CHROMOSOME NUMBER AND BEHAVIOUR IN THE GRASSHOPPER *PHOLIDOPTERA*

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1. INTRODUCTION

CHROMOSOME form and number in closely related animal groups are often remarkably constant. Indeed, for this reason, close similarity or great difference may be of interest taxonomically. In the sub-family Decticinæ (Orthoptera : Tettigoniidæ) four of the five Old World genera so far studied cytologically (*Decticus*, *Metrioptera*, *Gampsocleis* and *Platycleis*) are recorded as having a diploid number of 30+X rod-shaped chromosomes in the male (White, 1954). The American genera *Anabrus* (McClung, 1905, 1914), *Steiroxys* (Davis, 1908) and *Atlanticus* (White, 1941) differ considerably, however, having one or more pairs of autosomes metacentric. In the latter genus the X is also metacentric.

The single deviant in the Old World genera was *Pholidoptera*, where White (1941, 1954) claimed that an additional pair of autosomes was present in the species *Pholidoptera griseoaptera* (DeGeer), the new male number being $2n = 32+X$. Unfortunately, however, he gave no indication of the size range or centromere position of the new autosomal pair; neither did he publish any figures from which this could be deduced. Furthermore, he did not say how many meiotic cells of the two individuals he examined possessed clear counts of this number, no mitoses being observed.

A re-investigation of the cytology of this species was considered worthwhile, in view of the singular deviation of this record from those of the other related genera.

Five male imagines were collected near Welsh St Donat's, Glamorganshire, S. Wales, during the first week of August 1960. I am grateful to Mr T. Parsons for assistance on the collecting trip. The testes were vivisected in insect saline and, after removing the sparse fat body with needles, fixed in 1 : 3 acetic alcohol. Squash preparations were made, staining with acetic orcein.

2. OBSERVATIONS

(i) *The standard karyotype*

In some 1000 cells at all stages of meiosis, taken from the five individuals, the normal male count was $n = 15\pm X (2n = 31)$, the number possessed by the four other related Old World genera.

All members of the complement are rod-shaped, representing either an acro- or telocentric organisation (*cf.* Marks, 1957). The 15 pairs of autosomes may be grouped into three size ranges, large (L1-2), medium (M3-8) and small (S9-15) (plate, fig. 1). The large and medium chromosomes are readily distinguished from one another, but the smaller members of the complement are very close morphologically and often cannot be identified with certainty. The X chromosome is unpaired in the male, and, as in all other Tettigoniidæ, is extremely large. When at a comparable stage of contraction it approaches the size of the L1.

(ii) *Numerical deviants*

Seven cells were found in one individual exhibiting departures from the normal count, all apparently results of non-disjunction:

(*a*) Single cells at diplotene, diakinesis and first metaphase possessed two X chromosomes (*cf.* plate, fig. 5). Three early leptotene nuclei examined from the same individual also contained two X chromosomes (plate, fig. 9). All six cases must be a result of premeiotic non-disjunction. Indeed, though no mitoses were found in this individual, in one of the others about 50 mitotic anaphases were seen, in the majority of which, lagging separation of the X resulted in grossly elongated X bridges connecting daughter telophase groups. The large size of the X may at least play a part in this delayed separation. Such behaviour might be expected to lead, on occasion, to non-disjunction.

(*b*) In one other cell, two additional unidentifiable, small half-bivalents were apparently present at first anaphase, yielding a count of $2n = 32+X$ (text-fig. 1). Two possible explanations for this cell may be considered:

(1) The additional chromosomes represent a pair of B's, which had undergone normal bivalent formation and segregation (or been distributed to the two polar groups by chance). The complete absence of B's in all other cells of the five individuals makes this unlikely, however.

(2) The more likely alternative is that they arose by double mitotic autosomal non-disjunction, *i.e.* either non-disjunction of both homologues at the same premeiotic mitosis or non-disjunction of one or the other of a pair of homologues at two such successive mitoses, with subsequent bivalent formation and normal segregation (or else chance distribution).

Double mitotic non-disjunction has certainly been observed in the Acridid *Schistocerca gregaria* by John and Naylor (1961), where the largest pair of homologues has been found to be represented twice in one cell, with resulting quadrivalent formation.

