Phytochemical analysis, phytotoxic and insecticidal activities of medicinally important *Periploca hydaspidis*

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Abstract: The present study investigates the phyto-chemical analysis, phytotoxic activity and insecticidal activity of the stem and root tissues of medicinally important *Periploca hydaspidis.* All the extracted samples exhibited the phytotoxic activity. The data confirmed that water was the best solvent to extract the phytotoxic compounds from stem and root tissues. Roots extracted with different solvents exhibited better phytotoxic potential than samples obtained from the stem tissues. Samples extracted in methanol and water from both stems and roots tissues were active against the subject insects while hexane, ethyl acetate and butanol extracted samples in both parts exhibited no insecticidal activity. Water extracted samples of the stem and root exhibited better insecticidal activity compared with methanol extracted samples. *Tribolium castaneum* and *Rhizopertha dominica* were more sensitive in both the cases than *Callosobruchus maculates*. The plant contained alkaloids, flavonoids, carbohydrate, proteins and saponins.

Keywords: Phytochemistry, phtyto-toxic activity, insecticidal activity, *Periploca hydaspidis.*

INTRODUCTION

Plants having medicinal activities produce different bioactive chemical compounds which are used in the treatment of different human, animal, plant diseases and as antibiotics, insecticides, herbicides etc. Herbal medicine represents one of the most important fields of traditional medicine in rural areas. Thus, phytotherapy is practiced by a large section of people for the treatment of several physical, physiological, mental and social ailments. The study of medicinal plants is important to find out their potential as sources for new drug development (Schopen, 1983; El-Faky *et al*., 1995; Awadh *et al*., 2001). A renewed curiosity has occurred to investigate for phytochemicals of native and naturalized plants for pharmaceutical and nutritional purposes (Oktay *et al*., 2003; Wangensteen *et al*., 2004) with the identification that plant-derived products have great potential as sources of pharmaceuticals (Bakht *et al*., 2013 a,b; 2014 a, b,c; Amjad *et al*., 2016; Wajid *et al*., 2016; Bilal *et al*., 2017). Leaves, roots, flowers, whole plants, and stems are used for extracting phytochemicals which are used as medicines for treatment of many diseases.

There has been an intensive search for substances with considerable antimicrobial properties. Plants are known to synthesize certain chemicals which are naturally toxic to bacteria and fungi (Tshikalange *et al.,* 2005). In bryophytes, which are the simplest land plants, anatomical barriers are less effective and as a consequence, the synthesis of particular molecules, secondary metabolites with antimicrobial activity: the so called `chemical barrier' is the most effective defense mechanism (Harborne, 1973). Biflavonoids in mosses are also reported as possible chemical barriers against micro-organisms (Basile *et al*., 1999). Epidemiological studies have brought into being that intake of antioxidants such as vitamin C reduces the risk of coronary heart diseases and cancer (Marchioli *et al*., 2001). It is possible to reduce the risk of chronic diseases and to prevent disease progression by either enhancing the body's natural antioxidant defense or by supplementing with proven dietary antioxidants (Stanner *et al*., 2004). It has been reported that phenols, mainly flavonoids, from some medicinal plants have many biological activities (Ozgová *et al*., 2003).

Periploca hydaspidis belongs to the family *Asclepiadaceae*. It is a twining shrub, usually leafless; branches are smooth green and are 1.5 mm in diameter. Flowering is perennial and usually occurs in September-October. This plant is usually found in Swat, Pakistan. Other species of the family like *Periploca aphylla, Periploca calophylla* etc are found in Kashmir and India. It has been reported that other species like *Periploca aphylla* and *Periploca laevigata* possesses antibacterial and antioxidant activities (Mohamed *et al*., 2009; Iqbal *et al*., 2012).

MATERIALS AND METHODS

Collection and identification of plant material
The proposed study was conducted at the Institute of Biotechnology and Genetic Engineering, The University of Agriculture, Peshawar-Pakistan. Plant specimen was collected from different parts of Swat valley of Khyber Pukhtunkhwa (KP) province of Pakistan especially from the mountains of Madyan and Marghuzar area. The plant was identified by plant taxonomist, Prof. Mehboob-ur-Rahman of the Department of Botany at Government post Graduate Jahan Zeb College, Swat (KPK).

