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# Genetics and Morphology of 13 Albino Stocks in the *Rana nigromaculata* Group

By

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(With 29 Text-figures and 5 Plates)

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## INTRODUCTION

The discovery of albinos in the Japanese common pond frog, *Rana nigromaculata*, was first made in 1940 by KAWAMURA in his experiments on artificial parthenogenesis (KAWAMURA, 1949). By pricking eggs taken out of the uterus of a female with a needle, four albinic and two normal tadpoles were parthenogenetically obtained. Three of the albinos and the two normal tadpoles completed metamorphosis. The external characters, hypophysis, thyroid gland and sex of these albinos have been examined by TOKUNAGA (1949). The eyes of these albinos were black. As the diploidy of the frogs obtained by artificial parthenogenesis is due to duplication of chromosomes before the first cleavages, she considered that the mother of the albinos was heterozygous for a recessive albino gene.

A mature female albino of *Rana nigromaculata* was collected in 1952 from Osaka. The external and internal characters of this albino were anatomically and histologically observed by TANEGASHIMA (1957). A juvenile albinic frog probably belonging to this species was collected in 1963 from Handa, Aichi Prefecture, and reported by KONDO (1963). The eyes of these two albinos were red.

Although a fairly large number of albinos must have been observed around rice fields by farmers and others during a long period of time in Japan, they were not preserved or recorded. The present authors noticed that albinos are not always of extremely rare occurrence in the field. Since about twenty years ago, many albinos of various anuran species have been gathered mainly from Hiroshima Prefecture and its neighborhood. They and their offspring have been carefully reared in our laboratory.

The present authors (1977) have reported on ten albino stocks of *Hyla arborea japonica* collected from the field. In this report, they clarified for the first time in vertebrates that there are three different loci for albino genes and the ten albino stocks are classified into three groups on the basis of differences in albino locus. Recently, the present authors could demonstrate the presence of five different loci for albinos mainly in *Rana nigromaculata*.

The results of genetic and morphologic studies on these albinos in the *Rana nigromaculata* group will be presented in this paper.

## MATERIALS AND METHODS

### *I. Materials*

During the 13 years from 1967 to 1979, a total of 13 albino stocks were obtained in *Rana nigromaculata* HALLOWELL and *Rana brevipoda* ITO. Three of them were induced by irradiation of gametes in *Rana nigromaculata*, while the remaining 10 were collected from the field (Fig. 1).

1. Ex–145 stock (Ex)

This stock was derived from a *Rana nigromaculata* egg irradiated with 145 rads of X-rays in the breeding season of 1967 (NISHIOKA, 1977).

2. Sn–50 stock (Sn I)

This stock originated from a *Rana nigromaculata* spermatozoon irradiated with 50 rads of neutrons in the breeding season of 1967 (NISHIOKA, 1977).

3. Sn–130 stock (Sn II)

This stock was derived from a *Rana nigromaculata* spermatozoon irradiated with 130 rads of neutrons in the breeding season of 1967 (NISHIOKA, 1977).

4. *Rana brevipoda* stock (Br)

A mature male *Rana brevipoda* heterozygous for an albino gene was collected from Konko-cho, Okayama Prefecture in the breeding season of 1970. From this male an albino stock was established.

5. Yamaguchi I stock (Ym I)

Twelve albinic tadpoles of *Rana nigromaculata* were collected in June of 1973 by Professor SAMBUICHI, Faculty of Science, Yamaguchi University, from a rice field in Yabara, Yamaguchi Prefecture. These tadpoles were reared in his laboratory and became froglets, being 3～4 cm in total length, in December of the same year. Nine of them were given us and then reared in our laboratory

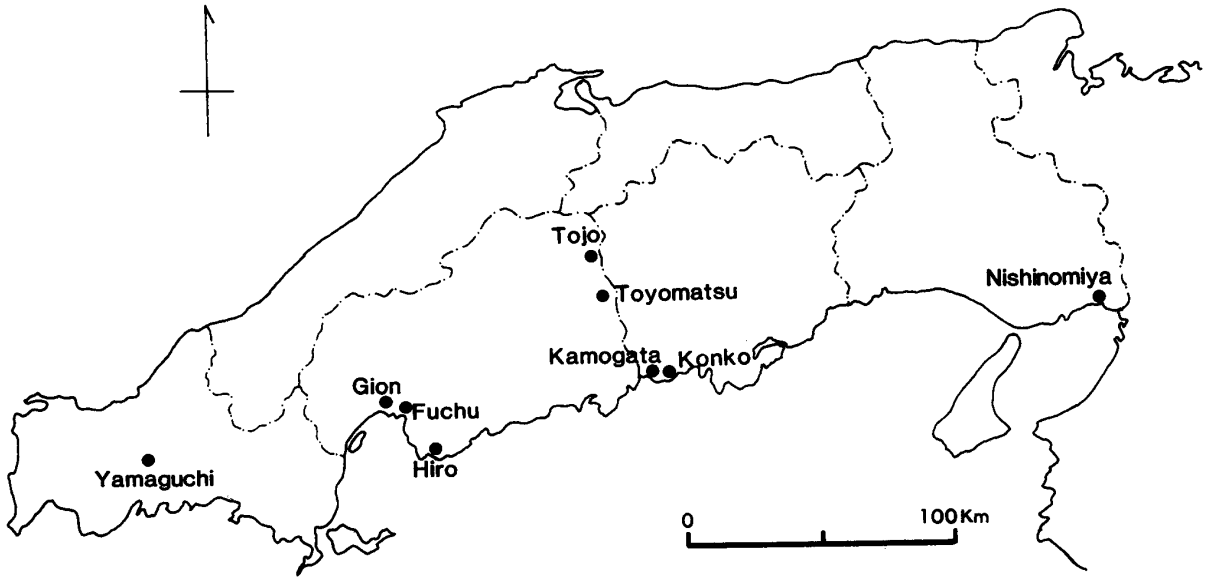


Fig. 1. Collecting stations of albino mutants in the *Rana nigromaculata* group.

until sexual maturity. Four were females and five were males.

6. Kamogata stock (Km)

An albinic froglet of *Rana nigromaculata* was collected in May of 1974 by Mr. Shirane, Faculty of Integrated Arts and Sciences, Hiroshima University, from Kamogata-cho, Asakuchi-gun, Okayama Prefecture. This froglet was 3 cm in body length and given to us at once. In attaining sexual maturity, it was a female.

7. Gion stock (Go)

This stock was derived from a mature female *Rana nigromaculata* collected in October of 1974 from Gion-cho, Hiroshima City. This female was heterozygous for an albino gene.

8. Tojo stock (Tj)

A total of 179 albinic tadpoles of *Rana nigromaculata* were collected in June of 1974 from Myoga's and Inoue's rice fields in Tojo-cho, Hiba-gun, Hiroshima Prefecture. All of them were reared until sexual maturity.

