Effect Ofpeganum Harmala on Seed Germination,Seedling Growth And Calcium Potassium Content of Triticum Aestivum and Hordeum Vulgare

Aseel Mohammed Imran
Babylone university , college of farmacy , department of pharmacognocy.
e-mail : biomaster2007@yahoo.com

Abstract

This study was designed to investigate the effect of the water extract of harmal (*Peganum harmala*) seeds on germination , seedling growth and Ca^{++} and K^{+} ions concentration of wheat (*Triticum aestivum*) and barley (*Hordeum vulgare*). It showed that the aqueous extracts of *P. harmala* seeds inhibition the germination, while the extracts increased the shoot length of *T. aestivum* and *H. vulgare* but this extracts decreased the root length of test species and increased the dry weight of the shoot, while decreased the weight of roots of the test species. The concentration 0.2% increased significantly the concentration of the potassium(k^{+}), while both concentrations (0.2%, 0.4%) cause insignificantly Decreased in potassium and calicium concentration.

the inhibitory effect on seed germination, the length and dry weight of shoot and root of test species were increased proportionally with increasing extract concentration. But the increase in shoot length and the concentration of k^{++} ion in *T. aestivum* occure only in concentration 0.2% .

Kyewords:*Peganum harmala,Triticum aestivum,Hordeam vulgare*,Calcium,Potassium,germination, growth.

الخلاصة

صممت هذه التجربة لمعرفة تاثير المستخلص المائي لبذور نبات الحرمل (*P.harmala*) في عملية الانبات و النمو و كذلك معرفة تاثير هذا المستخلص في تركيز ايوني البوتاسيوم و الكالسيوم في نباتي الحنطة (*T. aestivum*) و الشعير (*H. vulgare*) , و قد اجريت هذه التجربة في المختبر حيث زرعت بذور الحنطة و الشعير المعقمة في اطباق بتري , وقد اظهرت التجارب وجود انخفاض في النسبة المئوية للانبات لكلا نباتي التجربة في حين سبب التركيز 0.2% من المستخلص زيادة طول المجموع الخضري لنباتي الحنطة و الشعير , اما التركيز 0.4% فقد سبب انخفاض معنوي لطول المجموع الخضري لكلا النباتين , وانخفض طول المجموع الجذري بشكل معنوي لكلا نباتي التجربة مقارنة بالسيطرة و هذا الانخفاض يزداد بزيادة تركيز المستخلص و قد انخفض الوزن الجاف للمجموعين الخضري و الجذري لنباتي التجربة مقارنة بالسيطرة تحت تاثير المعاملة بكلا تركيزي المستخلص , كما و سبب المستخلص زيادة في تركيز البوتاسيوم (k^{+}) في نبات الحنطة في حين انخفض الكالسيوم (Ca^{++}) في نباتي الحنطة و الشعير بشكل غير معنوي .

الكلمات المفتاحية : الحرمل , الحنطة , الشعير, الانبات , النمو, الكالسيوم , البوتاسيوم .

Introduction :-

Peganum harmala commonly known as Syrian rue and Wild rue is flowering plant and is widly distributed in the central Asia, It has also been introduced in America and Australia.This plant is known as"Harmal"in North Africa.(Mahmoudian *et al* ., 2002) P. harmala is an medicinal plants (Patel *et al.*, 2012),and has been speculated to posses allelopathic properties (Farajollahi *et al.*,2012).It is suggested that P. harmala has an allelopathic effect on other plants, allelopathy refers to any direct and indirect harmful or beneficial effect by one plant on another through the production of chemical compounds that are released into the surrounding environmental (Rice, 1984) .

The commonly known phytochemical compounds from P. harmala are alkaloids, flavonoids and anthraquinones, amino acids and polysaccharides which isolated from its seeds, leaves, flowers, stems and roots (Pitre and Srivastava, 1987; Sharaf *et*

al.,1997; Buhkari *et al*., 2008; Movafeghi *et al*., 2009). The pharmacologically active compounds of P. harmala are several alkaloids, which are found especially in the seeds and the roots . These include β- carbolines such as: harmine, harmcaline (identical with harmidine), harmalol , harman and quinazoline derivatives: vasicine and vasicinone.(Mahmoudian *et a*l ,2002 ;Mosef *et al*., 2004 ; Pozzi *et al*.,2012 ;Asgarpanah and Ramezanloo,2012). Still, determination of phytotoxic substances in a certain plant is usually a necessary step to evaluate whether allelopathy exists, and the dependence of allelopathic effect occurring upon release of certain compounds into the environment (Gibson *et al*., 2011).

