



# Trophic Associations of Ants with Aphid Partners and New Distribution Records of some Ants in Pothwar Region of Pakistan

Ammara Gull-E-Fareen<sup>1,3</sup>, Imran Bodlah<sup>1\*</sup>, Muhammad Tariq Rasheed<sup>1</sup>, Yasir Niaz<sup>2</sup>, Muhammad Adnan Bodlah<sup>2</sup>, Muhammad Asif<sup>4</sup> and Nasir M. Khokhar<sup>5</sup>

<sup>1</sup>*Insect Biodiversity and Conservation Group, Department of Entomology, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi, Pakistan*

<sup>2</sup>*Fareed Biodiversity and Conservation Centre, Department of Agricultural Engineering, Khawaja Fareed University of Engineering and Information Technology, Rahim Yar Khan, Punjab, Pakistan*

<sup>3</sup>*Department of Environmental Sciences, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi, Pakistan*

<sup>4</sup>*Agricultural Linkages Program (ALP), Pakistan Agricultural Research Council, Islamabad, Pakistan*

<sup>5</sup>*Natural Sciences Linkages Programme, Pakistan Science Foundation Islamabad*

## ABSTRACT

Ants are the economically important and metropolitan insects on earth. They act as soil engineers, scavengers, decomposers, predators, pollinators and herbivores. The mutualistic relationship between ants and aphids is a very common and widespread phenomenon. It has been studied in various parts of the world. In this relationship, aphids provide food to the ants, while in return ants provide them protection from natural enemies. Ants also protect aphids from different diseases. Aphids (serious crop pests) can be divided into myrmecophilous and non- myrmecophilous. The main objective of this study was to determine trophic associations of ants associated with aphids on various host plants in Pothwar. For this purpose, seven ant species were selected, identified as *Camponotus compressus* (Fabricius, 1787), *Formica fusca* Linnaeus, 1758, *Formica clara* Forel, 1886, *Lepisiota frauenfeldi* (Mayr, 1855), *Myrmica aimonissabaudiae* Menozzi, 1939, *Tapinoma melanocephalum* (Fabricius, 1793) and *Tetraponera allaborans* (Walker, 1859). A lot of surveys were conducted during 2015-17 for the collection of ants associated with aphids on different host plants in different localities of Pothwar region. As a result of surveys, ant-aphid new mutualistic trophic associations were determined like *Camponotus compressus* with 12, *Formica fusca* 1, *Formica clara* 2, *Lepisiota frauenfeldi* 9, *Myrmica aimonissabaudiae* 4, *Tapinoma melanocephalum* 11 and *Tetraponera allaborans* with 1 new association in various localities of Pothwar on different host plants have been reported. All these associations are recorded for the first time from Pakistan. Trophic associations of studied ants with aphids on different host plants, their world distribution and comments on observed new associations have been given. New distribution records of ant's species have also been provided.

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### Authors' Contribution

AGF and MTR surveyed, collected and identified aphids and ants. IB planned and compiled the research. YN, NMK and MA wrote the manuscript. MAB helped in aphid species confirmation.

### Key words

Mutualism, Ant, Aphid, Distribution, Pakistan

## INTRODUCTION

Ants (Hymenoptera: Formicidae) are the economically important and metropolitan insects on earth. Formicidae is divided into 20 subfamilies (Bolton, 1994; Bolton *et al.*, 2006; Ward, 2007). Ants are good in seed dispersal (Hanzawa *et al.*, 1988). They improve soil aeration (Holldobler and Wilson, 1990). They are known as

ecosystem engineers as they are active in underground activities (Folgarait, 1998). They also perform different roles in ecosystem like scavengers, decomposers, predators, pollinators and herbivores. Harvester ants grow fungus in their nest by accumulating leaves and also feed on them (Holldobler and Wilson, 1990). Ants consume honey dew secretions of aphids in symbiotic association (Styrsky and Eubanks, 2007; Jahn and Beardsley, 1996). There is a mutualistic association between ants and aphids (Depa and Węgierek, 2011). In this relationship, aphids provide food to the ants, while in return ants provide them protection from natural enemies (Sudd, 1987; Cushman and Beattie,

\* Corresponding author: [imranbodlah@gmail.com](mailto:imranbodlah@gmail.com)  
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1991). Ants also protect aphids from different diseases e.g. *Formica podzolica* protect aphids, *Aphis asclepiadis*, from lethal fungal infections (Nielsen *et al.*, 2010). Flatt and Weisser (2000) reported that aphids associated with ants lived longer, had higher rate of reproduction and greater number of progeny (Yoo and Holway, 2011).

