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Seasonal Dynamics of Spiders and insect Pests in Citrus Orchards of District Sargodha, Pakistan

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Abstract.- Citrus leaf miner, citrus whitefly and citrus psylla are the major insect pests of citrus orchards in district Sargodha, Pakistan. Spiders, being natural predators are helpful to keep the insect pest populations below economic enjoy level. The diversity and guild structure of spiders in the citrus orchards was recorded in the study. Seasonal dynamics of spiders and insect pests and predator-pest (spider-pest) relationships were also studied. In total, 2665 spiders belonging to 12 families, 23 genera and 43 species were captured. Species and family composition of spiders varied on the foliage and ground. Abundance of spiders among sites differed significantly. However, non-significant difference was observed in the richness, diversity and evenness among sites. Abundance and infestation of pest was found to synchronize with the abundance of spiders. Maximum infestation of citrus leaf miner, citrus whitefly and citrus psylla was recorded in March, 2014 supporting maximum spider abundance in this month. There was strong positive correlation between abundance of insect pests and predators (spiders) which suggested that spiders could effectively control insect pest populations in the field.

Key worlds: spiders, leaf miners, whitefly, bio-control.

INTRODUCTION

Sargodha is an important agricultural area of Pakistan, producing about 650,000 metric tons of citrus each year; which is about 23% of Pakistan's total citrus production (http://www.phdeb.org). Citrus psylla *Diaphorina citri* Kuwayama (Hepner, 1993; Sohail *et al*., 2004; Hoddle, 2012), citrus leaf miner *Phyllocnistis citrella* Stainton (Ahmad *et al*., 2013; Kerns *et al*., 2002) and citrus red scale *Aonidiella aurantii* Maskell (Cardwell *et al*., 2006; Reeve and Murdoch, 1985) cause severe damage to the citrus crop, resulting in restricted share of Pakistan in global export market (http://www. tdap.gov.pk/docpdf).

Although traditional pest control, through pesticides use, is effective but causes damage to the population dynamics of useful invertebrates (Pekar, 1998; Tahir *et al*., 2010; Mukhtar *et al*., 2013). Insecticides contain toxic chemicals which have adverse impacts on agro-ecosystem and human health (Bukhari *et al*., 2012). Many pests have developed resistance against pesticides due to their excessive use (Khuhro *et al*., 2012; Rogers and Dewdney, 2012).

Efforts are being made to conserve natural predators in agro-ecosystems by using practices that do not damage population of natural predators (Sunderland and Samu, 2000). Several techniques like reduce tillage practice (Tahir *et al*., 2012), organic farming, mulching, intercropping (Sunderland and Samu, 2000) and preservation of over-wintering sites (Landis *et al*., 2000) have been used to increase the diversity and abundance of natural predators in agro-ecosystems. Use of natural predators as biological control agents is a part of integrated pest management technique (Amalin *et al*., 2001). Natural predators like ladybeetles (Michaud *et al*., 2002), lacewing *Chrysoperla carnea* (Rosenheim *et al*., 1993) and spiders (Nyffeler and Benz, 1987; Sunderland and Samu, 2000; Tahir and Butt, 2009) are effective bio-control agents for several pests in agro-ecosystems (Hodge, 1999; Ghavami, 2008).

Spider (Arachnidae: Aranae) is a most abundant, diverse and ecologically important group in many cropping systems (Hodge, 1999; Sunderland and Samu, 2000; Tahir *et al*., 2012).

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Hitherto 112 families, 3924 genera and 44540 species of spiders are described in the world (Platnick, 2014). They live in a variety of habitat like forest, fields, foliage, ground, and even some may live amphibious life (Bukhari *et al*., 2012). They have great importance in reducing and even preventing outbreaks of insect pests in agricultural fields (Sunderland *et al*., 1986). They are capable of limiting and equilibrating pest populations (Nyffeler, 1999; Sunderland and Samu, 2000), because of their abundance and high predatory potential (Tahir and Butt, 2009), prey specialization, polyphagy, top-down effect, numerical and functional responses, and wasteful killing (Maloney *et al*., 2003).

