



Title	A Chromosomal Survey of Some Indian Insects : I. Morphology of the Chromosomes in Eight Species of the Locustidae (With Plate VIII and 8 Text-figures)
Author(s)	ASANA, Jehangir J.; MAKINO, Sajiro; NIIYAMA, Hidejiro
Citation	北海道帝國大學理學部紀要, 6(3), 211-234
Issue Date	1938-12
Doc URL	<a href="http://hdl.handle.net/2115/27014">http://hdl.handle.net/2115/27014</a>
Type	bulletin (article)
File Information	6(3)_P211-234.pdf



[Instructions for use](#)

**A Chromosomal Survey of Some Indian Insects**  
**I. Morphology of the Chromosomes in Eight**  
**Species of the Locustidae<sup>1)</sup>**

By

**Jehangir J. Asana**

Gujarat College, Ahmedabad, India

**Sajiro Makino and Hidejiro Niiyama**

Zoological Institute, Faculty of Science,  
Hokkaido Imperial University, Sapporo, Japan

*(With Plate VIII and 8 Text-figures)*

**I. Introduction**

About a decade ago one of us (J.J.A.) started an investigation of the chromosomes of some Indian insects commonly found in the vicinity of Gujarat College, Ahmedabad, Western India. In this venture he has been receiving considerable help and guidance from such eminent investigators as Professor McClung of the University of Pennsylvania, U.S.A., Professor Kan Oguma and Dr. Makino of the Hokkaido Imperial University, Sapporo, Japan. Some of the results of this investigation which have already been recorded in Indian and foreign journals (Asana and others, 1930-1936) are so encouraging and the Indian Insect Fauna is so vast and varied that one may justifiably hope that insect cytology may prove to be one of the most interesting and fruitful fields of biological research in India.

This paper is the first of a series in which further observations on the chromosomes of several groups of Indian insects will be more

---

1) Contribution No. 132 from the Zoological Institute, Faculty of science, Hokkaido Imperial University, Sapporo, Japan.

systematically recorded. An attempt will be made, in the first place, to present the exact data with regard to the number, the configuration of chromosomes and the variety of karyotypes, if any. In subsequent papers we propose to discuss these data and their bearing on problems of general cytology. Accordingly in the present paper a karyogram study of the male germ cells of some eight species representing five subfamilies of the Locustidæ has been recorded.

Comparatively less progress has so far been made in the chromosome survey of the Locustidæ than in that of the Acrididæ. According to Handlirsch<sup>1)</sup>, the Locustidæ are subdivided into 14 or more subfamilies containing upwards of 3500 species. So far as we are aware chromosomal investigations have been carried out in about 35 species distributed among eight subfamilies. And one of the interesting results of these investigations is that the chromosomes of this group of Orthoptera as regards some of their morphological aspects, such as their shape and the number, vary considerably from species to species where they have been examined. This is noteworthy in view of the fact that this is not such a marked feature of the chromosomal complex in the Acrididæ which is almost similar in many subfamilies.

We feel greatly indebted to Professor Kan Oguma, who suggested this survey, for his highly valued advice and guidance.

## II. Material and Methods

Testes from young individuals obtained from the fields not far from the Ismail College, Jogeshwari, about 20 miles North of Bombay, and also from the insects collected in the vicinity of Gujarat College, Ahmedabad, Western India, constitute the material on which this study is based. All the specimens were collected and gonads fixed by one of us (J.J.A.) during four seasons between August 1930 to November 1933.

Most of the dissected specimens were identified by Dr. B. P. Uvarov of the British Museum to whom our grateful thanks are due, as it is often a very troublesome task to closely identify insects which have been dissected, and whose abdomens have been more or less mutilated in the process. Our thanks are also due to the Ento-

---

1) Schröder, Chr. 1925. *Handbuch der Entomologie*. Bd. III, Jena. S. 450.

mological Section of the Imperial Agricultural Research Institute, Pusa, which is now located near New-Delhi, India, and to Dr. H. Furukawa of the Tokyo Imperial University, who has very kindly assisted us in the taxonomical arrangement of the eight species listed below, which come under five subfamilies of the Locustidæ.

Subfam. Phaneropterinae

1. *Ducetia japonica* THUMBERG
2. *Elimaea securigera* BR. W.
3. *Holochlora* sp.

Subfam. Mecopodinae

4. *Mecopoda elongata* L.

Subfam. Pseudophyllinae

5. *Sathrophyllia* sp. (Probably *S. rugosa* L.)

Subfam. Conocephalinae

6. *Conocephalus* sp.

Subfam. Hexacentrinae (= Listroscelinae)

7. *Hexacentrus mundus* WALKER
8. *Hexacentrus annulicornis* STAL.

The material was fixed in Benda's fluid, Flemming's weak solution and Allen-Bouin's mixture. Sections were cut 12 to 15 micra in thickness and subjected to Heidenhain's iron-haematoxylin method of staining using lightgreen as the counter stain.

All the figures in the text were drawn by one of us, Makino, with the aid of a camera lucida using a Zeiss apoch. obj., 2 mm. and a compensating ocular K. 20 $\times$ , at a magnification of 3000 diameters. They were reduced to 2/3 in printing.

### III. Observations

#### Subfamily Phaneropterinae

The chromosomes observed in 12 species including 10 genera of this subfamily have already been reported upon in several papers (de Sinety '01; McClung '02, '14; Mohr '15; Pearson '27, '29; Ohmachi '35; Hareyama '37). In the present paper three species are examined, of which one has furnished new material on which observations have been made for the first time. As seen in the accompanying list of the chromosome numbers, the karyotype of this subfamily varies in a considerable degree, the elements composing it deviating in number

from 27 to 33. The only uniformity noticeable is that the X-chromosome in every species is always represented by the largest element in the complement and is quite conspicuous as a rod or V-shaped structure.

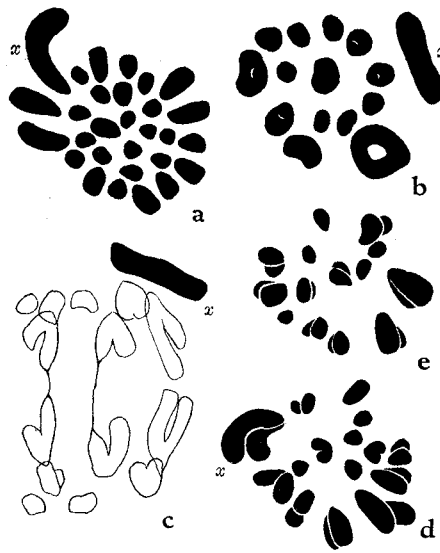
### 1. *Ducetia japonica* THUMBERG (Text-Fig. 1)

The testes from two individuals obtained in October 1932 at Jogeshwari constitute the present material. These were fixed in Allen-Bouin's solution. The specimens have been named by Dr. Uvarov.

The chromosomes of this species have recently been studied by Hareyama ('37). But his results are not in accord with our observations on the Indian form here recorded.

