



# Influence of Cultivar on Aphids (Hemiptera: Aphididae) and Associated Natural Enemies in Pakistani Wheat Ecosystems

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## ABSTRACT

The objective of this study was to determine the influence of cultivar on aphids and their natural enemies in wheat (*Triticum aestivum* L.). Thus, twenty Pakistani wheat cultivars were grown at the Agricultural Research Institute in Tarnab-Peshawar, Pakistan during the winter ('rabi' in local language) season. Plots were arranged in a randomized complete block design with three replicates per treatment/cultivar. Population of aphids, and numbers of parasitoids and predators were recorded at weekly intervals. *Rhopalosiphum padi* Linnaeus, *Schizaphis graminum* Rondani, and *Sitobion avenae* Fabricius were the predominant aphid species (Homoptera: Aphididae). Two species of parasitoids *Aphidius ervi* L. and *Aphidius colemani* Viereck were recorded. *Coccinella septempunctata* L. (Coleoptera: Coccinellidae), *Chrysoperla carnea* Stephens (Neuroptera: Chrysomelidae), and several species of Hover flies (Diptera: Syrphidae) were the most common predators. In general, aphids were reported in all wheat cultivars. Cultivar Saleem-2000 presented the highest number of aphids per tiller (9.1), while Gomal-2008 presented the lowest (1.9). Based on aphid presence per tiller, no cultivar was found to be Susceptible or Highly Susceptible, but 1 cultivar was found to be Moderate Susceptible, 8 Moderate Resistant, 9 Resistant, and 2 Immune. Rate of parasitism and predation increased when aphid density increased. In general, wheat cultivar influenced aphid population pressure, and indirectly influenced the natural population of parasitoids and predators.

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## Authors' Contribution

QZ designed, conducted the research and recorded the data. HB, RAS helped in data compilation. AK helped in data analysis. SIR helped in manuscript preparation and editing.

## Key words

Wheat, Aphids, Parasitoids, Predators, Wheat Aphids, Resistance, Ecosystems, Control

## INTRODUCTION

Wheat, *Triticum aestivum* Linnaeus (Poales: Family Poaceae), is a major staple food crop worldwide. In Pakistan, wheat is grown as an annual crop and is an important nutritional source in the Pakistani diet (Naz and Akmal, 2016). Wheat contributes about 96% of the total value of the country's agricultural sector, which represents 1.9% of the total Pakistani economy (Pakistan Economic Survey, 2017). In the 2016-17 growing season, wheat was sown on 9,052 thousand hectares, producing 26 million tons with an average yield of 2,845 kg ha<sup>-1</sup> (Pakistan Economic Survey, 2017). Depending on the region, there are two main crop-growing seasons in Pakistan. During winter or 'rabi' (1), in local language, crops like wheat, barley (*Hordeum vulgare* L.), chick pea (*Cicer arietinum* L.), and lentils (*Lens culinaris* Medikus) are grown from Oct to Dec, and harvested during Mar to May; summer or 'kharif' (2), were maize (*Zea mays* L.), rice (*Oryza sativa* L.), sorghum (*Sorghum bicolor* Moench), and millet

(*Panicum* spp.) are sown from Mar to Jul, and harvested around late Sept (Hatam, 1994). Both field seasons allow the presence of green crops that serve as host to many pest species.

Several arthropod species reduce wheat productivity in Pakistan. Species of aphids are present in wheat, and cause direct or indirect damage, by feeding or vectoring plant pathogens, respectively (Chapin *et al.*, 2001; Steffey and Gray, 2012). Aphid infestations have considerably affected productivity and profitability by reducing shoot growth, lowering chlorophyll concentrations, and overall root growth (Reidell and Kieckhefer, 1995). Different aphid species like *Rhopalosiphum maidis* Fitch, *R. padi* L., *Schizaphis graminum* Rondani, *Sitobion avenae* Fabricius, *Sipha maydis* Passerini, *Macrosiphum avenae* Fabricius, *Aphis nerii* Boyer de Fonscolombe, *Myzus persicae* Sulzer, *Brevicoryne brassicae* L., *Macrosiphum granarium* Kirby, and *R. rufiabdominalis* Sasaki prevail on wheat in Pakistan (Hamid, 1983; Leather *et al.*, 1989; Irshad, 2001; Mushtaq *et al.*, 2013). In the Peshawar valley of the Khyber Pakhtunkhwa province, *S. graminum*, *R. padi*, and *S. avenae* are dominant species (Hashmi *et al.*, 1983; Hachett *et al.*, 1987; Ali *et al.*, 2015).

