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STUDIES OF THE CHROMOSOMES OF FORMOSAN RHOPALOCERA

3. Nymphalidae and Libytheidae

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The first and second in this series of papers reported on the results of chromosomal investigation of 28 species of Papilionidae, Hesperiidae, Pieridae, Lycaenidae and Riodinidae in Formosan butterflies which were collected by Japanese entomologists in a trip during the period from March to April, 1965, as part of Japan-U. S. Co-operative Science Programs. The present report deals with the chromosomes observed in the male germ cells of 11 species of Nymphalidae and Libytheidae as the third of the serial papers on chromosomal survey of Formosan Rhopalocera.

In addition to the species described in this paper, we studied the chromosomes of the following two species: *Calinaga buddha formosana* Fruhstorfer and *Acraea issoria formosana* Fruhstorfer, but failed to obtain satisfactory findings because the materials were inadequate for chromosome research.

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Observations

A. NYMPHALIDAE

1. *Yoma sabina vasuki* Doherty

The haploid chromosome number is 31. Counts were made in 15 nuclei in the primary spermatocyte division and 8 nuclei in the secondary spermatocyte division from the testes of 4 males taken at the K'en-ting Park, P'ing-tung Province on April 3 and 4 (F-44, F-45, F-53) and at Kuan-tzu-ling, T'ai-nan Province on April 7 (F-94), 1965. There are 10 large, 4 small and 17 medium-sized chromosomes. Many dividing cells were observed in both primary and secondary spermatocytes.

2. *Kaniska canace drilon* (Fruh.)

The haploid chromosome number is 31. Counts were made in 16 nuclei (I) in the testes of one male (F-137) taken at Fen-ch'i-hu, Chia-i Province on April 12. There are two first largest, 14 second largest chromosomes and the others are small. No meiotic divisions were found in secondary spermatocytes.

3. *Symbrenthia hippoclus formosanus* Fruhstorfer

The haploid chromosome number is 31. Counts were made in 8 nuclei (I) and 5 nuclei (II) in the testes of one male (F-126) taken at Fen-ch'i-hu, Chia-i Province on April 11. Thirteen chromosomes are somewhat larger than the others.

4. *Neptis soma lutatia* Fruhstorfer

The haploid chromosome number is 29. Counts were made in 6 nuclei (I) in the testes of a single male (F-23) taken at K'en-ting Park, P'ing-tung Province on April 2. Seventeen chromosomes were distinctly larger than the others, and 4 chromosomes much smaller than the remaining medium elements.

5. *Neptis ananta taiwana* Fruhstorfer

The haploid chromosome numbers are 31, 32 and 33. Following nuclei were counted in the first meiosis in the testes of one male (F-162) taken at Ch'i-hsin-liao, Chia-i Province on April 15.

First metaphase	
n, 31	4 nuclei
n, 32	3 nuclei
n, 33	3 nuclei
Total	10 nuclei

The nuclei having *n*, 31 consist of 13 large, 16 medium and 2 small chromosomes. The *n*, 32 nuclei have an additional small element, and *n*, 33 nuclei have two small elements. Occasionally one or two small elements are found out of the equatorial plates at the metaphase.

6. *Pantoporia hordonia maligowa* (Fruh.)

The haploid chromosome number is 30. Counts were made in 6 nuclei (I) in the testes of a single male (F-148) taken at Ch'i-hsin-liao, Chia-i Province on April 14. There are apparently 17 large, 6 small chromosomes, the others being medium in size.

7. *Tacoraea perius perius* (Linnaeus)

The haploid chromosome number is 30. Counts were made in 5 nuclei (I, II) each in the testes of 3 males taken at K'en-ting Park, P'ing-tung Province (F-35, F-38) on April 3 and at Kuan-tzu-ling, T'ai-nan Province on April 7. In all of the 30-chromosome nuclei there are 9 large, 17 medium and 4 small elements.

8. *Tacoraea selenophora laela* (Fruh.)

