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Chromosomal Studies on Some Aphid Species from the Shimla Hills of Himachal Pradesh, India

[1]Meena Kumari, [2]Parul Sanaik*, and [3]Sarita Kumari

Author's Affiliation:
[1]Associate Professor in the Department of Biosciences, Himachal Pradesh University Summerhill, Shimla, Himachal Pradesh 171005, India.
E-mail: meenakchaudhary@gmail.com
[2]Research scholar, Department of Biosciences, Himachal Pradesh University Summerhill, Shimla, Himachal Pradesh 171005, India.
E-mail: parulsanaik111@gmail.com
[3]Research scholar, Department of Biosciences, Himachal Pradesh University Summerhill, Shimla, Himachal Pradesh 171005, India.
E-mail: koundalsarita123@gmail.com

***Corresponding author:**
Parul Sanaik
Research scholar in the lab of Cytogenetics and Molecular Genetics, Zoology, Department of Biosciences, Himachal Pradesh University Summerhill, Shimla, Himachal Pradesh 171005, India.
E-mail: parulsanaik111@gmail.com

ABSTRACT:
This paper presents the chromosomal analysis of three aphid species collected from orchards of Shimla, Himachal Pradesh, belonging to the following two subfamilies: Aphidinae and Calaphidinae. Chromosomal studies revealed the highest reported diploid number (2n) as 12 in *Myzus persicae* collected from the Apricot plant, and the lowest reported diploid number (2n) as 8 in *Aphis craccivora* and *Chromaphis juglandicola* infesting Apple and Walnut plant respectively. For, the construction of karyotypes and idiograms, detailed information regarding the mean actual length, total complement length, and relative lengths of chromosomes was calculated by using a scale and ocular micrometer. The karyotypes show a gradual decrease in the lengths of the chromosomes.

Keywords:
Aphids, Karyotypes, Chromosomes, Idiograms, Relative lengths, Calaphidinae.

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INTRODUCTION

Aphids are small soft-bodied insects, the most dreadful, phytophagous pests that feed on the phloem sieve elements (Tjalingii 1995). They are acting as an important model for studying hemipteran-plant interactions. About 5000 aphid species have been recorded worldwide, out of which 450 species are considered to infest crop plants (Blackman & Eastop2017). Aphids are not solely causing damage to the crops by feeding on the plant sap but also by serving as a vector for the transmission of various viral diseases (Kennedy *et al*. 1962). Their holocentric chromosomes and peculiar event of endomeiosis during continuous parthenogenesis (Cognetti, 1961) attribute genetic variability among them thus, make aphids a great subject of interest for research. Unlike most insects, their unique biology includes telescoping of generations,

holocyclic and anholocyclic life cycles, thelytoky, polyphenism, and polymorphism, which are responsible for their substantial increase in the rate of survival and reproductive potential (Mordvilko 1928, Akimoto 1983). Although a lot of studies have been done by many workers on the chromosomes of aphids. However, the polycentric nature of chromosomes and rapid karyotypic evolution open many doors for researchers to explore more about aphids. An attempt has been made here to perform karyotypic studies on the following three aphid species: *Aphis craccivora, Chromaphis juglandicola,* and *Myzus persicae*. This study expands the current cytogenetic information for further study on taxonomy and evolutionary relationships.

METHODS AND MATERIALS

Aphids were collected from the regions of different agro-climatic zone of Himachal Pradesh, Shimla (features a subtropical highland climate, and accounts for 80 percent of Himachal's total apple production).

For cytological studies, only small young embryos were selected and pretreated in a 0.7 % trisodium citrate solution followed by fixation in 1:3 glacial acetic acid and ethanol solutions. After fixation squashing in 45% acetic acid was followed by staining in 2% Giemsa. Then after mounting in xyloldibutyl phthalate (DPX) the permanent slides were inspected using a research binocular microscope.

For the identification of aphid species, keys developed by Blackman and Eastop (2000) were followed. Aphid mounts were prepared by boiling specimens in 95 % alcohol and then, specimens were boiled in a 10% KOH solution followed by 2 to 4 times washing in distilled water. After that, dehydration was done in subsequent higher grades of alcohol. At last, mounting was done in DPX, and species were identified by following the identification and information guide by Blackman and Eastop (2000).