9. Fuchu stock (Fc)

An albinic froglet of *Rana nigromaculata* immediately after metamorphosis was collected in July of 1974 by Mrs. Kashiwagi from Fuchu-cho near Hiroshima City. In attaining sexual maturity, it was a female.

10. Toyomatsu stock (Ty)

A total of 89 albinic tadpoles of *Rana nigromaculata* were collected in June of 1977 from Mr. Uchihi's rice field in Toyomatsu-mura, Jinseki-gun, Hiroshima Prefecture. All of them were reared in our laboratory until sexual maturity.

11. Nishinomiya stock (Ns)

Forty-two albinic tadpoles of *Rana nigromaculata* were collected in June of 1977 by Mr. Wakana, an undergraduate of Kwansei Gakuin University, from a rice field in Kitanatsugi-cho, Nishinomiya City. All of them were reared in our

laboratory until sexual maturity.

12. Yamaguchi II stock (YM II)

In the breeding season of 1977, about a dozen of albinic tadpoles were obtained from a mating between a wild-type female and a wild-type male of *Rana nigromaculata* collected by Professor SAMBUICHI, Faculty of Science, Yamaguchi University, from Sufu-cho, Yamaguchi City. These tadpoles became froglets, being 3～4 cm in body length, in January of 1978. Five of them were given us and thereafter reared in our laboratory until sexual maturity. Three were females and two were males.

13. Hiro stock (HR)

A total of 102 albinic tadpoles were collected in May of 1979 from Mr. KAMIFUKUURA's rice fields in Hiro-machi, Kure City. All of them were reared until sexual maturity.

## *II. Methods*

In each of the 13 albino stocks, matings were made between female and male albinos or between albinos and wild-type frogs in order to obtain albinic or heterozygous offspring. From matings betwcen albinos and heterozygous individuals or between heterozygous females and males, albinos were produced. Albinos were also produced by the method of diploid gynogenesis.

The method employed was as follows. Eggs of *Rana nigromaculata* were inseminated with UV-irradiated sperm of *Rana clamitans* and refrigerated for 2.5～3 hours at 1～2°C, 20～25 minutes after insemination. The UV-irradiation of sperm was done by exposing a small quantity of sperm suspension to a mercury lamp (GUL-5·J Type, Toshiba Co.) operated at 125 mA for 2 minutes at a distance of 20 cm from the lamp. The UV-ray was 2537 Å in main wave length and 2400 erg/cm$^2$/sec in energy level. As the nucleus of each spermatozoon is inactivated by UV-irradiation, the egg inseminated with such a spermatozoon develops gynogenetically after pseudofertilization. When the egg is refrigerated after insemination, it becomes a diploid owing to retention of the second polar body nucleus.

Ovulation was accelerated by injecting suspension of bullfrog pituitaries into the abdominal cavity. Fertilization was always performed artificially. Tadpoles were fed on boiled spinach or chard, while metamorphosed frogs were fed on crickets.

For electron microscopic observation, a piece of skin taken from the dorsal surface of the frog was put in 3% glutaraldehyde in 0.1 M phosphate buffer (pH 7.4) for prefixation, cut into smaller pieces at once and then kept for 2 hours by renewing the fixing fluid once more. After washed adequately in 0.1 M phosphate buffer, the pieces were postfixed in 2% osmic acid in the same buffer at 2～6°C. The piece of skin was then dehydrated in an ethanol series and embedded in Epon 812. Sections were cut on a Porter-Blum MT-1 ultramicrotome with a glass knife, double stained with saturated uranyl acetate and alkaline

lead citrate and photographed with a Hitachi Hs-8 electron microscope.

Dopa reaction for the purpose of light microscopic observation was mainly made according to the method by BLOCH (1917) as follows.

(1) A piece of about 5 mm×5 mm removed from a spot area of the dorsal skin of the body or hind limbs is fixed for 3 hours in cold 3% glutaraldehyde in 0.1 M phosphate buffer at pH 7.4.

(2) Sections are cut on a freezing microtome and washed for 5 seconds in distilled water.

(3) Sections are incubated for 3 hours at 37°C in fresh 0.1% *L*-dopa medium contained in 0.1 M phosphate buffer of pH 7.4.

(4) Sections are rinsed in water and then in alcohol. After covering with a coverslip, they are observed under a microscope.

Dopa reaction for electron microscopic observation was principally made according to the method by EPPIG and DUMONT (1972) as follows.

(1) A piece of the dorsal skin of the body or hind limbs is fixed in cold 3% glutaraldehyde in 0.1 M phosphate buffer at pH 7.4.

(2) After washing for about 15 minutes in 0.1 M phosphate buffer, the piece is incubated in 0.1% *L*-dopa medium contained in 0.1 M cacodylate buffer (pH 7.4) for 3 hours at 37°C.

(3) After washing in 0.1 M phosphate buffer, the piece is postfixed in 2% osmic acid contained in 0.1 M phosphate buffer (pH 7.4) for 2 hours.

(4) Dehydrated, embedded and sectioned. A part of each sample was incubated in *L*-dopa medium containing 0.01% PTU (phenylthiourea) as an inhibitor.

The developmental stages reported in this paper follow those of *Rana pipiens* established by SHUMWAY (1940) and TAYLOR and KOLLROS (1946) for convenience sake.

## OBSERVATION

### *I. Inheritance of 13 albino stocks*

#### 1. Ex–145 stock (Ex)

Eggs of a female *Rana nigromaculata* collected from the field in 1967 were irradiated with 145 rads of X-rays and fertilized with sperm of a normal male *Rana nigromaculata* collected from the field. As females obtained from this mating (Ex–145♀×W♂) attained sexual maturity in 1970, 12 (Nos. 1～12) of them were mated again with a normal male (W70♂, No. 2) collected from the field. Besides these matings, offspring were produced from the 12 females by the method of diploid gynogenesis. It was found that one of 16 hatched tadpoles obtained from female No. 11 by diploid gynogenesis was an albino. On the basis of this finding, it was assumed that about half of 53 offspring produced from a mating between this female, (Ex–145♀×W♂)♀, No. 11, and the male, W70♂, No. 2,

must be heterozygous for the albino gene (cf. NISHIOKA 1977, Tables 5 and 8). As these offspring attained sexual maturity in 1972, matings were made between four females (Nos. 1～4) and three males (Nos. 1～3) of them. The results showed that two (Nos. 2 and 3) of the females and one (No. 3) of the males were heterozygous for the albino gene. Then, 90 wild-type tadpoles and 30 albinic ones produced from a mating between one of the two females and the single male heterozygous for the albino gene, (Ex–145 W)$_{11}$W♀, No. 2×(Ex–145 W)$_{11}$W♂, No. 3, were reared in our laboratory (cf. Nishioka 1977, Table 10).