Farajollahi *et al* (2012), found that *P. harmala* extract inhibited the germination and growth of *Bromus tectorum* , Khan *et al*(2011), found that a methanol extract of *P. harmala* decreased seed germination of radish. Shao *et al*(2013) showed that *P. harmala* possessed significant inhibitory effect on dicot plants which being more sensitive than the monocot plants.

Potassium is a major osmotically active solute of plant cells(Mengel and Arncke, 1982). Leaf K^+ is thought to facilitate reduced osmotic potential leading to turgor maintenance of hydraulic conductivity gradients between leaves and soil water. Calcium has many important structural and physiological roles in plants. It is important in maintaining the stability of the cell walls, membranes and membrane bound proteins due to its ability to bridgechemical residues among these structures (Nayyar, 2003). Ca^{++} mediates several plant processes like cytoplasmic streaming, thigmotropism, cell division, cell elongation, cell differentiation, cell polarity, photomorphogenesis, plant defense, stress responses and stress protection (Nayyar, 2003) , so in our study we measured these minerals (k^+ and Ca^{++}) for there importance.

The aim of this sudy were to investigate the effect of *P. harmala* which is grown within the *T. aestivum* and *H. vulgare* field as aweed , by determination the germination , seedling growth , and measuring the contents of calcium and potassium of tested plants.It is suggested that little plant individuals. Could be grown around *P. harmala* due to its chemicals effects of compounds (allelochemicals) on other plants.

Materials and methods :

Preparation of aqueos extract:-

Ten grams of pouder seeds of *P. harmala* soaked in 200 ml distilled water (100 C^0 in conical flask) for 6 h at room temperature. After that put on horesontal shaker for 30 min and left it to stand for 1 h, then filterated and the final volume was dryied at 50 C^0 in oven untle became pouder. When prepare the concentration by taking 0.2 gm and 0.4 gm of this pouder in two flasks (each separate from other), after that add 50 ml d.w. and complet the volume to 100 ml d.w.

.**Treatments and experimental design:-**

Fifteen healthy and steriliezed seeds in sodium hypochloride, for 5 min (Martin,1990) of *T. aestivum* and *H. vulgare* were kept in steriliezed petri dishes on filter paper and moistered with 10ml extracts. Each treatment had 3 replicates each with 15 seeds. Control consisted of distilled water. The Petri-dishes were maintained under laboratory conditions at 25 C^0 temperature with diffused light during day.

Physiological parameters:-

After ten days, germinated was measured according to the low,(Fariman *et al*.;2011).

$$\text{germination (\%)} = \frac{\text{Number of germination seed}}{\text{Number of viable seeds initiated}} * 100$$

The length of shoot and root were measured by using rular(five randomize samples from each replicate) , and then these shoot and root dried in oven at55c for 15 min (Abdullah *et al.*,2011) ,then weighted by using sensitive balance.

The concentration of potasium (k^+) and calsium(Ca^{++}) measured in dried leaves by using flame photometer acording to Cresser and Parsons(1979).

Statistical analysis:-

The data obtained was subjected to three way analysis of variance complet randomized block design (CRBD). And the mean values were separated at $p<0.05$ applying least significant difference test (LSD).

Results:-

Effect on germination:-

The extracts of both concentrations (0.2% , 0.4%) inhibited germination of both test species (table 1) showed a significant effects at $p<0.05$ on germination , and this inhibition increase with increase the concentration (at 0.2% and 0.4% the percente of germination is 7.6% and 5.2% in sequence (in *T. aestivum*) in contrast with control(9.2%) and in *H. vulgare* become 8.8% and 9.2% in sequence with extract concentration (the percent of germination in control is 9.2%) , there was insignificant differse between the test species. And the comparison of the interaction between the concentrations and test species was insignificant .

