Sudan University of Science and Technology

College of Graduate Studies

Allelopathic Effect of Argel (*Solenostemma argel* L.) Additives on Seed Germination and Vigour of Two Snap Bean (*Phaseolus vulgaris* L.) Cultivars.

التأثير التضادي لإضافات الحرجل (Solenostemma argel) على انبات وقوة نمو بذور صنفين من الفاصوليا (*Phaseolus vulgaris* L.)

By

Wegdan Ibrahim Mohammed

B.Sc. (Agric.) Honours Sudan University of Science and Technology

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Supervisor: Prof. Dr. Ayoub Zeyada Elhag

Department of Horticulture

College of Agricultural Studies

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DEDICATION

This research is dedicated to souls of

Mother Father

As well as Sisters and Brothers

in particular Abd Ebagi Osman

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Abstract

Two laboratory and one field experiments were conducted at the laboratory of Seed Administration and at the Farm of the Horticulture Sector, Federal Ministry of Agriculture and Forestry, Khartoum, Sudan, respectively, to study the allelopathic effect of argel additives on snap bean seed germination and vigour. Seeds of two bean cultivars (Paulista and Star2000) were sown on filter paper in petri dishes (18cm in diameter) and on sand medium in plastic pots (18cm in diameter) at the laboratory. On filter paper they were moistened with four concentrations of argel shoot water extract (0.2, 0.4, 0.6 and 0.8% w/v of distilled water and pots they were mixed as powder with sand to have the same concentrations w/w of sand, and irrigated with tap water at 70% of sand water holding capacity, in addition to a control with distilled (on filter paper) or tap water (in pots). In the field experiment the seeds were mixed with four different amounts of argel powder (20, 40, 60 and 80% w/w of seeds), in addition to a control without powder. The experimental units were in completely randomized for laboratory and completely randomized block (in split units) designs for field experiments, with three and four replications, respectively. The allelopathic effects were evaluated as seed germination and emergence percentage, seed vigour, germination or emergence rate and uniformity and growth (seedling length, number of leaves per seedling and seedling shoot fresh and dry weight).The results showed that the cv. Star2000 was more sensitive to argel extracts under field conditions than controlled condition (lab.). The reverse was noticed with cv. Paulista. Argel shoot extract or powder of concentration of 0.2% in the lab.or argel powder at 40% in the field increased seed germination or emergence percentage, vigour and seedling growth, whereas the reverse was noticed with the higher extract or powder concentrations compared to control. It was also noticed that the percentage of fungal infected seeds decreased with increased argel

concentrations, where the lowest percentage of infected seeds was obtained by the highest argel concentration. It could be concluded that argel shoot extract or powder concentration 0.2% on filter paper or in pots (in lab), respectively, or powder at 40%.w/w of seed under field conditions had positive effect on snap bean seed germination, vigour and seedling growth, whereas, the higher ones had negative effect, irrespective of cultivar. Argel seed dressing of 40%w/w of seeds concentration could be used for improvement of field establishment of bean seedlings. Further studies are required to find which is more effective argel shoot extract or powder under field conditions.

المســـــتخلــص

التأثير التضادي لإضافات الحرجل (*Solenostemma argel*) على انبات وقوة نمو بذور صنفين من الفاصوليا (*Phaseolus vulgaris* L.)

أجريت تجربتان معمليتين وواحده حقلية بمعمل إدارة التقاوى ومزرعة إدارة القطاع البستاني، وزارة الزراعة والغابات الاتحادية، الخرطوم، السودان، على التوالي، لدراسة التأثير التضادي لإضافات الحرجل على إنبات وقوة نمو بذور الفاصوليا. زرعت بذور صنفين من الفاصوليا (باولستا واستار 2000) في ورق ترشيح في اطباق بترى (قطرها 18سم) و في بيئة رملية في اصص بلاستيكية (قطرها 18سم) في المعمل. ورطبت ب10 مل لأربعة تراكيز لمستخلص المجموع الخضري للحرجل (0,2%، 0,4% ،0,6% و0,8% وزن /حجم الماء المقطر) او خلطها كمسحوق مع الرمل (لتعطى نفس التراكيز من وزن الرمل)، وتم ريها بماء صنبور حتى 70% من السعه المائية للرمل بالإضافة لشاهد رُوي بماء مقطر او ماء صنبور(لورق الترشيح والاصص، على التوالى). في تجربة الحقل تم خلطت البذور بمسحوق الحرجل باربعة تراكيز (20%، 40%، 60% و 80% وزن/ وزن للبذور) بالإضافة لشاهد من غير مسحوق. الوحدات التجريبية كانت بتصميم عشوائي كامل بالمعمل بثلاث تكررات و تصميم قطاعات كاملة العشوائية (في وحدات منشقه) في تجربة الحقل وباربع تكرارات. قيم التأثير التضادى على نسبة إنبات او انبثاق البذور وقوة نموها (كسرعة انبات او انبثاق وتجانسهما) ونموالبادرات (كطول البادرة، عدد الاوراق للبادرة والوزن الرطب والجاف للمجموع الخضري للبادرة). أظهرت النتائج ان الصنف استار 2000 أكثر حساسية لمستخلص الحرجل تحت ظروف الحقل مقارنة بالجو المتحكم فيه (المعمل) ولوحظ العكس على الصنف بولستا . مستخلص او مسحوق الحرجل بتركيز 0,2% فى المعمل او مسحوقه بتركيز 40% وزن/ وزن البذور فى الحقل زاد نسبة