Though non-disjunction may sometimes be a random process, there are two possible modes of non-random behaviour. On the one hand it may be *chromosome* specific, in which case one might expect the same chromosome to repeat the error, at least on occasion. Alternatively,

TEXT-FIG. 1.—First anaphase with one additional small half-bivalent at each pole. Sister chromatid bridge present, involving the M5. × 1750.

it may, by virtue of some aspect of structural or genetic organisation, be *homologue* specific. Under such conditions, comparable behaviour of both homologues would be more likely than not.

(iii) *Bridges at first anaphase*

Recently, first anaphase bridges have been described in the grasshopper *Chorthippus brunneus* (John, Lewis and Henderson, 1960). They

TEXT-FIG. 2.—Four sister chromatid bridges in the same cell at first anaphase, involving (from left to right) the L2, M5, M6 and M4 bivalents. × 1750.

involve localised association of sister chromatids, most typically subterminally. Bridges of this type have also been observed here in *Pholidoptera*. As in *Chorthippus*, the largest members of the complement are most commonly involved, and the association is most characteristically subterminal (text-figs. 1, 2; plate, figs. 6, 7 10). Though it was not possible to determine the frequency from the small number of anaphases observed, up to four bridges have been seen in cells at this

stage (text-fig. 2). The bridges observed here possess two diametrically opposed characters not observed in *Chorthippus*:

1. The arm region adjacent to the centromere tends to be more readily uncoiled than the remainder of the arm, for this region was often found to be greatly elongated, the non-associated chromatid pulling away from its associated sister and taking its sister centromere with it. This tended to be asymmetrical, one side stretching considerably more than the other (plate, fig. 6). One bridge was found in which breakage of this elongating region had occurred, with complete loss of the centric end and the non-associated chromatid (plate, fig. 7).
2. Conversely, the non-bridging chromatids may on occasion remain tightly apposed to their bridging sisters, particularly at early anaphase (*cf.* plate, fig. 10).

The exact nature of these sister chromatid bridges has still not been elucidated, but they are in no way confined to *Chorthippus brunneus* and *Pholidoptera*. They have been observed in the following additional animal species:

(i) Orthoptera.		
(*a*) Acrididæ	*Chorthippus albomarginatus*	Henderson (unpublished)
	Chorthippus parallelus	John and Henderson (unpublished)
	Omocestus viridulis	John and Lewis (unpublished)
	Schistocerca gregaria	John and Naylor (1961)
(*b*) Tettigoniidæ		
	Tettigonia viridissima	Henderson (unpublished)
	Conocephalus dorsalis	Henderson (unpublished)
(ii) Diptera.		
Tipulidæ	*Tipula brevispina*	Henderson (unpublished)
	Tipula irrorata	Henderson (unpublished)

Furthermore, there are numerous figures in the literature from many different plant and animal groups which almost certainly represent this kind of bridge. Table 1 includes a few taken at random by a brief survey of some of the literature to indicate the range of organisms in which they may be present. A more exhaustive survey would doubtless reveal many more.

From this information it is readily apparent that close investigation may well reveal these bridges to be considerably widespread throughout both plant and animal kingdoms. The frequency with which they occur would be expected to vary from one organism to the next, possibly being higher in *Chorthippus* spp. than in the majority of animals and plants.

4. SUMMARY

1. The chromosome number in the male of *Pholidoptera griseoaptera* (DeGeer) (Orthoptera : Tettigoniidæ) is $2n = 30+X$, the number possessed by all other Old World genera of the sub-family Decticinæ.

