THE CHROMOSOMES OF SOME NEUROPTEROUS INSECTS OF THE FAMILY CHRYSOPIDAE[1]

BY

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(*With 5 Figures in Text*)

In spite of great advance in chromosome studies in the related orders, Lepidoptera, Trichoptera and Hemiptera, knowledge about the chromosomes of the order Neuroptera has remained quite limited until recently. So far as the literature shows, OGUMA and ASANA ('32) seem to be the first authors to deal with the Neuropteran chromosomes, finding the XY-complex in a species of *Palpares* of the Myrmeleonidae. Recently ITOH ('33, a, b) investigated the chromosomes of two species of the Sialidae which belong to the suborder Megaloptera of the Neuroptera, and he found male heterogamety in both of the species studied. Shortly later NAVILLE et de BEAUMONT ('33) published their extensive studies on the chromosomes of fifteen species of Neuroptera covering seven families, and established the chromosomal relationship between the allied orders. In the present paper the author wishes to give some account of the chromosomes of some Japanese species belonging to two genera of the family Chrysopidae, in comparison with the species studied by NAVILLE et de BEAUMONT ('33).

The author wishes to express his hearty thanks to Professor OGUMA for his kind guidance and valuable suggestions. Thanks are also due to Dr. S. KUWAYAMA of the Hokkaido Agricultural Experiment Station, for his troublesome work in identification of species and his helpful advice. Further, the author is greatly indebted to

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1) Contribution No. 75 from the Zoological Institute, Faculty of Science, Hokkaido Imperial University, Sapporo.

Mr. S. MAKINO for his constant help and encouragement during the course of the work.

Material and Methods

The material for the present study consisted of the testes from five species belonging to the Chrysopidae. Most of the material was captured in the vicinity of Sapporo (Hokkaido) from June to September 1933, but some were obtained from the vicinity of Mt. Fuji (Honshu), in June 1934.

For the study, the testes of imagoes were employed as the material to avoid error of identification of species, since in the pupal and larval stages it seems nearly impossible to determine the name of the insects taxonomically by their body structure.

The testes were taken out by vivisection and put into the fixatives as soon as possible. The paired testes, oval in shape, are found situated in the dorsal parts of the abdomen. They are yellow in color, about 1 mm. in length and covered by a thick coating of adipose tissue.

As the fixative, FLEMMING's solution and CAROTHERS' fluid were applied. The sections were cut 7 micra in thickness and stained with HEIDENHAIN's iron-haematoxylin using light green as the counterstain.

Observations

1. *Chrysopa japana* OKAMOTO

This is the most common species in Sapporo. The testes from about fifty males were sectioned and examined in the present study.

As shown in Fig. 1, *A–B*, the spermatogonium contains twelve chromosomes, an even number, all of which are telomitic and rod-shaped, showing gradational diminution in length. They arrange radially in the equatorial plate assuming a typical rosette, as generally observed in Orthoptera and Diptera. A chromosome of nearly the smallest size is always found at the central space of the equatorial plate, surrounded by the others.