Among mutualisms, ant-aphid interactions are among the most variable in terms of their outcomes whether or not the ant aphid interaction is beneficial to the aphids often depends upon both the spatial and ecological context. Aphids, the serious insect pests of various crops, fruits, vegetables etc. are myrmecophilous (tended by ants) and non- myrmecophilous (unattended). The great majority of ants taking part in such associations belong to phylogenetically advanced sub-families Dolichoderinae, Formicinae and Myrmicinae (Delabie, 2001). The mutualistic relationship between ants and aphids has been the subject of many studies on various aspects of this phenomenon in various regions of the world. In different regions like Iran, 21 ant species associated with 26 aphid species from 37 host plant species (Mortazavi *et al.*, 2015), 18 ants species associated with 34 species of aphids from Florida (Nielsson *et al.*, 1971), 10 species of ants associated with 24 aphid species from Russia (Addicott, 1979), 23 species of ants associated with 11 aphid species from Oceania (Idechiil *et al.*, 2007), 16 species of ants in association with 19 species of aphids from Ankara Province of Turkey (Özdemir *et al.*, 2008), 17 species of ants associated with aphids in Indonesia (Herwina *et al.*, 2013), 20 species of ants associated with 3 species of aphids in India (Kataria and Kumar, 2013) etc. But as far as Pakistan is concerned, recently a lot of work on both aphids (Bodlah *et al.*, 2013, 2017; Amin *et al.*, 2017a, b; Maryam *et al.*, 2019) and ants (Bodlah *et al.*, 2016, 2017, 2019) has been done but on mutualistic associations are still missing. Recently Bodlah *et al.* (2017) reported first association of a psyllid, *Trioza fletcheri* minor Crawford, 1912 with two ants species in Pothwar, there is no report on aphids and their mutualistic ants in Pakistan. To fulfill this gap and provide a base for further studies on interactions between aphids and ants, the present study was conducted.

## MATERIALS AND METHODS

### *Ant-aphid collection and preservation*

Surveys of Attock, Chakwal, Rawalpindi, Islamabad and Jehlum were conducted for the collection of ants and aphids from different host plants during 2015-17. Ants were collected from aphid infested crop plants, grasses, trees, weeds, ornamentals, fruits, vegetables etc. Only those ant-aphid pairs were collected where we observed ants stroke the aphids with their antennae, stimulating them

to release the honeydew. Ants were collected with aspirator and through hand picking, were killed and preserved in 75 % ethanol in small glass vials. Aphids were collected and preserved in 75 % ethanol in small glass vials. Longitude and latitudes of the surveyed areas were determined with the help of GPS device.

### *Identification of ant-aphid partners*

Ants were mounted on triangular cards and pinned. Large ants were preserved by using entomological pins. These specimens were identified by using compound microscope and identification keys by Bingham (1903). Aphids were identified using keys by Blackman and Eastop (1994, 2000). Host plants inhabiting ants and aphids were also collected and identified to develop the mutualistic association among ants, aphids and host plants of aphids. The voucher specimens have been deposited in the Museum collection of Department of Entomology, PMAS-AAUR.

## RESULTS AND DISCUSSION

Seven different species of ants belonging to seven different genera were associated with different aphid partners on various host plants in different areas of Pothwar region. All associations are new. New distribution records for ants have also been added.

### **1. *Camponotus compressus* (Fabricius, 1787)**

*Aphid partners in association reported during these studies in Pakistan*

Black bean aphid (*Aphis fabae* Scopoli, 1763), Fox glove aphid (*Aulacorthum solani* Kaltentbach, 1843), Pomegranate Aphid (*Aphis punicae* Passerini, 1863), Thistle aphid (*Brachycaudus cardui* Linnaeus, 1758), Woolly Apple Aphid (*Eriosoma lanigerum* Hausmann, 1802), Mustard Aphid (*Lipaphis erysimi* Kaltentbach, 1843), Ornate Aphid (*Myzus ornatus* Laing, 1932), Hemp aphid (*Phorodon cannabis* Passerini, 1860), Bird cherry-oat aphid (*Rhopalosiphum padi* Linnaeus, 1758), Wheat aphid (*Schizaphis graminum* Rondani, 1852), Grain Aphid (*Sitobion avenae* Fabricius, 1794), Safflower Aphid (*Uroleucon carthami* Hille Ris Lambers, 1948).

### *Material examined*

40♂, Rawalpindi: N33° 38.929' E073° 04.943' 1671 ft. elev., 02-12-2015 *Triticum aestivum* (Wheat), 2♀, N33° 38.929' E073° 04.943' 1645 ft. elev., 01-01-2016 *Parthenium hysterophorus* (Parthenium weed), *Duranta erecta* (Golden dewdrop), *Bougainvillea spectabilis* (Paperflower), *Moringa oleifera* (Moringa), *Cannabis* sp. (Hemp); 38♀, Islamabad: N33° 40.527' E073° 08.376'

1762 ft. elev., 03-09-2015 *Zea mays* (Maize), *Triticum aestivum* (Wheat), *Solanum nigrum* (Black nightshade), *Spinacia oleracea* (Spinach), *Parthenium hysterophorus* (Parthenium weed), *Capsicum annuum* (Green Chilly), *Abelmoschus esculentus* (Okara), *Asclepias* sp. (Milk weed), *Punica granatum* (Pomegranate), 3♀, N33° 42.558' E073° 01.330', 1686 ft. elev., 20-11-2015 *Bougainvillea spectabilis* (Paperflower), *Jasminum officinale* (Jasmine); 15♀, Murree: N33° 59.652' E073° 28.593' 4980 ft. elev., 28-11-2015 *Spinacia oleracea* (Spinach), *Jasminum officinale* (Jasmine), *Parthenium hysterophorus* (Parthenium weed), *Debregisia silicifolia* (Siharu); 5♀, N33° 55.341' E073° 24.216' 6302 ft. elev., 09-01-2017 *Fragaria ananassa* (Strawberry); 2♀, N33° 55.016' E073° 23.699' 6415 ft. elev., 28-11-2015 *Carthamus oxycanthus* (Wild Safflower), *Quercus ilex* (Holly oak); 6♀, N33° 59.342' E073° 28.573' 5020 ft. elev., 09-01-2016 *Fragaria ananassa* (Strawberry), *Viola odorata* (Sweet violet), *Artemisia absinthium* (Wormwood), *Helianthus annuus* (Sunflower), *Bischofia javanica* (Bishop wood); 14♀, Chakwal: N32° 46.160' E072° 42.299', 2209 ft. elev., 29-09-2015 *Eriobotrya japonica* (Loquat), *Parthenium hysterophorus* (Parthenium weed); 10♀, Jhelum: N32° 58.118' E073° 41.601', 859 ft. elev., 15-04-2016 *Parthenium hysterophorus* (Parthenium weed), *Rosa indica* (Rose).