TABLE I

Author	Date	Organism	Figure	Reference
(i) *Plants*				
Afify, A.	1933	*Aconitum stærkianum*	26*a*	*J. Genet.*, *27*, 293-318
Åkerman, Å. and Hagberg, A.	1954	*Avena sativa*	36, 27, 38	*Hereditas*, *40*, 438-452
Darlington, C. D.	1939	*Fritillaria kamskatchensis*	3*b*, *c*, *e*	*J. Genet.*, *37*, 341-364
Dodds, K. S.	1943	*Musa* spp.	9, 10, 11, 12	*J. Genet.*, *45*, 113-138
Doughty, L. R.	1936	*Agave amaniensis*	4A	*J. Genet.*, *33*, 198-205
Howard, H. W.	1938	*Raphanus sativus* × *Brassica oleracea*	13, 33	*J. Genet.*, *36*, 239-273
Huskins, C. L.	1927	*Avena sativa*	35	*J. Genet.*, *18*, 315-364
Larter, L. N. H.	1932	*Ranunculus acris*	32	*J. Genet.*, *26*, 255-283
		Ranunculus repens	35	
		Ranunculus ficaria	36, 37	
Rees, H.	1955	*Secale cereale*	2*a*, *b*, *c*	*Heredity*, *9*, 93-116
Ribbands, C. R.	1937	*Lilium* × *testaceum*	52	*J. Genet.*, *35*, 1-24
Richardson, M. M.	1936	*Lilium Martagon album* × *Lilium Hansonii*	52	*J. Genet.*, *32*, 411-450
Upcott, M.	1937	*Tulipa gesneriana* (Inglescombe Yellow)	28	*J. Genet.*, *34*, 339-398
		Tulipa Orphanidea	34*a*	
		Tulipa gesneriana spathulata	50	
(ii) *Animals*				
Ahmed, I. A.	1941	*Canis familiaris*	18	*Proc. Roy. Soc. Ed. B*, *61*, 107-118
Belar, K.	1929	*Stenobothrus lineatus*	9, 10, 68, 69, 70	*Roux' Arch. Ent. Mech.*, *118*, 359-484
Carothers, E. E.	1913	*Dissosteria carolina*	59	*J. Morphol.*, *24*, 487-512
Crew, F. A. and Koller, P. C.	1938	*Sus scrofa*	10*a*, *b*	*Proc. Roy. Soc. Ed. B*, *57*, 194-214
Darlington, C. D.	1932	*Chorthippus elegans*	13	*Biol. Bull.*, *63*, 357-367
Koller, P. C.	1937	*Homo sapiens*	8*c*, *d*	*Proc. Roy. Soc. Ed. B*, *59*, 163-175

2. In seven cells from one individual, numerical deviants were found: (*a*) Six cells, three of which were at different meiotic stages, contained an extra X chromosome, a result of premeiotic non-disjunction. (*b*) One cell at first anaphase contained an additional small pair of half-bivalents, which probably resulted from non-disjunction of one of the small pairs of homologues.

3. First meiotic anaphase sister chromatid bridges of a type previously described in *Chorthippus brunneus* were found in many of the larger members of the complement. It is suggested that these bridges may well prove to be of widespread occurrence.

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5. REFERENCES

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Plate

FIG. 1.—Late diplotene : 15AA+X. ×1250.

FIG. 2.—First metaphase : 15AA+X. ×1250.

FIG. 3.—Second metaphase, X absent : 15A. ×1250.

FIG. 4.—Diakinesis : 15AA+X. ×1250.

FIG. 5.—First metaphase : 15AA+XX. ×1250.

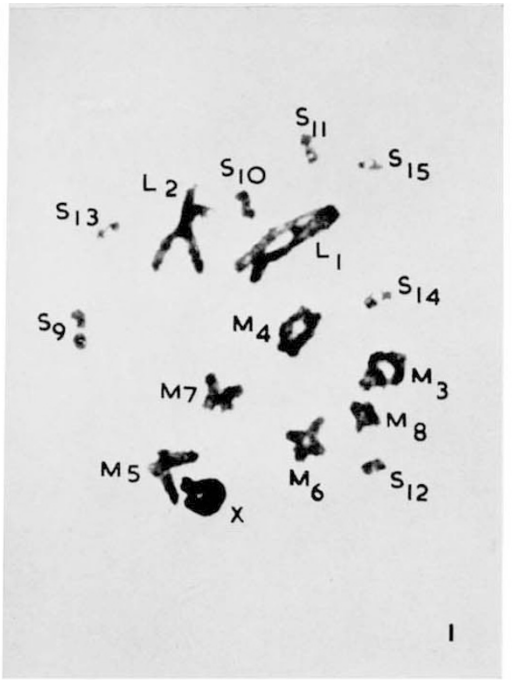
FIG. 6.—Asymmetrical elongation of the associated arms in a sister chromatid bridge at late first anaphase. ×1250.

FIG. 7.—First anaphase bridge in which upper sister chromatid and centromere of associated chromatid have been detached by breakage. ×2000.

FIG. 8.—Spiral structure of X chromosome from early diplotene nucleus. ×2000.

FIG. 9.—Early leptotene nucleus with two heteropycnotic X chromosomes. ×1250.