In Pakistan, aphids have mostly been controlled

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by insecticide application (Irshad, 2001). However, we recognize that excessive pesticide use may cause pest-resurgence, insect resistance, and disturb the natural balance of pest and natural enemies; they can also create human health problems and environmental pollution (Zacharia, 2011). In addition, chemical control can cause adverse effects on non-target organisms such as birds, fish, and other aquatic life; insecticides can also reduce the number of pollinators and adversely affect the soil microorganism composition that are involved in nutrient recycling (Iyaniwura, 1991; Baskar *et al.*, 2017). Due to recent outbreaks of aphids in Pakistan, producers have heavily relied on the use of pesticides (Punjab Agriculture Research Board, 2011; Wains *et al.*, 2014). Until recently, aphids were not a threat to wheat and populations were under control potentially by natural enemies (Hamid, 1983). Several botanical insecticides derived from oils coming from leaf and seeds have been used to control aphids in Pakistan (Iqbal *et al.*, 2011). A few examples include neem *Azadirachta indica* Juss, sweet orange *Citrus sinensis* Osbeck, balsam pear *Momordica dioica* Roxb. ex Willd, wild garlic *Allium vineale* L., chili pepper *Capsicum frutescens* L., *Moringa oleifera* Lam, and tobacco *Nicotiana tabacum* L. Neonicotinoid insecticides have been widely used in the region including several formulations such as thiamethoxam 25GW, imidacloprid 20% EC, and acetamiprid 20SP (Iqbal *et al.*, 2011; Zeb *et al.*, 2016; Shah *et al.*, 2017).

Host plant resistance along with natural enemies and cultural control provide an effective, environmentally friendly, and economical aphid control strategy. The premise is the alteration of pest behavior on insect host selection, oviposition and feeding, reduce insect survival and development, and recovery from injury (Smith, 1989). The general thought is that the use of plants with resistance traits will help reduce insecticide applications, and aid in the increase of biological control activity (Sharma and Ortiz, 2002). Certain host genotypes are known to affect performance of herbivores and community structure including predators and parasitoids (Kennedy, 2003; Schadler *et al.*, 2010). For instance, quack grass, *Elytrigia repens* L., affects the interaction of *R. padi*, the parasitoid wasp *Aphidius colemani* Viereck, and the predatory lacewing *Chrysoperla carnea* Stephens (Schadler *et al.*, 2010). Schadler *et al.* (2010) reported that the number of *R. padi* offspring's, and presence of its parasitoids differed considerably when exposed to different plant genotypes and the behavior was related to the nutritional contribution of each plant type which influence natural enemies' preference. Effects of any particular plant defense trait on parasitoids and predators depends on the specific attributes of the plant trait, and the detail of

the physical, biochemical, and behavioral interaction between natural enemies, its host (prey), and the plant (Kennedy, 2003). Givovich and Niemeyer (1995) and Eleftherianos *et al.* (2006) indicated that the presence of chemical compounds such as phenolics and hydroxamic acids, are involved in wheat plants resistance against *R. padi* and *S. avenae*. More specifically, aphids feeding caused reduction in the percentage of epicuticular wax, dry weight, sugar, amino acid level and slows the uptake of minerals like P, K, Ca, Mg, and Fe. In cabbage, ascorbic acid, proline, phenol peroxidases, oxidases, and minerals like Ca and K play a role against aphids (Khatab, 2007).  $\alpha$ -Tomatine, an alkaloid in tomato plants, is indirectly toxic to endoparasite of lepidopterous pest in tomatoes, since parasitoids acquire the alkaloid while parasitizing the pest who previously fed on the alkaloid (Campbell and Duffy, 1979). Leaf pubescence also could be one of the resistance factors against various wheat aphid species (Gholami *et al.*, 2013); for example, wheat cultivars with high trichome density reduced *D. noxia* incidence (Bahlmann *et al.*, 2003). Similarly, ecological aspects can be affected; such as the searching ability of *Encarsia formosa* Gahan, a parasitoid of whiteflies, whose numbers increase three-fold on hairless cucumber compared to hairy varieties (Van Emden, 1986). The searching and walking speed of *Trichogramma exiguum* L., egg parasitoids of *Heliothis zea* Boddie, are influenced by differences in leaf structure, particularly trichome form and density (Keller, 1987).

Because screening and identification of resistant germplasm are fundamental for long-term sustainable production, the impact of several Pakistani wheat cultivars on aphid species populations and their associated natural enemies' populations was studied in the Peshawar valley of the Khyber Pakhtunkhwa province in Pakistan. Natural enemies were included in our study since their impact in our region is under study (Adisu *et al.*, 2002).

