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STUDIES OF THE CHROMOSOMES OF FORMOSAN RHOPALOCERA
2. Pieridae, Lycaenidae and Riodinidae

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The first of this series of papers dealt with 8 species of Papilionidae and 5 species of Hesperiidae from Formosa, giving a description of the source of the material and the research methods (Maeki & Ae, 1968). The present paper describes the chromosomes observed in the male germ cells of 15 species belonging to Pieridae, Lycaenidae and Riodinidae of Formosa, and the related species are phylogenetically discussed in the light of the cytological data obtained.

In addition to the species described in this paper, we studied the chromosomes of the following species: *Gonepteryx mahaguru taiwana* Paravicini, *Deudorix eryx horiella* (Matsumura), *Syntarucus plinius* (Fabricius), *Jamides celeno celeno* (Cramer), *Nacaduba kurava asakusa* Fruhstorfer, *Zizeeria maha okinawana* (Matsumura), *Celastrina puspa myla* (Fruhstorfer), and *Megisba malaya volubilis* Fruhstorfer, but failed to obtain satisfactory findings because the materials were inadequate for chromosome research.

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Observations

A. PIERIDAE

1. *Leptosia nina niobe* (Wallace)

The haploid chromosome number is 19. Counts were made in each 10 nuclei (I, II) in testes of 3 males collected at Kuan-tzu-ling, T'ai-nan Hsien on April 7 (F-83, F-101) and at Ch'i-hsin-liao, Chia-i Hsien on April 15 (F-172). There are apparently 2 big, 4 first large, 5 second large, 7 third large, and 1 small chromosomes.

2. *Cepora coronis cibyra* (Fruhstorfer)

The haploid chromosome numbers are 23, 24 and 25. Following nuclei were counted in the first meiosis in the testes of 3 males (F-30, F-47, F-64) collected at K'en-ting-kung-yuan, P'ing-tung Hsien on April 2, 3 and 4.

