A COMPARATIVE STUDY OF THE CHROMOSOMES IN THE INDIAN DRAGONFLIES[1)]

BY

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Introduction

Barring the pioneer work of OGUMA ('15, '30, '32) and a paper by SMITH ('16) singularly few investigators have devoted their attention to chromosome studies on this very ancient order of insects, Odonata. In a paper published by OGUMA and ASANA ('32) attention was drawn to a very interesting observation, whose significance is not yet quite clear, that the m-chromosome presents every grade of size reduction among testicular cells of a single individual belonging to a species of Odonata, *Tramea chinensis*, collected in the vicinity of Gujarat College, Ahmedabad, about 300 miles north of Bombay, Western India.

This observed fact of size gradation of the m-chromosome in the testicular cells of a single specimen of the dragonfly, *T. chinensis*, coupled with the fact that one of us happened to be transferred on duty to a place farther south, nearer Bombay, to a locality richer in dragonfly fauna, acted as a stimulus for collecting additional material for further investigation. However, it must at once be admitted that the account presented in this paper throws little light, if any, on the bearing of this remarkable fact, upon the taxonomic studies

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of the Indian dragonflies, nor does it give any clue to any other significance of this cytological observation.

This paper is written with the purpose of making a comparative examination of the chromosomes in the male germ cells of the ten species of Indian dragonflies so far collected.

The authors are greatly indebted to Prof. KAN OGUMA for his encouragement and helpful suggestions in this study. They also express their thanks to Colonel F. C. FRAZER, Indian Medical Service, who kindly identified all the specimens. Special thanks are also extended to Mr. HIDEJIRO NIIYAMA for his kind assistance in the preparation of sections.

Material and Methods

All the specimens for the present study were collected from a very small, restricted area of slightly rising ground situated in the neighbourhood of the Ismail College, Jogeshwari, about 20 miles north of Bombay, in the Salsette sub-division of the Thana district, Western India. Salsette lies quite close to the Arabian sea at the foot of the hills, the Western Ghats. The country offers for the most part an intermixture of wood and village, and contains several lakes and reservoirs. All the dragonflies furnishing the material for the present study were collected during a period of five months, between July 7 and December 7, 1932.

All the material studied was obtained from specimens belonging to ten species of dragonflies which, according to FRAZER ('24), represent two families of the Sub-order Anisoptera and one family of the sub-order Zygoptera as listed below:

Sub-order Anisoptera
Family Libellulidae
Sub-family Libellulinae

1. *Pantala flavescens*
2. *Tramea limbata*
3. *Trithemis pallidinervis*
4. *Diplacodes trivialis*

5. *Brachythemis contaminata*
6. *Crocothemis servilia*
7. *Potamarcha obscura*
8. *Orthetrum sabina*

Family Aeschnidae
Sub-family Gomphinae
9. *Ictinus rapax*

Sub-order Zygoptera
Family Coenagrionidae
10. *Ceriagrion rubiae*

To avoid error of identification of species the testes used in this study were taken from maturing adults as was done by OGUMA ('30). Immediately after the specimens were captured, the male gonads were dissected out by vivisection in normal saline and dropped into the fixatives. The fixatives used were, (1) FLEMMING's strong solution with reduced acetic, (2) the same diluted with distilled water, (3) ALLEN-BOUIN's solution, (4) modified BOUIN's solution (P.F.A.$_3$). All these fixatives gave good results, FLEMMING's solution proving excellent for the preservation of spermatogonial chromosomes. Sections were cut eight micra in thickness and subjected to HEIDENHAIN's iron-haematoxylin method of staining using light green as the counter-stain.

All the figures in the text were drawn with the aid of the camera lucida using a Zeiss apochromatic objective 1.5 mm and a compensating ocular, K 20, under the magnification of 4200 diameters.

Observations

a.—Family Libellulidae

Sub-family Libellulinae

The chromosomes observed in representatives of six genera namely, *Orthetrum, Sympetrum, Libellula, Somatochlora, Trithemis* and *Tramea* of this sub-family have already been reported upon in

two papers previously published, OGUMA ('30) and OGUMA and ASANA ('32). In the present study specimens from eight genera of Indian dragonflies have been examined, five of which have furnished new material for the first time. However, with regard to the morphological characters of the chromosomes, their number, arrangement and mode of division, the present observations do not markedly differ from the results obtained in the previous studies above mentioned. All the species here recorded show uniformly 25 chromosomes as their diploid number, while their haploid number is 13. In most cases the chromosomes arrange themselves concentrically at the metaphase both in spermatogonial and primary spermatocyte divisions as in other dragonflies. The individual chromosomes when viewed from the pole, appear rod-like in the diploid group, lozenge or dumbbell shaped in the primary and rounded in the secondary spermatocytes. In the spermatogonial and primary spermatocyte metaphase the chromosomes lie in the equatorial plate with their axes tangential to the spindle axis; in the secondary spermatocyte at the same stage they lie with their long axes parallel to the spindle axis.