## MATERIALS AND METHODS

The experiment was conducted at the Agricultural Research Institute (ARI) Tarnab-Peshawar in Pakistan (34.0040° N, 71.6970° E). Twenty wheat cultivars (n=20) were planted in a randomized complete block with three replicates per treatment/cultivar (Table I). Each plot was 2.5 m wide  $\times$  2 m in length with a buffer area of 1 m between plots. Approximately 56.3 g of seed per variety per plot was sown by using a locally made hand manual single line seed drill hoe. Standard 30-cm row spacing, and general local agronomic practices were followed. Fertilizers at the rate of 120 kg of N, and 90 kg of P ha<sup>-1</sup> came from urea and diammonium phosphate (DAP). Half of the N, and full dose of K were applied at soil preparation, while the

**Table I. Parentage and pedigree of wheat varieties used on this experiment.**

Varieties	Parentage	Pedigree
Attahabib-2010	INQALAB 91*2/TUKURU	CGSS99B00015F-099Y-099M-099Y-099M-31Y-0B
Bakhtawar-92	JUP/BJY//URES or JUP/BJYG//URES	CM67458-4Y-1M-3Y-1M-5Y-0B
Bathoor-2008	URES/JUN//KAUZ	CM96818-1-0Y-0M-0B-2Y-2Y-0M
Barsat-2010	FRET2	CGSS96Y00146T-099B-099Y-099B-16Y-0B-0SY
Dera-98	F12-71/COC/CNO 79	CM76688-9Y-03M-02Y-2B-0Y
Daman-98	BOWS/3/CAR853/COC//VEES	CP02274-4C-0C-0Y-5M-ORES
Fakhr-e-Sarhad	PFAU'S/SERI//BOW'S'	CM85295-0101TOPY-2M-0Y-0M-3Y-0M
Gomal-2008	Atila	CM85836-4Y-0M-0Y-14M-0Y-5M-0Y-15J-0Y-0AP
Hashim-2008	JUP/ALD'S//KLT'S/3/VEE'S/6/BEZ//TOB/8156/4/ ON/3/6*TH/KF//6*LEE/KF/5	ICW91-0321-2AP-0TS-1AP-2AP-0L-0AP
Khyber-87	KAVKAZ/TORIM-73//POTAM-70/ANAHUAC-75	CM43903-H-4Y-1M-1Y-3M-2Y-0B
KT-2000	GEN#WHETON	SWMI11508-LAP-1AP-1AP-4AP-1AP-5AP=0AP
Pirsabak-2004	KAUZ/STAR	CMBW90Y3058-74M-015Y-015M-1Y-0B
Pirsabak-2008	KAUZ/PASTOR	CMSS93B00025S-48Y-010M-010Y-4Y-0M
Pirsabak-2005	MUNIA/CHTO/AMSEL	CMSS93B00729S-23Y-010M-010Y-010M-7Y-1M-0Y
Saleem-2000	CHAM-6/KITE/PAPAGO-86	ICW93-0032-7F-0K-0F.
Sehar-2006	CHILEROL/2* STAR/4/BOBWHITE//BUCKBUK/ PAVON-76/3/2*VEERY-10	CMSS95Y00645-100Y-200M-17Y-10M-0Y-0PAK
Siran-2010	PBW343*2/KUKUN	CGSS99B00041F-099Y-099M-099Y-099M-34Y-0B
Tatara-97	JUPATECO-73(SIB)ALONDRA//(SIB)KINGLET/3/ VEERY-S	CM79510-024Y-2M-05Y-01M-1Y-0B
Uqab-2000	CROW'S/NAC//BOW'S'	PB 222138-3A-0A-0A-231A-0A
Zam-2004	KAUZ* 2/OPATA//KAUZ	

Source of origin of the used wheat varieties is CIMMYT/ICARDA; This information was obtained from the Cereal Crop Research Institute (CCRI), Pirsabak, Nowshera-Kyber Pakhtunkhwa.

remaining half N was applied with first irrigation 3-wks after sowing (Inamullah *et al.*, 2011).

#### *Sampling aphids and natural enemies*

After seed germination until crop maturity, 9 tillers per plot were regularly inspected at weekly interval, and number of aphid species (alate and apterous), aphid mummies, and predators per tiller were counted. Aphids were collected from wheat plants using a fine camel hairbrush; samples were placed in vials in 80% ethyl alcohol, and transported to the laboratory for identification. Permanent slides of a sub-sample of the aphids collected were prepared in Hoyer's medium (Manya, 1987). Aphids were identified using morphological characteristics according to Blackman and Eastop (1984, 2000) and Manya (1987) identification keys. Percent parasitism was calculated by the following formula: [No. mummified aphid/ (No. mummified aphid + No. non-mummified aphid)] $\times$ 100 (Shahid *et al.*, 2012). For the identification of

the parasitoids, a sub-sample of mummies collected were placed in petri dishes (10 cm  $\times$  1.5 cm) in the laboratory at a temperature of 20  $\pm$  5°C, 50-65% relative humidity, and a photoperiod of 16:8 h D: L until hatching. Adult parasitoids were identified using Tobias (1995) identification keys. Also, most common predators groups were identified.