The haploid chromosome number is 30. Counts were made in 6 nuclei (I) and 8 nuclei (II) in the testes of one male (F-106) taken at Kuan-tzu-ling, T'ai-nan Province on April 7. All chromosomes are similar in size.

9. *Tacoraea cama zoroastes* (Butler)

The haploid chromosome number is 30. Counts were made in 8 nuclei (I) and 5 nuclei (II) in the testes of a single male (F-105) taken at Kuan-tzu-ling, T'ai-nan Province on April 7. Apparently four chromosomes are much smaller than the other elements.

10. *Timelaea maculata formosana* Fruhstorfer

The haploid chromosome number is 30. Counts were made in 15 nuclei (I) and 10 nuclei (II) in the testes of two males taken at K'en-ting Park, P'ing-tung Province on April 3, and at Kuan-tzu-ling, T'ai-nan Province on April 7. One chromosome is apparently outsize, 11 chromosomes are larger, and 5 chromosomes are smaller than the rest.

B. LIBYTHEIDAE

1. *Libythea celtis formosana* Fruhstorfer

The haploid chromosome number is 31. Counts were made in 10 nuclei (I) and 5 nuclei (II) in the testes of one male (F-84) taken at Kuan-tzu-ling, T'ai-nan Province on April 7. There are 12 first largest, 14 second largest and 5 somewhat small elements.

Discussion

Table 3 shows the chromosome numbers of 19 species of Formosan Nymphalidae and Libytheidae for which counts have been recorded. Chromosomes of eight out of the nineteen species listed in Table 3 have already been studied by Maeki, Ogata and Shirôzu (1965) in the previous survey of chromosomes of Formosan butterflies. The remaining eleven species are given in roman antiques in the Table, which have been cytologically studied for the first time.

The 5 *Neptis* species studied previously, namely *N. hylas, N. philyra, N. pryeri, N. columella* and *N. aceris*, have the number of *n*. 30 in common. However, the two species of Formosan *Neptis* reported in the present paper have a different number : *n*, 29 in *N. soma* and *n*. 31, 32, 33 in *N. ananta*. The karyotype of *N. soma* (*n*, 29) is unique in the number and sizes of its chromosomes. Its 29 chromosomes consist of 17 large, 8 medium and 4 small chromosomes, while *N. ananta* has a karyotype which vary from *n*, 31 to *n*, 33 at first meiosis. The *n*, 31 nuclei consist of 13 large, 16 medium and 2 small chromosomes and the nuclei of the *n*, 32 and *n*, 33 contain one or two supernumerary chromosomes besides the elements of *n*, 31. Namely, *N. ananta* has a basic number of *n*. 31,