Although the number of prey in the field fluctuate with season, time of day, microhabitat and foraging strategy (Uetz, 1991), but different spiders residing same field may exploit same resources (Hodge, 1999). Hunting and web-weavers are the two major clustors of spiders (Uetz 1977; Nyffeler 1982; Uetz *et al*., 1999) which are further divided into six to eight guilds (*i.e*., foliage runners, ground runners, stalkers, ambushers, sheet web weavers, wandering sheet weaver, orb weaver and space web builders) based on web use, web type and microhabitat (Uetz *et al*., 1999).

Most of the work done on spiders of Pakistan is restricted to taxonomy. Studies on agro-ecology and bio-control potential of spiders started in early 2000’s. In rice fields, Tahir and Butt (2009) estimated 33.33% damage to pest population by just three species of spiders *i.e*., *Lycosa terrestris*, *Pardosa birmanica* and *Oxyopes javanus*. Additionally, many other predators may have reducing effects on pest population. Mohsin *et al*. (2010) studied spider guild structure of oilseed crops in Pakistan. Mostly experiments revealing pest control properties of spiders are performed under laboratory conditions (Sunderland and Samu, 2000; Tahir and Butt, 2009; Khuhro *et al*., 2012). But there is a need to explore interaction between spider and pest in fields conditions before recommending spiders for pest control (Harwood *et al*., 2001).

Keeping in view the negative impacts of non-selective insecticides on beneficial arthropods, there is utmost need of using some eco-friendlier agricultural practices and reducing agrochemical applications. There is a serious need to explore spider fauna of Pakistan for its implementation in Integrated Pest Management programme. Unfortunately, this group of great ecological significance as biological control agent is overlooked in Pakistan (Tahir *et al*., 2011). Present study was aimed at to record the spider diversity in citrus orchards and to study the relationship of population dynamics of spiders and their major insect pests. We also studied the guild structure of spiders in citrus orchards.

MATERIALS AND METHODS

Study areas

Study was conducted in citrus orchard at Chak No. 87 N.B Tehsil, Sargodha (32° 5′ 1″ N, 72° 40′ 16″ E). Four fields of one acre each, located almost two hundred meter apart, were randomly selected for the study. All fields were under regular agricultural practices. Citrus orchards were present on three sides of field I, while in North fodder barseem was cultivated. Field II was surrounded by citrus from two sides whereas; wheat was cultivated on East and maize on Southern side. Field III was surrounded by citrus from North and West and by wheat on East side.

Fodder barseem was grown as refuge crop in field I from November to March. Field was irrigated almost monthly throughout the year and ploughed at regular intervals. Insecticide Talstar was sprayed in April, 2013. Fertilizer nitrophos was applied in October 2013.

Field II was regularly ploughed and irrigated almost every 15-20 days throughout the year. Brassica and fodder barseem were cultivated from August, 2013 through January, 2014. Insecticide Talstar and Copper Oxychloride were applied in April, 2013 and October, 2013 for the control of citrus insect pests. Fertilizers nitrophos and ammonium nitrate were applied in April 2013.

Wheat was cultivated in field III in October 2013. Insecticide Talstar and copper oxychloride and fertilizers *i.e*., nitrophos and ammonium nitrate, were applied in April, 2013. Moreover, fertilizers diammonium phosphate and urea was applied in October, 2013 in field III for newly sown wheat crop. However, ploughing and irrigation practices were on regular intervals throughout the year except during the month of December.

No refuge crop was cultivated in field IV throughout the year. Insecticide Talstar was sprayed in April, 2013. Nitrophos was applied in October, 2013 in field whereas fertilizer potash was applied in March, 2013.

Sampling

Sampling was carried out from ground and foliage on monthly basis from April, 2013 to March, 2014. During sampling, information about environmental factors (temperature, rainfall and relative humidity) and agricultural practices (*i.e.*,, irrigation, ploughing, tillage and insecticide spray) was noted. According to Tahir and Butt (2009b) spiders were regarded as most dominant if, constituted more than 10% of the total collection, dominant if more than 1% and rare if less than 5 in number.