The smallest pair of autosomes, the so-called m-chromosomes of OGUMA ('30), is found in all the specimens examined and they vary in magnitude from species to species. Taking the size of the X-chromosome, which seems to be nearly the same in all species at least of the sub-family Libellulinae, as a standard of measurement, an attempt has been made in the present investigation to compare it with the m-chromosomes in different species studied, with a view to estimating the variation in magnitude of the m-chromosome from species to species.

In all cases the X-chromosome is unpaired and it invariably takes a peripheral position in the equatorial plate, both in the diploid and in the haploid complex of chromosomes. As is the case with the majority of dragonflies so far investigated, it divides equationally at the metaphase of the primary spermatocyte, but goes undivided to one pole in the secondary division, showing a conspicuous precession in the course of this division. It is rather small and its relative

magnitude seems to be almost equal in every species examined just as has been reported for the Japanese forms studied by OGUMA ('30). If it be not actually of the same size, it varies but little in its magnitude from species to species. In a diploid group at the metaphase the X-chromosome seems nearly equal in size to the next to the smallest autosome. Consequently in the bivalent group it appears to be approximately half the size of the next to the smallest bivalent. In no case was the X-chromosome in its magnitude undistinguishable from the other autosomes, as was the case with *Gomphus unifasciatus* reported by OGUMA ('30).

In the mode of separation of the chromosomes of the secondary spermatocytes the Indian forms differ in no way from those of the Japanese forms (OGUMA '30). As the chromosomes separate, their middle portions gradually become narrower till the two halves are constricted off.

1. *Pantala flavescens*

Two pairs of testes from two specimens obtained in July and August 1932 constitute the material, a part of which was fixed in FLEMMING'S solution and the remainder in ALLEN-BOUIN'S solution.

From the taxonomic standpoint this species seems to be allied to some members of the genus *Tramea,* one species of which has already been dealt with cytologically by OGUMA and ASANA ('32), while the observations on one more species of the same genus, *Tramea,* are recorded in the next succeeding section of this paper.

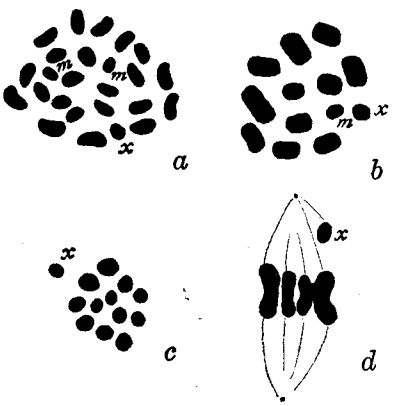

Fig. 1. *Pantala flavescens.* *a*, spermatogonium. *b*, primary spermatocyte. *c*, secondary spermatocyte. *d*, side view of secondary spermatocyte division. 4200×.

As seen in the equatorial plate at the metaphase of the spermatogonium there are 25 small rod-shaped, slightly curved chromo-

somes which vary in length (Fig. 1, *a*). Their arrangement on the spindle is in no way different from that of the chromosomes of *Trithemis* and *Tramea* as described by OGUMA and ASANA ('32).

Among these 25 univalents there is one almost constantly situated in the outer circle of the metaphase. This element assumes the shape of a condensed rod slightly pointed at its inner end, which, though other elements can be paired as homologous chromosomes, is found to have no mate. This is regarded as the solitary X-element (*x* in Fig. 1, *a*). A homologous pair of the smallest chromosomes in the univalent group is the pair of m-chromosomes (*m* in Fig. 1, *a*), which in magnitude seem to be slightly smaller than the X when viewed from the pole.

The primary spermatocyte metaphase shows 13 bivalents composed of 12 autosome tetrads and an X-element (Fig. 1, *b*), the latter, always occupying a peripheral position at this stage, assumes a conspicuous form in striking contrast to other autosome bivalents. In the first division all the chromosomes including the X-element are divided into equal halves, with the result that the daughter cells thus produced, the secondary spermatocytes, are all of one kind having a similar complex of chromosomes. Therefore, at the metaphase of the secondary spermatocytes 13 chromosomes are uniformly observed, of which 12 are the autosome dyads and one is the X-chromosome (Fig. 1, *c*). The latter always stands a little apart from the autosome group (*x* in Fig. 1, *c*). In the ensuing division the X, unlike the autosomes, does not divide but goes to one of the two poles entire, advancing towards it much earlier than the autosome group of its own side as seen in the side view of the spindle (Fig. 1, *d*). Thus are obtained two kinds of spermatids, one having the X-element, the other without X.

