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Genetic code complement and the distribution of constitutive heterochromatin of grasshoppers

Pooja Clara and Anupam M. D

Department of Zoology, Guru Nanak Dev University, Amritsar, Punjab, Pin Code: 143005, India.

Abstract

Chromosomes with detailed karyotypic information (nature, number, size, relative length, length of X-chromosome, nature of X-chromosome) and C-banding patterns of six species of grasshoppers belonging to sub-families- Tryxalinae, Oedipodinae and Catantopinae are discussed. The karyotypes comprises of acrocentric chromosomes with complement number 2n=23 (male). Constitutive heterochromatin distribution was found at centomeric, interstitial, terminal sites along with thick and thin bands among all the species except in *Acrida turrita*, which possessed only centromeric C-bands. The number and location of C-bands in Acridids exhibit both intra – and interspecific variations. In the present communication the chromosome complement and C-banding patterns are analyzed for further differences between congeneric species and among genera belonging to the same sub-family.

Keywords: Acrididae, Orthoptera, chromosome complement, C-banding.

INTRODUCTION

Orthoptera has been considered as a classical material for karyological investigations. The size and number of their chromosomes are such that both qualitative and quantitative studies on chromosomal anomalies can be detected easily (Turkoglu and Koca, 2002). The karyotype is found to have a cytotaxonomic value. Acridoid group is known for its karyotypic uniformity or conservatism (Aswathanarayana and Ashwath, 2006). The introduction of C-banding technique offers a simple mean of defining constitutively heterochromatic regions.

C-banding technique have made it easier to assess the changes in constitutive heterochromatin and have revealed the existence of remarkable degree of C-band variations within species (King and John, 1980; Lopez-Fernandez and Gosalvez, 1981). Many studies on the C-bands have been done in Acridoids which are known to possess high level of chromosomal variations. The number and location of C-bands in Acridids exhibit both intra- and interspecific variations (Yadav and Yadav, 1993). The present communication deals with the chromosome complement and the distribution of constitutive heterochromatin, along with the differences between congeneric species and among genera belonging to same sub-family are discussed.

MATERIALS AND METHODS

The males of 6 species Acrididae were collected in and around Guru Nanak Dev University campus, Amritsar (Punjab). The testes were excised following standard Colchicine-Hypotonic-Cell suspension-Flame dry technique (Yadav and Yadav, 1983). The flame dried slides were treated for C-banding following method of Sumner (1990) with slight modifications. The chromosomes were classified after Levan et al. (1964) and appropriate karyotypes were prepared.

RESULTS

The perusal of Table 1 revealed that, among the total six species studied; 4 species belong to sub-family Tryxalinae and these are *Acrida turrita* Linn., *Acrida exaltata* Walk., *Phlaeoba infumata* Brunn., *Phlaeoba antennata* Brunn. and one species *Oedaleus abruptus* Thunb. belong to sub-family Oedipodinae and *Oxya velox* (Fabr) belong to sub-family Catantopinae. The diploid chromosome number was found to be 23 and

Table 1. Nature of chromosomes and morphometric characters in 6 species studied.

S.No.	Species	Male 2n number	Chromosome size L	M	S	*Range of relative length of autosomes	Length of X- chromosomes	Nature of chromosomes m sm a	Nature of X-chromosome
1	Sub-family: Tryxalinae *Acrida turrita*	23	3	6	2	19.9 ± 0.39 144.2 ± 0.48	199.0 ± 2.26	__ __ All	Largest
2	*Acrida exaltata*	23	3	6	2	35.1 ± 0.36 128.9 ± 1.32	148.4 ± 0.42	__ __ All	Largest
3	*Phlaeoba infumata*	23	3	6	2	38.0 ± 0.72 128.2 ± 0.46	141.0 ± 1.18	__ __ All	Largest
4	*Phlaeoba antennata* Sub-family: Oedipodinae	23	3	6	2	23.2 ± 0.32 139.0 ± 1.02	151.1 ± 1.05	__ __ All	Largest
5	*Oedaleus abruptus*	23	3	6	2	22.7 ± 0.38 113.6 ± 1.02	136.3 ± 1.27	__ __ All	Largest
6	Sub-family: Catantopinae *Oxya velox*	23	3	6	2	26.3 ± 0.6 149.0 ± 2.52	175.4 ± 1.52	__ __ All	Largest