To capture the ground active spiders, pitfall traps were used. Wide mouth glass jars of 12 cm depth and 6 cm width were used as pitfall jars. For sampling, the jars were buried in the soil so that their rims were at level with the ground. At each site, traps were set into 3x3 m grid pattern at ten localities (4 grids in the center of the field and 6 grids at the margins of the fields). Each grid was consisted of four traps. One hundred and fifty ml of 70% alcohol and few drops of 5% liquid detergent were added to each jar. At each location traps were placed for 5 consecutive days (while changing the chemicals of traps after every 70-72 h).

Foliage spiders were collected by jarring and hand picking methods. For jarring, 4 plants were randomly selected from all sides of each field. Four branches of each tree were selected randomly. A white cloth of approximately 2.5x1.5 m was spread under the tree before jerking. Selected branches were jerked for three times for 5-10 seconds. Spiders dropped during jerking from branches were collected from the white cloth and placed individually in the glass vials.

Pest record

Data for citrus pests *i.e.*, citrus leaf miner, citrus whitefly and citrus psylla was recorded from April, 2013 to March, 2014. For the observation of citrus pest infestation, five plants were randomly selected from each field.

To record citrus leaf miner four branches from all sides of each selected plant were selected to record the infestation rate at shoot apex. Hundred leaves were selected from apex for data record.

To record citrus psylla and citrus whitefly five leaves from each plant were randomly selected for recording the abundance of citrus whitefly and citrus psylla. Total number of pest present on both sides of leaves was counted.

Collected animals were brought to the laboratory, washed with alcohol and preserved in absolute alcohol with the proper labeling of collection site, date, and scientific names. Collected specimens were identified to the lowest possible rank with the help of available keys and catalogue available *i.e.*, Dyal (1935), Tikader and Malhotra (1980), Tikader (1982), Tikader and Biswas (1981), Biswas and Biswas (1992), Gajbe (2008), Barion and Listinger (1995), Changmin *et al.* (1997), Platnick (2014) and other available literature.

Data analyses

Diversity, richness and evenness of the data were computed using widely used indices *i.e.*, Shannon-Weiner index (H'), Simpson's index (λ), Margalef index and modified Hill's ratio (E5). One way-ANOVA followed by Tukey's test was applied to compare the abundance, richness, evenness and diversity among sites and trapping sessions by using SPSS Software (version 13.0).

RESULTS

In total 2665 spiders belonging to 12 families, 23 genera and 43 species were sampled. Of total catch, 1395 spiders were collected from foliage and 1270 from ground (Table I). Relative abundance of spiders varied per trapping session. Highest number of spiders was collected in March, 2014. However, in February number of individuals was less as depicted in Figure 1. Overall, the most abundant family recorded from citrus orchards was Salticidae (29.49%), followed by Lycosidae (21.27%) and Araneidae (20.07%). These three families constituted 70.84% of total collection. Remaining 29.16% was represented by nine families *i.e.*, Clubionidae, Miturgidae, Gnaphosidae, Hersiliidae, Thomisidae, Oxyopidae, Tetragnathidae, Linyphiidae and Sparassidae (Fig. 2).

Table I.- Abundance of spiders on foliage and ground of citrus orchards.

