

A Comparative Study of the Chromosomes in Marine
Gastropods, with Some Remarks on
Cytotaxonomy and Phylogeny*

(With 3 Tables and 144 Figures in 12 Plates)

By

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Introduction

At present, there is no defense for the fact that chromosomes—their numbers, structure and behavior—constitute criteria of critical significance for an evolutionarily significant taxonomy. The differentiation of species in higher categories has perforce gone hand in hand with the diversification of the chromosomal mechanism. The record of evolution is partially reserved in the chromosomes of species now living (HUGHES-SCHRADER 1956).

The rise of genetics during the first thirty years of this century had a rather unfortunate effect on the prestige of systematics. A welcome improvement in the mutual understanding between geneticists and systematists has occurred phenomena tending to push systematics into the background. The importance of systematics in the study of evolution has been well accepted by the efforts of geneticists during the past fifty years (MAYR 1949). In order to discover true relationship and origin, it not only makes use of the general morphological characters, but also is quick to seize upon evidence afforded by physiological behavior, ecological relations, geographical distribution, fossil records, serological interactions, cytological characters, and genetical behavior in crosses. It is recently that genetics has influenced taxonomy: genetical and cytological analyses gave a new meaning and impetus. It is therefore very difficult to isolate taxonomy from modern experimental biology (MAYR 1949, SIMPSON 1945).

In recent years, chromosome cytology has made tremendous contributions to the development of animal taxonomy, particularly in the field of insects and related groups. Cytologists are concerned with the cytotaxonomic differences which exist between related species. They rarely involve major differences in the genetic mechanism, but often deal with difference in chromosome number or in the sizes and shapes of some of the chromosome complement. Such differences are sometimes useful to distinguish cryptic species that cannot be separated morphologically by non-cytologists (WHITE 1954).

Thus, the behavior and morphological changes of chromosomes in relation to the taxonomic arrangement have become a subject of prime importance to both cytologists and taxonomists. According to WHITE (1954), evolution involves two main aspects: 1) morphological and physiological changes, and 2) the formation of species. SHARP (1934) has pointed that cytological evidence of value to the taxonomy is of four chief kinds: chromosome number, chromosome morphology, chromosome behavior in crosses and aberrations in reproduction. McCLUNG (1914) put forward three lines of investigation: they are 1) comparison of karyotypes in related species, 2) comparison between the karyotypes of abnormal individuals and the normal chromosome set, and 3) experimental alteration of the karyotype.

McCLUNG (1917) and ROBERTSON (1916) have assumed that the union of rod-shaped chromosomes two by two at their inner ends producing V-shaped multiples constitutes a source of definite variation in the chromosome number within certain

related groups of animals. The rearrangements of this type in which two rod-shaped ones give rise to a large V-shaped element have occurred rather frequently in the phylogeny of many groups of animals such as *Drosophila*, grasshoppers, fishes, reptiles and birds (ROBERTSON 1916, PATTERSON and STONE 1952, MATTHEY 1948, NOGUSA 1960, YAMASHINA 1943, *etc.*). They are referred to as centric fusions (WHITE 1954). A centric fusion will lead to a decrease in number of chromosomes, and inversely a centric segregation will produce an increase in the chromosome number. Thus it is evident that the centric fusion will play important role in the variation of the chromosome numbers in related animals.

WHITE (1954) has presented three important evidences that deal with heritable change: they are gene mutation, structural change of chromosomes and polyploidy. The importance of polyploidy in the evolution of animals is very limited. In the field of cytology, the chromosomal changes which involve alteration in the sequence of the genes are of importance (WHITE 1957). SHARP (1934) has mentioned that, in cytotaxonomy, cytological characters, chiefly the number, morphology, and behavior of chromosomes are employed in the task of determining true natural relationships of species.

Along with those lines of investigations, a considerable contribution has been done in many groups of animals such as *Drosophila* (PATTERSON and STONE 1952, DOBZHANSKY 1941), Heteroptera (PFALER-COLLANDER 1941, da CUHNA 1945, MANNA 1951, *etc.*), Coleoptera (YOSIDA 1951, SMITH 1950, 1953, TAKENOUCI 1953, 1955, *etc.*), Odonata (OGUMA 1930, ASANA and MAKINO 1935), Lepidoptera (FEDERLEY 1938, BELIAJEFF 1930, LORKOVIC 1941, *etc.*), Orthoptera (McCLUNG 1914, ROBERTSON 1916, OHMACHI 1934, WHITE 1949~1957, HUGHES-SCHRADER 1943, 1951, 1956, HAREYAMA 1941, *etc.*), Neuroptera (NAVILLE and BEAUMONT 1936, HIRAI 1956, *etc.*), Crustacea (NIJYAMA 1959), Arachnida (HACKMAN 1948, SUZUKI 1954, *etc.*), Pisces (NOGUSA 1960), Reptillia (MATTHEY 1948), Aves (UDAGAWA 1951~1958) and Mammalia (MAKINO 1952), with valuable results in analyzing the evolutionary significance of the chromosomes.

The present investigations have been undertaken by the author with an aim to obtain, on the one hand, critical information on the bearing of cytology upon the mechanism and process of evolution, and on the other hand, to make a new approach to the problems of taxonomy in the Gastropoda (Mollusca), including the following orders, Archaeogastropoda, Mesogastropoda and Neogastropoda. In reference to the literature, it is evident that the chromosome survey has extensively been made in the pulmonate, while the gastropods have remained unexplored cytologically (*cf.* MAKING's monograph 1956). The basic data for understanding the chromosomal mechanisms of evolution in the Gastropoda have thus been in incomplete status. Under this situation an accurate survey of the chromosomes is quite desirable in order to formulate some ideas on the mechanism of evolution from both cytological and taxonomical standpoints.

INABA (1953, 1959 a, b) has carried out an extensive chromosome survey in some 60 species of the subclass Pulmonata and subclass Opithobranchia: he discussed

the basis of his cytological observations on taxonomical relationships of those animals with special regard to phylogeny. Recently, BURCH (1960) reported the chromosomes of 36 species of the pulmonate of order Basommatophora, a group of aquatic snails, with critical views on their systematic relations. In striking contrast to the Pulmonata, the knowledge on the chromosomes of the Gastropoda is very meagre. There are several classical papers appeared in this field by earlier investigators such as BOVERI (1890), ALEXENKO (1896), McMURICH (1896), AUERBACH (1896), CONKLIN (1902), KUSECHAKEWIFSCH (1913), SCHITZ (1920), ANKEL (1924, 1925), TUZET (1927, 1930), LAMS (1939) and POLISTER & POLISTER (1940). The majority of them have dealt with the study of atypical spermatozoa. STAIGER (1950, 1951, 1954) investigated the chromosomes of oocytes with particular concern to the question of male heterogamety.

The present author has investigated the chromosomes in male germ-cells of 53 species of the following three gastropod orders, Archaeogastropoda, Mesogastropoda and Neogastropoda. The results of this investigation constitute the data presented in the present paper.

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Historical review

On looking upon the literature, the author is accessible to references to several classical papers on the cytology of the Gastropoda (*cf.* MAKINO's monograph 1956). So far as the author is aware, BOVERI (1890) was the first to report the chromosomes of gastropods. He reported in eggs of *Carinaria mediterranea* 32 chromosomes in 2n

and 16 elements in n . AUERBACH (1896) studied the chromosomes of *Paludina vivipara* reporting 16 chromosomes in $2n$, and 16 and 8 elements in n . ALEXENKO (1896) reported 19 chromosomes in males and 20 chromosomes in females, and the occurrence of an X—O sex mechanism. McMURRICH (1896) reported the chromosomes of *Fulgur carica* to be 16 in eggs. In cleavage of *Crepidula plana*, CONKLIN (1902) showed that there were 60 in $2n$, and 30 in n . In *Murex brandaris*, 11~12 haploid number was reported by SCHITZ (1920). TUZET (1930) studied the 9 species belonging mainly to Neogastropoda and male heterogamety having X—O or X—Y mechanisms. KUSCHAKEWITSCH (1913) studied the chromosomes of *Conus mediterranea* reporting 14 chromosomes. He (1921) also reported 30 chromosomes in $2n$, 16 in n in *Cerithium vulgatum*, showing an X—O mechanism in males. ANKEL (1923) showed 34 diploid chromosomes and 17 haploid ones in *Bythini atentaculata*, and he (1925) also reported the chromosomes of *Cochlostoma septemspirale* as 12~13 in haploid, LAMS (1934) studied the chromosomes of *Murex brandaris* and reported 28 haploid chromosomes in males. POLISTER (1934) and POLISTER & POLISTER (1940) studied the chromosomes of several forms of *Viviparus*: the number of chromosomes varied with species from 18 to 26. ANKEL (1924) however reported 14 and 34 chromosomes in 2 species of *Viviparus*. Recently, STAIGER (1950, 1951, 1954) dealt with the chromosomes of 8 species of marine Neogastropoda, mostly in egg-maturation division and failed to observe the occurrence of heterotypic chromosomes, in contrary to the view of TUZET (1930) who argued the existence of an X—O condition in males. Very recently, INABA (1958) observed 34 diploid and 17 haploid chromosomes in the male of *Amalthea conica*. The reported data by previous authors so far obtained in the literature on the chromosomes of marine gastropods are tabulated in the form of a check-list in Table 1.

Due mainly to technical difficulties, the majority of earlier works have very inadequately investigated, and therefore no accurate knowledge have been collected on the morphology of the chromosomes. From the present-day-standards, those classical data seem to be far from the quality available for the current discussion. Particularly, the problems of male heterogamety claimed by KUSCHAKEWITSCH (1921) and TUZET (1930) in several forms of Neogastropoda merit attention of recent authors. Detailed investigations recently made by STAIGER (1950, 1951, 1954) and INABA (1958) have failed to observe the occurrence of heteromorphic chromosomes.

In contrast to marine gastropods, the chromosomes of pulmonate snails have rather extensively investigated by a number of authors (*cf.* MAKINO's monograph 1956). Recently, INABA (1953, 1959 a, b) has studied the chromosomes of about 60 species of land snails. Very recently, BURCH (1960) has reported the chromosomes of aquatic snails in 36 species. The reported evidence of the pulmonate chromosomes has been reviewed in detail in his paper (1960).

The present author has studied the chromosomes of 53 species of marine gastropods, through the ordinary paraffin method and supplement with the water-pretreatment squash method according to MAKINO and NISHIMURA (1952). Reviewed the author's own results, it becomes evident that the number of chromosomes ranges

Table 1. Chromosome number in marine gastropods studied by previous authors.

Species	2n	n	Note	Author
Order Archaeogastropoda				
Family Neritidae				
<i>Neritina fluviatilis</i>	19s	10 (I)	X-O	ALEXENKO '96
"	20o			
"	18s	9 (II)	X-Y	TUZET '30
Order Mesogastropoda				
Family Turritellidae				
<i>Turritella triplicata</i>		9-11	X-O?	SCHITZ '20
Family Cerithiidae				
<i>Cerithium vulgatum</i>	ca. 30	16 (I)	X-O	KUSCHAKEWTSCH '21
"		16 (I, II)		TUZET '30
Family Amaltheidae				
<i>Amalthea conica</i>	34s	17 (I, II)		INABA '58
Family Calptraeidae				
<i>Crepidula plana</i>	60m	30 (I, II)		CONKLIN '02
Family Pterotracheidae				
<i>Pterotrachsa mutica</i>	32m	16 (I, II)	sperm nuclei	BOVERI '90
		16		
Order Neogastropoda				
Family Muricidae				
<i>Murex brandaris</i>		28 (I)		LAMS '34
<i>M. trunculus</i>		11-12 (I)		SCHITZ '20
"		14 (I)	X-O	TUZET '30
"		35 (I)		STAIGER '50,
<i>Ocenebra erinaceus</i>		35 (I)		" '51
<i>Purpura lapillus</i>		18 (I)		"
"		13 (I)	isolated	"
<i>Nassa nitida</i>		14 (I)	X-O	TUZET '30
Family Pyrenidae				
<i>Columbella rustica</i>		16+ (I)		SCHITZ '17
"	ca. 20		X-O	TUZET '30
"		34 (I)		STAIGER '50
Family Buccinidae				
<i>Pisania maculosa</i>		16 (I)	X-O	TUZET '27
"		35 (I)		STAIGER '50
<i>Euthria cornea</i>		35 (I)		" '50
<i>Buccinum undatum</i>		35 (I)		" '50, '51
<i>Fulgur carica</i>		ca. 16 (I)		MCMURICH '96
Family Fasciolaridae				
<i>Fasciolaria tulipa</i>		31?	X-O	HYMAN '23
<i>F. lignaria</i>		35 (I)		STAIGER '50

Abbreviations in the columns "2n" and "n"

s : spermatogonium, o : oogonium, m : somatic mitosis,

I : primary spermatocyte and II : secondary spermatocyte

in haploid from 9 to 36 according to species, and that there is no evidence for the occurrence of particular chromosomes heteromorphic in shape and behavior.

Material and methods

The species studied in the present paper are listed below according to the taxonomical arrangement under the order, family, subfamily and species. They are 53 in table, being represented by 3 orders, 16 families.

The specimens used for this study were collected mainly from sea-shore adjacent to the College which is located in Nagata Honmachi, Yoshimi, Shimonoseki City, Yamaguti Prefecture. Shimonoseki City is situated at the westernmost end of the Main Island of Japan. The shell-fish fauna was studied by YOSHIDA, AMIO and NOJIMA (1957) in the vicinities of the city: it was reported that the sea around the College contained the fauna of both south and north sea. The method of observations was just the same as that reported in the author's previous papers (1961 a, 1961 b,) on the chromosomes of *Balanus amphitrite albicostatus*, *Hemicentrotus pulcherrimus*, *Anthocidaris crassispira* and *Mitella mitella*. The materials for observation were brought to the laboratory, and the gonads were picked out by breaking the shells. Then their sexes were examined, and only the males were used as material. A part of the testes was cut into small pieces, on the slide glass, and treated with water, or with N/100 KBr, for 10 to 15 minutes. After getting rid of the solution, they were fixed and stained with acetic orcein and acetic dahlia for 10 to 20 minutes.

The parts of the materials stained with acetic dahlia were stained again with acetic gentian violet. After staining, they were mounted with a cover-glass. Under some sheets of filterpapers, the slides were squashed by pressing with the finger-thumb. If it be necessary, they were squashed again by rolling a glass tube about 1.5 cm across upon them.

In supplement, often parts of materials were fixed in the usual way with CHAMPY's original solution, 1/2 to 1/3 diluted CHAMPY's solution or LOBIANCO's mixture. After the usual paraffin imbedding, the materials were sectioned into from 6 to 8 micra in thickness, and stained with HEIDENHAIN's iron-haematoxylin.

All the figures were made by the aid of an Abb's drawing apparatus. The magnification of all the figures was 4,000 times in diameter.

In the application of the squash technique it was rather difficult to get preparations available for the study of the spermatogonial chromosomes. The chromosomes showed a tendency of swelling with the elapse of time, making especially V-shaped chromosomes undistinguishable. With these preparations, therefore, the chromosomes of the 1st spermatocyte were mostly observed in this study.

Table 2.

