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The Chromosomes of a Japanese Spiny Lobster, *Panulirus japonicus* (v. Siebold)

By

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(*With 1 Table and 12 Textfigures*)

The result dealt with in the present paper constitutes the third report in the author's serial studies on chromosomes of the decapod Crustacea, which have been continuously carried on for several years.

Reviewing the literature that has appeared on the chromosomes of the Crustacea, there is found no reference concerning the subject in the group Palinura, in which the present material, *Panulirus japonicus* is included. The chromosome complex of this species is polymorphic in nature, a garniture being constituted of various kinds of telomitic and atelomitic chromosomes. In this respect, the present species seems to be quite unique among the Crustacea so far studied, and exhibits, at the same time, a striking difference from those in which only an isomorphic complex of chromosomes has been known. From the morphological view point, such a condition of chromosomes in the present species is very interesting when one considers the evolution of the chromosome complex in the Crustacea.

The investigation has been carried on at the Mitsui Institute of Marine Biology under the direction of Prof. Kan Oguma to whom the author wishes to express his sincere gratitude for his examination of findings and important suggestions. Further thanks are due to Dr. Sajiro Makino, under whose guidance and advices the present work could be completed.

Material and methods

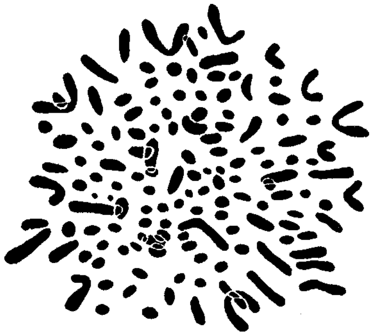
Panulirus japonicus (v. Siebold) is well known as the edible spiny lobster of the Palinuridae in our country. It distributes widely along the coasts of the Pacific ocean of Japan. The lobsters from which the material of the present work was obtained were collected by nets in waters adjacent to the Mitsui Institute of Marine Biology, at intervals from March to August in the years, 1933 and 1935. The testes were taken out in living state immediately after capture and put into the fixative as soon as possible. Flemming's weak solution showed the best result in preservation of the chromosomes in favourable condition. For staining, iron-haematoxylin method after Heindenhain was exclusively employed. Detailed treatment for preparation has been described in the author's previous paper (Niiyama, '34).

All the textfigures, except figs. *9* to *11* (photomicrographs), were drawn with the aid of an Abbe's drawing apparatus at the level of the desk on which the microscope was set.

Observations

The spermatogonium. In the testis, the cells contained in a cyst are of the same phase; some cysts are filled with only the spermatogonia, while others are provided with spermatocytes of different kinds, primary or secondary. This condition quite differs from that observed in *Paralithodes,* in which the spermatogonial cells are found lining the inner walls of the cyst while the deeper portions are occupied by the maturating cells in different grades of development in a manner similar to the mammals.

The chromosomes assume a rosette form in their metaphase arrangement, as they lie radially against the centre of the equatorial plate. They are found well apart from one another with sharply defined contour, so as to make counting of the chromosome number very easy in spite of their large number. In order to determine the number and form of the chromosomes, many equatorial plates were examined from their polar aspect. Examples are given in Textfigs. *1* to *3,* in every one of which overlapping of chromosomes occurs to a slight degree. After careful counting, it was decided that the diploid complex consists of 140 chromosomes. As is recognizable in the figures, the spermatogonial garniture shows polymorphic nature; of 140 chromosomes, 12 are atelomitic V-shaped elements while the remaining ones are all telomitic rods showing gradual diminution of size. According to the size and shape the atelomitic V-shaped chromosomes are arranged into 6 homologous pairs, of which three



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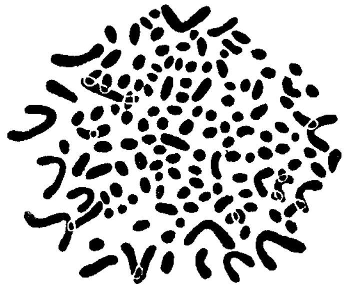

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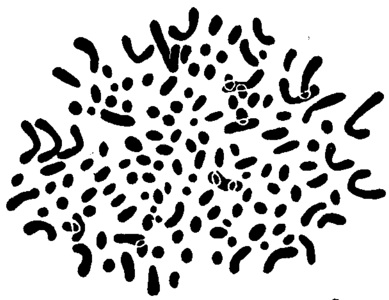

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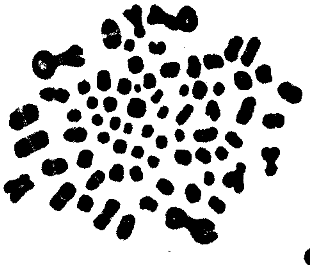
Textfigs. *1–3*. Metaphase groups of spermatogonia, 140 chromosomes in each. 3700×.

