Türk. Bit. Kor. Derg. (1984) 8 : 3-16

Karyological investigations on seven species of Dytiscidae (Adephaga : Coleoptera)

Joginder Singh YADAV* Kondal KARAMJEET* Abhay Singh YADAV*

Summary

The present investigation deals with the structure and behaviour of chromosomes during mitosis and meiosis during spermatogenesis in seven species of Dytiscidae. The cytogenetic picture was found to be: **Gaurodytes consperus** March. (Colymbetinae) 2n=40 (19+Xy) ; **Cybister tripunctatus** Oliv. (Dytiscinae) 2n=43 (21+X0) ; **Hydaticus leander** Rossi (Hydatiscinae) 2n=41 (20+X0) ; **H. vittatus** F. 2n=45 (22+X0) ; **H. luczonicus** Aube. 2n=41 (20+X) and **Eretes** sp. 2n=43 (21+X0). Mean chiasma frequency was comparatively low. Whereas a few ring bivalents were observed in dytiscines a single chiasma per bivalent seemed to be the rule in the three species of **Hydaticus**. **Eretes** sp., however, showed as many as five ring bivalents. A single anaphase bridge without fragment was observed in **H. leander**.

Introduction

Smith and Virkki (1978) listed the chromosomal data of 178 species of Adephaga. However, recently karyological studies on these beetles have caught the attention of cytogeneticists (Serrano 1980a, b, 1981a, b and unpublished; Yadav and Karamjeet 1980, 1981a ,b) and the number of cytologically known adephagan Coleoptera has rised to about 350 species. In spite of this increase and the fact that a dytiscid **Cybister roesolii** was among the earliest insects analysed cytologically (Voinov, 1903) the family Dytiscidae remains almost neglected as far as its karyology is concerned.

Yadav and Karamjeet (1980, 1981a) reported the chromosome number of seven species of Indian dytiscids. The present communication deals with the details of structure and behaviour of chromosomes during mitosis and sex chromosome mechanism in these species.

(*) Cytogenetics Lab., Dept. Zoology, Kurukshetra University, Kurukshetra-132119, India.
Alınış (Received) : 18.4.1983

Materials and Methods

The water beetles **Gaurodytes consperus** March., **Cybister tripunctatus** Oliv., **C. sugillatus** Er., **Hydaticus vittatus** F., **H. leander** Rossi, **H. luczonicus** Aube. and **Eretes** sp. constituted the materials for the present investigations. All these beetles are photopositive and were collected from tube lights from the campus of Kurukshetra University in the months of May-August during 1979 and 1980. Adult males were injected with 0.01 % colchicine. After 2-3 hours the beetles were vivisected in physiological saline. The testes were removed and pretreated with 1 % sodium citrate solution for 10 minutes and fixed in 1:3 acetic alcohol for 15 minutes. Slides were prepared by air drying technique, stained in 6 % Giemsa solution and mounted in DPX. Photomicrographs were taken and karyograms were prepared and analysed according to Yadav and Pillai (1973). Exact magnification is given alongwith the figures.

Observations

Numerical data on chromosome measurements of six species are presented in Table 1.

Gaurodytes consperus March. (Figs. 1, 2, 13)

2n=40 (Figs. 1-2) contain 10 pairs of metacentric (pairs 4, 6-11, 15-17), seven pairs of submetacentric (pairs 1-3, 5, 12-14) and two pairs of acrocentric (pairs 18, 19) autosomes showing a gradual decrease in size and the sex chromosomes a submetacentric X and metacentric y. The bivalents at metaphase I were rod or ring-shaped mean chiasma frequency and terminalization co-efficient per nucleus being 21.9 and 0.97 respectively. The sex bivalent usually lies away from the main plate. Metaphase II counts confirm the number of chromosomes in the karyotype (Fig. 13).

The male chromosomal formula is 19AA+Xy.