First metaphase

n, 23	17 nuclei
n, 24	2 nuclei
n, 25	3 nuclei
Total	22 nuclei

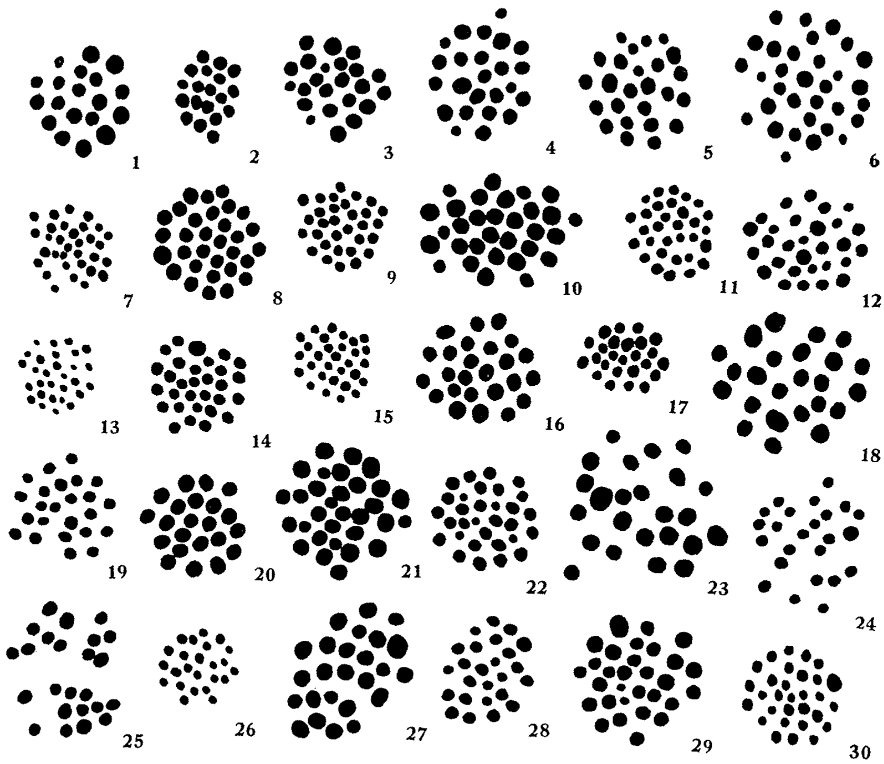
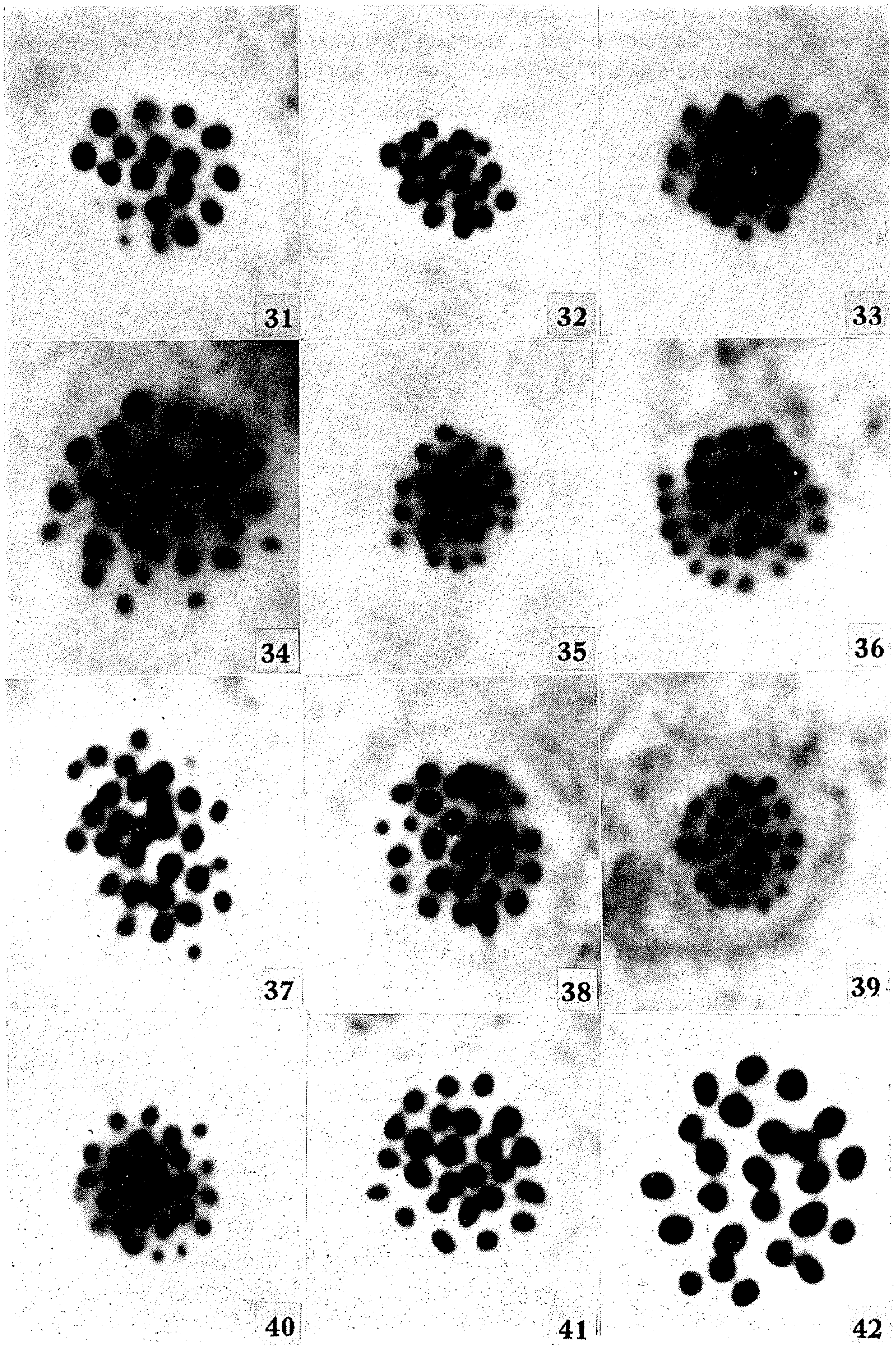


