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Comparative Karyotypic Analysis of Four Aphid Species Infesting Economically Important Horticultural Plants

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ABSTRACT:
Chromosomes of four aphid species infesting different horticultural crops were investigated from the North-Western Himalayas. The aphid species studied included *Aphis pomi* de Geer, a destructive pest of apple plants, *Brachycaudus helichrysi* (Kaltenbach), associated with peach *Chromaphis juglandicola* (Kaltenbach), infesting walnut and *Eriosoma lanigerum* (Hausmann), a well-known pest of apple orchards. The karyotypic studies deduced the diploid chromosome number (2n) of 8 in *A. pomi* and *C. juglandicola* , and 12 in *E. lanigerum* and *B. helichrysi.* . The chromosomes in all studied species were holocentric in nature. Comprehensive measurements of metaphase chromosomes were made to determine the mean actual length, total complement length, and relative length of individual chromosomes. Based on relative length data, idiograms were constructed for each species to illustrate their chromosomal organization and facilitate comparative cytogenetic analysis.

Keywords:
Aphid, Karyotype, Idiogram, Diploid Chromosome Number.

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INTRODUCTION

Aphids are a group of phytophagous pest insects with approximately 5000 species worldwide and about 450 species known to infest crop plants, with particular prevalence in the temperate zone of the northern hemisphere (Blackman & Eastop, 2000; Favret C & Aphid Taxon Community, 2025). Among these, about a hundred species of aphids are implicated with substantial damages to the agricultural and horticultural crops with significant drop in yield and quality depletion (Blackman & Eastop, 1984; Van Emden & Harrington, 2007). Not only these tiny soft bodied insects cause destruction by feeding directly on the phloem content of the host plant, but also inflict indirect damage by vectoring plant viral diseases (Kennedy et al., 1962). Due to several peculiar attributes of their life cycle such as such as cyclic parthenogenesis,

vivipary and telescoping of generations (Dixon, 1985) these tiny insects have drawn global interest.

Aphid life cycle show great variability in response to the environmental conditions, and can be either Holocyclic, if both parthenogenetic generations and sexual generation are involved in the annual cycle or anholocyclic, with only parthenogenetically reproducing generations existing throughout the year (Blackman, 1974; Wohrmann & Tomiuk, 1988; Moran,1989,1992).

Aphid cytogenetics is very complex due to their holocentric chromosomes and occurrence of endomeiosis during continuous parthenogenesis (Cognetti, 1961) The presence of diffused centromere, thereby fusion and fragmentation of chromosomes, results in frequent karyotypic variations (Steffan, 1968; Kuznetosva, 1968).

The rapid changes in the karyotypic configuration of the aphid genome serves as an important factor in the evolution of new biotypes. The karyotypic analysis of the aphid species from various temperate regions will be a valuable tool in taxonomic identifications and ascertaining the evolutionary scale of aphid species as the chromosome number is closely associated with physical features of aphid species. Cytogenetic investigations on aphids have been reported by several workers from different regions of the Northwestern Himalayas (Dutta & Gautam, 1993; Gautam & Kumari, 2003; Potan & Gautam, 2018; Kumari et al., 2022; Kumari et al., 2023; Kumari & Kumari, 2023; Sharma et al., 2024; Choudhary et al., 2024). However, no cytogenetic studies have been reported from the selected region of the Northwestern Himalayas, District Sirmour. Keeping this in view, chromosomes of four aphid species infesting plants of economic importance were investigated in the present study.

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MAETRIALS AND METHODS

The aphid species were collected from apple, peach, and walnut orchards located in the North-Western Himalayas regions, specifically the Shivalik Hills of Sirmour district, Himachal Pradesh (Table 1). The collected aphid species were identified using standard taxonomic keys developed by Blackman and Eastop (1984). Apterous, parthenogenetic, viviparous females were selected for cytogenetic analysis. The apterous, parthenogenetic and viviparous females were subjected to the cytogenetic slide preparation. The posterior end of the abdomen of the female was carefully punctured to bring out the embryos, followed by pretreatment of the embryos in 0.7% sodium citrate solution and fixation in 1:3 acetic acid-ethanol solution. The embryos were then placed in a drop of 45% acetic acid on a glass slide, squashed gently and stained in 2% Giemsa solution after drying. From the well-spread metaphase plates, the actual lengths of the chromosomes were recorded using an ocular micrometer. The data was utilized to calculate total complement length (TCL) and relative lengths. Idiograms were constructed from the relative length data

