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From the Institute of Genetics, University of Helsinki, Finland

ON THE CHROMOSOMES OF THE GEOMETRID MOTH GENUS CIDARIA*

By

ESKO SUOMALAINEN

With 54 Figures in the Text

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I. Introduction

Although the chromosomal conditions of the *Lepidoptera* have been rather intensively investigated, most of the investigations have so far dealt with the chromosomes of butterflies, hawk-moths, and bombycids. The chromosomes of such large groups of the *Lepidoptera* as noctuids and geometrids have been very poorly studied, not to mention the so-called micro-Lepidoptera where the chromosome number has been determined in quite isolated cases only. It is also notable that although several problems concerning lepidopteran chromosomes have been extensively investigated, certain important ones lack a final answer so far.

In what follows, it is my aim to give an account of my investigations on the chromosomal conditions in the geometrid moth genus *Cidaria*. I have chosen this genus of *Lepidoptera* for cytological investigations because it is a taxonomically clear-cut and at the same time a genus rich in species. The species can be classified into distinct groups, according to which the genus *Cidaria* is nowadays often split into several different genera.

The study of the genus *Cidaria* is greatly handicapped by the circumstance that — in contrast to the butterflies for instance — meiosis has already taken place in adult males; spermatogenesis is already completed in the caterpillar. Therefore, although the work is very laborious I have mainly confined my studies to the oogenesis. There is a further reason for this, namely that, in comparison with spermatogenesis, oogenesis has been rather poorly studied in the *Lepidoptera*. An additional disadvantage has been that the chromosomes of most *Cidaria* species are, as lepidopteran chromosomes in general, small-sized and globular in form. This renders the study of the bivalent structure difficult.

In spite of the difficulties mentioned above, it seemed that a cytological analysis of as many *Cidaria* species as possible could be profitable in view of tracing the chromosomal evolution in the *Lepidoptera*, and of

* Dedicated to Professor H. BAUER on the occasion of his sixtieth birthday.

clarifying certain other problems connected with the chromosomal conditions in this group.

Some of the results have been published as a preliminary report (SUOMALAINEN, 1963).

II. Material and Methods

All my material has been collected in Finland. The main part of the material was collected in the years 1936—1963 on the southern coast of Finland in the vicinity of the town of Porvoo 40 km east of Helsinki. A smaller body of data is derived elsewhere from Finland, inclusive Lapland.

Of the females, ovaries with mature eggs were fixed in Carnoy's fluid (6:3:1) and sectioned at a thickness of 15 μ. The ovaries were embedded in paraffin via butyl alcohol. The preparations were stained in Heidenhain's iron-hematoxylin or by the Feulgen method (H or F in the legends of the figures). To a lesser extent, also Gomori's hematoxylin stain was employed (G in the legends).

Spermatogenesis was studied in two species only. The testes were taken from adult male caterpillars, fixed in Carnoy's fluid (6:3:1) and sectioned at a thickness of 10 μ. The preparations were stained in Heidenhain's iron-hematoxylin or with Feulgen.

The drawings were made at bench level with an Abbe camera lucida or a Leitz drawing ocular, using a $\times 100$ immersion objective and a $\times 25$ ocular. After reduction the final linear magnification of the text figures is approximately $\times 2800$. The photographs were taken on Agfa Agepe 35 mm film with the Leitz Orthomat camera and magnified to 2100 $\times$.

III. Chromosome numbers of the investigated *Cidaria* species

It is not always easy to obtain usable metaphase plates from the ovaries of *Cidaria*. It is illustrative that although about 450 ovaries were examined in sectioned preparations, only 125, or 25 to 30 per cent, contained plain metaphase plates. Nevertheless, the chromosome numbers of 44 species of *Cidaria* were determined exactly, and a fairly good estimate was obtained for 4 species. This amounts to slightly more than 60 per cent of the *Cidaria* species recorded from Finland. In some species, however, the determination of the chromosome number is based on relatively few metaphases.

Table 1 shows the chromosome numbers of the *Cidaria* species investigated, as well as the collecting localities and the numbers of specimens. The nomenclature corresponds, with minute exceptions, to that of SEITZ (1915).

In addition to the numbers listed in Table 1, I have been able to determine the approximate chromosome number of four species (the material of all of them has been collected in Porvoo):

Cidaria (*Xanthorhoë*) *montanata* SCHIFF. In three females, three not quite plain first metaphase plates with 28—30 bivalents.