Fig. 1: *Leptosia nina niobe* (I). Fig. 2: same (II). Fig. 3: *Cepora coronis cibyra* *n*, 23(I). Fig. 4: same, *n*, 24 (I). Fig. 5: same, *n*, 25 (I). Fig. 6: *Appias indra aristoxenus* (I). Fig. 7: same (II). Fig. 8: *Catopsilia pomona pomona* (I). Fig. 9: same (II). Fig. 10: *Gonepteryx amintha formosana* (I). Fig. 11: same (II). Fig. 12: *Eurema andersoni godana* (I). Fig. 13: same (II). Fig. 14: *Eurema esakii* (I). Fig. 15: same (II). Fig. 16: *Eurema blanda arsakia* (I). Fig. 17: same (II). Fig. 18: *Rapala varuna formosana* (I). Fig. 19: same (II). Fig. 20: *Horaga onyx moltrechti* (I). Fig. 21: *Spindasis syama* (I). Fig. 22: same (II). Fig. 23: *Jamides bochus formosanus* (I). Fig. 24: same (II). Fig. 25: *Jamides alecto dromicus* (I). Fig. 26: same (II). Fig. 27: *Celastrina limbata himilcon* (I). Fig. 28: same (II). Fig. 29: *Abisara burnii etymander* (I). Fig. 30: same (II).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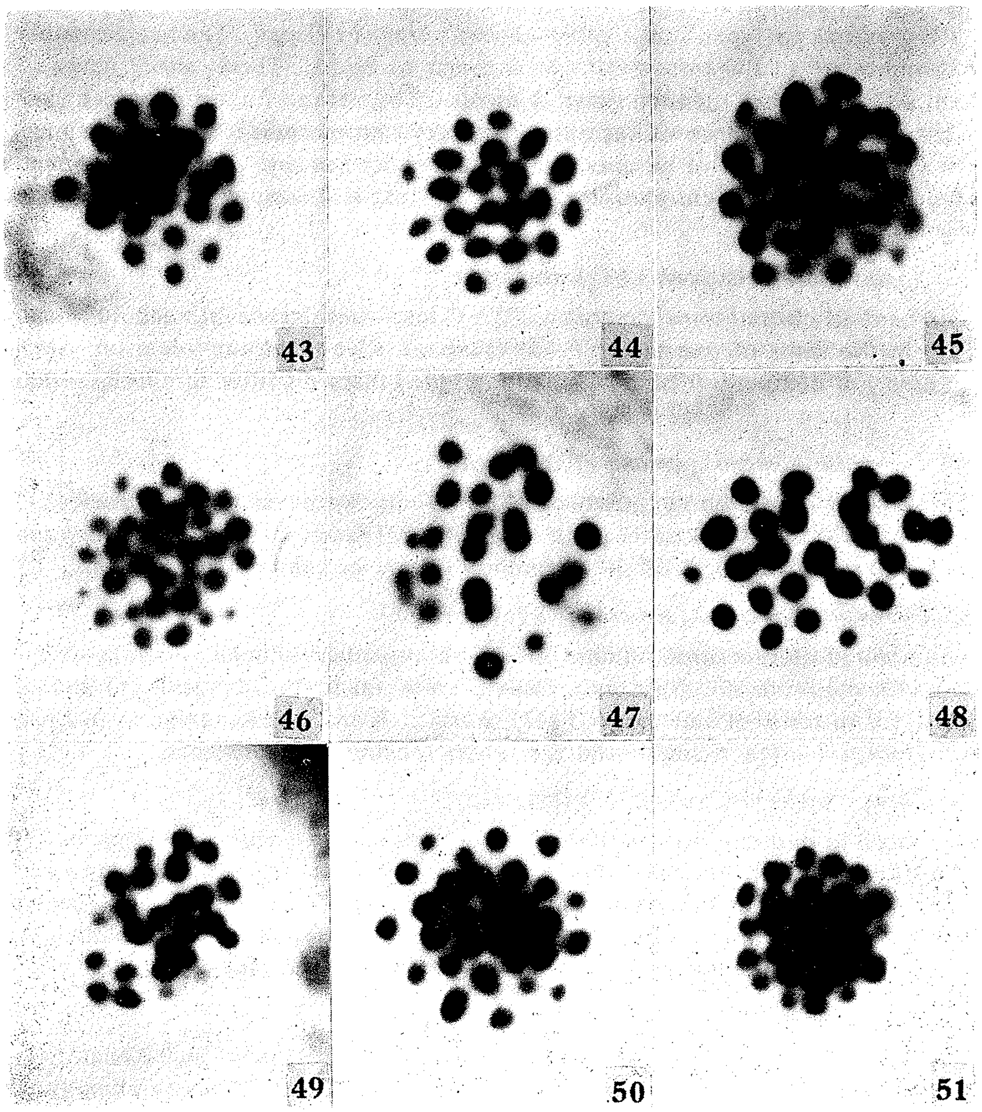