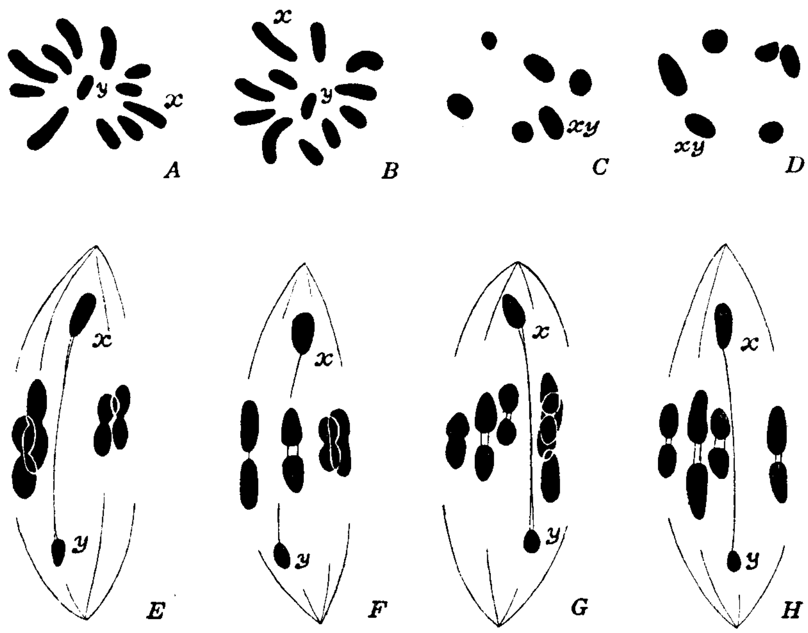

Fig. 1. *Chrysopa japana*. *A*, *B*, spermatogonial metaphase with 12 chromosomes; *C*, *D*, polar view of primary spermatocyte metaphase, showing 6 chromosomes; *E*, *F*, *G*, *H*, side view of primary spermatocyte meta-anaphase. ×3700.

As already mentioned the number of chromosomes is even; this will readily suggest the presence of a pair of sex-chromosomes. To find out the sex-chromosome in the garniture, every two homologous mates are checked out according to their size and shape as is the general custom. Thus we can distinguish an unequal pair of chromosomes in addition to five homologous pairs. The former is comprised of two chromosomes of different size so they may be considered as an XY-pair (see Fig. 1, *A–B*, *xy*). The difference of size between X- and Y-chromosome is very striking. The former attains a length intermediate between the first and the second largest autosomes while the latter is represented by the smallest chromosome. The X-element always takes a peripheral position and the Y-chromosome, on the contrary, is found at the central space of the equatorial plate.

The chromosomes found in the primary spermatocyte division constitute the metaphase plate in which every chromosome is widely separated from each other, owing probably to the existence of such a small number as six (Fig. 1, *C–D*). As seen in Fig. 1, *E–H*, the chromosomes are elongated dumb-bell shaped in lateral aspect and dispose themselves in the equatorial plate with their long axes parallel to the spindle axis. Consequently when observed from a pole of the spindle they appear like chromosomes of simple structure with round or oblong outline (Fig. 1, *C–D*). Generally they arrange to form a ring in the equatorial plate surrounding a space free from chromosomes at the centre. Of these six chromosomes, five are autosome bivalents and the remaining one, which occupies the peripheral position, is the XY-complex. The latter, differently from the autosomes, assumes an asymmetrical tetrad composed of unequal X- and Y-element, which are separated precociously in striking contrast to the others (Fig. 1, *E–H, xy*). In polar aspect, therefore, they can hardly be detected unless one changes the focus in considerable degree in both sides of the equator. The difference of size between X- and Y-component is very striking as already mentioned in case of the spermatogonium. In both elements the spindle fibre is constantly attached to their free ends, opposite the point where they come in connection.

As the result of the present division the X is completely separated from the Y and becomes involved into one of the daughter cells of which the other aquires the Y-chromosome. Hence, there should be produced two kinds of secondary spermatocytes with different chromosome garnitures in respect to the sex-chromosome components, in spite of the equal number of chromosomes.

2. *Chrysopa sapporensis* OKAMOTO

The present species is one of the closest allies to the preceding species, but is not so abundant in fields as the latter. In field collection, for instance, one can find only 2 or 3 specimens of *Ch. sapporensis* against one hundred of *Ch. japana*. About fifteen males were used as the material of the study. In parallel to the close taxonomic characteristics these two species much resemble each other in general features of the chromosomes.

The spermatogonium possesses twelve chromosomes as is clearly seen in metaphase (Fig. 2, *A–B*). They are all telomitic rod in shape and arrange themselves in a rosette form. By checking out every two homologous chromosomes, according to their size and shape, the X- and Y-element can be determined with facility (Fig. 2, *A–B*, *xy*).