Cybister tripunctatus Oliv. (Figs. 3-4, 14-15)

The 2n=26 reported earlier (Yadav and Karamjeet, 1980) is erroneous being a case of misreporting. The spermatogonial metaphase revealed 2n=43 (Figs. 3-4) differentiated into 14 pairs of metacentric (pairs 2, 4-11, 13-16, 19), four pairs of submetacentric (pairs 1, 3, 12, 17) and three pairs of acrocentric (pairs 18, 20, 21) autosomes and an acrocentric X chromosome which is larger than the last autosomal pair. The last two pairs of autosomes being equal-sized all others show a gradation in size. The X chromosome shows positive heteropycnosis during prophase of the first meiotic division up to diplotene (Fig. 14) but looses it onwards. Chiasmate were mostly terminalized by metaphase I the mean chiasma frequency and terminalization coefficient per nucleus being 23.37 and 0.89 respectively. Metaphase II plates with X chromosome (Fig. 15) and without it were seen. Morphology of the karyotype was further confirmed due to separation of chromatids marking exact location of the centromeres.

The male chromosomal formula is 21AA+X0.

Cybister sugillatus Er. (Figs. 5, 6, 16-17)

2n=43 (Fig. 6). The karyotype comprises 15 pairs of metacentric (pairs 4-14, 16-19), four pairs of submetacentric (pairs 1-3, 15) and two pairs of acrocentric (pairs 20, 21) autosomes and a submetacentric X chromosome which, according to size, lies between fourth and fifth autosomal pairs (Fig. 5). Ring and dumb-bell-shaped autosomal bivalents and the univalent X constitute metaphase I plate (Fig. 16). The mean chiasma frequency and terminalization coefficient per nucleus were 26.5 and 0.8 % respectively. The chromosome count was confirmed at metaphase II (Fig. 17).

The male chromosomal formula is 21AA+X0.

Hydaticus vittatus F. (Figs. 7, 8, 18)

2n=45 (Figs. 7, 8) contain 12 pairs of metacentric, 10 pairs of acrocentric autosomes and the metacentric X chromosome which is the largest element of the diploid complement. The autosomes are comparatively small and vary little as far as their size is concerned and it is rather difficult to arrange these according to size. Metaphase I revealed 23 elements, 22 autosomal bivalents which are in the form of rods simple or dumb-bells and the univalent X (Fig. 18). The mean chiasma frequency and terminalization coefficient per nucleus were found to be 22.0 and 0.91 respectively.

The male chromosomal formula is 22AA+X0.

Hydaticus leander Rossi (Figs. 19-22)

2n=41 differs from 2n=45 reported earlier by Joneja (1960). The autosomes are categorized into 13 pairs of metacentric and seven pairs of acrocentric autosomes of overlapping sizes. The sub-metacentric X chromosome is the largest element of the diploid complement. A single chiasma per bivalent seems to be the rule in this species. Even during diakinesis the chiasmata were mostly terminalized the mean terminalization coefficient being 0.88. The chiasmate were fully terminalized by metaphase I. The bivalents were much condensed and the X chromosome was distinct due to its large size and univalent nature (Fig. 19). Occasionally during anaphase I a single bridge, without fragment, was observed (Fig. 20). The metaphase II plates differs in the presence (Fig. 21) or absence of the X chromosome, 20 autosomes always being present. Due to separation of chromatids morphology of the karyotype was confirmed at this stage.

The male chromosomal formula is 20AA+X0.

Hydaticus luczonicus Aube. (Figs. 9, 10, 23, 24).

2n=41 (Figs. 9, 10) comprises 16 pairs of metacentric (pairs 1, 2, 4-15, 17, 18), one pair of submetacentric (pair 3) and three pairs of acrocentric (pairs 16, 19, 20) autosomes which gradually decrease in size and an exceptionally large submetacentric X chromosome showing differential staining. Metaphase 1 depicted 20 rod-shaped autosomal bivalents and a univalent X. The mean terminalization coefficient per nucleus was 0.98. Metaphase II plates (Figs. 23, 24) confirmed the number and morphology of chromosome noted earlier.

The chromosomal formula is 20AA+X0.