انبات او انبثاق وقوة نمو البذور ونمو البادرات ، فى حين ان التراكيز الاعلى لها تاثير سلبى. لوحظ ايضا ان نسبة البذور المصابة بالفطريات انخفضت بزيادة تراكيز الحرجل، حيث حصل على اقل نسبة بذور مصابة عند اعلى تركيز. يمكن ان يخلص ان مستخلص او مسحوق المجموع الخضرى للحرجل بتركيز 0.2% على ورق ترشيح او فى اصص فى المعمل او مسحوقه بتركيز40% وزن/وزن البذور تحت ظروف الحقل له تاثير ايجابى عل ىنسبة الانبات اوالانبثاق وقوة النمو ونمو بادرات الفاصوليا، فى حين ان التراكيز الاعلى لها تاثير سلبى، بغض النظر عن الصنف. يمكن معاملة بذور الفاصوليا ب 40% وزن/ وزن البذور مسحوق حرجل لتحسين تاسيس البادرات. المطلوب دراسات اضافية لمعرفة ايهما اكثر فاعلية مستخلص ام مسحوق الحرجل تحت ظروف الحقل.

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Chapter one

Introduction

Snap bean (*Phaseolus vulgaris* L), a herbaceous annual, belongs to the family Fabaceae. It is grown world widely for both green pods and dry seeds (Broughton *et al.* 2003). It is a good source of calcium and vitamins A and C. The world average area of bean is 70,000 hectares, with an average yield of 20.7 million tons of green pods (Anonymous, 2012).

In Sudan snap bean is one of the most important vegetable crops grown for export. The area under snap bean is 9.5 thousand fedans which is limited on the silty loamy soils of the Nile banks (in River Nile and Kartoum States) with an average yield of 28.5 thousand tons of seeds and green pods (Anonymous, 2013). Recently, many efforts has been made to extend the area under beans on the high traces (heavy clay or saline soils). However, its yield and quality are very low due to its sensitivity to water logging and salinity in such soils. Moreover, it is high susceptible to fungal diseases and so poor germination under such condition.

To overcome such problems and to improve seed germination seed dressing with some herbs (*Solenostemma argel*) and shrubs (*Calotropis procera*) extracts or powder was tested by many researchers (Abd–Alla *et al*., 2001, Aqil and Ahmed, 2003 and Shafique *et al*., 2007). Moreover, Allelopathic potential of many crop plants and weeds have been investigated against different crops (Kato-Noguchi and Tanaka, 2003) and. Anderesen and Cedergreen (2010).

Argel (*Solenostemma argel*) is one of these plants which had been tested for its allelopathic relationship with other crop plants. It is a member of the family *Asclepiadaceae*. It is an aromatic and medicinal plant, originated in tropical Africa. It occurs in the desert area of Mali, Niger, Chad and Sudan. Also it is widely distributed in Algeriga, Egypt and Saudi Arabia. It contains acylated phenolic glycosides, namely argelin and argelosid, choline, flavonoids, monoterpene, glucoside, sitosterol saponin. It has anti–inflammatory and triterpenoid,

antimicrobial and larvicidal activities (Abdel Aziz and EzzEldein, 2007). Mahmmed *et al*. (2015) found that argel or neem (*Azadirachta indica* and *Aristolochia bracteolate*) extract and powder are considered one of the herbs which might had positive effects on seed germination of some crops. Generally under such condition of water logging and salinity of high terraces and reclaimed soils bean establishment and yield will be affected due to poor germination. This study was objected to investigate the allelopathic effect of argel additives on field establishment of snap bean (seed germination and vigour).

Chapter two

Literature review

2.1. The crop in general

Snap bean (*Phaseolus vulgaris* L.) is a dicotyledonous plant, and a member of the family, *Fabaceae*. It is originated in Peru and spread to South and Central America. It was introduced to Europe by the Spanish explorers around the 16th century, and spread further throughout the world by Spanish and Portuguese traders ((WWW.STARKEAYRES.CO.ZA.2016).

It has several common names (French, snap, green, kidney, haricot and dwarf beans). It is cultivated for either dry seeds or immature green pods. (Raymond, 2010) The green pods are rich in protein, vitamins A, C, calcium and iron.

2.2. Varieties:

Over 130 varieties of snap bean are known. Varieties (of succulent and flavored pods) are used of their immature green and they are usually grown in home gardens and also in large areas for export. Pods are of various shapes from thin "fillet" types to wide "Romano" types and colours, either green, purple, red, or streaked ((WWW.STARKEAYRES.CO.ZA.2016).

2.3. Cultivars known in Sudan:

These are Paulista, Star2000, Giza 3, Venty, Star2052 and Ambassdor (Ahmed, 2014).

2.4. Environmental requirements:

The optimum maximum and minimum temperature stor growth and yield between 26 –28 °C and 14 –16°C, respectively. Temperature above 36°C or below 10°C results in poor pod setting (Sidhu, 2010).

2.4.1. Soil:

Bean can be grown on a wide range of soils types varying from sand to heavy clay but well drained sandy loams or red loams or slit loam with pH of 6.0 to 7.0 are optimum (Sidhu, 2010).

2.5. Cultural practices:

2.5.1. Seed bed preparation:

The field should be deep ripped to around 0.5m depending on type of soil and presence of sub-soil layers. Then it should be harrowed to ensure a fine tilth with no excessive clods. This is very important to have a deep and fine seed bed. ((WWW.STARKEAYRES.CO.ZA.2016).

2.5.2. Sowing date and method:

Beans are mostly grown in Sudan in autumn and winter seasons (November to January). Generally It is possible to produce beans the year round in frost-free areas ((WWW.STARKEAYRES.CO.ZA.2016).