Order Archaeogastropoda	
Family Haliotidae	<i>Haliotis (Sulculus) japonica</i> REEVE
Family Fissurellidae	<i>Clypidina (Montfortula) picta</i> (DUNKER) <i>Macroschisma sinensis</i> A. ADAMS <i>Macroschisma dilatata</i> A. ADAMS
Family Patellidae	<i>Cellana toreuma</i> (REEVE) <i>Cellana eucosmia</i> (PILSBRY) <i>Cellana nigrolineata</i> (REEVE)
Family Acmaeidae	<i>Patelloida (Collisellina) saccharina lanx</i> (REEVE) <i>Patelloida (Asteracmea) pygmaea</i> (DUNKER) <i>Patelloida (Asteracmea) pygmaea lampanicola</i> HABE <i>Notoacmea schrenckii</i> (LISCHKE) <i>Notoacmea concinna</i> (LISCHKE) <i>Notoacmea fuscoviridis</i> TERAMACHI
Family Trochidae	
Subfamily Margaritinae	<i>Stomatella lyrata</i> PILSBRY
Subfamily Monodontinae	<i>Cantharidus callichroa</i> (PHILIPPI) <i>Thalotia japonicus</i> (A. ADAMS) <i>Monodonta labio</i> (LINNÉ) <i>Monodonta neritoides</i> (PHILIPPI) <i>Tegula (Chlorostoma) lischkei</i> (TAPPARONE-CANEFRI) <i>Tegula (Omphalius) nigerrima</i> (GMELIN) <i>Tegula (Omphalius) rustica</i> (GMELIN) <i>Tegula (Omphalius) pfeifferi carpenteri</i> (DUNKER)
Family Turbinidae	
Subfamily Turbininae	<i>Turbo (Batillus) cornutus</i> SOLANDER <i>Lunella coronata coreensis</i> (RÉCLUZ) <i>Astrarium haematragum</i> (MENKE)
Family Neritidae	<i>Puperita (Heminerita) japonica</i> (DUNKER)
Order Mesogastropoda	
Family Littorinidae	<i>Littorina (Littorivaga) brevicula</i> (PHILIPPI) <i>Littoraria (S. S.) strigata</i> (PHILIPPI) <i>Nodilittorina granularis</i> (GRAY) <i>Nodilittorina picta</i> (PHILIPPI)
Family Potamididae	<i>Cerithidea (Cerithidea) rhizophorarum</i> A. ADAMS <i>Cerithidea (Cerithideopsilla) cingulata</i> (GMELIN) <i>Cerithidea (Cerithideopsilla)</i> <i>djadjariensis</i> (K. MARTIN)

Family Cerithiidae	<i>Batillaria zonalis</i> (BRUGUIÈRE)
Subfamily Cerithiinae	<i>Batillaria multiformis</i> (LISCHKE)
	<i>Proclava kochi</i> PHILIPPI
	<i>Contumax kobelti</i> (DUNKER)
Family Naticidae	<i>Neverita (Glossaulax) didyma</i> (RÖDING)
Order Neogastropoda	
Family Muricidae	<i>Chicoreus asianus</i> KURODA
	<i>Bedevina birileffi</i> (LISCHKE)
	<i>Purpura (Mancinella) bronni</i> DUNKER
	<i>Purpura (Mancinella) clavigera</i> KÜSTER
	<i>Purpura (Mancinella) luteostoma</i> (HOLTEN)
Family Pyrenidae	<i>Pyrene testudinaria tylerae</i> (GRIFFITH et PIDGEON)
	<i>Pyrene (Mitrella) bicincta</i> (GOULD)
	<i>Anachis (Anachis) misera</i> (SOWERBY)
	<i>Columbella (Euplica) versicolor</i> (SOWERBY)
Family Buccinidae	<i>Babylonia japonica</i> (REEVE)
	<i>Pisania (Japeuthria) ferrea</i> (REEVE)
	<i>Cantharus (Pollta) subrubiginosus</i> (SMITH)
Family Nassariidae	<i>Tritia (Hinia) festiva</i> (POWYS)
	<i>Nassarius (Niotha) livescens</i> (PHILIPPI)
Family Mitridae	<i>Pusia hizenensis</i> (PILSBRY)

Results of observations

Order Archaeogastropoda

The knowledge on the chromosomes of the species belonging to this order has remained very limited. Referring to the literature, only 1 species, *Neritina fluviatilis*, was cytologically studied by authors: ALEXENKO (1896) reported $2n$, 20 and n , 10, (X—O) while TUZET (1930) gave $2n$, 18s and n , 9 (II), showing an X—Y mechanism of sex-determination in the male. The author has studied the chromosomes of 26 species which cover 14 genera and 7 families, such as Haliotidae, Fissurellidae, Patellidae, Acmaeidae, Trochidae, Turbinidae and Neritidae.

A. Halictidae

According to INO (1952), there has been described 9 species belonging to this family. The author investigated the chromosomes of 1 species. Formerly no species of this family has cytologically been studied so far, and therefore the present report is the first on the chromosomes of this family.

1. *Haliotis (Sulculus) japonica* REEVE

This species is distributed all over the world. It is an ear-shell which is distinguished from other species by its many suction holes. This specimen was obtained by a fisherman who gathered it from the shore a little off Yoshimi district, Shimonoseki City.

The spermatogonial metaphase plate showed the chromosome number of $2n$, 34 which varied in length ranging from dot-like to short rod-like ones, but there seem to occur several pairs of J-shape (Fig. 1).

In the 1st spermatocyte metaphase there were 17 bivalents of either dot-like or rod-like form: they were somewhat larger than the spermatogonial chromosomes in outline (Fig. 2).

In the 2nd spermatocyte metaphase plate there were 17 chromosomes, smaller in outline than those of the 1st spermatocyte (Fig. 3).

The chromosome number of this species was determined as $2n$, 34 and n , 17.

B. Fissurellidae

With respect to the chromosomes of the species of this family, there is accessible no paper in the literature. The author studied the chromosomes of 3 species representing 2 genera.

2. *Clypina (Montfortula) picta* (DUNKER)

This species is distributed over the regions south of central district of Japan Proper, and it clings to the reefs below the high tidal line. They are small limpets with shells incised lightly on the edge. They were collected from the shore behind this College in June, 1961.

In the 1st spermatocyte metaphase there were 17 bivalents of somewhat angular dot-like outline. In the periphery of the equatorial plate there existed one or two larger bivalents (Figs. 4~5).

3. *Macroschisma sinensis* A. ADAMS

This species attaches itself to either the top or base of the reef on the shore south of central district of Japan Proper. It has a large sized slug-like body, on the top of which a small, flat dish like shell is located. The apex of the shell is provided with a slender aperture. The specimens were collected from the shore behind the College in June, 1961.

In the 1st spermatocyte metaphase there were 16 bivalents, rather angular dot-like in form; they varied in size in a considerable degree (Figs. 6~7).

4. *Macroschisma dilatata* A. ADAMS

The range of distribution and the form of this species resemble those of *M. sinensis* as described above, but the former is smaller than the latter, showing a variation of the location of the small aperture on the top of the shell. They were collected from the shore behind the College, in June, 1961.

In the spermatogonial metaphase there were 32 chromosomes containing several

pairs of V-shaped ones. The V-shaped chromosomes seemed to consist of 2 pairs. The other chromosomes varied in length, ranging from dot-like form to rod-like one, which were considered as J-shaped chromosomes, though not very distinct (Fig. 8).

In the 1st spermatocyte metaphase there were 16 angular bivalents which varied in size in considerable degree. Large-sized bivalents have a tendency to arrange in the periphery of the equatorial plate (Figs. 9~10).

In the 2nd spermatocyte metaphase, there were observed n , 16 chromosomes of dot-like form, far smaller in size than the 1st spermatocyte (Fig. 11).

The chromosome number of this species was decided as $2n$, 32 and n , 16.

C. Patellidae

Previously no species of this family has cytologically been researched. The author investigated the chromosomes of 3 species belonging to 1 genus of this family. They are small limpets, whose cells are also small in all the species studied here.

5. *Cellana toreuma* (REEVE)

This species is distributed all over the country, inhabiting on the shore within the high tidal zone. It is a small, flat limpet. The specimens for study were collected from the shore behind the College, in May and November, 1960.

The 1st spermatocyte metaphase showed 9 dot-shaped bivalents. Surrounding 1 or 2 bivalents in the center, 7 to 8 bivalents were found arranged in the peripheral area (Figs. 12~13).

6. *Cellana eucosmia* (PILSBRY)

Ranging from the regions south of Hokkaido to Kyushu, this species is distributed. The shell is taller than that of *C. toreuma* and is distinguished from others by the coloration in side of its shell. The material was collected from the shore behind the College in June, 1960.

The spermatogonial metaphase showed 18 chromosomes consisting of short rod-shaped or dot-shaped elements. They were arranged radially (Figs. 14~15).

In the 1st spermatocyte metaphase, 9 dot-shaped bivalents were observed (Figs. 16~17).

The chromosome number of this species was confirmed to be $2n$, 18 and n , 9.

7. *Cellana nigrolineata* (REEVE)

This species is distributed all over the country and adheres to the shore reef about the high tidal line. Though it resembles *C. eucosmia*, it is easily distinguished from others by its reddish brown radiant color on the surface of the shell. The specimens were collected from the shore behind the College in February, 1960.

In the 1st spermatocyte metaphase there were observed 9 dot-like or short rod-like bivalents which varied in size (Figs. 18~19).

The 2nd spermatocyte metaphase contained n , 9 chromosomes of dot-shape, smaller in size than the former.

The arrangement of chromosomes in any of the above 3 species was similar to that of *C. toreuma* (Figs. 20~21).

Through the course of the meiotic divisions, there was found no special chromosome regarded as the sex-determining element, either in shape or in behavior.

D. Acmaeidae

No paper on the chromosomes of the species belonging to this family have been published so far by any author. The author observed the chromosomes of 6 species covering 2 genera in this family, as given below.

8. *Patelloida (Collisellina) saccharina lanx* (REEVE)

This species attaches itself to the shore reef around the high tidal line in all parts south of Hokkaido. It is a star-shaped limpet. Though it resembles *Siphonaria japonica* of the Basommatophoric Pulmonata, both of them are distinguishable from each other by the such characters as its radial ribs on the surface of the shell and the apex of the shell. The material for this study was collected from the shore behind the College in June, 1960.

In the 1st spermatocyte metaphase, 9 bivalents with dot-like forms varying in size were observed (Figs. 22~23).

9. *Patelloida (Asteracmea) pygmaea* (DUNKER)

This species and is an oval limpet is distributed on the high tidal zone of southern part of Japan Proper, Shikoku, Kyushu, Korea and Formosa.

The 1st spermatocyte metaphase was found to contain 9 bivalents with a dot-like form varying in size (Figs. 24~25).

10. *Patelloida (Asteracmea) lampanicola* HABE

This species is a small limpet about 5 to 6 mm long and clings to the surface of the shells of roll-shells such as *Batillaria multiformis* and *Cerithidea (Cerithideopsis) cingulata* which inhabit the dry beach in all parts of this country. The specimens collected from the shore in front of the College in June, 1960 provided the present material.

The 1st spermatocyte metaphase showed 9 dot-like bivalents which varied in size (Figs. 26~27).

11. *Notoacmea schrenckii* (LISCHKE)

This species is distributed every part of this country and is a limpet whose shell is oval, flat plate-shaped. The specimens as material were collected from the shore in front of the College in June, 1960.

In the 1st spermatocyte metaphase 9 dot-like bivalents were observed (Figs. 28~29).

12. *Notoacmea concinna* (LISCHKE)

Though the distribution and form of this species resemble those of *N. schrenckii* described above, both of them are distinguishable from each other by the difference

between the figures on the surface of the shells. The specimens for this study were collected from the shore behind the College in February, 1960.

In the 1st spermatocyte metaphase, 9 dot-like bivalents varying in size were observed (Figs. 30~31).

13. *Notoacmea fuscoviridis* TERAMACHI

The distribution and the form of this species resemble those of *N. schrenchii*, but the surface of its shell is speckled with green coloration. The material for this study was collected from the shore behind the College in March, 1960.

In the 1st spermatocyte there occurred 9 dot-like bivalents which varied in size considerably (Figs. 32~33).

The above 6 species are common in the fact that the chromosome number and arrangement of the bivalents at the metaphase are similar. Thus, it is evident that the 6 species here studied are characterized by $n, 9$.

E. Trochidae

Although there are a great number of species in this family, no chromosome survey has been made as far. Here the author has had an opportunity to study the chromosomes of 9 species represented by 4 genera in this family.

14. *Stomatella lyrata* PILSBRY

This species adheres to the base of the reef below the tide mark of the regions south of the central district of Japan Proper. It is a small sized roll-shell which is equipped with a shell globular and rod-shell-like. The specimens for this study were collected from the shore behind the College in May, 1961.

In the 1st spermatocyte metaphase there were observed 21 dot-shaped bivalents of nearly equal size (Figs. 34~35).

15. *Cantharidus callichroa* (PHILIPPI)

This species clings to the seaweeds in shallow waters of the high tidal zone in all the parts of this country. It is a conical small sized roll-shell with a mouth obstructed by folds. The surface of its shell is speckled beautifully. The specimens collected from the shore behind the College in January and February, 1961 furnished the material for study.

In the spermatogonial metaphase 18 pairs of chromosomes were observed. Their forms, except a few pairs of V-shaped ones varying in length, range from the dot-shape to the rod-shape. They were radially arranged on the equatorial plate (Fig. 36).

The 1st spermatocyte metaphase contained 18 bivalents. Except several pairs of large-sized dot-like bivalents, the remaining elements were somewhat smaller dot-like one. The large-sized bivalents are considered to have been derived from the V-shaped chromosomes which occurred in the spermatogonial metaphase (Figs. 37~38).

Based on the above observations it is evident that the chromosome number of this species is $2n, 36$ and $n, 18$.

16. *Thalotia japonica* (A. ADAMS)

This species is distributed in all parts of this country and inhabits the reef in the high tidal zone. It is a smaller conical roll-shell, as compared with *C. callichria* described above. The specimens collected from the shore behind the College in February, 1961 were used as material.

It was found that the 1st spermatocyte metaphase contained 18 bivalents. Among them several large-sized chromosomes are remarkable. Their forms were dot-shaped and nearly identical in size (Figs. 39~40).

17. *Monodonta labio* (LINNÉ)

This species is distributed on the high tidal zone in all the parts of this country and is a small roll-shell with a hard globular shell. On the dark green surface of the shell were arranged square granules in checker. The specimens were collected for the present study twice or three time every month during the period from 1958 to 1960 from the shore both in front of and behind the College.

The 1st spermatocyte metaphase was found to contain 18 dot-shaped bivalents which varied in size in considerable degree (Figs. 41~42).

In the 2nd spermatocyte metaphase there were 18 dot-like chromosomes, being smaller in size than the former (Figs. 43~44).

It was observed that the chromosome number of this species was n , 18 and that through the course of meiotic divisions, there occurred no special chromosome either in shape or in behavior.

18. *Monodonta neritoides* (PHILIPPI)

This species is similar to *M. labio* as mentioned above in the distribution. It is a roll-shell with a shell the surface of which is smooth, rather flat and globular. It is easily distinguishable by the coloration on the surface of the shell, being dark green. The individuals on the shore behind the College in June, 1960, provided the material.

In the 1st spermatocyte metaphase there occurred 18 dot-like bivalents varying in size (Figs. 45~46).

19. *Tegula (Chlorostoma) lischkei* (TAPPARONE-CANEFRI)

This species is a conical roll-shell distributed on the high tidal zone in most parts of this country. The specimens for study were collected twice or three times every month from December, 1959 to May, 1960 from the shore both in front of and behind the College.

The 1st spermatocyte metaphase was found to possess 18 angular dot-shaped bivalents which varied in size (Figs. 47~48).

20. *Tegula (Omphalius) nigerrima* (GMELIN)

This species is almost similar to *T. (C.) lischkei* in the distribution but is smaller as compared with *T. (C.) lischkei*. The upper convoluted part of the spiral assumes an obtuse angle. The materials for study were collected from the shore behind the College in July, 1960.

