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Original article (Orijinal araştırma)

Karyotype analysis of *Phauloppia lucorum* (Koch, 1841) (Oribatida: Oribatulidae)

Phauloppia lucorum (Koch, 1841)'un karyotip analizi (Oribatida: Oribatulidae)

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Abstract

Currently, about 11,000 oribatid mites have been described, but chromosome numbers have been reported for only a few species. Furthermore, the oribatid mites are a valuable model for holocentric chromosomes in cytogenetic studies. The diploid chromosome number is generally 2*n* = 18 in oribatid mites, though some have 2*n* = 16 or 30. Although there are many studies on the morphology and systematics of *Phauloppia lucorum* (Koch, 1841) (Oribatida: Oribatulidae), there is no information about its karyotype or chromosome numbers. The aim of this study is to investigate the chromosome number, monoploid ideogram and detailed chromosomal measurements of *P. lucorum*. The diploid chromosome number of *P. lucorum* was found to be 2*n* = 12. The total haploid chromosome length and the average chromosome length were 7.39 µm and 1.23 µm respectively. The chromosome lengths varied from 0.91 to 1.67 µm. In conclusion, these results are the first for the chromosome numbers and karyotype analysis for *P. lucorum*.

Keywords: Acari, holocentric chromosome, karyotype, Oribatida, *Phauloppia lucorum*

Öz

Günümüzde, 11,000 oribatid akar türü tanımlanmıştır fakat çok az sayıda türün kromozom sayısı bildirilmiştir. Ayrıca oribatid akarlar sitogenetik çalışmalarda holosentrik kromozomlar için değerli bir modeldir. Genellikle oribatid akarlarda diploid kromozom sayısı 2*n* = 16, 30 gibi bazı istisnalar dışında 2*n* = 18 şeklindedir. *Phauloppia lucorum* (Koch, 1841) (Oribatida: Oribatulidae) üzerine çok sayıda morfolojik ve sistematik çalışmalar bulunmasına rağmen karyotip ve kromozom sayısı hakkında bilgi yoktur. Bu çalışmanın amacı *P. lucorum*'un kromozom sayısı, monoploid ideogram ve detaylı kromozom ölçümlerini araştırmaktır. *P. lucorum*'un diploid kromozom sayısı 2*n* = 12 olarak bulundu. Toplam haploid kromozom uzunluğu ve ortalama kromozom uzunluğu sırasıyla 7.39 µm, 1.23 µm'dir. Kromozom uzunluğu 0.91-1.67 µm aralığında değişmektedir. Sonuç olarak, *P. lucorum*'un kromozom sayısı ve karyotip analizi ilk kez bildirilmiştir.

Anahtar sözcükler: Acari, holosentrik kromozom, karyotip, Oribatida, *Phauloppia lucorum*

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Introduction

Oribatid mites are the dominant microarthropod group in forest soil-litter habitats. They play an important role in the decomposition of the soil organic matter. Oribatid mites are compose of 11,036 described species and subspecies found throughout the world (Subías, 2004, updated 2018; Gulvik, 2007; Ayyıldız & Toluk, 2016).

The species, *Phauloppia lucorum* (Koch, 1841) (Oribatida: Oribatulidae), is distributed in the Holarctic Region (and frequently in the Western Palearctic) and in Nepal (Subías, 2004, updated 2017). The diagnostic characteristics of this species are: integument color yellowish brown, length of body 690 (680-710) µm, width 480 (460-510) µm (n = 6), rostrum rounded, rostral setae setiform, lamella reduced as a weak line, costula thin, sensillus short and head claviform, pteromorpha not developed, notogaster with four pairs of porose areas and 14 pairs of setae, epimeral setal formula 3-1-3-3, four pairs of genital setae, two pairs of anal setae, all legs tridactylous (Figure 1).