Bean seed are usually sown 2 -3 cm deep, deep enough to give good coverage and sufficient moisture to promote fast germination and growth. An spacing of 60x20cm is recommended for better growth and to minimize rapid spread of foliar diseases (Sidhu, 2010).

2.5.3. Irrigation:

In winter, generally it is irrigated every 10 -12 days according to soil type, canopy and growth stage. As with the application of fertilizer the water requirements of the bean plant are crucial to achieve maximum yields. It is sensitive to water logging, particularly at seed emergence. Pre-irrigation is preferred to direct post irrigation to avoid water logging, especially in heavy clay and saline soils. The greatest need for water is during the flowering and pod set stages. Depending on the prevailing climatic conditions during growth the water requirement of a bean crop can vary considerably, however, for planning purposes 650 - 750 mm of water may be need. ((WWW.STARKEAYRES.CO.ZA.2016).

2.6. Nutrition requirement:

2.6.1. Nitrogen

Nitrogen fertilization should be based on the results of a proper soil analysis. A leaf analysis can be done on plants, where nitrogen composition of less than 2.5% would indicate a deficiency. Under normal conditions, a total nitrogen application of 100-120 kg N /ha applied in various splits is seen as the norm. 60% of the total Nitrogen can be applied prior to planting and the remainder needs to be applied by week 4 after planting (WWW.STARKEAYRES.CO.ZA.2016).

2.6.2. Phosphorus:

Usually all the Phosphorus is applied at planting. Under normal conditions a total phosphorous application of 30-65 kg/ha is adequate. Where the phosphorus status of the soil has been built up over several years, as little as 10kg P/ha should be adequate. Soil analysis should be the basis of this decision (WWW.STARKEAYRES.CO.ZA.2016).

2.6.3. Potassium:

Under normal conditions a potassium application of 50-95 kg/ha is adequate. (WWW.STARKEAYRES.CO.ZA.2016)

2.6.4. Calcium:

Calcium analysis should be above 200mg/kg with a total percentage of 65% in the total cat ion exchange capacity (CEC). Sufficient calcium will be available for a green bean crop when adhering to a recognized liming protocol. In case of deficiency, an application of Ca $(NO_3)_2$ equivalent of 35-55 kg Ca/ha would be sufficient (WWW.STARKEAYRES.CO.ZA.2016).

2.7. Weed control:

Weed control on any farm must be addressed with a holistic approach and begins with correct land preparation but encompasses the correct use of pre- and post-

emergence herbicides, manual hoeing and crop rotation. Weeds need to be adequately controlled because they are efficient competitors with the crop for nutrients, moisture and sunlight. Some of them might be hosts of pests and diseases, or they might provide shelter for insect pests. It is very important that weeds be controlled in the early stages of crop development, because early competition can more seriously affect plant growth and result in the low yields (WWW.STARKEAYRES.CO.ZA.2016).

2.8. Harvest:

Green beans can be harvested manually or mechanically. The exact timing of the harvest depends greatly on the method of harvesting. Green beans for the fresh market are generally harvested by hand. Only pods that are physically ready are picked, meaning that a field may be picked as many as four times or more per season.. Picking usually start around 50 days after emergence. Depending on variety, picking can last from 10-25 days. (WWW.STARKEAYRES.CO.ZA.2016)

2.9. Post-harvest:

Beans should be harvested when the pods are bright green, fleshy and seeds are small and green. After that period, seed development reduces quality and the pod becomes pithy and tough and loses green color. Beans should be well formed and straight, bright in color with a fresh appearance, and tender but firm. They should easily snap when bent. Leaves, stems, broken beans, blossom remains, and insect damage should not be present. Decreased quality during post-harvest handling is usually often associated with water loss, chilling injury and decay. Rapid transport and cooling of fresh market crops is important to maintain quality. Hydro cooling and pressure cooling techniques are preferred for immediate removal of field heat. Night and early morning harvesting is preferred (WWW.STARKEAYRES.CO.ZA.2016)

2.10. Storage:

Green beans have a short shelf-life and are not well suited for storage. At 5-7.5°C a shelf-life of 8-12 days is expected. Very good quality can be maintained for a few

days at temperatures below 5°C but chilling injury will be induced. Some chilling may occur at the recommended storage temperature of 5°C after 7-8 days. (WWW.STARKEAYRES.CO.ZA.2016).

2.11. Pests:

The most important pests are cut worms, leaf hoppers, coleoptera, aphids, white fly And jassids

2.12. Diseases:

The most important diseases (Hagedorn and Inglis, 1986) are:-

2.12.1. Fungal diseases:

These are powdery mildew, blight, alternaria leaf spot and anthracnose,

2.12.2. Bacterial diseases:

These are bacterial brown spot, bacterial wilt and common blight.

2.12.3. Virus diseases:

These are common and yellow mosaic viruses.