As these tadpoles attained sexual maturity in 1974, matings were made between two females, Ex. Het. 72♀, Nos. 1 and 2, and two males, Ex. Het. 72♂, Nos. 1 and 2, heterozygous for the albino gene. The results showed that 147 of 201 hatched tadpoles were of the wild-type and 54 were albinos (Table 1).

TABLE 1

Production of tadpoles homo- or heterozygous for an albino gene in the Ex stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of hatched tadpoles (St. 21) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1974 | Ex. Het. 72, Nos. 1, 2 | Ex. Het. 72, Nos. 1, 2 | 242 | 230 (95.0%) | 201 (83.1%) | 147 | 54 |
| 1978 | Ex. Het. 72, Nos. 3, 4 | Ex. Alb. 74, Nos. 3, 4 | 761 | 659 (86.6%) | 602 (79.1%) | 289 | 313 |
| | Ex. Alb. 74, No. 2 | Ex. Alb. 74, Nos. 3, 4 | 218 | 204 (93.6%) | 174 (79.8%) | 0 | 174 |
| | Wild 78, Nos. 1～3 | Ex. Alb. 74, Nos. 3, 4 | 455 | 422 (92.7%) | 394 (86.6%) | 394 | 0 |

In 1978, matings were made between two females (Ex. Het. 72♀, Nos. 3 and 4) which had been produced in 1972 and were heterozygous for the albino gene and two male albinos (Ex. Alb. 74♂, Nos. 3 and 4) obtained in 1974. It was found that 289 of 602 hatched tadpoles produced from these matings were of the wild-type and 313 were albinos. Matings were also made between a female albino (Ex. Alb. 74♀, No. 2) obtained in 1974 and the above two male albinos (Ex. Alb. 74♂, Nos. 3 and 4). A total of 174 hatched tadpoles produced from these matings were all albinos. On the other hand, 394 hatched tadpoles produced from three normal females (W78♀, Nos. 1～3) collected from the field by mating with the above two male albinos (Ex. Alb. 74♂, Nos. 3 and 4) were all of the wild-type (Table 1).

These findings evidently indicate that the albinos are mutants due to a single recessive gene induced in an egg by irradiation with 145 rads of X-rays.

## 2. SN–50 stock (SN I)

Eggs of a normal female collected from the field in 1967 were fertilized with sperm irradiated with 50 rads of neutrons. As females raised from these eggs attained sexual maturity in 1970, offspring were produced from five (Nos. 1～5) of them by the method of diploid gynogenesis. It was found that female No. 5 produced six albinic tadpoles. As female No. 5 was heterozygous for the albino

gene, this female was mated with a normal male collected from the field. About half of 157 offspring produced from this mating, (W♀×Sn–50♂)♀, No. 5×W70♂, No. 3, was considered to be heterozygous for the albino gene. Then, matings were made in 1972 between three mature females (Nos. 1～3) and three mature males (Nos. 1～3) of the offspring. The results showed that two (Nos. 1 and 2) of the females and one (No. 1) of the males were heterozygous for the albino gene. Of the tadpoles obtained from one of these two matings, (W×Sn–50)$_5$W♀, No. 1×(W×Sn–50)$_5$W♂, No. 1, 46 wild-type individuals and 20 albinos were reared (cf. Nishioka 1977, Tables 6, 8 and 11).

As these tadpoles obtained in 1972 attained sexual maturity in 1974, brother and sister matings were made between two females and two males heterozygous for the albino gene, Sn I. Het. 72♀, Nos. 1 and 2×Sn I. Het. 72♂, Nos. 1 and 2. It was found that 109 of 143 tadpoles produced from these matings were of the wild-type and 34 were albinos. In 1975, a mating between another heterozygous female and a male albino, Sn I. Het. 72♀, No. 3×Sn I. Alb. 72♂, No. 1, was made. The result showed that 96 of 191 tadpoles obtained from this mating were of the wild-type and 95 were albinos.

TABLE 2

Production of tadpoles homo- or heterozygous for an albino gene in the Sn I stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of hatched tadpoles (St. 21) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1974 | SnI. Het. 72, Nos. 1, 2 | SnI. Het. 72, Nos. 1, 2 | 255 | 178 (69.8%) | 143 (56.1%) | 109 | 34 |
| 1975 | SnI. Het. 72, No. 3 | SnI. Alb. 72, No. 1 | 269 | 231 (85.9%) | 191 (71.0%) | 96 | 95 |
| 1976 | SnI. Alb. 74, No. 1 | SnI. Alb. 74, No. 2 | 335 | 282 (84.2%) | 197 (58.8%) | 0 | 197 |
| 1977 | SnI. Het. 75, No. 4 | SnI. Alb. 74, No. 4 | 274 | 256 (93.4%) | 202 (73.7%) | 107 | 95 |

In 1976, a brother and sister mating between a female albino and a male albino obtained in 1974, Sn I. Alb. 74♀, No. 1×Sn I. Alb. 74♂, No. 2, was made. A total of 197 tadpoles produced from this mating were all albinos. In 1977, a mating between a heterozygous female and a male albino, Sn I. Het. 75♀, No. 4×Sn I. Alb. 74♂, No. 4, was made. Of 202 tadpoles produced from this mating, 107 were of the wild-type and 95 were albinos (Table 2).

It is evident from these findings that the albinos are mutants due to a single recessive gene induced in a spermatozoon by irradiation with 50 rads of neutrons.

### 3. Sn–130 stock (Sn II)

In 1967, eggs of a normal female collected from the field were fertilized with sperm irradiated with 130 rads of neutrons. As females raised from these eggs attained sexual maturity in 1970, offspring were produced from 10 (Nos. 1～10) of the females by the method of diploid gynogenesis. The results showed that four albinic tadpoles were produced from female No. 3 of the 10 females. As

female No. 3 was heterozygous for the albino gene, offspring were produced from a mating between this female and a normal male collected from the field, (W♀ × Sn–130♂)♀, No. 3 × W70♂, No. 3. It was considered that about half of 112 tadpoles produced from this mating was heterozygous for the albino gene. As 90 of these tadpoles completed metamorphosis and 30 frogs attained sexual maturity in 1972, brother and sister matings were made between eight mature females (Nos. 1～8) and six mature males (Nos. 1～6) of these frogs. The results indicated that females Nos. 4, 5, 6 and 8 and males Nos. 1, 2 and 4 were heterozygous for the albino gene. Of the offspring of these heterozygous frogs, 72 wild-type and 24 albinic tadpoles obtained from a mating between female No. 6 and male No. 4, $(W \times Sn\text{–}130)_3W$♀, No. 6 × $(W \times Sn\text{–}130)_3W$♂, No. 4, were reared (cf. Nishioka 1977, Tables 6, 8 and 12).

