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Population dynamics of walnut aphid, *Chromaphis juglandicola* Kaltenbach, 1843 (Hemiptera: Aphididae) on *Juglans regia* (Walnut) in Kashmir

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Abstract
Regarded as one of the economically important crops of Kashmir, walnut trees are vulnerable to insect pests and diseases leading to great economic losses every year, thus to have knowledge about various factors that leads to it, is indispensable. The objective of the present study was to evaluate the population dynamics in terms of abundance and distribution of *Chromaphis juglandicola* in response to biotic and abiotic factors. Sites were monitored for two years from January 2017 to December 2018, in which five trees were randomly selected from two sites and each site was visited fortnightly. The counted collected specimens were identified and sampling protocol was established by seasonal richness peaks. The data depicted that *C. juglandicola* showed definite seasonal trend with highest population in summer and lowest in autumn with peak population in the month of June. Further, it was observed aphidophagous coccinellids also played a significant role in controlling aphid population. Six beetles were reported during two years of study out of which *Calvia punctata* and *Harmonia diamadata* were most abundant.

Keywords: Walnut, *Chromaphis juglandicola*, biotic, abiotic and aphidophagous coccinellids

Introduction
Walnut is one of the cash crops of Kashmir valley and is important source of foreign exchange. So, it is imperative to understand the biological complexities of walnut ecosystem to improve output yield by pest control and prevent crop loss. Walnuts are prone to various insect pests among which aphids act as the major pests [1]. There is no above and below plant on the ground which is not infested by aphids as they have astounding characteristics to increase ability to act as phytophagous insect pests. Walnut aphid, *Chromaphis juglandicola* is the major pest of walnut worldwide as reported by various scientists and is pest of concern [2, 3, 4]. Aphids deteriorate the plant health by changing the nutrient amount leading to reduced photosynthetic capacity resulting in leaf yellowing, premature death and spotting [5]. Incessant feeding of walnut aphid leads to reduced tree vigour, quality, size of nut and mostly the yield [6, 7]. Further, they discharge copious amount on leaves, nuts & shoots in the form of honey dew which result in the development of black sooty mold fungus. This in turn results in dwindled photosynthetic rate due to reduced light penetration [3]. The population of aphids shows huge dynamics in different seasons and total abundance changes from year to year. They can have many generations with overlapping during vegetative season thus two important contributing factors are production and egg survival rates [8, 9]. The overall population is also restrained due to dynamic changes in environmental conditions in terms of abiotic factors, availability of resources, intraspecific competition and impact of natural enemies mostly predators and parasitoids [10, 11]. Aphidophagous beetles are most abundant biological control agents. The purpose of the present work was to study the population dynamics of *C. juglandicola* colonizing the walnut trees of Kashmir on the background of weather conditions and biological control agent coccinellid beetles.

Material and Methods
Field experiment was carried out in 2017 and 2018 in urban areas of district Srinagar in Kashmir. Two sites were selected, site one (S1) situated in the Kashmir University Botanical Garden (34° 7' 56.94" N & 74° 50' 15.67' E) and other site (S2), Batpora outskirts of Srinagar district (34° 9' 46.12" N & 74° 51' 3.32" E) located at the road side which had ongoing traffic and

human intervention. Sampling was carried out fortnightly in which five trees were randomly selected from both sites and the number of aphids was quantified from the lower lamina of leaves. 20 sub terminal leaves were randomly selected from the lower and middle canopy, which made a total of 100 leaflets/site on each sampling date [12, 3]. In each season mostly from early spring to late autumn winged and wingless specimens were counted in 14 day intervals.

Sampling for natural enemies was done by sweeping and handpicking methods [13]. The net used for collection was made of white muslin cloth with long handle. Hand picking method was mostly adopted for collection. Random sampling was done by choosing 5 walnut trees from each walnut orchard from each location fortnightly. The collected specimens were counted and kept in collection tubes and brought to Entomology Laboratory Department of Zoology for identification.

Identification

The collected specimens of each species were carefully studied for all details under binocular microscope and were identified by running keys and taxonomic work of following workers [14, 15, 16, 17, 18]. Further, during the whole research survey, collected specimens which could not be identified were subsequently sent to ZSI, Kolkata for taxonomic identification. Data was put to analysis by using SPSS Statistical software (Version 20). The data regarding average monthly temperature, rainfall and relative humidity was obtained from Weather Department, Srinagar Jammu and Kashmir.

Results

During the extensive survey at two sites of Srinagar district, the population dynamics of *C. juglandicola* feeding on walnut trees throughout two years (2017 and 2018) of study is presented in Fig.1 while data pertinent to weather parameters is given in Fig.2 (A&B).