2.2. Allelopathic effects of plant extracts:

2.2.1. On seed germination and vigour:

Many researchers (Wilson *et al*., 1997 and Abd –Alla *et al*., 2001). Reported that some plant extracts have antifungal activity against fungi and can substitute agrochemicals which may have undesirable biological effects on human beings and environments. Moreover, some may have growth regulating activities such as those containing jasomates, sulicylates and polyamins (Basra, 2000). Hasan *et al*. (2005) studied the effects of plant extracts on seed- borne fungi of wheat which effected seed germination, seedling health and vigour index they used ten plant extracts (*Zingber officinde, Allium sativum, Allium cepa, Adhatoda vesica, Lawsonia alba, Achyranthes aspera, Cuscuta reflexa, Vinca rosea, Nigella sativa*) *in vitro* against seed borne fungi. All plants extracts reduced the incidence of seed borne fungi, increased seed germination and vigour index. Bateman and Kwasna

(1999) and Khanzada *et al.* (2002) indicated that seed borne pathogens may cause seed abortion or seed rot, elimination of germination capacity and finally seedling damage. Also Silva *et al.* (2001) and Masum *et al*.(2009) reported that some herbaceous and medicinal plants have some antifungal properties against early seeds borne mycoflora

Özer (2005) reported that some pathogens are carried on seed or within seeds and could reduce seed germination and seedling emergence. Shafique *et al*. (2007) studied the effect of aqueous leaf extracts of 8 allelopathic tree species,(*Acacia nilotica, Alstonia scholaris, Azadirachta indica* L. , *Eucalyptus citriodora* ,*Ficus bengalensis* L., *Mangifera indica* L., *Melia azedarach* L. *and Syzygium cumini*L.) on wheat seed. They found no pronounced difference between the effectiveness of 10 and 20 minutes treatments. Generally the aqueous treatment for 10 minutes enhanced seed germination as compared to control.

Perelló *et al*., (2013) found that garlic juice improved poor germination of wheat seed caused by natural mycoflora of grain in addition to its growth promoting activities on seedling vigour. Moreover, Sahoo *et al.*(2015) stated that *Citrus reticulata* Blanco leaf extract had allelopathic potential which reduced the germination of chili, soybean and maize. Nevertheless, Seyyedi *et al*., (2013) found that sunflower and caster bean shoot aqueous extract allelochemicals showed substantial potential to the inhibition of germination percentage, germination rate, emergence rate and seedling length of dodder**.**

2.2.2. On plant growth:

Many researchers (Qasem and Foy, 2001 and Kadioglu *et al.*, 2005) reported the presence of allelopathic relationship among crops, weeds and soil micro organisms which they have defined as a mechanism that weeds and soil microorganisms often utilize to affect germination dynamics and growth of field crops. Kupidłowska *et al.* (2006) reported that allelochemicals produced by allelopathic plants showed directly negative influences on seed germination and

plant growth of other plants even in low concentrations. Andresen and Cedergreen (2010) concluded that tea seed extract showed pronounced and direct physiological effects on plants, which can both increase and decrease growth and yield depending on extract concentration. Ayeni and Kayode (2013) tested the allopathic effects of sorghum and maize residues on growth of E*uphorbia heterophylla* . They found that the extracts of both crops showed considerable inhibitions to all growth parameters of the extract treated seeds. Chukwuka *et al*.(2014) found that higher extract concentrations of maxican sunflower (*Tithmonia diversifolia*) and bitter leaf (*Vernonia amygdalina*) significantly inhibited radical and plumule growth of maize seedlings, whereas the lower concentration influence radical growth. The plant growth and yield were not significantly affected. Gulzar and Siddiqui (2015) reported that higher concentrations of calotropis shoot extract (60% and 80%) significantly reduced cauliflower seedling growth (radicle and plumule length, dry matter accumulation and relative water content) compared to control. The retardatory effect increased with the increase in the extract concentration, especially leaf followed by fruit and flower extracts.

Chapter three

Materials and methods

This study was conducted in three experiments to assess the allelopathic effects of argel shoots extract and powder on seed quality of two snap bean (P*haseolus valgaris*L.) cultivars ”Paulista and Star 2000”.

3.1. Location of experiments:

Two of the three experiments were conducted at the Seed Laboratory of the Seed Administration and the third one at the Experimental Farm of the Horticultural Sector Administration, of The Federal Ministry of Agriculture and Forestry, Khartoum, Sudan.

3.2. Materials

For the laboratory experiments petri dishes of 19cm in diameter (D), watmann filter paper (18cm in D), plastic pots of (20cm in D) and sand were used. The farm is clay soil with pH of 7.5. Argel shoots extract for petri dishes and argel shoot powder for the pot and field experiments were used, respectively. Seeds of the snap bean cultivars (Paulista and Star2000) were obtained from Shambat Research Station, Agricultural Research Corporation, Federal Ministry of Agriculture and Forestry, Sudan.

3.3. Methods

Preparation of Argel concentrations:

Argel dried shoots were finely milled using an electric mill. The powder is thoroughly mixed and part of it was used for the extract preparation. Each 2.0, 4.0, 6.0 and 8.0g of powder were dissolved in 500 ml of distilled water, thoroughly stirred and placed for 24hours, at room temperature. Then it was thoroughly stirred, sieved, filtered with Watmann filter paper and completed to one liter with distilled water to give extracts concentrations of 0.2, 0.4, 0.6 and 0.8% w/v of distilled water.

3.4. Laboratory experiments

3.4.1. Petri dishes experiment:

The seeds of the two cultivars were placed on top of filter paper in petri dishes (of 15 seeds each and in 3 replications). The filter paper was moistened with 10 ml of the extracts concentrations 0.2, 0.4, 0.6 and 0.8% w/v of distilled water, in addition to control with distilled water. They were covered with another filter paper and the dishes were covered and distributed randomly on the bench in the germination room at temperature (25°C). The filter paper was moistened with 5ml of the extract or distilled water for the control when required. The germinated seedlings were counted daily till the end of the germination period (21 days).

