christi* and *P. juliflora* grown in monocultures under two levels of nitrogen.

	Chlorophyll *ab* (nmol/ml)			
	Ziziphus spina-christi		***Prosopis juliflora***	
	Level of nitrogen		**Level of nitrogen**	
Density (plant/pot)	**High***	**Low****	**High***	**Low****
1	13.29 ±1.61	7.35 ±0.58	4.89 ±0.72	2.99 ±1.23
2	15.36 ±1.58	8.16 ±1.32	5.01 ±1.53	1.79 ±0.41
3	13.29 ±0.72	6.20 ±1.09	6.78 ±1.07	3.17 ±0.29
4	14.60 ±1.61	6.87 ±0.75	6.06 ±0.98	2.18 ±0.80
6	12.36 ±1.88	5.39 ±0.98	5.57 ±1.10	2.05 ±0.86

-* 100 ppm -**20 ppm

Table 23. t- test (upper values) and p-values (lower values) comparing least squares means for the interaction between nitrogen level and plant density on total chlorophyll *ab* for *P. juliflora* and *Z. spina-christi* grown in monocultures in high (100 ppm) and low (20 ppm) levels of nitrogen. Pr > |t|.

Density and level of N	1 (HLN)	1 (LLN)	2 (HLN)	2 (LLN)	3 (HLN)	3 (LLN)	4 (HLN)	4 (LLN)	6 (HLN)+	6 (LLN)
1 (HLN)		1.3891 0.1750	-0.0852 0.9327	2.2674 0.0307	-1.3715 0.1804	-1.2556 0.2190	-0.8463 0.4040	1.9833 0.0565	-0.4877 0.6293	2.0839 0.0458
1 (LLN)	3.2682 0.0027		-1.4744 0.1508	0.8783 0.3868	-2.7606 0.0097	0.1336 0.8946	-2.2355 0.0330	0.5942 0.5568	-1.8768 0.0703	0.6948 0.4925
2 (HLN)	-1.1410 0.2629	-4.4092 0.0001		2.3564 0.0254	1.2863 0.2082	1.3407 0.1901	-0.7612 0.4525	2.0685 0.0473	-0.4024 0.6902	2.1692 0.0381
2 (LLN)	2.8192 0.0084	-0.4489 0.6567	3.9602 0.0004		-3.6389 0.0010	1.0119 0.3197	-3.1138 0.0040	-0.2841 0.7783	-2.7551 0.0099	-0.1834 0.8557
3 (HLN)	-0.0025 0.9980	-3.2707 0.0027	1.1384 0.2639	-2.8218 0.0084		2.6271 0.0134	0.5252 0.6033	3.3548 0.0022	0.8839 0.3838	3.4555 0.0017
3 (LLN)	-3.8968 0.0005	-0.6287 0.5343	-5.0379 <0.0001	-1.0778 0.2898	-3.8994 0.0005		-2.1019 0.0441	0.7277 0.4724	-1.7432 0.0915	0.8284 0.4140
4 (HLN)	0.7249 0.4741	3.9930 0.0004	-0.4162 0.6803	3.5441 0.0013	0.7223 0.4757	4.6217 <0.0001		2.8297 0.0082	0.3587 0.7223	2.9304 0.0064
4 (LLN)	-3.5278 0.0014	-0.2596 0.7969	-4.6688 <0.0001	-0.7086 0.4841	-3.5303 0.0014	0.3690 0.7147	-4.2527 0.0002		-2.471 0.0194	0.1006 0.9205
6 (HLN)	-0.5117 0.6126	7.7564 0.0098	-1.6527 0.1088	3.3075 0.0281	-0.5142 0.6109	3.3852 0.0020	-1.2366 0.2258	3.0161 0.0062		2.5717 0.0153
6 (LLN)	-4.339 0.0001	-1.0708 0.2928	-5.4800 <0.0001	-1.5197 0.1390	-4.3415 0.0001	-0.4422 0.6615	-5.0639 <0.0001	-0.8112 0.4236	-3.8273 0.0006	