In 2017, first sporadic appearance of *C. juglandicola* was observed in the month of April with lowest mean of 18 aphids/100 leaves. With gradual increase in temperature, the number of aphids upturned and a peak mean of 262 aphids/100 leaves were observed in June. Aphid number decreased in first fortnight of July up to the first fortnight of August after which the number again started to increase in second fortnight of August. However, population disappeared in the last days of the second fortnight of October with a minimum mean number of 20.5 aphids/100 leaves. The calculated regression equation was Y= 14.1x−196 having a correlation coefficient (r) 0.290 and p-value (0.528). Positive correlation is exhibited between the temperature and aphid number while negative correlation was found between relative humidity and aphid number with correlation coefficient value of 0.058.

The incidence of *C. juglandicola* at site S2 in the first fortnight of April was 20 aphids/100 leaves. The month of April received incessant rainfall and relatively low temperature hampered the growth of aphids. However, on the onset of May, number of aphids showed a successive increase in population number with a mean abundance of about 157.5 aphids/100 leaves due to an increase in temperature and a small amount of rain. In the month of June, ensuing surge in the number of aphids occurred in the first fortnight which was about 412 aphids/100 leaves and the number dwindled to 230 aphids/100 leaves in the second fortnight while in the month of July plummet in number was observed with a mean of about 96 aphids/100 leaves. The profound decrease in number was due to the escalation of mean temperature of about 30.5 ºC and the minimum average rainfall of 0.9mm. In the month of August, the number showed an increasing trend till the second fortnight of October with a minimum mean number of 28 aphids/100 leaves. The calculated P values for temperature, relative humidity and rainfall were 0.384, 0.621 and 0.714 respectively. The overall result showed a positive significant correlation with temperature having correlation coefficient (r) value of 0.392 and regression equation Y=4.41x-76.7.

In 2018, at site S1 aphid population varied in different months of the year. The count of aphids in the month of April was low compared to last year (2017) with a mean number of 9 aphids/100 leaves. The maximum mean was 238 aphids/100leaves with a sharp plunge of 101.5 aphids/100 leaves in July. Further, the occurrence of aphids lasted up to the second fortnight of October with a mean number of 11aphids/100 leaves. The evaluated result showed that temperature and rainfall had a positive correlation with the aphid population having correlation coefficient (r) of 0.613 and 0.43 while P values were 0.143 and 0.928, respectively. The regression equations for temperature and rainfall were Y=28.4x− 559 and Y=0.19x +179 respectively. RH had a negative correlation with the aphid population (p>0.928) having correlation coefficient (r) of -0.421 and regression equation Y = -12.8x+1102.

At site S2 there was a variation in number of aphids when compared to previous year with a mean of 14 aphids/ 100 leaves in the month of April due to a decrease in temperature and rainfall. An increase in temperature and decrease in rainfall favoured aphid development in the months of May and June. The maximum number of aphids were 285 aphids/100 leaves in the first fortnight of June. With the advent of July, the number was relatively less and continued to decline till the second fortnight with mean number of 73 aphids/100 leaves. Further, in subsequent observations, August showed increased number of aphids compared to September and October. The aphid colonies thoroughly decreased till total wipe out of population in the last fortnight of October. The calculated P-values for temperature, RH and rainfall were 0.043, 0.534 and 0.925 respectively which depicted that temperature had a significant positive correlation with aphid number having regression equation Y= 37.6x−748 compared to RH and rainfall whose P-values were non-significant.

On analysing the population dynamics of aphids in two consecutive years 2017 & 2018 and its comparison to weather parameter (Fig.2 (A&B)), it was found that in both the years, aphids appeared in the first fortnight of April (early spring) while increase in temperature surged the population but beyond 30 ℃, dry periods and mostly rains show negative effect on their growth in the month July. On comparing the aphid population, highest number of aphids were found in the year 2018 while the lowest in the year 2017. Similarly, Site S2 located along the roadside showed higher number of aphids in both years as compared to site S1.

Simultaneously during the present investigation six coccinellid beetles were found on walnut trees. These beetles were identified as *Calvia punctata, Oenopia conglobata, Coccinella septempunctata*, *Harmonia dimidata*, *Adalia tetraspilota* and *Harmonia eucharis*. They were found to be fluctuating throughout the sampling period. *Calvia punctata* and *Harmonia dimidata* were the most encountered species during the present study. Ample population was noted throughout the growing period. Due to very low temperature, there was no occurrence/abundance of any species during the months of January, February, and December. This was observed for both the species in two years. Low population was observed in early march and April but later it gradually increased and attained maximum number during the months of June, July and August, then it again started declining from September until they undergo hibernation. Fig.3 (A-D) showed significant variation in the population index of two coccinellid beetles, *Calvia punctata* and *Harmonia dimidata* during 2017 and 2018.