#### *Rating host plant resistance*

Aphid data were classified in groups and analyzed as early-season (last week of Nov to Jan), mid-season (Feb to mid-Mar), and late-season populations (mid-Mar to first week of Apr). Aphids' parasitism was recorded weekly and data were analyzed per season as described above. This procedure of classification of data into group was adopted from Ronquim *et al.* (2004), Muhammad *et al.* (2013) and Wains *et al.* (2014). The following modifications were made: Immunity (I), 1-2 aphid tiller<sup>-1</sup>; Resistance (R), 3-4 aphid tiller<sup>-1</sup>; Moderately Resistant (MR), 5-6 aphid tiller<sup>-1</sup>; Tolerant (T), 7-8 aphid tiller<sup>-1</sup>; Moderately Susceptible

**Table II.** Mean number of aphids per tiller per variety during early, mid and late season in Agriculture Research Institute, Tarnab-Peshawar, Pakistan.

Wheat cultivars	Early (Nov to Jan)	Mid (Feb to mid-Mar)	Late (Mid-Mar to first week of Apr)	Seasonal mean	Rating
Saleem-2000	7.80 a	13.00 a	5.51 a	9.11a	MS
Tatara-97	4.00 b	9.36 cd	4.54 b	5.85b	MR
Zam-2004	4.16 b	10.20 b	2.51 e	5.81b	
FakhreSarhad-99	4.00 b	10.42 b	2.30 efgh	5.77bc	
KT-2000	4.06 b	9.40 c	2.31 efgh	5.47bcd	
Barsat-2010	3.41 c	8.92 cd	2.31 efgh	4.98bcde	
Dera-98	3.28 cd	9.12 cd	2.0 gh	4.93 bcde	
Attahabib-2010	2.93 d	8.72 de	3.04 cd	4.78cdef	
Daman-98	3.27 cd	8.21 ef	1.24 j	4.52def	
Bakhtawar-92	2.23 e	7.64 f	4.30 b	4.27efg	R
Khyber-87	2.01 ef	7.97 f	2.93 d	4.05efgh	
Seher-2006	3.00 cd	6.48 gh	1.33 j	3.82fgh	
Pirsabak-2008	1.59 fg	7.69 f	3.41 c	3.80fgh	
Bathoor-2008	1.65 fg	6.90 g	2.02 fgh	3.35ghi	
Siran-2010	1.61 fg	6.96 g	1.59 ij	3.29ghi	
Auqab-2000	1.65 fg	6.06 h	2.40 ef	3.14hi	
Hashim-2008	1.42 gh	5.23 i	1.30 j	2.61ij	
Pirsabak-2005	1.13 h	5.02 i	2.29 efgh	2.60ij	
Pirsabak-2004	0.64 i	5.04 i	2.36 fg	2.41ij	I
Gomal-2008	0.57 i	3.97 j	1.92 hi	1.85j	

Within columns, means followed by the same letters are not significantly different at 0.05 % level of significance. Rating scale: Immunity (I), 1-2 aphid tiller<sup>-1</sup>; Resistance (R), 3-4 aphid tiller<sup>-1</sup>; Moderately Resistant (MR), 5-6 aphid tiller<sup>-1</sup>; Tolerant (T), 7-8 aphid tiller<sup>-1</sup>; Moderately Susceptible (MS), 9-10 aphid tiller<sup>-1</sup>; Susceptible (S), 11-15 aphid tiller<sup>-1</sup>; and Highly Susceptible (HS), 16-20 aphid tiller<sup>-1</sup>.

(MS), 9-10 aphid tiller<sup>-1</sup>; Susceptible (S), 11-15 aphid tiller<sup>-1</sup>; and Highly Susceptible (HS), 16-20 aphid tiller<sup>-1</sup> (Table II).

#### Data analysis

Data were analyzed with analysis of variance using Statistix 8.1 (Analytical Software, 2005) for multiple comparisons; LSD-test was used for separation of means at  $\alpha=0.05$ . Standard errors of means of aphids (all aphids pooled together, and main aphid species individually; Table III) aphid parasitoids (Table IV), and predators (Table V) were calculated following previous parameters reported by Zeb *et al.* (2011). Correlation ( $r$ ) between aphid density and rate of parasitism per variety and aphid density and number of predator per variety were determined using statistical software (Table VI).

## RESULTS

As shown in Table II, the mean number of aphids

per tiller per cultivar differ early, mid, and late-season. Overall, statistically significant differences of the entire season mean were observed ( $F=517.49$ ;  $df=19$ ;  $P > 0.0000$ ). The highest mean number of aphids per tiller was recorded on Saleem-2000 (9.1 aphids/tiller), while the lowest was found on Gomal-2008 (1.9), followed by Pisabak-2004 (2.4), Pisabak-2005 (2.6), and Hashim-2008 (2.6). Early ( $F=396.56$ ;  $df=19$ ;  $P > 0.0000$ ), mid ( $F=93.86$ ;  $df=19$ ;  $P > 0.0000$ ), and late ( $F=68.79$ ;  $df=19$ ;  $P > 0.0000$ ) season, the cultivar Saleem-2000 presented the highest number of aphids/tiller (Table II) classifying this variety as moderately susceptible (MS). In addition, no cultivar was found to be susceptible or highly susceptible, but 1 cultivar was found to be moderate susceptible, 8 moderate resistant, 9 resistant, and 2 immune.

