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A new cytological record of two scarab beetle species from the genus *Gymnopleurus* Illiger, 1803 (Coleoptera: Scarabaeidae)

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ABSTRACT

Karyological investigations on two *Gymnopleurus* spp.; *Gymnopleurus miliaris* (F.) and *Gymnopleurus parvus* (MacL.) are under taken in the present study. Meioformula of *G. miliaris* (17AA+Xyp) and G. *parvus* (8AA+Xyp) are presented. None of these beetles exhibited a modal polyphagan karyotype. However, Xyp was found to be the male sex chromosome mechanism. Most chromosomes are acro and subtelocentric. Change in the number and size of chromosomes in the karyotypes is suggestive of the Robertsonian fusion having played some role in the evolution of karyotype in this genus. The evolutionary trends in *Gymnopleurus* have been discussed.

Keywords: *Gymnopleurus,* Karyotype, Chromosomal rearrangements, Scarab beetle.

1. Introduction

The cosmopolitan superfamily Scarabaeoidae is an ancient group which comprises about 35,000 different species [6]. It is a very complex taxon in which the impressive diversity among species and groups has been marked in morphological, ethological and ecological studies. On the contrary, data available about the chromosomes of Scarabaeoidae are very scanty, comprising the haploid or diploid numbers of about 200 species and karyotype description of some of them [11, 12, 16, 19]. The genus *Gymnopleurus* Illiger, 1803 (Scarabaeidae: Scarabaeinae), whose distribution is Palearctic, Afrotropical and Oriental [8] comprises about 50 species [3], but only five of them have been partly investigated from karyological point of view. Reports like 2n=18 for *G. geoffroyi* [1], 2n=20 for *G. sturmi* [6], haploid number n=10 for *G. koenigii* [7], 2n=20 for *G. cyaneus* [9] and the diploid number 2n=18 for *G. sinatus* [10] have been given. Since information on these insects were meagre as compared to other genera of polyphagan beetles, the present communication deals with the chromosomal analysis of two species of *Gymnopleurus* by means of Giemsa staining. This is a new cytological record of the subtribe Gymnopleurina, whose phylogeny is still controversial [4].

Table 1: Chromosomal analysis of *Gymnopleurus* spp.

S.no.	Species	Diploid number (2n)	Meioformula	Reference
1	*G. koenigii*	20	9AA+Xyp	Dasgupta, 1963
2	*G. cyaneus*	20	9AA+Xyp	Kacker, 1976
3	*G. sturmi*	20	9AA+Xyp	Colomba *et al.*, 2000
4	*G. sinatus*	18	8AA+Xyp	Manna & Lahiri,1972
5	*G. geoffroyi*	18	8AA+Xyp	Angus *et al.* 2007
6	*G. parvus*	18	8AA+Xyp	Present report
7	*G. miliaris*	36	17AA+Xyp	Present report

2. Materials and methods

Sexually mature male specimens of *G. miliaris* (F.) and *G. parvus* (MacL.) were collected from Seonti forest, Kurukshetra (Haryana, India) in months of August and September, 2009. All the specimens were identified following the guidelines laid down by Arrow [2]. Chromosomal preparations were obtained, using the air drying method [17] from male gonads. Evaluation of chromosomal morphology was based on ten meiotic metaphases. Percentage relative length of chromosomes was also calculated. Spermatogonial metaphase, meiotic stages and interphase nuclei were analysed, photographed and karyotypes have been prepared.

3. Results

3.1. *Gymnopleurus miliaris* (F.) 2n=36

Spermatogonial metaphase was characterised by the presence of 36 chromosomes (Fig.1a). The karyotype comprised of 17 pairs of autosomes and sex chromosomes X and y (Fig. 1b). Autosomal pairs 1to 3 are metacentric, pairs 4 to 6 are submetacentric and remaining 11 pairs are acro and subtelocentric. The autosomes showed a gradual decrease in size. The X chromosome is subtelocentric whereas y is metacentric and smallest. Percentage relative length of autosomes varied from 1.55 to 10.88 whereas that of X is 9.13 and y is 1.4 (Table 2). Metaphase I revealed 17 autosomal bivalents and the sex parachute (Fig. 2). During spermatogonial anaphase the chromosomes separate and move synchronously to the opposite poles (Fig. 3). The sex chromosomes also move along with autosomes and do not show any differential behaviour. Due to chromatid separation the morphology of the chromosomes is very clear at metaphase II (Fig. 4). The haploid number at metaphase II and number of bivalents at metaphase I tally with the diploid number counted at spermatogonial metaphase.

Table 2: Percentage relative length of chromosomes of *Gymnopleurus* spp.

Chromosomal pairs	**Species names with percentage relative length of chromosomes**	
Pair No.	***Gymnopleurus miliaris***	***Gymnopleurus parvus***
1	10.88	16.02
2	9.63	12.73
3	8.2	12.28
4	7.76	8.23
5	7.21	8.2
6	6.94	7.96
7	5.19	6.75
8	5.5	6.44
9	4.88	-
10	4.89	-
11	3.98	-
12	3.57	-
13	3.2	-
14	2.25	-
15	1.68	-
16	1.55	-
17	1.57	-
X	20.31	15.92
y	3.2	5.41

3.2. *Gymnopleurus parvus* (MacL.) 2n=18

Spermatogonial metaphase possessed 2n=18 (Fig. 1c). The karyotype comprises two pairs of metacentric (pair 1 & 3), one pair of submetacentric (pair 2) and five pairs of acro and subtelocentric (pairs 4-8) autosomes and subtelocentric X and metacentric y sex chromosomes (Fig. 1d). X chromosome is next in size to the first pair of autosomes whereas y being the smallest element of the component. Percentage relative length of autosomes varied from 6.44 to 16.02 whereas that of X is 15.92 and y is 5.41 (Table 2). During diplotene stage the bivalents become distinctly visible. They appeared as long and thick filaments, but chiasmata were not discernable (Fig. 5). At metaphase I the bivalents are in the form of rings and rods (Fig. 6). The parachute shape of sex bivalent is not very clear. Metaphase II plate with 8 autosomes and one X chromosome was encountered (Fig. 7). Due to chromatid separation the metacentric, submetacentric and acrocentric nature of the chromosomes is very clear at this stage.