RESULTS

Subfamily Aphidinae:

***Aphis craccivora*:**

A polyphagous species of aphid, which was mainly found to infest the members of the Fabaceae family. Species were collected from the infested soft, tender shoots and leaves of the Spur variety of the Apple plant. This species depicts the 2n=8 (Figure 1). The measurements of chromosomes showed that the actual mean length of the chromosomes ranged from 1.24µm ± 0.032 S.E. in the shortest chromosome to 3.29µm ± 0.023 S.E. in the longest chromosome. The total complement length was 16.86µm ± 0.324 S.E. The relative length of chromosomes ranged from 7.36µm ± 0.181 S.E. in the shortest chromosome and 19.55µm ± 0.101 S.E. in the longest chromosome (Table 2). The Karyotype and idiogram of this species showed a gradual decrease in the length of the chromosomes Figure 2, 3)

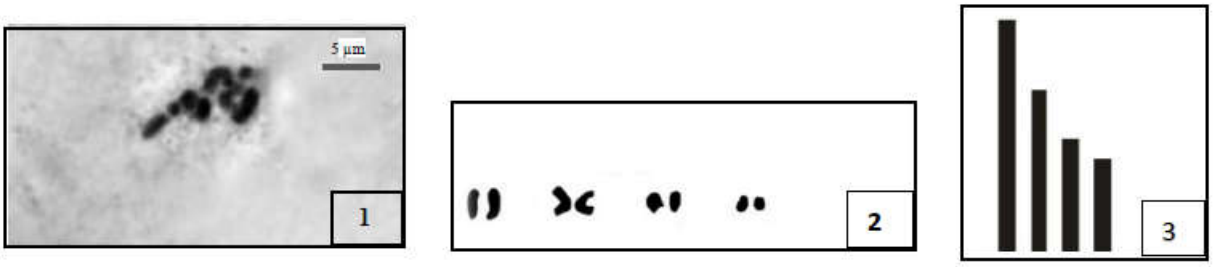
Table 1: Table showing studies of aphid species along with their host plant, Location (Altitude range), and chromosome number.

Sr.No.	Aphid Species	Host Plant	Chromosome number (2n)	Altitude Range	Location
1	*Aphis craccivora*	Apple (Spur Variety)	8	6900 ft.	Kotkhai (Shimla)
2	*Chromaphis juglandicola*	Walnut	8	6900ft.	Kotkhai (Shimla)
3	*Myzus persicae*	Apricot	12	6900ft.	Kotkhai (Shimla)

Table 2: Mean Actual lengths, Mean relative lengths, and total complement lengths of somatic metaphase plates of three aphid species.