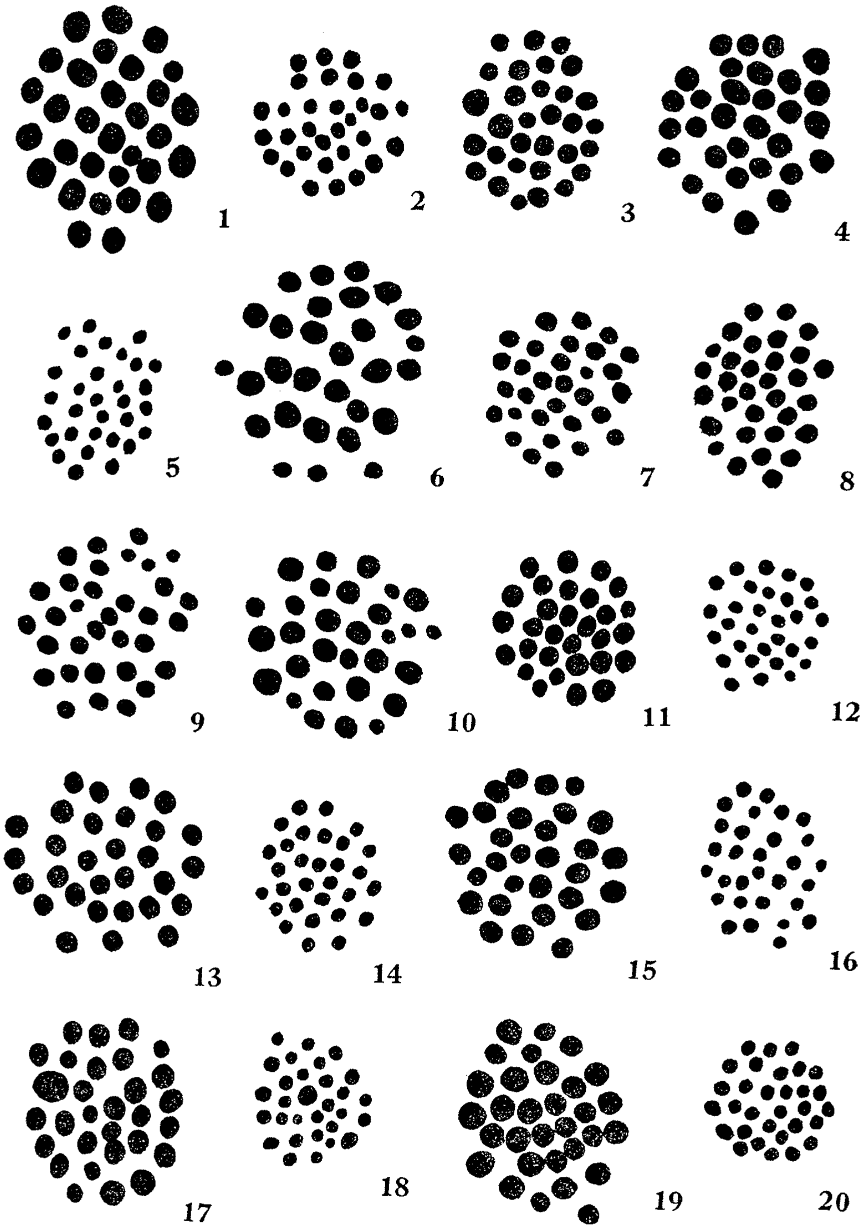

Fig. 1: *Yoma sabina vasuki* (I). Fig. 2: same (II). Fig. 3: *Kaniska canace drilon* (I). Fig. 4: *Symbrenthia hippoclus formosanus* (I). Fig. 5: same (II). Fig. 6: *Neptis soma lutatia* (I). Fig. 7: *Neptis ananta taiwana* *n*, 31 (I). Fig. 8: same *n*, 32 (I). Fig. 9: same *n*, 33 (I). Fig. 10: *Pantoporia hordonia maligowa* (I). Fig. 11: *Tacoraea perius perius* (I). Fig. 12: same (II). Fig. 13: *Tacoraea selenophora laela* (I). Fig. 14: same (II). Fig. 15: *Tacoraea cama zoroastes* (I). Fig. 16: same (II). Fig. 17: *Timelaea maculata formosana* (I). Fig. 18: same (II). Fig. 19: *Libythea celtis formosana* (I). Fig. 20: same (II).