- Shaded cells represent *P. juliflora* - Non-shaded cells represent *Z. spina-christi*

Appendix A

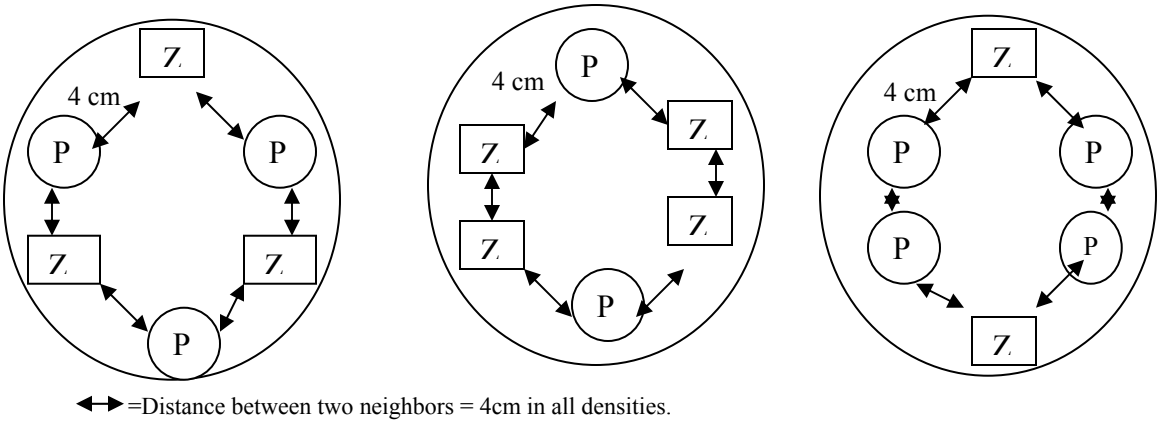

Figure 1. The order of *Z. spina -christi* and *P. juliflora* grown at mixed plantings 3:3, 4:2, and 2:4.

(4Z:2P)

(3Z:3P)

(2Z:4P)

Figure 2. Growth of *Z. spina-christi* and *P. juliflora* in high level of nitrogen (green color) and low level of nitrogen (yellow color) in mixed plantings in different proportions.

One plant

Two plants

Three plants

Four plants

Six plants

Figure 3. Growth of *Z. spina-christi* in high level of nitrogen (green color) and low level of nitrogen (yellow color) in monoculture plantings.

One plant

Two plants

Three plants

Four plants

Six plants

Figure 4. Growth of *P. juliflora* in high level of nitrogen (green color) and low level of nitrogen (yellow color) in monoculture plantings.

Synthesis and Conclusions

Regardless of the initial causes of plant invasion, non-native plants can alter plant interactions and local environmental conditions causing a change in native community composition and diversity via allelopathic and/or competitive effects. This research represents an effort to define and quantify these processes for two species, one an invasive plant (*Prosopis juliflora*) and the other native plant to Saudi Arabia (*Ziziphus spina-christi*).

The traditional view for allelopathy emphasizes direct effects of allelochemical upon the target plant. However, the separation of allelopathy from competition in the field is one of the many challenges of studying the interaction between plant species. It is logical that competition occurs through the removal of resources while allelopathy may occur even when resources are not limited. In plant communities, both allelopathy and competition are potentially affecting the structure of the community with respect to individual species. It is most likely that both strategies are important for plant species that dominate a community. However, allelopathic effects depend on the target plant as well as the allelopathic plant. In my study, I determined that an allelopathic response did occur, but not the one that was expected. Extracts from the invasive species actually stimulated growth of the native species, except at the highest concentrations. Furthermore, extracts from the native species inhibited the growth of the invasive species.

Competition for resources, such as nutrients, is part of the physiological and ecological strategy of sympatric species. However, outcomes of below-ground competition are a function of plant traits and these may change in response to resource availability. Where species are competitively superior when a resource is abundant they may become disadvantaged when the same resource is deficient. In the case of *P. juliflora* the species is superior when nitrogen is abundant which is indicated by the species gaining more biomass compared to *Z. spina-christi*. However, the deficiency of nitrogen altered the interaction to favor *Z. spina-christi* a species that seems able to gain more biomass under conditions of nitrogen deficiency.