sex-determining mechanism was found to be XO: XX type, among all the species investigated. Table 1 shows the various morphometric characters of all the species investigated. It was ascertained that, the chromosome morphology is acrocentric for all the species. X-chromosome is the marker, as it is the largest element among all the species studied. Figure 1 shows the constructed karyotypes of all the species respectively.

Table 2 and Figure 2 are showing the position of constitutive heterochromatin among male grasshoppers studied. It was observed that, the centromeric bands of constitutive heterochromatin were found among all the species under study. The interstitial bands were seen in *A. exaltata*, *P. infumata*, *P. antennata* only. Terminal bands were seen in all the species investigated except in *A. turrita*. Thick and thin bands were seen in all the species of hoppers except in A*. turrita*.

DISCUSSION

The karyology of every species is unique in itself and provides an identity to species (Channaveerappa and Ranganath, 1997). The short horned grasshoppers are characterized bypossessing acrocentric chromosomes. Due to great cytogenetic uniformity, the short horned hoppers are considered as an example of "Karyotypic conservatism" (Aswathanarayana and Ashwath, 2006). In the present study, 6 Acridids have been investigated among which 4 belong to sub-family Tryxalinae and one each belonging to sub-family Oedipodinae and Catantopinae, respectively. It is revealed that, hoppers belonging to family Acrididae have 23 chromosome number. The sex-determining mechanism is found to be XO/XX type among all the studied species. Yadav and Yadav (1986) reported similar results in relation to chromosome number and sex-mechanism among Haryana population of