The spermatogonial complement consists of 29 distinct elements of which 28 are autosomes, the unpaired, solitary body being the X-

chromosome (Text-fig. 1, a; Fig. 1, Pl.). Among the autosomes 6 pairs are comparatively larger than others, occupying a peripheral position in equatorial arrangement. They show a gradational diminution in length. All the remaining elements are small, nearly equal in size, and in shape they appear as condensed rods. The X-chromosome takes the form of a heavy conspicuous rod, the largest one among all the elements, and always lies at the periphery of the spindle.



Textfig. 1. Chromosomes of *Ductia japonica*. 2000 $\times$ . a, spermatogonial metaphase. b, primary spermatocyte metaphase. c, anaphase of the same. d, secondary spermatocyte metaphase, X-class. e, the same, no X-class. x: X-chromosome.

The metaphase of the primary spermatocyte shows indisputably and quite clearly 15 chromosomes (Text-fig. 1, b). Among them there are 14 bivalents, more or less ring-shaped, while the remaining

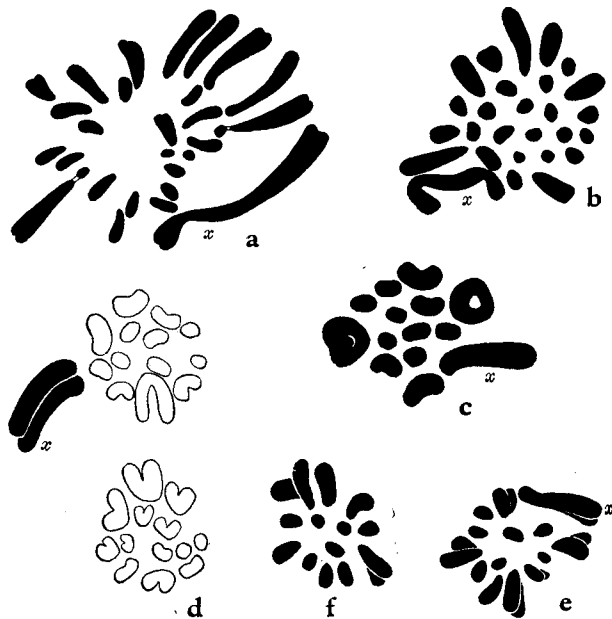
massive, rod-shaped element, lying at the periphery of the spindle and going ahead of the others to one pole, is the X-chromosome (Fig. 2, Pl.). This characteristic behaviour of the X-element makes it clearly demonstrable in the lateral aspect of the spindle, while in the polar view it can only be detected by changing the focus in which the equatorial plate is found. In division, as shown in Text-fig. 1, c, the autosome tetrads segregate into two equal halves, while the X goes undivided to one pole. As a result of this mode of division there arise two kinds of secondary spermatocytes relative to their chromosome complements. One shows 15 chromosomes at the metaphase including the X (Text-fig. 1, d), while the other contains 14 chromosomes in which no element corresponding to the X-chromosome is present (Text-fig. 1, e).

As seen in the accompanying list this species from India is the only representative in the Phaneropterinæ so far recorded which has 29 as the diploid chromosome number. Recently Hareyama ('37) has reported that the chromosome number in the Japanese form of the same species, *Ducetia japonica*, is 27 and not 29, the number in the Japanese form being two less than what obtains in the Indian form. The difference in the number of chromosomes among the individuals of the same species, except that they have been obtained from two widely separated regions, is rather difficult to account for. Two of the authors, Makino and Niiyama, have examined the slides independently of each other and they have counted the same number in many equatorial plates of spermatogonia, and their counts of the reduced number in the primary spermatocytes also agree. Our specimens have been identified by Dr. Uvarov and the same number of chromosomes, 29, has been observed in all the slides examined derived from two pairs of testes from two individuals. In view of the fact that Hareyama ('37) has given only brief descriptions of his observations without any figures we are not in a position to compare more closely the chromosomal complex of the Indian with that of the Japanese specimen and record anything beyond stating the fact that the difference in the number observed is not based upon the formation of any compound chromosomes. We shall await the publication of a fuller description of his observations by Hareyama.

2. *Elimaea securigera* BR. W. (Text-Fig. 2)

In a preliminary note Asana ('31) first reported upon the spermatogonial number of this species which is 27. The present material consists of testes from two specimens, one collected at Ahmedabad in August 1930 and the other at Jogeshwari in September 1932. They were fixed in Benda's solution.

The chromosome number as counted in the spermatogonium is without doubt 27 (Text-fig. 2, a-b; Fig. 10, Pl.). This chromosome complement is somewhat different from that of the foregoing species and from those of the other species of Phaneropterinae as well. The X-element here is quite distinctly the largest and is characterised by the presence of a subterminal constriction, which gives it the appearance of J. Among the autosomes there stand out quite clearly four pairs of longer elements arranged on the outer zone of the equatorial plate. These surround the remaining nine pairs of smaller



Textfig. 2. Chromosomes of *Elimaea securigera*.  $\times 2000$ . a-b, spermatogonial metaphases. c, primary spermatocyte metaphase. d, primary spermatocyte telophase. e, secondary spermatocyte metaphase, X-class. f, the same, no X-class. x: X-chromosome.

chromosomes. All these chromosomes show a gradational magnitude in size. In well differentiated preparations a distinct constriction in the terminal regions of the longest pair of autosomes is noticeable as shown in Text-fig. 2, a (Fig. 10, Pl.).

Well defined 14 chromosomes are observable in the metaphase of the primary spermatocyte (Text-fig. 2, c). They consist of 13 autosomal bivalents, four of which are larger than others, and a single X-element with its subterminal fibre attachment, the point of attachment being directed towards one of the poles. In the first division it goes to one pole ahead of others, entire, without segregation (Text-fig. 2, d and Fig. 3, Pl.). The secondary spermatocytes include two kinds of chromosome complements, one containing 13 elements plus an elongated J-shaped X, 14 in all (Text-fig. 2, e), while the other has only 13 chromosomes without X (Text-fig. 2, f).

Prior to our observations, the chromosome number, 27, was known to exist only in three species of Phaneropterinae, namely, *Ducetia japonica*, *Isotima japonica* and *Phaneroptera nigroantennata* as reported by Hareyama ('37) (see the accompanying list). However, it is not possible to compare the complexes of these species as Hareyama ('37), as mentioned above, has published only brief notes without figures. Nevertheless, there is some justification for the inference that *E. securigera* has a different chromosomal constitution, though the number of the chromosomes in the four species is the same. This deduction is made on considering the observations of Hareyama ('37), described in words, that the chromosomes of *Ducetia* and *Phaneroptera* are all telomitic and *Isotima* contains 16 rods, two V's, eight spheroids and a large V-shaped X-element, which is not the case with the Indian species. Finally, it is interesting to find that the X-element of *Elimaea securigera* shows a remarkable resemblance in shape to that of *Amblycorypha* studied by Pearson ('29).