#### Comments on ant-aphid associations

This ant has been reported in association with 5 aphid species namely *Aphis craccivora* (Kataria and Kumar, 2013; Rakhshan and Ahmad, 2015), *Aphis fabae* (Kataria and Kumar, 2013), *Aphis gossypii* (Vergheese and Tandon, 1987; Kataria and Kumar, 2013; Lokeshwari et al., 2015), *Aphis nerii* (Kataria and Kumar, 2013) and *Myzus persicae* (Kataria and Kumar, 2013) from various parts of the world on different host plants. Here we have added new associations of this species with 12 different aphid partners from Pakistan. *Camponotus compressus* was found in abundance in association with 12 aphid species on *Triticum aestivum* (Wheat), *Parthenium hysterophorus* (Parthenium weed), *Duranta erecta* (Golden dewdrop), *Bougainvillea spectabilis* (Paper flower), *Moringa oleifera* (Moringa) and *Cannabis* sp. (Hemp) from various localities of district Rawalpindi. On *Zea mays* (Maize), *Triticum aestivum* (Wheat), *Solanum nigrum* (Black night shade), *Spinacia oleracea* (Spinach), *Parthenium hysterophorus* (Parthenium weed), *Capsicum annuum* (Green Chilly), *Abelmoschus esculentus* (Okara), *Asclepias* sp. (Milk weed), *Punica granatum* (Pomegranate), *Bougainvillea spectabilis* (Paper flower) and *Jasminum officinale* (Jasmine) from different locations of Islamabad. On *Spinacia oleracea* (Spinach), *Jasminum officinale* (Jasmine), *Parthenium hysterophorus* (Parthenium

weed), *Debregisia silicifolia* (Siharu), *Fragaria ananassa* (Strawberry), *Carthamus oxycanthus* (Wild Safflower), *Quercus ilex* (Holly oak), *Viola odorata* (Sweet violet), *Artemisia absinthium* (Wormwood), *Helianthus annuus* (Sunflower) and *Bischofia javanica* (Bishop wood) from Murree, *Eriobotrya japonica* (Loquat) and *Parthenium hysterophorus* (Parthenium weed) from district Chakwal and *Parthenium hysterophorus* (Parthenium weed) and *Rosa indica* (Rose) from areas of district Jhelum. It was found on the tree trunks and barks tending aphids for honey dew secretion. All the collected specimens were identified and found similar to published description of Bingham (1903). *Camponotus compressus* is reported for the first time in association with any aphid species from Pakistan, so it is reported as new country record.

#### Comments on distribution of *camponotus compressus* in Pakistan

Umair et al. (2012) mentioned the distribution of this species only from Islamabad and Rawalpindi. We have added new distribution records in various localities of Pothwar.

## 2. *Formica fusca* Linnaeus, 1758

#### Aphid partners in association reported during these studies in Pakistan

Pine aphid (*Cinara orientalis* Takahashi, 1925)

#### Material examined

20♀, (Kuldana) N33° 55.315' E073° 24.212', 6324 ft. elev., 15-04-2017 *Pinus wallichiana* (Himalayan pine); 10♀, Murree: (Aliote) N33° 55.689' E073° 24.957', 6042 ft. elev., 15-04-2017 *Pinus wallichiana* (Himalayan pine); 13♀, (Kuldana) N33° 55.315' E073° 24.212', 6324 ft. elev., 04-05-2017 *Pinus wallichiana* (Himalayan pine); 15♀, Murree: (Chara Pani) N33° 51.013' E073° 19.156', 3809 ft. elev., 11-11-2016 *Pinus wallichiana* (Himalayan pine).

#### Comments on ant-aphid associations

This species has been recorded to be associated with 57 aphid species from various parts of the world as reviewed by Siddiqui et al. (2019). During our studies, 1 new trophic association of this species with aphid, *Cinara orientalis* was observed. This association is new country record from Pakistan. This ant species was observed and found in abundance on the pine tree *Pinus wallichiana* (Himalayan pine) in association with *Cinara orientalis*. Both the ants and aphids were present in the lenticels of the tree. Aphids were sucking sap from tree trunk and ants were getting honey dew from aphids. All the collected specimens were identified and found similar to the published description of species by Bingham (1903).