C. (*Xanthorhoë*) *spadicearia* SCHIFF. In one first metaphase plate, about 29 bivalents.

C. (*Psychophora*) *caesiata*, F. In one first metaphase, about 31 bivalents.

Table 1. *The chromosome numbers of the Cidaria species investigated*

Species	Locality	Haploid number	Number of specimens investigated[1]	Plain metaphases[2]
Subgenus *Lyncometra* PRT				
ocellata L. (Fig. 1)	Porvoo	31	2	2
Subgenus *Plemyria* HB.	Porvoo, Tvärminne	29	3	3 (+4)
bicolorata HFN. (Fig. 2)	Porvoo	31—32	1	1 (+1)
Subgenus *Thera* STPH.				
variata SCHIFF. (Figs. 3, 44)	Porvoo	13	6	14[3]
obeliscata HB. (Figs. 4, 45)	Porvoo	13	5	7
cognata THNBG (Fig. 5)	Hiittinen	20	3	1 (+3)
juniperata L. (Fig. 6)	Porvoo, Heinola	30	5	13
firmata HB. (Fig. 7)	Porvoo	19	2	3
Subgenus *Chloroclysta* HB.				
miata L. (Fig. 8)	Heinola	30	1	2 (+2)
Subgenus *Dysstroma* HB.				
truncata HFN. (Fig. 9)	Porvoo	29	3	2 (+2)
latefasciata STGR (Fig. 10)	Porvoo	29	3	1 (+3)
citrata L. (Fig. 11)	Porvoo	29	6	5 (+5)
Subgenus *Xanthorhoë* HB.	Porvoo	31	4	8 (+2)
fluctuata L. (Fig. 12)	Pernaja			
quadrifasciata CL. (Fig. 13)	Porvoo	31	3	4 (+3)
Subgenus *Colostygia* HB.				
aptata HB. (Fig. 14)	Porvoo	30	2	1 (+1)
olivata SCHIFF. (Fig. 15)	Porvoo	31	1	1 (+1)
pectinataria KNOCH (Fig. 16)	Porvoo	31	2	2
didymata L. (Figs. 18, 46)	Porvoo	27	4	5
parallelolineata RETZ (Fig. 17)	Porvoo	30?	1	1 (+1)
Subgenus *Lampropteryx* STPH.				
suffumata SCHIFF. (Fig. 19)	Porvoo	32	1	1[3]
minna BTLR (Figs. 20, 47)	Porvoo	17	2	4 (+3)
Subgenus *Psychophora* KIRBY				
sabini frigidaria GN. (Fig. 21)	Enontekiö: Kilpisjärvi	29	2	3
Subgenus *Coenotephria* PRT				
sagittata F. (Fig. 22)	Porvoo	12	2	3 (+2)
Subgenus *Euphyia* HB.				
cucullata HFN. (Fig. 23)	Porvoo	31?	2	1 (+1)
luctuata SCHIFF. (Fig. 25)	Porvoo, Enontekiö	30	2	1 (+2)
corylata THNBG (Fig. 24)	Porvoo	30	2	2 (+3)

[1] Only specimens with plain metaphase plates included.

[2] The figure in parentheses refers to the number of metaphase plates which are not fully indisputable.

[3] The same chromosome number has been observed in many cells during meiotic divisions in the male.

Table 1 (continuation)

Species	Locality	Haploid number	Number of specimens investigated[1]	Plain metaphases[2]
Subgenus *Camptogramma* STPH.				
bilineata L. (Fig. 26) . . .	Porvoo	30	2	4
Subgenus *Mesoleuca* HB.				
albicillata L. (Fig. 27) . . .	Porvoo	31	3	3
Subgenus *Eulype* HB.				
hastata L.	Porvoo	31	3	1 (+2)
subhastata NOLCK. (Figs. 28—29)	Porvoo	30	1	2
	Enontekiö: Kilpisjärvi	31	1	1
Subgenus *Epirrhoë* HB.				
tristata L. (Fig. 30)	Porvoo, Maarianhamina	30	2	3
alternata MÜLL. (Figs. 31, 48)	Porvoo	31	3	4 (+2)
	Pernaja			
galiata SCHIFF. (Fig. 32) . .	Hiittinen	31	2	2
Subgenus *Perizoma* HB.				
taeniata STPH. (Fig. 33) . . .	Porvoo	32	2	4 (+1)
affinitata STPH. (Fig. 36) . .	Porvoo	25	1	1
alchemillata L. (Fig. 34) . .	Porvoo	30	5	9 (+1)
hydrata TR. (Fig. 35)	Porvoo	25	2	2
blandiata SCHIFF. (Fig. 37) .	Porvoo	30	2	2
	Hiittinen			
flavofasciata THNBG (Fig. 38)	Porvoo	30	2	2 (+1)
Subgenus *Hydriomena* HB.				
furcata THNBG (Fig. 39) . .	Porvoo	28	3	3 (+1)
coerulata F. (Fig. 40) . . .	Porvoo	30	4	7 (+1)
ruberata FRR	Porvoo	30	1	1
Subgenus *Pelurga* HB.				
comitata L. (Figs. 41, 49) . .	Porvoo	32	4	11 (+3)
Subgenus *Hydrelia* HB.				
testaceata DON. (Fig. 42) . .	Porvoo	13	1	8
flammeolaria HFN. (Fig. 43) .	Porvoo	30	2	2 (+1)

C. (*Euphyia*) *unangulata* HW. In one first metaphase plate, about 28 bivalents.

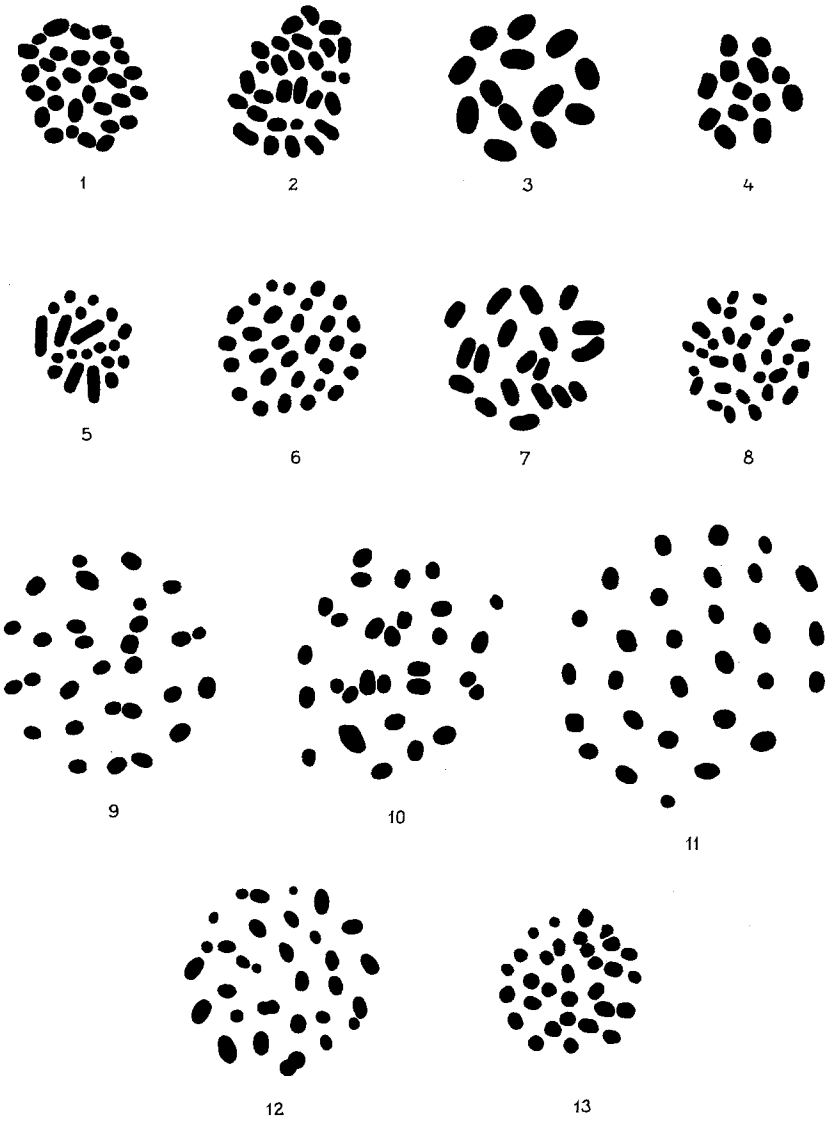
BELIAJEFF (1930) has found the same chromosome number as mentioned above in *Cidaria* (*Hydriomena*) *coerulata* (= *autumnalis*). I have myself earlier (SUOMALAINEN, 1953) reported the chromosome numbers of four species of the subgenus *Thera*.