Differences were observed when most common aphid species were analyzed separately (Table III). Close to 67% of aphids collected were *R. padi*, 16%, *S. graminum*, 15% *S. avenae*, and less than 2% 'other

aphids'. Consistently, cultivar Saleem-2000 presented the highest number of *R. padi* ( $F=28.77$ ,  $df=19$ ,  $319$ ;  $P > 0.0000$ ), *S. graminum* ( $F=2.74$ ,  $df=19$ ;  $P > 0.0003$ ), and *S. avenae* ( $F=23.41$ ,  $df=19$ ;  $P > 0.0000$ ) per tiller.

**Table III. Number of aphids (Mean±SE) per tiller per cultivar in Agricultural Research Institute, Tarnab, Peshawar, Pakistan.**

Wheat cultivars	<i>R. padi</i>	<i>S. graminum</i>	<i>S. avenae</i>
Saleem-2000	8.12 ± 0.78 a	2.02 ± 0.32 a	1.64 ± 0.12 a
Tatara-97	5.39 ± 0.73 b	0.64 ± 0.23 fg	1.89 ± 0.12 a
Zam-2004	5.22 ± 0.73 bc	1.20 ± 0.46 bcdefg	0.66 ± 0.17 bcd
Fakhr-e-Sarhad	5.01 ± 0.71 bcd	1.55 ± 0.41 abcd	0.26 ± 0.07 fgh
KT-2000	4.35 ± 0.77 de	1.63 ± 0.44 abc	0.89 ± 0.99 b
Barsat-2010	4.17 ± 0.76 ef	1.13 ± 0.4 bcdefg	0.30 ± 0.03 efg
Dera-98	4.39 ± 0.76 cde	0.95 ± 0.29 defg	0.74 ± 0.18 bc
Attahabib-2010	3.75 ± 0.73 efg	1.32 ± 0.35 bcde	0.74 ± 0.13 bc
Daman-98	4.24 ± 0.71 de	0.59 ± 0.28 g	0.57 ± 0.18 cde
Bakhtawar-92	3.40 ± 0.78 fgh	1.25 ± 0.35 bcdefg	0.67 ± 0.11 bcd
Khyber-87	3.16 ± 0.72 ghi	1.28 ± 0.36 bcdef	1.64 ± 0.09 a
Sehar-2006	2.99 ± 0.51 ghi	1.08 ± 0.29 cdefg	0.10 ± 0.05 g
Pirsabak-2008	3.05 ± 0.76 ghi	1.38 ± 0.45 abcde	0.13 ± 0.06 g
Bathoor-2008	2.33 ± 0.71 ijk	1.31 ± 0.4 bcde	0.37 ± 0.12 defg
Siran-2010	2.12 ± 0.57 jkl	1.74 ± 0.4 ab	0.09 ± 0.07 g
Auqab-2000	2.78 ± 0.59 hij	0.83 ± 0.28 efg	0.10 ± 0.06 g
Hashim-2008	2.08 ± 0.49 jkl	0.78 ± 0.44 efg	0.50 ± 0.09 cdef
Pirsabak-2005	1.64 ± 0.36 kl	1.41 ± 0.46 abcde	0.31 ± 0.09 efg
Pirsabak-2004	1.92 ± 0.6 kl	0.80 ± 0.28 efg	0.20 ± 0.07 fg
Gomal-2008	1.39 ± 0.4 l	1.06 ± 0.26 cdefg	0.22 ± 0.07 fg

Within columns, means followed by the same letters are not significantly different at 0.05 % level of significance.

*Aphidius ervi* and *A. colemani* were the most common parasitoids. Early in the season ( $F=2.92$ ;  $df=19$ ;  $P > 0.0001$ ), the mean rate of parasitism was close to zero (Table IV); however, as season progressed, parasitism gradually increased (mid-season  $F=5.38$ ;  $df=19$ ;  $P > 0.0001$ ; late season,  $F=28.37$ ;  $df=19$ ;  $P > 0.0001$ ). The highest seasonal mean rate of parasitism was recorded on Pirsabak-2005 (20) followed by Siran-2010 (20), and Gomal-2008 (19) (Table IV).

*Coccinella septempunctata* L (Coleoptera: Coccinellidae) ( $F=11.32$ ,  $df= 19$ ,  $P= 0.000$ ), *Chrysoperla carnea* Stephens (Neuroptera: Chrysopidae) ( $F=4.07$ ;  $df=19$ ;  $P=0.001$ , and several species of Hover flies ( $F=2.44$ ,  $df=19$ ,  $P=0.0095$ ) were the dominant predators (Table V). Total predator population (all predators combined) per

tiller<sup>-1</sup> was significantly higher ( $F= 7.03$ ,  $df= 19$ ,  $P=0.0000$ ) on Attahabib-2010 followed by Pirsabak-2005 (Table V). Lower mean predators' population was recorded on Siran-2010 and Auqab-2000.