2. *Tramea limbata*

A pair of testes from a single specimen secured in November 1932 comprises the material for this study. The fixative used was modified BOUIN's solution (P.F.$A._3$).

In all respects the chromosomes of this species show a close resemblance to those of the taxonomically related species, *T. chinensis* described by OGUMA and ASANA ('32). As seen in Fig. 2, *a–c* the diploid number is 25 and the haploid 13. In this species as compared with the preceding one the m-chromosomes in the diploid set are of conspicuously smaller size in relation to the other autosomes (Fig. 2, *a*), and this condition is again observed in the primary and secondary spermatocytes (Fig. 2, *b–c*). When viewed from the pole the m-chromosome seems to be about half the size of the X-element.

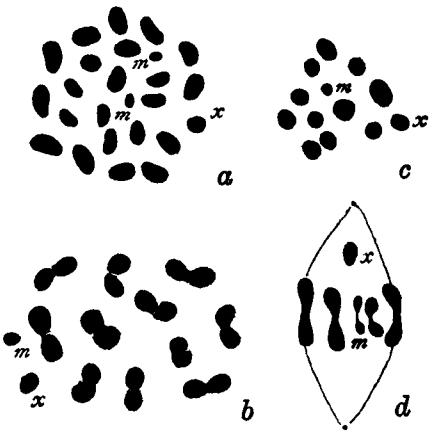

Fig. 2. *Tramea limbata*. *a*, spermatogonium. *b*, primary spermatocyte. *c*, secondary spermatocyte. *d*, side view of secondary spermatocyte division. 4200×.

The autosome bivalents, except the m-bivalent, appear dumb-bell shaped and show a transverse suture in the middle in polar view (Fig. 2, *b*). In a superficial examination this suture may seem to be a characteristic of the tetrads in this species in contrast to the tetrads described in the preceding section. But that it is not so and that this suture is merely an artifact will be presently seen when we consider the next case, the chromosomes of *Trithemis pallidinervis*. Furthermore, this conclusion is borne out by the fact that if the preparations of *Tramea limbata* are strongly destained then only the tetrads show this suture, otherwise they do not. The m-bivalent never exhibits such a suture and assumes an appearance similar to the X-dyad. The X-chromosome separates into two equal halves in the first division, but goes to one pole entire, without separation, in advance of other chromosomes in the succeeding division of the secondary spermatocytes (Fig. 2, *d*).

3. *Trithemis pallidinervis*

Specimens of this species were secured in November 1932 and the fixative was modified FLEMMING's solution.

In number, form and other morphological features, the chromosomes of this species resemble those of the dragonfly, *Trithemis aurora,* reported upon by OGUMA and ASANA ('32). There are 25 chromosomes in the spermatogonium (Fig. 3, *a*), and 13 are seen at the metaphase in both the primary and secondary spermatocytes (Fig. 3, *b–d*). In its magnitude relative to the X-element the m-chromosome of this species shows no marked difference from that of *Trithemis aurora,* and when viewed from the pole it is a little less than half as large as the X-element.

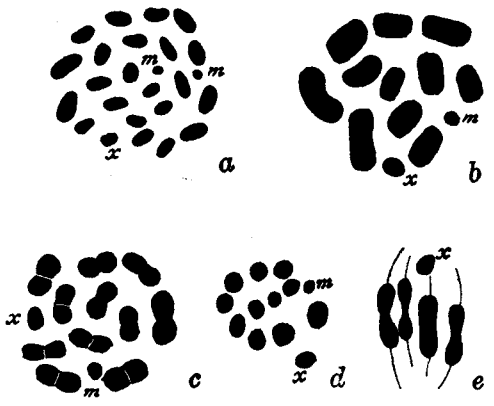

Fig. 3. *Trithemis pallidinervis*. *a*, spermatogonium. *b* & *c*, primary spermatocytes. *d*, secondary spermatocyte. *e*, side view of secondary spermatocyte division. 4200×.

When one examines the bivalent chromosomes at the metaphase of the primary spermatocyte as drawn in Fig. 3, *b* and *c* the bivalents of one group, excepting the X and m-bivalent, look very different from those of the other, so much so that one may be misled into believing that Fig. 3, *b* and *c* represent two different types of primary spermatocytes. As noted previously, these conditions are not fundamentally different from each other. When deeply stained, the chromosomes appear more condensed and massive as in Fig. 3, *b*, when strongly destained they show a conspicuous transverse constriction as in Fig. 3, *c*. Regarding the nature of this transverse suture an explanation has been offered by OGUMA and ASANA ('32) with reference to a similar condition in *Trithemis aurora.*

4. *Diplacodes trivialis*

The following observations are based on the study of a pair of testes obtained from an individual captured in September 1932; the fixative was modified FLEMMING's solution.