### 3. *Holochlora* sp. (Text-Fig. 3)

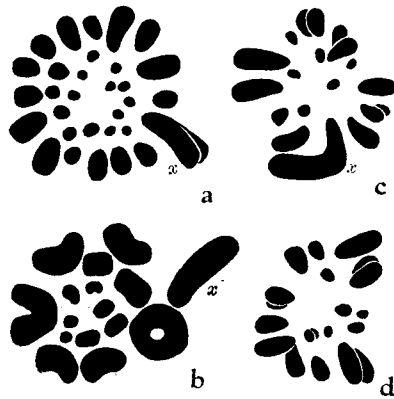
The specimen from which the testes have been derived was identified at Pusa, the headquarters of the Imperial Agricultural Research Institute of India. The insect came from Jogeshwari and the testes were fixed in Benda's solution.

The spermatogonium contains 31 well-defined chromosomes, composed of 30 autosomes and a heavy rod-shaped solitary X-element,



which is conspicuous by its large size (Text-fig. 3, a; Fig. 4, Pl.). Among the autosomes two groups can be distinguished, one of which consists of 16 more or less elongated rod-shaped elements occupying the peripheral zone of the equatorial plate, while the other group is made up of 14 more or less rounded chromosomes, which lie scattered in the central space. This distinction in shape between the two groups of chromosomes, the outer rods and the inner spheroids, is quite clear-cut.

At the metaphase of the primary spermatocytes the above mentioned elements get converted into 15 autosomal bivalents, the pre-



Textfig. 3. Chromosomes of *Holochlora* sp. 2000 $\times$ . a, spermatogonial metaphase. b, primary spermatocyte metaphase. c, secondary spermatocyte metaphase, X-class. d, the same, no X-class. x: X-chromosome.

ceding solitary X-chromosome remaining as such (Text-fig. 3, b). In correspondence with the spermatogonial chromosomes eight of these bivalents have a conspicuously large size, in shape resembling a ring or a thick V. They lie at the periphery of the spindle surrounding the remaining seven small bivalent chromosomes. The X-chromosome in this stage maintains its previous heavy rod-shape appearance, lying always at the periphery of the spindle with its fibre attachment directed towards one of the poles and going to that pole ahead of the others (Fig. 5, Pl.). It, therefore, lies obliquely to the equatorial plate and is demonstrable in the polar view only

by changing the focus in which the equatorial plate is seen. Due to this asymmetrical distribution of the X in the first division, two kinds of secondary spermatocytes are produced, the representative of one class consisting of 15 autosomes and an X-element and that of the other class has 15 autosomes only (Text-fig. 3, c-d).

In a brief note Ohmachi ('35) has reported upon the chromosomes of a related Japanese species, *Holochlora japonica* with similar results. It appears that the *Holochlora* karyotype is of fairly common occurrence in some groups of the Locustidæ. Besides *Holo-*

*chlora*, *Xiphidion melanum* of the Xiphidiinæ (Hareyama '32), *Decticus verrucivorus* (Mohr '19), *D. verrucosus* (Buchner '09), *D. albifrons* (de Winiwarer '31), *Gampsocleis bürgeri* (Hareyama '32), *Metrioptera japonica* (Hareyama '27; Kichijo '34) and *M. bonneti* (Hareyama '27), the last nine forms belonging to the Decticinæ, all uniformly show a closely similar karyotype.

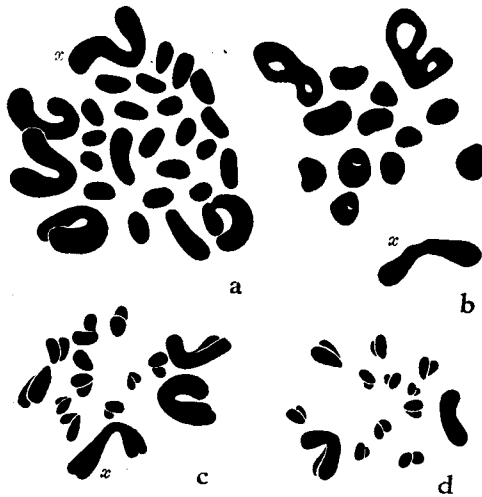
### Subfamily Mecopodinae

Comparatively very little is known of the chromosomes of this subfamily. Our knowledge in this direction is limited to a short account of the chromosomes of *Mecopoda elongata nipponensis* given by Hareyama ('32).

#### 4. *Mecopoda elongata* L. (Text-Fig. 4)

Two pairs of testes derived from two individuals collected at Jogeshwari in September 1932 furnish the material for our observations. The testes were fixed in P.F.A. 3 (modified Bouin), and the specimens were identified by Dr. Uvarov.

The diploid number of chromosomes as observed in the spermatogonial metaphase is, without doubt, 27. This chromosome complement consists of two pairs of large V-shape and 11 pairs of somewhat smaller and rod-shaped univalents with a single V-shaped X-element, which shows a submedian insertion (Text-fig. 4, a). One of the two pairs of V-shaped elements among the autosomes is almost



Textfig. 4. Chromosomes of *Mecopoda elongata*. 2000 $\times$ . a, spermatogonial metaphase. b, primary spermatocyte metaphase. c, secondary spermatocyte metaphase, X-class. d, the same, no X-class. x: X-chromosome.

submedian in attachment constriction, while the other pair is distinctly subterminal. Among the rod-shaped elements, barring a pair of fairly long chromosomes, a gradual diminution in size is evident. No elements, distinctly spheroidal in form, have been discovered in this complex.

The chromosomal constituents of the metaphase equatorial plate of the primary spermatocyte are 13 bivalents and an X-element (Text-fig. 4, b). Two elements among these autosomal bivalents are conspicuously large and appear as compound ring-tetrads originating from the two pairs of large atelomitic chromosomes of the spermatogonium. The X-element, in which the submedian insertion still persists, always lies in a plane a little above the equatorial plate where the autosomal bivalents are scattered. In division, the X runs undivided to one pole, showing as usual a conspicuous precocious behaviour (Fig. 11, Pl.). As a result of this division two kinds of spermatocytes with differing chromosomal constitutions come into being, one containing 14 chromosomes including a submedian X-element and the other has 13 autosomes without the X (Text-fig. 4, c-d). In every case two large atelomitic chromosomes are constantly observable, the others, excepting the X, being provided with a telomitic fibre attachment.

As the notes published by Hareyama ('32) on the chromosomes of the closely allied Japanese form *Mecopoda elongata nipponensis* are too brief and unaccompanied by figures, we are not in a position to make any detailed comparison between the chromosomes of these two related forms beyond observing that they have the same number of chromosomes. However, the karyotype of *Mecopoda* alone among all the species of Locustidæ so far examined presents some strange features. For one thing, although the same number of chromosomes exists in the four forms, *Mecopoda*, *Elimaea*, *Isotima* and *Phaneroptera*, the chromosome complex of the first form mentioned is materially different from the three latter species (cf. the accompanying list). Besides the above noteworthy feature, we find that *Mecopoda* shows some resemblance to certain species of Xiphidiinæ and of Conocephalinæ in having four V-shaped elements in its chromosomal complex as they have in their complements. But the resemblance ends here, as the chromosomal complements of these species as compared with that of *Mecopoda* are much complex in their constitution.