Discussion

Out of the two aphids namely *P. juglandis and C. juglandicola* infesting walnut trees, *C. juglandicola* was observed dominating species on walnut foliage at both the sites during the study period [19, 20]. Site (S2) located along roadside had maximum aphid population compared to site (S1) which could be attributed to pollution due to vehicular movement compared to parks where the pollution level is low, Wani and Ahmad (2014) [19] and Czechowska *et al*., (1979) [21] suggesting that air pollution had a positive correlation with the development of Hemipteran insects [22]. Roadside trees in urbanized areas had a large number of hemipteran pests infesting them [21, 23]. While other researchers observed that the number of aphids feeding on park trees is always less than the number of aphids feeding on road trees which is in congruity with our results [17, 19, 24]. Sucking pests show high activity on roadside trees when exposed to pollution, causing an increase in nitrogen and amino acid content in their tissues which is very conducive for their development [25]. *C. juglandicola* was found highest in number during the year 2018 due to favourable weather parameters mostly temperature. For the entire study period, population showed changeability concerning weather conditions which played a significant role in the occurrence of the pests. With the advent of spring, they showed sporadic occurrence and warm spring helped in population increase in the month of April. Aphids were recorded from spring to autumn which is consistent with the observations made by Sluss (1967) [26], Cichocka, (1980), [17] Karczmarz (2012) [4] and Wani and Ahmad (2014) [19]. The peak population was observed in the last fortnight of May and June which is in line with the inferences made by Jaskiewicz (2004) [27]. Further, it was observed that with the peak population in June, the aphids undergo population crash which is in corroboration with the results of Frazer and Bosch (1973) [28], Wani and Ahmad (2014) [19] who found that after growth in exponential phase, they endure population breakdown. An increase in temperature in mid-summer had a negative effect on aphid development. Similar reports were made by Kmiec (2007) [29] and Karczmarz (2012) [4]. The results also depicted the decrease in population size when there was a rise in temperature during summer season which was according to the findings of Sluss (1967) [26] and Karczmarcz (2012) [4] who found that temperature above 30℃ limits population development. Temperature and little rainfall showed a positive correlation with the growth of walnut aphids and relative humidity showed a negative role in population change [30, 19]. Walnut aphid population is greatly affected by changing weather parameters as early spring enhances growth. However, the advent of high temperature above 30 ºC impedes further growth. Humidity showed a negative role and little rainfall showed a positive correlation in population dynamics.

In present investigations, the variation in the population index of different coccinellids varied with the change in the locations, temperature and observational years. These findings are in accordance with Ali and Rana (2012) [31], Honek (1985) [32], Kieckhefer and Elliott (1990) [33], Elliott *et al.* (1996) [34] and Hodek and Michaud (2008) [35]. However, other workers working on coccinellid beetles also reported their variation due to population change in aphids [36, 37]. In another experiments variation in population of coccinellids also depend on climate change [38, 39, 36, 40]. However, few reports are also available on variation with change in the plant characteristics [35,41,42]. During present study, on comparing the number of beetles during both the years at two sites, S1 showed good abundance of both beetles than S2. The study also reveals that the predatory coccinellid beetles abundance is negatively correlated with *C. juglandicola* population (Fig. 3 (A-D)). Current study showed site S2 had more number of C. *juglandicola* but low number of predatory beetles which is attributed to the location of S2 which had pollution., as already discussed above walnut trees at site 2 were present along road side which is highly subjected to pollution. The pollution showed negative effect on predatory beetles that resulted in decreased population of beetles. The increase in C. *juglandicola* population is not accompanied by a simultaneous increase of their predatory beetles, as the road side environment is not conducive to their development. These findings are in accordance with Minoranskij and Wojciechowski (1988) and Lubiarz *et al*. (2017) [43, 44] which also reported that with an increasing urbanization pressure the numbers of saprophages, zoophages and some phytophages with biting mouthparts were diminishing, while the numbers of herbivores with piercing-sucking mouthparts were on the increase. Khaliq *et al.* (2014) [45] also showed biotic and abiotic environmental effects on insects and their population dynamics.