This species in its taxonomic characters is closely related to the members of the genus *Sympetrum,* the chromosomes of which have been examined by OGUMA ('30).

The chromosomes of *D. trivialis* resemble in all important particulars such as number, form and arrangement, the chromosomes of the foregoing species already described. The spermatogonium contains 25 chromosomes (Fig. 4, *a*). In both the primary and the secondary spermatocytes the number of chromosomes is 13 (Fig. 4, *b–d*). The X-element as in Fig. 4, *e,* takes a peripheral position on the spindle and shows post-heterokinesis.

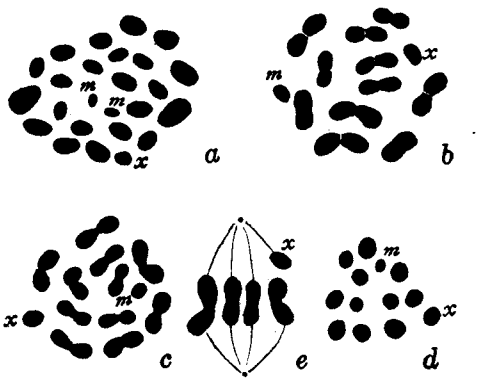

Fig. 4. *Diplacodes trivialis.* *a,* spermatogonium. *b* & *c,* primary spermatocytes. *d,* secondary spermatocyte. *e,* side view of secondary spermatocyte division. 4200×.

The comparison of the chromosomes between the present species and the representatives of *Sympetrum* furnishes striking evidence in regard to the relative magnitude of the m-chromosome. According to OGUMA ('30), the m-chromosome in *S. pedemontanum* is very minute, while in *S. frequense* the reduction of the m-chromosome has led to its total disappearance from the complex, with the result that only 12 chromosomes constitute the haploid number. In *Diplacodes,* the m-chromosome is invariably about half the size of the X-element in polar view.

5. *Brachythemis contaminata*

Testes of three specimens were used in this study. These were captured in September 1932. A part of the testes was fixed in ALLEN-

BOUIN's solution and the remainder in modified BOUIN's solution (P.F.A.$_3$).

Though this species is closely allied to *D. trivialis* just treated, its chromosomes individually seem to be larger in size when compared with those of the latter. This observation is reinforced by the fact that while the m-chromosome in *Diplacodes* species is about half the size of the X, in the present species it is nearly one-third (See Fig. 5, *a–c*).

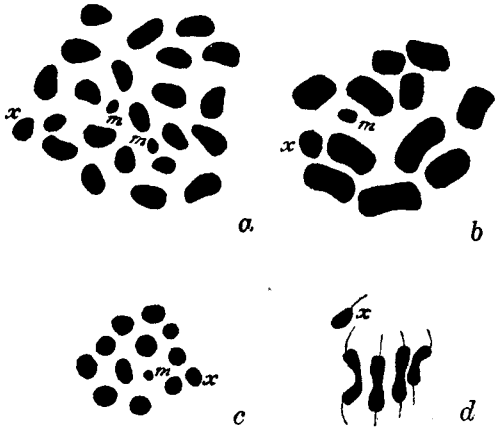

Fig. 5. *Brachythemis contaminata*. *a*, spermatogonium. *b*, primary spermatocyte. *c*, secondary spermatocyte. *d*, side view of secondary spermatocyte division. 4200×.

The diploid number is 25 in the spermatogonium, the haploid 13 as seen in the primary and secondary spermatocytes (Fig. 5, *a–c*). The X-chromosome always takes a peripheral position in the equatorial arrangement and shows post-heterokinesis (*x* in Fig. 5, *a–d*).

6. *Crocothemis servilia*

The testes from one individual were fixed in September 1932 and those from the other in November of the same year. ALLEN-BOUIN's solution and modified BOUIN's solution (P.F.A.$_3$) were employed as the fixatives. A large number of division figures and other phases of spermatogenesis were observed.

Fig. 6, *a* shows 25 chromosomes in the spermatogonium, of which the autosomes are in 12 pairs, and the remaining one is the X-element. Among the autosomes, the m-chromosomes stand out quite clearly because of their strikingly small size as compared with the rest. Thirteen chromosomes in the primary spermatocytes are shown in Fig. 6, *b–c*, and the same number is depicted in the metaphase of

the secondary spermatocyte (Fig. 6, *d*). The X-element, as in all the cases mentioned above, lies on the outer circle of the spindle, divides equationally in the first division and runs ahead towards one pole without separation in the second division (Fig. 6, *e*). In relative magnitude the m-chromosome is about one-third the size of the X-element.