IV. Discussion

1. Chromosome numbers in the genus Cidaria

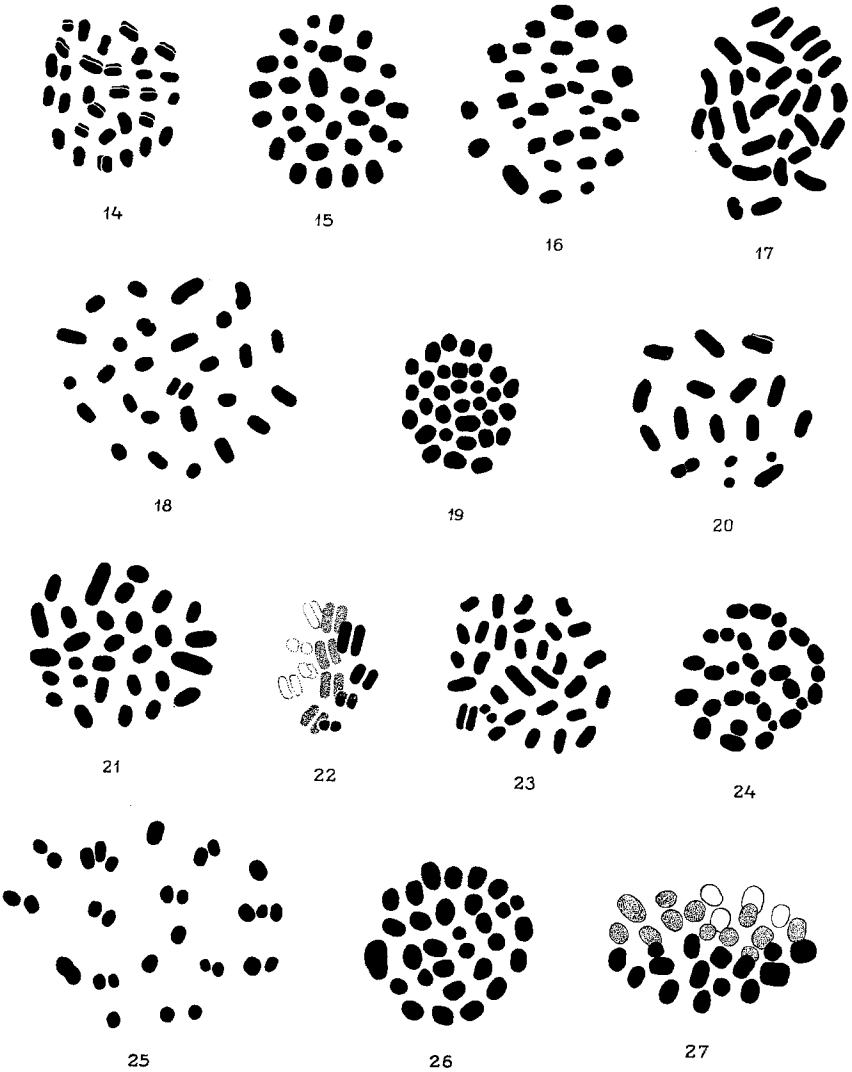
The most common haploid chromosome number in the genus *Cidaria* is 30; fifteen species have this number (Fig. 50). The chromosome number of 33 species — 75 per cent of the species investigated — falls

between 29 and 32. This agrees well with our earlier knowledge of the chromosome numbers of the *Lepidoptera* in general. According to WHITE (1954, p. 177) 30 is the number second in commonness among the *Lepidoptera* as a whole — the most common number is 31 — and the chromosome number of most species falls between 28 and 32.