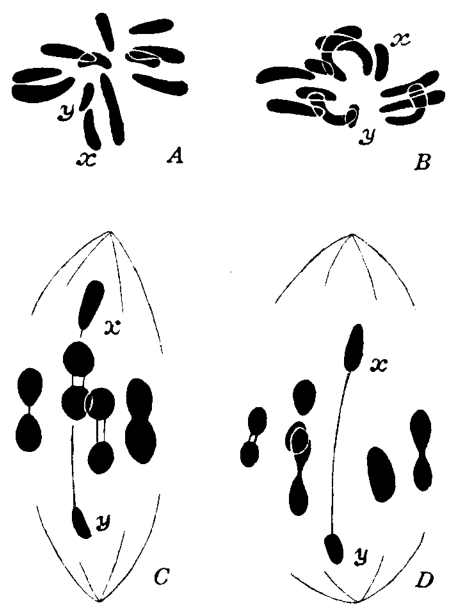

Fig. 2. *Chrysopa sapporensis*. *A*, *B*, spermatogonial metaphase with 12 chromosomes; *C*, *D*, side view of primary spermatocyte meta-anaphase. ×3700.

The chromosomes in the primary spermatocyte could be observed merely in the profile of the spindle, but not in the polar views, due to insufficient amount of material. Still we could find six bivalents, of which one forms a heteromorphic tetrad (Fig. 2, *D*). The latter is possibly the XY-complex showing precocious segregation (Fig. 2, *C–D*). The size difference between X and Y seems as great as in the preceding species.

3. *Chrysopa intima* MacLachlan

Most of material of the present species was collected in the vicinity of Mt. Fuji in June, 1934[1]. About twenty males were used, almost all being preserved in Carothers' fluid.

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1) For collection of the material the author is greatly indebted to Mr. M. Yago of the Shizuoka Agricultural Experiment Station.

According to OKAMOTO ('19), the present species is closely allied to, *Ch. perla*, which was already investigated by NAVILLE et de BEAUMONT ('33). The general features of the chromosomes closely resemble those of the latter species. Fig. 3, *A* and *B*, show the polar views of spermatogonia in metaphase, in each of which twelve chromosomes are clearly observed. In contrast to the case of *Ch. japana* and *Ch. sapporensis*, the chromosomes of this species are remarkably thick and short, which is probably due to the fixative, but they do not show any fundamental difference in structure as

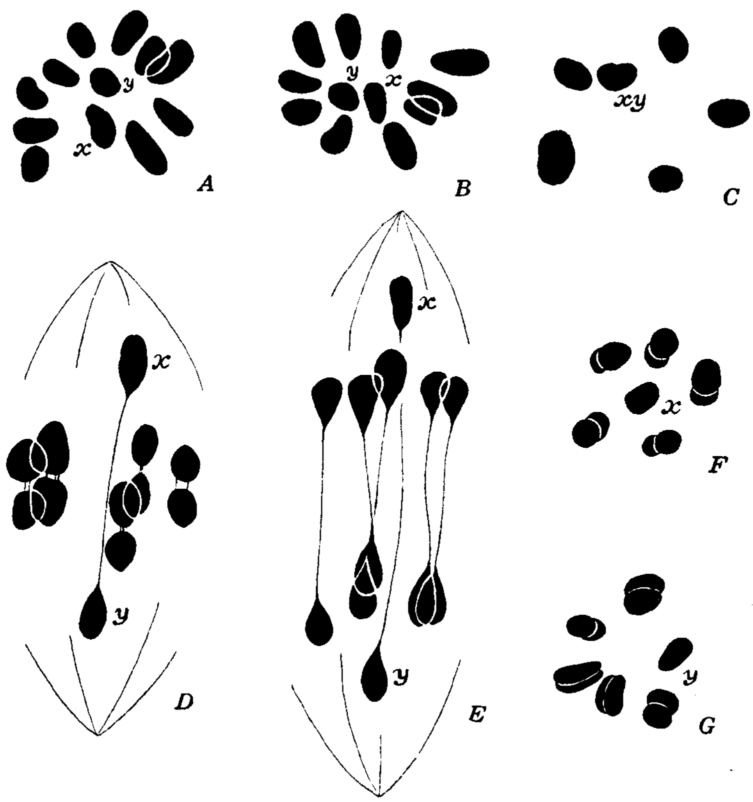

Fig. 3. *Chrysopa intima*. *A*, *B*, spermatogonial metaphase with 12 chromosomes; *C*, polar view of primary spermatocyte metaphase, showing 6 chromosomes; *D*, side view of primary spermatocyte metaphase; *E*, side view of primary spermatocyte anaphase; *F*, *G*, polar view of secondary spermatocyte metaphase, showing 6 chromosomes, *F*; X-class, *G*; Y-class. ×3700.

they are all rod-shaped and telomitic in fibre attachment. The X- and Y-chromosome are always found in the central space of the equatorial plate. Similar to *Ch. perla* but sharply distinguishable from any species investigated in the present study, the size difference between X- and Y-element is very small. This is also clearly demonstrated in the side view of the primary spermatocyte division (Fig. 3, *D–E*).