Effect on shoot and root growth:-

The extract at concentration 0.2% significantly increase the shoot length of *T. aestivum* (8.9cm) in contrast with control (6.5cm), while *H. vulgare* showed insignificant differse among the concentration 0.0% , 0.2% and 0.4% (8.3cm, 8.4cm, 8.3cm) in sequence, (table 2),and there was significant differse between the test plant (6.6 cm in *T. aestivum* and 8.3 cm in *H. vulgare*), and there were asignificant increase in shoot lengthat concentration 0.2% (8.6 cm) in contrast with c0ncentration 0.4% wich significantly decrease the shoot length (6.4 cm) and control (7.4 cm.).

Tble-3- showed significant reduce in root length in both test species and this reduce a length increase with increasing extract concentration (in *T. aestivum* root length is 5.9, 1.4, 0.3 and in *H. vulgare* the root length is 5.6 , 3.7 , 2.8 in 0.0% , 0.2% 0.4% in sequence), and there was asignificant difference between the test plants (T. aestivum 2.5 cm while in H. vulgare 4.0 cm). .

Effect on shoot and root dry weight:-

The extract at concentration 0.2% showed insignificant effect on dry weight of shoot at $p< 0.05$ in *T. aestivum* ((7.4 , in contrast with control (7.6 ,)) and showed a significant decrease in shoot length of *H. vulgare* (6.5) in contrast with control (7.7) , while the concentration 0.4% showed a signoficant inhibitory effect (4.0 in *T. aestivum* and 6.3 in *H. vulgare*) (table 4).

Table -5- showed asignificant increase in dry weight of root in concentration 0.2%((6.4 , 6.9 in *T. aestivum* and *H. vulgare* in sequence in contrast with control (3.8 and 6.6 in sequence)) while it was founed asignificant decrease in dry weight of root in concentration 0.4% (1.6 in *T. aestivum* and 5.5 in *H. vulgare*) .and there was a a significant defference between test plants(*T. aestivum* 3.9mg while in *H.vulgare* 6.3mg)

Effect on Ca^{++} and K^{+} ions concentration:-

Table -6- showed insignificant differse in the concentration of Ca^{++} at 0.2 concentration in *T. aestivum*(3.5ppm) and *H. vulgare* (3.8ppm) in contrast with control(3.5 , 3.9) in sequence , but in 0.4 the concentration of Ca^{+} showed a significant decrease in*T.aestivum* (3.0ppm) and in *H. vulgare* (3.1ppm) in contrast with control.

While the concentration of potassium(K^{+}) (table 7) showed asignificant increase at concentration 0.2% and 0.4% in *T.eastivum* (37.9ppm , 37.2ppm) in contrast with control wich give 30.8ppm , but in *H. vulgare* showed insignificant decrease in K^{++} oncentration (47.0 ppm , 45.5 ppm , 44.8 ppm) at all concentration.

Table 1. Effect of *P. harmala* extract on germination percent (%) of *T. aestivum* and *H. vulgare* .

concentration / plant	o.o% (control)	0.2%	0.4%	mean
T. aestivum	92	76	52	73.3
H. vulgare	95	88	92	91.6
mean	93.5	77.5	72.0	81.7

LSD(0.05)
Concentration =1.150
Plant =0.813
Entraction =0.813

Table 2. Effect of *P. harmala* extract on shoot length (cm) of *T.aestivum* and *H. vulgare* .

concentration / plant	o.o% (control)	0.2%	0.4%	mean
T. aestivum	6.5	8.9	4.5	6.6
H. vulgare	8.3	8.4	8.3	8.3
mean	7.4	8.6	6.4	8.18

LSD(0.05)
Concentration =0.880
Plant =0.508
Entraction =0.508

Table 3. Effect of *P. harmala* extract on root length (cm) of *T.aestivum* and *H. vulgare*

concentration / plant	o.o% (control)	0.2%	0.4%	mean
T. aestivum	5.9	1.4	0.3	2.5
H. vulgare	5.6	3.7	2.8	4.0
mean	5.7	2.5	1.5	3.24