In the 1st spermatocyte metaphase there were observed 18 angular dot-shaped bivalents varying in size in considerable degree (Figs. 49~50).

21. *Tegula (Omphalius) rustica* (GMELIN)

This species is distributed on the high tidal zone over the regions south of Hokkaido. The shell bears a close resemblance to *T. (C.) lischkei* as mentioned above, but it is distinguished from others by the minute morphological character in umbilical aperture. The specimens as material were collected from the shore behind the College in May, 1960.

The spermatogonial metaphase showed 36 chromosomes, the forms of which ranged from the dot-like to rod-like. A few seemed to be V-shaped or J-shaped chromosomes, though not very certainly (Fig. 51).

It was observed that the 1st spermatocyte metaphase contained angular dot-shaped bivalents varying in size (Figs. 52~53).

The chromosome number of this species was found to be $2n$, 36 and n , 18.

22. *Tegula (Omphalius) pfeifferi carpenteri* (DUNKER)

This species is distributed on the high tidal zone in most districts along the coast of the Japan Sea and Kyushu. Although it resembles *T. (O.) rustica* as mentioned above, it is distinguished from the other by its shell having the spiral vein at the base. The material for study was collected from the shore behind the College, in June, 1960.

In the 1st spermatocyte prophase there occurred 18 large-sized bivalents which were variable in form (Figs. 54~55).

The 1st metaphase plate showed 18 angular dot-shaped bivalents (Figs. 56~57).

F. Turbinidae

According to OKADA and FUJITA (1933), the native top-shells in Japan were represented by 22 species. Most of them, however, have their habitats either in the depth of the sea or in the waters south of Japan, so that it is difficult to obtain them. In the present investigation the author investigated the chromosomes of 3 species covering 3 genera, which are comparatively easily obtainable. There has been accessible no paper on the chromosomes of the species belonging to this family. The morphological feature common to all of this family is that they have a calcareous operculum.

23. *Turbo (Batillus) cornutus* SOLANDER

This species is the commonest one inhabiting the shallows below the low water line in all the parts south of Hokkaido in Japan. The spine type has been reported by many investigators as the "Inland Sea" species and the non-spine type as the "Open Sea" species. In that respect the author tried to investigate the difference, if any, of the chromosomes between them. The materials used were collected from the shore behind the College in June, 1960 and June, 1961, and those obtained by a fisherman who caught them from the shore off the Yoshimi district.

(a) Non-spine form

In the 1st spermatocyte metaphase there existed 18 dot-shaped bivalents varying in size rather remarkably (Figs. 58~59).

(b) Spine form

The results of the 1st spermatocyte metaphase obtained in this form wholly agreed with the findings of the results in the former form (Figs. 60~61).

It is thus evident that both non-spine and spine forms are characterized by the same cytological feature, the chromosome number being n , 18.

24. *Lunella coronata coreensis* (RÉCLUZ)

This species is a roll-shell assuming a form of an irregular deflected globe. The specimens for study were collected from the shore behind the College once or twice every month during the period from December, 1959 to May, 1960.

In the 1st spermatocyte metaphase, 18 dot-shaped bivalents varying in size were observed (Figs. 62~63).

At the periphery of the nucleus plate there were 2 or 3 somewhat large-sized bivalents.

25. *Astraliium haematragum* (MENKE)

This species is a conical aculeous small roll-shell distributed on the high tidal zone, ranging from the regions the central district of Japan Proper, Shikoku, Kyushu to Chira. It is distinguished from others by the purple coloration at the periphery of its calcareous operculum. The individuals collected from the shore behind the College in July, 1960 and in June, 1961 furnished the present material.

It was observed that the 1st spermatocyte metaphase contained 18 dot-shaped bivalents which were variable considerably in size (Figs. 64~65).

G. Neritidae

In this family, ALEXENKO(1928) studied the chromosomes of *Neritina fluviatillis* reporting $2n$, 19s—20o and n , 10 (I), and TUZET (1930) reported $2n$, 18s and n , 9 (II) in the same species. The former author claimed an X-O sex-mechanism, whereas the latter, X-Y. It is doubtful that the chromosome number in the species differs with authors. The present author studied the chromosomes of 1 species as below.

26. *Puperita (Heminerita) japonica* (DUNKER)

This species inhabits, clinging to the shore reef above the high tidal line. It is a horseshoe-shaped roll-shell speckled with white or brown coloration on the black ground. The material was collected from the shore behind the College in November, 1959, February, 1960 and March, 1961.

It was found that the spermatogonial metaphase showed 22 chromosomes. They are apparently of dot-like type, some having the outline of J-shape, though the exact morphology has remained uncertain due to poor preservation of chromosomes (Figs. 66~67).

In the 1st spermatocyte metaphase there were 2 large-sized bivalents, 5 to 6 medium-sized ones and 2 to 3 small-sized ones, being 11 in total (Figs. 68~69).

In the 2nd spermatocyte metaphase there were observed 11 chromosomes which were smaller size than the former (Fig. 70).

Based on the above findings, it was evident that the chromosome number of this species was $2n$, 22 and n , 11 without doubt, and that through the course of the meiotic divisions, there was found no special chromosome either in shape or in behavior.

Order Mescgastropoda

In reference to the literature, it is evident that the chromosomes of 5 species covering 5 genera and 5 families have so far been reported by previous authors. The author has had an opportunity to observe 12 species which are represented by 7 genera and 4 families.

H. Littorinidae

Any of the previous authors has not observed the chromosomes of the species belonging to this family. The author investigated chromosomes of 4 species which cover 2 genera in this family.

27. *Littorina (Littorivaga) brevicula* (PHILIPPI)

This species is distributing every part of high tidal line in this country and is a small-sized globular roll-shellfish which inhabits the reef. The individuals gathered from the beach in front of this College in January, 1961, constituted the material for study.

In the metaphase of the 1st spermatocyte, 17 dot-like bivalents varying in size considerably were observed (Figs. 71~72).

28. *Littoraria (s.s.) strigata* (PHILIPPI)

This species is a small roll-shellfish which takes a form of a tower and the surface of which is speckled with tiger's stripes. The specimens were gathered from the beach in front of this College in July, 1960.

The metaphase of the 1st spermatocyte showed 17 bivalents, which consisted of some large-sized ones and small-sized dot-like ones (Figs. 73~74).

In the metaphase of the 2nd spermatocyte there were 17 dot-like chromosomes, smaller in size than the former (Fig. 75).

29. *Nodilittorina granulais* (GRAY)

Ranging on the shores south of Japan Proper, Shikoku and Kyushu, this species adheres to the reefs near the high tidal line. It is a roll-shellfish far smaller than *L. strigata*. The material for study obtained from the shore behind this College in July, 1960.

The metaphase of the 1st spermatocyte was found to contain 18 bivalents which

were of dot-type in general appearance varying in size (Figs. 76~77).

In the metaphase of the 2nd spermatocyte there occurred 18 chromosomes of a dot-like appearance, smaller than the former (Fig. 78).

30. *Nodilittorina picta* (PHILIPPI)

This species is distributed below the high tidal line over the regions south of the southern part of Japan Proper. It is a roll-shellfish smaller than *N. granularis* and the surface of the shell is speckled with granular forms. The individuals gathered from the beach behind this College in July, 1960 constituted the material for study.

The metaphase of spermatogonium was found to contain 30 chromosomes in 15 pairs. They were of dot- or rod-type and vary in size considerable degree (Fig. 79).

In the metaphase of the 1st spermatocyte, the nucleus plate consisted of 15 bivalents assuming various shapes and sizes (Figs. 80~81).

The nucleus plate of the 2nd spermatocyte showed 15 chromosomes of dot-like appearance, smaller than the former (Fig. 82).

Based on the above findings, it is evident that the chromosome number of this species is $2n$, 30 and n , 15.

I. Potamididae

On the chromosomes of this family, no report has formerly been appeared. The author studied the chromosomes of 5 species which cover 2 genera in this family.

31. *Cerithidea (Cerithidea) rhizophorarum* A. ADAMS

Ranging from the regions south of Japan Proper, to the southwestern Islands and the Republic of China, this species inhabits a dry beach at the estuary, subjecting to the effect of fresh-water. It is a cylindrical roll-shell, the top of its shell being eroded entirely. The specimens obtained in the dry beach of the mouth of the River Yoshida, Ozuki-machi, Shimonoseki City, in July, 1961 constituted the present material.

In the 1st spermatocyte metaphase there were observed 18 bivalents assuming dot-like appearance and varying in size in a considerable degree. On the circumference of the nucleus plate there were scattered large-sized bivalents surrounding the smaller ones in the central space (Figs. 83~84).

32. *Cerithidea (Cerithideopsilla) cingulata* (GMELIN)

This species inhabits the sandy mud of the high tidal zone of every part of this country. It is a conical roll-shell provided with brown fasciae. The material was gathered from the shore in front of this College in June, 1960 and in June, 1961.

The nucleus plate of the 1st spermatocyte was large in size, with large bivalents. In the nucleus plate in the metaphase there were 18 bivalents of various sizes. At the periphery of the nucleus plate was found occupied large bivalents (Figs. 85~86)

33. *Cerithidea (Cerithideopsilla) djadjariensis* (K. MARTIN)

This species is distributed over the regions ranging from the central district

of Japan Proper and southwards to Formosa and is a conical roll-shell. The surface of its shell is dirty brown and all the spiral lines, being crossed by the longitudinal grooves, form granular lines, which, at first sight, look like a file. Its habitat is similar to that of *C. (C.) rhizophorum*. The specimens for study were collected from a dry beach of the estuary of the River Yoshida.

The 1st spermatocyte metaphase showed 18 dot-shaped bivalents with a considerable size-variation. In the peripheral area of the nucleus plate larger bivalents were scattered (Figs. 87~88).

34. *Batillaria zonalis* (BRUGUIÈRE)

This species inhabits a dry beach throughout Japan. It is a conical roll-shell on the surface of which there are protuberant projections. The material for study was collected from the shore behind the College in June, 1960.

The spermatogonial metaphase was found to contain 36 chromosomes. They varied in size from dot-like to short rod-like ones (Fig. 87). Further details of the chromosome morphology were difficult to examine, because of technical difficulty in fixation.

In the 1st spermatocyte metaphase 18 bivalents varying in size were observed. In the periphery of the nucleus plate there were 1 to 2 larger bivalents (Figs. 90~91).

The chromosomes at the 2nd spermatocyte metaphase assumed dot and short rod-shape being smaller in size than in the former. The number was decided as 18 (Figs. 92~93).

It was concluded that the chromosome number of this species was $2n$, 36 and n , 18.

35. *Batillaria multiformis* (LISCHKE)

This is a common species distributed over all parts of the country as *B. zonalis* is. It is a short tower-like roll-shell with considerable individual as well as geographical variations. Some of them have a dark grey coloration, while others show white belts around their sutures. The materials for this study were collected from the shore in front of the College in June, 1960 and May, 1961.

The spermatogonial metaphase showed 36 chromosomes in 18 pairs. The metaphase plate shows several pairs of V- or J-shaped chromosomes in addition to certain numbers of dot-like and short rod-like ones (Fig. 94). Further details have remained uncertain, because of unfavorable preservation of the chromosomes.

The 1st spermatocyte metaphase was found to contain 18 bivalents with a considerable size-variation. In the periphery of the nucleus plate there occurred 1 to 2 larger bivalents (Figs. 95~96).

In the 2nd spermatocyte metaphase, were found 18 chromosomes varying in size from dot-like to short rod-like (Figs. 97~98).

Based on the above observations, it is evident that the chromosome number of this species is undoubtedly $2n$, 36 and n , 18.

J. Cerithiidae

Looking upon the literature, the author is referable to two papers in which 3 species belonging to this family have cytologically been investigated. KUSCHAKEWITSCH (1921) reported on *Cerithium vulgatum*, $2n$ about n , 16 (I) with an X-O sex-determining mechanism, whereas TUZET (1930) in the same species described n , 16 (I, II). Here the author studied the chromosomes of 2 species represented by 2 genera in this family.

35. *Proclava kochi* PHILIPPI

This species inhabits the sandy mud about 1 meter below the low water mark in the regions south of the central district of Japan Proper. It is a long tower-like roll-shell the surface of which assumes a greyish brown coloration and a goose skin-like aspect with belt around. The material for study was collected from the shore in front of the College in July, 1960.

The spermatogonial metaphase showed 36 chromosomes, among which 1 or more pairs of V- or J-shaped chromosomes are remarkable (Fig. 99).

In the 1st spermatocyte metaphase there occurred 18 bivalents variable in size in considerable degree (Figs. 100~101).

It is therefore evident that the chromosomes number of this species is $2n$, 36 and n , 18.

37 *Contumax kobelti* (DUNKER)

This species inhabits the gravel on the high tidal zone. It is a tower-shaped roll-shell, the external appearance of its surface showing a form of polygon. The specimens for this observation were obtained from the shore behind the College in May, 1961.

In the 1st spermatocyte prophase there were observed 18 bivalents some of which were ring-like in form (Fig. 102). The same number of chromosomes was found to occur in the metaphase plate of the 1st division; the bivalents were dot-like in appearance with a considerable size-variation (Figs. 103~104).

K. Naticidae

On the chromosomes of the species belonging to this family no reports had been published hitherto. The author studied the chromosomes of 1 species as given below.

38. *Neverita (Glossaulax) didyma* (RÖDING)

Concealing itself under the sandy mud below the lowwater mark, this species moves about. It is a semispherical roll-shell the surface of which, assuming a brown or light yellowish brown color, is smooth and lustrous. The specimens collected from the shore in front of the College in June, 1961, served as the material for study.

The 1st spermatocyte metaphase was found to contain 16 comparatively large-sized bivalents which varied in size considerably (Figs. 105~106).

Order Neogastropoda

Reference to the literature indicates that in this order the chromosomes of 12 species represented by 10 genera and 4 families have been reported by the previous authors. Among the three orders dealt with here, the chromosome survey has been most extensively carried out in the order Neogastropoda. The present author has had an opportunity to investigate the chromosomes of 15 species which cover 12 genera and 5 families, such as Muricidae, Pyrenidae, Buccinidae, Nassaridae and Mitridae.

L. Muricidae

Previously, the chromosome studies in this family have made by several authors. LAMS (1934) reported on *Murex brandaris* n, 28 (I). In *M. trunculus*, SCHITZ (1920) reported n, 11 or 12 (I), TUZET (1930) described n, 14 (II) and an X-O sex mechanism, and STAIGER (1950, 1951) claimed n, 35 (I). Further STAIGER (1950, 1951) reported n, 35 in *Oxinebra erinaceus* and n, 18 (I) in *Purpura lapillus*. TUZET (1930) showed n, 14 and an X-O mechanism in *Nassa nitida*. The present author studied the chromosomes of 5 species represented by 3 genera in this family.

39. *Chicoreus asianus* KURODA

This species is distributed over the waters around Japan Proper, Shikoku and Kyushu and lives in the sea at the depth of 10 meter or thereabout. It is a short spindle shaped roll-shell which is furnished with large-sized prickles. The material for study was collected from the shore behind the College in July, 1960.

The spermatogonial metaphase was found to show some pairs of V- or J-shaped chromosomes in addition to dot- and rod-like ones, variable in size (Figs. 107—108). The chromosome number (2n) was found to be 68.

In the 1st spermatocyte metaphase 34 bivalents varying in size were observed (Figs. 109~110).

The chromosome number of this species therefore was determined as 2n, 68 and n, 34.

40. *Bedevina birileffi* (LISCHKE)

This species is a spindle-shaped small sized roll-shell the surface of whose shell assumes a dark green color. The specimens collected from the shore in front of the College in June, 1961 furnished the present material.