Table 1: Aphid species along with host plant, altitude and place of collection

S. No.	Aphid species	Host plant	Altitude	Chromosome number (2n)	Place of collection
1	*Aphis pomi*	Apple (Spur Variety)	1700m	8	Ghanduri, Sirmour
2	*Brachycaudus helichrysi*	Peach	1700m	12	Ghanduri, Sirmour
3	*Chromaphis juglandicola*	Walnut	1100m	8	Kharkan, Sirmour
4	*Eriosoma lanigerum*	Apple (Spur Variety)	1700m	12	Ghanduri, Sirmour

RESULTS

Aphis pomi de Geer

The young apple sapling and the abaxial (lower) surface of the leaves of the apple tree, *Malus domestica*, were both heavily colonised by green apple aphids. Apterae individulas were bright green to yellowish green in colour, characterised by dark siphunculi and cauda. Karyotypic studies reveal holocentric chromosomes with 2n = 8 (Fig. 1 & 2). The actual length of the chromosomes varied from 0.90 µm ± 0.09 for the shortest chromosome to 2.21 µm ± 0.12 for the longest chromosome. The mean total complement length was 12.64 µm ± 0.76. The relative length of chromosomes ranged from 7.04 ± 0.36 for the shortest chromosome to 17.75 ± 1.10 for the longest chromosome (Table 2). The idiogram of this species is marked by a gradual decrease in the length of the chromosomes (Fig. 3).

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

Sr. No.	Aphid Species	Chromosome Numbers													TCL***(µm) ± S.E.
			1	2	3	4	5	6	7	8	9	10	11	12	
1	*Aphis pomi*	A.L.* (µm) ± S.E.	2.21 ± 0.12	2.21 ± 0.12	1.83 ± 0.12	1.83 ± 0.12	1.39 ± 0.16	1.39 ± 0.16	0.90 ± 0.09	0.90 ± 0.09					12.64 ± 0.76
		R.L.** (µm) ± S.E.	17.75 ± 1.10	17.75 ± 1.10	14.47 ± 0.49	14.47 ± 0.49	10.74 ± 0.79	10.74 ± 0.79	7.04 ± 0.36	7.04 ± 0.36					
2	*Brachycaudus Helichrysi*	A.L.* (µm) ± S.E.	1.82 ± 0.11	1.82 ± 0.11	1.48 ± 0.09	1.48 ± 0.09	1.37 ± 0.07	1.37 ± 0.07	1.24 ± 0.05	1.24 ± 0.05	0.92 ± 0.03	0.92 ± 0.03	0.70 ± 0.03	0.70 ± 0.03	15.07 ± 0.71
		R.L.** (µm) ± S.E.	12.03 ± 0.27	12.03 ± 0.27	9.80 ± 0.17	9.80 ± 0.17	9.07 ± 0.11	9.07 ± 0.11	8.26 ± 0.12	8.26 ± 0.12	6.17 ± 0.20	6.17 ± 0.20	4.68 ± 0.11	4.68 ± 0.11	
3	*Chromaphis juglandicola*	A.L.* (µm) ± S.E.	1.95 ± 0.26	1.95 ± 0.26	1.57 ± 0.18	1.57 ± 0.18	1.28 ± 0.13	1.28 ± 0.13	0.93 ± 0.10	0.93 ± 0.10					11.45 ± 1.25
		R.L.** (µm) ± S.E.	16.67 ± 0.73	16.67 ± 0.73	13.73 ± 0.24	13.73 ± 0.24	11.31 ± 0.34	11.31 ± 0.34	8.29 ± 0.69	8.29 ± 0.69					
4	*Eriosoma lanigerum*	A.L.* (µm) ± S.E.	1.70 ± 0.12	1.70 ± 0.12	1.30 ± 0.09	1.30 ± 0.09	1.16 ± 0.05	1.16 ± 0.05	1.03 ± 0.03	1.03 ± 0.03	0.92 ± 0.05	0.92 ± 0.05	0.76 ± 0.06	0.76 ± 0.06	13.72 ± 0.61