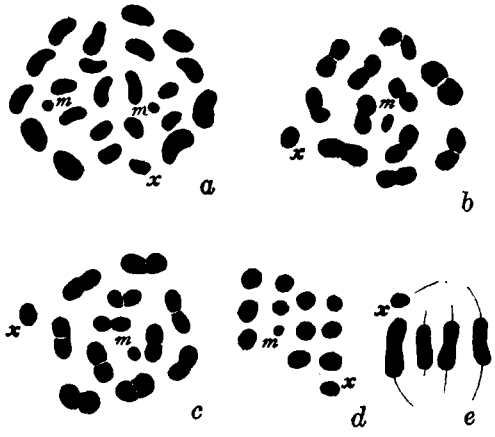

Fig. 6. *Crocothemis servilia*. *a*, spermatogonium. *b* & *c*, primary spermatocytes. *d*, secondary spermatocyte. *e*, side view of secondary spermatocyte division. 4200×.

7. *Potamarcha obscura*

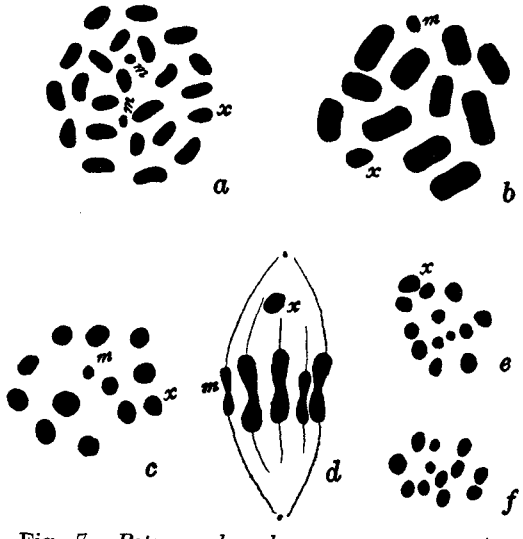

Fig. 7. *Potamarcha obscura*. *a*, spermatogonium. *b*, primary spermatocyte. *c*, secondary spermatocyte. *d*, side view of secondary spermatocyte division. *e* & *f*, sister groups of secondary spermatocyte division anaphase. 4200×.

The material for this study consists of two pairs of testes fixed during July and August 1932, one pair in modified BOUIN's solution (P.F.A.$_3$) and the other in FLEMMING's solution.

This specimen is closely related to the above, *Crocothemis servilia*. No marked difference is seen as regards the morphological characters and behaviour of the chromosomes between these two related species.

The diploid number is 25, the haploid 13 as seen in Fig. 7, *a–c*. The X-element is peripheral in position in both the diploid and the haploid group. As in the other species described above, it goes undivided to one pole in the secondary spermatocyte division as in Fig. 7, *d* and *e–f*. The m-chromosome, when observed from a pole, appears to be smaller than about one-third of the X-element in magnitude.

8. *Orthetrum sabina*

A pair of testes from a specimen of this species was fixed in modified BOUIN's solution (P.F.A.$_3$) in September 1932, which served as the material for investigation.

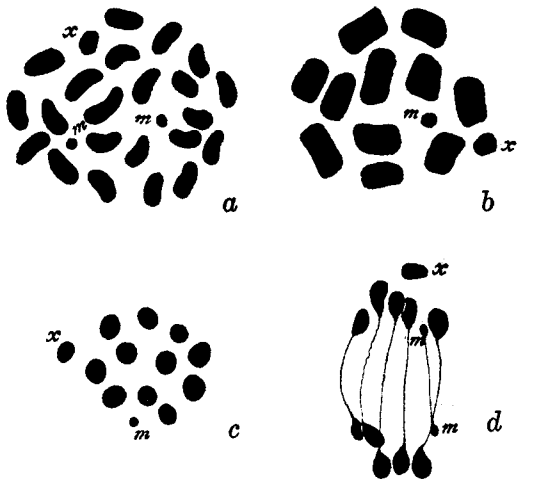

Fig. 8. *Orthetrum sabina*. *a*, spermatogonium. *b*, primary spermatocyte. *c*, secondary spermatocyte. *d*, side view of secondary spermatocyte division. 4200×.

The chromosomes of *Orthetrum albistylum* and *O. japonicum*, two Japanese species of this genus, have been examined by OGUMA ('30). The Indian form, *O. sabina*, is a close ally of the Japanese species, and it is probably therefore that the number, morphological characters and behaviour of the chromosomes of these three allied forms show a marked resemblance. The diploid number in *O. sabina* is 25 and the haploid 13 (Fig. 8, *a–c*). However, a noteworthy feature when these species are compared is a graded difference in the relative magnitude of the m-chromosome as compared with the X-element from species to species. In other words, the m-chromosome is nearly equal to the X-element in *O. albistylum;* in *O. japonicum* it is smaller than half the X (see OGUMA '30) ; while in *O. sabina*, it is much smaller, less

than one-third of the X-chromosome (Fig. 8, *a–d*). Thus, this very interesting character of the m-chromosome may perhaps prove useful in confirming specific distinctions in the taxonomy of this group.