Although many oribatid mites have been described, chromosome numbers have been reported for only a few species. Furthermore, the oribatid mites are a valuable model with holocentric chromosomes in cytogenetic studies, although not the only model, as about 800 species have been reported to have holocentric chromosomes including plants, nematodes, arachnids and insects (Melters et al., 2012). In general, the diploid chromosome number in oribatid mites is $2n$ = 18 (Oliver, 1977; Norton et al., 1993) with some exceptions having $2n$ = 16 or 30 (Heethoff et al., 2006; Eroğlu & Per, 2016). Although there are many studies on morphologicy and systematics of *P. lucorum*, there is no information about the karyotype or chromosome numbers in the literature. The aim of this study was to investigate the chromosome number, monoploid ideogram and detailed chromosomal measurements of *P. lucorum*.

Material and Methods

Material

The specimens were collected from Turkey: Sakarya, Kılıçkaya Hill, 40º28.214' N, 30º25.027' E, 551 m, in soil under *Pinus* sp., 22.IV.2015, 2 exs (mounted on aluminum stubs and gold-coated for scanning electron microscopy); 40º30.257' N, 30º28.830' E, 1302 m, in soil, 19.VI.2015, 3 exs; 40º30.204' N, 30º27.484' E, 1170 m, in soil, 19.VI.2015, 1 ex.; 40º29.200' N, 30º25.860' E, 896 m, in soil under *Pinus* sp., 21.VI.2015, 1 ex.; 40º29.388' N, 30º23.028' E, 463 m, in lichen on *Pinus* sp., 01.XI.2015, 1 ex.; 40º28.809' N, 30º23.872' E, 721 m, in lichen on *Pinus* sp., 06.XI.2015, 1 ex.; 40º29.207' N, 30º23.763' E, 660 m, in soil under *Pinus* sp., 06.XI.2015, 2 exs; 40º28.940' N, 30º23.523' E, 606 m, in lichen on *Pinus* sp., 06.XI.2015, 1 ex.; 40º28.900' N, 30º23.510' E, 598 m, in lichen on *Pinus* sp., 06.XI.2015, 1 ex. All materials were collected by Sedat Per (Figure 2).

Cytogenetic procedure

The cytogenetic procedure was conducted using the method developed by Imai et al. (1988) with substantial modifications by Gokhman & Quicke (1995). The procedure used on 10 specimens for which sex was not determined: (i) the hypotonic sodium citrate solution (1%) with colchicine (0.005%) for pretreatment and crushing; (ii) the fresh hypotonic solution for incubation; (iii) the fixative series for fixation, fixative 1 (glacial acetic acid-ethanol-distilled water, 3-3-4), fixative 2 (glacial acetic acid-ethanol 1-1), fixative 3 (glacial acetic acid); (iv) Giemsa staining.

At least 10 mitotic plates were assessed to determine the number of diploid chromosomes. A qualified photomicrograph was taken using a DP72 digital camera mounted on an Olympus BX-53 light microscope. The holocentric chromosomes were measured in micrometers using KaryoType software (Altınordu et al., 2016). The ideogram was drawn based on total chromosome lengths in order from largest to smallest.