**Table IV. Mean rate of parasitism per tiller per variety during early, mid and late season in Agricultural Research Institute, Tarnab, Peshawar, Pakistan.**

Wheat cultivars	Early (Nov to Jan)	Mid(Feb to Mar)	Late (mid-Mar to first week of Apr)	Seasonal mean
Pirsabak-2005	2.96 a	15.46 b	46.98 cde	20.21 ± 7.49 a
Siran-2010	0.00 f	9.82 d	56.53 a	20.2 ± 10.87 a
Gomal-2008	0.00 f	16.30 a	45.15 de	19.04 ± 8.19 a
Hashim-2008	0.80 d	11.75 c	50.50 bc	18.34 ± 8.53 ab
Daman-98	0.00 f	6.19 gh	51.40 b	16.53 ± 9.55 abc
Sehar-2006	0.00 f	4.16 j	48.39 bcd	15.35 ± 1.57 abcd
Zam-2004	0.00 f	9.67 d	43.80 e	15.14 ± 7.26 abcd
Barsat-2010	0.00 f	5.23 i	39.76 f	13.45 ± 8.44 abcde
Attahabib-2010	1.54 b	7.62 e	29.80 gh	11.54 ± 5.35 bcde
Bakhtawar-92	0.57 e	4.90 i	31.33 g	10.99 ± 5.82 cde
Uqab-2000	1.10 c	6.05 gh	29.05 gh	10.84 ± 4.97 cde
Tatara-97	0.00 f	4.93 i	27.93 ghi	10.17 ± 6.02 cde
Dera-98	0.53 e	6.75 f	26.76 hij	10.09 ± 4.52 cde
Pirsabak-2004	0.00 f	4.12 j	30.65 g	10.08 ± 6.11 cde
Bathoor-2008	1.1 c	6.25 fgh	25.32 ij	10.04 ± 4.52 cde
KT-2000	0.56 e	6.40 fg	25.11 ij	9.89 ± 4.47 cde
Saleem-2000	0.00 f	6.58 fg	26.50 hij	9.20 ± 4.48 cde
Fakhr-e-Sarhad	0.00 f	5.24 i	24.33 ij	8.62 ± 4.46 de
Pirsabak-2008	0.00 f	3.40 k	23.41 j	8.13 ± 4.71 e
Khyber-87	0.00 f	5.85 h	16.87 k	6.61 ± 2.54 e

Within columns, means followed by the same letters are not significantly different at 0.05 % level of significance.

Table VI shows a correlation between aphid density and rate of parasitism and between aphid density and number of predators per tiller on each variety. Varieties such as Saleem-2000 that consistently showed the highest number of aphids per tiller was negative correlated to rate of parasitism but positive correlated to predator density. The only significant correlations were observed between aphids per tiller and predators present in cultivars Gomal-2008 and Pirsabak-2008.

**Table V. Number of predators (Mean±SE) per tiller per variety in Agricultural Research Institute, Tarnab, Peshawar, Pakistan.**