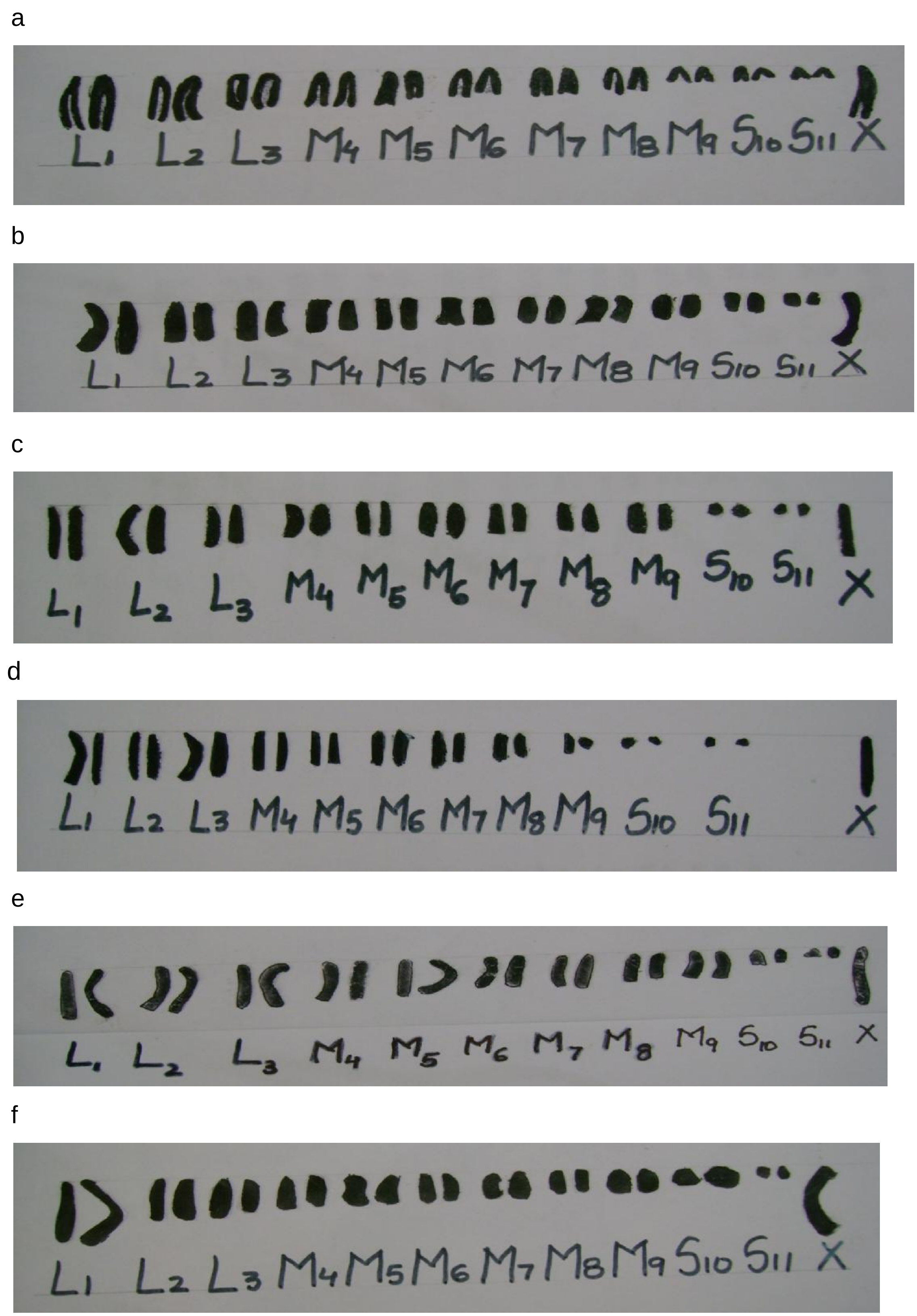

Figure 1. Karyotypes of (a) *A. turrita* (b) *A. exaltata* (c) *P. infumata* (d) *P. anteenata* (e) *O. abruptus* (f) *O. velox*.

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Table 2. Showing the position of C-heterochromatin in male grasshopper species under present investigation.

Species	**C-banding sites**				
Super-family: Acridoidea	**Centromeric**	**Interstitial**	**Terminal**	**Thick band**	**Thin band**
Sub family: Tryxalinae *Acrida turrita*	1-11,X	——	——	1,4,6,7	——
Acrida exaltata	1-11,X	1,X	1,2,4	1,2,3,4,5,6,7,8,9,X	3,4
Phlaeoba infumata	1-11,X	3,5,7,X	1,6,7	1,2,3,5,6,7,X	1,4
Phlaeoba antennata	1-11,X	2	4,5	1,2,3,4,X	1,3,6
Sub family: Oedipodinae *Oedaleus abruptus*	1-11,X	___	4,5	3,5,6,8,9,X	1,2,5,6,7
Subfamily: Catantopinae *Oxya velox*	1-11,X	___	1,5,X	1,2,3,4,6,7,X	2,3,5,7,8

Acridoideans. While studying the chromosomes of 11 species of grasshoppers from Simla (H.P), Sharma and Gautam (2002) also revealed similar results. So, the short horned grasshoppers of different regions are showing cytogenetic uniformity, regarding chromosome number and sex-determining mechanism.