Family and species name	Foliage	Ground
Araneidae		
Araneus elliptica (Tikader & Bal, 1981)	16	-
Araneus mitificus (Simon, 1886)	47	-
Cyrtophora citricola (Forsskal, 1775)	32	-
Eriovixia excelsa (Simon , 1889)	184	-
Neoscona bengalensis (Tikader & Bal, 1981)	21	-
Neoscona mukerji (Tikader, 1980)	120	40
Neoscona rumpfi (Thorell, 1878)	14	-
Neoscona theisi (Guerin, 1838)	40	21
Clubionidae		
Clubiona drassodes (Cambridge, 1874)	70	84
Clubiona analis (Thorell, 1895)	16	-
Gnaphosidae		
Gnaphosa jodhpurensis (Tikader & Gajbe, 1977)	-	71
Gnaphosa sp.	-	12
Zelotes sp.	-	11
Hersiliidae		
Hersilia savignyi	3	-
Linyphiidae		
Linyphiidae sp. 1	25	17
Linyphiidae sp. 2	-	14
Lycosidae		
Hippasa mudhae (Tikader & Malhotra, 1980)	-	25
Lycosa maculata (Butt, Anwar & Tahir, 2006)	-	38
Lycosa madani Pocock	-	27
Lycosa terrestris (Butt, Anwar & Tahir, 2006)	-	71
Pardosa birmanica (Simon, 1884)	-	205
Pardosa oakleyi (Gravely, 1924)	-	39
Pardosa sumatrana (Thorell, 1890)	-	148
Evippa sp.	-	14
Miturgidae		
Cheiracanthium inclusum (Hentz, 1847)	126	-
Cheiracanthium sp.	14	-
Oxyopidae		
Oxyopes javanus (Thorell, 1887)	19	72
Oxyopes sp.	8	-
Salticidae		
Marpissa tigrina (Tikader, 1965)	6	16
Myrmarachne maratha (Tikader, 1973)	39	64
Phintella versicolor (C.L. Koch 1846)	51	-
Phintela vittata (C.L.Koch, 1846)	107	-
Plexippus paykulli (Audouin, 1826)	19	11
Rhene indica (Tikader, 1973)	5	-
Telamonia dimidiata (Simon, 1899)	11	-
Thyene imperialis (Rossi, 1846)	177	106
Epeus sp.	39	-
Salticidae sp.	54	81
Sparassidae		
Olios mahabangkawitus (Barrion & Listinger, 1995)	19	-
Tetragnathidae		
Leucauge decorata (Blackwel, 1864)	16	-
Tetragnatha javana (Thorell, 1890)	51	-
Thomisidae		
Thomisus pugilis (Stolickza, 1869)	18	28
Thomisus spectabilis (Doleschall, 1859)	28	55
Total	**1395**	**1270**

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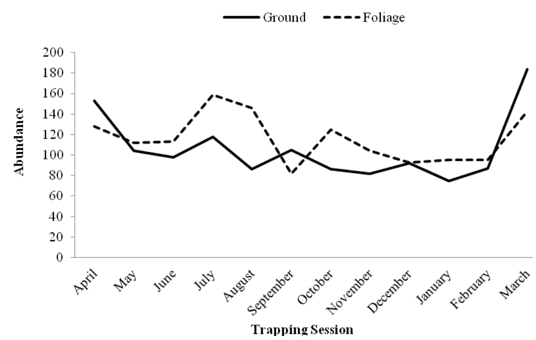

Fig. 1. Seasonal dynamics of spiders collected from ground and foliage from April, 2013 to March, 2014.

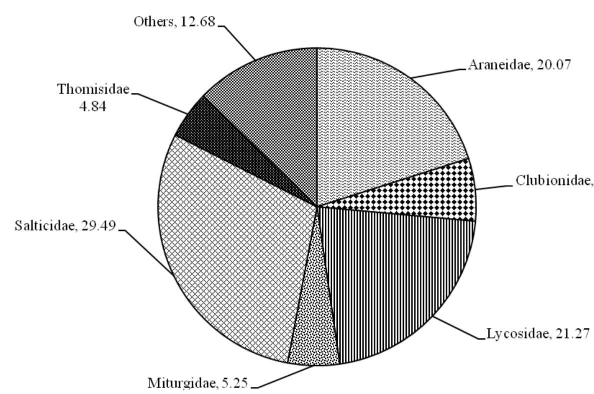

Fig. 2. Percent composition of all families collected from all sampling sites of citrus orchards.