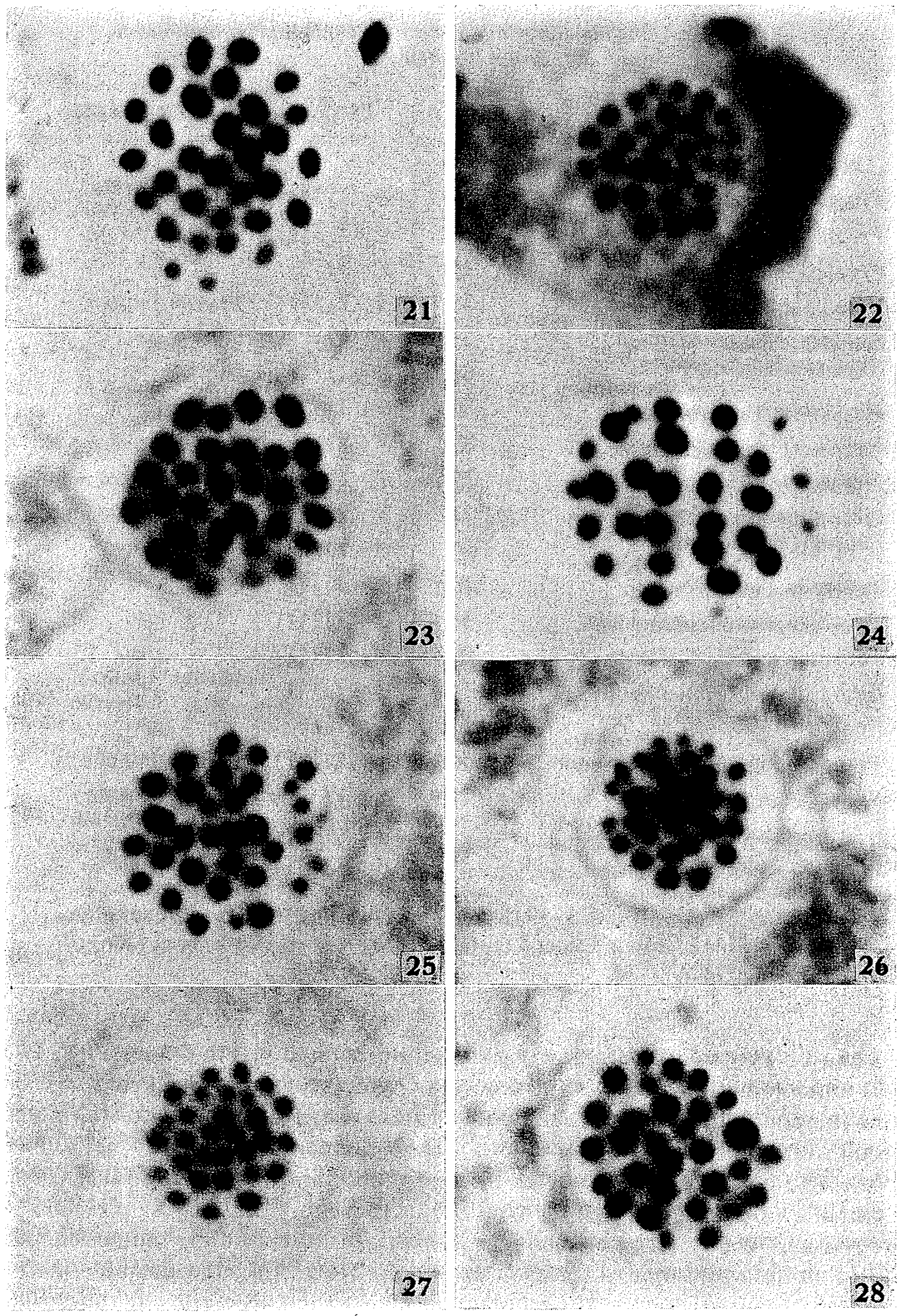

Fig. 21: *Yoma sabina vasuki* (I). Fig. 22: same (II). Fig. 23: *Symbrenthia hippoclus formosanus* (I). Fig. 24: *Neptis soma lutatia* (I). Fig. 25: *Neptis ananta taiwana* (I, n=33). Fig. 26: *Tacoraea perius perius* (I). Fig. 27: *Tacoraea selenophora laela* (II). Fig. 28: *Timelaea maculata formosana* (I).