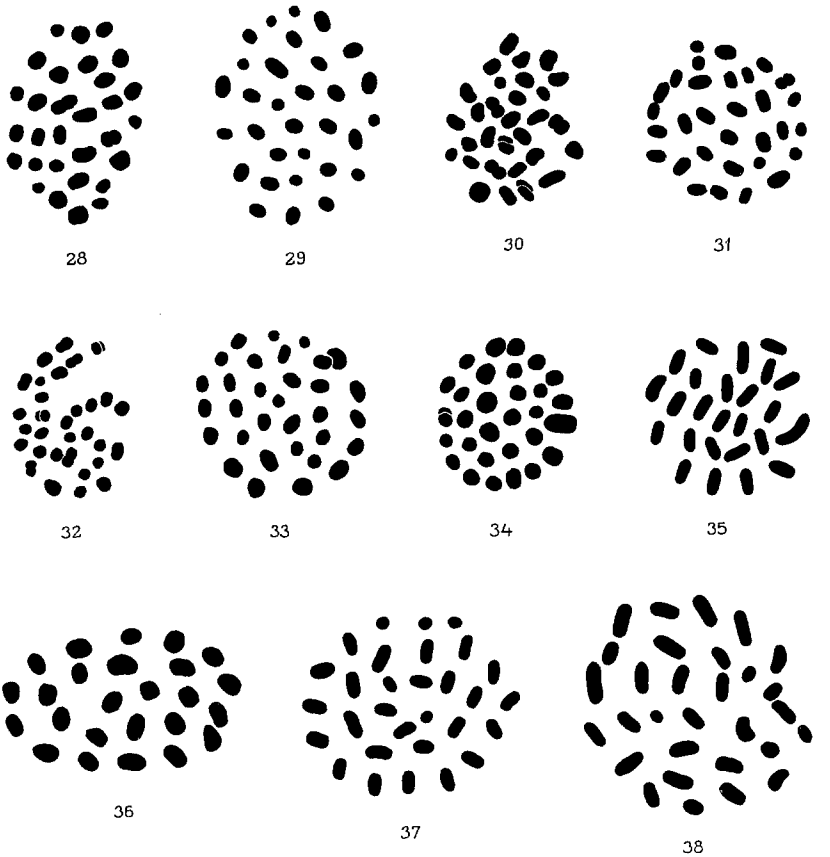
Figs. 1—13. First meiotic division metaphase plates in oocytes in 13 *Cidaria* species. 1 *ocellata*. 2 *bicolorata*. 3 *variata*. 4 *obeliscata*. 5 *cognata*. 6 *juniperata*. 7 *firmata*. 8 *miata*. 9 *truncata*. 10 *latefasciata*. 11 *citrata*. 12 *fluctuata*. 13 *quadrifasciata*. — 1—3, 6—7 and 10—12 H, 4—5, 8 and 13 F, 9 G

However, deviations from the modal numbers just mentioned, are of the greatest interest. From Fig. 50 we see that my material contains several chromosome numbers which are subnormal, namely 28, 27, 25, 20, 19, 17, 13 and 12. The number 13 has been detected in as many as three species. On the other hand, chromosome numbers greater



Figs. 14—27. First meiotic division metaphase plates in oocytes in 14 *Cidaria* species. 14 *aptata*. 15 *olivata*. 16 *pectinataria*. 17 *parallelolineata*. 18 *didymata*. In the centre of the plate, both component chromosomes of one bivalent are seen separately. 19 *suffumata*. 20 *minna*. 21 *sabini frigidaria*. 22 *sagittata*. Prometaphase. 23 *cucullata*. 24 *corylata*. 25 *luctuata*. 26 *bilineata*. 27 *albicillata*. — 16, 21 and 24—27 H, 14—15, 17—18, 20 and 23 F, 19 and 22 G

than normal were wholly absent in my material. In this respect my *Cidaria* material differs, for instance, from the butterflies investigated, in which LORKOVIĆ (1941) and DE LESSE (1952, 1953, 1954, 1960, 1961)

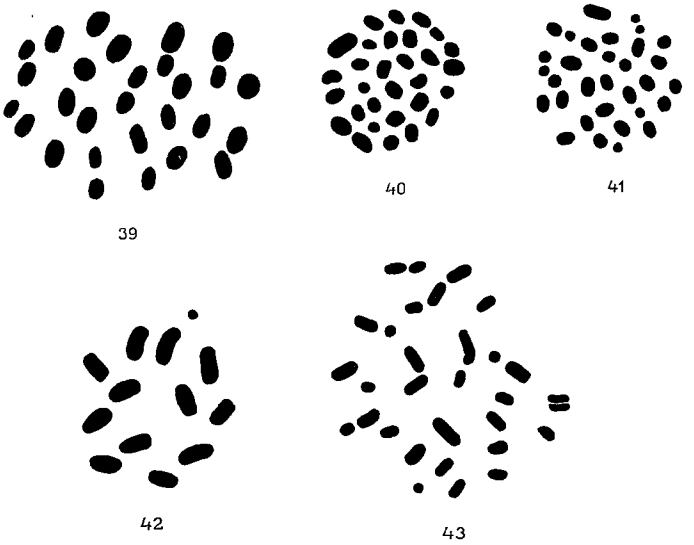


Figs. 28—38. First meiotic division metaphase plates in oocytes in 10 *Cidaria* species. 28 *subhastata* from Porvoo; 30 bivalents. 29 *subhastata* from Enontekiö, Kilpisjärvi; 31 bivalents. 30 *tristata*. 31 *alternata*. 32 *galiata*. 33 *taeniata*. 34 *alchemillata*. 35 *hydrata*. 36 *affinitata*. 37 *blandiata*. 38 *flavofasciata*. — 29—30, 33, 36 and 38 H, 28, 31—32, 34—35 and 37 F

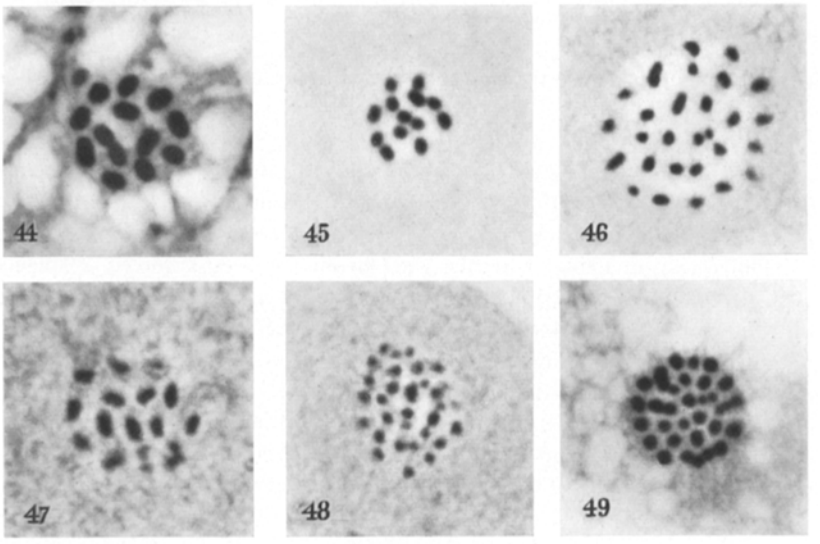
have reported some very high chromosome numbers. However, the lack of high numbers in my material may well be ascribable to chance alone.