3.4.2. Pots experiment.

Different amounts (2, 4, 6 and 8g) of argel shoots powder were thoroughly mixed with sandy soil (each with 1kg sand) to have concentrations of 0.2, 0.4, 0.6 and 0.8% w/w of sand, in addition, to control without argel powder. Then it was packed in plastic pots at 1kg/pot. The seeds of both cultivars were sown in the plastic pots at 15seeds/pot. The pots were distributed at random on the bench in the germination room at temperature of (25°C) and in 3 replications. They were irrigated with equal amounts of water (70% of the sand water holding capacity) every second day and the emerged seedlings were counted every day till the end of the experiment (21days).

3.5. Fields experiment.

After land preparation (ploughing, harrowing, leveling and ridging) the field was divided into 2x 2.8m plots each with two 70cm ridges. The seeds of both cultivars were thoroughly mixed with argel shoot powder to have concentrations 20, 40, 60 and 80% w/w of seed, in addition, to a control without powder. The seeds were sown on the north side of each ridge at 15 cm within row spacing (2seeds/hole) and in 4 replications. They were irrigated immediately and every week. The emerged

seedlings were counted every day till the end of germination period (21days from sowing).

3.6. Data collected.Germination (G %) or emergence (E %) percentage

It was calculated as follows (ISTA, 2013):-

3.6.1. Germination percentage

$$x = \frac{ta}{a} * 100$$

x: G % or E %.

ta: total number of germinated or emerged seedlings

a: total number of seeds.

3.6.2. Vigour

It was evaluated as germination or emergence rate and uniformity and they were calculated as follows (Nichols and Heydecker, 1968):-

3.6.2.1. Germination rate (GR) or emergence rate(ER) (days)

$$x = \frac{a_1 n_1 + a_2 n_2 + \ldots\ldots + a_m x_m}{ta}$$

x: GR or ER.

a: number of germinated or emerged seedlings per day.

n: number of days from sowing.

ta: total number of germinated or emerged seedlings.

3.5.2.2. Germination uniformity (GU) or emergence uniformity (EU) (seeds/day)

$$x = \frac{ta}{d}$$

x: GU or EU

ta: total number of germinated or emerged seedlings

d: number of days

3.7. Seedlings growth

It was evaluated as length and fresh and dry weight of seedlings

3.7. 1. Seedling length (cm).

It was measured from the shoot base to tip of the last leaf .randomly selected seedlings and the average length was calculated

3.7. 2. Seedling fresh weight (g)

Randomly selected seedlings were weighted and the average seedling fresh weight was calculated.

3.7.3. Seedling dry weight (g)

The same seedlings were dried in an oven at 85°C for 24 hours and their dry weight was recorded. The average dry weight was calculated

3.8. Experimental design and data analysis:

The experimental units for the two laboratory experiments were in completely randomized design with three replications while those of the field experiment were in randomized complete block (in split plots) design in four replications, with the cultivars as main and argel concentrations as sub plots, respectively. The collected data were analyzed using MSTAT computer program, (version 3).The means were separated by Duncan's Multiple Range Test. (DMRT) at $p \leq 0.05$(Steel *et al*.,1997).

Chapter four

Result

4.1. Lab experiments:

4.1.1. Petri dishes experiment:

4.1.1.1. Allelopathic effect of argel shoot extract on seed germination% and vigour (germination rate and uniformity).

The cv. Star 2000 showed high germination percentage and vigour than cv. Paulista under controlled conditions (Table 1). Addition of argel up to 0.2% concentration increased germination % and vigour (germination rate and uniformity) of bean seeds. Higher concentrations had negative effect on germination% and vigour of both cultivars. .However, the cv. Paulista showed the highest negative response due to argel concentration above 0.2% than cv. Star 2000.

4.1.2. Pots experiment:

4.1.2.1. Allelopathice effect of argel shoots powder on seedling emergence% and vigour (emergence rate and uniformity).

As in Table 2, addition of argel up to 0.2% improved emergence% and rate, whereas emergence uniformity was negatively affected. Higher extract concentrations above 0.2% reduced both seed emergence and vigour. No significant differences were noticed between the two cultivars. The cv. Paulista, however, showed better emergence and vigour in sand medium than cv. Star2000.

4.1.2.2. Effect of argel on seedling growth (seedling length and fresh and dry weight).

In sand medium (Table3), the cv. Paulista gave the tallest seedlings and the highest seedling fresh and dry weight than cv. Star2000. Addition of argel powder up to 0.2% increased both seedling length and fresh and dry weight, irrespective of cultivar. No significant differences in seedling growth were noticed due to cultivar

Table 1.The allelopathic effect of argel shoot extract on seed germination % and vigour (germination rate and uniformity) of two bean cultivars

Argel concentration % w/v of distilled water	Germination percentage (%)			Germination rate (day)			Germination uniformity (seeds/day)		
	Paulista	Star2000	Mean	Paulista	Star2000	Mean	Paulista	Star2000	Mean
0.0 control	93.2^{a}	97.8^{a}	95.5^{A}	1.6^{ab}	1.3^{b}	1.4^{A}	2.8^{b}	7.5^{a}	5.2^{A}
o.2	97.8^{a}	97.8^{a}	97.8^{A}	1.5^{ab}	1.8^{ab}	1.7^{A}	2.7^{b}	3.5^{b}	3.1^{B}
0.4	91.1^{a}	100.0^{a}	95.6^{A}	1.7^{ab}	1.3^{b}	1.5^{A}	2.4^{b}	6.3^{a}	4.4^{A}
0.6	88.9^{a}	95.0a	92.2^{A}	2.0^{a}	1.5^{ab}	1.8^{A}	2.7^{b}	3.1^{b}	2.9^{B}
0.8	93.2^{a}	100.0^{a}	96.6^{A}	1.5^{ab}	1.3^{b}	1.4^{A}	2.6^{b}	6.7^{a}	4.7^{A}
Mean	$92.8^{(a)}$	$98.2^{(a)}$	-	$1.6^{(a)}$	$1.4^{(a)}$	-	$2.6^{(b)}$	$5.4^{(a)}$	-
CV			6.4			21.4			17.8

Means having the same alphabetical letters within the same column or row were not significant using DMRT at $P \leq 0.05$

Table 2 .The allelopathic effect of argel shoot powder on seed emergence % and vigour (emergence rate and uniformity) of two bean cultivars.