Photographs of chromosomes in spermatocyte divisions.

Fig. 31: *Leptosia nina niobe* (I). Fig. 32: same (II). Fig. 33: *Cepora coronis cibyra* (I). Fig. 34: *Appias indra aristoxenus* (I). Fig. 35: same (II). Fig. 36: *Catopsilia pomona pomona* (I). Fig. 37: *Gonepteryx amintha formosana* (I). Fig. 38: *Eurema andersoni godana* (I). Fig. 39: same (II). Fig. 40: *Eurema esakii* (I). Fig. 41: *Eurema blanda arsakia* (I). Fig. 42: *Rapala varuna formosana* (I). Fig. 43: same (II). Fig. 44: *Horaga onyx moltrechti* (I). Fig. 45: *Spindasis syama* (I). Fig. 46: same (II). Fig. 47: *Jamides bochus formosanus* (I). Fig. 48: *Celastrina limbata himilcon* (I). Fig. 49: same (II). Fig. 50: *Abisara burnii etymander* (I). Fig. 51: same (II).

This is a remarkable situation and quite resemble the previously known cases of *Pieris melete* in Japan and *Cepora coronis phryne* in Nepal. The basic number of chromosome in *Cepora coronis cibyra* seems to be 23. There are 3 large, 3 small, and the others medium-sized elements. The cause of the numerical variation lies in the presence of the supernumerary chromosome. The supernumeraries vary from 1 to 2 in number and are characterized by a minute element. The nuclei having 23 chromosomes are most frequent, being 77% in 22 nuclei observed.

3. *Appias indra aristoxenus* Fruhstorfer

The haploid chromosome number is 32. Counts were made in each 10 nuclei (I, II) in the testis of one male (F-46) taken at K'en-ting-kung-yuan on April 3. There are distinctly 9 large, 4 small, and the others medium in chromosomal size.

4. *Catopsilia pomona pomona* (Fabricius)

The haploid chromosome number is 31. Counts were made in 10 nuclei (I) and 10 nuclei (II) in the testis of one male (F-41) taken at K'en-ting-kung-yuan on April 3. Apparently 2 of the chromosomes are larger than the others.

5. *Gonepteryx amintha formosana* (Fruhstorfer)

The haploid chromosome number is 31. Many suitable nuclei were found in first and second meiotic divisions. Counts were made in 15 nuclei (I) and 10 nuclei (II) in testis of one male (F-42) taken at K'en-ting-kuang-yuan on April 3. There are 4 large, 6 small, and the others medium chromosomes.

6. *Eurema andersoni godana* (Fruhstorfer)

The haploid chromosome number is 29. Counts were made in 12 nuclei (I) and 5 nuclei (II) in the testis of one male (F-28) taken at K'en-ting-kung-yuan on April 2. The specimen showed many dividing cells in both first and second meiosis. Six chromosomes are distinctly larger than the others and seven chromosomes are much smaller than the several medium-sized elements.