b.—Family Aeschnidae

Sub-family Gomphinae

OGUMA ('30) has studied three species of the genus *Gomphus*, belonging to this sub-family. An account of the chromosomes of an Indian species, *Ictinus rapax*, of the same sub-family is presented below. As far as the mode of chromosome arrangement, behaviour of the X-chromosome and type of the division of the secondary spermatocyte chromosomes are concerned, the present observations on *Ictinus* are in agreement with those of OGUMA ('30) on the Japanese genera of the same sub-family, excepting the remarkable fact that the X-chromosome in the Indian form assumes such an enormous size that we know nothing comparable to it in all the cases hitherto described.

9. *Ictinus rapax*

The male gonads of this species as compared with those of other species are very large in volume, and in this respect the *Ictinus* under investigation can clearly be distinguished from all the members of the family Libellulidae of the sub-order Anisoptera. The testes from three different specimens obtained in July and September 1932 were fixed, some in FLEMMING's strong solution and some in modified BOUIN's solution (P.F.A.$_3$).

The metaphase spindle in the present species is extremely large and the individual chromosomes also attain an uncommonly large size as shown in Fig. 9, *c–d*. In none of the dragonflies of the families Libellulidae and Aeschnidae so far investigated, are these unusual features met with as regards the size of the chromosomes and the spindle. But, OGUMA ('30) records a case, that of *Mnais strigata* belonging to Calopterygidae, in which the chromosomes are of very large size; however, the chromosomes of *Ictinus rapax* are still larger.

By a careful examination of many excellent division figures, it was ascertained that there are 23 chromosomes in the spermatogonia (Fig. 9, *a–b*) and 12 in the primary and secondary spematocytes (Fig. 9, *c–e*). Therefore, in respect to the number of chromosomes the Indian species does not materially differ from the Japanese form, *Gomphus suzukii*, studied by OGUMA ('30).

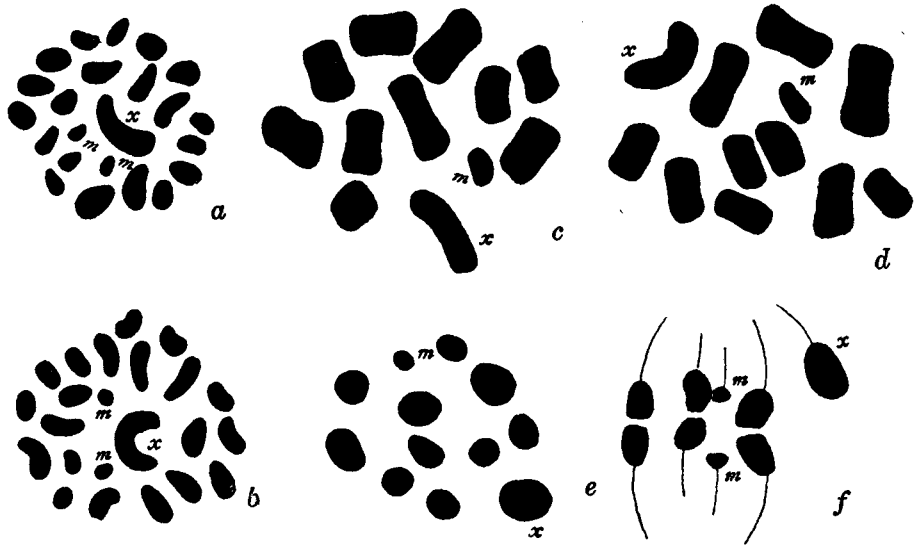

Fig. 9. *Ictinus rapax*. *a* & *b*, spermatogonia. *c* & *d*, primary spermatocytes. *e*, secondary spermatocyte. *f*, side view of secondary spermatocyte division. 4200×.

In all the metaphase sections of the spermatogonia the presence of a chromosome of enormous size, gently curved and rod-shaped, offers a striking contrast to the remaining, short, compact chromosomes by which it is surrounded. This, the largest element, almost always lies in a central region of the spindle; and by picking out every two homologous members of a pair in accordance with their shape and size, it can easily be determined that this huge chromosome remains unpaired and is the X-chromosome (*x* in Fig. 9, *a–b*). Again, one finds this conclusion to be fully warranted by observing the behaviour of this element in the maturation division (*x* in Fig. 9,

c–f). Moreover, this remarkably large size of the X-chromosome makes the relative magnitude between itself and the smallest m-chromosome most conspicuous.

When the primary spermatocyte division is observed it is found that this distinctive appearance of the X-element is still maintained. It is the largest rod, somewhat slender and curved, while the autosomes are of various sizes and all assume an irregular lozenge shape. The X-element is always situated in the outer circle of the spindle and divides equationally, as do the other autosome bivalents, in the anaphase (*x* in Fig. 9, *c–d*).