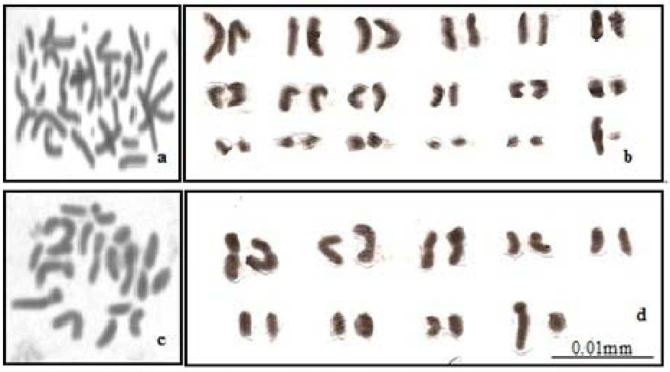
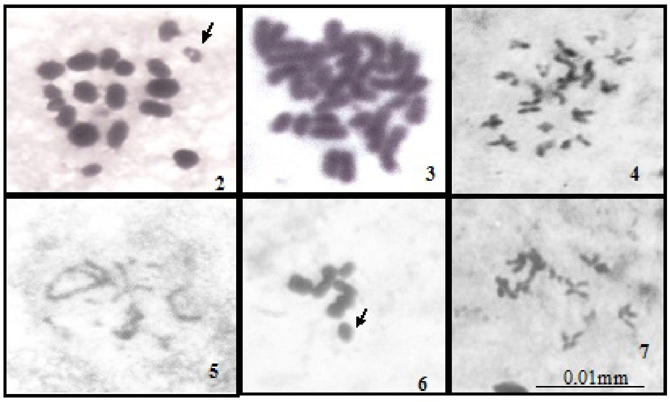


Fig: 1a,b *G. miliaris*; 1c,d: *G parvus*. a&c: spermatogonial metaphase; b&d: karyotypes.



Figs 2-4. *G. miliaris* **Fig. 2.** Metaphase I, arrow shows Xyp; **Fig 3.** Spermatogonial anaphase I; **Fig. 4.** Metaphase II; **Figs. 5-7** *G.parvus* **Fig. 5.** Diplotene stage; **Fig. 6.** Metaphase I, arrow indicates Xyp; **Fig. 7.** Metaphase II.

4. Discussion

According to some of the reports, Scarabaeinae (Coprinae) is less conservative subfamily of Scarabaeidae. Unlike other scarabs there are much more variations in the form and size of chromosome numbers [18]. But as in most of other scarabs the sex chromosome mechanism in Scarabaeinae is mainly Xyp type. In the present chromosomal analysis two species under genus *Gymnopleurus* are new cytological record of Scarabaeinae. A perusal of literature on *Gymnopleurus* in Table 1, indicated the chromosome number of *G. parvus* (2n=18) matched with that of *G. geoffroyi* [1] and *G. sinatus* [10]. The diploid number of *G. koenigii* [7], *G. cyaneus* [9] and *G. sturmi* [6] is 2n=20 which is the modal number of polyphagan beetles. But unlike all other species, *G. miliaris* is characterised by presence of a large number of chromosomes with 2n=36. The chromosome number, however, shows a wide variation from 2n= 18 to 2n=36. Consequently, both increase and decrease in chromosome number has occurred during evolutionary history. Species with especially large number of chromosomes, about double the base number, as occur in *G. miliaris* (present report), the question has been raised by some of the researchers, whether polyploidy has played a role in such numerical increase [13, 15]. But both of these reports were rejected due to the failure to encounter neo Xy systems which constitute stepping stone to multiple formations.

The karyotype of *G. miliaris* possessed 11 pairs of acrocentric chromosomes and shows much difference in percentage relative length between the largest, RCL=10.88 and smallest chromosome RCL=1.57 (Table 2). This condition was also reported by Vorontsov [14] in species *Philonthus intermedius* with 2n=56, of family Staphylinoidae which is another phyletic branch of the superfamily Scarabaeoidae, thus suggesting that such a feature is quite common within the taxon. The variability in autosome number in such species may suggest the occurrence of extensive karyotypic reorganisation leading to a more stable karyotype at least at autosomal level.

Acrocentry being the primitive evolutionary behaviour of the chromosomes, the karyotype of *G. miliaris* with many pairs of acrocentric chromosomes can be interpreted as ancestral to the karyotype of other species of this genus. In the evolution of karyotype in the genus *Gymnopleurus*, Robertsonian fusion of autosomes as well as more complicated rearrangements of chromosomal material seem to have played a leading role in decrease in the number of acrocentric pairs and increase in the meta or submetacentric pairs of chromosomes in the karyotypes of species with diploid number 20 or 18 and with conserved sex chromosome mechanism of Xyp type [5].

5. Conclusion

At last we have concluded that the possibility of polyploidy playing a vital role in the numerical increase of chromosomes during evolution of genus *Gymnopleurus*. Robertsonian fusion of autosomes played a major role in decrease in the transformation of acrocentric pairs in some species into metacentric pairs in other species of the same genus. The situation, however, will become clearer when more species will be investigated cytologically and by the use of banding techniques to ascertain the involvement of constitutive heterochromatin.

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