Wheat cultivars	Coccinellid beetles	Green lace wing	Syrphid flies	Total predators population
Pirsabak-2005	0.25 ± 0.09 a	0.37 ± 0.03 a	0.05 ± 0.01 cd	0.44 ± 0.15 a
Attahabib-2010	0.28 ± 0.09 a	0.13 ± 0.03 cdefgh	0.12 ± 0.02 ab	0.40 ± 0.10 ab
Bathoor-2008	0.19 ± 0.5 b	0.32 ± 0.04 ab	0.10 ± 0.01 bcd	0.36± 0.12 abc
Zam-2004	0.17 ± 0.02 b	0.25 ± 0.03 abcd	0.14 ± 0.03 ab	0.36 ± 0.08 abc
Barsat-2010	0.10 ± 0.02 cdefg	0.27 ± 0.02 abc	0.22 ± 0.05 a	0.34 ± 0.10 abcd
Sehar-2006	0.15 ± 0.02 bcd	0.24 ± 0.02 abcde	0.13 ± 0.01 bc	0.32 ± 0.06 abcd
KT-2000	0.16 ± 0.06 bc	0.22 ± 0.17 bcdef	0.07 ± 0.04 bcd	0.29 ± 0.06 bcde
Tatara-97	0.14 ± 0.04 bcde	0.15 ± 0.04 cdefgh	0.09 ± 0.03 bc	0.26 ± 0.07 cdef
Khyber-87	0.08 ± 0.02 fghi	0.20 ± 0.03 bcdefg	0.12 ± 0.03bc	0.23 ± 0.03 defg
Dera-98	0.11 ± 0.04 cdef	0.12 ± 0.03 fgh	0.04 ± 0.02 cd	0.19 ± 0.05 efg
Daman-98	0.08 ± 0.01 fgh	0.10 ± 0.00 fgh	0.09 ± 0.04 bc	0.18 ± 0.05 efg
Pirsabak-2008	0.10 ± 0.01 defg	0.07 ± 0.02 gh	0.09 ± 0.05 bc	0.17 ± 0.05 fgh
Fakhr-e-Sarhad	0.10 ± 0.03 defg	0.11 ± 0.03 efg	0.04 ± 0.02 cd	0.17 ± 0.05 fgh
Pirsabak-2004	0.08 ± 0.01 fgh	0.03 ± 0.02 h	0.12 ± 0.06 bc	0.16 ± 0.03 fgh
Saleem-2000	0.03 ± 0.01 hi	0.13 ± 0.03 cdefgh	0.10 ± 0.01 bc	0.14 ± 0.05 gh
Gomal-2008	0.10 ± 0.02 defg	0.02 ± 0.02 h	0.06 ± 0.03 bcd	0.13 ± 0.11 gh
Hashim-2008	0.08 ± 0.03 def	0.12 ± 0.04 defgh	0.00 ± 0.00 d	0.13± 0.02 gh
Bakhtawar-92	0.05 ± 0.01 ghi	0.07 ± 0.04 gh	0.06 ± 0.00 bcd	0.11 ± 0.06 gh
Siran-2010	0.05 ± 0.01 ghi	0.08 ± 0.02 fgh	0.06 ± 0.02 bcd	0.12 ± 0.05 gh
Auqab-2000	0.02 ± 0.01 i	0.12 ± 0.04 defgh	0.00.05±0.04 cd	0.10 ± 0.03 h

Within columns, means followed by the same letters are not significantly different at 0.05 % level of significance.

## DISCUSSION

Aphids are present in all wheat Pakistani cultivars early, mid, and late season. Following a modification of Ronquim *et al.*, 2004, Muhammad *et al.* (2013) and Wains *et al.* (2014) ranking scales, the cultivar Saleem-2000 was found Moderately Susceptible; 8 cultivars including Tatara-97, Zam-2004, FakhreSarhad-99, Barsat-2010, KT-2000, Dera-98, Attahabib-2010, and Daman-98 were found Moderately Resistant; 9 cultivars, including Bakhtawar-92, Khyber-87, Sehar-2006, Pirsabak-2008, Bathoor-2008, Siran-2010, and Uqab-2000 were categorized Resistant; while Pirsabak-2004 and Gomal-2008 showed immune response (Table II). This ranking scale, based on the number of aphids per tiller, is widely used. For instance, Muhammad *et al.* (2013) conducted field experiments to study the population dynamics of wheat aphids on 16 wheat cultivars where Sehar-2006, which was also included in our study, was the least 'preferred' cultivar considered "Immune", based on mean number of aphids per tiller; in our study this cultivar performed somewhere in the middle classifying as "Resistant". Khan and Gul (2012)

reported the screening of 20 wheat cultivars where 4 of those, CT-0492, CT-07043, WL-01869 and NIFA V-16, were highly infested or "Susceptible" (11 aphids per tiller) while others such Bathoor-2008 (8 aphids per tiller) was found to be "Tolerant". Wains *et al.* (2010) tested 12 wheat lines including Shafaq-2006, Sehar-2006, FSD-08 and Lasani-2008 classified as "Resistant". Akhtar *et al.* (2009) studied Inqilab-91, Chakwal-97, MH-97, Margalla-99, Chenab-2000, Iqbal-2000, Saleem-2000 and Wafaq-2001 concluding that Saleem-2000 and Iqbal-2000 were "Susceptible" varieties to wheat aphids. Iqbal *et al.* (2008) screened 28 wheat varieties/lines finding that Iqbal-2000 (8 aphids per tiller) was "Susceptible" while the advance line V00147 was "Resistant" (3 aphids per tiller). Other varieties such as Suliman-96, RT-5, Khaniwal, K2-L, Takbeer, Nowshera-96, Pirsabak-85, Blue Silver, KRL-1.4, Ghaznawi-98, Bakhtawar-92, Kim, Fakhr-e-Sarhad, Dera-98, Saleem-2000, ICARDA, SARC-3, RT-10, Tatara-97, KT-2000 and Inqilab have been continuously tested (Akhtar and Mujahid, 2006; Khan *et al.*, 2007a). Some of these cultivars, varieties or lines are common in local commercial production (Pakistani Economic Survey, 2017).