*Comments on distribution of Formica fusca in Pakistan*

Menozzi (1939) reported this species from Karakorum (Province Gilgit-Baltistan). During our surveys, new distribution pattern in Murree (Province Punjab) has been added.

### 3. *Formica clara* Forel, 1886

*Aphid partners in association reported during these studies in Pakistan*

Pine aphids (*Cinara confinis* (Koch, 1856), (*Cinara orientalis* (Takahashi, 1925)

*Material examined*

15♂, Murree: (Kuldana) N33° 55.315' E073° 24.212', 6324 ft. elev., 15-04-2017 *Abies pindrow* (Pindrow fir); 3♀, (Hill Dhuloo): N33° 58.513' E073° 29.703', 5198 ft. elev., 21-4-2017 *Abies pindrow* (Pindrow fir); 5♀, (Kuldana): N33° 55.315' E073° 24.212', 6324 ft. elev., 15-04-2017 *Pinus wallichiana* (Himalayan pine); 20♀, Murree: (Aliote) N33° 55.689' E073° 24.957', 6042 ft. elev., 15-04-2017 *Pinus wallichiana* (Himalayan pine); 5♀, (Kuldana): N33° 55.315' E073° 24.212', 6324 ft. elev., 04-05-2017 *Pinus wallichiana* (Himalayan pine); 7♀, Murree (Chara Pani): N33° 51.013' E073° 19.156', 3809 ft. elev., 11-11-2016 *Pinus wallichiana* (Himalayan pine).

*Comments on ant-aphid associations*

Siddiqui *et al.* (2019) mentioned this ant species to be in association with 8 aphid species from the world. During these studies, new associations with 2 aphid species have been added.

*Formica clara* was found in association with two aphid species namely, *Cinara confinis* and *Cinara orientalis* on *Abies pindrow* (Pindrow fir) and *Pinus wallichiana* (Himalayan pine) trees. Both aphids and ants were present in the lenticels on the tree trunks. All the collected specimens were identified and found similar to the published species description of Bingham (1903). *Formica clara* is reported for the first time in association with any aphid species from Pakistan.

*Comments on distribution of Formica clara*

Seifert and Schultz (2009) mentioned only its distribution in Pakistan but not the exact localities were reported. Here we have added exact localities of its distribution in Pothwar region of Pakistan.

### 4. *Lepisiota frauenfeldi* (Mayr, 1855)

*Aphid partners in association reported during these studies in Pakistan*

Loquat aphid (*Aphis eugeniae* van der Goot), Giant bark willow aphid (*Tuberolachnus salignus*

(Gmelin, 1776), Wild fig aphid (*Greenidea* (*Greenidea*) *ficicola* (Takahashi, 1921), *Greenidea* aphid (*Greenidea* (*Trichosiphum*) *formosana* (Maki, 1917), *Greenidea* aphid (*Greenidea* (*Greenidea*) *decaspermi* Takahashi), Apple aphid (*Aphis pomi* de Geer), Crepe myrtle aphid (*Tinocallis kahawaluokalani* (Kirkaldy), Chrysanthemum aphid (*Macrosiphoniella sanborni* (Gillette), Mint aphid (*Ovatus crataegarius*), Cotton Aphid (*Aphis gossypii* Glover, 1877), Black bean aphid (*Aphis fabae* Scopoli, 1763).

*Material examined*

10♀, Rawalpindi: N33° 38.612' E073° 04.476' 1733 ft. elev., 09-02-2016 *Eriobotrya japonica* (Loquat); 5♀, Islamabad: N33° 39.656' E073° 23.047', 3327 ft. elev., 19-03-2016 *Eriobotrya japonica* (Loquat); 4♀, N33° 39.516' E073° 23.007', 2153 ft. elev., 01-03-2016 *Eriobotrya japonica* (Loquat); 7♀, Chakwal: N32° 46.160' E072° 42.299', 2209 ft. elev., 29-03-2016 *Eriobotrya japonica* (Loquat); 4♀, N33° 40.527' E073° 08.376' 1762 ft. elev., 03-03-2016 *Eriobotrya japonica* (Loquat); 5♀, Rawalpindi: N33° 38.929' E073° 04.943' 1645 ft. elev., 11-03-2016 *Salix* sp. (Willow); 4♀, Islamabad: N35° 40.527' E072° 08.376' 1763 ft. elev., 13-04-2016; 10♀, Rawalpindi: N33° 38.612' E073° 04.476' 1733 ft. elev., 09-03-2016 *Salix* sp. (Willow); 15♀, Attock: N32° 56.655' E072° 51.312', 1663 ft. elev., 09-03-2016 *Salix* sp. (Willow); 6♀, Rawalpindi: N33° 39.516' E073° 23.007', 2153 ft. elev., 24-04-2016; *Ficus* sp. (Fig): 2♀, Rawalpindi: N33° 38.612' E073° 04.476' 1733 ft. elev., 09-04-2016 *Ficus* sp. (Fig); 7♀, Islamabad: N33° 43.929' E073° 02.179' 3836 ft. elev., 17-04-2016 *Ficus* sp. (Fig); 5♀, Rawalpindi: N33° 38.929' E073° 04.943' 1671 ft. elev., 14-04-2016 *Eugenia jambolana* (Jaman); 2♀, N33° 39.516' E073° 23.007', 2153 ft. elev., 24-04-2016 *Eugenia jambolana* (Jaman); 8♀, Murree: N33° 55.341' E073° 24.216' 6302 ft. elev., 09-04-2016 *Eugenia jambolana* (Jaman); 4♀, Kalar-Kahar: N32° 46.138' E072° 42.537', 2153 ft. elev., 12-04-2016 *Eugenia jambolana* (Jaman); 6♀, Jhelum: N32° 58.119' E073° 41.602', 859 ft. elev., 15-04-2016 *Eugenia jambolana* (Jaman); 3♀, Islamabad: N33° 40.527' E073° 08.376' 1762 ft. elev., 23-02-2016 *Eugenia jambolana* (Jaman); 2♀, Rawalpindi: N33° 38.929' E073° 04.943' 1645 ft. elev., 12-03-2016 *Psidium guajava* (Guava); 12♀, Islamabad: N33° 40.527' E073° 08.376' 1762 ft. elev., 03-04-2016 *Psidium guajava* (Guava); 5♀, Rawalpindi: N33° 38.612' E073° 04.476' 1733 ft. elev., 09-02-2016 *Eriobotrya japonica* (Loquat); 3♀, Islamabad: N33° 39.656' E073° 23.047', 3327 ft. elev., 19-03-2016 *Eriobotrya japonica* (Loquat); 2♀, N33° 39.516' E073° 23.007', 2153 ft. elev., 01-03-2016 *Eriobotrya japonica* (Loquat); 6♀, Chakwal: N32° 46.160' E072° 42.299', 2209 ft. elev., 29-03-2016 *Eriobotrya japonica*