Eretes sp. (Figs. 11, 12, 25)

2n=43 (Figs. 11, 12) were differentiated into 11 pairs of metacentric (pairs 1-3, 6-12, 15), three pairs of submetacentric (pairs 4, 13-14) and seven pairs of acrocentric (pairs 5, 16-21) autosomes and the submetacentric X chromosome. Whereas X is the largest element of the diploid complement the autosomes show gradual decrease in size. As many as five ring bivalents were observed at metaphase I. The mean chiasma frequency and terminalization coefficient per nucleus were 26.8 and 0.71, respectively. Metaphase II plates differed in the presence or absence of the sex chromosome.

The male chromosomal formula is 21AA+X0.

Discussion

A perusal of data in Table 1 reveals that **Gaurodytes consperus** has the maximum genome size, the smallest occurring in **Hydaticus vittatus.** The chromosomes showed maximum size variation in **Cybistes tripunctatus** their relative percentage length varying from 10.14 to 1.74. Size variation is, however, minimum in **H. vittatus** (5.80 to 3.10). Although X chromosome is the largest element of the karyotype in **H. Vittatus, H. luczonicus** and **Eretes** sp. its actual size is maximum in **G. consperus.**

Mitosis and meiosis are typically coleopteran in the dytiscids under present investigations. Occasionally a single anaphase I bridge, without fragment, was observed in **H. leander** which might have resulted due to stickyness, an established character of heterochromatin. Dasgupta (1963) also reported the occurrence of such bridges in **Cybister limbatus.** The chiasma frequency was low as most of the bivalents were rod-shaped. This genetic rigidity seems to have been given up by **Eretes** sp. in which as many as five ring bivalents were reported.

Cytological data of 32 species belonging to 10 genera from six tribes of three subfamilies are on record to-date (Table 2). However, from the data published prior to 1910 only that of Schäfer (1907) has been retained since the other authors were either themselves not certain about their counts (Voinov, 1903; Henderson, 1907) or their studies were confined to oogonial stages giving no details of chromosome numbers and of the course of meiosis. These authors made only casual reference to the chromosome number (Giardina, 1901; Günthert, 1910; Debaisieux, 1909). Consequently we are left with the chromosomal data of 28 species the number of higher taxa remaining unaltered. Suortti (1971) reported polymorphism in **Colymbetes paykulli. In Hydaticus leander** the number of chromosomes observed during the course of present investigations differed from the earlier report of Joneja (1960). As a result the histogram (Fig. 26) depicts the distribution of haploid chromosome number in 31 species of Dytiscidae. The chromosome number varies from 2n=19 in colymbetine **Platambus maculatus,** 9+X (Suortti, 1971) to 2n=45 in hydatiscines **Hydaticus leander** and **H. vittatus,** 22+X (Joneja 1960, present report). However, most of the species show a closer distribution n=18 to n=22 (Fig. 26). There are two peaks n=18 shown by seven species and n=27 by nine species. A closer examination, however, reveals that the first peak is due to the species of subfamily Colymbetinae alone whereas species of subfamilies Colymbetinae and Dytiscinae contribute towards the formation of the second peak. So none of these numbers is representative of the family as a whole. The histogram also does not show a «model number». It is not advisable also to derive a modal number for the family from such a meagre data.

Dytiscids possess simple sex-chromosome mechanisms. Of the 27 species for which the sexchromosome mechanism is known 17 (63 %) possessed only X chromosome in the males whereas 10 species (37 %) possessed a Y chromosome also. Association of the sex chromosomes during first meiotic division is definitely chiasmate in five of these species and has been designated as XY or Xy. Suortti (1971) reported Xyp association in five species including **Agabus bipustulatus**. In **A. sturmi** the author has put a ? with Xyp. However, Smith (1953) in congeneric **A. confinis** has described the sex chromosome association as 'XY'. Suortti (1971) analysed sectioned material and drew camera lucida drawings of which only **Rantus notatus** depicts a clear Xyp. This author also recorded 10 as the number of "Bivalents in MI" in **Platambus maculatus** where the sex chromosome mechanism was recorded as 'XO' and the number of "Chromosomes in MII" as 9 and 10. In the light of these observations the reports of Xyp do not appear to be beyond doubt and the species need reinvestigations.