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Preparation of crude extracts of stems and roots

The plant material was dried in a dark shaded place to prevent the degradation of the active phytochemical constituents due to direct sunlight. The plant was then divided into stems and roots and made powdered with grinder (Germany model 3240) separately. One kilogram of powdered plant material was taken in separatory funnel and three litres of methanol (Analytical grade) was poured into it. The funnel was kept for 7 days at room temperature and stirred regularly for thorough mixing. The seperatory funnel was kept in dark to protect it from direct sunlight. The methanol extracted samples were filtered through Whatman No. 1 filter paper and fresh methanol was added to the plant residue for further extraction and was soaked again for another seven days. The methanolic solution having soluble compounds was transferred to rotary evaporator for evaporation of the methanol. The methanol was recovered under pressure at 40ºC leaving the semi-solid extract, recognized as crude methanolic extract. The whole process was repeated three times. The crude methanolic extract was weighed and divided into two portions. One portion (10g) was tested as crude methanolic extract, while the other portion (43g) was dissolved in 300ml distilled water and transferred to separatory Funnel for further fractionation with different solvents. For this purpose n-hexane was added and lightly shaken. After shaking, the separatory funnel was allowed in the stand in order to separate the water and n-hexane in to two layers. The upper organic layer was removed and the lower layer was again extracted with fresh n-hexane. The process was repeated three times. All extractions of n-hexane were collected in a single flask and transferred to rotary evaporator for evaporation of n-hexane to obtain semi-solid material. The semi-solid extract was further dried at 45°C on water bath and preserved in glass vials. Similar procedure was adopted for ethyl acetate and butanol. The aqueous part at the end was collected and dried using rotary evaporator and water bath. Finally five different extracted samples namely crude methanol, n-hexane, ethyl acetate, butanol and water were obtained. The described procedure was carried out for the stems and roots of *Periploca hydaspidis,* separately.

Phytotoxic activity of the plant extracted with different solvents

Stock solution of the different extract was prepared in ethyl acetate. From the stock solution, different concentrations were prepared in falcon tubes and kept open for 24 hours to evaporate ethyl acetate. After 24 hours, 20 ml of the enriched medium with slightly basic pH was added to sterilized flasks having ten healthy plants of *Lemna minor* with three fronds each and kept in growth cabinet/chamber for seven days (at 30 ^{0}C; light intensity of 9000 lux and 60% humidity). On eighth day the fronds was measured and parquet was used as positive control. For survival analysis the data was analyzed by IBM SPSS statistics20 (De Almeida *et al*., 2010) and the percent growth was calculated according to the following formula (Rehmanullah *et al*., 2012).

$$\text{Growth inhibition } \% = \frac{\text{Fronds in sample}}{\text{Fronds in ve control}} \times 100$$

Insecticidal activity of different extracts of the plant

Preparation of test samples

For the preparation of test samples 5, 25 and 125 mg of each extract was solubilized in methanol and the concluding dilution of 5, 25 and 125 mg/ml was obtained.

Rearing of the insects

Three species of stored grain pests namely *Tribolium castaneum, Rhizopertha dominica* and *callosobruchus maculatus* were acquired from The Research Laboratory of Entomology section, Nuclear Institute of Food and Agriculture (NIFA) Peshawar Pakistan. Fresh grain and flour was infested with these insects. After the whole life cycle, 5 g part of the infested grain which consisted of eggs of the insects was transferred to more fresh stored grains. After 7 days, the eggs were hatched and small insects began to develop. For our experiment we selected one day old culture following the methods of Rehman-ullah *et al*. (2012).

Procedure for the insecticidal activity

Filter papers were cut roundly in the size of 90mm and were sterilized through autoclave. They were kept in petri plates. The prepared test samples were loaded on these petri plates. Five ml of the test sample was loaded to each petri plate and kept open for 24hrs in order to completely evaporate the methanol. Ten insects were added to each petri plate and kept at 27°C for 24hrs in a growth chamber. On the third day readings were noted following the methods of Rahmanullah *et al*. (2012). For survival analysis the data was analyzed by IBM SPSS statistics20. Permethrin was used as standard insecticide.