Such species groups or subgenera are of interest where closely related species have wholly different chromosome numbers. There are three such subgenera in my material. It is notable that these abnormal chromosome numbers are not confined to certain subgenera in such a way that all species of a subgenus have the deviating number. These exceptions occur usually as isolated cases among normal numbers.

In Figs. 3—7 we see the first metaphase of the female of five species belonging to the subgenus *Thera*. Two species, namely *variata* and *obeliscata*, have only 13 bivalents. One species, *firmata*, has 19, one,



Figs. 39—43. First meiotic division metaphase plates in oocytes in 5 *Cidaria* species. 39 *furcata*. 40 *coerulata*. 41 *comitata*. 42 *testaceata*. 43 *flammeolaria*. — 39—40 and 43 H, 42 F, 41 G



Figs. 44—49. First meiotic division metaphase plates in oocytes in 6 *Cidaria* species. 44 *variata* (the same cell as in Fig. 3). 45 *obeliscata* (the same cell as in Fig. 4). 46 *didymata* (the same cell as in Fig. 18). 47 *minna* (the same cell as in Fig. 20). 48 *alternata*. 49 *comitata*. — 44 and 49 H, 45—48 F

cognata, 20, and the fifth one, *juniperata*, has the most common number among *Cidaria*, namely 30. All these five species resemble each other very much morphologically. It also appears from the figures that in those species with a small number of chromosomes, the chromosomes are markedly bigger than in species with a high chromosome number. This is typical of all species of *Cidaria*, and has already been observed in other *Lepidoptera*.

Figs. 19—20 show the chromosome sets of the species pair *minna* — *suffumata*, belonging to the subgenus *Lampropteryx*. These species

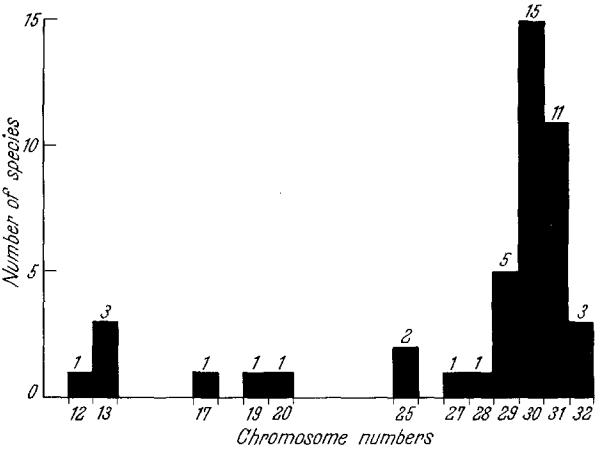


Fig. 50. Haploid chromosome numbers of the *Cidaria* species investigated

resemble each other so much that the former was long considered to be a variety of the latter. However, *minna* has only 17 chromosomes, of which three are much smaller than the others, whereas *suffumata* has 32 chromosomes. Figs. 42—43 show the chromosomes of two species of the subgenus *Hydrelia*, *testaceata* has 13 chromosomes — one of them is much smaller than the others — and *flammeolaria* 30.

The question arises whether the chromosome number of a species is subject to local variation. My material is not appreciably enlightening on this point, since it has been collected mainly on a single locality, namely Porvoo in South Finland. What arouses attention, however, is that the two *subhastata* individuals, derived from localities widely separated geographically, have different chromosome numbers. The specimen collected from Porvoo, at the southern coast of Finland, has the haploid number 30 (Fig. 28), whereas the other one, collected from Kilpisjärvi, in northern Lapland, has 31 (Fig. 29). The difference may be spurious, however, since only one specimen represents each of the two local populations. It is not wholly beyond possibilities that the

subhastata populations of North and South Finland would differ in chromosome number, as is the case in the populations of e.g. *Phragmatobia fuliginosa* in different parts of Central Europe (SEILER, 1925).

2. Size differences within a chromosome set

It is characteristic of the chromosome sets of most *Cidaria* species studied that, within a species, the chromosomes are approximately equal in size, a circumstance typical of the *Lepidoptera* in general (see e.g. BELIAJEFF, 1930, p. 377; FEDERLEY, 1938, p. 460; LORKOVIĆ, 1941, p. 166). In about 80 per cent of the *Cidaria* species investigated the chromosomes within each species are either of equal size (e.g. Figs. 3 and 13), or, if differences exist, all intergrades occur (e.g. Figs. 12 and 43).

Species having chromosomes widely differing in size are relatively rare in my material. Four species, *bilineata* (Fig. 26), *alchemillata* (Fig. 34), *coerulata* (Fig. 40) and *comitata* (Figs. 41, 49), possess a bivalent which is clearly bigger than the others; this is typical of many butterflies as well (DE LESSE, 1960); *sabini frigidaria* (Fig. 21) shows two such bivalents in the first metaphase; of the 13 bivalents of *testaceata* (Fig. 42) one is much smaller than the others, while three such ones are found among the 17 bivalents of *minna* (Figs. 20, 47); *cognata* (Fig. 5) possesses the most peculiar chromosome set in this respect, as the chromosomes fall into two distinct size groups: five of the chromosomes are very big and 15 small.