		R.L.** (µm) ± S.E.	12.35 ± 0.68	12.35 ± 0.68	9.41 ± 0.33	9.41 ± 0.33	8.49 ± 0.18	8.49 ± 0.18	7.53 ± 0.21	7.53 ± 0.21	6.72 ± 0.26	6.72 ± 0.26	5.50 ± 0.35	5.50 ± 0.35	

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

***Brachycaudus helichrysi* (Kaltenbach)**

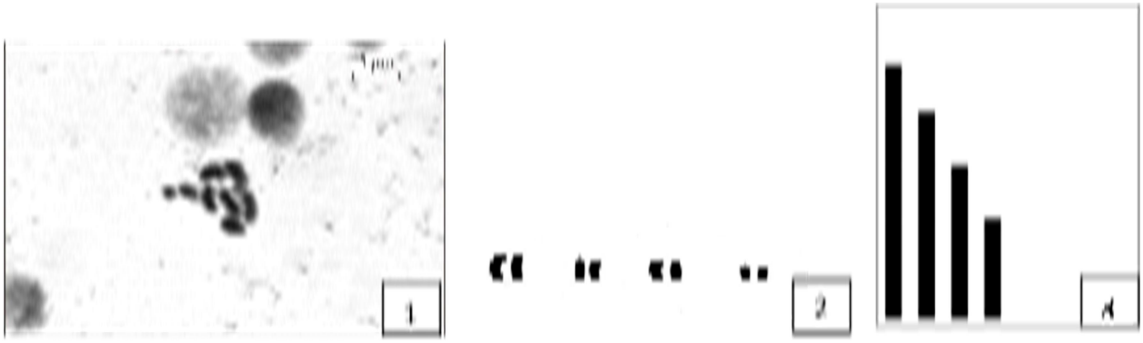
The aphid infestation *of B. helichrysi* was recorded on the leaves of peach plants ,*Prunus persica*. Apterae were pale yellow to greenish in colour, and colonies induced characteristic pseudogall formation on hoist leaves. The diploid chromosome number was found to be 12 (Fig. 4 & 5). The mean actual lengths of the shortest and the longest chromosomes were 0.70 µm ± 0.03 and 1.82 µm ± 0.11, respectively, and the total complement length was 15.07 µm ± 0.71. The relative length of chromosomes ranged from 4.68 ± 0.11 to 12.03 ± 0.27 (Table 2). The idiogram in Fig. 6 shows a gradual decrease in chromosomal length.

***Chromaphis juglandicola* (Kaltenbach)**

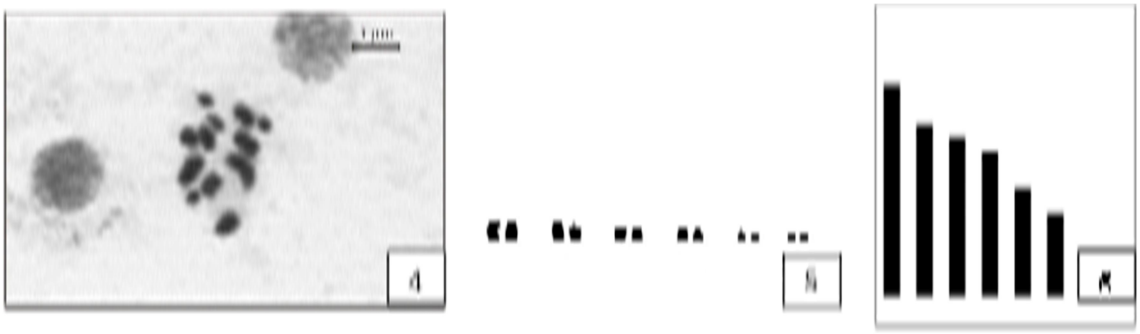
The alate viviparae of *C.juglandicola* were found on the undersides of the leaves of the walnut, *Juglans regia*. The aphids were observed pale yellow and scattered all over the surface of the leaf. The diploid chromosome number observed was 8 (Fig. 7 & 8), and the mean actual lengths of the shortest and the longest chromosomes were 0.93 µm ± 0.10 and 1.95 µm ± 0.26, respectively, and the total complement length was 11.45 µm ± 1.25. The relative length of chromosomes ranged from 8.29 ± 0.69 to 16.67 ± 0.73 (Table 2). A gradually decreasing pattern is observed for chromosomal lengths in the idiogram (Fig. 9)