## Subfamily Pseudophyllinae

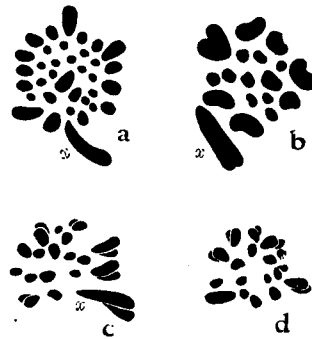
Three species of *Jamaicana* belonging to this subfamily have been cytologically studied by Woolsey ('15). The main result of this investigation is the ascertaining of the fact that numerical variation observed in these species was consequent upon the formation of multiple chromosomes. Our observations on the chromosomes in this subfamily are confined to the study of a single species of *Sathrophyllia*.

5. *Sathrophyllia* sp. (Text-Fig. 5)

Though the species name of this locustid has not been determined with certainty, Dr. Uvarov is of opinion that the two specimens, one obtained at Ahmedabad in August 1930 and the other at Jogeshwari in November 1932, probably belong to *Sathrophyllia rugosa* L. The testes were fixed in Benda's solution.

The specimens are comparatively much larger and thicker in size than the preceding ones. And, therefore, it is rather astonishing to find that the germ cells and the contained chromosomes of this species are remarkably smaller than those of the foregoing ones. This is well brought out in the accompanying microphotographs (Fig. 6 and others, Pl.). The diploid garniture of chromosomes consists of 35 elements as follows: 9 pairs of medium sized rods showing slight differences of length among them; 8 pairs of spheroidal chromosomes of nearly equal size, and a solitary X, which appears as a remarkably huge rod (Text-fig. 5, a; Fig. 6, Pl.). There is an obvious distinction between the rods and spheroids.

The primary spermatocyte metaphase shows 18 well-defined chromosomes (Text-fig. 5, b). Seventeen of these elements are bivalent autosomes derived from 17 pairs of spermatogonial chromosomes. A thick, huge rod-shaped element, always appearing at the periphery of the spindle, is the X-chromosome. Its point of attach-



Textfig. 5. Chromosomes of *Sathrophyllia* sp. 2000 $\times$ . a, spermatogonial metaphase. b, primary spermatocyte metaphase. c, secondary spermatocyte metaphase, X-class. d, the same, no X-class. x: X-chromosome.

ment to the spindle fibre is directed towards one of the two poles, hence it always lies obliquely to the equatorial plane of the spindle (Fig. 7, Pl.), and, therefore, in the polar view it can only be traced through its whole length by changing the focus of the microscope. As usual, this chromosome does not divide at this stage and it runs to one of the two poles ahead of the other chromosomes in the first division. Thus there are produced two kinds of secondary spermatocytes, one with 18 chromosomes including the X-element, which is conspicuous by its large size, and the other with 17 autosomes as seen in Text-fig. 5, c and d.

The chromosome number, 35, found in this species is the largest so far recorded for the Locustidæ. The general configuration of chromosomes in this species seems to exhibit no marked difference from the ordinary complement of *Jamaicana* reported by Woolsey ('15) when their figures are compared. In none of our specimens of *Sathrophyllia* we have been able to detect any multiple chromosomes.

#### Subfamily Conocephalinae

The chromosomes of only three species of this subfamily have hitherto been reported upon. McClung ('14) has given a brief account of the diploid number of a species of *Conocephalus* which is 33. Hareyama ('32) and Ohmachi ('35) have studied the chromosomes of *Homoeocoryphus lineosus* where the diploid number is 25. But in another species, *Euconocephalus varius*, recently studied by Hareyama ('37) he reports 21 as the diploid number of chromosomes. The observations recorded below have been based on a pair of testes derived from a single specimen of *Conocephalus*, the species name of which is unfortunately not known. But it is interesting to note that its chromosomal complement differs from those of *Euconocephalus* and *Homoeocoryphus* in the number and some other respects.

#### 6. *Conocephalus* sp. (Text-Fig. 6)

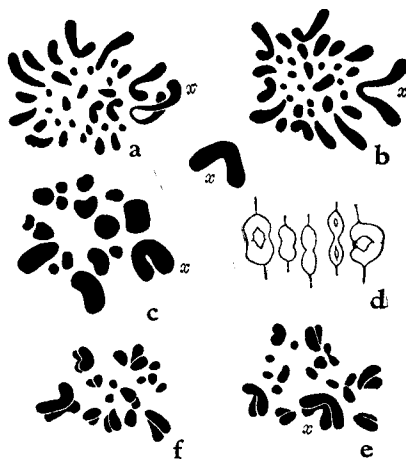
The specimen was collected at Ahmedabad in August 1930. The testes were fixed in weak Flemming. Dr. Uvarov has given us the generic name.

As in the previous case, the size of the germ cell in this species too is small, the individual chromosomes being rather small and

slender. The elements of the diploid complement are variegated in shape and size, as there are V-shaped, rod-shaped and spheroidal chromosomes in this motley garniture. After a critical examination of several metaphase plates, it was decided that there are 33 chromosomes in the spermatogonium, there being six pairs of minute spheroidal, eight pairs of rod-shaped and two pairs of V-shaped elements, besides a solitary X-chromosome, which is very large and V-shaped (Text-fig. 6, a-b; Fig. 8, Pl.). The spheroidal chromosomes are all of nearly equal magnitude. Among the rod-shaped elements there is a remarkably long pair, the others showing a gradual diminution in length. Of the two atelomitic pairs, one is comparatively much larger and has a subterminal constriction, while the other smaller pair is almost submedian in constriction. The X-chromosome, because of its extremely large size stands out quite clearly in the garniture and has a nearly median fibre attachment.

Sixteen bivalent chromosomes and an X-element constitute the metaphase plate of the primary spermatocyte (Text-fig. 6, c). The X is typically V-shaped, has its apex always directed towards one pole and runs ahead of the others (Text-fig. 6, d; Fig. 9, Pl.). Therefore, it can only be detected in polar view by changing the focus of the microscope in which the equatorial plate is viewed. Due to the asymmetrical distribution of the X in the first division, there are produced as usual two kinds of secondary spermatocytes with respect to their chromosomal contents.

As shown in Text-fig. 6, e and f, one of these contains 17 chromosomes including the large V-shaped X, and the other has 16 elements as there is no X in it. The autosomes in this stage show a complete half set of the spermatogonial



Textfig. 6. Chromosomes of *Conocephalus* sp. 2000 $\times$ . a-b, spermatogonial metaphases. c, primary spermatocyte metaphase. d, side view of the same in meta-anaphase. e, secondary spermatocyte metaphase, X-class. f, the same, no X-class. x: X-chromosome.

complex, being composed of two atelomitic, eight telomitic and six minute spheroidal chromosomes.