No clear picture with regard to a primitive dytiscid karyotype and the direction it might have taken during the course of evolution emerges from the available data. However, the number of chromosomes, metacentric nature of most of the elements constituting the karyotype and their large size do not conform to the 18+Xyp male primitive formula proposed by Smith (1950). More data must be acquired before any conclusion is drawn in respect of these issues.

Özet

Dytiscidae (Adephaga: Coleoptera) familyasına ait yedi tür üzerinde karyolojik araştırmalar

Bu araştırma, Dytiscidae familyasına ait yedi tür üzerinde spermatogenesis süresince mitosis ve meiosis sırasında kromozomların yapısal ve davranışsal özelliklerini ortaya koymayı amaçlamaktadır. Sitogenetik incelemeler sonucunda saptanan kromozom sayıları şöyledir: **Gaurodytes consperus** March, 2n=40 (19+Xy) ; **Cybister tripunctatus** Oliv., 2n=43 (21+X0) ; **Hydaticus leander** Rossi, 2n=41 (20+X0); **H. vittatus** F., 2n=45 (22+X0) ; **H. luczonicus** Aube., 2n=41 (20+X) ve **Eretes** sp. 2n=43 (21+X0). Ortalama chiasma frekansı, nispeten düşük bulunmuştur. An-

cak Dytiscinae altfamilyasında, birkaç tane halka bivalent saptanmış olup **Hydaticus** cinsine bağlı 3 türde, bivalent başına tek bir chiasma'nın bulunması, genel bir kural olarak düşünülmüştür. Bununla birlikte, **Eretes** sp.'de 5 kadar bivalent halkasının olduğu tespit edilmiştir. **H. leander**'de, herhangi bir bağlantıya sahip olmayan tek bir anaphase köprüsünün bulunduğu görülmüştür.

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Table 2. Chromosomal analysis of the family Dytiscidae

Species	Diploid chromosome number	Male chromosomal formula	Reference
Colymbetinae			
AGABINI			
1. **Agabus confinis** Gyll.	——	20+XY	Smith 1953
2. **A. bipustulatus** L.	——	20+Xyp	Suortti 1971
3. **A. sturmi** Gyll.	——	20+Xyp ?	″ ″
4. **Ilybius aenescens** Thoms.	——	17+X	″ ″
5. **I. ater** DeG.	——	17+X	″ ″
6. **I. fenestratus** F.	35 ♂	17+X	″ ″
7. **I. fuliginosus** F.	——	17+X	″ ″
8. **I. guttiger** Gyll.	——	17+X	″ ″
9. **I. subaeneus** Er.	——	17+X	″ ″
10. **Gaurodytes consperus** March.	40 ♂	19+Xy	Present Report
COLYMBETINI			
11. **Colymbetes fuscus**	35—37 ♀ *	——	Günthert 1910
12. **C. paykulli** Er.	——	? 17+Xyp	Suortti 1971
13. **C. striatus** L.	——	19—21+Xyp ?	″ ″
14. **Platambus maculatus** Thoms.	——	9+X	″ ″
15. **Rantus exoletus** Forst.	——	20+Xyp	″ ″
16. **R. notatus** F.	44 ♂	21+Xyp	″ ″
Dytiscinae			
DYTISCINI			
17. **Dytiscus marginalis** L.	36—41 (40) ♂ *	—	Henderson 1907
	38 ♂	18+XY	Schäfer 1907
	40 ♀ *	—	Debaisieux 1909
18. **D. circumcinctus**	38 ♂	18+XY	Schäfer 1907

Table 2. (continued)