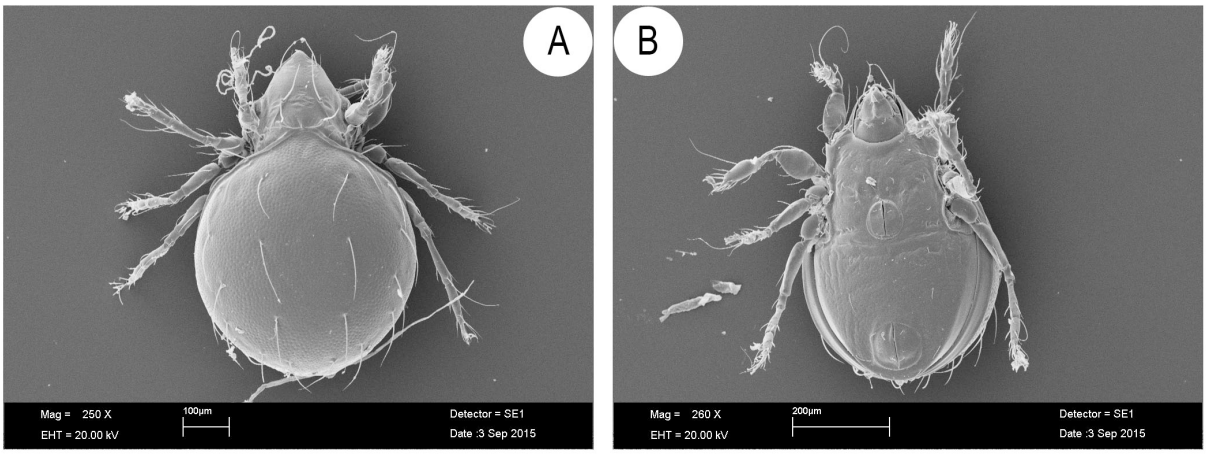

Figure 1. *Phauloppia lucorum*: A) dorsal view, and B) ventral view.