Fig. 3, *C* shows the polar view of the primary spermatocyte metaphase, where six bivalent chromosomes are to be discovered in circular arrangement. The XY-complex separated into composing elements in this division, just like in the other species studied, (Fig. 3, *D–E*). The secondary spermatocytes, thus produced, as shown in Fig. 3, *F* and *G*, present two kinds in respect to the sex-chromosome contained.

4. *Chrysopa kurisakiana* OKAMOTO

Together with the previous species, a male specimen of the present species was obtained at the same locality. As the study was carried out merely on this one pair of testes, it was impossible to follow every developing phase of the chromosomes in the material. Thus in the material only the primary spermatocyte chromosomes were to be observed. Fig. 4, *A* and *B*, show the polar view of the metaphase plates, in which six chromosomes in haploid condition are clearly demonstrated. The

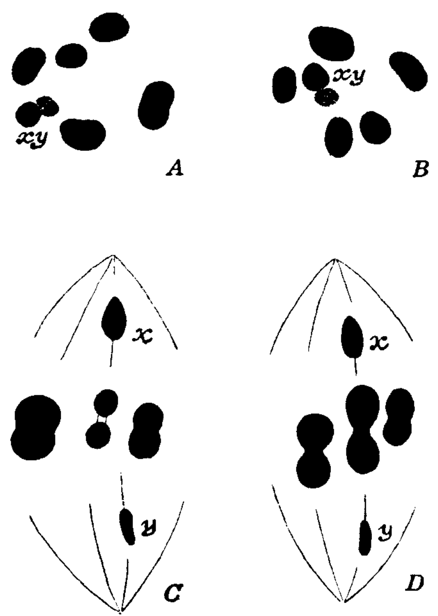

Fig. 4. *Chrysopa kurisakiana*. *A*, *B*, polar view of primary spermatocyte metaphase, showing 6 chromosomes; *C*, *D*, side view of primary spermatocyte metaphase. ×3700.

XY-complex segregates precociously as usual (Fig. 4, *C–D*, *xy*). From what is known about the primary spermatocyte, the spermatogonial complex of chromosomes is expected to be constituted of ten autosomes and an XY-pair, and the size ratio of the latter seems to be the same as in the case of *Ch. japana*.

5. *Chrysotropia japonica* NAKAHARA[1]

The species here described belongs to a different genus from the foregoing four species. This is rather rare in Sapporo. The testes from three males were employed as the material. It is of great importance that there was found in the present species, in correspondence with the taxonomical difference, a marked characteristic concerning the chromosome. It is the presence of V-shaped chromosomes, which have never been observed in any species of *Chrysopa* studied by the present author.

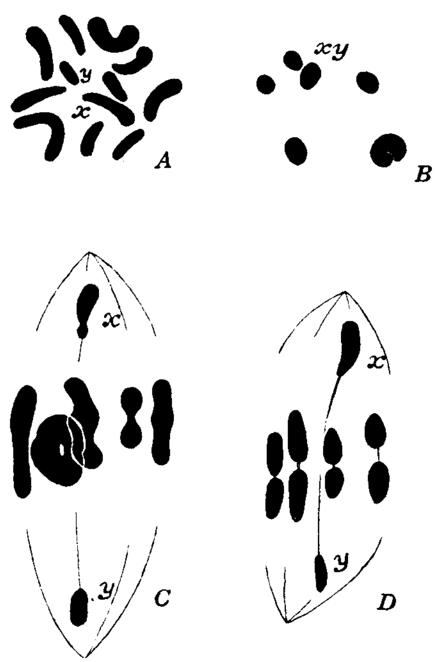

Fig. 5. *Chrysotropia japonica*. *A*, spermatogonial metaphase with 12 chromosomes; *B*, polar view of primary spermatocyte metaphase, showing 6 chromosomes; *C*, *D*, side view of primary spermatocyte meta-anaphase. ×3700.