Argel powder % w/w of soil	Emergence %			Emergence rate (day)			Emergence uniformity(seeds/day)		
	Paulista	Star2000	Mean	Paulista	Star2000	Mean	Paulista	Star2000	Mean
0.0 control	93.3^{a}	88.0^{a}	90.7^{A}	2.6^{e}	3.8^{abc}	3.2^{BC}	5.4^{a}	4.4^{ab}	4.9^{A}
0.2%	98.7^{a}	93.0^{a}	96.0^{A}	2.6^{e}	3.3^{cd}	2.9^{C}	4.7^{ab}	4.4^{ab}	4.5^{A}
0.4%	96.0^{a}	86.7^{a}	91.3^{A}	3.2^{d}	3.8^{abc}	3.6^{AB}	5.4^{a}	4.3^{abc}	4.9^{A}
0.6%	94.7^{a}	90.7^{a}	92.7^{A}	3.1^{de}	4.0^{ab}	3.6^{AB}	4.8^{ab}	4.9^{ab}	4.9^{A}
0.8%	90.7^{a}	97.3^{a}	94.0^{A}	3.4^{bcd}	4.1^{a}	3.8^{A}	4.5^{ab}	6.2^{a}	5.4^{A}
Mean	$94.7^{(a)}$	$91.2^{(a)}$	-	$3.0^{(a)}$	$3.8^{(a)}$	-	$4.9^{(a)}$	$4.8^{(a)}$	-
CV	6.7			8.9			11.6		

Means having the same alphabetical letters within the same column or row were not significant using DMRT at $P \leq 0.05$

Table3. The allelopathic effect of argel shoot powder on seedling growth (seedling length and shoot fresh and dry weight) of two bean cultivars.

Argel powder %w/wof soil	Seedling length(cm)			Seedling fresh weight(g)			Seedling dry weight(g)		
	Paulista	Star2000	Mean	Paulista	Star2000	Mean	Paulista	Star2000	Mean
0.0 control	30.9a^{b}	25.9^{cd}	28.4^{A}	53.5^{b}	33.3^{d}	43.4^{B}	3.3^{a}	2.0^{d}	2.7^{A}
0.2	33.1^{a}	25.1^{d}	29.1^{A}	64.5^{a}	34.0^{cd}	49.7^{A}	3.3^{a}	2.2^{cd}	2.8^{A}
0.4	31.2^{ab}	27.8^{bcd}	29.5^{A}	51.3^{b}	40.2^{c}	45.8^{A}	3.0^{abc}	2.3^{bcd}	2.7^{A}
0.6	29.8^{abc}	27.3^{bcd}	28.6^{A}	52.5^{b}	38.0^{cd}	45.3^{A}	3.2^{ab}	2.3^{bcd}	2.8^{A}
0.8	29.2^{bcd}	27.8^{bcd}	28.5^{A}	54.0^{b}	38.5^{cd}	46.3^{A}	3.2^{ab}	2.5^{abcd}	2.8^{A}
Mean	30.8$^{(a)}$	26.8$^{(b)}$	-	55.2$^{(a)}$	36.9$^{(b)}$	-	3.2$^{(a)}$	2.3$^{(b)}$	-
CV	7.4			6.4			17.9		

Means having the same alphabetical letters within the same column or row were not significant using DMRT at $P \leq 0.05$.

and argel interaction, however, the cv. Star2000 responded positively (seedling length and seedling fresh and dry weight) to argel up to 0.4% concentration.

4.1.2: Field experiment

4.1.2.1. Effect of argel shoots powder on seedling emergence% and vigour (emergence rate and emergence uniformity).

The results (Table4) showed that addition of argel powder up to 40%w/w of seeds showed significant positive effect on both seedling emergence percentage and uniformity, whereas the emergence rate was only positively improved at the lowest argel concentration (20%).irrespective of cultivar. The cv. Paulista showed the highest values of the three attributes than cv. Star 2000 under field conditions. However, the highest seedling emergence percentage and vigour of both cultivars were obtained at 60% argel powder concentration.

4.1.2.2. Effect of argel shoots powder on seedling growth (seedling length and number of leaves).

As in Table5, no significant differences in seedling length were noticed between the two cultivars. The cv. Paulista, however, showed significant higher number of leaves per seedling than the cv. Star 2000. Addition of argel powder showed no significant effect on seedling growth. However the tallest seedlings and the highest number of leaves per seedling were obtained at 80% argel powder concentration by cultivars Star2000 and Paulista, respectively.