The chromosomes, when viewed from the pole, at the metaphase of the secondary spermatocytes are found to vary in size and appear to be more or less oval in shape. Here too, the X-element is the largest of all, and occupies without exception an eccentric position (*x* in Fig. 9, *e*). The ensuing division of all the autosomes is initiated by the appearance of a clear transverse split in the middle of each chromosome, and as the division proceeds the two straight, truncated ends of each chromosome segregate into two daughter elements derived from each member (Fig. 9, *f*). This mode of division of the secondary spermatocyte is what is called the aeschnid type by OGUMA ('30) to distinguish it from the libellulid type. The X-chromosome in the secondary spermatocyte, unlike the autosomes, does not divide but migrates to one pole without division as seen in Fig. 9, *f*.

Finally, it must be emphasized that the X-chromosome of *Ictinus rapax*, the Indian form, in assuming such an uncommonly large size as a member of the chromosome complex of Odonata furnishes an exceptional, almost unique example among the chromosomes of the dragonflies. It is true that the X-chromosome of *Gomphus unifasciatus* studied by OGUMA ('30) shows likewise a comparatively large size. But in that case, in respect to its magnitude, there is so little difference from the autosomes that it is almost impossible to point it out. And in all the other dragonflies so far studied it is no where so large. In almost all these forms, the X-element as seen in the

metaphase of the primary spermatocyte is about half as large as the next to the smallest autosome.

c.—Family Coenagrionidae

So far as the literature shows, no species belonging to this family has been studied cytologically up to the present time. The following observations have been made on the chromosomes of the male gonads of only one species from India.

10. *Ceriagrion rubiae*

Specimens were obtained in August and September 1932 and the testes were fixed in modified BOUIN'S solution (P.F.A.$_3$) and also in modified FLEMMING'S solution.

The equatorial metaphase plates of all the spermatogonial cells show without exception, 27 chromosomes (Fig. 10, *a*). This number

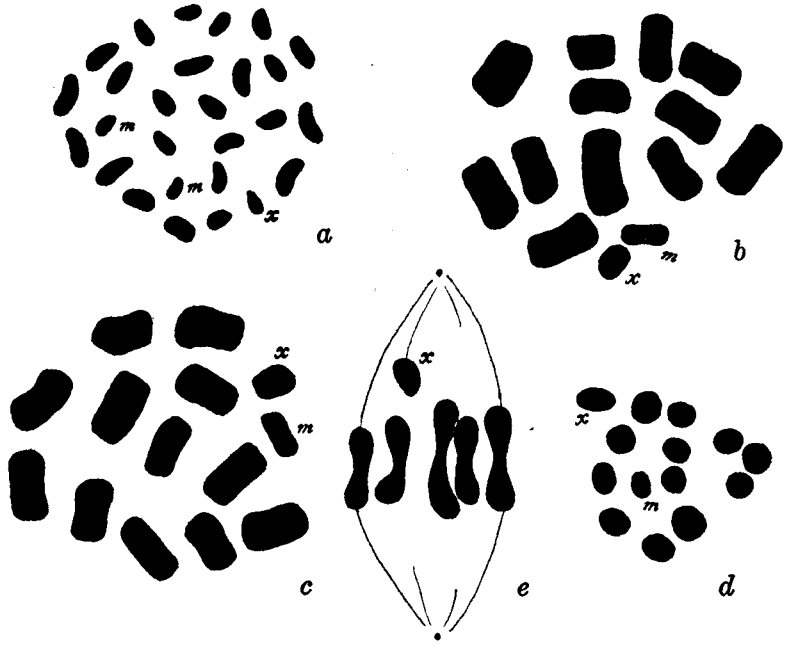

Fig. 10. *Ceriagrion rubiae*. *a*, spermatogonium. *b* & *c*, primary spermatocytes. *d*, secondary spermatocyte. *e*, side view of secondary spermatocyte division. 4200×.

seems to be rather rare, as, besides this Indian form, the same number has been reported only in one other, among all the species of dragonflies so far studied. This is *Anax junius*, a species of Aeschnidae studied by LEFEVRE and MCGILL ('08).

A pair of m-chromosomes is found towards the middle of the equatorial plate, lying among other autosomes. In the polar view of the metaphase, the X-element cannot be easily distinguished, because the difference in size of the individual chromosomes composing the complex is almost inappreciably small, though the slender form and the characteristic peripheral position of the X-chromosome may serve to some extent for its identification (x in Fig. 10, a).