Within the scope of this study, it has not been possible to decide whether any of the different-sized chromosomes are sex chromosomes. BAUER (1943) and WHITE (1946, 1954, 1957a), in fact, have supposed that in such *Lepidoptera* species at least which have a very high chromosome number, the frequently existing big chromosome is a sex chromosome.

The facts presented above indicate that the chromosomal evolution in *Cidaria* and in the *Lepidoptera* in general usually favours chromosome sets with equal-sized chromosomes.

3. The chromosomal evolution in the Lepidoptera

Most authors discussing the phylogeny of lepidopteran chromosome complements (e.g. BELIAJEFF, 1930; FEDERLEY, 1938; LORKOVIĆ, 1941; WHITE, 1954, 1957a) agree that the most common haploid chromosome numbers of the *Lepidoptera* (29—31) are phylogenetically original, and that other chromosome numbers have probably been derived from them. As the lepidopteran chromosome numbers vary widely around the numbers 29—31 — the lowest number known being 8 (*Erebia tyndarus*; LORKOVIĆ, 1949) and the highest one 191 (*Lysandra nivescens*; DE LESSE,

1954, 1960) — both decrease and increase in chromosome number have taken place among the *Lepidoptera*; the former change has been more usual, however. Various hypotheses have been put forward to explain the origin of often so widely differing chromosome numbers among the *Lepidoptera*.

LORKOVIĆ (1941, 1949) has suggested that deviating chromosome numbers of butterflies, which in some cases are really multiples of each other, are of polyploid origin. This view has been strongly opposed by WHITE (1946; 1954, p. 208; 1957a). I have also (SUOMALAINEN, 1953, 1959) been rather definitely of the opinion that polyploidy must be out of the question. My reason for this is, among others, the fact that although the chromosome numbers are multiples of each other in some cases, many numbers between the multiples are also known. This also excludes the possibility that differences in the degree of polyteny would be involved, such as those detected by SCHRADER and HUGHES-SCHRADER (SCHRADER and HUGHES-SCHRADER, 1956; HUGHES-SCHRADER and SCHRADER, 1956; HUGHES-SCHRADER, 1957) in certain *Hemiptera*. Also FEDERLEY (1938, 1943, 1945) and DE LESSE (1960) consider polyploidy improbable among *Lepidoptera*.

Table 2
Average DNA-contents in four Cidaria species

Species	Haploid number	DNA-content
Subgenus *Thera* .		
obeliscata	13	6.70
juniperata	30	6.13
Subgenus *Lampropteryx*		
minna	17	5.94
suffumata	32	7.45

In order to settle the problem, photometric measurements of the DNA-content of the chromosome sets of *Cidaria* species were performed. Table 2 illustrates the results of these measurements. Again, closely related species with markedly different chromosome numbers have been chosen as objects of study. The measurements have been made of the first meiotic metaphase of the female by means of two-wavelength photometry. Each value represents a mean of three to five metaphase measurements. A greater number of metaphase plates stained approximately simultaneously was not available.

We see that in the species pair *obeliscata* — *juniperata*, where the former has 13 and the latter 30 chromosomes, the means of DNA-content are 6.70 and 6.13, respectively. In the other species pair studied, *minna* (n = 17) — *suffumata* (n = 32) the DNA-contents of the chromosome sets are 5.94 and 7.45. The results show that the DNA-content of closely related species is almost equal, in spite of great differences in chromosome numbers. This is also true of the subgenera investigated. In any case, the numbers are not multiples of each other. Admittedly, in both species pairs investigated one has a "normal" chromosome

number and the other a much lower number, but it is presumable that also species with supernormal chromosome number will give a similar result. The results of the DNA-measurements confirm the view that in *Cidaria* species and other *Lepidoptera*, a large chromosome of a species with a low chromosome number corresponds to two or even more chromosomes of one with a high number.

The findings presented above confirm the opinion expressed already earlier by several authors (BELIAJEFF, FEDERLEY, WHITE, SUOMALAINEN, op.c.), that the "abnormal" lepidopteran chromosome numbers are due to "fragmentations" and "fusions". Owing to the small size and spherical form of lepidopteran chromosomes, it is difficult to make observations of the underlying cytological details. This question is, however, closely connected with the problem of the character of the kinetochore of lepidopteran chromosomes.

As late as 1945, WHITE (p. 177) remarked that we do not know anything about the kinetochore in the *Lepidoptera*. FEDERLEY (1943, 1945), however, basing his opinion on several arguments, came to the conclusion that in the *Lepidoptera* a diffuse kinetochore is to be found. Also Professor HANS BAUER suggested, in a lecture in Berlin-Dahlem in 1941, that the *Lepidoptera* possess holokinetic chromosomes. I have myself (SUOMALAINEN, 1953) come, on many grounds, to the conclusion that "it is more than probable that the *Lepidoptera* in fact have a non-localized, or diffuse, kinetochore" (p. 90). (For details see SUOMALAINEN, op.c.). Also VIRKKI (1963, p. 118) expresses the opinion that his observations on the moth *Diatraea saccharalis* "favor a diffuse centromere". Although the holokinetic nature of the lepidopteran chromosomes so far lacks experimental proof, many authors regard it certain or at least probable (see e.g. WHITE, 1957a, p. 126; DARLINGTON, 1958, p. 92; SMITH, 1960, p. 70).