(Loquat); 3♀, N33 40.527' E073 08.376' 1762 ft. elev., 03-03-2016 *Eriobotrya japonica* (Loquat), 3♀, Rawalpindi: N33 39.655' E073 23.047', 3323 ft. elev., 09-03-2016 *Malus pumila* (Apple); 5♀, Islamabad: (Pir Suhawa) N33 43.929' E07302.179' 3836ft. elev., 17-3-2016 *Malus pumila* (Apple); 6♀, Rawalpindi: N33 38.929' E073 04.943' 1645 ft. elev., 17-08-2016 *Lagerstromia indica* (Crepe myrtle); 8♀, Islamabad: N33 43.929' E07302.179' 3836ft. elev., 27-07-2016 *Lagerstromia indica* (Crepe myrtle); 5♀, Chakwal: N32 46.160' E072 42.299', 2209 ft. elev., 29-09-2015 *Lagerstromia indica* (Crepe myrtle); 3♀, Jhelum: N32 58.118' E073 41.601', 850 ft. elev., 15-07-2016 *Lagerstromia indica* (Crepe myrtle), 8♀, Rawalpindi: N32 38.928' E074 04.943' 1640 ft. elev., 21-03-2016 *Chrysanthemum indicum* (Chrysanthemum flower); 5♀, Islamabad: N33 40.527' E072 08.375' 1760 ft. elev., 3-04-2016 *Chrysanthemum indicum* (Chrysanthemum flower); 2♀, Rawalpindi: N34 38.612' E073 04.470' 1730 ft. elev., 09-03-2016 *Chrysanthemum indicum* (Chrysanthemum flower); 9♀, Attock: N32 56.655' E072 51.312', 1663 ft. elev., 09-03-2016 *Chrysanthemum indicum* (Chrysanthemum flower); 4♀, Murree: N33 59.652' E073 28.593' 4980 ft. elev., 28-04-2016; 7♀, Chakwal: N32 46.160' E072 42.299', 2209 ft. elev., 29-09-2015 *Chrysanthemum indicum* (Chrysanthemum flower), 4♀, Murree: N33 55.016' E073 23.699' 6415 ft. elev., 28-03-2016 *Mentha longifolia* (Mint).

#### Comments on ant-aphid associations

According to Shiran *et al.* (2013), this species has trophic association with 11 different aphid partners. Siddiqui *et al.* (2019) mentioned this ant species to be in association with 2 aphids.

In our studies, 9 new ant aphids' trophic associations have been added. This ant was most abundantly found in association with aphids on a large number of host plant species. It was found associated with 11 aphid species on different plant species including *Mentha longifolia* (Mint), *Chrysanthemum indicum* (Chrysanthemum flower), *Lagerstromia indica* (Crepe myrtle), *Malus pumila* (Apple), *Eriobotrya japonica* (Loquat), *Psidium guajava* (Guava), *Eugenia jambolana* (Jaman), *Ficus* sp. (Fig) and *Salix* sp. (Willow) from different areas of district Rawalpindi, Jhelum, Chakwal, Attock, Murree and Islamabad. It was the most active species found in association with aphids. All the collected specimens were identified and found similar to the published description of species by Bingham (1903). *Lepisiota frauenfeldi* is reported for the first time in association with any aphid species from Pakistan, so it is added as new country record.

#### Comments on distribution of *Lepisiota frauenfeldi* in Pakistan

Umair *et al.* (2012) recorded this species only from Rawalpindi and Islamabad. New locality records have been added during these studies.