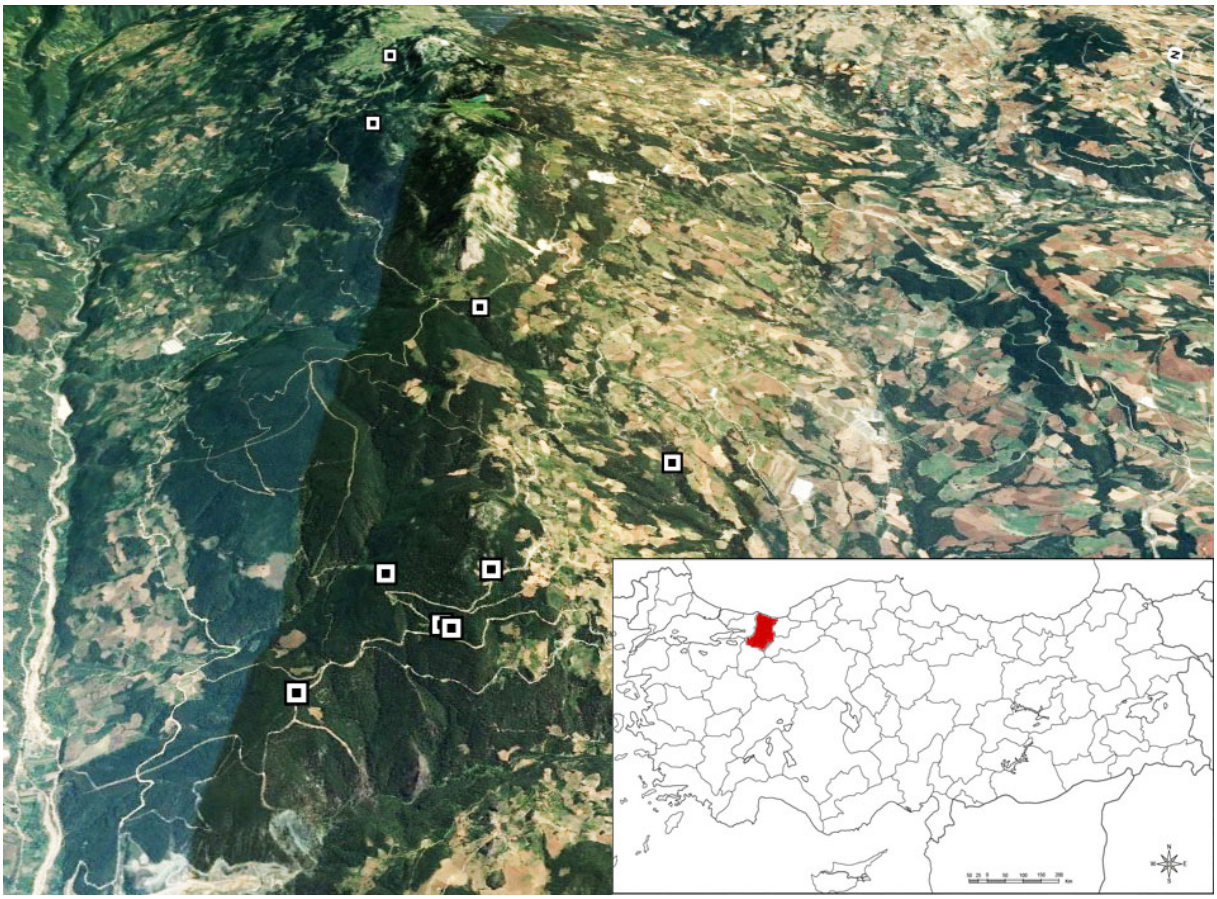

Figure 2. Collection locations of *Phauloppia lucorum* on the Kılıçkaya hill (Sakarya Province, Turkey).

Results

The diploid chromosome number of *P. lucorum* was determined to be 2*n* = 12 and the mitotic metaphase chromosomes are shown in Figure 3. The chromosome lengths and monoploid ideogram are given in Table 1 and Figure 4, respectively. The karyotype consists of holocentric chromosomes. The karyotype formula could not be determined due to the holocentric chromosomes.

The total haploid chromosome length and the average chromosome length were 7.39 and 1.23±0.27. The chromosome lengths varied from 0.91 to 1.67. The satellite was not observed in the chromosomal observations.