In the 1st spermatocyte metaphase 30 bivalents varying in size were observed: 1 to 2 larger bivalents were located in the peripheral zone of the nucleus plate (Figs. 111—112).

41. *Purpura (Mancinella) bronni* DUNKER

This species is distributed over Japan Proper, Shikoku, Kyushu and the Loochoo Islands and below the low water mark. It is a short spindle-shaped roll-shell with a hard shell the surface of which is marked with 4 tubercloids striped and the

interior of whose mouth assumes an orange color. It was known to cause a damage to cultured shell-fish such as oysters. The specimens for observation were collected from the shore behind the College in July, 1960.

The 1st spermatocyte metaphase showed 30 bivalents which varied in size and shape (Figs. 113~114).

42. *Purpura (Mancinella) clavigera* KÜSTER

This species is distributed over Hokkaido, Japan Proper, Shikoku, Kyushu, Korea and China. Though its form resembles that of *P. (M.) bronni* its knotted stripes on the surface are not striking, and the lipsedge assumes dark brown color. This species is known to be injurious to the cultured shell-fish. The material for study was collected from the shore in front of and behind the College in July, 1960 and in June, 1961.

The 1st spermatocyte nucleus plate was composed of dot-shaped bivalents varying in size. The chromosome number was found to be n , 30 (Figs. 115~116).

43. *Purpura (Mancinella) luteostoma* (HULTEN)

This species is distributed from the northeastern districts to Shikoku, Kyushu and the Loochoo Islands and inhabits in the shallows below the low water mark. It is a spindle-shaped roll-shell. Compared with *P. (M.) clavigera*, its tubercles are smaller, assuming a dark brown color and the mouth of the shell a light yellow color. The specimens obtained in the fishing harbor, Yoshimo-cho, Shimonoseki City, in July, 1960, provided the present material.

The spermatogonial metaphase showed several pairs of V- or J-shaped chromosomes in addition to dots or rods variable in size. The chromosome number was observed to be 60 (Figs. 117~118).

In the 1st spermatocyte metaphase there were 30 dot-shaped bivalents which varied in size. There is tendency that large-sized bivalents are located at the periphery of the nucleus plate (Figs. 119~120).

The chromosome number of this species is therefore $2n$, 60 and n , 30.

M. Pyrenidae

In *Columbella rustica* belonging to this family, SCHITZ (1917) reported n , 16 (I), TUZET (1930) n , about 20 and an X-O mechanism, and STAIGER (1950) n , 34 (I). The author investigated the chromosomes of 4 species covering 3 genera.

44. *Pyrene testudinaria tylerae* (GRIFFITH et PIDGEON)

This species is distributed over the regions south of central district of Japan Proper and Shikoku and inhabits below the sea-water's edge. It is a small spindle roll-shell with a conical spiral tower and the surface of the shell speckled with meshes interlaced with zigzag vertical lines. The specimens obtained from the shore behind the College in April, 1961 consisted the material for this study.

In the 1st spermatocyte metaphase there were observable 35 large-sized bivalents (Figs. 121~122). The bivalents and the nucleus plate of this species were the largest

in size, among the species so far dealt with in the present study. The nucleus plate showed 12 to 13 large-sized bivalents located at the periphery of the equatorial plate surrounding the small-sized bivalents in the center.

45. *Pyrene (Mitrella) bicincta* (GOULD)

This species inhabits on the high tidal zone of Japan Proper, Shikoku, Kyushu, Korea and the tropics. It is a spindle shaped fragile small roll-shell. The surface of its shell is smooth rich in variations of speckles. This species is well differentiated between the two sexes by the size and external shape. The material for study was collected from the shore behind the College in March, 1960 and in March, 1961.

In the 1st spermatocyte metaphase there were observed n , 34 dot-like bivalents which varied in size. In the periphery of the nuclear plate there were found several larger bivalents (Figs. 123~124).

Through the course of the meiotic divisions, there was found no special chromosome either in shape or in behavior.

46. *Anachis (Anachia) misera* (SOWERBY)

This species is distributed on the high tidal zone throughout Japan, resembling *P. (M.) bicincta* as mentioned above. It is a small-sized spindle shaped roll-shell with a small tubercle. The specimens collected from the shore behind the College in February and in July, 1960 were used as material.

The spermatogonial metaphase showed 64 chromosomes varying in size from dot to rod. The nuclear plate seemed to contain V- or J-shaped chromosomes, though their number was not certainly determined (Fig. 125).

In the 1st spermatocyte metaphase there were observed 32 dot-shaped bivalents variable in size. The nuclear plate contained more than 10 large-sized bivalents located at the periphery of the nuclear plate, surrounding small-sized ones in the center (Figs. 126~127).

The chromosome number of this species was determined as $2n$, 64 and n , 32.

Though the course of the meiotic divisions, there occurred no special chromosome either in shape or in behavior.

47. *Columbella (Euplica) versicolor* (SOWERBY)

This species is distributed over warm waters south of Japan Proper, Shikoku and Kyushu and inhabits below the low water mark. It is a small short-spindle shaped snail the shell of which is speckled with dots or zigzag lines on the white ground. The material used here was collected from the shore behind the College in July, 1960.

In the spermatogonial metaphase 56 chromosomes were observable. There were found 2 pairs of remarkably large V-shaped chromosomes located at the periphery of the nuclear plate, in addition to certain pairs of small V-shaped ones, and dot- and rod-like elements of varying in size (Fig. 128).

In the 1st spermatocyte metaphase there were observable 28 bivalents. In the peripheral zone of the nuclear plate there were 2 larger bivalents. The other

chromosomes were relatively large in size (Figs. 129~130).

It is apparent that the present form is characterized by the chromosome number of $2n$, 56 and n , 28.

N. Buccinidae

In this family, TUZET (1927) reported on *Pisania maculosa* n , 16 (I) and an X-Y mechanism, while STAGER (1950) 35 (I). Further STAIGER (1950, 1951), working with *Euthria cernea* and *Buccinum undatum*, reported n , 35 (I) in each. Further more, McMURRICH (1896) described n , about 16 in *Fulgur carica*. The present author made observations of the chromosomes in 3 species belonging to 3 genera, as given below.

48. *Babylonia japonica* (REEVE)

Inhabiting the sandy mud below the lowwater mark on the regions south of the central district of Japan Proper, this species is an oval roll-shell with a strong thick shell the surface of which is speckled with many brown circles. The specimens collected from a dry beach of Chofu-cho, Shimonoseki City and from the shore in front of the College in May, 1960 and in July, 1961, respectively were employed as material for this study.

In the 1st spermatocyte metaphase there were observable 36 dot-like bivalents which varied in size in considerable degree (Figs. 131~132).

49. *Pisania (Japeuthria) ferrea* (REEVE)

This species is distributed in all the parts of Japan and is a conical roll-shell either a smooth surfaced shell. The material was collected from the shore behind the College in February, 1960 and in February, 1961.

In the 1st spermatocyte metaphase there were observable 35 dot-shaped bivalents varying in size (Figs. 133~134).

Through the course of the meiotic divisions, there were found no special chromosome either in shape or in behavior.

50. *Cantharus (Pollia) subrubiginosus* (SMITH)

Inhabiting around the low water mark on the shore ranging from the central district of Japan Proper to Kyushu, this species is a long spindle roll-shell with the yellowish brown shell with a constricted structure, and a round bulgy whorl striped markedly lengthwise. The specimens collected from the shore behind the College in April, 1961 constituted the material for this study.

The spermatogonial metaphase was to show 70 chromosomes: they consist of 2 pairs of large V-shaped chromosomes, several pairs of small V-shaped ones, and the remaining dot- to rod-like ones variable in size (Fig. 135).

In the 1st spermatocyte metaphase there were observable 35 bivalents forming the nuclear plate. In the periphery there were distributed several large-sized bivalents (Figs. 136~137).

The chromosome number of this species is therefore $2n$, 70 and n , 35.

O. Nassariidae

There is accessible no paper regarding to the chromosomes of the species belonging to this family. The author examined the chromosomes of 2 species in this family.

51. *Tritia (Hinia) festiva* (POWYS)

This species is inhabiting in the neighborhood of *Zostera* which grows gregariously below the low water mark in all parts of this country and is a light green short-spindle-shaped roll-shell the surface of which is covered with square coarse granules. The specimens employed here were collected from the shore in front of the College in June, 1960, and in June, 1961.

The 1st spermatocyte metaphase plate showed 34 bivalents, each of which was dot-like in appearance, variable in size in considerable degree (Figs. 138~139).

52. *Nassarius (Niotha) livescens* (PHILIPPI)

This species resembles *T. (H.) festiva*, but the surface of the shell assumes a brown coloration and the form of the edge of the shell aperture is different from the other. The material was collected from the shore in front of the College in July, 1960.

In the 1st spermatocyte prophase there occurred 34 chromosomes, some of which were ring-shaped in appearance (Fig. 140).

At the 1st spermatocyte metaphase the nuclear plate showed also 34 bivalents dot-like in outline and variable in size considerably (Figs. 141~142).

P. Mitridae

No paper is accessible in the literature on the chromosomes of the species belonging to this family. The author investigated the chromosomes of 1 species.

53. *Pusia hizenensis* (PILSBRY)

This species inhabits clinging to the reef near the low water mark of the region south of the central district of Japan Proper and Kyushu. It is a long-spindle-shaped roll-shell with the shell having a greyish white band on the greyish brown ground. The specimens collected from the shore behind the College in May, 1951, supplied the material.

In the 1st spermatocyte metaphase there were 30 dot-shaped bivalents varying in size (Figs. 143~144). The larger bivalents were arranged in the peripheral zone of the nuclear plate surrounding the small-sized ones.

Discussion and Conclusion

No defense is need for that the number, structure and behavior of the chromosomes constitute criteria of critical significance for an evolutionarily significant taxonomy. The chromosomes represent the physical basis of the evolutionary mecha-

nism, and therefore any significant alteration in their structure or behavior represents an evolutionary change. The differentiation of species in higher categories has perforce gone hand in hand with the diversification of the chromosomal mechanism. The differences in chromosome number and chromosome shape which frequently distinguish one species from its relatives throw new light on the problems of taxonomy, while the cytological characteristics of whole group of organisms bear the problem on the differentiation of the higher categories of classification and the problems of their evolutionary patterns, plasticity and adaptiveness (WHITE 1954, HUGHES-SCHRADER 1950, 1956, da CUNHA 1960). It is thus evident that cytology will prove a useful tool in elucidating evolutionary relationships in animals, and that cytological data serve as valuable criteria for the diagnosis of species.

Chromosome cytology has contributed to insect systematics in several ways. Generally cytologists are concerned with the cytotaxonomic differences which exist between the related species. These only rarely involve major difference in the genetic mechanism, but often consist of differences in chromosome numbers or in the sizes and shapes of some of the chromosomes in the set. Such differences may sometimes be used to distinguish sibling or cryptic species that cannot be separated at all, or only with difficulty or uncertainty, on external characters. These differences are the result of chromosomal rearrangements which have arisen spontaneously and established themselves in phylogeny. One type of rearrangement is called a centric fusion in which two rod-shaped chromosomes give rise to a V-shaped multiple through translocations, and it leads to a diminution in chromosome numbers. Another is a dissociation or a centric segregation and produces an increase of the chromosome number. Deletions lead to loss of genes, duplications to acquisition of extra genetic material, inversions to changes of gene-sequence within the individual chromosomes, and translocations to interchanges of genetic material between nonhomologous chromosomes (WHITE 1954, 1957).

The present author has studied the chromosomes of 53 species of marine gastropods, with special consideration on the significance of cytotaxonomical characters. The data were given in the foregoing pages. In the following, a discussion will be made on the cytological relationship in allied species from the cytotaxonomical standpoint, though basic data for understanding the taxonomical relation in Gastropoda are very incomplete, since the chromosomes have been studied in only a very small part of the species. The total number of described species of Gastropoda is said to be more or less than 60,000. Out of this total, about 100 or more species have been studied by chromosome cytologists to more or less degree. The chromosome numbers of marine gastropods reported by previous authors are given in Table 1, and those established by the present author are tabulated in Table 3. The following discussion will be devoted in reference to the data given in Table 3.

In the superfamily Pleurotomariacea of the order Archaeogastropoda, the chromosome numbers were reported in 1 species of the Haliotidae, and 3 species of the Fissurellidae, being 4 species in total (Table 3). They are n , 17, 17, 16 and 16, respectively. Viewed from the shape of chromosomes, it seems probable that they

Table 3. The species under investigation and their chromosome numbers.

Species	2 n	n	Note
Order Archaeogastropoda			
Superfamily Pleurotoariacea			
Family Haliotidae			
<i>Haliotis (Sulculus) japonica</i>	34s	17 I, II	
Family Fissurellidae			
<i>Clypidina (Montfortula) picta</i>		17 I	
<i>Macroschisma sinensis</i>		16 I	
<i>M. dilatata</i>	32s	16 I, II	
Superfamily Patellacea			
Family Patellidae			
<i>Cellana toreuma</i>		9 I	
<i>C. eucosmia</i>	18s	9 I	
<i>C. nigrolineata</i>		9 I, II	
Family Acmaeidae			
<i>Patelloida (Collisellina) saccharina lanx</i>		9 I	
<i>P. (Asteracmea) pygmaea</i>		9 I	
<i>P. (A.) lampanicola</i>			
<i>Notoacmea schrenckii</i>		9 I	
<i>N. concinna</i>		9 I	
<i>N. fuscoviridis</i>		9 I	
Superfamily Trochacea		9 I	
Family Trochidae			
Subfamily Margaritinæ			
<i>Stomatella lyrata</i>		21 I	
Subfamily Monodontinae			
<i>Cantharidus callichroa</i>	36s	18 I	
<i>Thalotia japonicus</i>		18 I	
<i>Monodonta labio</i>		18 I, II	
<i>M. neritoides</i>		18 I, II	
<i>Tegula (Chlorostoma) lischkei</i>		18 I	
<i>T. (Omphalius) nigerrima</i>		18 I	
<i>T. (O.) rustica</i>	36s	18 I	
<i>T. (O.) pfeifferi carpenteri</i>		18 I	
Family Turbinidae			
Subfamily Turbininae			
<i>Turbo (Batillus) cornutus</i>		18 I	
<i>Lunella coronata coreenis</i>		18 I	
<i>Astralium haematragum</i>		18 I	
Superfamily Neritacea			
Family Neritidae			
<i>Puperita (Heminerita) japonica</i>	22s	11 I, II	

Order Mesogastropoda		
Superfamily Littorinacea		
Family Littorinidae		
		17 I
		17 I, II
		18 I, II
	30s	15 I, II
Superfamily Cerithiacea		
Family Potamididae		
		18 I
		18 I
		18 I
	36s	18 I, II
	36s	18 I, II
Family Cerithiidae		
Subfamily Cerithiinae		
	36s	18 I
		18 I
Superfamily Naticaea		
Family Naticidae		
		16 I
Order Neogastropoda		
Superfamily Muricacea		
Family Muricidae		
	64s	34
		30 I
		30 I
		30 I
	60s	30 I
Superfamily Buccinacea		
Family Pyrenidae		
		35 I
		34 I
	64s	32 I
	56s	28 I
Family Buccinidae		
		36 I
		35 I
	70s	35 I
Family Nassariidae		
		34 I
		34 I
Superfamily Volutacea		
Family Mitridae		
		30 I

Abbreviations in the columns "2n" and "n"

s : spermatogonium, I : primary spermatocyte and II : secondary spermatocyte

are considered to have been derived from a common ancestral type seemed to be characterized by n , 18. For instance, *M. dilatata* possesses 2 pairs of the V-shaped chromosomes in the spermatogonial metaphase. If the V-chromosomes be assumed as multiples associated of rod-shaped chromosomes two by two, the total number of this species becomes 36 in $2n$ and 18 in n . Further, the diploid complement of this species contains certain numbers of long rod-like chromosomes. In agreement with the view of HIRAI (1956), the author considers that the long rod-type elements may be derived from the fusion of two short rod-like chromosomes. If such a postulate is permitted, it is highly probable that the ancestral type of Archaeogastropoda might have been evolved from a species with a considerably large number of chromosomes. The fact that the shape of bivalents in the 4 species here studied is somewhat angular being variable in size seems to lend support to the above view.