*Thyene imperialis (*Rossi, 1846) was the most dominant species (10.61%) followed by dominant species, *Pardosa birmanica* (7.69%), *Eriovixia excelsa* (6.90%), *Neoscona mukerjei* (6%) and

Clubiona drassodes (5.77%). These five species contributed 36.97%, whereas, the remaining thirty eight species comprised 63.03% of the total sample. Abundance of these species varied on foliage and ground. There were nineteen species present only on foliage and not on ground. Similarly twelve species, present on ground and were not represented on foliage. Collected specimen represented every life stage of spiders. Of all the specimen adult male were 25.92%, females 31.21% and 42.85% were juvenile. However, their relative abundance varied per trapping session (Fig. 3).

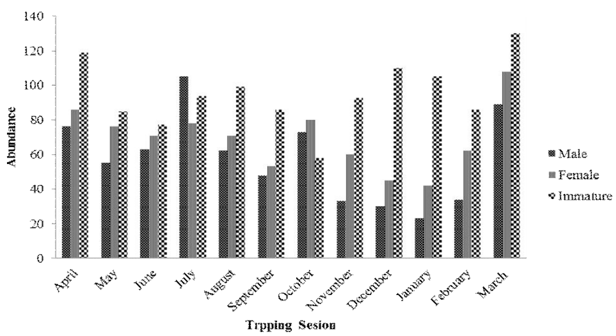

Fig. 3. Abundance of adult males, females and immature during different trapping sessions.

Spiders collected from ground and foliage of all sampling sites were divided into different guilds according to Uetz *et al.* (1999). Spider guilds included stalkers, foliage runners, ground runners, orb web weavers, sheet web weavers and ambushers. Overall, most dominant spider guild was of stalkers (33.2%), including Salticidae and Oxyopidae. Second most dominant guild was ground runners (24.8%) (Lycosidae, Gnaphosidae). Family Araneidae and Tetragnathidae of orb-web weavers constituted third most dominant guild (22.58%). Other guilds included foliage runner 7% (Clubionidae, Sparassidae), ambushers 4.8% (Thomisidae) and sheet web weavers 2.1% (Linyphiidae). However, a slight difference in guild structure of different sampling sites was observed. Generally stalkers were dominant taxa in all sites followed by ground runners and web-weavers but their percentages varied at each site (Fig. 4).

Results of analysis of variance (ANOVA) showed that four fields differed significantly for the abundance of spiders. However, there was no difference in the richness, diversity and evenness of spiders (Table II). Figure 1 is representing the seasonal dynamics of ground and foliage spiders during various trapping sessions. Highest number of spiders was recorded during the month of March, 2014. Spider abundance was found to be positively correlated with temperature (Pearson correlation= 0.91; $P < 0.001$). However, negative correlation was observed between spider abundance and rainfall (Pearson correlation = -0.43; $P = 0.211$)

Table II.- Results of Tukey's test showing abundance, richness, diversity and evenness among studied fields.

Sampling sites	Abundance	Richness	Diversity	Evenness
Field I	521^{a}	5.53^{a}	4.62^{a}	0.85^{a}
Field II	898^{d}	5.94^{a}	4.32^{a}	0.81^{a}
Field III	592^{b}	5.99^{a}	4.31^{a}	0.83^{a}
Field IV	654^{c}	5.90^{a}	4.09^{a}	0.88^{a}

Note: Values in the columns having different superscripts are significantly different.

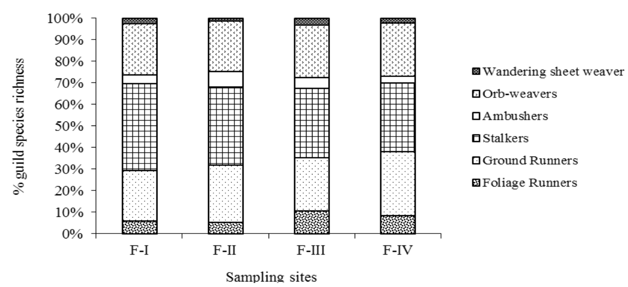

Fig. 4: Guild structure of spiders in citrus orchards.