The diploid number observed in this species is quite in accord with that reported by McClung ('14) for *Conocephalus* sp.; but a closer comparison of the two complement is not possible, since no detailed description is given in McClung's paper. On the other hand, when the Indian form is compared with *Homoeocoryphus lineosus* (Hareyama '32; Ohmachi '35) there is some agreement between the two, though there is difference in the number of chromosomes, as both have two pairs of atelomitic chromosomes. Moreover, the evidence that this species from India bears a close resemblance to *Xiphidion gladiatum* (Ohmachi & Sokame '35), *Orchelimum vulgare* and *O. concinnum* (King '24) belonging to the Xiphidiinæ, not only in its chromosome number but also in its constitution, is interesting when one considers the fact that the Conocephalinæ and the Xiphidiinæ are taxonomically closely related.

#### Subfamily Hexacentrinae

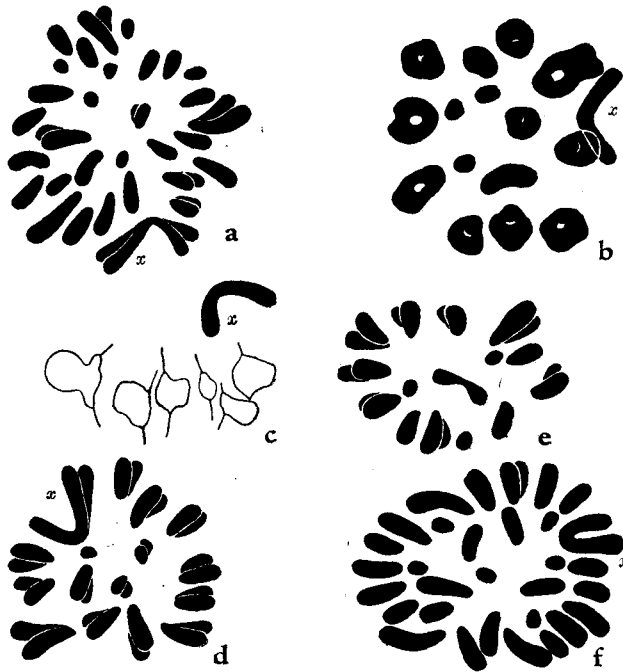
Our knowledge of the chromosomes in this subfamily was limited to a single species, *Hexacentrus japonicus* recently investigated by Hareyama ('37) before the investigations recorded below were undertaken. Hareyama ('37) reports 33 as the diploid number in the Japanese form, which is two more than the number in the two Indian species dealt with in the following description.

#### 7. *Hexacentrus mundus* WALKER (Text-Fig. 7)

Testes from about six individuals collected in two different localities, Ahmedabad (August 1930) and Jogeshwari (August 1933) furnished the present material. Some of these testes were fixed in diluted Flemming's solution and some in modified Bouin. The dissected specimens were identified by Dr. Uvarov.

As the testes were in an advanced stage of development the dividing spermatogonia were scarcely found. The metaphase plate drawn in Text-fig. 7, a (Fig. 12, Pl.) shows 31 chromosomes as the diploid number, which is two less than that observed in *H. japonicus*. An examination of the microphotograph (Fig. 12, Pl.) will leave no doubt in the mind of the observer that there are 31 chromosomes in

this plate. To support the validity of this count a search was made for mitosis in somatic cells. And fortunately many cells in the division process were found in the epithelium of the seminiferous duct of a certain individual. Two of the authors, Niiyama and Makino, have examined several equatorial plates independently of each other, and both the observers counted the same number, 31. One of the several mitotic metaphases is reproduced in Text-fig. 7, f.



Textfig. 7. Chromosomes of *Hexacentrus mundus*. 2000 $\times$ . a, spermatogonial metaphase. b, primary spermatocyte metaphase. c, side view of the same. d, secondary spermatocyte metaphase, X-class. e, the same, no X-class. f, somatic metaphase of the epithelial cell of the seminiferous duct. x: X-chromosome.

In no locustid so far studied such a strange chromosomal complex has been observed as appears in this species. This complement is remarkable in having the following constituents: three pairs of small spheroidal chromosomes of nearly equal size, 12 pairs of rod-elements showing a gradational diminution in size, and a solitary V-shaped X-chromosome with a submedian constriction.



The haploid number observed in the primary spermatocyte metaphase is invariably 16 in every equatorial plate examined, 15 of which are autosomal tetrads, the remaining element being the X-chromosome with a distinct submedian constriction (Text-fig. 7, b). Among these tetrads are well distinguishable three small elements of nearly equal size, which obviously have descended from three pairs of chromosomes of the corresponding shape in the spermatogonium. As in the case of the several foregoing species, the X lies at the periphery of the spindle, has its apex directed towards one of its two poles and in anaphase it runs ahead of others (Text-fig. 7, c). Due to this asymmetrical distribution of the X-chromosome, there are produced as usual two kinds of secondary spermatocytes relative to their chromosomal contents. In one group, the X-class, we have 15 autosomes and the X-element, the latter having a submedian fibre attachment, while the other group has 15 autosomes only (Text-fig. 7, d-e). In both the cases the autosomal complement, as is expected, consists of a complete half set of the spermatogonial complex, the three smallest elements in each being quite evident.

As mentioned above in this species the number of chromosomes is 31, two less than what obtains in the Japanese form, *H. japonicus* as reported by Hareyama ('37). We feel confident that no mistake has been made, as in every one of the six specimens of *H. mundus* so far examined and which were collected in two widely separated localities the number invariably shown in primary spermatocytes is 16. In the chromosomal complex of the Indian form which two chromosomes of the Japanese form are lacking is obscure at present, because the notes of Hareyama ('37) on *H. japonicus* are too brief to allow a close comparison.

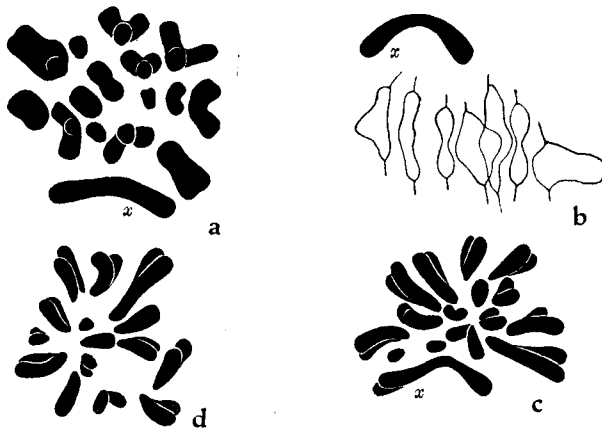
The chromosomal complex of *H. mundus* is somewhat unique in its constitution and, therefore, in this respect it furnishes a new example for the Locustidæ, though the number of chromosomes, 31, which is seen in this complex is rather common in the family Locustidæ.

#### 8. *Hexacentrus annulicornis* STAL. (Text-Fig. 8)

The following observations are based on a pair of testes from a single individual collected at Ahmedabad in August 1930. They were fixed in Benda's fluid. The specimen was identified at Pusa.