In the superfamily Patellacea, 3 species of the Patellidae and 6 species of the Acmaeidae have been cytologically studied by the author: they have the chromosome number of 9 (n). These 9 species are common in the fact that the metaphase plate shows 7 or 8 bivalents lying in the periphery, surrounding 1 or 2 bivalents in the center. On the basis of the above evidence, it is apparent that they are closely related with each other. Since there is no variation in their chromosome number, it is probable that the differentiation of these species may be to the structural differences of their chromosomes. HANE is of opinion that *N. fuscoviridis* may perhaps be an ecological form of *N. coninna* (personal communication), and the cytological features are alike between the above two forms. It is apparent that the basic number of chromosomes of the Patellidae and Acmaeidae is to be n , 9.

In the family Trochidae, the chromosomes of 9 species were studied (Table 3): all the species studied have n , 18, with an exceptional species, *S. lyrata*, which has n , 21. The bivalents found in the 1st spermatocyte nuclear plate look nearly identical between the 8 species having n , 18 and *S. lyrata* having n , 21. In agreement with the taxonomical consideration, *S. lyrata* may be primitive rather than other 8 species in the course of the evolutionary development. *S. lyrata* is placed in the subfamily Margaritinae, while the other 8 species are put in the subfamily Monodontinae.

Three species of the family Turbinidae are characterized by having n , 18. They are also remarkable for the presence of 3 to 4 comparatively large-sized bivalents located in the periphery. They have calcified operculum. Considered from the above facts it is highly probable that they are closely related and that they may be differentiated due to the structural differences of the chromosomes in the process of evolution. In *T. (B.) cornutus*, there are specimens with or without spines. But such morphological characters have no relation to the structure of the chromosomes.

The family Trochidae and the family Turbinidae as described above are included in the same superfamily. Chromosomally the 8 species of Trochidae studied here, except *S. lyrata*, and 3 species of the Turbinidae are alike in chromosome number, the arrangement of the bivalents in the nuclear plate, and the ratio of large-sized

bivalents and small-sized ones. In the light of the above findings it is probable that the above two families are in close affinity.

The 1st spermatocyte metaphase of *Puperita (Heminerita) japonica* of the family Neritidae, showed bivalents of wide size-variation. There was no evidence for the occurrence of any particular chromosome in behavior, shape and other morphological features. For *Neritina fluvitilis*, a related species, ALEXENKO (1896) reported $2n$, 19s and 20o, and n , 10 (I), and TUZET (1930) gave $2n$, 18s and n , 9 (II). The former author claimed the sex-determining mechanism of an X-O and the latter author showed an X-Y mechanism in males. The results of the present author's observations showed that the number was $2n$, 22 and n , 11. The author, however, observed the chromosome of 1 species only, in this family, and therefore hesitates to conclude any statement on the relationship of the chromosomes and taxonomy.

The chromosome numbers of species belonging to the order Archaeogastropoda were observed and relation between the chromosome number and taxonomy was considered. Important facts to be mentioned here are that all the species studied in this order may be divided, in a broad sense, into main groups—one group of animals is involved in superfamily Patellacea with the chromosome number of n , 9, and the other group is those with the chromosome number of n , 18 or related numbers. That a group of animals whose chromosome number is smaller than 18 belongs to taxonomically lower order, is an evidence unfavorable to the explanation of the evolutionary relationship of this order.

In conclusion, the number of chromosomes of the Archaeogastropoda ranges in variation from 9 to 21 in haploid. It is interesting to see that n , 9 and 18 are very frequently distributed in many species so far studied.

In the family Littorinidae of the order Mesogastropoda, both *Littorina brvicula* and *Littoraria (S.S) strigata* were found to have n , 17. From the same chromosome number found in the two species, they are supposed to be closely related. The taxonomical relation between *Nodilittorina granularis* and *N. picta* cytologically has been not clear cytologically at present, but HABA (1946) described that *N. picta* lies above *N. granularis* on the basis of the study of the radula. Chromosomally, *N. granularis* has n , 18, while *N. picta* has n , 15.

It was observed that 5 species of the family Potamididae had n , 18 in each. Considered from the facts that they all have the same chromosome number at n , 18, and the 1st spermatocyte metaphase contains several large-sized bivalents, chiefly located in the periphery of the plate, it is conceivable that they may be parallel in the course of evolution. The nuclear plate of the 5 species are remarkably different in size, though the difference may probably be due to some technical causes.

In the family Cerithiidae dealt with here, the following 2 species, *Proclava kochi* and *Contumax kobelti*, do not belong to the same genus, but have the same chromosome number of n , 18. By virtue of above fact, it is understood that they may be closely related with one another.

The two families, Potamididae and Cerithiidae, belong to the same superfamily

and have a similarity in the chromosome number. Therefore, it is conceivable that they may be pretty closely related, being parallel in the course of evolution.

In the family Naticidae, only 1 species, *Neverita (Glossaulax) didyma*, proved to have n , 16. INABA (1958) reported the chromosome number of *Amalthea conica* as $2n$, 34s and n , 17 (I, II). The author reported the chromosomes of 7 species of the Cerithiacea as 18 (n). *N. (G.) didyma* mentioned above belongs to the Naticacea, but has an approximate chromosome number. They are in the range of the chromosome number characteristic to the order of Mesogastropoda. In *Crepidula plana*, CONKLIN (1902) reported $2n$, 60 and n , 30. It is evident that from the chromosome numbers, *C. plana* is rather exceptional in the Mesogastropoda. Further, INABA and TANAKA (1953), working with *Semisuleospira libertina* of the family Pleuroceridae reported $2n$, 16 and n , 8.

In conclusion, the chromosome number of the Mesogastropoda varied from 15 to 18 in haploid, so far as 12 species were studied chromosomally by the author. It is remarkable that variation range of the chromosome number in this order is very small. Phylogenetically, the Archaeogastropoda is very distinct by separated from Mesogastropoda. So far as the author's cytological findings are concerned, the two orders are not very far separated, but are of the same strain.

On the chromosomes of order Neogastropoda, STAIGER (1951, 1954) reported the chromosomes of 8 species which were studied in oocytes. Generally speaking, it is very hard to count the oocyte chromosomes accurately as pointed out by MAKINO and NIYAMA (1947) in the study on chromosomes of echinoderms. STAIGER's findings are on the whole approximate to those of the present author.

In the family Muricidae, it was observed that *Chicoreus asianus* had n , 34, while the following 4 species, *Bedequina birileffi*, *Purpura (Mancinella) bronni*, *P. (M.) clavigera* and *P. (M.) luteostoma*, were characterized by n , 30. Viewed from their chromosome numbers and the shape of their shells, one may be allowed to make the statement that they are closely related with each other. Previously, the chromosomes of this family were reported by several authors: in *Murex trunculus*, SCHITZ (1920) reported n , 11—12, TUZET (1930), n , 18 and STAIGER (1950, 1951) n , 35, respectively, with different findings. So far as the findings of the present author are concerned, STAIGER's counts seem to be valid.

In *P. lapillus*, STAIGER (1950, 1951) reported n , 18 and n , 13, in its isolated race, whereas all the species of *Purpura* investigated by the author have n , 30. It is surprising to find a remarkably great difference in number of the chromosomes between the results of STAIGER (1950, 1951) and Those of the present author. Based on the above evidence presented, it is difficult to discuss the taxonomical relation of *Purpura* species studied by STAIGER (1950, 1951) and by the author. In *Nassa nitida*, TUZET (1930) reported n , 14 which, however, is a doubtful finding.

In the family Pyrenidae it was found that the chromosome numbers ranged between n , 35 in *Pyrene testudinaria tylerae* and n , 28 in *Columbella (Euplica) versicolor*. HABA (1945) made a comparative study on the radula in 7 species belonging to this family, and found no particular characteristics. Judging from the

chromosome number, the species of this family seem to be closely related with each other. The author, however, has some interest in finding that *P. t. tylerae* has something different from others in the morphology of chromosomes in the 1st spermatocyte metaphase. On this basis, a suggestion is possible that this species lies on the different branch of the systematic tree of evolution from others, though this view may not be agreed by taxonomists.

In *C. rustica*, SCHITZ (1917) reported $n, 16$, TUZET (1930), n , about 20 and STAIGER (1950) $n, 34$: thus there are considerable discrepancies in the number of chromosomes of this species by the authors. As shown above, the results of the present author regarding 4 species in this family render possible $n, 34$ reported by STAIGER as valid.

In the family Buccinidae, it was observed that *Babylonia japonica* had $n, 36$, and *Pisania (Japeuthria) ferrea* and *Cantharus (Pollia) subrubiginosus* possessed an identical number of $n, 35$. Further, a similarity in shape of the bivalents supports the view that they are closely related. In *Pisania maculosa*, TUZET (1927) reported $n, 16$ while STAIGER (1950) $n, 35$; the present author wonders that their materials are different species. Further, STAIGER (1950) reported that *Euthria corna* and *Buccinum undatum* have $n, 35$. Comparison of the author's findings on the same genus and STAIGER's ones revealed that they are characterized by an identical chromosome complement. Concerning *Fulgur carica* McMURRICH (1896) reported about 16 in n which, in reference to the modern authors' findings, is doubtful.

In the family Nassaridae the author has observed that *Tritia (Hinia) festiva* and *Nassarus (Niotha) livesceus* are characterized in each by $n, 34$: it is apparent that they are considerably related with one another, considered cytologically.

In the family Mitridae, the author has found that *Pusia hizenensis* possesses 30 chromosomes in haploid. On this basis, this species is characterized by the chromosome complex specific to this order. In conclusion, the present author has studied the chromosomes of 15 species in the order Neogastropoda.

It is evident that the number shows a rather wide range of variation from 28 to 36. The frequent numbers are 30 and 34, the former being found in 5 species, and the latter in 4 species. From the data reported by previous authors and by the present author, it seems probable that the basic number of this order lies in 35 to 36. Formerly the orders Mesogastropoda and Neogastropoda are thought to be one order by some systematists. In the light of the chromosomal findings, the two orders should be clearly divided.

The sex-determining mechanism of the Gastropoda is the subject which merits special attention of many investigators in the past and now, and discrepant claims have been afforded by several authors. Some authors expressed the view of an X-O mechanism, while other an X-Y. According to EGAMI (1949), the sexuality of the Gastropoda is of very complicated one. TUZET (1930) reported on 4 species an X-O mechanism and an X-Y mechanism in one species. ALEXENKO (1896) observed an X-O mechanism in one species (for details, see Table 1). The present author has

observed the chromosomes of 53 species of the Gastropoda. So far as the findings of the author are dealt with there is no evidence to show the existence of any particular chromosome which is heterotypic in behavior and morphology characteristic to the sex chromosome of other animals. In reference to the figures presented in the paper of TUZET (1930), it becomes evident that the X chromosome designated by him is inconstant in outline by cells, and further its orientation in meta-anaphase spindle is extremely variable. To the author's view, the chromosome indicated as X by TUZET may be the ordinary bivalent which is mechanically displaced through the effect of fixation. INABA (1958) has reported that in *Amalthea conica* a V-shaped bivalent derived from a pair of V-shaped spermatogonial chromosomes splits slightly behind others, but he did not regard it as the X-element. So far as the findings of the present author and INABA (1958) there is no special element morphologically distinguishable from others.

Summary

The present investigation deals with the chromosomes in male germ cells of 53 species of marine gastropods which are represented by 16 families and three orders, mostly carried out with the water pre-treatment squash technique. The species under study and their chromosome numbers established are given summarizingly in Table 3. The descriptions of the data were presented according to the arrangement of current taxonomy.

The results of the author's comparative study of the chromosomes in 53 species and those reported by previous investigators were discussed from standpoints of the cytotaxonomical and phylogenetical significance of cytological characters.

The number of chromosomes of the order Archaeogastropoda ranges in variation from 9 to 21 in haploid, so far as the results derived from 26 species are concerned. It is interesting to find that n , 9 and 18 are very frequent in the species so far studied. In the light of the chromosome conditions obtained here, the order Archaeogastropoda is to be divided into two main groups: one is represented by the species having n , 9 and the other by members having n , 18.

In the order Mesogastropoda, the chromosomes of 12 species were investigated. It was found that the number of chromosomes varied from 15 to 18 in haploid. Remarkable is that the range of variation in the chromosome number is very small in this order. So far as the cytological data obtained here are dealt with, the Mesogastropoda is not very far distantly separated from the Archaeogastropoda in the phylogenetical system, contrary to the common view of systematists.

Based on the study of chromosomes in 15 species of the order Neogastropoda, it becomes evident that the number of chromosomes shows a rather wide range of variation from 28 to 36. The frequent numbers are 30 and 34. From the data by previous authors and by the present author, it is probable that the basic number of this order lies between 35 and 36. Formerly, the orders, Neogastropoda and

Mesogastropoda are included in one and the same order by some systematists. In view of the chromosomal findings, the two orders may be of two separated ones.

So far as the findings by the present author are concerned, there is no evidence showing the presence of particular chromosome which is heterotypic in behavior and morphology characteristic to the sex-chromosome observed in other animals. According to the author's view, the X-element designated by some previous authors is no other than the chromosome which is mechanically displaced unusually by the influence of technical procedures.

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P L A T E

PLATE I

- Figs. 1—3. *Haliotis (Sulculus) japonica*. 1, spermatogonium. 2, first metaphase. 3, second metaphase.
- Figs. 4—5. *Clypidina (Montfortula) picta*. first metaphase.
- Figs. 6—7. *Macroschisma sinensis*. first metaphase.
- Figs. 8—11. *Macroschisma dilatata*. 8, spermatogonium. 9—10, first metaphase. 11, second metaphase.