As is well-known the diffuse kinetochore makes it possible for chromosome fragments which have resulted from splitting of the chromosome to divide and function as autonomous wholes. Conversely, if chromosomes unite with other chromosomes there is no autonomous behaviour of separate kinetochores within one chromosome to create disturbances during the division of the new chromosome. As already mentioned, the details of the process by which the "fragmentations" and "fusions" in the *Lepidoptera* may take place are unknown. It is obvious that most "fusions" are so-called centric fusions. A diffuse kinetochore, one is willing to assume, could render their formation easier, in the sense that the breaks need not, at least theoretically, be so strictly localized as in chromosomes with a localized kinetochore. WHITE (1957a, p. 126—128) has presented a more detailed theoretical discussion of the chromosomal evolution in the *Lepidoptera*. He con-

cludes that, for instance, "in the ancestry of *L.* (*Lysandra*) *nivescens* (species with n = 191) we must assume that at least 166 structural rearrangements, each involving a gain of 2 telomeres, have occurred... Each of these rearrangements was most likely a translocation whereby a "donor" chromosome provided the 2 required telomeres together with some adjacent material". In another genus of butterflies, *Erebia*, where both increase and decrease of chromosome number have occurred "the species *E. ottomana*, with n = 40, must have acquired at least 10 "fragmentations"; while the very closely related *E. tyndarus* with n = 8, has presumably accumulated about 20 fusions."

When we examined the chromosome numbers of *Cidaria* species, we found that 75 per cent of the species studied had the number typical of the genus and of the *Lepidoptera* in general, and only 25 per cent had a deviating number. This applies to other lepidopteran groups as well. Consequently, it is obvious that the chromosome numbers of the *Lepidoptera* are less variable than the diffuse kinetochore would, at least theoretically, allow. The same holds true with respect to the *Hemiptera* and *Homoptera*, which also have a diffuse kinetochore. Among them, either, deviations from the modal number are not very common (HALKKA, 1957, 1959, 1960; SUOMALAINEN and HALKKA, 1963).

If we examine these conditions in the plant genus *Luzula*, which also possesses a diffuse kinetochore, the situation is clearly different. The cytology and chromosomal evolution of this genus has been studied in detail by, for example, NORDENSKIÖLD (1951, 1956, 1961, 1962, in the press). In the genus *Luzula*, the chromosome number is clearly more variable and frequently exhibits the so-called agmatoploidy or "endonuclear polyploidy". That differences in the degree of polyteny are not involved in an agmatoploid series is shown by the fact that in the hybrids, one large chromosome conjugates with several small ones. This clearly indicates fragmentation. However, photometric studies on a number of *Luzula* species (MELLO-SAMPAYO, 1961; HALKKA, in the press) have shown that within this genus besides agmatoploidy both true polyploidy and differential polyteny are encountered. It is obvious that the animals with a diffuse kinetochore possess some kind of system rendering changes in chromosome numbers more difficult than in the plant genus *Luzula*. I and Dr. HALKKA (SUOMALAINEN and HALKKA, 1963, p. 508) have put forward the suggestion "that in animals the presence in both sex chromosomes and autosomes of genes influencing sex determination interferes with transverse fragmentation of chromosomes. Such sex-determining genes never, of course, occur in the monoecious *Luzula*. In animals the mechanism of sex determination may often be so nicely balanced that any systemic fragmentations might upset

it to a degree fatal to the organism". This view is supported by the assumption by BAUER and WHITE, that the large, unfragmented chromosome often found in the butterflies, is a sex chromosome (see p. 175).

4. Bivalent structure in lepidopteran females

Genetic experiments performed with *Lepidoptera* have revealed that crossing over takes place in the males but not in the females. This has been observed both in the silkworm *Bombyx mori* and the wax moth *Galleria mellonella* (see e.g. WHITE, 1945, p. 193, and TAZIMA, 1964, p. 47).

MAEDA (1939) has observed distinct chiasmata in the males of the silkworm, but is of the opinion that chiasmata are not formed in the females. Although FEDERLEY (1943, p. 242) mentioned having now and then found distinct cross and ring bivalents in species with large chromosomes, such as *Dicranura vinula*, he was still in 1943 of the opinion that "Chiasmata kommen überhaupt nicht in der Oogenese der Lepidopteren vor, und in den kugeligen Chromosomen der Spermatogenese ist dies noch weniger der Fall" (p. 245). However, a little later (1945) he was able to find distinct chiasmata in the spermatogenesis of *Trichiura crataegi*, a moth species with large chromosomes, and to analyse them. In the spermatogenesis, both rod and ring bivalents are formed. I have been able to observe a similar formation of bivalents, and distinct chiasmata, in the spermatogenesis of the *Cidaria* (For details see SUOMALAINEN, 1953). Also DEODIKAR and THAKAR (1958) as well as SRIVASTAVA and GUPTA (1962) have found chiasmata in the spermatogenesis of the Indian silkworm, *Philosamia ricini*.

We already discussed the observation of MAEDA (op.c.) that no chiasmata are formed in the lepidopteran oogenesis. FEDERLEY (1943) was of the same opinion, but after having observed distinct chiasmata in the spermatogenesis of *Trichiura* (1945) he assumed that they occur also in the oogenesis of *Lepidoptera*. BAUER (1939, p. 587) concluded from the photographs published by FEDERLEY (1938) of the oogenesis of certain butterflies — mainly of *Erebia medusa polaris* — that "diese Bilder FEDERLEYs zeigen... klar, daß Chiasmata in der Oogenese nicht gebildet werden". This opinion that the chiasmata are absent in the lepidopteran oogenesis he has expressed also later (BAUER, 1953, p. 505). WHITE (1945, p. 193; 1954, p. 230), on the contrary, interpreted the oogenesis pictures by MAEDA, mentioned above, so that "a single chiasma occurs in each bivalent, near one end. This chiasma may be completely terminalized, so that the two chromosomes which form the bivalent are merely in end-to-end contact. No such strict localization occurs during spermatogenesis, so that the difference in genetical behaviour between the sexes is explained by MAEDA'S work.

although not in the way that he has claimed." — That entirely contrary opinions concerning the formation of chiasmata in the oogenesis of the *Lepidoptera* have been expressed depends at least in part on the small size of the chromosomes and their spherical form which make it difficult to observe the details in the bivalents.