**Table VI. Correlation of aphid density and its predators and percent parasitism per tiller per variety.**

Wheat cultivars	Correlation coefficient (r) between aphid density and predator	Correlation coefficient (r) between aphid density and parasitism
Pirsabak 2005	0.302	0.145
Atta habib 2010	0.37	-0.196
Zam 2004	0.318	-0.328
Bathoor 2008	-0.002	-0.067
Barsat 2010	0.303	-0.340
Seher 2006	0.217	-0.340
Saleem 2000	0.217	-0.346
Dera 98	-0.032	-0.318
FakhreSarhad 99	-0.185	-0.385
Tatara 97	0.066	-0.306
Daman 98	0.144	-0.451
Auqab 2000	0.192	-0.099
Pirsabak 2004	-0.584	-0.101
Khyber 87	0.283	-0.034
Bakhtawar 92	0.386	-0.038
Hashim 2008	-0.150	-0.069
Gomal 2008	0.762*	0.177
KT 2000	-0.120	-0.042
Pirsabak 2008	0.662*	-0.135
Siren 2010	0.208	-0.195

\*Significant ( $P \leq 0.05$ )

The parental and pedigree of each cultivar tested are present in Table I showing the indirect linkage of some cultivars; some cultivars are coming from long standing breeding programs mainly disease resistant oriented. It was not part of this study to evaluate the morphological or chemical characteristics of each cultivar tested that could help us understand why aphids choose one cultivar versus another one. Plant morphological characteristic such as growth habit, plant height, leaf color and growth stage of various wheat genotypes could affect the aphid seasonal dynamic (Markova and Tomchev, 2013). It was part of our study to determine presence of aphids in given cultivars to be able to determine action control methods. Based on our study, alate aphids can start colonizing as soon as plants emerged (personal observation). Previously, Ali *et al.* (2011, 2015), and Aheer *et al.* (2006) reported similar results. Ronquim *et al.*, 2004 indicated that population differences based on number of individuals occurring on the host plant is one way to verify host resistance to

insects. It is known that the seasonal distribution of aphids depends upon the climatic condition of the region, host plant quality, natural enemies and agricultural practices (Brewer and Elliot, 2004).

Although unknown how many species of aphids are present in Pakistani wheat fields, it is not surprising to find *R. padi* as the predominant specie; *S. graminum*, and *S. avenae* were also predominant in wheat fields (Table III). Gianoli (2000) and Honek *et al.* (2006) previously reported the three species aforementioned as the main species found in wheat fields. In our study, *R. padi* was found to colonize the leaves (1-2 leaf stage) and stems early in the season, while *S. graminum* infestation occurred at the 5-6 leaf stage mainly on leaves but later on spikes; *S. avenae* appeared late in the season but only present on spikes suggesting some sort of species displacement or specialization which was not evaluated on this study.

Insect-parasitoid-predator interactions are important when evaluating susceptibility or resistance of cultivars/varieties/lines. For instance, in our study the cultivars characterized as “Immune” based on the number of aphids per tiller such as Pirsabak-2004 and Gomal-2008 (Table II) presented one of the lowest (Pirsabak-2004) and highest (Gomal-2008) parasitism rate (Table IV); in contrast one of the highest (Pirsabak-2004), and lowest (Gomal-2008) predator rate (Table V) suggesting a more complex relationship. In an early study by Schuster and Starks (1975), this complexity was demonstrated studying the preference of *Lysiphlebus testaceipes* L. for greenbug in small grain. Similarly, Biswas and Singh (1998) indicated that host plant resistance and bio-control should be further study in well control settings. In our study, *A. ervi* and *A. cloemani* were identified as the main two parasitoids. In other studies, Adisu *et al.* (2002) reported *A. colemani*, *A. rhopalosiphi* and *A. uzbekistanicus* in Germany; Chambers *et al.* (1982) and Charlet *et al.* (2002) reported *L. testaceipes*, *Aphelinus mali*, and *A. avenaphis* in North Dakota, USA; Khan *et al.* (2007b) also reported *A. ervi* and *A. colemani* in Pakistan, while Ali *et al.* (2015) reported *Diaeretiella rapae*. In our study, parasitism rates started low, however, as season progressed, the mean rate of parasitism increased (Table IV). This observation is similar to the one noted by Tomanovic *et al.* (2008).

## CONCLUSION AND RECOMMENDATION

A good degree of resistance was observed in varieties, Gomal-2008, Hashim-2008, Pisabak-2004 and Pirsabak-2005 against whet aphid. Furthermore, these varieties shown high compatibility for natural parasitism. Pirsabak-2005 was the only variety among the tested wheat varieties which provided easy access to parasitoids

and predators for parasitism or consumed his prey (aphid). The susceptible variety Saleem-2000 in spite of having more aphid shown less compatible for bio-control agents. It is further suggested that resistance mechanism of these varieties should be studied to explore the physiochemical and physiological base of resistance. This information will provide a baseline data to breeder to develop high yielding wheat varieties with adaptation to different agro ecological condition and compatible with biological control agents.

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#### Statement of conflict of interest

The authors stated no conflicts of interest.

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