Qualitative analysis

Confirmatory test of alkaloid

The alkaloid in the extracts was confirmed through Mayer's Test (Tiwari *et al*., 2011). Solvent free extracts of about 50 mg were stirred separately with 10 ml of dilute hydrochloric acid and then filtered it. After filtration, one or two drops of Mayer's reagent were added to side of test tube containing 3ml of filtrate. A white or creamy precipitate showed the presence of alkaloid in the extracts.

Confirmatory test of proteins

Protein concentrations in different solvent extracted samples from *Periploca hydaspidis* were determined through Millon's Test. For the preparation of Millon's solution, 1 gram of mercury was dissolved in 9 ml of fuming nitric acid. After completion of the reaction, an equal amount of distilled water was added. Hundred mg of the different extracts were dissolved separately in 10 ml of distilled water and filtered through Whatman No.1

filter paper. Few drops of Millon's reagent were added to 2 ml filtrate, the white precipitate indicated the presence of protein (Tiwari *et al*., 2011).

Confirmatory test for carbohydrate

Carbohydrates in the extracts were confirmed through Fehling's Test. Fehling's solution A was prepared by dissolving 34.66 gram of $CuSO_4$ in distilled water and final volume was made upto 500 ml. Fehling's solution B was made by dissolving 173 grams of potassium sodium tartarate and 50 grams of sodium hydroxide was dissolved in distilled water and final volume was made upto 500 ml solution. Hundred mg of the extracts were dissolved separatly in 5 ml of distilled water and filtered through Whatman No.1 filter paper. After filtration, one ml of filtrate was boiled with one ml each of Fehling's solution A and solution B, the appearance of red precipitate indicated the presence of sugar (Tiwari *et al*., 2011).

Confirmatory test of fixed oil and fats

Saponification test

Hundred mg of extracts were mixed with few drops of 0.5 N alcoholic potassium hydro oxides along with a drop of phenolphthalein. Mixture was then heated on water bath for 2 hrs. The soap formation and partial neutralization of alkali was observed which indicated the presence of fixed oil and fats (Tiwari *et al*., 2011).

Tannins test

One ml of distilled water and 1-2 drops of ferric chloride solution was added to 0.5 ml of extracts solution. The appearance of blue color indicated the presence of gallic tannins and green black for catecholic tannins (Lyengar, 1995).

Saponins test

Half gram of the plant extracts were shaken in a test tube for about 5 minutes, appearance of persistent foam indicated test positive for Saponins (Harborne, 1973).

Confirmatory test of sterols

The presence of the phytosterols in the extract was confirmed through Libermann-Burchard's test. Fifty mg of the extract was dissolved in 2ml acetic anhydride. Two drops of sulphuric acid were added slowly. Red color in the sulphuric acid region indicated the presence of phytosterol (Tiwari *et al*., 2011).

Flavonoid test

One and half ml of 50% methanol solution was added to four milliliters of the extract solution and was warmed. Magnesium and 5-6 drops of concentrated Hydrochloric acid was added to the solution. Red color showed the presence of Flavonoids (Siddiqui and Ali, 1997).

STATISTICAL ANALYSIS

Data are presented as mean values of three replicates. Statistix 8.1 (Analytical computer software) software was used to carry out statistical analysis. The least significant difference among means was computed using Least Significant Difference (LSD) test (Steel *et al*., 1997).

RESULTS

Phytotoxic activity of different solvent extracted samples from the stems of Periploca hydaspidis

The data regarding phytotoxic potential of different extracts obtained from the stem of *Periploca hydaspidis* is shown in fig. 1. The results revealed that maximum phytotoxic activity of 70% was exhibited by water extracted samples at 1000 ppm/ml followed by 63.30% of the same sample and butanol extracted sample respectively. The data also suggested that minimum phytotoxic potential of 10% was demonstrated by n-hexane extracted sample at 800ppm/ml. It can be seen from the data that water, butanol and crude methanolic extracted samples showed good inhibitory effects on the proliferation of the *lemna minor* while the inhibitory effects of n-hexane and ethyl acetate extracted samples were moderate. The FI_{50} values calculated by SPSS software indicated that FI_{50} value for crude methanolic extracts was 934.845 while FI_{50} values for n-hexane, ethyl acetate, butanol and water extracted sample were 979.844, 998.713, 917.036 and 882.988 respectively. The order of the phytotoxic potential of different solvent extracted samples obtained from the stem tissues water > butanol > methanol >n-hexane >ethyl acetate.