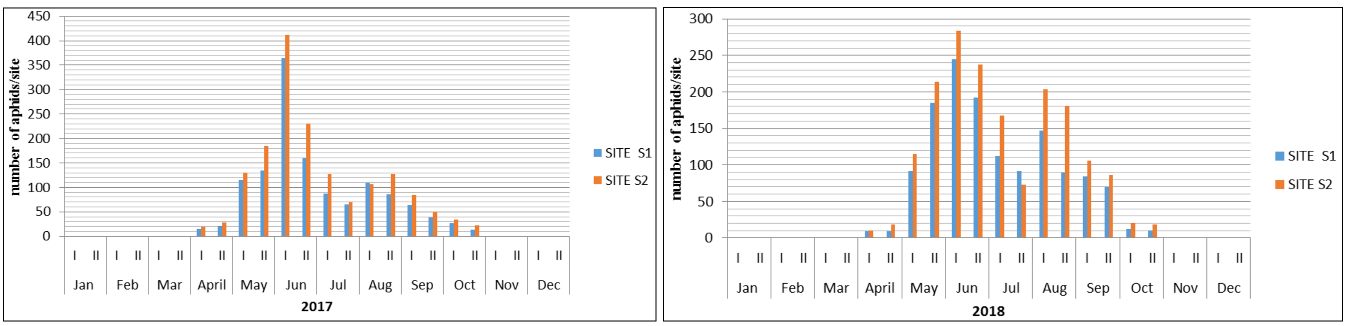

Fig 1: Abundance of *C. juglandicola* on walnut trees in district Srinagar in year 2017 and 2018

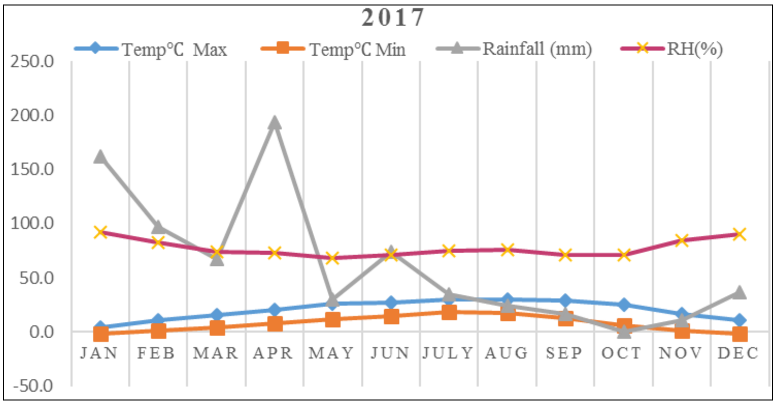

Fig 2 A): Monthly weather report (Temp., Rainfall & RH) of the year 2017 (Source: Meteorological Department, Srinagar J&K)

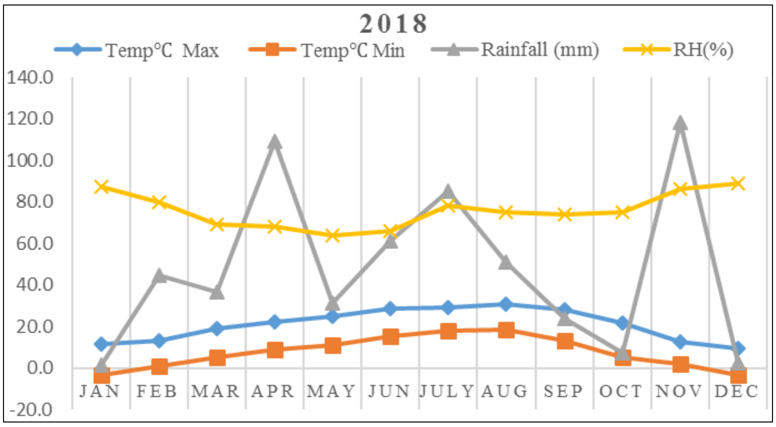

Fig 2 B): Monthly weather report (Temp., Rainfall & RH) of the year 2017 (Source: Meteorological Department, Srinagar J&K)