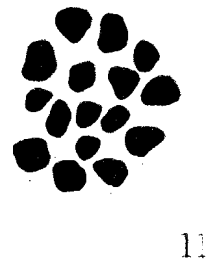
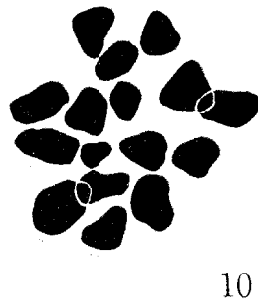
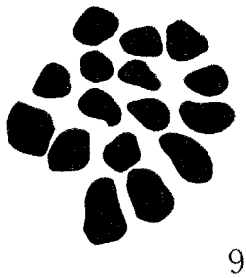
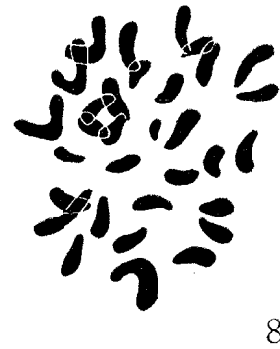
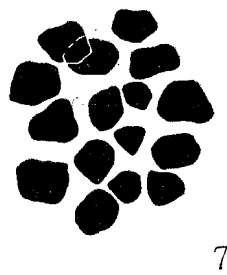
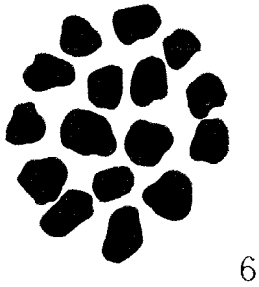
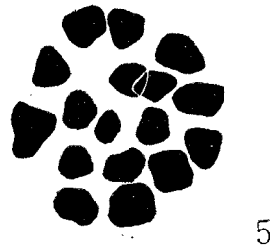
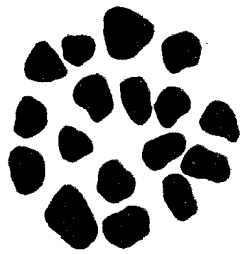
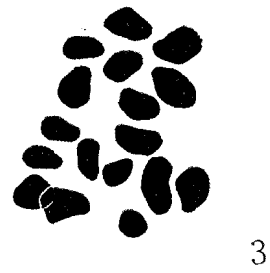
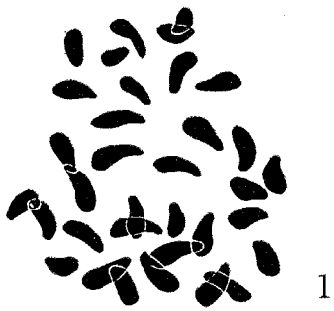


PLATE II

- Figs. 12—13. *Cellana toreuma*. first metaphase.
- Figs. 14—17. *Cellana eucosmia*. 14—15, spermatogonium. 16—17, first metaphase.
- Figs. 18—21. *Cellana nigrolineata*. 18—19, first metaphase. 20—21, second metaphase.
- Figs. 22—23. *Patelloida (Collisellina) saccharina lanx*. first metaphase.
- Figs. 24—25. *Patelloida (Asteracmea) pygmaea*. first metaphase.
- Figs. 26—27. *Patelloida (Asteracmea) lampanicola*. first metaphase.
- Figs. 28—29. *Notoacmea schrenckii*. first metaphase.
- Figs. 30—31. *Notoacmea concinna*. first metaphase.
- Figs. 32—33. *Notoacmea fuscoviridis*. first metaphase.
- Figs. 34—35. *Stomatella lyrata*. first metaphase.
- Figs. 36—38. *Cantharidus callichroa*. 36, spermatogonium. 37—38, first metaphase.

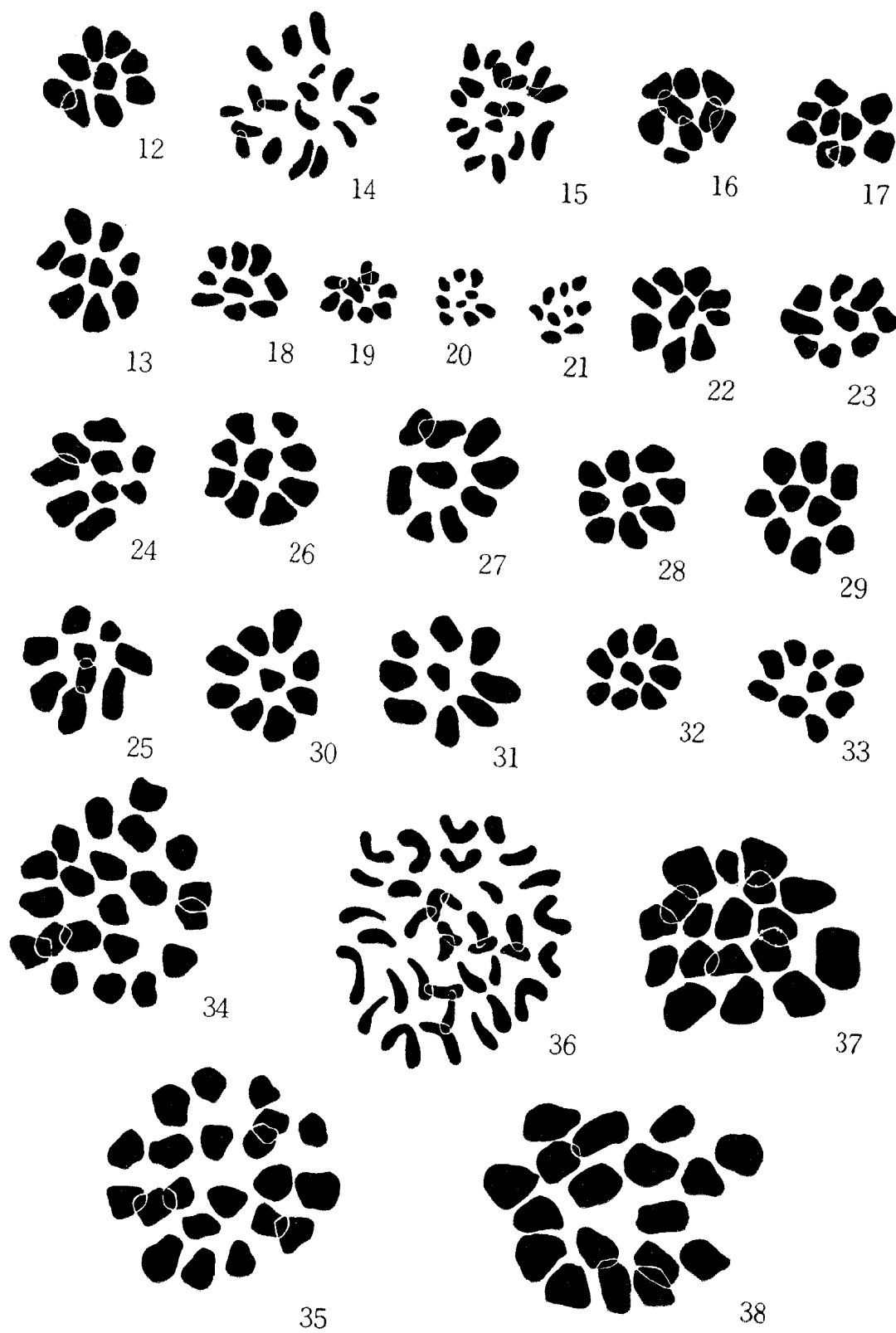


PLATE III

- Figs. 39—40. *Thalotia japonicus*. first metaphase.
- Figs. 41—44. *Monodonta labio*. 41—42, first metaphase. 43—44, second metaphase.
- Figs. 45—46. *Monodonta neritoides*. first metaphase.
- Figs. 47—48. *Tegula (Chlorostoma) lischkei*. first metaphase.
- Figs. 49—50. *Tegula (Omphalius) nigerrima*. first metaphase.
- Figs. 51—53. *Tegula (Omphalius) rustica*. 51, spermatogonium. 52—53, first metaphase.
- Figs. 54—57. *Tegula (Omphalius) pfeifferi carpenteri*. 54—55, first prophase. 56—57, first metaphase.

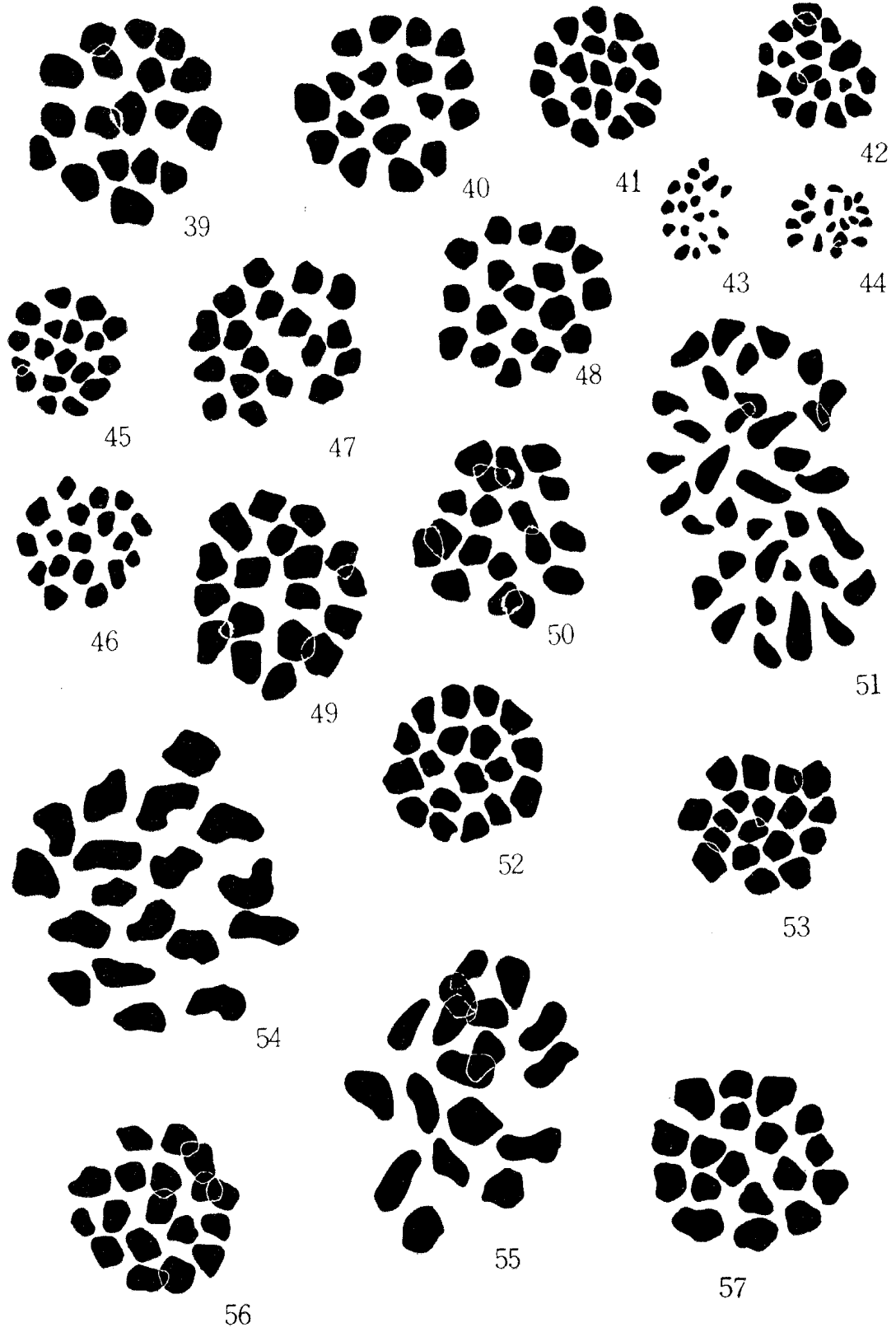


PLATE IV

- Figs. 58—61. *Turbo (Batillus) cornutus*. first metaphase. 58—59, non-spine type. 60—61, spine type.
- Figs. 62—63. *Lunella coronata coreenis*. first metaphase.
- Figs. 64—65. *Astraliium haematragum*. first metaphase.
- Figs. 66—70. *Puperita (Heminerita) japonica*. 66—67, spermatogonium. 68—69, first metaphase. 70, second metaphase.
- Figs. 71—72. *Littorina brevicula*. first metaphase.
- Figs. 73—75. *Littoraria (S.S.) strigata*. 73—74, first metaphase. 75, second metaphase.
- Figs. 76—78. *Nodilittorina granularis*. 76—77, first metaphase. 78, second metaphase.

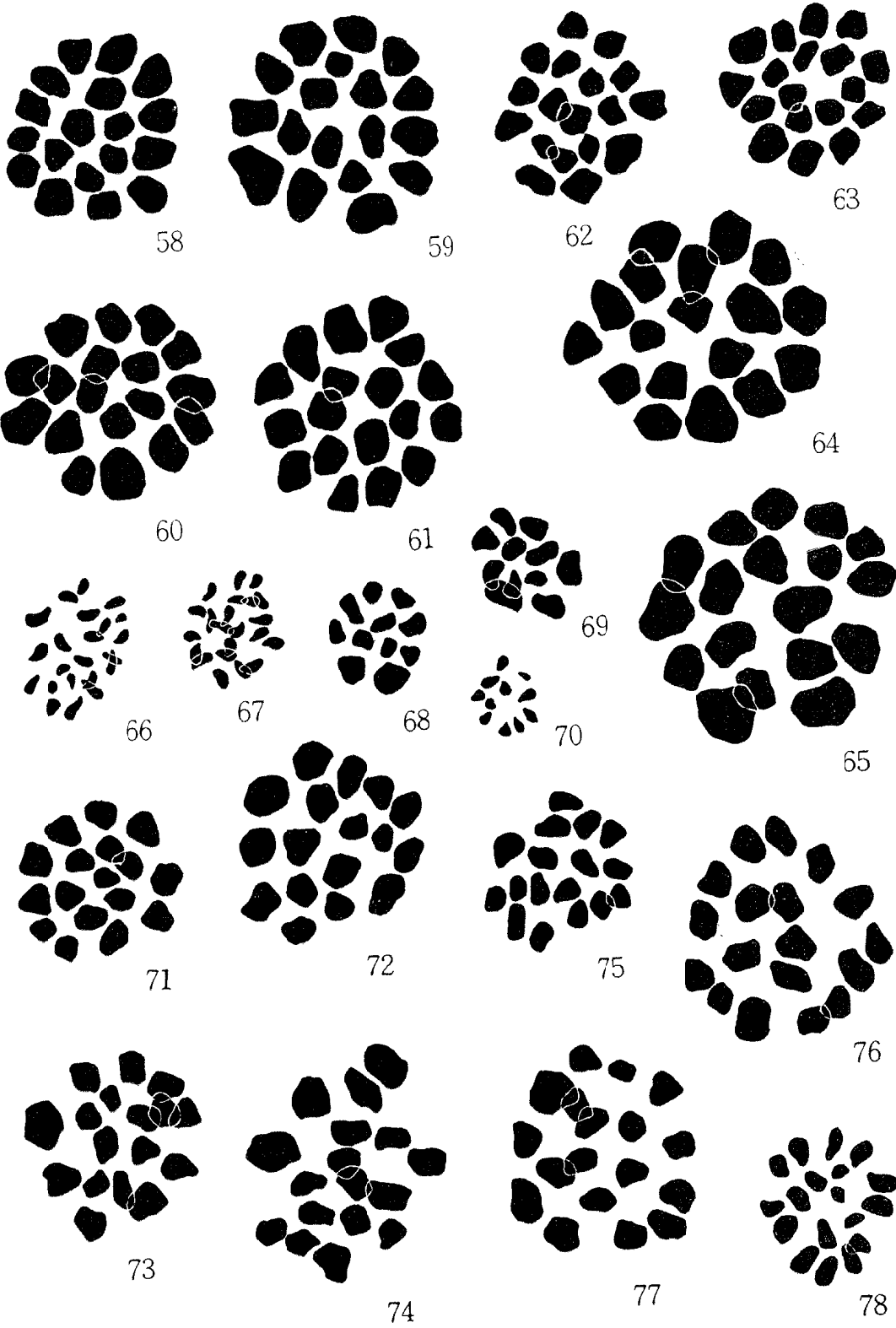


PLATE V

- Figs. 79—82. *Nodilittorina picta*. 79, spermatogonium. 80—81, first metaphase. 82, second metaphase.
- Figs. 83—84. *Cerithidea (Cerithidea) rhizophorarum*. first metaphase.
- Figs. 85—86. *Cerithidea (Cerithideopsilla) cingulata*. first metaphase.
- Figs. 87—88. *Cerithidea (Cerithideopsilla) djadjariensis*. first metaphase.