Since the germ cells in this specimen too were much advanced in the spermatogenetic process, the spermatogonial divisions were entirely absent in our material. Therefore, our observations are confined to primary and secondary spermatocytes. And here a close similarity is noticeable between *H. mundus* and this species with regard to two main features, firstly, in the number of chromosomes and secondly, in the general morphological characters of their elements, barring a slight difference mentioned below.

The number of chromosomes in the primary spermatocyte metaphase is 16 without exception (Text-fig. 8, a). The V-shaped X has a submedian constriction and goes undivided to one pole (Text-fig. 8, b). The secondary spermatocytes that are produced as a result of the division of primary spermatocytes are, as usual, of two kinds,



Textfig. 8. Chromosomes of *Hexacentrus annulicornis*. 2000 $\times$ . a, primary spermatocyte metaphase. b, side view of the same. c, secondary spermatocyte metaphase, X-class. d, the same, no X-class. x: X-chromosome.

one with the X-element and 15 autosomes, the other without it and consisting only of 15 autosomes (Text-fig. 8, c-d). From this result it may be legitimately inferred that in all probability there are 31 chromosomes in the diploid complement of this species, as in the case of *H. mundus*. However, a closer comparison between the secondary spermatocyte chromosomes of the two species reveal a difference, which may well serve as a diagnostic feature under the microscope

between *H. mundus* and *H. annulicornis*. The rod-shaped elements of *H. annulicornis* are apparently larger than those of *H. mundus* (compare Text-fig. 8, c-d with 7, d-e), though in other respects these two species are similar, there being in each complement 12 rods and three small autosomes. The difference is most noticeable when the longest elements in the secondary spermatocyte chromosomes are compared between the two species. The evidence of some difference is likewise observable in the primary spermatocyte chromosomes also. As seen in Text-fig. 8, a, the tetrads of *H. annulicornis* are better differentiated in form than those of *H. mundus*, probably due to the increase of chiasmata, which is proportional to the length of chromosomes.

#### IV. Review of the chromosome numbers in the Locustidæ

So far as the literature shows the chromosomes of the locustid Orthoptera have been investigated in 43 species covering eight subfamilies. And those include the results presented in this paper. The following list records the number of chromosomes in the Locustidæ hitherto investigated.

##### List of the chromosome numbers in the Locustidæ

Species	Diploid	Haploid	Shape of X-chrom.	Reference
<b>Phaneropterinae</b>				
1. <i>Amblycorypha rotundifolia</i>	33 spg	17 (I) 16, 17 (II)	V	PEARSON, '27, '29
2. <i>Amblycorypha oblongifolia</i>	33 spg	—	V	PEARSON, '27, '29
3. <i>Ducetia japonica</i>	27 spg	14 (I) 13, 14 (II)	R	HAREYAMA, '37
<i>Ducetia japonica</i>	29 spg	15 (I) 14, 15 (II)	R	This paper
4. <i>Elimaea securigera</i>	27 spg	14 (I) 13, 14 (II)	V	This paper
5. <i>Holochlora japonica</i>	31 spg	—	R	OHMACHI, '35

spg: spermatogonium. oog: oogonium. (I): primary spermatocyte. (II): secondary spermatocyte. V: V-shaped chromosome. R: rod-shaped chromosome.

(Continued)

Species	Diploid	Haploid	Shape of X-chrom.	Reference
6. <i>Holochlora</i> sp.	31 spg	16 (I) 15, 16 (II)	R	This paper
7. <i>Isotima japonica</i>	27 spg	14 (I) 13, 14 (II)	V	HAREYAMA, '37
8. <i>Leptophyes punctatissima</i>	31 spg 32 oog	— —	V	MOHR, '15
9. <i>Microcentrum</i> sp.	33 spg	17 (I) 16, 17 (II)	—	MCCLUNG, '02
10. <i>Phaneroptera nigroantennata</i>	27 spg	14 (I) 13, 14 (II)	R	HAREYAMA, '37
11. <i>Orphania denticauda</i>	31 spg	16 (I) 15, 16 (II)	R	DE SINETY, '01
12. <i>Scudderia</i> sp.	33 spg	17 (I) 16, 17 (II)	—	MCCLUNG, '02 '14
<b>Mecopodinae</b>				
13. <i>Mecopoda nipponensis</i>	27 spg	14 (I) 13, 14 (II)	V	HAREYAMA, '32
14. <i>Mecopoda elongata</i>	27 spg	14 (I) 13, 14 (II)	V	This paper
<b>Pseudophyllinae</b>				
15. <i>Jamaicana flava</i>	35 spg	18 (I) 17, 18 (II)	R	WOOLSEY, '15
16. <i>Jamaicana subguttata</i>	35 (34) spg	18 (I) 17, 18 (II)	R	WOOLSEY, '15
17. <i>Jamaicana unicolor</i>	35 (33) spg	18 (I)	R	WOOLSEY, '15
18. <i>Sathrophyllia</i> sp. ( <i>S. rugosa</i> L.?)	35 spg	18 (I) 17, 18 (II)	R	This paper
<b>Xiphidiinae</b>				
19. <i>Orchelimum concinnum</i>	33 spg 34 oog	—	V	KING, '24
20. <i>Orchelimum vulgare</i>	33 spg 34 oog	— —	V	KING, '24
21. <i>Xiphidion gladiatum</i>	33 spg	—	V	OHMACHI & SOKAME, '35
22. <i>Xiphidion maculatum</i>	21 spg	—	V	OHMACHI & SOKAME, '35
23. <i>Xiphidion melanum</i>	31 spg	16 (I) 15, 16 (II)	R	HAREYAMA '32
24. <i>Xiphidium fasciatum</i>	33 spg	—	V	MCCLUNG, '99, '08
25. <i>Xiphidium</i> sp.	33 spg	17 (I) 16, 17 (II)	—	MCCLUNG, '99, '02, '03, '14

(Continued)