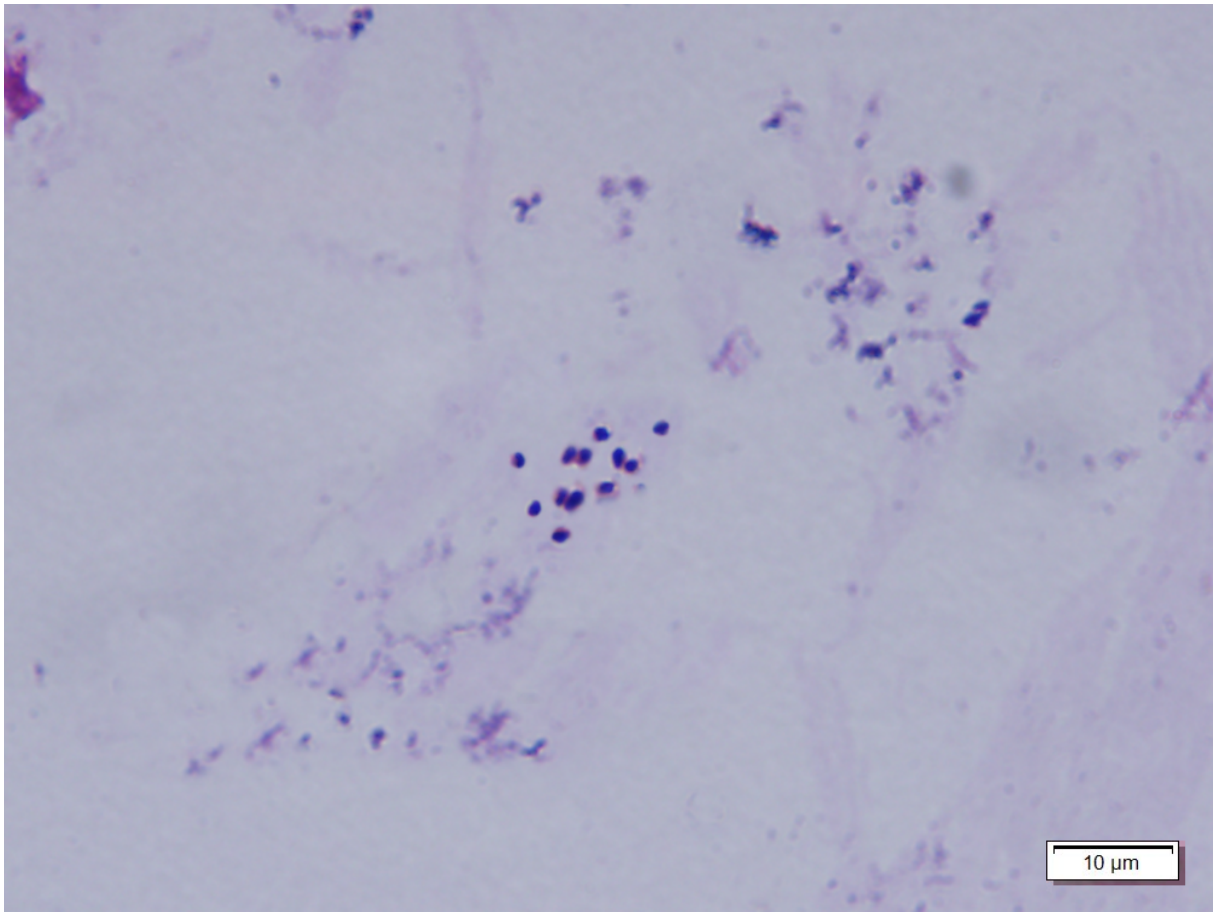

Figure 3. Photomicrograph of mitotic metaphase chromosomes in *Phauloppia lucorum*.

Table 1. The total chromosome lengths of *Phauloppia lucorum*

Chromosome Pair	1	2	3	4	5	6
Length (µm)	1.67	1.40	1.24	1.14	1.03	0.91

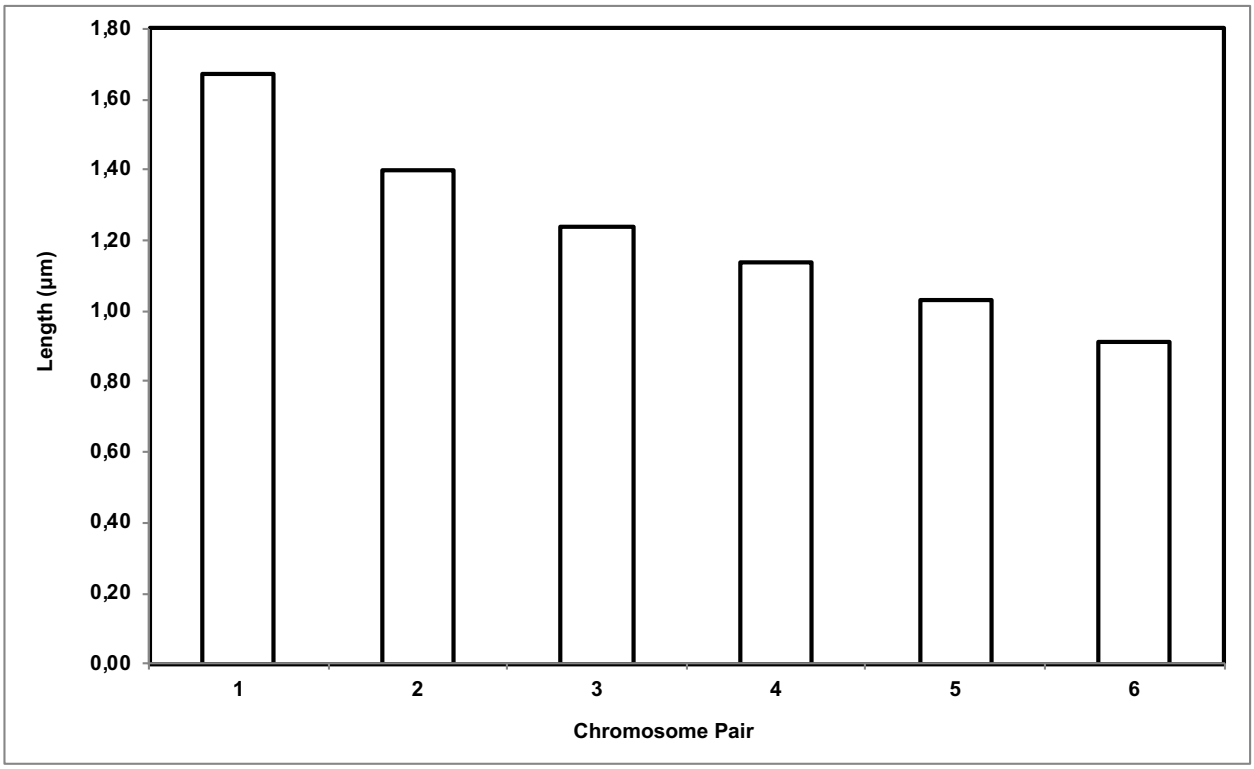

Figure 4. The monoploid ideogram of *Phauloppia lucorum*.