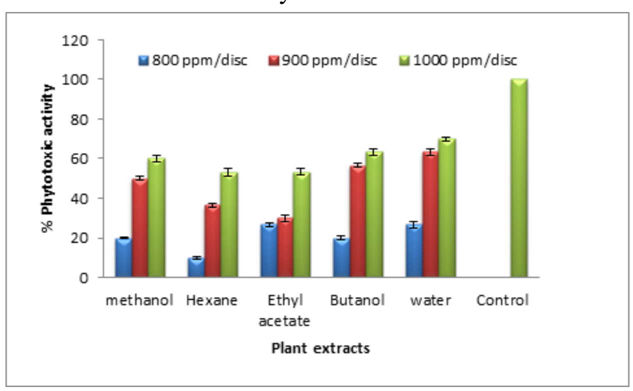

Fig. 1: Phytotoxic activity (%) of different solvent extracted samples from the stems of *Periploca hydaspidis* against *Lemna minor* (Bar represent LSD at $P<0.05$).

Phytotoxic activity of different solvent extracted samples from the roots of periploca hydaspidis

Fig. 2 presents data regarding phytotoxic potential of different extracts from the roots of *Periploca hydaspidis*. The data showed that highest phytotoxic activity was measured by water extracted samples at 1000 ppm/ml which was calculated to be 86.6%, followed by 76.60% of the butanol, 66.60% of ethyl acetate and 66.59% of n-hexane extracted samples. The lowest phytotoxic potential was shown by crude methanolic extracts at 800 ppm/ml which was calculated to be 16.60%. Water and butanol extracted samples demonstrated good inhibitory effects on the proliferation of the *lemna minor* while moderate inhibitory activity was noted for crude methnol, n-hexane and ethyl acetate extracted samples. The data also suggested that FI_{50} for methanol extracted sample was 955.281 while the FI_{50} values for n-hexane, ethyl acetate, butanol and water extracted samples were 922.854, 922.854, 914.614 and 885.9386 respectively. The order of the phytotoxic potential of the different solvent extracted samples was water >butanol >ethyle acetate >n-hexane >methanol.

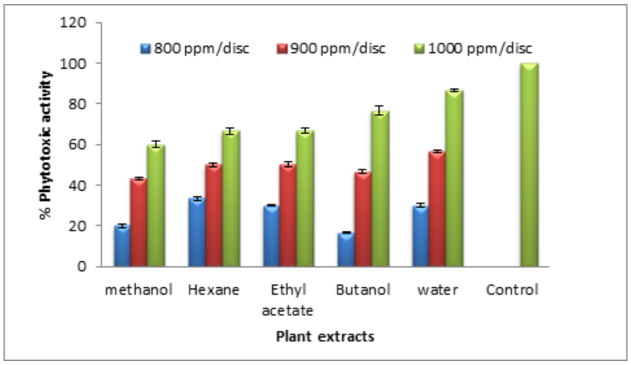

Fig. 2: Phytotoxic activity (%) of different solvent extracted samples from the roots of *Periploca hydaspidis* against *Lemna minor* (Bar represent LSD at P<0.05).

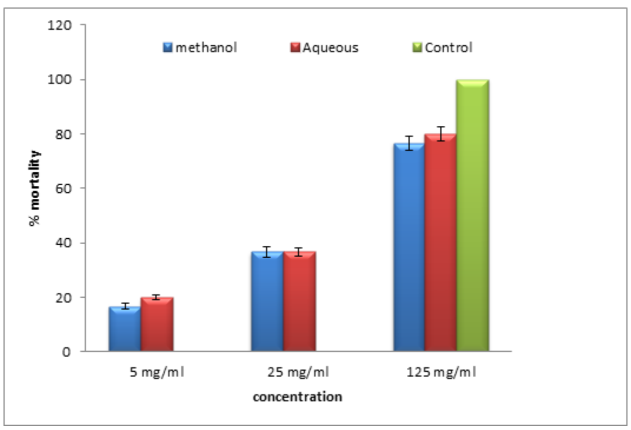

Fig. 3: Insecticidal activity (%) of water and butanol extracted samples from the stem of *Periploca hydaspidis* against *Tribolium castaneum*(Bar represent LSD at P<0.05).