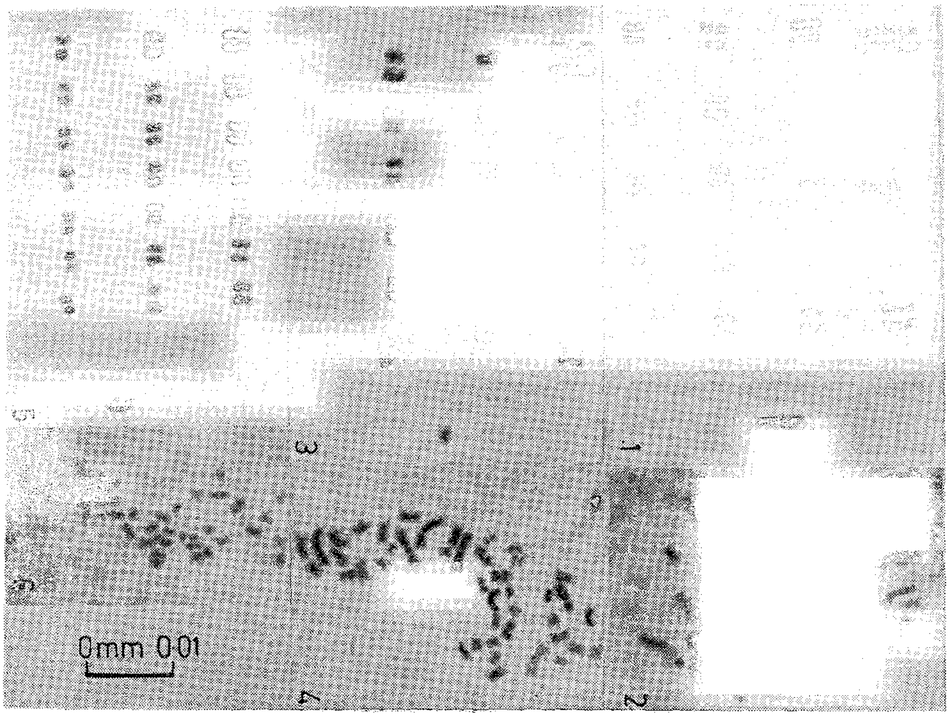
Species	Diploid chromosome number	Male chromosomal formula	Reference
19. **Dytiscus** sp.	40 ♀ *	—	Giardina 1901
20. **Dytiscus** sp.	38—40 ♀ *	—	Günthert 1910
21. **Dytiscus** sp.	34 ♂	—	Smith 1960
CYBISTERINI			
22 **Cybister roeselii**	22 ♂ *	11—15 (I), 12 (II)	Voinov 1903
23. **C. limbatus** F.	——	21+X	Dasgupta 1963
24. **C. tripunctatus** Oliv.	43 ♂	21+X	Present Report
25. **C. sugillatus** Er.	43 ♂	21+X	" "
Hydatiscinae			
HYDATICINI			
26. **Hydaticus fabricii** MacL.	41 ♂	20+X	Joneja 1960
27. **H. leander** Rossi	45 ♂	22+X	" "
	41 ♂	20+X	Present Report
28. **H. vittatus** F.	45 ♂	22+X	" "
29. **H. luczonicus** Aube.	—	20+X	" "
ERETINI			
30. **Eretes sticticus** L.	——	20+X	Joneja 1960
31. **Eretes** sp.	43 ♂	21+X	Present Report
32. Unidentified Dytiscid	—	18+X	Smith 1960