As these frogs attained sexual maturity in 1974, brother and sister matings between two heterozygous females and two heterozygous males obtained in 1972, Sn II. Het. 72♀, Nos. 1 and 2 × Sn II. Het. 72♂, Nos. 1 and 2, were made. It was found that 153 of 198 tadpoles produced from these matings were of the wild-type and 45 were albinos. In 1975, a brother and sister mating between a female and a male albino obtained in 1972, Sn II. Alb. 72♀, No. 1 × Sn II. Alb. 72♂, No. 1, was made. All 466 tadpoles produced from this mating were albinos. Of 176 tadpoles produced from a mating between the above female albino and a heterozygous male obtained in 1972, Sn II. Alb. 72♀, No. 1 × Sn II. Het. 72♂, No. 3, 90 were of the wild-type and 86 were albinos. On the other hand, 118 tadpoles produced from a mating between the above female albino and a male collected from the field as well as 152 tadpoles from a mating between a female collected from the field and the above male albino were all of the wild-type.

During the years from 1977 to 1979, matings were made between female

TABLE 3

Production of tadpoles homo- or heterozygous for an albino gene in the Sn II stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of hatched tadpoles (St. 21) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1974 | SnII. Het. 72, Nos. 1, 2 | SnII. Het. 72, Nos. 1, 2 | 224 | 208 (92.9%) | 198 (88.4%) | 153 | 45 |
| 1975 | SnII. Alb. 72, No. 1 | SnII. Alb. 72, No. 1 | 800 | 756 (94.5%) | 466 (58.3%) | 0 | 466 |
| | | SnII. Het. 72, No. 3 | 235 | 220 (93.6%) | 176 (74.9%) | 90 | 86 |
| | | Wild 75, No. 1 | 214 | 212 (99.1%) | 118 (55.1%) | 118 | 0 |
| | Wild 75, No. 1 | SnII. Alb. 72, No. 1 | 158 | 155 (98.1%) | 152 (96.2%) | 152 | 0 |
| 1977 | SnII. Alb. 74, Nos. 2, 3 | SnII. Alb. 74, No. 3 | 349 | 341 (97.7%) | 211 (60.5%) | 0 | 211 |
| 1978 | SnII. Alb. 74, Nos. 4, 5 | SnII. Het. 75, No. 4 | 447 | 306 (68.5%) | 289 (64.7%) | 147 | 142 |
| 1979 | SnII. Alb. 74, Nos. 6, 7 | SnII. Alb. 74, No. 5 | 231 | 221 (95.7%) | 168 (72.7%) | 0 | 168 |
| | SnII. Het. 75, No. 3 | SnII. Alb. 74, No. 5 | 190 | 187 (98.4%) | 185 (97.4%) | 95 | 90 |

and male albinos and between albinos and heterozygous frogs obtained in 1974 and 1975. It was found that 211 and 168 tadpoles produced from matings between female and male albinos, Sn II. Alb. 74♀, Nos. 2 and 3 × Sn II. Alb. 74♂, No. 3, and Sn II. Alb. 74♀, Nos. 6 and 7 × Sn II. Alb. 74♂, No. 5, were all albinos. Of 289 tadpoles produced from matings between two female albinos and a heterozygous male, Sn II. Alb. 74♀, Nos. 4 and 5 × Sn II. Het. 75♂, No. 4, 147 were of the wild-type and 142 were albinos. Of 185 tadpoles produced from a mating between a heterozygous female and a male albino, Sn II. Het. 75♀, No. 3 × Sn II. Alb. 74♂, No. 5, 95 were of the wild-type and 90 were albinos (Table 3).

These findings indicate that the albinos are mutants due to a single recessive gene induced in a spermatozoon by irradiation with 130 rads of neutrons.

### 4. *Rana brevipoda* stock (Br)

In 1970, Mr. Ohtani found that five of 370 tadpoles produced from a mating between a normal female *Rana brevipoda* and a male *Rana brevipoda*, Br. Het. 70♂, No. 1, collected by him from Konko-cho, Okayama Prefecture were albino-mosaics. A part of wild-type tadpoles obtained from this mating was reared by him until sexual maturity. In 1973, several brother and sister matings were made among these frogs and it was found that many albinos were produced from one of the matings. The present authors received three female and three male albinos from Mr. Ohtani. These albinos were reared until sexual maturity.

TABLE 4

Production of tadpoles homo- or heterozygous for an albino gene in the Br stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of hatched tadpoles (St. 21) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1975 | Br. Alb. 73, Nos. 1~3 | Br. Alb. 73, No. 3 | 228 | 128 (56.1%) | 82 (36.0%) | 0 | 82 |
| | | Wild *bre.* 75, No. 1 | 326 | 211 (64.7%) | 190 (58.3%) | 190 | 0 |
| 1978 | Br. Het. 75, Nos. 1, 2 | Br. Alb. 75, No. 5 | 587 | 441 (75.1%) | 326 (55.5%) | 165 | 161 |
| 1979 | Wild *bre.* 79, Nos. 1, 2 | Br. Alb. 75, No. 6 | 330 | 283 (85.8%) | 276 (83.6%) | 276 | 0 |
| | Br. Het. 75, No. 3 | Br. Alb. 75, No. 6 | 320 | 299 (93.4%) | 253 (79.1%) | 128 | 125 |

In 1975, 82 tadpoles produced from brother and sister matings between female and male albinos, Br. Alb. 73♀, Nos. 1~3 × Br. Alb. 73♂, No. 3, were all albinos. On the other hand, 190 tadpoles produced from the same three female albinos by mating with a normal male *Rana brevipoda*, Wild *bre.* 75♂, No. 1, collected from the field were all of the wild-type. In 1978 and 1979, matings between three heterozygous females and two male albinos obtained in 1975, Br. Het. 75♀, Nos. 1~3 × Br. Alb. 75♂, Nos. 5 and 6, were made. The results showed that 293 of 579 tadpoles produced from these matings were of the wild-type and 286 were albinos. The tadpoles produced from normal female *Rana brevipoda*

collected from the field by mating with a male albino were all of the wild-type (Table 4).

These findings evidently indicate that the albinos are mutants due to a single recessive gene.