During the present investigation, the chromosomes are found to be acrocentric in nature. Up to six metacentrics through fusions have been reported in Tryxalines *Myrmeleotettix maculates* (John and Hewitt, 1966) and *Stauroderus scalaris* (John and Hewitt, 1968). Meanwhile, Aswathanarayana and Ashwath (2006) observed a series of structural changes involving 6th, 7th and 9th pair, exhibiting hetero and homomorphism in *Gastrimargus africanus* orientalis. Mayya et al. (2004) reported short arms in chromosomes of *Aiolopus thalessimus* tumulus and *Acrotylus humbertianus*. Whereas, no such change have been reported in present study. The X-chromosome is found to be largest of all the other chromosomes among the 6 species investigated. But in the population of *O. velox* from Himachal Pradesh, the length of X-chromosome was found to be between L1 and L2 (Sharma and Gautam, 2002). Mayya et al. (2004) also reported the X-chromosome to be largest in all the species except in *A. thalessimus* tumulus and *Spathosternum prasiniferum*. Yoshimura et al. (2005) explored 4 species of Oxya (*Oxya hyla* intricate, *Oxya japonica* japonica, Oxya *chinensis* formosana and *Oxya yezoensis*) and reported that X- chromosome was the longest of medium sized pairs of chromosomes in all the *Oxya* sp. But in our studies, X- chromosome is the largest and marker in *O. velox*. The C-band represents the constitutive heterochromatin in the homologous chromosome of a karyotype. This type of DNA consist of short repeated polynucleotide sequences. C-bands exhibit centromeric, interstitial and terminal sites. It is a variant state of chromatin and said to be genetically inert (Yunis and Yasminieh, 1971). C-banding pattern in various species of grasshoppers provide important clues that have occurred during the course of evolution. Many studies have shown a remarkable degree of C-band variation.

In the present study, C-bands are also found to be present in three locations, that is centromeric, interstitial and terminal. But their extent is found to vary among the species studied. All the nine species of Acridids studied possess centromeric C-bands. Yadav and Yadav (1993) and Mayya et al. (2004) reported the prevalence of centromeric bands in short horned grasshoppers as a very common feature. Kumaraswamy and Rajasekarasetty (1976) reported centromeric C-bands in *A. turrita*. Aswathanarayana and Ashwath (2006) also revealed centromeric bands in *A. turrita*. According to Yadav and Yadav (1993), restriction of C-heterochromatin to centromeric regions is considered to facilitate whole arm translocation. C-bands found within the centre of the body of chromosome is termed as interstitial C-band. In the present study, interstitial C-bands are seen in all the species of grasshoppers exceptin *A*. *turrita*, *O. abruptus* and *O*. *velox*. The interstitial C-bands were also encountered in 10 species of their study by Yadav and Yadav (1993). Mayya et al. (2004) revealed the presence of interstitial C-bands at different locations on chromosomes among Acridoid species. These interstitial bands are found to be inactivated centromere in some species of *Hieroglyphus nigrorepletus* (Yadav and Yadav, 1993). These interstitial C-bands might have an effect on the expression of the flanking euchromatic segment (Aswathanarayana and Ashwath, 2006). Terminal bands were exhibited by all the 6 species except *A*. *turrita*. Similarly, the absence of terminal bands were also reported in *A*. *turrita* by Yadav