Table 3.* *Chromosome numbers of Formosan Nymphalidae, and Libytheidae.*

Species	Number (n)	Phase	Reference
A. Nymphalidae			
Precis almana almana	31	♂ (I, II)	Maeki, Ogata, Shirôzu, 1965
Precis orithya orithya	31	♂ (I, II)	〃
Precis iphita iphita	31	♂ (I)	〃
Yoma sabina vasuki	31	♂ (I, II)	Present paper
Kaniska canace drilon	31	♂ (I)	〃
Symbrenthia hippoclus formosanus	31	♂ (I, II)	〃
Hypolimnas bolina kezia	31	♂ (I, II)	Maeki, Ogata, Shirôzu, 1965
Neptis hylas luculenta	30	♂ (I)	〃
Neptis soma lutatia	29	♂ (I)	Present paper
Neptis ananta taiwana	31, 32, 33	♂ (I)	〃
Pantoporia hordonia maligowa	30	♂ (I)	〃
Tacoraea perius perius	30	♂ (I, II)	〃
Tacoraea selenophora laela	30	♂ (I, II)	〃
Tacoraea cama zoroastes	30	♂ (I, II)	〃
Euthalia thibetana insulae	14	♂ (I, II)	Maeki, Ogata, Shirôzu, 1965
Cyrestis thyodamas formosana	31	♂ (II)	〃
Timelaea maculata formosana	30	♂ (I, II)	Present paper
Sephisa daimio taiwana	31	♂ (I)	Maeki, Ogata, Shirôzu, 1965
B. Libytheidae			
Libythea celtis formosana	31	♂ (I, II)	Present paper

* Tables 1 (Papilionidae, Hesperiidae), and 2 (Pieridae, Lycaenidae, Riodinidae) are in Parts 1 and 2 of this series of papers (Kontyû 36 : 120, 130).

and one or two small elements are sometimes present in the nuclei (n, 32 and n, 33) and sometimes in the extra-nuclear cytoplasm (n, 31). From the above facts the supernumeraries are supposed to have derived from ancestral elements through chromosomal fragmentation. The fragmentation theory is supported by the size of large chromosomes in *Neptis* species, and by the fact *N. ananta* has smaller sizes of 13 large elements than those of the 17 large elements in *N. soma* and the 11 large elements in *N. hylas*. In view of the numerical variation of the chromosomes in *Neptis ananta*, it seems that the unstable status of karyotype offers an important cytological clue to the speciation of butterflies.

Chromosomes of *Tacoraea* species (*T. perius*, *T. selenophora* and *T. cama*) constantly number 30 in haploid set. While the above three species of *Tacoraea* completely agree in the number of chromosomes, their patterns of chromosome sizes are different. The nuclei of *T. perius* contain 4 small elements as are present in those of *T. cama,* while *T. selenophora* possesses an identical size for all chromosomes.

Timelaea maculata has been placed in Apaturinae on the basis of a comparative study of the male genitalia (Shirôzu, 1960). The findings relative to the early stages have corroborated this classification (Kubo & Muroya, 1967). The butterflies belonging to the Apaturinae seem to have the characteristic of having 1 or 2 huge elements. Namely, *Hestina japonica* (*n*, 30) has 1 huge chromosome, and *Sasakia charonda* (*n*, 29) has 2 huge elements, although *Apatura ilia* (*n*, 31) has an identical size for all chromosomes. The chromosomes of *T. maculata* (*n*, 30) reported in present paper contain 1 huge element. Therefore, present authors recognize it as a species of Apaturinae from the similarity in their karyotype.

Butterflies belonging to the Libytheidae probably have a common characteristic number of *n*, 31, as already confirmed in 2 species, *Libythea celtis* (Japan) and *Libytheana bachmanii* (Mexico). The newly examined *Libythea celtis formosana* also has 31 chromosomes in haploid set. However, the above species show slight difference in karyotype. *L. bachmanii* has 4 chromosomes smaller than the others, *L. celtis celtoides* has 4 or 5 small chromosomes and *L. celtis formosana* has apparently 5 small ones.

Summary

1. In this report, the number and relative sizes of chromosomes are described regarding 10 species of Nymphalidae, 1 species of Libytheidae, all from Formosa.

2. It is noteworthy that the karyotype has supernumerary chromosomes in *Neptis ananta taiwana* (*n*, 31, 32, 33) from Formosa.

3. The present authors cytologically support the conclusion regarding of the classification of *Timelaea maculata* introduced by Shirôzu and Saigusa (1960). Judging from the karyotype of this species having a huge chromosome it should be regarded as a species belonging to Apaturinae.

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