## 5. Yamaguchi I stock (YM I)

Nine albinic froglets of *Rana nigromaculata* given by Professor SAMBUICHI in 1973, attained sexual maturity in 1974. Four of them were females and five were males. Two of the male albinos, YM I. Alb. 73♂, Nos. 1 and 2, produced 396 heterozygous tadpoles by mating with two normal female *Rana nigromaculata*, W74♀, Nos. 1 and 2, collected from the field. As normal ovulation occurred in two of the four female albinos, YM I. Alb. 73♀, Nos. 1 and 2, in 1975 and in another female albino, YM I. Alb. 73♀, No. 3, in 1976, these females were mated with the remaining three male albinos, YM I. Alb. 73♂, Nos. 3~5, obtained in 1973. The results indicated that 936 tadpoles produced from these matings were all albinos. When matings were made between two (Nos. 1 and 2) of the above three female albinos and two heterozygous males, YM I. Het. 74♂, Nos. 1 and 2, obtained in 1974, 728 tadpoles were produced. Of these tadpoles, 354 were of the wild-type and 374 were albinos. When these two female albinos were mated with two normal males, W75♂, Nos. 2 and 3, 703 heterozygous tadpoles were produced. All these tadpoles were of the wild-type.

TABLE 5

Production of tadpoles homo- or heterozygous for an albino gene in the YM I stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of hatched tadpoles (St. 21) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1974 | Wild 74, Nos. 1, 2 | YmI. Alb. 73, Nos. 1, 2 | 469 | 432 (92.1%) | 396 (84.4%) | 396 | 0 |
| 1975 | YmI. Alb. 73, Nos. 1, 2 | YmI. Alb. 73, Nos. 3, 4 | 1206 | 1107 (91.8%) | 862 (71.5%) | 0 | 862 |
| | | YmI. Het. 74, Nos. 1, 2 | 944 | 917 (97.1%) | 728 (77.1%) | 354 | 374 |
| | | Wild 75, Nos. 2, 3 | 929 | 878 (94.5%) | 703 (75.7%) | 703 | 0 |
| 1976 | YmI. Alb. 73, No. 3 | YmI. Alb. 73, No. 5 | 188 | 134 (71.3%) | 74 (39.4%) | 0 | 74 |
| 1977 | YmI. Het. 75, Nos. 1, 2 | YmI. Alb. 75, No. 6 | 688 | 310 (45.1%) | 260 (37.8%) | 121 | 139 |
| | | YmI. Het. 75, No. 3 | 674 | 654 (97.0%) | 556 (82.5%) | 427 | 129 |
| 1978 | YmI. Alb. 75, Nos. 5, 6 | YmI. Alb. 75, No. 7 | 356 | 244 (68.5%) | 201 (56.5%) | 0 | 201 |
| 1979 | YmI. Alb. 75, Nos. 7, 8 | YmI. Het. 75, No. 4 | 369 | 359 (97.3%) | 207 (56.1%) | 102 | 105 |
| | YmI. Het. 75, No. 3 | YmI. Het. 75, No. 4 | 2110 | 2067 (98.0%) | 1996 (94.6%) | 1478 | 518 |

During the three years from 1977 to 1979, matings were made between female and male albinos, between albinos and heterozygous individuals and between heterozygous females and males, by using four female albinos, YM I. Alb. 75♀, Nos. 5~8, two male albinos, YM I. Alb. 75♂, Nos. 6 and 7, three heterozygous

females, YM I. Het. 75♀, Nos. 1~3, and two heterozygous males, YM I. Het. 75♂, Nos. 3 and 4. Of 260 tadpoles produced from matings between two heterozygous females and a male albino, YM I. Het. 75♀, Nos. 1 and 2×YM I. Alb. 75♂, No. 6, 121 were of the wild-type and 139 were albinos. Of 2552 tadpoles produced from matings between three heterozygous females and two heterozygous males, YM I. Het. 75♀, Nos. 1~3×YM I. Het. 75♂, Nos. 3 and 4, 1905 were of the wild-type and 647 were albinos. Of 207 tadpoles produced from matings between two female albinos and a heterozygous male, YM I. Alb. 75♀, Nos. 7 and 8×YM I. Het. 75♂, No. 4, 102 were of the wild-type and 105 were albinos. All 201 tadpoles produced from matings between two female albinos and a male albino, YM I. Alb. 75♀, Nos. 5 and 6×YM I. Alb. 75♂, No. 7, were albinos (Table 5).

These findings evidently indicate that the albinos are mutants due to a single recessive gene.

## 6. Kamogata stock (KM)

An albinic froglet of *Rana nigromaculata* was collected in May of 1974 by Mr. SHIRANE from Kamogata-cho, Asakuchi-gun, Okayama Prefecture. The present authors received this animal from Mr. SHIRANE and reared it for one year until sexual maturity. This albino was a female, KM. Alb. 74♀, No. 1, and then mated in the breeding season of 1975 with a normal male *Rana nigromaculata*, W75♂, No. 1, collected from the field. From this mating 536 wild-type tadpoles heterozygous for the albino gene were produced.

TABLE 6

Production of tadpoles homo- or heterozygous for an albino gene in the KM stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. II) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1975 | KM. Alb. 74, No. 1 | Wild 75, No. 1 | 553 | 550 (99.5%) | 536 (96.9%) | 536 | 0 |
| 1977 | KM. Het. 75, Nos. 1, 2 | KM. Het. 75, No. 5 | 974 | 931 (95.6%) | 308 (31.6%) | 235 | 73 |
| 1978 | KM. Alb. 77, Nos. 2, 3 | KM. Alb. 77, No. 1 | 464 | 403 (86.9%) | 385 (83.0%) | 0 | 385 |
| 1979 | KM. Alb. 77, Nos. 4, 5 | KM. Alb. 77, No. 2 | 357 | 151 (42.3%) | 103 (28.9%) | 0 | 103 |
| | KM. Alb. 77, Nos. 6, 7 | KM. Het. 75, No. 6 | 498 | 491 (98.6%) | 454 (91.2%) | 224 | 230 |
| | KM. Het. 75, No. 3 | KM. Alb. 77, No. 2 | 1825 | 1517 (83.1%) | 1328 (72.8%) | 670 | 658 |

In 1977, these tadpoles attained sexual maturity. Then, brother and sister matings between two females and one male, KM. Het. 75♀, Nos. 1 and 2× KM. Het. 75♂, No. 5, were made. Of 308 tadpoles obtained from these matings, 235 were of the wild-type and 73 were albinos.

In 1978 and 1979, matings were made between female and male albinos and between albinos and heterozygous frogs by using albinos and heterozygous frogs obtained in 1975 and 1977. It was found that 488 tadpoles produced from

matings between four female and two male albinos, Km. Alb. 77♀, Nos. 2～5× Km. Alb. 77♂, Nos. 1 and 2, were all albinos. Of 454 tadpoles produced from matings between two female albinos and a heterozygous male, Km. Alb. 77♀, Nos. 6 and 7×Km. Het. 75♂, No. 6, 224 were of the wild-type and 230 were albinos. Of 1328 tadpoles produced from a mating between a heterozygous female and a male albino, Km. Het. 75♀, No. 3×Km. Alb. 77♂, No. 2, 670 were of the wild-type and 658 were albinos (Table 6).