Coming to the primary spermatocytes, one is astonished to find that both the chromosomes and the metaphase spindle show unexpectedly large dimensions, which is reminiscent of the state of things observed in the preceding species, *Ictinus rapax*, coming from the same geographical region. Therefore, so far as the size of the chromosomes is concerned, the chromosomes of *I. rapax* and of *Ceriagrion* seem to be the largest among those of all the dragonflies described hitherto by previous investigators.

The 14 bivalent chromosomes constitute the equatorial plate at the metaphase of the primary spermatocyte (Fig. 10, b–c). Excepting the X-element and m-bivalent all the tetrads are lozenge shaped and somewhat elongated horizontally. They do not markedly differ in size from one another. While the X-chromosome appears as a compact oval body, taking its usual peripheral position on the spindle, the m-bivalent seems to be thinner and lies in the outer circle of the spindle. Though the latter is somewhat elongated, in the polar aspect it seems to be equal to the X in absolute magnitude. In the anaphase, all the chromosomes, including the X-element, separate into two equal halves.

All the secondary spermatocytes show, without exception, 14 chromosomes at the metaphase (Fig. 10, d). As was the case in the primary spermatocytes, the chromosomes of the secondary also fail to exhibit any conspicuous size difference among themselves as seen

in Fig. 10, *d*. The identification of the m-chromosome is, therefore, rather difficult at this stage; nor is the magnitude of the X-chromosome such as to enable one to pick it out from among the rest, were it not for its eccentric position (*x* in Fig. 10, *d*). In the ensuing division the autosomes of the secondary spermatocytes separate equally, while the X-element goes to one pole without division as shown in Fig. 10, *e*.

Thus it will be easily seen that these peculiarities in size and number of the chromosomes of *Ceriagrion rubiae* described above are remarkable characteristics which distinguish the chromosomes of this Indian form from the chromosomes of all the other species of dragonflies studied by previous investigators.

Summary

1) The numerical relation between the chromosomes of ten species of dragonflies from a very restricted region in Western India dealt with in the present paper is given in the following table:

Species	Haploid	Diploid
Libellulidae		
Pantala flavescens	13	25
Tramea limbata	13	25
Trithemis pallidinervis	13	25
Diplacodes trivialis	13	25
Brachythemis contaminata	13	25
Crocothemis servilia	13	25
Potamarcha obscura	13	25
Orthetrum sabina	13	25
Aeschnidae		
Ictinus rapax	12	23
Coenagrionidae		
Ceriagrion rubiae	14	27

As will be seen in the above table, all the members of the Libellulidae show a constancy in number, 25 in diploid and 13 in haploid,

through all the species studied. In *Ictinus* belonging to another group, Aeschnidae, the diploid is 23, the haploid 12. The chromosome number in *Ceriagrion* (Coenagrionidae), is the largest among all the dragonflies studied so far.

2) Despite this variation in the chromosome number, there is present an unpaired X-chromosome, without exception, in all species investigated. In every species, the X-chromosome always takes a peripheral position on the outer circle of the spindle in the spermatogonial and primary spermatocyte metaphases and acquires a peculiar eccentric position in the metaphase of the secondary spermatocyte. It separates into two equal halves in the primary spermatocyte division, but migrates to one pole entire, without separation, ahead of other chromosomes in the secondary spermatocyte division.

In Libellulidae, on the whole, the X-chromosome seems to be of almost uniform size in every species examined. It looks nearly, though not exactly, equal in magnitude to the next to the smallest autosome univalent in the spermatogonial complex. In the bivalent group, it appears approximately half as large as the next to the smallest bivalent.

In *Ictinus rapax* (Aeschnidae) the circumstances are different. The largest element in the chromosome complex of this species is the X-chromosome. It is remarkably large and occupies a central position in the spermatogonial metaphase. These are the most distinctive characteristics of the chromosomes of this species.

In *Ceriagrion rubiae* (Coenagrionidae), the condition of the X-chromosome resembles that of the same element in the members of Libellulidae. In the primary spermatocyte metaphase, the X-chromosome appears to be nearly half as large as the next to the smallest bivalent.

3) In all the Indian dragonflies representing the three groups above mentioned the smallest autosome, the so-called m-chromosome, is present without exception. It varies in magnitude from species to species. The varying magnitude of the m-chromosome relative to the size of the X-element in each species is given in the following table:

The size relation between the m-chromosome and the X-chromosome

Species	The m-chromosome
Pantala flavescens	slightly smaller than X
Tramea limbata	about a half of X
Trithemis pallidinervis	smaller than half of X
Diplacodes trivialis	about a half of X
Brachythemis contaminata	nearly one-third of X
Crocothemis servilia	about one-third of X
Potamarcha obscura	smaller than one-third of X
Orthetrum sabina	smaller than one-third of X
Ictinus rapax	very much smaller than X (X being very large)
Ceriagrion rubiae	about equal to X in absolute magnitude

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