LSD(0.05)
Concentration =0.501
Plant =0.354
Entraction =0.354

Table 4. Effect of *P. harmala* extract on dry weight of shoot (mg) of *T.aestivum* and *H. vulgare* .

concentration / plant	o.o% (control)	0.2%	0.4%	mean
T. aestivum	7.6	7.4	4.0	6.3
H. vulgare	7.8	6.5	6.3	6.8
mean	7.7	6.9	5.15	6.58

LSD(0.05)
Concentration =0.706
Plant =0.499
Entraction =0.499

Table 5. Effect of *P. harmala* extract on dry weight of root (mg) of *T.aestivum* and *H. vulgare*

concentration / plant	o.o% (control)	0.2%	0.4%	mean
T. aestivum	3.8	6.4	1.6	3.9
H. vulgare	6.6	6.9	5.5	6.3
mean	5.2	6.6	3.5	5.1

LSD(0.05)
Concentration =1.114
Plant =0.643
Entraction =0.643

Table 6. Effect of *P. harmala* extract on potasium concentration (ppm) of *T.aestivum* and *H. vulgare* .

concentration / plant	o.o% (control)	0.2%	0.4%	mean
T. aestivum	30.8	37.9	37.2	35.3
H. vulgare	47.0	45.5	44.8	45.7
mean	38.9	41.7	41	40.5

LSD(0.05)
Concentration =0.298
Plant =0.727
Entraction =0.727

Table 7. Effect of *P. harmala* extract on calcium concentration (ppm) of *T.aestivum* and *H. vulgare*

concentration / plant	o.o% (control)	0.2%	0.4%	mean
T. aestivum	3.51	3.5	3.0	3.3
H. vulgare	3.9	3.8	3.1	3.6
mean	3.7	3.6	3.0	3.4

LSD(0.05
Concentration =0.298
Plant =0.211
Entraction =0.211

Discussion :-

Aqueous extract significantly reduced the germination and this reduced inarease with the increasing extract concentration (table 1). This reduce in germination may be due to the chemicals which found in *P. harmala* like alkaloids , glycosides , and tannins(Buhkari et al., 2008; Movafeghi et al., 2009). When

susceptible plants are exposed to allelochemicals, germination, growth and development may be affected (Xuan *et al.,* 2004). Allelopathic compounds may decrease cell turgor (El-Khawas and Shehala, 2005), mitosis division, DNA replication (Roshchina, 2001)and finally decreasing cell growth. Our reselts finding agree with resulting of Farajollahi et al(2012) who found that the allelopathic effect of *P. harmala* extract resulted in negative effect on germination properties of *Bromus tecorum.*

Furthermore the comparison of extract concentrations gave a significant inhibitory effect with the increase of extract concentrations. The present findings corroborate the earlier results reported by Farajollahi *et al* (2012) who found that in all treatments, the negative allelopathic effect of *P. harmala* on germination characteristics of *B. tectorum* was increased as the pouder weight of *P. harmala* was increased .

Table 2 showed increase in shoot length for both test species at 2% concentration,our results agree with resulting of El hassan *et al* (2013) who found an increase in shoot length of *Cucumis melo* when treated by *P.harmala* seed extract, Shao *et al* (2013) found that *P. harmala* possessed significant growth inhibitory effect on dicot plants (lettuce and amaranth) more than monocot plants(wheat and ryegrass). *P. harmala* extract contain alkaloids , flavonoids , glycosides (Sharaf et al.,1997; Buhkari et al., 2008; Movafeghi et al., 2009) and this compounds have allelopathic effects which are effect on cell division and cell elongation by reduse auxin values (Tawaha *et al*., 2007) these compounds decrease plant growth by preventing of nutrients absorption or direct interference into respiration or phosphorelation oxidative.