These findings evidently show that the albinos are mutants due to a single recessive gene.

## 7. Gion stock (Go)

This stock was derived from a mature wild-type female *Rana nigromaculata* collected in October of 1974 from Gion-cho, Hiroshima City. In the breeding season of 1975, 21 wild-type and seven albinic tadpoles were produced from eggs of this female by the method of diploid gynogenesis after accelerating ovulation by injecting bullfrog pituitaries. Although 16 wild-type and five albinic tadpoles completed metamorphosis, all of them had abnormal limbs. When the female parent, Go. Het. 74♀, No. 1, was mated with a normal male, W75♂, No. 3, collected from the field, 137 tadpoles produced from this mating were all of the wild-type. Of these tadpoles, 94 completed metamorphosis, whereas most of these had abnormal limbs. It was believed that about half of these wild-type frogs was heterozygous for the albino gene.

TABLE 7

Production of tadpoles homo- or heterozygous for an albino gene in the Go stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. 25) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1975 | Go. Het. 74, No. 1 | Wild 75, No. 3 | 171 | 170 (99.4%) | 137 (80.1%) | 137 | 0 |
| | | GD | 446 | 138 (30.9%) | 28 ( 6.3%) | 21 | 7 |
| 1976 | Go. Het. 74, No. 1 | Go. Het. 75, Nos. 1～5 | 543 | 459 (84.5%) | 370 (68.1%) | 297 | 73 |
| 1978 | Go. Alb. 76, Nos. 1～3 | Go. Alb. 76, No. 3 | 561 | 501 (89.3%) | 270 (48.1%) | 0 | 270 |
| | Go. Het. 74, No. 1 | Go. Alb. 76, No. 3 | 552 | 549 (99.5%) | 445 (80.6%) | 232 | 213 |
| | Wild 78, Nos. 2～4 | Go. Alb. 76, No. 3 | 563 | 509 (90.4%) | 460 (81.7%) | 460 | 0 |
| 1979 | Go. Alb. 76, Nos. 4～6 | Go. Alb. 76, No. 5 | 523 | 195 (37.3%) | 102 (19.5%) | 0 | 102 |
| | Het. ♀ × Het. ♂ Wild 76, Nos. 1～4 | Go. Alb. 76, No. 6 | 977 | 921 (94.3%) | 645 (66.0%) | 314 | 331 |
| | Het. ♀ × Het. ♂ Wild 76, Nos. 5, 6 | Go. Alb. 76, No. 6 | 399 | 196 (49.1%) | 184 (46.1%) | 184 | 0 |
| 1980 | Go. Het. 78, No. 2 | Go. Het. 78, No. 7 | 855 | 813 (95.1%) | 791 (92.5%) | 593 | 198 |

GD, diploid gynogenesis

In 1976, 31 frogs not having serious abnormality of the limbs attained sexual maturity. When 10 males of these frogs were mated with the female parent,

Go. Het. 74♀, No. 1, no albinic tadpoles were produced from five of the 10 males, while 297 of 370 tadpoles produced from the remaining five males, Go. Het. 75♂, Nos. 1～5, were of the wild-type and 73 were albinos. Of these tadpoles, 259 wild-type and 64 albinic tadpoles completed metamorphosis. Nearly all of these frogs showed abnormal limbs. In the breeding season of 1978, 97 wild-type and 31 albinic frogs which did not have serious abnormality of the limbs were living. In 1978 and 1979, matings were made between six female albinos, Go. Alb. 76♀, Nos. 1～6, and two male albinos, Go. Alb. 76♂, Nos. 3 and 5, which were slightest in abnormality of the limbs. The results showed that 372 tadpoles produced from these matings were all albinos. When the female parent, Go. Het. 74♀, No. 1, was mated with a male albino, Go. Alb. 76♂, No. 3, 232 of 445 tadpoles produced were of the wild-type and 213 were albinos. When these tadpoles were continuously reared, nearly all of them became froglets passing over normal metamorphosis. However, abnormality of the limbs was found in considerably many individuals.

In 1978, matings were made between three normal females of *Rana nigromaculata*, W78♀, Nos. 2～4, collected from the field and a male albino, Go. Alb. 76♂, No. 3, obtained in 1976. All 460 tadpoles produced from these matings were of the wild-type. Of these tadpoles, 230 were reared and 217 completed metamorphosis. When these froglets were continuously reared, a slight abnormality appeared in the limbs of about half number of them. Of 97 mature wild-type frogs produced from matings of heterozygous females and males which were obtained in 1976, 47 were females and 50 were males. Of these females, six, (Het. ♀ × Het. ♂)W76♀, Nos. 1～6, were mated with a male albino, Go. Alb. 76♂, No. 6. It was found that 314 of 645 tadpoles produced from four (Nos. 1～4) of the six females were of the wild-type and 331 were albinos, while no albinos were produced from the remaining two females (Nos. 5 and 6). Of the tadpoles obtained from the four females (Nos. 1～4), 314 wild-type and 331 albinic tadpoles were continuously reared; 277 and 310 completed metamorphosis, respectively. Of these froglets, 100 wild-type and 100 albinic were continuously reared. A slight abnormality was found in the limbs of about one-fourth of these frogs.

In 1980, a mating was made between a heterozygous female and a heterozygous male, Go. Het. 78♀, No. 2 × Go. Het. 78♂, No. 7, obtained in 1978, from a mating between a normal female, W78♀, No. 2, collected from the field and a male albino, Go. Alb. 76♂, No. 3. These heterozygous female and male were selected as the best grown and the most normally shaped frogs among their mates. They produced 791 tadpoles, of which 593 were of the wild-type and 198 were albinos. Of these tadpoles, 100 wild-type and 198 albinic tadpoles were continuously reared, 97 and 191 normally metamorphosed and 84 and 166 attained sexual maturity, respectively (Table 7). There were no frogs having abnormal limbs.

These findings evidently indicate that the albinos are mutants due to a single recessive gene.

## 8. Fuchu stock (Fc)

In July of 1974, an albinic froglet of *Rana nigromaculata* was collected by Mrs. KASHIWAGI from a dry riverbed in Fuchu-cho near Hiroshima City and given to the present authors. This froglet attained sexual maturity in 1976 and became a female. When this female albino was mated with a normal male, W76♂, No. 1, collected from the field, 85 wild-type tadpoles heterozygous for the albino gene were produced. As these tadpoles attained sexual maturity in 1977, a male of them, Fc. Het. 76♂, No. 1, was mated with the female albino, Fc. Alb. 74♀, No. 1, collected in 1974. Of nine tadpoles produced from this mating, four were of the wild-type and five were albinos. A mating between the same female, Fc. Alb. 74♀, No. 1, or male, Fc. Het. 76♂, No. 1, and a normal male or female collected from the field produced 33 or 30 tadpoles, all of which were of the wild-type.