More than ten years ago (SUOMALAINEN, 1953) my attention was drawn to the observation that the bivalents in the spermatogenesis of the *Lepidoptera* appear very different from those in the oogenesis. This is especially conspicuous in species having fewer chromosomes,

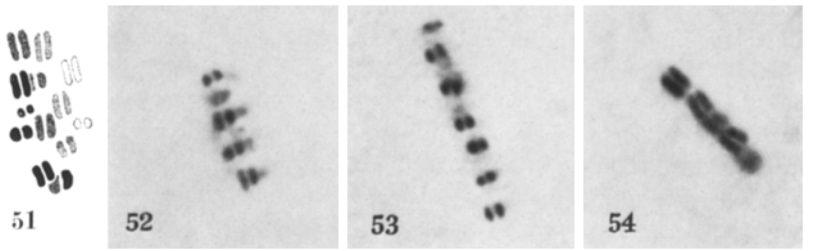


Fig. 51. First meiotic division metaphase plate in the side view in the female of *Cidaria* (*Lampropteryx*) *minna*. Part of the bivalents omitted. — F. 52

Figs. 52—54. First meiotic division metaphase plates in the side view in oocytes in 3 *Cidaria* species. 52 *minna*. 53 *subhastata*. 54 *testaceata*. — 52—54 F

which then are large. In polar view, the form of the bivalents at the first metaphase of the oogenesis is more elongated than in the spermatogenesis. In my oogenesis preparations, the bivalents showed structures which I had earlier (op.c.) considered to be chiasmata. Consequently, the difference of the bivalents in the spermatogenesis and the oogenesis seemed to be best explained by assuming that the bivalents have different modes of orientation in the two sexes; in this case co-orientation in the male and auto-orientation in the female. In view of this, absence of crossing over in lepidopteran females would be explained by the chiasmata being localized at the ends, as also WHITE has supposed (see above).

All earlier preparations of cidarian oogenesis were stained in Heidenhain's iron-hematoxylin. It appeared later that this stain does not give an adequate picture of the structure of the bivalents in lepidopteran females, since it stains also the elimination chromatin. Having studied the bivalents in the oogenesis of *Cidaria* species in more detail and also in well-stained Feulgen preparations, I have come to the conclusion that no chiasmata are formed in the oogenesis of the *Lepidoptera* as already earlier suggested by MAEDA and BAUER (see above). In Feulgen preparations, the elimination chromatin characteristic of lepidopteran females is not stained at all (cf. RIS and KLEINFELD, 1952), and chromo-

somes are visible as parallel threads without any sign of chiasma (Figs. 51—54). This was well observable in very many *Cidaria* species investigated.

A closer study of the structures which I earlier interpreted as chiasmata, in the diakinesis of several females, revealed that they are not always situated midst in the bivalent. The situation should, of course, be so if one terminal chiasma were present. This also indicates that chiasmata are excluded.

Such an achiasmatic meiosis is known in some other insect groups, namely in certain mantids and roaches, in *Panorpa*, and in several dipterans; it is also likely among certain scorpions (BAUER, 1953, p. 505; WHITE, 1954, p. 222 ff.; 1957b, p. 78; 1961, p. 100—101; ULLERICH, 1961). Recently, achiasmatic meiosis has been detected also in the copepod *Tigriopus* (AR-RUSHDI, 1963). It is clear that the absence of chiasmata in one sex has rather far-reaching genetic consequences, since it reduces recombination to a half. In the *Lepidoptera*, the exceptionally high chromosome number (the modal haploid number 29—31) is a means for compensating for this, and is perhaps partly explained just by this condition.

Summary

1. The oogenesis of 44 (48) species of the Geometrid moth genus *Cidaria* has been studied. Spermatogenesis has been studied in two species only.

2. The most common (haploid) number among the *Cidaria* species so far investigated is 30, which is likewise the number second in commonness among the *Lepidoptera* as a whole. Most (33, or 75 percent) of the species have a chromosome number between 29 and 32, like most of the other groups of *Lepidoptera*.

3. No species has more than 32 chromosomes, whereas eleven have less than 29. The smaller chromosome numbers found are 28, 27, 25, 20, 19, 17, 13 and 12.

4. The great differences in chromosome numbers between closely related species are of interest. Such discrepancies are found in the subgenera *Thera* (*variata* and *obeliscata* 13, *firmata* 19, *cognata* 20 and *juniperata* 30), *Lampropteryx* (*minna* 17 and *suffumata* 32), and *Hydrelia* (*testaceata* 13 and *flammeolaria* 30).

5. The chromosomes are clearly bigger in the species with a low chromosome number than in those with a high one. In the chromosome sets of most *Cidaria* species studied the chromosomes are approximately equal in size.

6. Photometric measurements revealed that the DNA-content of closely related species is almost equal, in spite of great differences in

chromosome numbers. This is also true of the subgenera investigated. This indicates that one chromosome in a species with a low number of chromosomes corresponds to two or more chromosomes of another one with a high chromosome number.

7. The discrepancies in the chromosome numbers among *Lepidoptera* have not arisen from polyploidy or differences in the degree of polyteny. They are due to "fragmentations" or "fusions", which are rendered easier by the diffuse kinetochore.

8. It is obvious that in animals with a diffuse kinetochore some mechanism, possibly the location of sex-determining genes, reduces the role of chromosomal rearrangements in chromosomal evolution from what the diffuse kinetochore otherwise would allow.

9. Contrary to earlier assumptions of the author, chiasmata are not formed in the bivalents during oogenesis in the *Lepidoptera*. This is especially evident in preparations stained with Feulgen, when the elimination chromatin contained by the bivalents in the female remains unstained.

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Prof. Dr. ESKO SUOMALAINEN
Institute of Genetics, University of Helsinki
P. Rautatiekatu **13**, Helsinki (Finland)