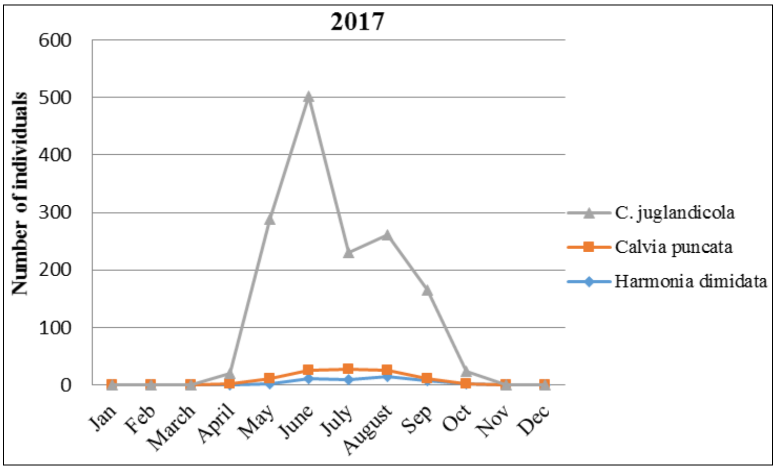

Fig 3 A): Population density of *C. juglandicola* and its predators at site 1

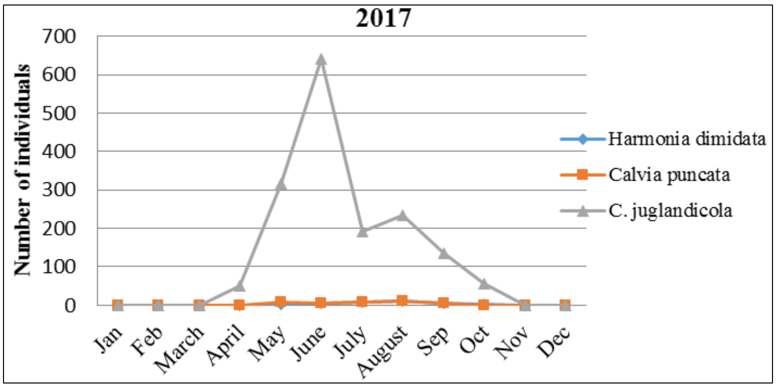

Fig 3 B): Population density of *C. juglandicola* and its predators site 2

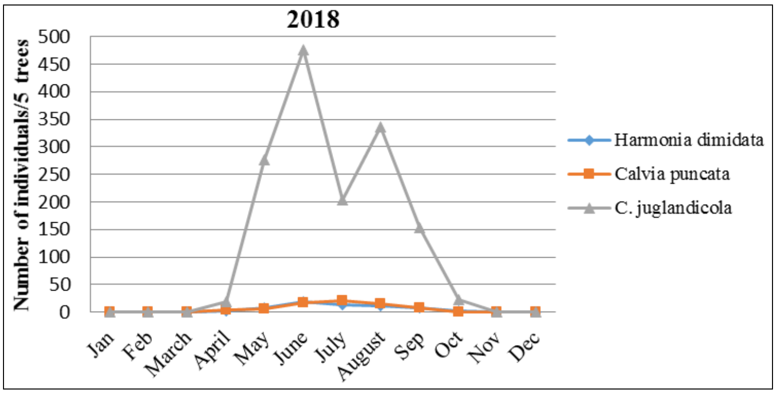

Fig 3 C): Population density of *C. juglandicola* and its predators site 1

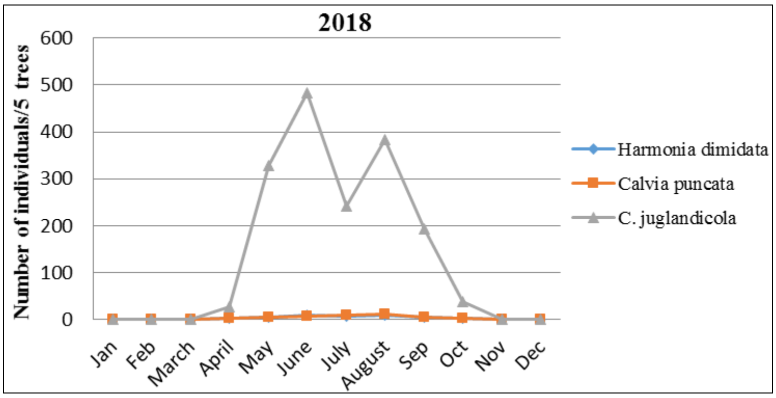

Fig 3 D): Population density of *C. juglandicola* and its predators site 2.

Conclusions

Walnut aphid population is greatly affected by changing weather parameters as early spring enhances growth however, with the advent of high temperature above 30 ˚C impedes further growth. Humidity showed negative role and little rainfall showed positive correlation in population dynamics. The present study revealed the existence of good number of coccinellid species on walnut orchards, which can be used best as biological control against aphid species, *Chromaphis juglandicola.* Therefore, releases should be made in such periods when aphid populations are abundant. The present work also shows the extreme richness of Coccinellid beetles in pollution free site of walnut orchards as compared to polluted site, which depicts that pollution not only affects pests but also natural enemies. Aphids develop rapidly in polluted site and require repeated application of insecticides to control this dreaded pest, thereby developing several side effects *viz.*, environment pollution, low population of natural enemies and toxic residue problems.

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