Insecticidal activity of different solvent extracted samples from the stems and roots of Periploca hydaspidis

Data regarding the insecticidal potential of water and methanol extracted samples from the stem of *Periploca hydaspidis* against *Tribolium castaneum* is shown in fig. 3. The data revealed that variable effect was shown by different extracts and was dose and insect dependent. Maximum mortality of 80.66% was shown by water extracted samples at 125 mg/ml followed by methanol extracted samples (76.66%). Similarly, minimum mortality of 16.66% was demonstrated by methanol extracted samples at 5mg/ml when compared with controls. The survival (Probit) analysis of the data revealed that the LC_{50} value for methanol extracted samples was 65.55 while LC_{50} value for water extracted samples was calculated to be 59.61. The LCL for methanol extracted samples was 43.241 and the UCL was found to be 94.273, while the LCL and UCL for water extracted sample was 37.37 and 86.09 respectively.

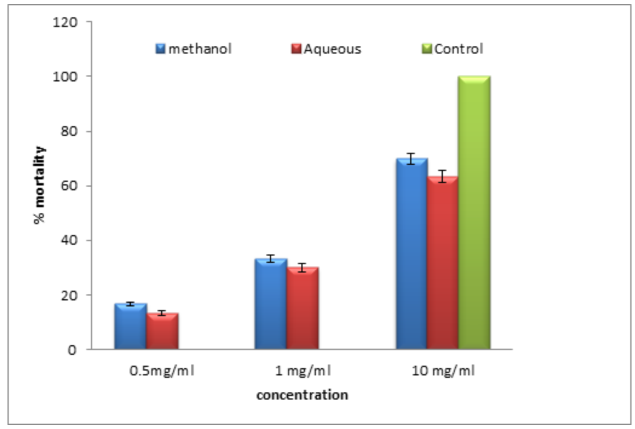

Fig. 4: Insecticidal activity (%) of water and butanol extracted samples from the stem of *Periploca hydaspidis* against against *Rhizopertha dominica* (Bar represent LSD at P<0.05).

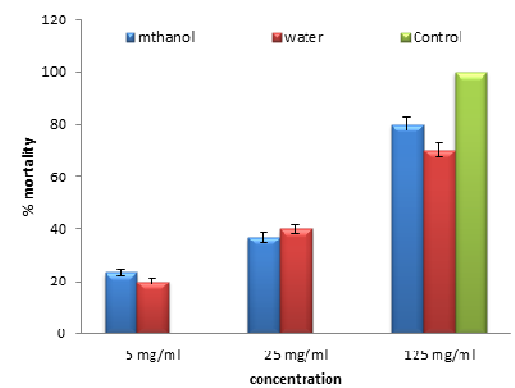

Fig. 5: Insecticidal activity (%) of water and butanol extracted samples from the stem of *Periploca hydaspidis* against *Callosobruchus maculates* (Bar represent LSD at P<0.05).

Fig. 4 indicates the insecticidal activity of water and methanol extracted samples from the stems of *Periploca hydaspidis* extracted against *Rhizopertha dominica.* Maximum mortality of 70.00% was shown by methanol extracted samples at 125mg/ml followed by water extracted samples (63.33%) when compared to control. The data also indicated that minimum mortality (13.33%) was calculated for water extracted samples at 5 mg/ml. The survival (Probit) analysis of the data revealed that the LC_{50} value for butanol extracted samples was 76.42 while LC_{50} value for water extracted samples was calculated to be 90.59. The LCL for methanol extracted samples was 51.67 and the UCL was found to be 113.79, while the

LCL and UCL for water extracted sample was 63.93 and 138.75.