Species	Diploid	Haploid	Shape of X-chrom.	Reference
<b>Conocephalinae</b>				
26. <i>Conocephalus</i> sp.	33 spg	—	—	MCCLUNG, '14
27. <i>Conocephalus</i> sp.	33 spg	17 (I) 16, 17 (II)	V	This paper
28. <i>Euconocephalus varius</i>	21 spg	11 (I) 10, 11 (II)	V	HAREYAMA, '37
29. <i>Homocoryphus lineosus</i>	25 spg	13 (I) 12, 13 (II)	V	HAREYAMA, '32; OHMACHI, '35
<b>Locustinae</b>				
30. <i>Locusta orientalis</i>	35 spg	—	V	HAREYAMA '32; OHMACHI, '35
31. <i>Locusta viridissima</i>	33 spg	17 (I) 16, 17 (II)	R	OTTE, '06, '07
<i>Locusta viridissima</i>	29 spg 30 oog	15 (I) 14, 15 (II)	V	MOHR, '16
<b>Decticinae</b>				
32. <i>Anabrus</i> sp.	33 spg	17 (I) 16, 17 (II)	—	MCCLUNG, '02, '05, '14
33. <i>Decticus (Tettigonia) albifrons</i>	31 spg	16 (I)	R	DE WINIWARTER, '31
34. <i>Decticus verrucivorus</i>	23 spg	12 (I) 11, 12 (II)	V	VEJDOWSKY, '12
<i>Decticus verrucivorus</i>	31 spg	—	R	MOHR, '19
35. <i>Decticus verrucosus</i>	31 spg	—	R	BUCHNER, '09
36. <i>Gampsocleis bürgeri</i>	31 spg	16 (I) 15, 16 (II)	R	HAREYAMA, '32
37. <i>Metrioptera japonica</i>	31 spg	16 (I) 15, 16 (II)	R	HAREYAMA, '32; KICHIJO, '34
38. <i>Metrioptera bonneti</i>	31 spg	16 (I) 15, 16 (II)	R	HAREYAMA, '32
39. <i>Metrioptera brachyptera</i>	31 spg	16 (I)	R	WHITE, '36
40. <i>Steiroxys trilineata</i>	29 spg	15 (I) 14, 15 (II)	R	DAVIS, '08; MEEK, '13
<b>Hexacentrinae</b>				
41. <i>Hexacentrus japonicus</i>	33 spg	17 (I) 16, 17 (II)	V	HAREYAMA, '37
42. <i>Hexacentrus mundus</i>	31 spg	16 (I) 15, 16 (II)	V	This paper
43. <i>Hexacentrus annulicornis</i>	—	16 (I) 15, 16 (II)	V	This paper

In comparison with the Acrididae the chromosome survey in the Locustidae has made little progress.

All the species recorded are in agreement in the evidence they furnish that the male is heterogametic. The X-element constantly occurs in the male in all species without exception. In every case the X is represented by the largest element, which is either V-shaped or like a rod. The X is quite conspicuous because of its shape and size, and further, it is extremely typical in the precocious behaviour it exhibits in the first division. This very conspicuous behaviour of the X-element is highly characteristic of the Locustidae.

The number of chromosomes varies according to species. In comparison with the Acrididae this variation in the number extends over a wider range. If we disregard for a moment the difference in the morphological nature of the chromosome complements in this family and review the number alone, it will be found that two groups, 31 and 33, predominate over other numbers (cf. the accompanying list).

Another noticeable feature is that inconstancy either in the number of the chromosomes or in the nature of the garniture or complex within a single subfamily is not infrequently found. And this is clearly in evidence in the Phaneropterinae of which many species have been investigated.

Finally, our knowledge of the chromosomes of the Locustidae is yet too scanty to justify any generalization being attempted at this stage. To establish correlations between chromosomes and taxonomical determinations is a task fraught with many difficulties, which demands the accumulation of accurate cytological data on an extensive scale in a very large number of species, before one can attempt to formulate any rule for general application.

### Literature

- ASANA, J. J. 1930. Studies on the chromosomes of Indian Acrididae. Proc. of the 17th Ind. Sci. Congress. 1930.
- 1931. Studies on the chromosomes of Indian Orthoptera. Proc. of the 18th. Ind. Sci. Congress. 1931.
- (with K. OGUMA) 1932. Additional data to our knowledge on the dragon-fly chromosomes, with a note on the occurrence of X-Y chromosome in the ant-lion (Neuroptera). Jour. Fac. Sci. Hokkaido Imp. Univ. Ser. VI, Zool. Vol. I, No. 4.

- ASANA, J. J. (with C. E. McCLUNG) 1933. Chromosomes of *Schizodactylus monstruosus*. Jour. Morph., 55.
- 1934. Studies on the chromosomes of Indian Orthoptera. IV. The idiochromosomes of *Hierodula* sp. Current Science, 2, No. 7.
- (with S. MAKINO) 1934. The idiochromosomes of an earwig, *Labidura riparia*. Jour. Morph., 56.
- (with C. E. McCLUNG) 1934. The auxocytes of *Schizodactylus monstruosus*. Cytologia, 6.
- (with S. MAKINO) 1934. The chromosomes of *Poecilocera picta*, FABR. (a short-horned grasshopper). Jour. Univ. Bombay, Vol. 2. pt. 5.
- (with S. MAKINO) 1936. A comparative study of the chromosomes in Indian dragon-flies. Jour. Fac. Sci. Hokkaido Imp. Univ. Ser. VI. Zool. Vol. 4. No. 2.
- (with H. KICHIJO) 1936. The chromosomes of six species of ant-lions (Neuroptera). Jour. Fac. Sci. Hokkaido Imp. Univ. Ser. VI. Zool. Vol. 5. No. 2.
- BUCHNER, P. 1909. Das accessorische Chromosom in Spermatogenese und Oogenese der Orthoptera, zugleich ein Beitrag zur Kenntnis der Reduktion. Arch Zellf., 3.
- DAVIS, H. S. 1908. Spermatogenesis in Acrididae and Locustidae. Bull. Mus. Comp. Zool., Harvard, 53.
- HAREYAMA, S. 1927. On the chromosome of *Metrioptera japonica*. (Abstract in Japanese). Zool. Mag. (Japan), 39.
- 1932. On the spermatogenesis of an Orthopteran *Gampsocleis bürgeri* D. H. Jour. Sci. Hirosima Univ. Ser. B, vol. 1.
- 1932. On the chromosomes of some insects belonging to the Locustidae. (Abstract in Japanese). Zool. Mag. (Japan), 44.
- 1937. On the chromosomes in the spermatogenesis of some insects of the Locustidae. (Abstract in Japanese). Zool. Mag. (Japan), 49.
- KICHIJO, H. 1934. On the giant spermatogonia with polyploid chromosome numbers in *Metrioptera japonica* Bolivar. (Japanese). Zool. Mag. (Japan), 46.
- KING, R. L. 1924. Material for demonstration of accessory chromosomes. Sci., 60.
- McCLUNG, C. E. 1899. A peculiar nuclear element in the male reproductive cells of insects. Zool. Bull., 2.
- 1902. The spermatocyte divisions of the Locustidae. Kansas Univ. Bull., 1.
- 1905. The chromosome complex of Orthopteran spermatocytes. Biol. Bull., 9.
- 1908. The spermatogenesis of *Xiphidium fasciatum*. Kansas Univ. Sci. Bull., 4.
- 1914. A comparative study of the chromosomes in Orthopteran spermatogenesis. Jour. Morph., 25.
- MEEK, C. F. U. 1913. A metrical analysis of chromosome complexes, showing correlation of evolutionary development and chromatin thread-width throughout the animal kingdom. Phil. Trans. Roy. Soc. London, 203 B.