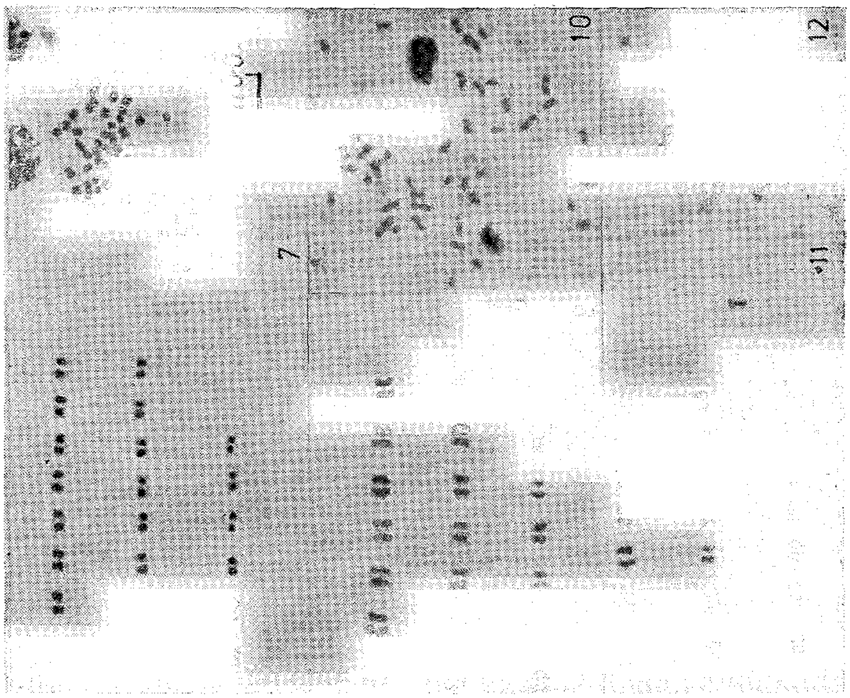
* Data not included in the histogram (see text)

Explanation to figures

Gaurodytes consperus (Figs. 1-2, 13), **Cybister tripunctatus** (Figs. 3-4, 14-15), **C. sugillatus** (Figs. 5-6, 16-17), **Hydaticus vittatus** (Figs. 7-8, 18), **H. leander** (Figs. 19-22), **H. luczonicus** (Figs. 9-10, 23-24), **Eretes** sp. (Figs. 11-12, 25).

Fig.1	Karyotype
Fig.2	Spermatogonial metaphase
Fig.3	Karyotype
Fig.4	Spermatogonial metaphase
Fig.5	Karyotype
Fig.6	Spermatogonial metaphase
Fig.7	Karyotype
Fig.8	Spermatogonial metaphase
Fig.9	Karyotype
Fig.10	Spermatogonial metaphase
Fig.11	Karyotype
Fig.12	Spermatogonial metaphase
Fig.13	M. II with X
Fig.14	Diplotene
Fig.15	M II without X
Fig.16	M I
Fig.17	M II with X
Fig.18	M I
Fig.19	M 1
Fig.20	Anaphase I showing a bridge
Fig.21	M II with X
Fig.22	M II without X
Fig.23	M II with X
Fig.24	M II without X
Fig.25	M 1



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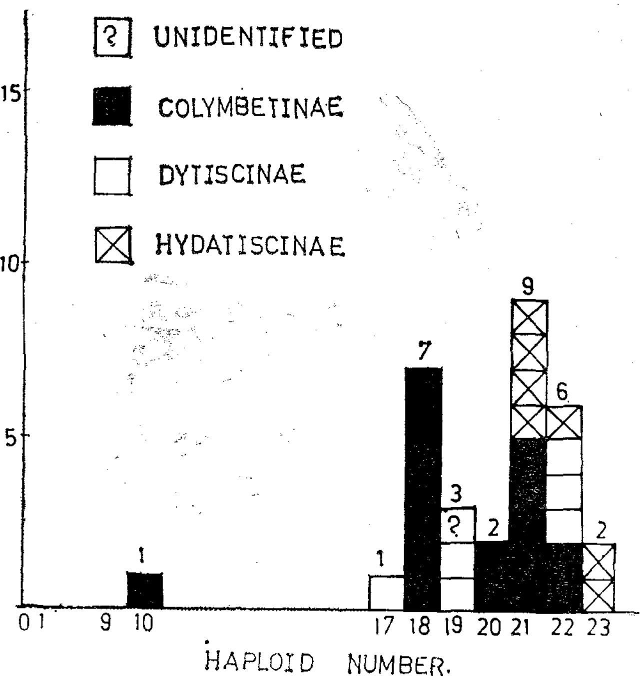
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