FIG. 10.—First anaphase with sister chromatid bridges involving the L1, L2 and M6 (one in M3 having recently broken). Close lateral association of the non-associated sister chromatids, with rotation, is exhibited by the L2. ×1250.



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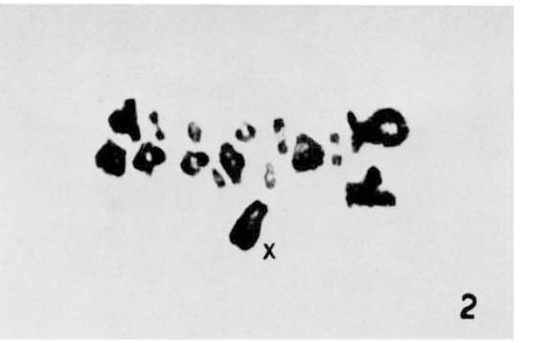

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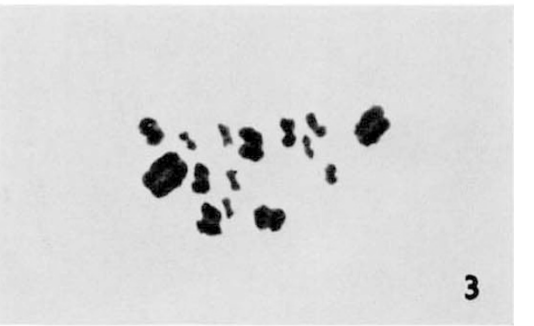

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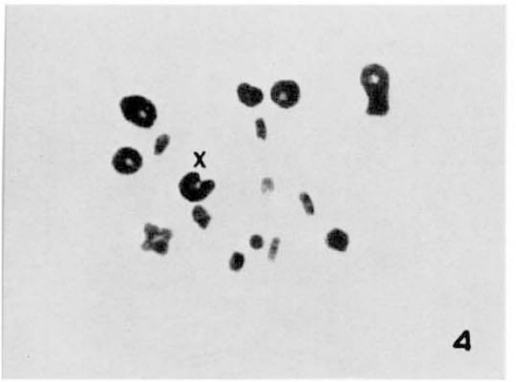

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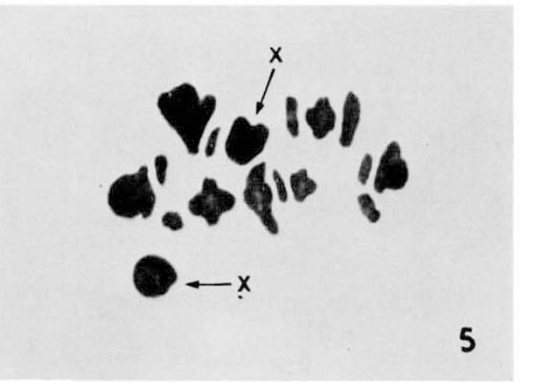

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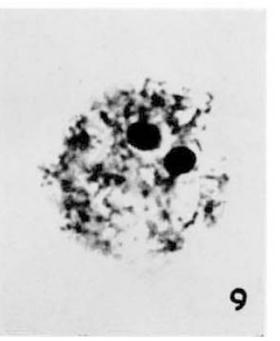

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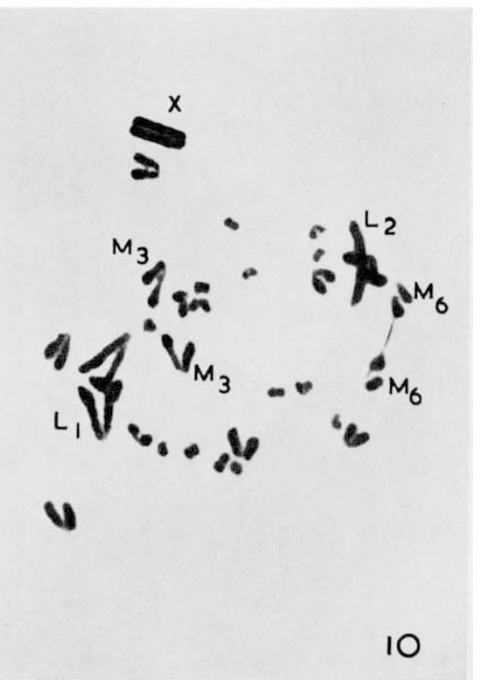

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