pairs are rather large as compared with the remaining three. They all have nearly submedian attachment of the spindle fibres. Among the telomitic chromosomes, it seems impossible to know which two constitute the homologous pairs, except for some longer ones. As a whole, the larger chromosomes occupy a peripheral position in the equatorial plate surrounding the smaller ones in the central part, though occasionally, there appear some cases in which a certain number of larger chromosomes are found lying at the central region. As can readily be noticed, in such condition of chromosomes as thus observed in the spermatogonium, the present species hardly shows resemblance to any species hitherto dealt with by the present author (*Cambaroides* and *Paralithodes*), nor to any one reported by several authors in different kinds of Decapoda, as *Cambarus* (Fasten, '14), *Cancer* (Fasten, '18, '24), *Lophopanopeus* (Fasten, '26), *Telphusa* (Delpino, '34) and *Pandalus* (Leopoldseder, '34). The present species, therefore, constitutes only one representative of Decapoda, in which the polymorphic nature of chromosomes is, at present, known.

The primary spermatocytes. The chromosomes of the primary spermatocyte are observed with absolute clearness in the polar view of the metaphase plates, as they scatter evenly and distinctly separated from each other. As the usual case may be, the larger



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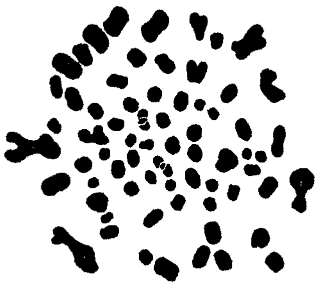

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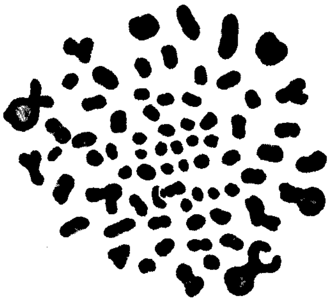

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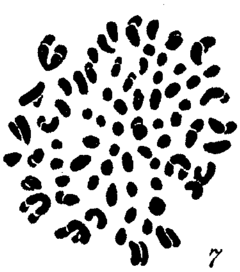

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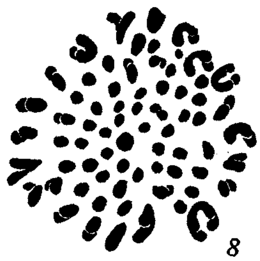

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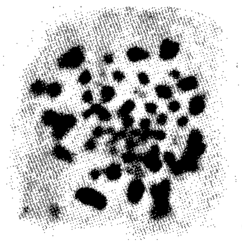

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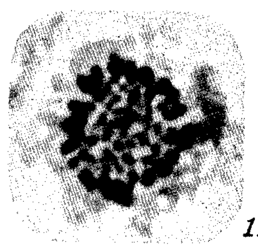

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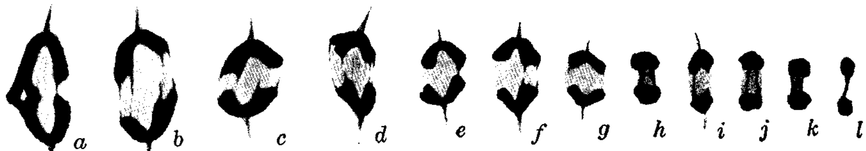

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Textfigs. *4–6*. Metaphase groups of primary spermatocytes, 70 tetrads in each. 3700×. Textfigs. *7–8*. Metaphase groups of secondary spermatocyte, 70 dyads in each. 3700×. Textfigs. *9–11*. Photomicrographs, 1800× ; *9*, Metaphase of primary spermat cyte ; *10*, side-view of the same ; *11*, Metaphase of secondary spermatocyte. Textfig. *12*. Separation of tetrads in the primary spermatocyte division anaphase (not all elements shown). 4200×.

ones take the peripheral position with those of smaller size in the central region of the spindle. Since the tetrads are distinctly separated from one another, as already mentioned, counting of their number is accomplished without any confliction. As expected from the count of the spermatogonial chromosomes, the primary spermatocyte contains invariably 70 tetrads (Textfigs. *4, 5, 6* and *9*).