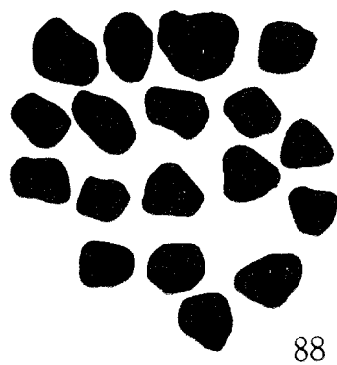
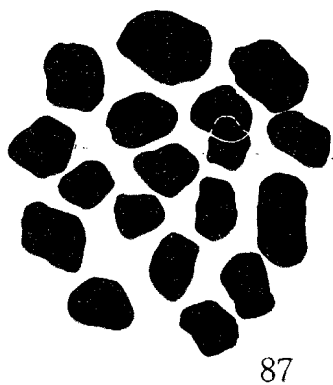
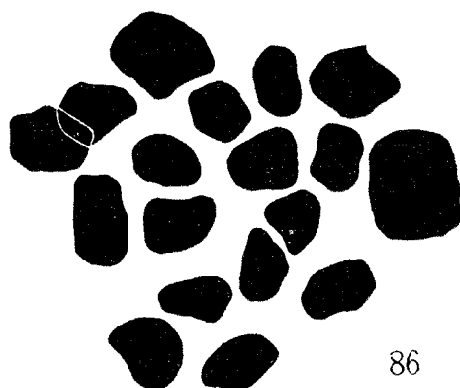
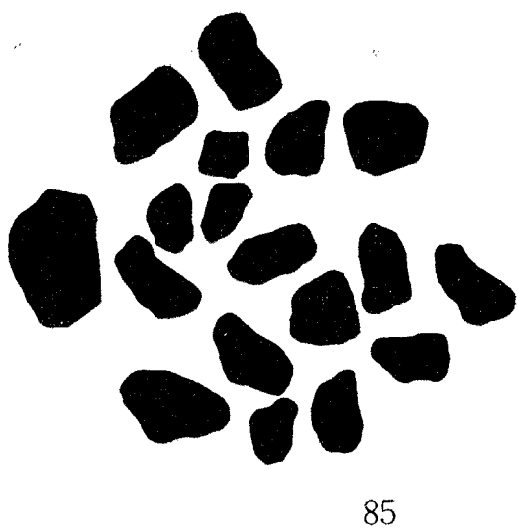
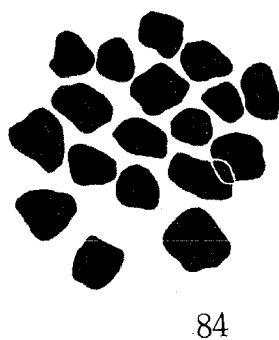
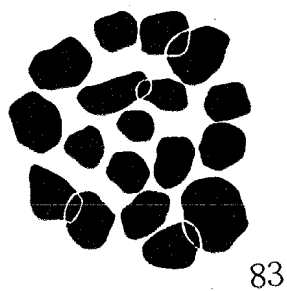
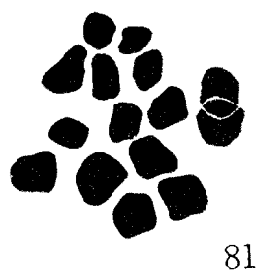
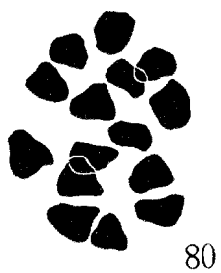
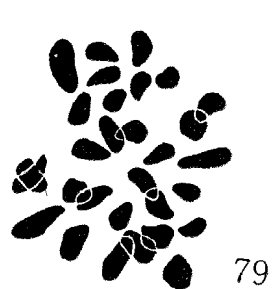


PLATE VI

- Figs. 89—93. *Batillaria zonalis*. 89, spermatogonium. 90—91, first metaphase. 92—93, second metaphase.
- Figs. 94—98. *Batillaria multiformis*. 94, spermatogonium. 95—96, first metaphase. 97—98, second metaphase.
- Figs. 99—101. *Proclava kochi*. 99, spermatogonium.
100—101, first metaphase.
- Figs. 102—104. *Contumax kobelti*. 102, first prophase. 103—104, first metaphase.

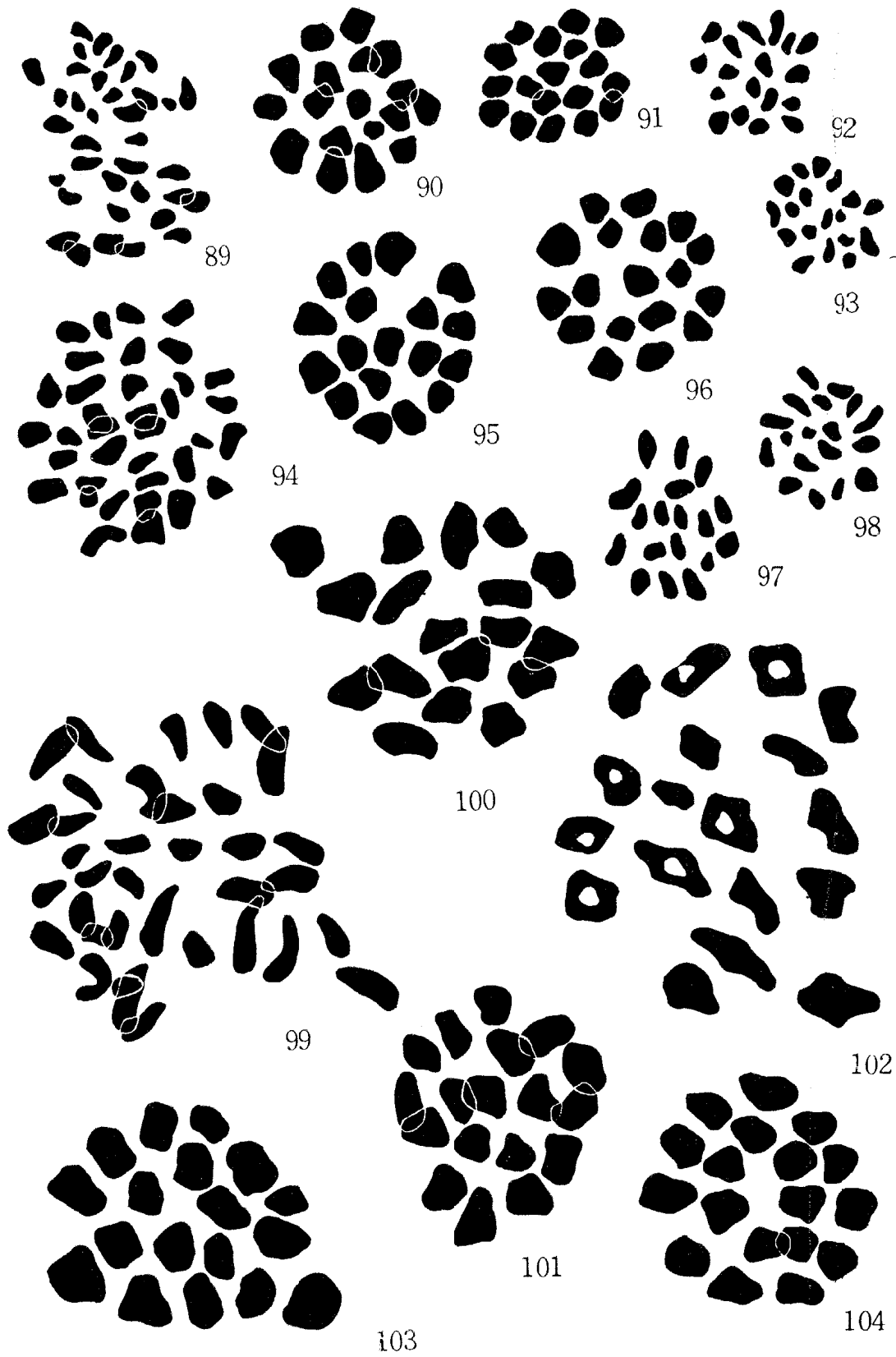


PLATE VII

Figs. 105—106. *Neverita (Glossaulax) didyma*. first metaphase.

Figs. 107—110. *Chicoreus asianus*. 107—108, spermatogonium.
109—110, first metaphase.

Figs. 111—112. *Bedequina birileffi*. first metaphase.

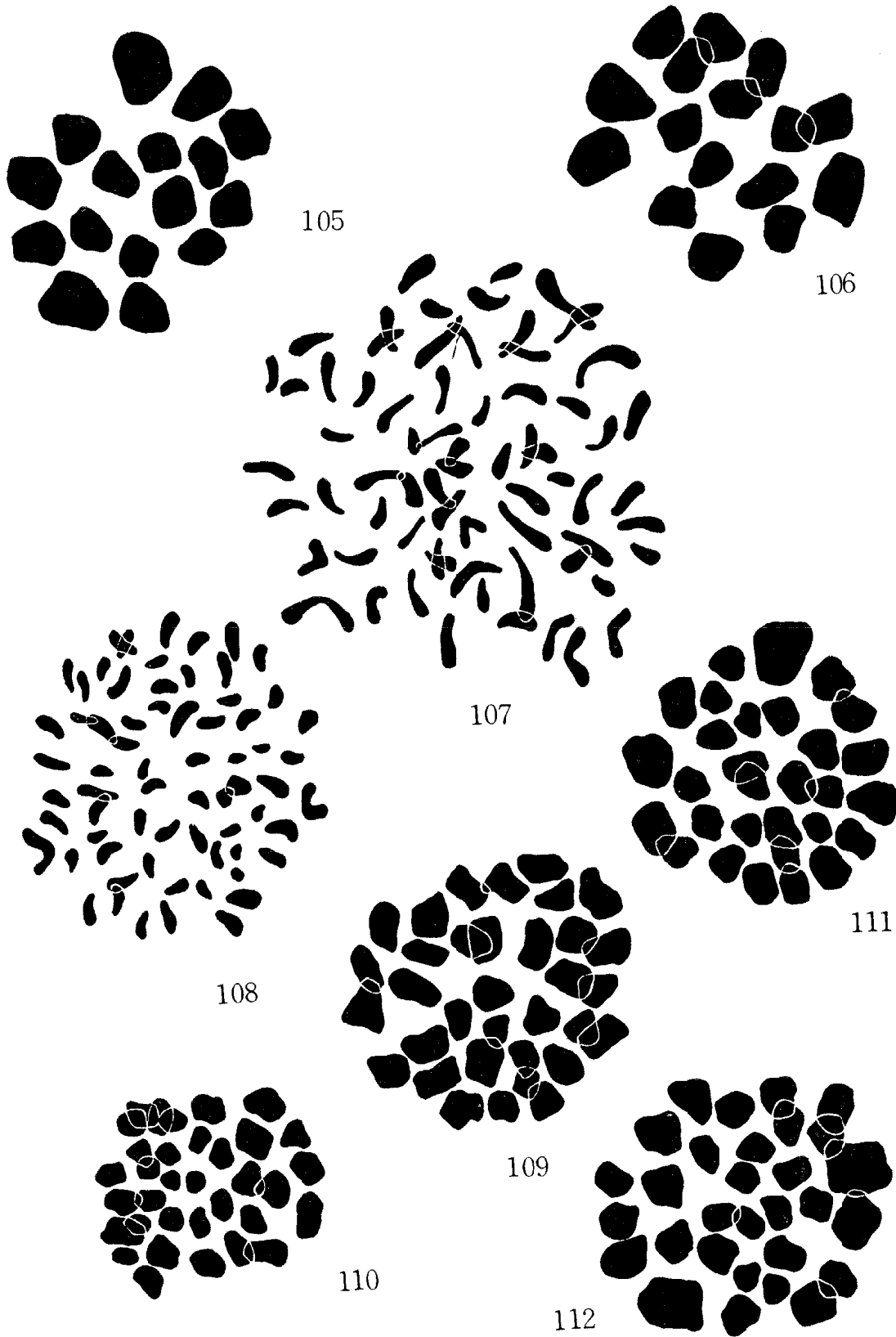


PLATE VIII

Figs. 113—114. *Purpura (Mancinella) bronni*. first metaphase.

Figs. 115—116. *Purpura (Mancinella) clavigera*. first metaphase.

Figs. 117—120. *Purpura (Mancinella) luteostoma*. 117—118,
spermatogonium. 119—120, first metaphase.

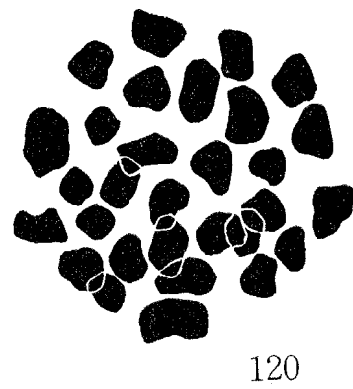
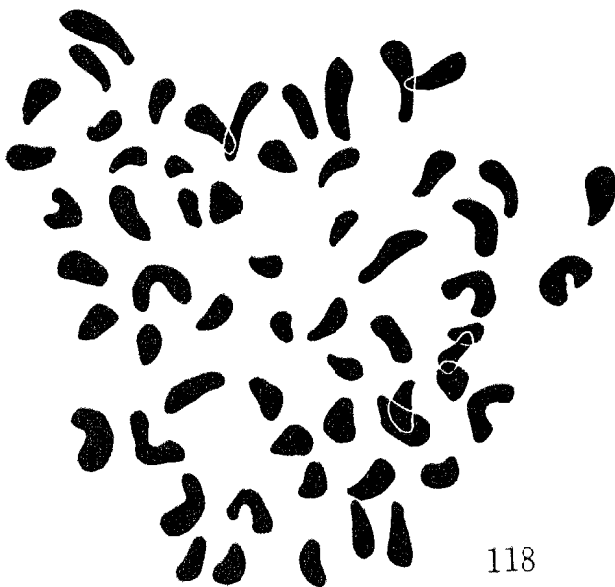
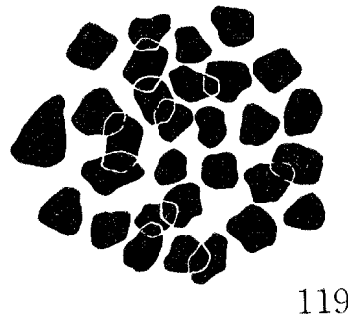
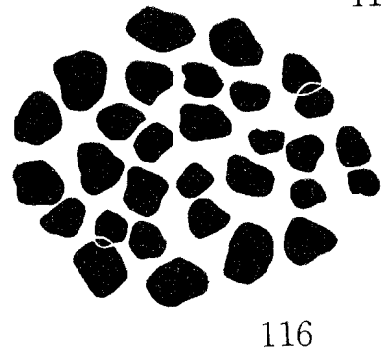
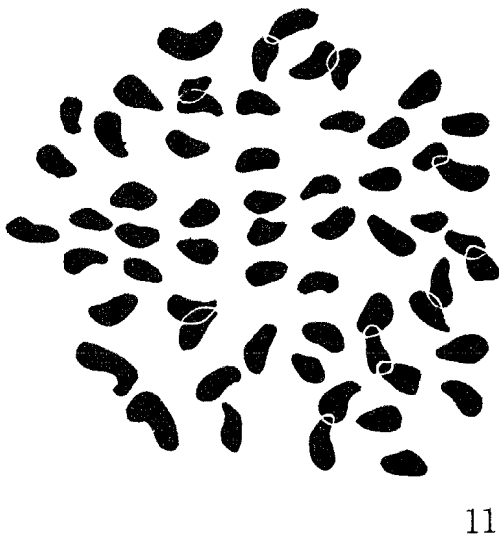
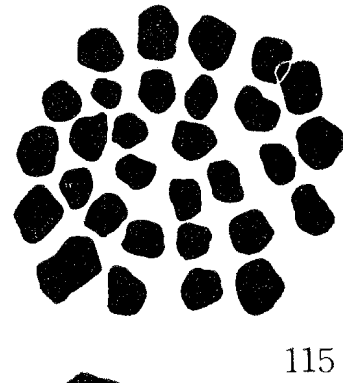
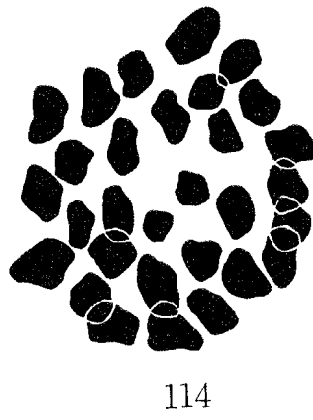
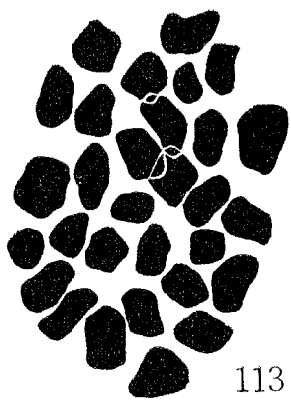
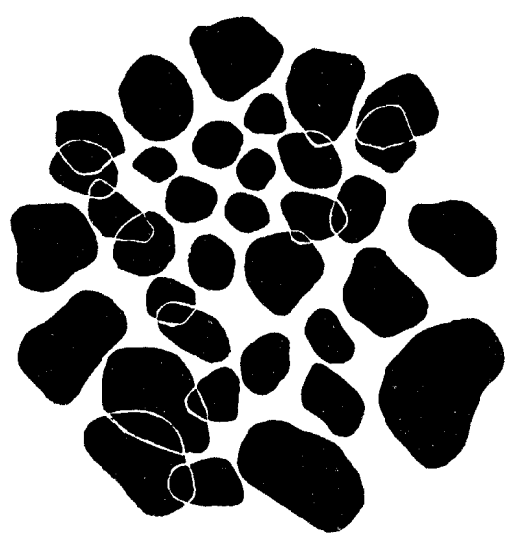