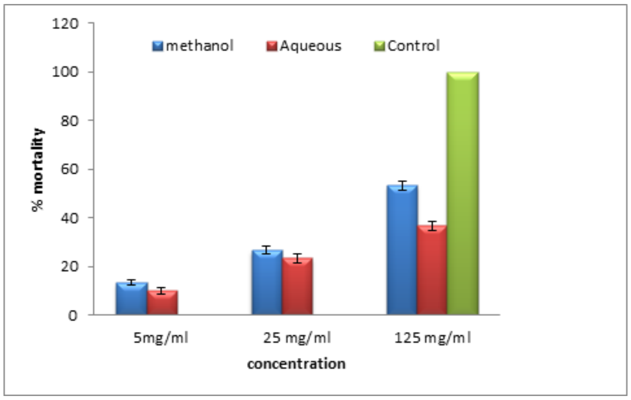

Fig. 6: Insecticidal activity (%) of water and butanol extracted samples from the roots of *Periploca hydaspidis* against *Tribolium castaneum* (Bar represent LSD at P<0.05).

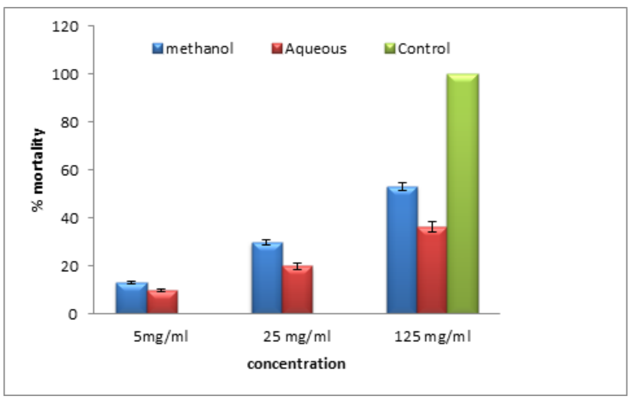

Fig. 7: Insecticidal activity (%) of water and butanol extracted samples from the roots of *Periploca hydaspidis* against *Rhizopertha dominica* (Bar represent LSD at P<0.05).

Data relating to the insecticidal activity of water and methanol extracted samples from the stem of *Periploca hydaspidis* against *Callosobruchus maculatus* is shown in fig. 5. Maximum mortality of 80.00% was noted by methanol extracted samples at 125 mg/ml followed by water extracted samples (70.00%) compared with control. Minimum mortality rate (20.00%) was established by water extracted samples at 5 mg/ml. The survival (Probit) analysis of the data revealed that the LC_{50} for methanol extracted samples was 57.35 while LC_{50} for water extracted samples was calculated to be 69.41. The LCL for methanol extracted samples was 34.34 and the UCL was found to be 84.60, while the LCL and UCL for water extracted sample was 41.60 and 110.06 respectively.

Data concerning the insecticidal potential of water and methanol extracted samples from the of roots of *Periploca hydaspidis* against *Tribolium castaneum* is shown in fig. 6. Maximum mortality percentage rate was demonstrated by methanol extracted sample at 125 mg/ml which was 53.33% followed by water extracted sample which was 36.66%. The minimum percent mortality rate was established by water extracted sample which was calculated to be 10.00% at 5 mg/ml. The survival (Probit) analysis of the data revealed that the LC_{50} for methanol extracted was 113.05 while LC_{50} for water extracted samples was calculated to be 175.70. The LCL for methanol extracted samples was 78.79 and the UCL was found to be 206.96, while the LCL and UCL for water extracted sample was 110.97 and 997.25 respectively.

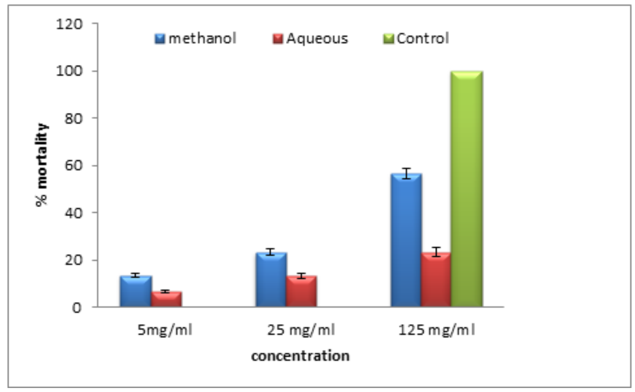

Fig. 8: Insecticidal activity (%) of water and butanol extracted samples from the roots of *Periploca hydaspidis* against *Callosobruchus maculatus* (Bar represent LSD at P<0.05).