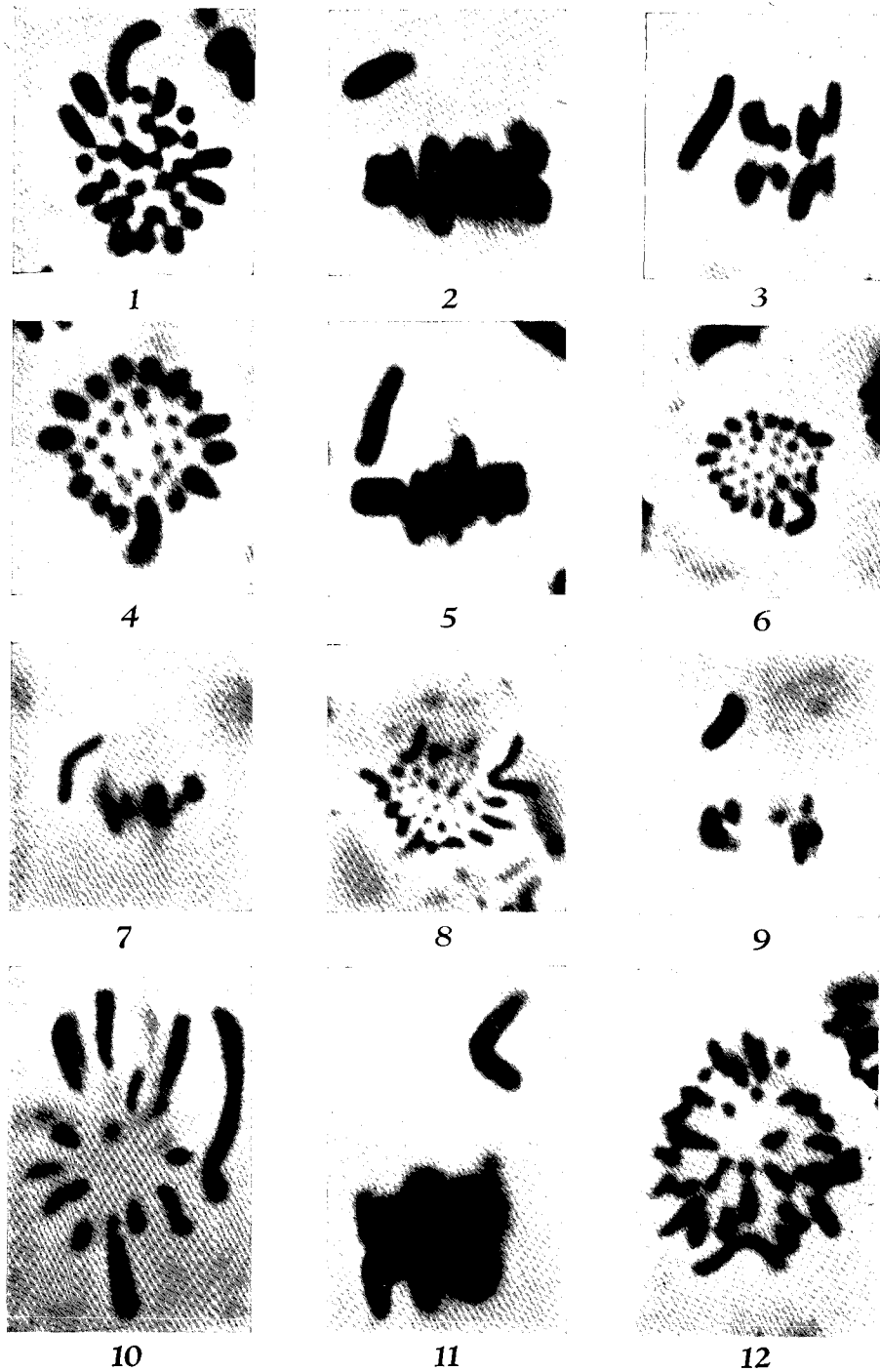
- MOHR, O. L. 1915. Sind die Heterochromosomen wahre Chromosomen? Arch. Zellf., 14.
- 1916. Studien über die Chromatinreifung der männlicher Geschlechtszellen bei *Locusta viridissima*. Arch. Biol., 29.
- 1919. Mikroskopische Untersuchungen zu den Experimenten über den Einfluss der Radiumstrahlen und der Kälterwirkung auf die Chromatinreifung und das Heterochromosom bei *Decticus verrucivorus*. Arch. mikr. Anat., 92.
- OTTE, H. 1906. Samenreifung und Samenbildung von *Locusta viridissima*. Zool. Anz., 30.
- 1907. Samenreifung und Samenbildung von *Locusta viridissima*. Zool. Jahrb., 24.
- OHMACHI, F. 1935. On the relation between the chromosomes and taxonomy in the Locustoidea (A review in Japanese). Zool. Mag. (Japan), 47.
- OHMACHI, F. and SOKAME, C. 1935. On the taxonomical significance of chromosomes in the genus *Xiphidion*. Jap. Jour. Genet., 11.
- PEARSON, N. E. 1927. A study of gynandromorphic katydids. Am. Nat., 61.
- 1929. The structure and chromosomes of three gynandromorphic katydids. Jour. Morph., 47.
- SINÉTY, M. R. de 1901. Recherches sur la biologie et l'anatomie des Phasmes. La Cellule, 19.
- VEJDOWSKY, F. 1912. Zum Problem der Vererbungsträger. Böhm. Ges. Wiss. Prag.
- WHITE, M. J. D. 1936. Chiasma-localization in *Mecostethus grossus* L. and *Metrioptera brachyptera* L. (Orthoptera). Zeits. Zellf. mikr. Anat., 24.
- WINIWARTER, H. de 1931. Evolution de l'hétérochromosome chez *Tettigonia (Decticus) albifrons* (Fab.). Arch. Biol., 42.
- WOOLSEY, C. 1915. Linkage of chromosomes correlated with reduction in numbers among the species in a genus, also within a species of the Locustidae. Biol. Bull., 28.



### Explanation of Plate VIII

All are microphotographs taken with Leitz-Makam using a 2 mm apochr. obj. and 10 × periplan oc. at the magnification of 900 times and then enlarged into about 2000 times.

- Fig. 1. Spermatogonial metaphase of *Ducetia japonica*. The same as Textfig. 1, a.
- Fig. 2. Side view of the primary spermatocyte meta-anaphase of *Ducetia japonica*.
- Fig. 3. Side view of the primary spermatocyte anaphase of *Elimaea securigera*.
- Fig. 4. Spermatogonial metaphase of *Holochlora* sp. The same as Textfig. 3, a.
- Fig. 5. Side view of the primary spermatocyte meta-anaphase of *Holochlora* sp.
- Fig. 6. Spermatogonial metaphase of *Sathrophyllia* sp.
- Fig. 7. Side view of the primary spermatocyte meta-anaphase of *Sathrophyllia* sp.
- Fig. 8. Spermatogonial metaphase of *Conocephalus* sp. The same as Textfig. 6, b.
- Fig. 9. Side view of the primary spermatocyte meta-anaphase of *Conocephalus* sp.
- Fig. 10. Spermatogonial metaphase of *Elimaea securigera*. The same as Textfig. 2, a.
- Fig. 11. Side view of the primary spermatocyte meta-anaphase of *Mecopoda elongata*.
- Fig. 12. Spermatogonial metaphase of *Hexacentrus mundus*. The same as Textfig. 7, a.



*S. Makino photo.*

*J. J. Asana, S. Makino & H. Niiyama: Chromosome morphology  
in eight species of the Locustidae*