***Eriosoma lanigerum* (Hausmann)**

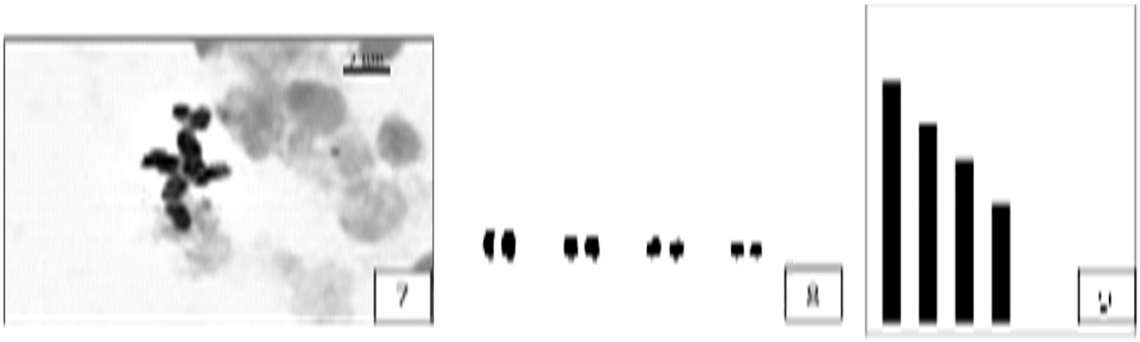
Dense woolly colonies of *E. lanigerum* were observed on nodes, branches, and trunk of apple (*Malus domestica,*spur variety), where the infestation resulted in localized deformation of plant tissues. The apterae were brown in color and covered with a dense layer of waxy coating. The diploid chromosome number for this species was found to be 12 (Fig. 10 & 11). The mean actual lengths of the shortest and the longest chromosomes were 0.76 µm ± 0.06 and 1.70 µm ± 0.12, respectively and the total complement length was found to be 13.72 µm ± 0.61. The relative length of chromosomes ranged from 5.50 ± 0.35 to 12.35 ± 0.68 (Table 2). A gradually decreasing pattern is observed for chromosomal lengths in the idiogram (Fig. 12)



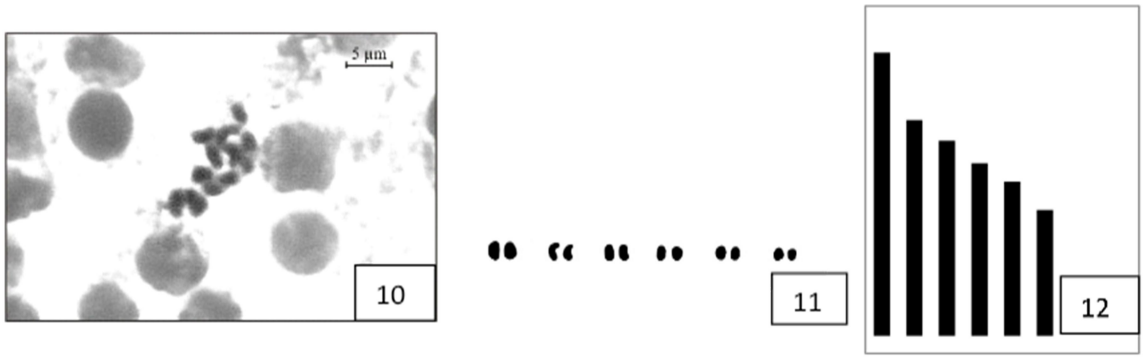
Figures 1-3: ***Aphis pomi*****. 1. Somatic chromosomes. 2. Karyotype. 3. Idiogram.**



Figures 4-6: ***Brachycaudus helichrysi*****. 4. Somatic chromosomes. 5. Karyotype. 6. Idiogram.**



Figures 7-9: ***Eriosoma lanigerum*****. 7. Somatic chromosomes. 8. Karyotype. 9. Idiogram.**



Figures 10-12: ***Chromaphis juglandicola*****. 10. Somatic chromosomes. 11. Karyotype. 12. Idiogram.**