In my greenhouse experiment, the allelopathic effect appears to be less important for the invasive plant species than competition. Under field condition the effects of allelopathic substances could be more important in facilitating the invasive species since

low concentrations of allelochemicals coupled with environmental stress from drought and high temperatures may enhance the effect of allelopathic materials. Furthermore, accumulation of *P. juliflora* litter in the dry environment of Saudi Arabia may produce a concentrating effect in which toxic levels of allelochemicals are released during the infrequent rains. It is also important to identify the stage of development during which allelopathy occurs in a plant. For example, allelopathy could be effective throughout the life of a species, or during a particular stage of development or after achieving a particular size or nutrient status.

Studies of plant competition, competitive ability for species such as *P. juliflora* can be separated into two mechanisms: effect and response. Competitive effect refers to a species' ability to suppress the growth of other species at a certain level of resource availability through depletion of resources such as nitrogen. Competitive response refers to a species' ability to tolerate resource levels that have been depleted as a result of competition. Successful invasive plants often exhibit both competitive effects and competitive responses as they interact with native vegetation. For the interaction between *P. juliflora* and *Z. spina-christi* the mechanism for success seems to be one of effect more than response.

The success of an invader not only depends on having access to resources unused by the resident species, but also on the ability of the species to acclimate to changing conditions. *P. juliflora* seemed more adaptive in biomass allocation to different tissues depending on resource availability, being able to allocate most of its biomass to roots under the low nitrogen, but in high nitrogen levels the allocation was mostly to stem. Conversely, *Z. spina-christi* maintained a fairly consistent R:S ratio at both low and high nitrogen levels. This indicates that *P. juliflora* is more adaptable to changing environmental conditions than the native species. These contrasting patterns of biomass allocation suggest that *P. juliflora* posesses the amplitude for resource partitioning, depending on the type of environmental stress.

Size limitation of pots in the greenhouse experiment could affect my conclusions because pot size can limit resources and alter the potential of the species to compete. The more extensive root development in *P. juliflora* may be advantageous in the field in obtaining the limited supply of resources. This, in turn, may change the outcome of

competition where fast growing plants are likely to be more efficient at capturing nutrients because of a large root mass. But in a constricted environment, such as in my study the large root system may become a disadvantage.

Field studies do not easily permit the separation of allelopathy from competition since both species occupy the same habitat and the two mechanisms may be working together. But field studies may be necessary since it is diffecult to duplicate many of the complexities that occur in nature in a controlled environment. My study has illucidated some of the mechanisms by which the two species interact and has eliminated some of the possible causes of invasive behavior.

From an applied perspective, if the introduced species success is due to an unusual capacity for resource competition then control is a matter of keeping it from acquiring the particular resources. I have determined that the introduced species is more aggressive in high levels of nitrogen therefore care must be taken to avoid introducing the species to habitats with high nitrogen levels to avoid the development of monospecific stands. It is unclear as to the role of nitrogen fixation on the process of invasion by *P. juliflora* and the displacement of *Z. spina-christi*. But it could potentially play a large role and is a subject that deserves further study

This study suggests that future research efforts should evaluate the interaction of the exotic plants and native species in the field. It is possible that allelopathy may play a role as well as competition under field conditions. In Saudi Arabia, the ability of *P. juliflora* to invade and dominate local plant habitats is unquestionable since the invasive species performs extremely well, displacing the native species. Field studies with different soil nutrient levels show that *Z. spina-christi* may reduce the spread of *P. juliflora* in habitats with low nitrogen. The small nutrient difference in arid and semi-arid systems may have important biological consequences in plant competition and succession. Finally, field studies on *P. juliflora*'s ability to add nitrogen to the soil under field conditions are particularly needed as well as studies to determine the dynamics of allelochemicals in leaf litter under arid conditions.