Discussion

Chromosomal parameters are important contributors to the understanding of evolutionary relationships, when used in conjunction with morphological or molecular techniques. Some important chromosomal parameters are basic chromosome number, diploid chromosome number (2*n*), karyotype formula, total haploid length and karyotype asymmetry. The S/AI formula is used to calculate karyotype asymmetry in highly organized animals (Eroğlu, 2015). The karyotype formula and the karyotype asymmetry could not be determined due to the holocentric chromosomes in *P. lucorum*. They are parameters specific to monocentric chromosomes. Although the diploid chromosome number of *P. lucorum* is 2*n* = 12, this not common in the oribatid mites, it is close to the general karyotype reports of the order as *Archegozetes longisetosus* Aoki, 1965, *Galumna* sp. It is reported that the common chromosome number is 2*n* = 18 in oribatid mites (Norton et al., 1993; Heethoff et al., 2006). As a broader example, mites and ticks have two to 36 chromosomes (Oliver, 1977). Eroğlu & Per (2016) found that the chromosome number of one oribatid mite, *Zygoribatula cognata* (Oudemans, 1902), is 2*n* = 30.

The chromosomes of *P. lucorum* are small holocentric chromosomes. The holocentric chromosomes do not show a localized centromere, which is the thin waist-like structure seen in eukaryotic chromosomes. Although monocentric chromosomes are much more common, the holocentric chromosomes have the broad phylogenetic distribution. Melters et al. (2012) reported that the holocentric chromosomes evolved at least nine different times in animals and four different times in plants. The holocentric chromosomes are small-sized chromosomes ranging from 0.5 to 2.0 µm (Wrensch et al., 1994). *Phauloppia lucorum* has small holocentric chromosomes (range 0.91-1.67). Many other arthropods, such as Lepidoptera, Hemiptera and Odonata, also have holocentric chromosomes (White, 1973; Heethoff et al., 2006). Holocentric chromosomes can provide some advantages. For example, sensitivity to radiation infertility is lower in butterflies compared to other insect groups. The main reason for this durability is that butterflies have holocentric chromosomes (North, 1967). After radiation, each fragment that separates from the holocentric chromosomes acts as a separate chromosome and will not

be lost in the anaphase (Lachange, 1967). Another advantage is that very different meiotic adaptations are needed for organisms to adopt holocentric chromosomes. Some of these adaptations are restriction of kinetochore activity, inverted meiosis and asymmetric meiosis (Melters et al., 2012).

The sex chromosomes could not be determined in *P. lucorum*. In generally, the oribatid mites have weak sexual dimorphism in size; and strong dimorphism is rare (Behan-Pelletier, 2015). The sexual dimorphism is a physical difference between two sexes of the same species other than in the sexual organs. There are three reproduction models in the Acari; thelytoky, haplodiploidy and diplodiploidy. It has been reported that the diplodiploidy is the ancestoral reproduction model in mites (Norton et al., 1993; Wrensch et al., 1994). Generally, in diplodiploidy, the young mites (male and female) are produced from fertilized eggs and the sex ratio is almost equal (1:1). However, the order Oribatida is characterized by a similar karyotype with the absence of sex chromosomes (Heethoff et al., 2006). Unlike sexual oribatid mites, the male proportion is very low (males rare) in parthenogenetic species and the rare males are generally sterile. Some factors may affect the proportion of rare male in parthenogenetic species. Environmental conditions are one of the most important of these factors and may induce the production of males (Chang et al., 2017).

This study reports for the first time the chromosome numbers and karyotype of *P. lucorum*. As there are many species for which chromosome data is unknown among oribatid mites, more chromosomal data are needed to support to the cytotaxonomy of oribatid mites.

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