The data relating to the insecticidal activity of water and methanol extracted samples from the roots of *Periploca hydaspidis* against *Rhizopertha dominica* is shown in fig. 7. Maximum mortality percentage rate was demonstrated by methanol extracted sample at 125 mg/ml which was 53.33% followed by water extracted samples (36.66%). Minimum mortality rate of 10.00% measured by water extracted samples (10.00%) at 5 mg/ml. The survival (Probit) analysis of the data indicated that the LC_{50} value for methanol extracted samples was 111.49 while LC_{50} value for water extracted samples was calculated to be 173.27. The LCL and UCL for methanol extracted samples was 76.24 and 213.96 respectively. Similarly, the LCL and UCL for water extracted samples was 112.46 and 687.26 respectively.

Fig. 8 presents data relating to the insecticidal potential of water and methanol extracted samples from the root of *Periploca hydaspidis* against *Callosobruchus maculatus*. The data indicated that highest mortality percentage was demonstrated by methanol extracted samples at 125 mg/ml which was 56.66% followed by water extracted samples (23.33%). The lowest mortality was noted by water extracted samples (6.66%) at 5 mg/ml. The survival (Probit) analysis of the data revealed that the LC_{50} value for methanol extracted samples was 106.94 while LC_{50} value for water extracted sample was calculated to be 258.44. The LCL for methanol extracted sample was 76.85 and the UCL was found to be 258.44. The plant

Table 1: Qualitative phytochemical analysis of different solvent extracted samples from the stem (+ shows presence and – presents absence)

Phytochemicals	Name of the extract				
	methanol	n-hexane	Ethyl acetate	Butanol	Water
Alkaloids	+	+	+	-	+
Carbohydrates	+	+	+	-	+
Flavonoids	+	-	-	-	+
Proteins	+	+	+	+	+
Saponins	-	-	-	+	+
Sterols	+	-	-	+	+
Tannins	-	+	-	+	+

Table 2: Qualitative phytochemical analysis of different solvent extracted samples from the root (+ shows presence and – presents absence)

Phytochemicals	Name of the extract				
	methanol	n-hexane	Ethyle acetate	Butanol	Water
Alkaloids	+	+	+	-	+
Carbohydrates	+	+	+	-	+
Flavonoids	-	-	-	+	+
Proteins	-	+	+	+	+
Saponins	-	+	-	+	-
Sterols	+	-	-	+	+
Tannins	-	+	-	-	-

extracted with other solvents did not show any insecticidal activity.

Phytochemical analysis of different solvent extracted samples from the stems and roots of periploca hydaspidis

The results obtained from different qualitative phytochemical analysis of the stem tissues of the plant is given in table 1. The data revealed that alkaloids and carbohydrates were present in all the extracts except butanol extracted samples. Similarly, flavonoids were present only in methanol and water extracted samples and proteins in all the extracted samples while saponins were present only in butanol and water extracted samples. table 2 indicates the qualitative phytochemical analysis of the root tissues. Analysis of the data showed that alkaloids and corbohydrates were confirmed in all the extracts except butanol extracted samples, flavonoids were present only in butanol and water extracted samples. Proteins were found in all the extracted samples except methanol extracted sample while saponins were present only in butanol and n-haxnae extracted samples. The presence of tannins was noted only in n-hexane extracted samples and sterols were found to be present in methanol, butanol and water extracted samples.

DISCUSSION

Famers usually face the problems of weeds infestation in their crops and therefore extensively use chemical herbicides which are environmentally offensive, especially to water, soil and food (Wahab *et al*., 2012). In the present study we carried out phytotoxic activity to develop new herbicides which are cheap, safe and environmentally passive. Phytotoxic potential of different extracts of the stem and root of *Periploca hydaspidis* revealed that the maximum activity was exhibited by water extracted sample at the highest concentration followed by butanol extracted samples. The water, butanol and crude methanolic extracted samples showed good inhibitory effects on the proliferation of the *Lemna minor* while the inhibitory effects of n-hexane and ethyl acetate extracted samples were moderate. The order of the phytotoxic potential of the stem extracted samples were water >butanol >methanol >n-hexane >ethyl acetate. The order of the phytotoxic potential of the roots extracted samples were water >butanol >ethyl acetate >n-hexane >methanol. Rahmaullah *et al*. (2012) and Rauf *et al*. (2012) concluded similar results for different plants while the present results are contradictory to Grisi *et al*. (2013) who reported better phytotoxic activity for hexane and ethyl acetate extracted samples. This may be due to the fact that in the present study non polar to polar solvent extraction procedures was used while they carried out polar to non polar solvent extraction process (Macias, 2010).