The spermatogonium contains twelve chromosomes (Fig. 5, *A*), of which ten are rod-shaped with graduational diminution in size, but the remaining two are evidently V-shaped constituting one homologous pair. When the homologous pairs are checked

1) According to OKAMOTO ('19), the present species is the sole representative of the genus *Chrysotropia* in Japan.

out, one will soon notice two chromosomes of different size, which occupy the central space of the equatorial plate. These two should be an unequal pair of sex-chromosomes, X and Y (Fig. 5, *A*, *xy*).

In the polar view of the primary spermatocyte metaphase, are found six distinct chromosomes of bivalent nature as expected from the number in the spermatogonium (Fig. 5, *B*). One bivalent out of six is thick V-shaped in polar view while nearly lozenge-shaped in lateral aspect, in striking contrast to the remaining five, which are round or oblong in polar view while dumb-bell in profile. This chromosome is evidently the one descended from the two V-chromosomes found formerly in the spermatogonium.

The XY-complex extraordinarily widely separates into components as in other cases (Fig. 5, *C–D*, *xy*). Occasionally one finds a distinct constriction near one end of the X-element, where it comes in connection with the Y-element, as seen in Fig. 5, *C*, *x*. The significance of this constriction in the X-element is obscure at present.

Unfortunately, the chromosome of the secondary spermatocyte was not actually observed. It is very probable, however, that there are two kinds of spermatocytes, of which one contains X and the other Y.

Some Remarks

In their paper concerning the Neuropteran chromosomes, NAVILLE et de BEAUMONT ('33) described the chromosomes of six species of Chrysopidae. We know from their descriptions that four of the studied species, i.e., *Chrysopa perla*, *Ch. vulgaris*, *Ch. prasina* and *Ch.* (*Chrysotropia*) *alba*, have the chromosome formula 10N+XY for the male cell. In detail, it is composed of five homologous pairs plus an unequal pair of X- and Y-element and all are rod-shaped with telomitic fibre attachment. So far as the number and form of the chromosomes are concerned, therefore, the results obtained from these four species are in perfect accordance with those obtained in the Japanese species, *Ch. japana*, *Ch. sapporensis*, *Ch. intima* and

Ch. kurisakiana, as reported in the present paper. The matter is quite otherwise, however, in the remaining two species, *Ch. septempunctata* and *Ch.* sp. In the former species there are found 10 chromosomes which are to be formulated as 8N+XY, and two of the autosomes are represented by V-shaped chromosomes instead of rods. If we take the hypothesis of linkage between two different chromosomes of rod-shaped so as to produce a V, the numerical difference of this species can easily be accounted for, as NAVILLE et de BEAUMONT ('33) so consider.

The same formula 8N+XY will be also applied to the latter species, but there exists an important difference concerning the chromosome constitution, as there are found four V-shaped (two large and two small), four rod-shaped autosomes and X-Y elements. So the matter becomes very complex in this case upon considering linkage as in the preceding species. For 4 V's ought to be considered as derived from 8 rods, thereby the whole 10 chromosomes should account for 14 rod chromosomes.

Here we find one more different formula 10N+XY in *Chrysotropia japonica*, in which are present 2 large V-shaped and 8 rod-shaped autosomes plus X-Y chromosomes. This formula seems to be at first quite unique among species of the family Chrysopidae, but it can be brought into line as in the case of *Chrysopa* sp., just mentioned above, if the theory of linkage is adopted for V-shaped chromosomes.

NAVILLE et de BEAUMONT ('33) noted the size variation of the Y-chromosome according to the different individuals in *Chrysopa vulgaris*, just as in *Oecanthus* (MAKINO, '32). In every species studied in the present investigation, almost no size variation of the Y-element among individuals has been observed. Furthermore, the size ratio of the X- and Y-element is very conspicuous with the exception of the case of *Ch, intima*, as compared with the six species described by NAVILLE et de BEAUMONT ('33).

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