TABLE 8

Production of tadpoles homo- or heterozygous for an albino gene in the Fc stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. I) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1976 | Fc. Alb. 74, No. 1 | Wild 76, No. 1 | 123 | 105 (85.4%) | 85 (69.1%) | 85 | 0 |
| 1977 | Fc. Alb. 74, No. 1 | Wild 77, No. 1 | 134 | 70 (52.2%) | 33 (24.6%) | 33 | 0 |
| | | Fc. Het. 76, No. 1 | 120 | 19 (15.8%) | 9 ( 7.5%) | 4 | 5 |
| | Wild 77, No. 1 | Fc. Het. 76, No. 1 | 215 | 41 (19.1%) | 30 (14.0%) | 30 | 0 |
| 1978 | Fc. Het. 76, Nos. 1, 2 | Fc. Het. 76, Nos. 2, 3 | 474 | 435 (91.8%) | 349 (73.6%) | 264 | 85 |
| 1980 | Fc. Alb. 78, Nos. 4, 5 | Fc. Alb. 78, No. 2 | 1023 [2] | 956 [2] (93.5%) | 871 [1] (85.1%) | 1 [1] | 870 |

[ ] Numbers of normal-colored eggs and tadpoles raised from these eggs

In the breeding season of 1978, brother and sister matings between two heterozygous females and two heterozygous males obtained in 1976, Fc. Het. 76♀, Nos. 1 and 2 × Fc. Het. 76♂, Nos. 2 and 3, produced 349 tadpoles, of which 264 were of the wild-type and 85 were albinos. In 1980, two female albinos, Fc. Alb. 78♀, Nos. 4 and 5, raised from the above albinic tadpoles were injected with bullfrog pituitaries, as they attained sexual maturity. The results showed that one female laid 925 light-colored eggs and a normal-colored (brown-black) egg, while the other female laid 868 light-colored eggs and a normal-colored egg. Of a total of 1795 eggs of the two female Fc-type albinos, two (0.11%) were normally colored. When 1023 of these eggs including the normal-colored eggs were inseminated with sperm of a male albino, Fc. Alb. 78♂, No. 2, 871 hatched normally. Of these tadpoles, 870 were albinos and the remaining one (0.11%) was of the wild-type. This wild-type tadpole was raised from one of the two normal-colored eggs (Table 8).

These findings seem to indicate that the albinos are mutants due to a single recessive gene and that reverse mutation of the albino allele to the wild-type

allele occurred in a frequency of about 0.11% in the eggs of the two female Fc-type albinos.

## 9. Tojo stock (TJ)

A total of 179 albinic tadpoles of *Rana nigromaculata* were collected in June of 1974 from rice fields in Tojo-cho, Hiba-gun, Hiroshima Prefecture. They were brought to our laboratory and then reared until sexual maturity. In the breeding season of 1975, a mating between a female *Rana nigromaculata* collected from the field and a male of the above albinos, W75♀, No. 1×TJ. Alb. 74♂, No. 1, was made. From this mating, 205 wild-type heterozygous tadpoles were produced. In 1976, 109 albinic tadpoles were produced from a mating between a female albino, TJ. Alb. 74♀, No. 1, and a male albino, TJ. Alb. 74♂, No. 4. When this female albino was mated with a normal male *Rana nigromaculata*, W76♂, No. 2, collected from the field, 49 wild-type heterozygous tadpoles were produced.

In 1977, from two kinds of matings between two female albinos and a heterozygous male, TJ. Alb. 74♀, Nos. 2 and 3 ×TJ. Het. 75♂, No. 1, and between two heterozygous females and a male albino, TJ. Het. 75♀, Nos. 1 and 2× TJ. Alb. 74♂, No. 5, 137 and 343 tadpoles were produced, respectively. Of these tadpoles, 70 and 163 were of the wild-type and 67 and 180 were albinos, respectively. Of 325 tadpoles produced from brother and sister matings between two heterozygous females and a heterozygous male obtained in 1975, TJ. Het. 75♀, Nos. 1 and 2×TJ. Het. 75♂, No. 2, 245 were of the wild-type and 80 were albinos. As 72 of 163 wild-type tadpoles and 70 of 180 albinos obtained in 1977 from matings, TJ. Het. 75♀, Nos. 1 and 2×TJ. Alb. 74♂, No. 5, attained sexual

TABLE 9

Prdocution of tadpoles homo- or heterozygous for an albino gene in the TJ stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. 25) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1975 | Wild 75, No. 1 | TJ. Alb. 74, No. 1 | 215 | 210 (97.7%) | 205 (95.3%) | 205 | 0 |
| 1976 | TJ. Alb. 74, No. 1 | TJ. Alb. 74, No. 4 | 255 | 164 (64.3%) | 109 (42.7%) | 0 | 109 |
| | | Wild 76, No. 2 | 194 | 82 (42.3%) | 49 (25.3%) | 49 | 0 |
| 1977 | TJ. Alb. 74, Nos. 2, 3 | TJ. Het. 75, No. 1 | 227 | 203 (89.4%) | 137 (60.4%) | 70 | 67 |
| | TJ. Het. 75, Nos. 1, 2 | TJ. Alb. 74, No. 5 | 533 | 508 (95.3%) | 343 (64.4%) | 163 | 180 |
| | | TJ. Het. 75, No. 2 | 573 | 506 (88.3%) | 325 (56.7%) | 245 | 80 |
| 1979 | TJ. Alb. 77, Nos. 4~6 | TJ. Alb. 77, No. 6 | 532 [4] | 515 [3] (96.8%) | 274 [1] (51.5%) | 1 [1] | 273 |

[ ] Numbers of normal-colored eggs and tadpoles raised from these eggs

maturity, three female albinos, TJ. Alb. 77♀, Nos. 4~6, of them were injected in 1979 with suspension of bullfrog pituitaries. The results showed that two of 1586 eggs from female No. 4, one of 1401 eggs from female No. 5 and one of

962 eggs from female No. 6 were normal-colored eggs, while all the other eggs were light-colored. Of a total of 3949 eggs of the three female TJ-type albinos, four (0.10%) were normally colored. When 532 of these eggs including the four normal-colored eggs were inseminated with sperm of a male albino, TJ. Alb. 77♂, No. 6, 515 eggs cleaved normally and 274 became feeding tadpoles. While 273 of them were albinos, the remaining one was of the wild-type. This tadpole completed metamorphosis and became a wild-type frog (Table 9).