#### 5. *Myrmica aimonissabaudiae* Menozzi, 1939

##### Aphid partners in association reported during these studies in Pakistan

Black bean aphid (*Aphis fabae* Scopoli, 1763), wild rose aphid (*Chaetosiphon (Pentatrachopus) glabrum* David, Rajasingh and Narayanan, 1971), Pine aphids (*Cinara confinis* (Koch, 1856), (*Cinara orientalis* (Takahashi, 1925).

##### Material examined

11♀, (Kuldana) N33 55.315' E073 24.212', 6324 ft. elev., 15-04-2017 *Rumex dentatus* (Jangli Palak); 4♀, (Aliote) N33 55.689' E073 24.957', 6042 ft. elev., 15-03-2017 *Rosa brunonii* (Himalayan Musk Rose); 3♀, (Osia) N33 55.318' E073 24.209', 5027 ft. elev., 25-03-2017 *Rosa brunonii* (Himalayan Musk Rose); 18♀, Murree: N33 43.255' E073 02.150', 4861 ft. elev., 02-03-2016; N33 59.652' E073 28.593' 4980 ft. elev., 28-03-2016 *Rosa brunonii* (Himalayan Musk Rose); 3♀, N33 55.315' E073 24.212', 6324 ft. elev., 11-03-2017 *Rosa brunonii* (Himalayan Musk Rose); 2♀, N33 55.318' E073 24.209', 5027 ft. elev., 15-03-2017 *Rosa brunonii* (Himalayan Musk Rose); 12♀, Murree: (Kuldana) N33 55.315' E073 24.212', 6324 ft. elev., 15-04-2017 *Abies pindrow* (Pindrow Fir); 2♀, (Hill Dhuloo) N33 58.513' E073 29.703', 5198 ft. elev., 21-4-2017 *Abies pindrow* (Pindrow Fir); 2♀, (Kuldana) N33 55.315' E073 24.212', 6324 ft. elev., 15-04-2017 *Pinus wallichiana* (Himalayan Pine); 22♀, Murree: (Aliote) N33 55.689' E073 24.957', 6042 ft. elev., 15-04-2017 *Pinus wallichiana* (Himalayan Pine); 3♀, (Kuldana): N33 55.315' E073 24.212', 6324 ft. elev., 04-05-2017 *Pinus wallichiana* (Himalayan Pine); 7♀, Murree: (Chara Pani) N33 51.013' E073 19.156', 3809 ft. elev., 11-11-2016 *Pinus wallichiana* (Himalayan Pine).

##### Comments on ant-aphid associations

This species has not been recorded from the world in association with aphids. During our studies 4 novel association of this species were observed for the first time in different localities of Pakistan.

*Myrmica aimonissabaudiae* was found in association with *Aphis fabae*, *Chaetosiphon (Pentatrachopus) glabrum*, *Cinara confinis* and *Cinara orientalis* on a number of host plant species including *Pinus wallichiana* (Himalayan Pine), *Abies pindrow* (Pindrow Fir), *Rosa brunonii* (Himalayan Musk Rose) and *Rumex dentatus*

(Jangli Palak) from different areas of Chara Pani, Kuldana, Aliote, Hill Dhuloo and Osia from district Rawalpindi. All the collected specimens were identified and found similar to the published description of Bingham (1903). *Myrmica aimonissabaudiae* is reported for the first time in association with any aphid species from Pakistan as well as from the whole world, so it is new country as well as new to science association.

*Comments on distribution of Myrmica aimonissabaudiae in Pakistan*

Menozi (1939) reported this species only from 2 localities, Karakorum (Gilgit Baltistan Province), Gund (Sindh Province) of Pakistan. Here we reported it for the first time from Pothwar region of Pakistan.

### 6. *Tapinoma melanocephalum* (Fabricius, 1793)

*Aphid partners in association reported during these studies in Pakistan*

Loquat aphid (*Aphis eugeniae* van der Goot), Giant bark willow aphid (*Tuberolachnus salignus* (Gmelin, 1776), Wild fig aphid (*Greenidea (Greenidea) ficicola* Takahashi, 1921, *Greenidea* aphid (*Greenidea (Trichosiphum) formosana* (Maki, 1917), *Greenidea* aphid (*Greenidea (Greenidea) decaspermi* Takahashi), Apple aphid (*Aphis pomi* de Geer), Crepe myrtle aphid (*Tinocallis kahawaluokalani* (Kirkaldy), Chrysanthemum aphid (*Macrosiphoniella sanborni* (Gillette), Black Bean Aphid (*Aphis fabae solanella* Theobald, 1914), Cotton Aphid (*Aphis gossypii* Glover, 1877), Mint aphid (*Ovatus crataegarius*), Black bean aphid (*Aphis fabae* Scopoli, 1763).