Generally, in all experiments it was noticed (no data is shown) that the number of fungal infected seeds of both cultivars were reduced by increasing of argel up tom the highest concentration (80%)

Table 4.The allelopathic effect of argel shoot powder on seedling emergence % and vigour (emergence rate and uniformity) of two bean cultivars

Argel powder g/g of seeds	Emergence %			Emergence rate (day)			Emergence uniformity(seeds/day)		
	Paulista	Star2000	Mean	Paulista	Star2000	Mean	Paulista	Star2000	Mean
0.0 control	53.9^{bcd}	48.7^{bcd}	51.3^{B}	5.0^{bcd}	6.2^{ab}	5.6^{A}	1.9^{abcd}	1.3^{cd}	1.6^{B}
0.2	57.9^{bc}	45.5^{d}	51.7^{B}	3.7^{e}	4.1^{de}	3.9^{B}	2.8^{a}	1.0^{d}	$1.9A^{B}$
0.4	58.3^{bc}	51.7^{bcd}	64.5^{A}	4.9^{cd}	6.9^{a}	5.9^{A}	2.8^{a}	2.1^{abc}	$2.5A^{B}$
0.6	72.7^{a}	56.2^{bcd}	53.4^{B}	$5.8^{a\,bc}$	5.8^{abc}	5.8^{A}	2.9^{a}	2.3^{ab}	2.6^{A}
0.8	59.1^{b}	47.7^{cd}	51.4^{B}	5.1^{bcd}	6.8^{a}	5.9^{A}	2.1^{abc}	1.8^{bcd}	1.9^{AB}
Mean	$60.4^{(a)}$	$49.9^{(b)}$	-	$4.9^{(b)}$	$5.9^{(a)}$	-	$2.5^{(a)}$	$1.7^{(a)}$	-
CV			12.0			13.2			27.9

Means having the same alphabetical letters within the same column or row were not significant using DMRT at $P \leq 0.05$.

Table 5.The allelopathic effect of argel shoot powder on seedling growth (seedling length and leaf number) of two bean cultivars.

Argel powder g/g of seeds	Seedling length (cm)			Number of leaves/seedlings		
	Paulista	Star2000	Mean	Paulista	Star2000	Mean
0.0 control	9.8^{b}	10.6b	10.2^{A}	5.8^{cde}	4.9^{def}	5.3^{A}
0.2	10.7^{b}	8.8^{b}	9.7^{A}	6.0^{cd}	4.8^{def}	5.3^{A}
0.4	10.5^{b}	10.1^{b}	10.0^{A}	7.5^{ab}	3.7^{f}	5.6^{A}
0.6	11.5^{b}	10.4^{b}	10.0^{A}	6.3^{bc}	4.0^{f}	5.2^{A}
0.8	8.8^{b}	15.9^{a}	12.4^{A}	7.9^{a}	4.6^{ef}	5.2^{A}
Mean	$10.3^{(a)}$	$11.1^{(a)}$	-	$6.7^{(a)}$	$4.4^{(b)}$	-
CV	18.9			14.8		

Means having the same alphabetical letters within the same column or row were not significant using DMRT at $P \leq 0.05$.

Chapter Five

Discussion

Allelopathic effect of argel on seed germination vigour and seedling growth

The cv. Star 2000 showed high germination percentage than cv. Paulista under controlled conditions, whereas the reverse was observed under field conditions Addition of argel increased germination or emergence %, and vigour (germination or emergence rate and uniformity) of bean seeds up to 0.2% argel extract (w/v of distilled water) or powder(w/w of soil) in the laboratory (controlled conditions). Higher concentration had negative effect on germination% and vigour of both cultivars. However, the cv. Star 2000 showed the highest negative effect due to argel concentration above 0.2% than cv. Paulista. Moreover, under field conditions similar positive response (improved emergence rate and vigour) to argel up to 60%w/w of seeds, was shown by both cultivars. It was also noticed that addition of argel reduced the percentage of fungal infected seedlings. Similar results were obtained by Kadioglu and Asav (2005). They reported a stimulation effect of some weed extracts (up to 95%concentration) on seed germination and vigour of chick pea seeds and an inhibitory effect of extract of some other weeds on germination and vigour of tomato, common bean, onion, barley and wheat. Moreover, Velu *et al*., (1996) and Sahoo. (2013) reported the allelopathic effects of a number of trees species such as mango and acacias. Sahoo *et al*. (2015) tested the effect of *Citrus reticulata* Blanco shoot extract (leaf, flower and fruit) on seed germination using petri dishes. They found that higher extract concentrations (above 55%) had inhibitory effects on seed germination and vigour of some crops (chilli, soybean, okra and maize).The positive allelopathic effects of low extract concentrations might be attributed to the hormone –like properties of allelo-chemicals of plants extracts such as jasmonates salicylates and polyamines amines (Basra, 2000 and Sahoo *et al*., 2015), or to their inhibitory effect on seed- borne

fungi which might affect seed germination and vigour (Hasan *et al*. , 2005 and Shafique *et al*.,2007).

Addition of argel up to 0.2% w/w of soil or 40%w/w of seeds in pots or in the field, respectively, increased seedling growth (seedling length and fresh and dry weight, irrespective of cultivar. The number of leaves per plant was not affected by argel addition. The seedling growth of cv. Star 2000 was negatively affected even by the lowest argel amount. Andresen and Cedergreen (2010) concluded that tea seed extract showed pronounced and direct physiological effects on plants, which can both increase and decrease growth and yield depending on extract concentration. Ayeni and Kayoda (2013) found that the extracts of sunflower and sorghum showed considerable inhibitions to all growth parameters of the extract treated seeds. Gulzar and Siddiqui (2015) reported that higher concentrations of *calotropis* shoot extract (60% and 80%) significantly reduced cauliflower seedling growth. Moreover, Sahoo *et al*. (2015) concluded that citrus shoot extracts had allelopathic potential which suppressed growth and development of the tested crops (chilli, okra and soybean).

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Appendices

Petri dishes experiment:

Appendix .1 analysis of variance (Anova)on germination rate.