7. *Eurema esakii* Shirôzu

The haploid chromosome number is 30. Counts were made in 10 nuclei (I) and 6 nuclei (II) in the testis of one male (F-26) taken at K'en-ting-kung-yuan on April 2. One chromosome is apparently big in size and 15 chromosomes are larger than the other elements.

8. *Eurema blanda arsakia* (Fruhstorfer)

The haploid chromosome number is 25. Counts were made in 10 nuclei (I) and 5 nuclei (II) in the testes of 2 males (F-51, F-52) taken at K'en-ting-kung-yuan on April 3. Eight chromosomes are distinctly larger than the others.

B. LYCAENIDAE

1. *Rapala varuna formosana* Fruhstorfer

The haploid chromosome number is 24. Counts were made in 18 nuclei (I) and 15 nuclei (II) in the testes of 2 males taken at Kuan-tzu-ling on April 7 (F-80) and at Chi-hsin-liao on April 14 (F-151). Three chromosomes are distinctly larger than the others and 16 chromosomes were sowewhat smaller than the several medium-sized elements.

2. *Horaga onyx moltrechti* Matsumura

The haploid chromosome number is 22. Counts were made in 15 nuclei (I) in the testes of 2 males (F-82, F-89) taken at Kuan-tzu-ling on April 7. No meiotic divisions were found in secondary spermatocytes. Apparently eight chromosomes are large in chromosomal size.

3. *Spindasis syama* (Horsfield)

The haploid chromosome number is 31. Counts were made in 10 nuclei (I) and 5 nuclei (II) in the testes of 3 males (F-69, F-77, F-85) taken at Kuan-tzu-ling on April 6 and 7. Apparently five chromosomes are much smaller than the other large elements.

4. *Jamides bochus formosanus* Fruhstorfer

The haploid chromosome number is 24. Counts were made in 11 nuclei (I) and 6 nuclei (II) in the testes from 3 males (F-145, F-146, F-147) taken at Chu-ch'i on April 13. There are big 1, large 6, and the others somewhat small in chromosomal size.

5. *Jamides alecto dromicus* (Fruhstorfer)

The haploid chromosome number is 24. Counts were made in 10 nuclei (I) and 5 nuclei (II) in the testes of 2 males taken at Kuan-tzu-ling on April 7 (F-99) and at Ch'i-hsin-liao on April 15 (F-174). The karyotype not include a big chromosome which observed in *J. bochus formosanus*. The chromosomes of *J. alecto dromicus* are quite similar to that of *J. bochus* except huge element, there are 6 large and the other all small in chromosomal features.

6. *Celastrina limbata himilcon* (Fruhstorfer)

The haploid chromosome number is 25. Counts were made in 13 nuclei (I) and 7 nuclei (II) in the testes of 2 males (F-93, F-119) taken at Kuan-tzu-ling on April 7 (F-93), and at Fen-ch'i-hu, Chia-i Hsien on April 10 (F-119). There are apparently 1 big, 18 large, and 6 small chromosomes.

C. RIODINIDAE

1. *Abisara burnii etymander* (Fruhstorfer)

The haploid chromosome number is 30. Counts were made in 16 nuclei (I) and 10 nuclei (II) in the testes from 3 males (F-122, F-123, F-136) taken at Fen-ch'i-hu on April 11 and 12. In all of the 30-chromosome nuclei, there are 1 huge, 6 very large, 14 moderately large, 7 medium, and 2 small elements.

Discussion

Chromosomes of eight out of the twenty-three species of Pieridae, Lycaenidae and Riodinidae listed in Table 2 have already been studied by Maeki, Ogata and Shirôzu (1965) in the previous survey of chromosomes in Formosan butterflies. The remaining fifteen species are given in roman antiques in the Table, which have been cytologically studied for the first time.