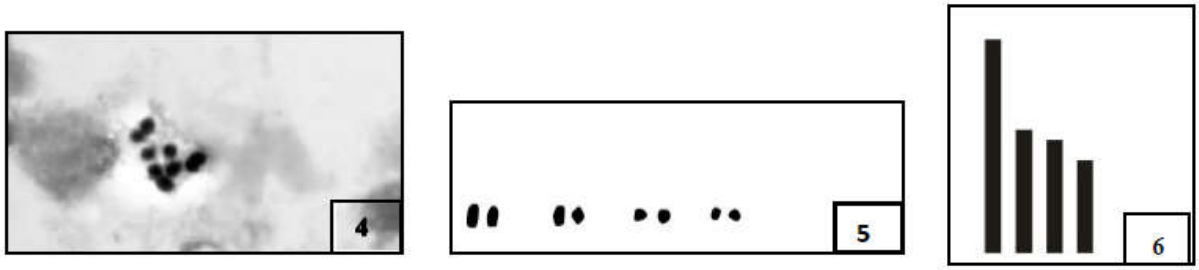
Sr. No.	Aphid Species		Chromosome Numbers												TLC***µm ± S.E
			1	2	3	4	5	6	7	8	9	10	11	12	
1	*Aphis craccivora*	A.L.* (µm) ±S.E.	3.29 ±0.02	3.27 ±0.01	2.29 ±0.03	2.34 ±0.03	1.54 ±0.03	1.72 ±0.02	1.14 ±0.04	1.24 ±0.03					16.86 ±0.32
		R.L.** (µm) ±S.E.	19.55 ±0.10	19.41 ±0.09	13.62 ±0.12	13.88 ±0.16	9.74 ±0.17	10.20 ±0.15	6.77 ±0.23	7.36 ±0.18					
2	*Chromaphis juglandicola*	A.L. (µm) ±S.E.	2.23 ±0.02	2.17 ±0.03	1.29 ±0.02	1.25 ±0.03	1.18 ±0.01	1.14 ±0.01	0.96 ±0.01	0.95 ±0.01					11.21 ±0.10
		R.L. (µm) ±S.E.	19.89 ±0.22	19.38 ±0.20	11.57 ±0.18	11.16 ±0.20	10.58 ±0.08	10.19 ±0.17	8.62 ±0.09	8.53 ±0.14					
3	*Myzus persicae*	A.L. (µm) ±S.E.	3.26 ±0.02	3.06 ±0.02	2.25 ±0.01	2.19 ±0.01	2.09 ±0.01	2.07 ±0.01	1.45 ±0.01	1.42 ±0.01	1.37 ±0.01	1.37 ±0.01	1.15 ±0.01	1.12 ±0.01	22.85 ±0.11
		R.L. (µm) ±S.E.	14.27 ±0.10	13.37 ±0.08	9.85 ±0.06	9.61 ±0.07	9.17 ±0.03	9.07 ±0.05	6.36 ±0.03	6.21 ±0.06	6.01 ±0.06	6.01 ±0.06	5.04 ±0.06	4.90 ±0.04	

* = Actual length,**= Relative length, ***= Total complement length,

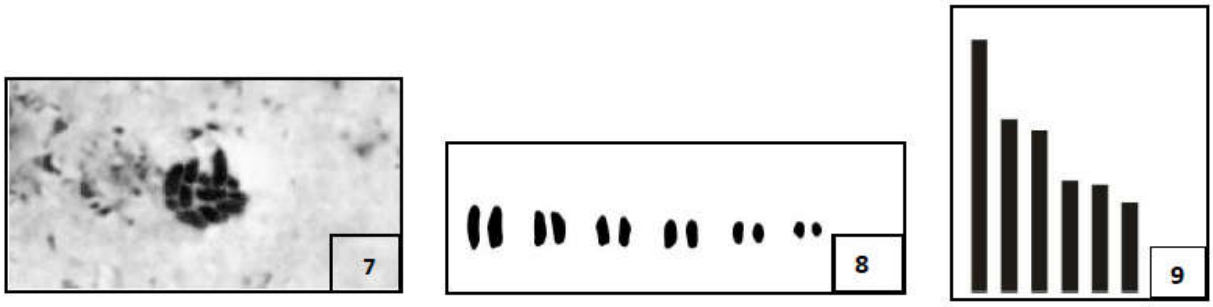


Figures 1-3: ***Aphis craccivora*** **(2n=8). 1. Somatic metaphase plate. 2. Karyotype. 3. Idiogram.**

***Myzus persicae*:**
Light-Green colored aphids of this species were collected from the young leaves of *Pyrus communis*. This ant-attended species was extremely polyphagous in nature. Revealed chromosome number (2n) in *Myzus persicae* was 2n=12 (figure 7). The measurements of somatic metaphase chromosomes showed, that the actual mean length of the chromosomes ranged from 1.12µm ± 0.011 S.E. in the shortest chromosome to 3.26µm ± 0.029 S.E. in the longest chromosome. The mean total complement length was 22.85µm ± 0.112 S.E. The relative length of chromosomes ranged from 4.905µm ± 0.048 S.E. in the shortest chromosome to14.27µm ± 0.101 S.E. in the longest chromosome (Table 2). The idiogram of this species showed a gradual decrease in the length of the chromosomes (Figure 9).