The stored grain insects cause serious problems. If the crop is not completely damaged, the nutritional value is adversely affected due to infestation. These pests especially eat the embryos of the grains, thus reducing the protein content of the stored grain and minimize the percentage of seeds to germinate. For this reason novel insecticides which are cost effective and environmentally friendly should be developed to control the pest problems. The insecticidal potential of the stem extracted samples of *Periploca hydaspidis* against *Tribolium castaneum* revealed dose dependent effect (Chen *et al*., 2007; Chu *et al*., 2012). Maximum mortality percentage was shown by water extracted sample at highest concentration compared with other extracts. The activity of the stem butanol and water extracted samples against *Rhizopertha dominica* was also dose dependent as demonstrated for *Tribolium castaneum.* Maximum mortality rate (%) was shown by butanol extracted sample at the highest concentration when compared with other samples. The toxicity of the extracts against *Callosobruchus maculatus* revealed that maximum mortality percent rate was demonstrated by butanol extracted sample at the highest concentration and minimum activity by water extracted sample at 5 mg/ml. Chen *et al*. (2012) demonstrated similar results for *Periploca sepium* who found considerable activity against the test insects. Root extracted samples were also screened for their insecticidal activity against *Tribolium castaneum*. Maximum mortality rate (%) was noted by butanol extracted samples at the highest concentration compared with minimum mortality rate of water extracted samples at the lowest concentration (Chu *et al*., 2012). The data also indicated that maximum mortality rate (%) against *Rhizopertha dominica* was demonstrated by butanol extracted samples at 125 mg/ml compared with the minimum percent mortality rate of water extracted samples at 5 mg/ml. Our results agree with Saleem *et al*. (2004) and Wang *et al*. (2010). The same extracts were also screened for their insecticidal potential against *Callosobruchus maculates.* Our results suggested that maximum mortality rate (%) was demonstrated by butanol extracted sample at the highest concentration when compared with other samples (Rahmanullah *et al*., 2012). It can be concluded from these results that polar solvents like butanol and water can be used for the isolation of insecticidal compounds for the development of commercially available insecticides (Wang *et al*., 2010; Chu *et al*., 2012).

The extensive utilization of plants to treat a wide range of diseases is accredited to the existence of many bioactive compounds (Ayodele, 2003). These bioactive compounds can be derived from any fraction of a plant like root, stem, bark, leaves, flowers, seeds etc (Ara and Nur, 2009).

Flavonoids, phenols and tannins are antioxidants produced by the plants are reported to have many biological (Vinson *et al*., 1995; Tiger, 1980Agbor *et al*., 2004). Tannins are attributed to antibacterial properties and trigger immune response in the body against many parasites (Tiger, 1980). Flavonoids are strong anti-cancerous compounds and have free radical scavenging activity (Okwu, 2006). The alkaloids in many extracts could be allied with the antimicrobial activities (Ramkumar *et al*., 2007). The existence of these compounds in *P. hydasipdis* confirms the medicinal value of the stems and roots of the plant. The results obtained from the qualitative phytochemical analysis of the stem of the plant revealed that alkaloids and corbohydrates were present in all extracts except butanol extracted samples, flavonoids only in methanol and water extracted samples, proteins in all the extracted samples while saponins were present only in butanol and water extracted samples. Qualitative phytochemical screening of the root tissues of the plant indicated that alkaloids and carbohydrates were present in all the extracts except butanol extracted samples, flavonoids in butanol and water extracted samples, proteins in all the extracted samples except methanol extracted samples while saponins were present only in butanol and n-haxnae extracted samples. Tannins were present only in n-hexane extracted samples and sterols were found to be present in methanol, butanol and water extracted samples.

CONCLUSION

It can be concluded from the present study that maximum phytotoxic activity was noted by aqueous extracted samples from stem and root tissues. Crude methanolic and water extracts from both stems and roots tissues showed good insecticidal activities compared with other extracts. The plant contained alkaloids, flavonoids, carbohydrate, proteins and saponins which show the medicinal importance of the tested plant.

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