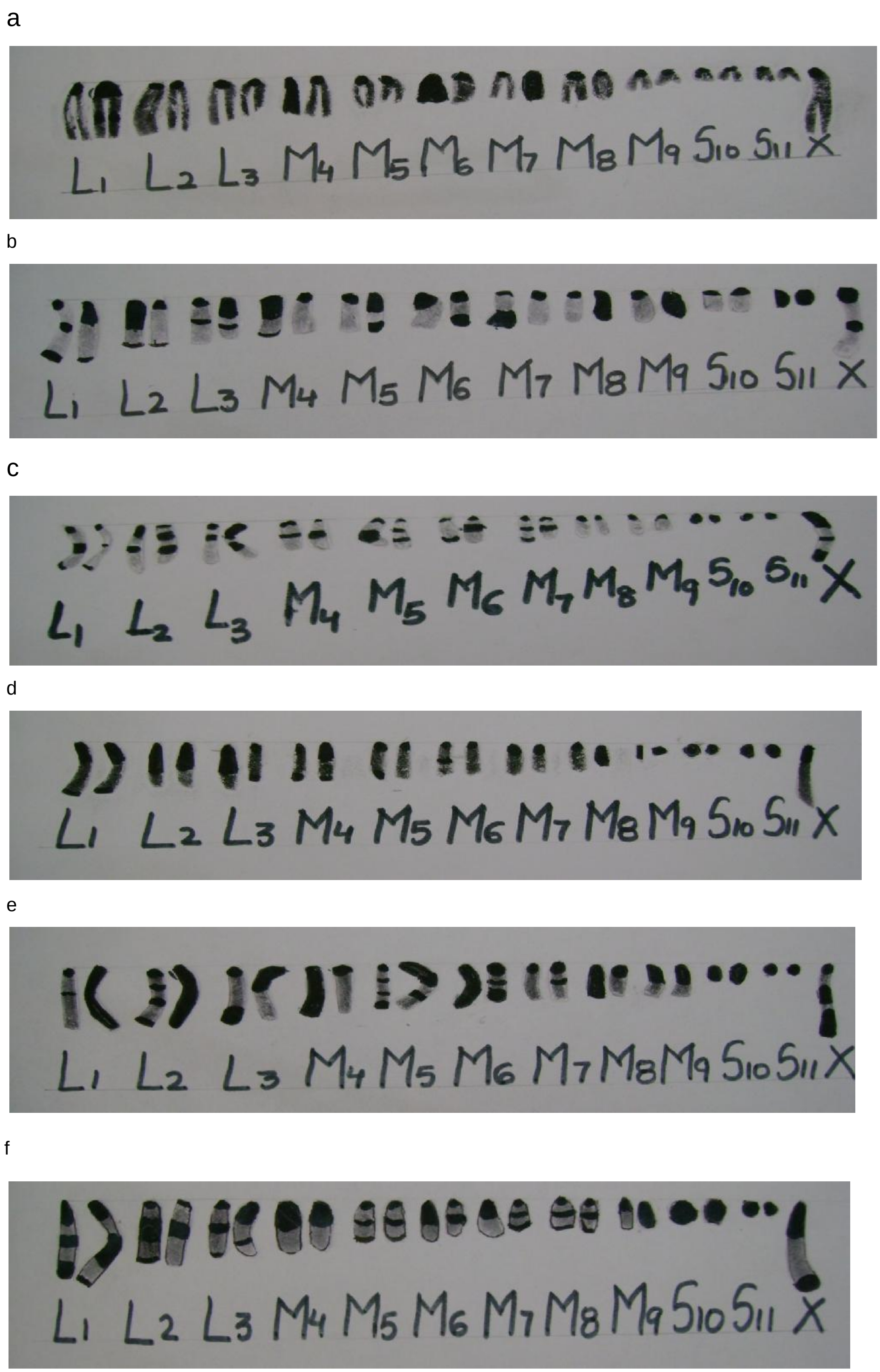

Figure 2. C- Banding karyotypes of (a) *A. turrita* (b) *A. exaltata* (c) *P. infumata* (d) *P. antennata* (e) *O. abruptus* (f) *O. velox*.

and Yadav (1993). On the other hand, our studies revealed the presence of centromeric, interstitial, terminal bands along with thick and thin bands.

The comparison of interspecific C-banding patterns of the same sub-family has no clear correlation. The species from the same genus have not shown uniformity in their C-banding patterns (John and King, 1977; Santos and Giraldez, 1982) which has attributed to dynamic nature of heterochromatin (Yadav and Yadav, 1993). Same situation has been seen in present study where genus coming under different sub-families show some C-banding similarities (*P*. *antennata*, *O*. *abruptus*). The species from the same sub-family differ in their C-band distribution (*A*. *turrita*, *A*. *exaltata*). Likely, such comparisons are such that, one cannot be sure that chromosomes of similar relative lengths are necessarily homologous in all genomes (King and John, 1980). Perhaps, the only exceptions are the X and the megameric chromosomes which presumably have a common origin within the Acrididae (White, 1973). The immediate tendency for C-heterochromatin to vary in grasshoppers has been considered by many reports (Santos et al., 1983; Yadav and Yadav, 1983; present report). The pattern and distribution of C-heterochromatin distribution varies among *Acridoid taxa*, especially karyologically conservative ones. These variations are to be governed by some hidden mechanism of change, other than gross chromosome rearrangements operating in the process of speciation.

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