Root length was more affected compared to length of shoot (table-3-) , and the decrease in root length increase with increasing the concentration of extract , this results agree with resulting of Shao *et al* (2013) who found that increase in *P. harmala* extract concentration cause increase the inhibition of root length of dicot and monocot plants, Farajollahi et al (2012) found that the root length of *Bromus tectorum* increase with increasing weight value of *P. harmala*. this study agree with the study of Assaeed & Al-Doss, (1996) which found that the extract of *R. stricta* (which also contain alkaloids) cause less effect on shoot length compared to root length at initial growth of seedling of *Achillia fragrantission , Farsetia aegyptia , Lasiurus scidicus.*

The reduce in the dry weight in shoot and root of both test species belong to the allelopathic effect of *P. harmala* extracts which inhibiting the growth of test species except in concentration 0.2% which showed increase the dry weight of roots of both test species , that may be occure because of the chemicals that found in the extract which decrease the cell elongation but increase the concentration of materials in the cells or increase of mitotic index , Mutawakil (2012) found that the aqueous extract of *R. stricta* ((which also contain alkaloids(Al- Yahya *et al,* 1990)) increase of mitotic in root tip of *Vicia faba*.

Our resulting about increase of potassium concentration at 0.2% in *T. aestivum* and decrease of calcium agree with resulting of Majid et al (2007) who found an increase in potassium concentration and decrease in calsium concentration in *T. aestivum* when trasfered to drout stress. Asghari *et al(*2001) suggested that higher ratio of K^{+}/Ca^{++} in response to drought stress in two lines demonstrated the tolerance of wheat cultivars against stress.

Conclusion:-

This study conducted that:-

1- *P.harmala* extract inhibited germination of *T. aestivum* and *H. vulgare*, while *T. aestivum* more sensitive than *H. vulgare.*

2- The inhibition in growth parameters increased with increasing in concentration of extract.

3- there was increasing in K^+ content of 0.2% and 0.4% concentration in *T. aestivum.* This may indicate the ability of this plant to accumulate K^+ in its tissue.

References:-

Abdullah S, Ahmad,M S, Shaari A, Johar H M, Noor N F. 2011. Drying characteristics and herbal metabolites composition of misia kucing (*Orthosiphon staminiues Benth*.) International Conference on Food Engineering and Biotecnology. IPCBEEvol.9; 305-309.

Al- Yahia MA ,Al-Mashall A, Mossa JS, Al –Badr AA,Tariq M. 1990.Saudi plants: Aphytochemical and Biological Approach.KACST,Riyadh:King Saud University press; 123-129. .

Asgarpanah J and Ramezanloo F 2012. Chemistry, pharmacology medicinal properties of Peganum harmala.African journal of pharmacy and pharmacology Vol.6(22),pp. 1573-1580.

Asghari, R. H.; Zadeh,E and A.R. Khabiri.2001. Effect of drought stress on abscisic acid, mannitol, K^+ and Ca^{++} content in two lines of wheat (*Triticum aestivum* L) . Pak. J.Bot., 33:197-202.

Assaeed AM. and Al-Doss AA. 1996. Effect of Rhazya stricta Leachate on Seedling Growth and Survival of Some Range Plant Species. JKAU: Met., Env., Arid Land Agric. Sci., vol.7. pp. 13-20.

Bunkari, N.;Choi, J.H.; Jeon CW.; Park HW.; Kim ,W.H.; Khan, M.A and Leet , SH 2008 Phytochemical Studies of the Alkaloids from Peganum harmala . Appi.Chem., 12 (1): 101-104.

Cresser, M.E. and I. Parsons 1979 Sulpharic, perichloric and digestion of plant material for magnesium. Analytical chemical A., 109: 431-436.

El- Khawas, S.A.;Shehala, M.M. 2005. The allelopathic potentialities of Acacia and Eucalyptus prostrate on monocot (Zea mays L.) and dicot (Phasiolus vulgaris L.) plants. Biotechnology, 4: 23-34.

Farajollahi, A.; Tavili, A.; Gholinejad, B.; Darini ,J and Hossein Pouzesh 2012. Investigation and Compare the Allelopathic Effect for Different Tissues of Peganum harmala in Different Amounts on the Bromus tectorum Germination and Growth Characreristics. Ecopersia 1 (3) , 217- 226.

Fariman, Z.K., Azizi, M. and Noori, S. 2011. Seed Germination and Dormancy Techniques for Echinacea purpurea L. J. BIOL.ENVIRON. SCI. 5(13) : 7-10.