Source of variation	df	SS	Ms	f	Prb
Replication	2	0.21	0.10	0.97	
paulista	1	0.49	0.49	4.50	0.0480
Star2000	4	0.57	0.14	1.28	0.3131
interaction	4	0.51	0.13	1.15	0.3653
Error	18	1.99	0.11		
Total	29	3.77			

Appendix. 2 analysis of variance(Anova)on germination uniformity.

Source of variation	df	SS	Ms	f	Prb
Replication	2	0.02	0.01	0.01	
paulista	1	57.46	57.46	111.75	0.0000
Star2000	4	23.49	5.87	11.42	0.0001
interaction	4	23.12	5.78	11.24	0.0001
Error	18	9.26	9.26		
Total	29	113.36			

Appendix 3 analysis of variance(Anova) Argel on germination %

Source of variation	df	SS	Ms	f	prb
Replication	2	48.51	24.25	0.66	
paulista	1	217.13	217.13	5.89	0.0259
Star2000	4	103.22	25.80	0.70	
interaction	4	68.37	17.09	0.46	
Error	18	662.65	36.81		
Total	29				

Pots experiment:

Appendix.4. Analysis of Variance(Anova) on emergence rate

Source of variation	df	SS	Ms	f	prb
Replication	2	0.81	0.40	4.32	0.02
Variety1	1	4.93	4.92	52.49	0.0000
Variety2	4	2.47	0.61	6.56	0.0019
interaction	4	0.23	0.05	0.62	
Error	18	1.69	0.09		
Total	29	10.13			

Appendix .5 Analysis of Variance(Anova)on emergence uniformity.

Source of variation	df	SS	Ms	f	prb
Replication	2	1.49	0.74	2.28	0.13
Variety1	1	0.10	0.10	0.32	
Variety2	4	2.00	0.50	1.53	
interaction	4	7.41	1.85	5.66	0.23
Error	18	5.89	0.33		0.oo
Total	29	16.90			

Appendix .6analysis of variance(Anova)on emergence %.

Source of variation	df	SS	Ms	f	prb
Replication	2	81.07	40.53	1.03	0.3771
Variety1	1	90.13	90.13	2.29	0.1475
Variety2	4	109.87	27.47	0.69	
interaction	4	216.53	54.13	1.38	0.2817
Error	18	708.27	39.35		
Total	29	1205.27			

Appendix .7 Analysis of Variance(Anova) on seedling length.

Source of variation	df	SS	Ms	f	prb
Replication	2	4.16	2.08	0.46	
paulista	1	123.14	123.14	27.09	0.0001
Star2000	4	5.85	1.46	.32	
interaction	4	39.71	9.93	2.18	0.1119
Error	18	81.79	4.54		
Total	29	254.67			

Appendix .8 analysis of variance(Anova) on dry weight.

Source of variation	df	SS	Ms	f	prb
Replication	2	70.38	35.19	4.08	0.0346
paulista	1	2488.85	2488.85	288.57	0.0000
Star2000	4	126.03	31.50	3.65	0.0239
interaction	4	309.20	77.30	8.96	0.0004
Error	18	155. 25	8.62		
Total	29	3149.71			

Appendix.9 analysis of variance(Anova)on dry wieght.

Source of variation	df	SS	Ms	f	prb
Replication	2	0.31	0.16	0.65	
paulista	1	6.53	6.53	27.03	0.0001
Star2000	4	0.12	0.03	0.12	
interaction	4	0.55	0.14	0.57	
Error	18	4.35	0.24		
Total	29	11.87			

Field experiment:

Appendix .10 analysis of variance (Anova) for the emergence rate.

Source of variation	df	SS	Ms	f	prb
Replication	3	0.516	0.172	0.3317	
Variety1	1	11.396	11.396	21.9952	0.0001
Variety2	4	24.029	6.007	11.5951	0.0000
interaction	4	5.671	1.418	2.7367	0.0495
Error	27	13.989	0.518		
Total	39	55.600			

Appendix .11 analysis of variance (Anova) on emergence uniformity.

Source of variation	df	Ss	Ms	f	prb
Replication	3	1.878	0.626	1.7750	0.1757
Variety1	1	6.344	6.344	17.9910	0.0002
Variety2	4	5.277	1.319	3.7412	0.0151
interaction	4	2.401	0.600	1.7021	0.1786
Error	27	9.521	0.353		
Total	39	25.421			

Appendix.12 analysis of variance(Anova)on emergence %

Source of variation	df	SS	Ms	f	prb
Replication	3	98.95	32.98	0.75	
Variety1	1	1089.10	1089.10	24.60	0.0000
Variety2	4	931.54	232.89	5.26	0.0029
interaction	4	166.82	41.70	0.94	
Error	27	1195.29	44.27		
Total	39	3481.693			

Appendix13 analysis of variance(Anova) on seedling highest %

Source of variation	df	SS	Ms	f	prb
Replication	3	2.88	0.960	0.118	
Variety1	1	589.82	589.83	72.47	0.0000
Variety2	4	53.72	13.43	1.65	0.1907
interaction	4	308.94	77.23	9.49	0.0001
Error	27	219.75	8.14		
Total	39	1175.104			

Appendix.14 analysis of variance (Anova) on number of leaves.

Source of variation	df	SS	Ms	f	prb
Replication	3	4.83	1.61	2.41	0.0886
Variety1	1	5.67	5.67	78.88	0.0000
Variety2	4	5.44	1.36	2.04	0.1173
interaction	4	12.15	3.04	4.55	0.0061
Error	27	18.03	0.67		
Total	39	93.127			