*Material examined*

8♂, Rawalpindi: N33 38.612' E073 04.476' 1733 ft. elev., 09-02-2016 *Eriobotrya japonica* (Loquat); 5♂, Islamabad: N33 39.656' E073 23.047', 3327 ft. elev., 19-03-2016 *Eriobotrya japonica* (Loquat); 2♂, N33 39.516' E07323.007', 2153 ft. elev., 01-03-2016 *Eriobotrya japonica* (Loquat); 7♂, Chakwal: N32 46.160' E072 42.299', 2209 ft. elev., 29-03-2016 *Eriobotrya japonica* (Loquat); 3♂, N33 40.527' E073 08.376' 1762 ft. elev., 03-03-2016 *Eriobotrya japonica* (Loquat); 5♂, Rawalpindi: N33 38.929' E073 04.943' 1645 ft. elev., 11-03-2016 *Salix* sp. (Willow); 3♂, Islamabad: N35 40.527' E072 08.376' 1763 ft. elev., 13-04-2016; 6♂, Rawalpindi: N33 38.612' E073 04.476' 1733 ft. elev., 09-03-2016 *Salix* sp. (Willow); 2♂, Attock: N32 56.655' E072 51.312', 1663 ft. elev., 09-03-2016 *Salix* sp. (Willow). 5♂, Rawalpindi: N33 39.516' E07323.007', 2153 ft. elev., 24-04-2016; *Ficus* sp. (Fig): 2♂, Rawalpindi: N33 38.612' E073 04.476' 1733 ft. elev., 09-04-2016 *Ficus* sp. (Fig); 5♂, Islamabad: N33

43.929' E07302.179' 3836ft. elev., 17-04-2016 *Ficus* sp. (Fig); 4♂, Rawalpindi: N33 38.929' E073 04.943' 1671 ft. elev., 14-04-2016 *Eugenia jambolana* (Jaman); 4♂, N33 39.516' E07323.007', 2153 ft. elev., 24-04-2016; *Eugenia jambolana* (Jaman); 7♂, Murree: N3355.341' E073 24.216' 6302 ft. elev., 09-04-2016 *Eugenia jambolana* (Jaman); 5♂, Kalar-Kahar: N32 46.138' E072 42.537', 2153 ft. elev., 12-04-2016 *Eugenia jambolana* (Jaman); 3♂, Jhelum: N32 58.119' E073 41.602', 859 ft. elev., 15-04-2016 *Eugenia jambolana* (Jaman); 7♂, Islamabad: N33 40.527' E073 08.376' 1762 ft. elev., 23-02-2016 *Eugenia jambolana* (Jaman); 3♂, Rawalpindi: N33 38.929' E073 04.943' 1645 ft. elev., 12-03-2016 *Psidium guajava* (Guava); 6♂, Islamabad: N33 40.527' E073 08.376' 1762 ft. elev., 03-04-2016 *Psidium guajava* (Guava); 2♂, Rawalpindi: N33 38.612' E073 04.476' 1733 ft. elev., 09-02-2016 *Eriobotrya japonica* (Loquat); 8♂, Islamabad: N33 39.656' E073 23.047', 3327 ft. elev., 19-03-2016 *Eriobotrya japonica* (Loquat); 2♂, N33 39.516' E07323.007', 2153 ft. elev., 01-03-2016 *Eriobotrya japonica* (Loquat); 4♂, Chakwal: N32 46.160' E072 42.299', 2209 ft. elev., 29-03-2016 *Eriobotrya japonica* (Loquat); 4♂, N33 40.527' E073 08.376' 1762 ft. elev., 03-03-2016 *Eriobotrya japonica* (Loquat); 5♂, Rawalpindi: N33 39.655' E073 23.047', 3323 ft. elev., 09-03-2016 *Malus pumila* (Apple); 2♂, Islamabad: (Pir Suhawa) N33 43.929' E07302.179' 3836ft. elev., 17-3-2016 *Malus pumila* (Apple); 6♂, Rawalpindi: N33 38.929' E073 04.943' 1645 ft. elev., 17-08-2016 *Lagerstroemia indica* (Crepe myrtle); 3♂, Islamabad: N33 43.929' E07302.179' 3836ft. elev., 27-07-2016 *Lagerstroemia indica* (Crepe myrtle); 6♂, Chakwal: N32 46.160' E072 42.299', 2209 ft. elev., 29-09-2016 *Lagerstroemia indica* (Crepe myrtle); 4♂, Jhelum: N32 58.118' E073 41.601', 850 ft. elev., 15-07-2016 *Lagerstroemia indica* (Crepe myrtle), 8♂, Rawalpindi: N32 38.928' E074 04.943' 1640 ft. elev., 21-03-2016 *Chrysanthemum indicum* (Chrysanthemum flower); 2♂, Islamabad: N33 40.527' E072 08.375' 1760 ft. elev., 3-04-2016 *Chrysanthemum indicum* (Chrysanthemum flower); 7♂, Rawalpindi: N34 38.612' E073 04.470' 1730 ft. elev., 09-03-2016 *Chrysanthemum indicum* (Chrysanthemum flower); 10♂, Attock: N32 56.655' E072 51.312', 1663 ft. elev., 09-03-2016 *Chrysanthemum indicum* (Chrysanthemum flower); 5♂, Murree: N33 59.652' E073 28.593' 4980 ft. elev., 28-04-2016; 7♂, Chakwal: N32 46.160' E072 42.299' 2209 ft. elev., 29-09-2016 *Chrysanthemum indicum* (Chrysanthemum flower); 3♂, Islamabad: N33 40.527' E073 08.376' 1762 ft. elev., 03-09-2016 *Solanum nigrum* (Black nightshade); 6♂, Rawalpindi: N32 38.928' E074 04.943' 1640 ft. elev., 21-05-2016 *Cassia fistula* (Amaltas); 8♂, Islamabad: N33 40.527' E073 08.376' 1762 ft. elev., 03-09-2016 *Parthenium hysterophorus* (Parthenium weed),