Gibson, D.M.; Krasnoff, S.B.; Biazzo, J.; Milbrath, L. 2011. Phytotoxicity of antofine from invasive swallow- worts. J. Chem. E col. 37, 871- 879.

Khan, M. Hussain1, F. Musharaf, S. and Imdadullah 2011 Allelopathic effects of *Rhazya stricta* decne on seed germination and seedling growth of maize African Journal of Agricultural .

Mahmoudian, M.;Jalilpour,H. and Salehian, P.2002. Toxicity of *Peganum harmala* :Riview and a Case Report. Iranian Journal of Pharmacology & Therapeutics. IJPT1: 1-4.

Majid, S.A.; Asghar, R and Murtaza, G. 2007. Potassium-Calcium Interrelationship Linked to Drought Tolerance Wheat (*Triticum aestivum*) .Pak. J.Bot., 39(5) :1609-1621.

Martin, V.L.; Mccoy, E.L. and Dick, W.A .1990. Alellopathy of crop residues influences corn seed germination and early growth. Agron , J. 82: 555-560.

Mengel, K. and Arnk, M.M. 1982. Effect of potassium on the water potential, the pressure potential , the osmotic potential and cell elongation in leaves of Phasiolus vulgaris. Physiol. Plant , 54: 402-408.

Monsef, H.R.,Ghobadi, A., Iranshahi, M. and Abdollahi, M.2004. Antinociceptive effects of Peganum harmala L. alkaloid extract on mousse formalin test. J. Pharm. Sci. 7(1): 65-69.

Movafeghi, A.;Abidini, M.;Fathoazad, F.; Aliasgharpour, M.; Omidi, Y. 2009 Floral nectar Composition of Peganum harmala L. Nat. Prod. Res. 23, 301-308.

Munns, R. and Termaat, A. 1986. Whole-plant response to salinity. Australian Journal of Plant Physiology, 13: 143-160.

Mutawakil, H.Z. Mohammed. 2012. Effect of Rhazya stricta Leaves Extract in root Tip Meristems of VIcia faba . New York science journal; 5(4) 11-17.

Nayyar, H. 2003. Calcium as environmental sensor in plants. Curr. Sci., 84(7): 893-902.

Patel, K., Gadewar, M., Tripathi, R. and Prasad,S.K. 2012. A review on medicinal importance, pharmacological activity and bioanalytical aspects of beta- carboline alkaloid "Harmine" .Asian Pacific Journal of Tropical Biomedicine. 660- 664.

Pitre , S and Srivastava, S.K. 1987 Two new anthraquinons from the seeds of Peganum harmala . Planta Medica, 53(1): 106- 107.

Pozzi, F., Shibayama, N., Leona, M. and Lombardi, J.R. (2012). TLC-SERS study of Syrian rue (*Peganum harmala*) and its main alkaloid constituents. J. Raman Spectrosc.44, 102- 107.

Rice, L.E. 1984. Allelopathy. 2nd Ed. New York: Academic Publishers, pp. 424.

Roshchina, V.V. 2001. Molecular – cellular mechanisms in pollen allelopathy. Allelopathy Journal 8: 11- 28.

Shao, H., Hung, X., Zhang, Y.. and Zhang, C. 2013. Main Alkaloids of *Peganum harmala* L. and Their Different Effect on Dicot and Monocot Crops. Molecules, 18, 2623- 2634.

Sharaf, M., El- Ansari, M.A.., Matlin, S.A. Saleh, N.A. 1997. Four flavonoid glycosides from Peganum harmala. Phytochem., 44(3): 533- 536.

Tawaha, K.H., Alali, F., Gharaibeh, M., Mohammad, M. and El- Elimat, T. 2007. Antioxidant activity and total phenolic content of selected Jordanian plant species, Food Chem . 104: 1372- 1378.

Xuan, T.D, Tawata, S, Hong, N.H,., Khanh, T.D, Chung, i.m. 2004. Assessment of phytotoxic action of Ageratum conyzoides L. (billy goat weed) on weeds. Crop Protection, 23: 915- 922.

Zahran, M.A. 1983. Introduction to plant ecology and vegetation types of Saudi Arabia. Faculty of Metrology and Environmental Studies, King Abduaziz Univercity.Jedah.