(Scale = 5 µm)

Figsure 1- 12: Aphid Karyotypes

DISSCUSSION

The present investigation focused on karyotypic analyses of four aphid species infesting major horticultural crops, namely, *A. pomi* de Geer, the green apple aphid, *B. helichrysi* (Kaltenbach), the leaf-curling plum aphid, *C. juglandicola* (Kaltenbach), the small walnut aphid, & *E. lanigerum* (Hausmann), the woolly apple aphid. All species examined exhibited holocentric chromosomes, a characteristic cytogenetic feature of aphids. The diploid chromosome number was determined as 2n = 8 in *A. pomi* and *C. juglandicola*, whereas 2n = 12 was recorded in *B. helichrysi* and *E. lanigerum*. The results obtained corroborate the conserved chromosomal arrangement commonly observed in these aphid species. *A. pomi* De Geer is a widely distributed pest of apple, where it causes considerable damage to the fruit directly and indirectly (Blackman & Eastop, 1984; Oatman & Legner, 1961; Hamilton et al., 1986; Gautam & Kumari, 2004). Gautam and Kumari (2003) published the first cytogenetic studies of *A. pomi* in India, reporting a diploid chromosomal number of 2n = 8. The current study's chromosome count is entirely consistent with these previous findings, verifying the species' chromosomal stability across geographical regions.

B. helichrysi (Kaltenbach) is a polyphagous species known to infest a wide variety of plants species especially members of the families Compositae and Boraginaceae, and commonly associated with waxy colonies on *Prunus* spp. (Favret C & Aphid Taxon Community, 2025). In the present study, individuals of this species were collected from curled leaves of peach plants and diploid chromosome number was determined as 2n = 12 which is in agreement with the previous reports by Kulkarni & Kacker (1981a), Gautam & Sharma (1990) & Dutta & Gautam (1993), indicating chromosomal consistency within the species.

The active members of C. juglandicola (Kaltenbach) are predominantely alate & monophagous, feeding specifically on walnut hosts. The Diploid Chromosome number (2n) recorded in this study for *C. juglandicola* is eight, the same has been reported by other workers (Dutta & Gautam, 1993; Blackman & Eastop, 1994; Kumari et al., 2023). This congruence provides additional evidence that the number of chromosomes in this species is stable across geographic areas. *Eriosoma lanigerum* (Hausmann) is a globally distributed pest of apple, characterized by the formation of dense woolly colonies on aerial parts and roots of host plants. The present study confirmed a diploid chromosome number of 2n = 12 for *E. lanigerum*, demonstrating strong conformity with earlier cytogenetic studies conducted by Robinson and Chen (1969a), Kulkarni and Kacker (1980), and Gautam and Verma (1982). The consistency of chromosome number across independent studies highlights the cytogenetic uniformity of these economically important species. Thus, the karyotypic consistency found in this work provides important cytogenetic information for aphid populations living in the regions of Sirmour District, Himachal Pradesh . These studies are essential for comprehending species stability, taxonomic connections among aphid groups, and chromosomal evolution. They also lay the groundwork for further genetic and ecological research.

CONCLUSION

Aphids constitute economically significant pests of horticultural crops and exhibit distinctive cytogenetic characteristics, particularly the presence of holocentric chromosomes. The present investigation provides cytogenetic insights into four aphid species infesting horticultural crops of different regions of Sirmour District, Himachal Pradesh. Detailed karyotypic analyses revealed the presence of holocentric chromosomes in all studied species, with stable and species-specific diploid chromosome numbers. The diploid chromosome number (2n) was recorded as 8 in *Aphis pomi* de Geer and *Chromaphis juglandicola* (Kaltenbach), while 2n = 12 was observed in *Brachycaudus helichrysi* (Kaltenbach) and *Eriosoma lanigerum* (Hausmann). For sustainable horticulture production, these results provide a scientific basis for improving species identification and creating pest management plans specific to individual regions.

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Authors Contribution
Reeba conducted the laboratory work, Meenakshi and Swati Mahajan assisted in field work and Meena Kumari and Reeba drafted the manuscript. All authors reviewed and endorsed the final version of the manuscriopt.

Conflict Of Interest
The authors declare that no competing interests exist.

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