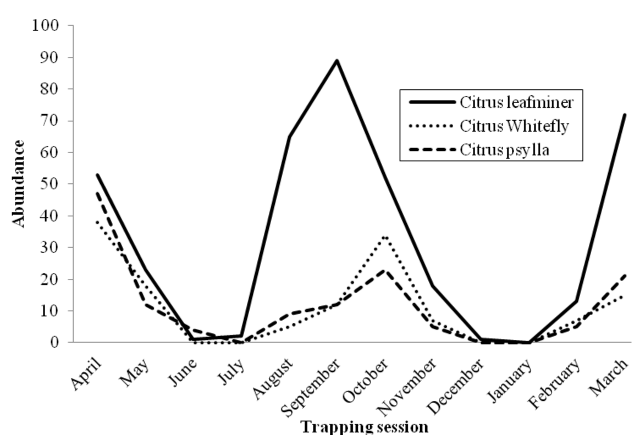

Fig. 5. Seasonal fluctuations of citrus leaf miner, citrus whitefly and citrus psylla.

Table III.- Results of Tukey's test showing abundance, richness, diversity and evenness among trapping sessions from April, 2013 to March, 2014.

Trapping session	Abundance	Richness	Diversity	Evenness
April	281 [c]	6.55 [c]	4.45 [c]	1.01 [b]
May	216 [b]	6.18 [c]	4.00 [b]	0.99 [b]
June	211 [b]	5.86 [b]	3.60 [a]	1.02 [b]
July	277 [c]	6.41 [c]	3.71 [a]	0.84 [a]
August	232 [b]	5.84 [b]	4.65 [c]	0.86 [a]
September	187 [a]	5.57 [a]	3.52 [a]	0.88 [a]
October	211 [b]	5.93 [b]	4.72 [a]	1.01 [b]
November	186 [a]	5.27 [a]	4.32 [b]	1.06 [b]
December	185 [a]	6.32 [c]	4.14 [b]	1.02 [b]
January	170 [a]	6.14 [c]	3.94 [b]	1.05 [b]
February	182 [a]	6.19 [c]	3.59 [a]	1.04 [b]
March	327 [d]	6.63 [d]	5.62 [d]	0.92 [a]

Note: Values in the columns having different superscripts are significantly different.

Abundance of citrus leaf miner, citrus whitefly and citrus psylla revealed variation per trapping session. Citrus leaf miner was most devastating pest and was found almost throughout the year. It showed population peaks twice a year, once from August to October and later from March to May. In present study maximum leaf miner infestation was recorded during September, 2013. Citrus psylla showed maximum infestation in April and later in September and October. Maximum infestation was measured during April, 2013 followed by October. Citrus whitefly was also most abundant during the month of April and October. Figure 5 gives an account on relative abundance of various insect pests of citrus per trapping session.

DISCUSSION

Present study resulted in 43 species, 23 genera and 12 families from citrus orchards. Same number of species was previously reported by Vetter *et al.* (2013) from citrus orchards of Faisalabad. Moreover, they reported 38 genera and 17 families. Population dynamics of spiders varied per trapping session both on foliage and ground. An obvious decline was observed in the abundance of prey and predators after insecticide spray in the field. Spider catch was high during the month of April. Whereas, abundance of spiders dropped in successive trapping sessions *i.e.*, May and June. This might be due to application of insecticide (Talstar and copper oxychloride) and fertilizers *i.e.*, nitrophos and ammonium nitrate, at the end of April but before next trapping session. Application of fertilizers (diammonium phosphate, nitrophos and urea), and insecticides *i.e.*, Talstar and copper oxychloride at the end of October might have played a role in the reduction of predator and pest population in successive trapping sessions (Rodrigues *et al.*, 2013). Field management practices such as insecticide application cause disturbance in the field resulting in less number of spiders. Almost same influence of specific insecticides on spider abundance have been reported in pear orchards of Czech Republic (Pekar, 1998) and guava orchards of Lahore (Tahir *et al.*, 2010). Fluctuations in predator and pest abundance per trapping session might also be due to seasonal variations, natural disasters and instabilities in environmental factors like temperature, humidity and rainfall (Mahalakshmi and Jeyaparvathi, 2014).