Figures 4-6: ***Chromaphis juglandicola*** **(2n=8). 4. Somatic metaphase plate. 5. Karyotype. 6. Idiogram.**



Figures 7-9: ***Myzus persicae*** **(2n=12). 7. Somatic metaphase plate. 8. Karyotype. 9. Idiogram. Scale = 5µm.**

Subfamily Calaphidinae:

Pale-yellow-colored monophagous species of aphids was collected from the abaxial surface of the leaf. All the members of the species were found to be alate and attended by ants. Reported diploid chromosome number (2n) in *Chromaphis juglendicola* was eight (Figure 4). The measurements of chromosomes in ten somatic metaphase plate showed, that the actual mean length of the chromosomes ranged from 0.95µm ± 0.016 S.E. in the shortest chromosome to 2.23µm ± 0.025 S.E. in the longest chromosome. The mean total complement length was 11.21µm ± 0.107 S.E. The relative length of chromosomes ranged from 8.53µm ± 0.149 S.E. in the shortest chromosome to 19.89µm ± 0.220 S.E. in the longest chromosome (Table 2). The idiogram of this species showed a gradual decrease in the length of the chromosomes (Figure 6).

DISCUSSION

The present work has been undertaken to study the diploid chromosome number and karyotypes of three species of the following two subfamilies: Aphidinae and Calaphidinae.

Subfamily Aphidinae

Gosh, (1989) from the Indian region reported 464 species in 105 genera lying under the subfamily Aphidinae. Of the three species reported in the present study, 2 species belong to this subfamily. Out of 2 species, one belongs to the genus *Aphis*, which is considered the largest aphid genus comprising nearly 500 species (Eastop and Hille Ris Lambers, 1976).

In the present study, *Aphis craccivora* was found to infest the Spurr variety of Apple plants. This species has characteristic tongue-shaped cauda with tubular siphunculi which are almost equal to cauda in length. The head vertex has a smooth capsule and poorly developed antennal tubercles. The chromosome number in *Aphis craccivora* was found to be eight, the same number of chromosomes was also reported by other workers on the different host plants (Kurl, 1978 on *Vigna catjang* and *Lathyrus sativus*;

Kulkarni and Kacker, 1979 on *Cassia fistula*; Dutta and Gautam, 1993 on *Citrus* species; and Guleria and Gautam, 2015 on *Punica granatum*).

The genus *Myzus* comprises of 55 species (Blackman and Eastop, 1984). Several workers such as Shinji, 1941; Coling, 1955; Gut, 1976; Robinson and Chen, 1969; Kuznetsova, 1969; Blackman, 1980b; Khuda-Bukhsh and Pal, 1985, reported chromosome numbers in the twenty-two species belonging to the genus *Myzus*, where in seventeen species they found same chromosome number (2n) i.e. twelve.

The present study revealed diploid chromosome number 2n=12 in the *Myzus persicae* collected from the *Pyrus communis,* which is in accordance with the reports of earlier workers (Sethi & Nagaich, 1972; Blackman, 1971, 1980a; Kurl, 1985; Dutta & Gautum, 1993; Guleria & Gautum, 2015). The species was characterized by the presence of prominent converging antennal tubercles, finger-shaped cauda, and cylindrical siphunculi which were longer than cauda.

Subfamily Calaphidinae

There are 358 described species belonging to 62 genera of this subfamily. Members of the Calaphidinae have an annual life cycle without any host alternation, Most aphid species of this subfamily feed on woody angiosperms.

Only a single species of this subfamily *Chromaphis juglandicola* has been discussed here. All the members of *Chromaphis juglandicola* are alate which is in conformity to a characteristic feature of this subfamily that is in summer, all the tree-dwelling species occur entirely as active alate morphs. They have distinctive long antennae (dark brown bands) which are about half of their body length, cauda is pale and knobbed and siphunculi are conical in shape. Diploid Chromosome number (2n) is eight, which was in agreement with the earlier reports of Dutta & Gautam (1993) and Blackman & Eastop (1994).

CONCLUSION

Aphids are gregarious pests that are affecting agricultural and horticultural crops and causing potential damage to crop plants. Keeping their complex cytogenetics in view, three species of aphids *Aphis craccivora* (2n=8), *Chromaphis juglandicola* (2n=8), and *Myzus persicae* (2n=12) have been observed to study their karyomorphology. The presence of centromeric activity all along the chromosome contributes to the karyotypic variation in their chromosome number or the total complement length of chromosomal plates. These studies help ascertain the correct taxonomic position of closely related species.

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