PLATE IX

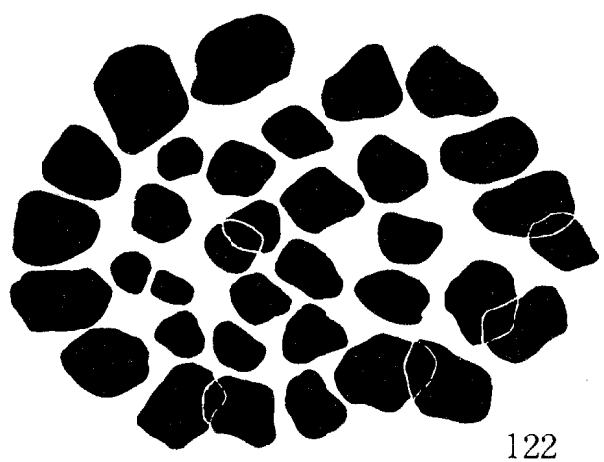
Figs. 121—122. *Pyrene testudinaria tylerae*. first metaphase.

Figs. 123—124. *Pyrene (Mitrella) bicincta*. first metaphase.

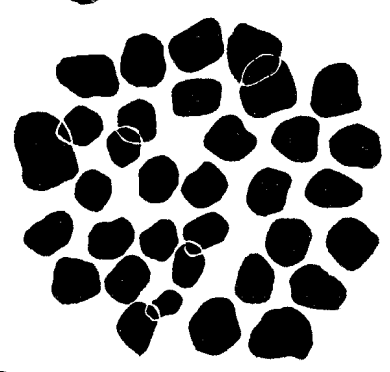
Figs. 125—127. *Anachis (Anachis) misera*. 125, spermatogonium.
126—127, first metaphase.



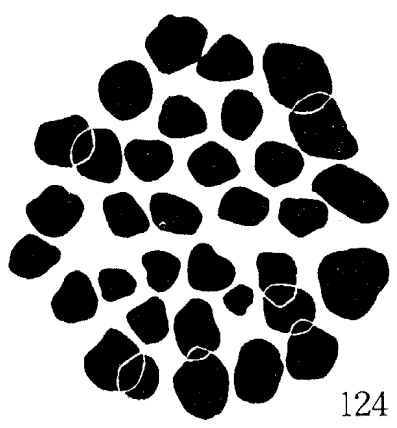
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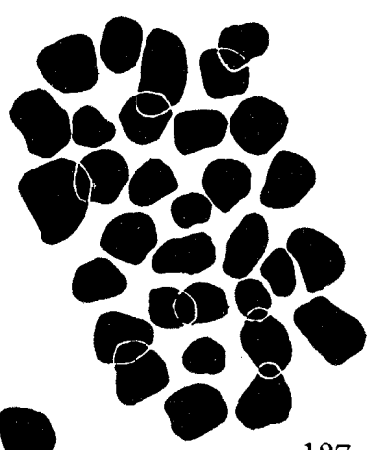
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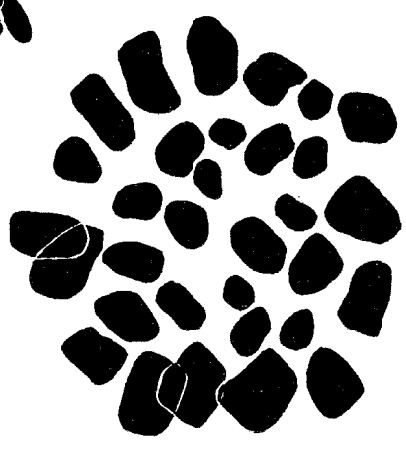
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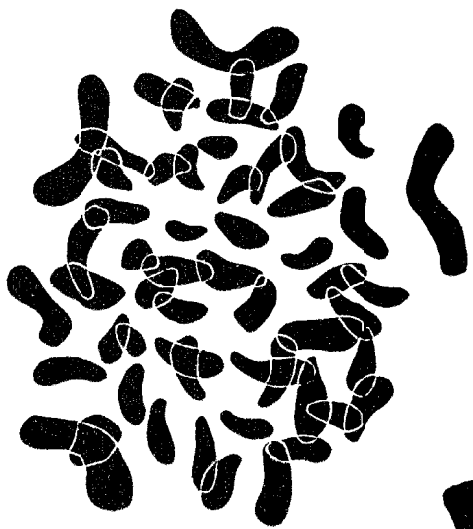
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PLATE X

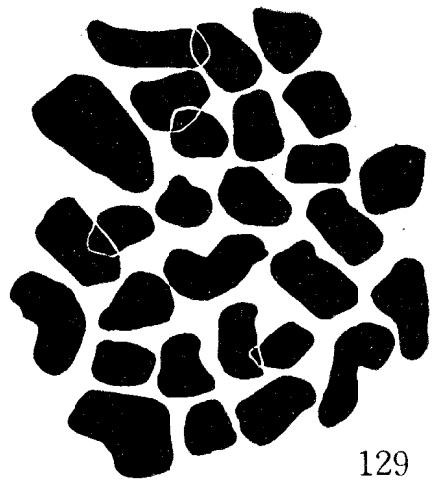
Figs. 128—130. *Columbella (Euplica) versicolor*. 128, spermatogonium.
129—130, first metaphase.

Figs. 131—132. *Babylonia japonica*. First metaphase.

Figs. 133—134. *Pisania (Japeuthria) ferrea*. First metaphase.



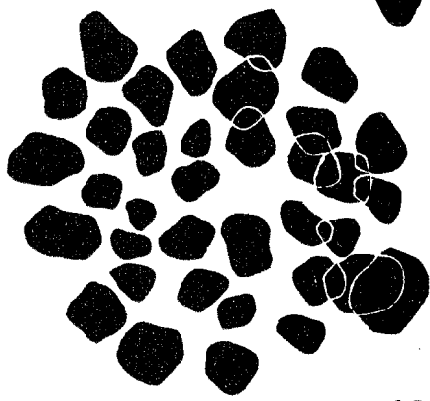
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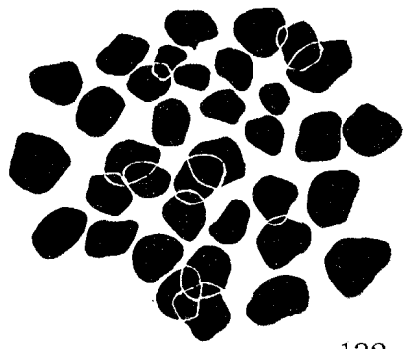
129



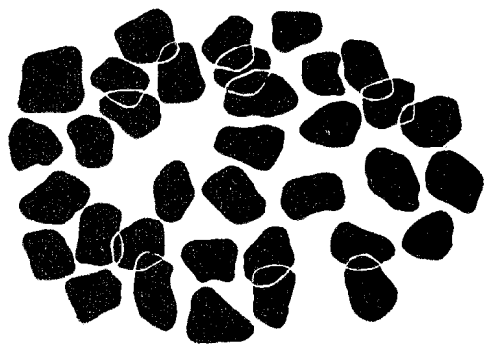
130



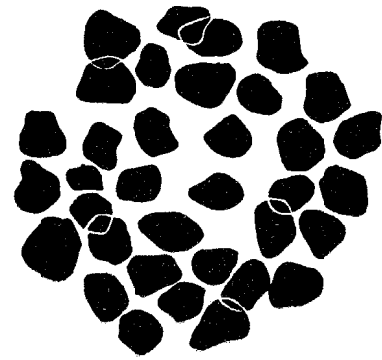
131



132



133



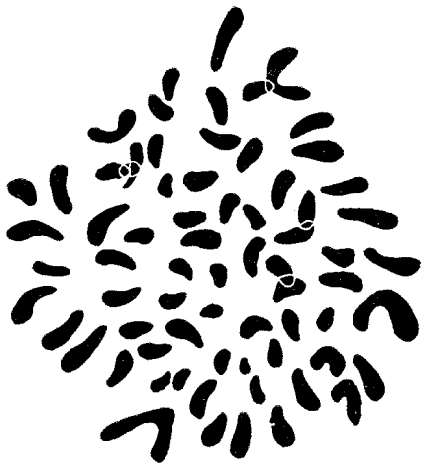
134

PLATE XI

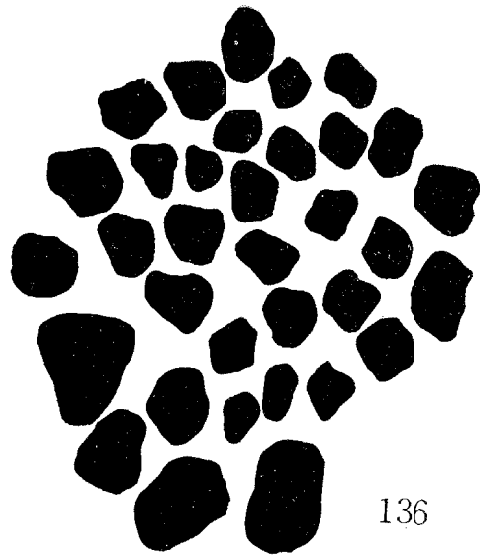
Figs. 135—137. *Cantharus (Pollia) subrubiginosus*. 135, spermatogonium.

136—137, first metaphase.

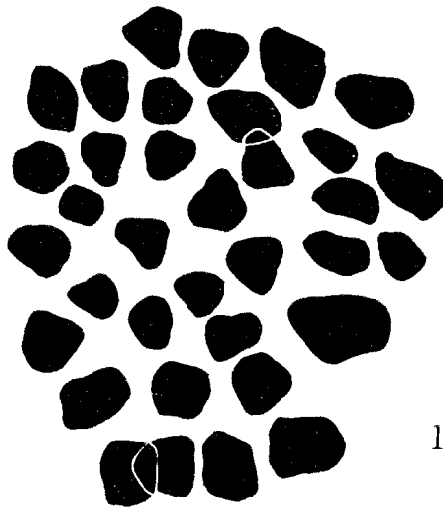
Figs. 138—139. *Tritia (Hinia) festiva*. first metaphase.



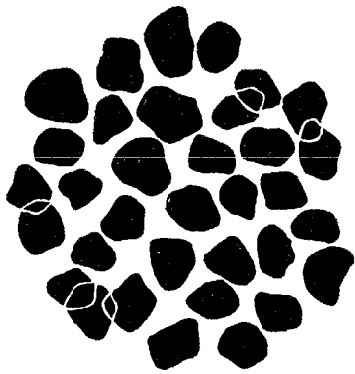
135



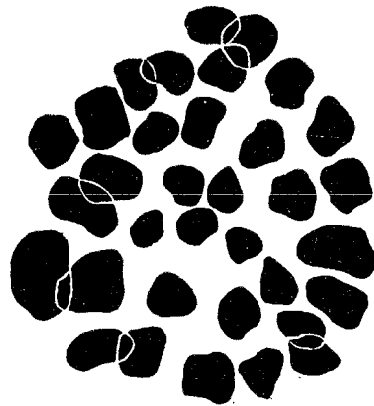
136



137



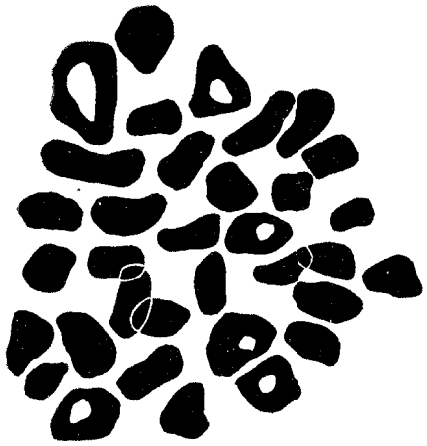
138



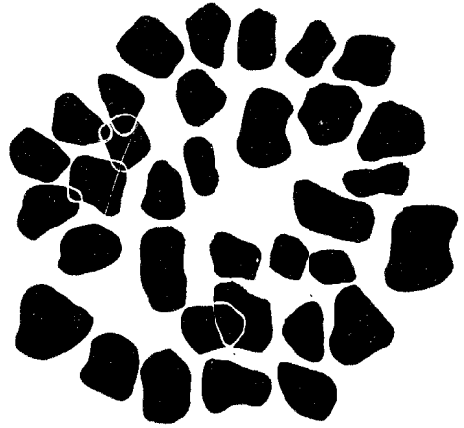
139

PLATE XII

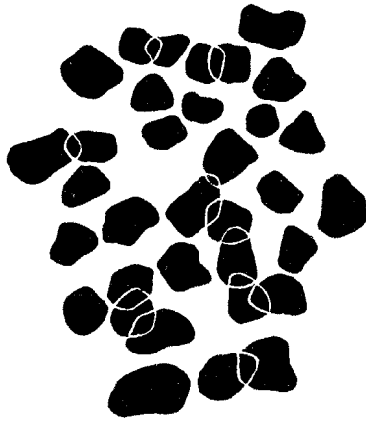
- Figs. 140—142. *Nassarius (Niotha) livescens*. 140, first prophase.
141—142, first metaphase.
- Figs. 143—144. *Pusia hizenensis*. first metaphase.



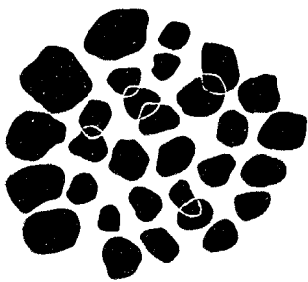
140



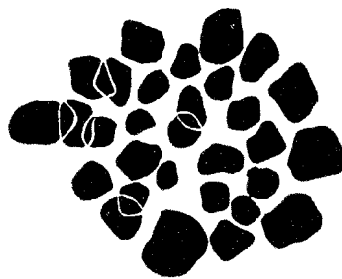
141



142



143



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