It was not surprising that there was positive correlation between spider abundance and temperature. Increase in temperature and humidity favors spider population (Ghafoor and Mahmood, 2011). High abundance of spiders during the months of March and April might also because pest flush hit the highest point during these months resulting in availability of more food for predators (Vetter *et al.*, 2013). Minimum spider catch during the month of January might be owing to lowest average temperature in this month resulting in limited food resources as compared to other trapping sessions (Marchetti, 2014). Peak abundance of spiders in various sessions appeared to be synchronous with the time when there was maximum pest infestation. Spider abundance was also influenced by rainfall, as it disturbs spider habitat structure causing less spider catch. Among citrus pests, maximum infestation by citrus psyllid was observed during the month of March, April and October. Citrus psylla also showed maximum infestation in April, 2013. These findings are in accordance with experimental results of Vetter *et al.* (2013). Whereas, citrus leaf miner was present almost round the year. But peaked populations were observed during March to May and then again from August to October. Populations of citrus pest were also affected by seasonal variations. Summer and spring are most favorable season to support higher leaf miner populations, because more leaf flush and younger shoots are available (Pena *et al.,* 1996; Wang *et al.,* 1999; Diez *et al.,* 2006). Higher population of citrus leaf miner was during September and March in the present study might also promote spider growth in respective months by providing more food resources (Maloney *et al.,* 2003). A yellow sac spider *Cheiracanthium inclusum* is found to be important predator of citrus leaf miner (Amalin *et al.,* 2003). In accordance to this, *C. inclusum* was found second most abundant species in April, 2013; when there was maximum leaf miner infestation.

Spiders usually occur in the form of spider assemblages in agro-ecosystems; by this, they can utilize shared resources efficiently (Uetz *et al.,* 1999). Moreover, agricultural diversification results in complexity in habitat structure and provides more abundant and diverse resources (Sunderland and Samu, 2000). Availability of resources results in an increased growth and abundance of spiders (Wardle, 1995). This phenomenon of spider assemblages and resource sharing raises the guild concept. Cultivation of various crops such as wheat, maize and barseem in addition to citrus caused agricultural diversification in sampling sites. This might have supported occurrence of several guilds *i.e.*, stalkers, ground runners, orb-weavers, foliage runners, ambushers and wandering sheet-weavers in the study area.

Salticidae was the most dominant spider taxa on foliage reported during present study. *T. imperialis* (10.61 %) was overall most dominant species, followed by *P. birmanica* (7.69%), *E. excelsa* (6.90 %), *N. mukerjei* (6%) and *C. drassodes* (5.77%). Similar results were reported by Tahir and Butt (2009). These findings are also in accordance with the foregoing studies of Vetter *et al*. (2013). This similarity in results might be due to the fact that the two sampling sites are closely located.

Present experiment yielded less number of ground dwelling species of citrus than previously reported by Tahir *et al.* (2011) which may be because of different climatic conditions and crop culture. However, Lycosidae was found most abundant family on ground as previously reported by various researchers. Monzo *et al.* (2009) and Tahir *et al.* (2011) reported Lycosidae as most dominant family in citrus orchards. Species and family composition of study area was different from that of Tahir *et al.* (2011) as they reported 38 species from citrus. Non-significant difference was observed for diversity and richness values per trapping session in all sampling sites for ground. This might be due to the fact that sampling sites were located in close proximity (almost 200 meter apart from each other).

More specimens were captured from foliage than ground. Variation in the species samples depends upon sampling techniques. Spiders from foliage were collected by jarring method, hand-picking and sometime by tap sampling method. Use of multiple sampling techniques may disturb spider's habitat and provide chance for capturing various species of different habits and habitat (Vetter *et al.,* 2013).

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