*Capsicum annuum* (Green Chilly), *Abelmoschus esculentus* (Okara); 3♀, Rawalpindi: N33 38.929' E073 04.943' 1671 ft. elev., 02-11-2015 *Eriobotrya japonica* (Loquat); N33 38.929' E073 04.943' 1645 ft. elev., 01-01-2017 *Parthenium hysterophorus* (Parthenium weed), *Duranta erecta* (Golden dewdrop); 6♀, Chakwal: N32 46.160' E072 42.299', 2209 ft. elev., 29-09-2016 *Eriobotrya japonica* (Loquat); 10♀, Murree: N33 55.016' E073 23.699' 6415 ft. elev., 28-02-2016 *Mentha longifolia* (Mint).

#### Comments on ant-aphid associations

This ant species has been recorded in association with 4 aphid species namely, *Aphis gossypii*, *Hysteroneura setariae*, *Myzus persicae* and *Pentalonia nigronervosa* as reviewed by Siddiqui *et al.* (2019). Here we added 11 new trophic associations of this ant with different aphids on different host plants. *Tapinoma melanocephalum* was most abundantly found in association with aphids on a large number of host plants. It was found associated with 12 aphids species on *Mentha longifolia* (Mint), *Parthenium hysterophorus* (Parthenium weed), *Duranta erecta* (Golden dewdrop), *Capsicum annuum* (Green Chilly), *Abelmoschus esculentus* (Okara), *Cassia fistula* (Amaltas), *Solanum nigrum* (Black nightshade), *Chrysanthemum indicum* (Chrysanthemum flower), *Lagerstroemia indica* (Crepe myrtle), *Malus pumila* (Apple), *Eriobotrya japonica* (Loquat), *Psidium guajava* (Guava), *Eugenia jambolana* (Jaman), *Ficus* sp. (Fig) and *Salix* sp. (Willow), from different locations of District Rawalpindi, Islamabad, Attock, Jhelum and Chakwal. All the collected specimens were identified and found similar to the species description of Bingham (1903). *Tapinoma melanocephalum* is reported for the first time in association with any aphid species from Pakistan, so it is added to the country's fauna as new record.

#### Comments on distribution of *Tapinoma melanocephalum* in Pakistan

Wetterer (2009) recorded this ant from Karachi (Sindh Province) of Pakistan. During our studies new distribution records have been added from various localities of Pothwar (Punjab Province). This species is reported for the first time in Punjab.

#### 7. *Tetraponera allaborans* (Walker, 1859)

*Aphid partners in association reported during these studies in Pakistan*

Willow aphid (*Chaitophorus* sp.)

#### Material examined

8♀, Rawalpindi (Neela Sand): N33' 54.762'E072' 28.237' 993 ft, 1846 ft. elev., 13-04-2016 *Salix* sp. (Willow); 5♀, Rawalpindi (Neela Sand): N33' 54.762'E072'

28.237' 993 ft, 1846 ft. elev., 3-05-2016 *Salix* sp. (Willow plant); 15♀, Rawalpindi (Neela Sand): N33' 54.762'E072' 28.237' 993 ft, 1846 ft. elev., 13-04-2017 *Salix* sp. (Willow plant); 5♀, Rawalpindi (Neela Sand): N33' 54.762'E072' 28.237' 993 ft, 1846 ft. elev., 23-05-2017 *Salix* sp. (Willow plant).

#### Comments on ant-aphid associations

This species has been reported in new association with aphid for the first time in the world. Previously it was not reported with any association with aphids. *Tetraponera allaborans* was found associated with Willow aphid (*Chaitophorus* sp.) on *Salix* sp. (Willow plant) from Neela Sand area of district Rawalpindi. This ant along with aphid was present on the branches of willow plant. Ants were collecting honey dew from aphids. All the collected specimens were identified and found similar to the description of Bingham (1903). *Tetraponera allaborans* is reported for the first time in association with any aphid species from Pakistan, so it is added as new country record.

## DISCUSSION

Mutualistic association of ants and aphids has been a topic of interest by various ecologists and taxonomists of the world in the history and recent years. Mutualism is a fundamental element in aphid-ant communities, found within various aphid-ant groups and formulates the species richness, abundance and distribution (Styrsky and Eubanks, 2007). Understanding of the aphid-ant associations generally provides patterns of aphid-ant species complexes in different ecological zones. There is a need of more studies related with taxonomy and ecology for better understanding of aphid-ant associations (Siddiqui *et al.*, 2019).

*Camponotus compressus* with 12, *Formica fusca* with 1, *Formica clara* with 2, *Lepisiota frauenfeldi* with 9, *Myrmica aimonissabaudiae* with 4, *Tapinoma melanocephalum* with 11 and *Tetraponera allaborans* with 1 new associations on various host plants have been reported for the first time from Pakistan. Prior to our studies, only faunal work on aphids and ants separately has been done. These studies will provide baseline for further studies on patterns of ant-aphid mutualistic associations in various ecological zones of Pakistan leading towards their interactions with other communities of ecosystem. New distribution of studied ant species have been added in Punjab province, previously recorded from other provinces.

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*Statement to conflict of interest*

The authors declare there is no conflict of interest.

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