Generally, species belonging to Pieridae have *n*, 25–26 or *n*, 31 as the haploid set. *Leptosia nina niobe* (*n*, 19) from Formosa is unique in the number and sizes of its chromosomes. Its 19 chromosomes consist of 2 largest, 4 second largest, 5 third largest, 7 medium and 1 small chromosomes. This seems to suggest the chromosomal fusion of ancestral elements because larger elements are present. *Leptosia nina niobe* with 19 chromosomes is ranked as a third exceptional spe-

Table 2*. *Chromosome numbers of Formosan Pieridae, Lycaenidae, and Riodinidae.*

Species	Number (*n*)	Phase	Reference
A. Pieridae			
Leptosia nina niobe	19	♂(I, II)	Present paper
Delias aglaia curasena	25	♂(I, II)	Maeki, Ogata, Shirôzu, 1965
Pieris canidia canidia	25	♂(I, II)	〃
Cepora nadina eunama	25	♂(I)	〃
Cepora coronis cibyra	23, 24, 25	♂(I)	Present paper
Appias lyncida formosana	32	♂(I, II)	Maeki, Ogata, Shirôzu, 1965
Appias indra aristoxenus	32	♂(I, II)	Present paper
Ixias pyrene insignis	28	♂(I, II)	Maeki, Ogata, Shirôzu, 1965
Hebomoia glaucippe formosana	17	♂(I, II)	〃
Catopsilia crocale crocale	31	♂(I, II)	〃
Catopsilia pomona pomona	31	♂(I, II)	Present paper
Gonepteryx amintha formosana	31	♂(I, II)	〃
Eurema andersoni godana	29	♂(I, II)	〃
Eurema esakii	30	♂(I, II)	〃
Eurema blanda arsakia	25	♂(I, II)	〃
B. Lycaenidae			
Rapala varuna formosana	24	♂(I, II)	〃
Horaga onyx moltrechti	22	♂(I)	〃
Spindasis syama	31	♂(I, II)	〃
Heliophorus epicles matsumurae	25	♂(I, II)	Maeki, Ogata, Shirôzu, 1965
Jamides bochus formosanus	24	♂(I, II)	Present paper
Jamides alecto dromicus	24	♂(I, II)	〃
Celastrina limbata himilcon	25	♂(I, II)	〃
C. Riodinidae			
Abisara burnii etymander	30	♂(I, II)	〃

* Table 1, for the Papilionidae and Hesperiidae, is in Part 1 of this series of papers (Kontyû, 36 : 120).

cies in Pieridae, the first being *Pieris brassicae* (*n*, 15), and second, *Hebomoia glaucippe* (*n*, 17), in having smaller number of chromosomes.

Among of the 6 *Eurema* species studied previously, only *Eurema laeta* has *n*, 30 chromosomes, other 5 species, namely *E. hecabe*, *E. proterpia*, *E. mexicana*, *E. nicippe* and *E. lisa* have the constant number of *n*, 31. However, the three species of Formosan *Eurema* reported in the present paper have each a different number *n*, 25 in *E. blanda*, *n*, 29 in *E. andersoni*, and *n*, 30 in *E. esakii*. The karyotype of *E. esakii* (*n*, 30) has one huge chromosome, and it may account for the 1+1 fusion of two elements in the ancestral karyotype (*n*, 31). Regarding the other two species, *E. andersoni* has 6 large elements and *E. blanda* has 8 large elements in their karyotypes. Therefore, it seems difficult to determine the cause of the numerical variation from the ancestral karyotype in these two species, unlike in *E. esakii*, from the standpoint of chromosomal fusion. The chromosome number in these species of Formosan *Eurema*, especially that of *E. blanda*, is much different from a general concept of the *Eurema* species reported so far.