Of large sized tetrads found in the peripheral part of the spindle, three are sharply distinguishable from others in having a striking characteristic in their morphology. They belong to the compound ring-tetrads, composed of two or three rings connected in a series, with atelomitic fibre attachment. It is certain that these tetrads are the descendants of three pairs of larger V-shaped chromosomes in the spermatogonium. Three more tetrads with atelomitic fibre attachment, but small in size, are always found in addition to those mentioned above. They are evidently descendants of three pairs of smaller atelomitic chromosomes in the spermatogonium, but whether they have ring structure is not known at present. When viewed from the pole of division, the remaining tetrads exhibit, at least in larger ones, a dumbbell shape though different in size, showing a remarkable transverse suture in each middle region. Considered from their shape and structure, it is evident that these tetrads are similar in nature to those previously described in *Cambaroides* and *Paralithodes* by the present author (Niiyama, '34, '35). In the smaller tetrads, however, the actual presence of the middle suture is not proven for certain.

In the anaphase of the first division, the atelomitic chromosomes separate into two identical double V's (Textfig. *12, a* to *f*). Of the telomitic tetrads, the larger ones also present a V-figure in their separated halves, but the V in this case is always single in nature (Textfig. *12, g* to *i*). In the remaining smaller tetrads the separating halves usually show no further transformation of form but remain still as spheres (Textfig. *12, j* to *l*).

The separation of chromosomes of the first division always takes place synchronously as may be seen in Textfig. *10*.

The secondary spermatocyte. As naturally expected from the mode of separation of chromosomes in the previous division, only one kind of secondary spermatocyte is produced in respect to the chromosome complex. There are also counted 70 dyads without exception in the metaphase equatorial plate of the secondary spermatocyte

(Textfig. *7*, *8* and *11*). The chromosomes are quite similar in arrangement to those of the spermatogonium and primary spermatocyte, as larger dyads are arranged at the periphery of the spindle while dot-like ones are in the central region. Further, all chromosomes corresponding to those of the spermatogonium are to be pointed out too in the garniture of the secondary spermatocyte: 6 atelomitic V-shaped chromosomes and the remaining telomitic ones from rod to spherical shape.

Remarks

There have hitherto been published some sixteen memoirs concerning the chromosome number of the Decapoda. In Table I the

TABLE I.

Suborder	Series	Species	Diploid	Haploid	Author
Natantia	Eucyphidea	**Crangon cataphractus*		40–44	Carnoy, '85
		Pandalus borealis		34	Leopoldseder, '34
Reptantia	Palinura	*Panulirus japonicus*	140	70	Niiyama, '36
	Astacura	**Astacus fluviatilis*		ca. 58	Prowazek, '02
		**Homarus*		18	Labbé, '04
		Cambarus virilis	200	100	Fasten, '14
		Cambarus immunis (?)		104	,,
		Cambaroides japonicus	196	98	Niiyama, '34
	Anomura	**Eupagurus prideoxii*		12*f.*	Weismann & Ishikawa, '88
		**Hippa talpoides*		60	Nichols, '09
		Paralithodes camtschatica	208	104	Niiyama, '35
	Brachyura	**Carcinus menas*		30–40	Carnoy, '85
		**Menippe mercenaria*	51–80	25–28*f.*	Binford, '13
		Cancer magister		60	Fasten, '18
		Cancer productus		58	Fasten, '24
		Cancer oregonensis		56	,,
		Cancer gracilis		52	,,
		Lophopanopeus bellus		62	Fasten, '26
		Telphusa fluviatilis		39	Delpino, '34

f.: female.

chromosome number in the decapod Crustacea is arranged in systematic order after Kükenthal ('27). Since in the species with asterisks in this table the chromosome number seems not to be settled absolutely, it is better to disregard them in course of the discussion.

Looking over these contributions, there are found only a few in which exact studies have been made on the morphology of chromosomes. Though the chromosomes of several species belonging to Natantia and Reptantia in the Decapoda have been extensively investigated by Leopoldseder ('34), Fasten ('14, '18, '24, '26), Delpino ('34) and the present author ('34, '35), most of the studies are confined to the chromosomes in the haploid complex and those in the diploid are unknown, except *Cambarus virilis* (Fasten, '14), *Cambaroides japonicus* (Niiyama, '34), *Paralithodes camtschatica* (Niiyama, '35) and the present *Panulirus*. As is to be supposed from the figures of the haploid complex indicated, the diploid complexes in *Pandalus, Cambarus, Cancer, Lophopanopeus* and *Telphusa* seem to be made up of rods and spherules of various sizes. In contrast to all the above cases, in which the chromosome shows such simplicity in form, the present case (*Panulirus*) exhibits a rather complicated nature, in that there are present a number of V-shaped chromosomes additionally to ordinary rods and spherules, thus expressing the polymorphic type. Summarizing these data obtained from the species above mentioned, it is suggested that in the development of the chromosome complex of Decapoda, fragmentation and association may play an important role for variation of chromosome number, as the V at least is to be accounted for as an association of two rods.

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