The present survey has revealed that one or two supernumerary chromosomes were found to occur in the nuclei of the Formosan *Cepora coronis cibyra* (*n*, 23, 24, 25). On the other hand, a similar situation exists in the Himalayan *Cepora coronis phryne* (*n*, 22, 23, 24) studied by the present authors (1966). The nuclei of the Himalayan form contain the same number of supernumerary chromosomes as are present in those of the Formosan form. But there is a definite difference in the basic numbers between the two subspecies. The number is consistently *n*, 22 in *C. c. phryne* of Nepal and *n*, 23 in *C. c. cibyra* of Formosa. However, their karyotypes are remarkably similar to each other. In view of the numerical variation of the chromosomes in the two subspecies of *Cepora coronis*, it may be concluded that the species shows an unstable status of the karyotype due to geographic isolation. The Himalayan *C. c. phryne* is probably a more differentiated form than the Formosan *C. c. cibyra* from the cytological viewpoint, because it is generally considered that the number of chromosomes in butterflies tend toward reduction and supernumerary chromosomes arise from chromosomal fragmentation.

Chromosomes of *Appias* species (*A. drusilla*, *A. lyncida*, and *A. indra*) are constantly *n*, 32, which is an uncommon number in Rhopalocera. While the above three *Appias* species completely agree in the number of chromosomes, their patterns of chromosome sizes are different. Maeki and Remington (1960) reported an identical size for all chromosomes in *A. drusilla* from North America, Maeki, Ogata and Shirôzu (1965) pointed out 2 small elements in *A. lyncida* from Formosa, while *A. indra* dealt with in this paper is remarkable in having 9 large and 4 small elements.

In the Lycaenidae the characteristic number of chromosome is generally *n*, 24. There are many interesting individual variants from the basic type, especially in the genera *Agrodiaetus* (*n*, 10, 13, 15, 21, 23, 28, 41, 45, 78, 82, 84, 88, 90, 123, 131, 147, 190), *Satyrium* (*n*, 41), *Taraka* (*n*, 15), and *Ussuriana* (*n*, 47). Among the numerous species of Lycaenidae so far studied cytologically, *n*, 31 chromosomes have only been observed in *Spindasis syama*, which was examined in this study. The chromosome number of *Jamides bochus* (*n*, 24) of Formosa is the same as that of *J. alecto* (*n*, 24) of Formosa, but differs from the latter in the sizes of chromosomes. In *J. bochus*, the chromosomes are all larger than those of *J. alecto*. Butterflies belonging to the genus *Celastrina* probably have a characteristic number of *n*, 25, as already confirmed in *C. argiolus* of Europe and Japan (Federley '38, Lorković '41, Maeki and Makino '53) and *C. puspa* of Hong Kong (Maeki and Ae '68), and the newly examined *C. limbata* also has 25 chromosomes in the haploid set. The karyotype of 25 chromosomes in *C. puspa* include 9 large and 5 small elements, while *C. limbata* reported in this paper has 25 chromosomes, consisting of 1 huge, 18 large and 6 small elements.

On the other hand, the karyotype of *Abisara burnii etymander* which was reported in this paper was composed of chromosomes of various sizes, that is, 1 largest, 6 second largest, 14 third largest, 8 medium and 2 small chromosomes. Up to date, 3 species belonging to Riodinidae have been reported by previous workers. *Calephelis virginiensis* of North America has *n*, 45 and 9 chromosomes are distinctly larger than the others (Maeki and Remington, 1961a). *Abisara echerius* and *Zemeros flegyas* of Hong Kong both have *n*, 31, in which 24 chromosomes are larger than the remaining 7 chromosomes (Maeki and Ae, 1968). The 4 species thus far studied are respectively from North America, Hong Kong, and Formosa. Since the karyotypes are so different in these species of Riodinidae, it may offer an indication for the complicated character of this family.

Summary

1. In this report, the number and relative sizes of chromosomes are described regarding 8 species of Pieridae, 6 species of Lycaenidae and 1 species of Riodinidae, all from Formosa.

2. All species here have furnished new material for cytology. It is noteworthy that *Leptosia nina* has *n*, 19, *Eurema blanda n*, 25, *Spindasis syama n*, 31, and *Abisara burnii*, Riodinidae, *n*, 30.

3. The karyotype having supernumerary chromosomes of *Cepora coronis cibyra* (*n*, 23, 24, 25) from Formosa differs from that of the Himalayan *C. c. phryne* (*n*, 22, 23, 24).

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