These findings seem to indicate that the albinos are mutants due to a single recessive gene and that reverse mutation of the albino allele to the wild-type allele occurred in a frequency of about 0.10% in the eggs of the three female TJ-type albinos.

## 10. Toyomatsu stock (TY)

In June of 1977, 89 albinic tadpoles of *Rana nigromaculata* were collected from Mr. UCHIHI's rice field in Toyomatsu-mura, Jinseki-gun, Hiroshima Prefecture and then reared until sexual maturity in our laboratory. As male albinos were sexually matured in the breeding season of 1978, one of them, TY. Alb. 77♂, No. 1, was mated with three normal females of *Rana nigromaculata*, W78♀, Nos. 2～4, collected from the field. From these matings, 285 wild-type feeding tadpoles were produced. As female albinos collected in 1977 and heterozygous frogs obtained in 1978 attained sexual maturity in the breeding season of 1979, matings were made between female and male albinos, between heterozygous females and male and between the same females and a male albino. When four female albinos, TY. Alb. 77♀, Nos. 1～4, were injected with bullfrog pituitaries, normal ovulation occurred in all of them. A total of 149 of 1728 eggs obtained from the four females, including 56 of 772 eggs from female No. 1, 51 of 751 eggs from female No. 2, 12 of 123 eggs from female No. 3 and 30 of 82 eggs from female No. 4, were larger, being 2.5～2.6 mm in diameter, while the other eggs were of normal size, being 1.8～1.9 mm in diameter. The eggs of the females of the TY stock were slightly colored in contrast to those of the other kinds of albino stocks. However, the larger eggs were somewhat paler than the normal-sized ones in the TY stock. When matings were made between these four female albinos and a male albino, TY. Alb. 77♂, No. 5, none of the larger eggs cleaved normally, while 521 (67.0%) of 778 normal-sized eggs cleaved normally, 379 (48.7%) became hatching tadpoles (stage 20), and 268 (34.4%) became normally feeding tadpoles (stage III).

Brother and sister matings were made between four one-year-old heterozygous females obtained in 1978 and a heterozygous male, TY. Het. 78♀, Nos. 1～4× TY. Het. 78♂, No. 1. From these matings, 1031 hatching tadpoles (stage 20) were produced. While albinic tadpoles were indistinguishable from the wild-type at the hatched stage (stage 21) in the TY stock, they were slightly distinguishable at the stage of tail-fin circulation (stage 22) owing to fading of the eye color. Fourteenth days after they began to eat (stage III), the albinos were easily distinguishable from the normal ones. When counting was made at this stage, 730 of 983 normally feeding tadpoles were of the wild-type and 253 were

TABLE 10

Production of tadpoles homo- or heterozygous for an albino gene in the Ty stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1978 | Wild 78, Nos. 2~4 | Ty. Alb. 77, No. 1 | 420 | 300 (71.4%) | 285 (67.9%) | 285 | 0 |
| 1979 | Ty. Alb. 77, Nos. 1~4 | Ty. Alb. 77, No. 5 | 778 | 521 (67.0%) | 268 (34.4%) | 0 | 268 |
| | Ty. Het. 78, Nos. 1~4 | Ty. Het. 78, No. 1 | 1039 | 1031 (99.2%) | 983 (94.6%) | 730 | 253 |
| | | Ty. Alb. 77, No. 6 | 642 | 638 (99.4%) | 468 (72.9%) | 239 | 229 |

albinos. From matings between four heterozygous females and a male albino, Ty. Het. 78♀, Nos. 1~4×Ty. Alb. 77♂, No. 6, 468 normally feeding tadpoles (stage III) 14 days after beginning to eat were produced. Of these tadpoles, 239 were of the wild-type and 229 were albinos (Table 10). Whereas the albinos of the Ty stock were completely normal in shape and structure at the tadpole stage, abnormality frequently appeared in the eyes of metamorphosed frogs; right and left eyes were remarkably asymmetric in size in a considerable number of albinos.

It is evident from these findings that the albinos are mutants due to a single recessive gene.

## 11. Nishinomiya stock (Ns)

In June of 1977, many albinic tadpoles of *Rana nigromaculata* were discovered by Mr. Wakana in a rice field of Kitanatsugi-cho, Nishinomiya City. Forty-two of them were collected from the rice field and brought to our laboratory. As males of these albinos attained sexual maturity in the breeding season of 1978, one of them, Ns. Alb. 77♂, No. 1, was mated with three normal females of *Rana nigromaculata*, W78♀, Nos. 2~4, collected from the field. From these matings, 153 wild-type feeding tadpoles 14 days after beginning to eat (stage III) were produced. As the heterozygous tadpoles attained sexual maturity in 1979, brother and sister matings between four females and one male, Ns. Het. 78♀, Nos. 1~4×Ns. Het. 78♂, No. 1, were made. While 743 hatching tadpoles were produced, albinos were indistinguishable from wild-type tadpoles at this developmental stage as found in the Ty stock. They were slightly distinguishable from the wild-type

TABLE 11

Production of tadpoles homo- or heterozygous for an albino gene in the Ns stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1978 | Wild 78, Nos. 2~4 | Ns. Alb. 77, No. 1 | 260 | 165 (63.5%) | 153 (58.8%) | 153 | 0 |
| 1979 | Ns. Het. 78, Nos. 1~4 | Ns. Het. 78, No. 1 | 877 | 798 (91.0%) | 702 (80.0%) | 546 | 156 |
| | | Ns. Alb. 77, No. 3 | 433 | 218 (50.3%) | 194 (44.8%) | 96 | 98 |

tadpoles at the stage of tail-fin circulation and became evidently distinguishable 14 days after beginning to eat (stage III). Of 702 feeding tadpoles, 546 were of the wild-type and 156 were albinos.

When the same four heterozygous females were mated with a male albino, Ns. Alb. 77♂, No. 3, collected from the field, 96 of 194 feeding tadpoles 14 days after beginning to eat (stage III) were of the wild-type and 98 were albinos (Table 11).

It is evident from these findings that the albinos are mutants due to a single recessive gene.

## 12. Yamaguchi II stock (YM II)

In the breeding season of 1977, about one dozen of albinic tadpoles were obtained by Professor SAMBUICHI, Faculty of Science, Yamaguchi University, from a mating between a normal female and a normal male of *Rana nigromaculata* collected from Sufu-cho, Yamaguchi City. In January of 1978, the present authors received from him five of these albinos which were already froglets, being 3～4 cm in body length. These albinic froglets attained sexual maturity in our laboratory in 1979. Three of them were females and two were males. When matings between the three females and one of the two males, YM II. Alb. 78♀, Nos. 1～3×YM II. Alb. 78♂, No. 1, were made, 301 hatching tadpoles obtained from these matings were all albinos. When this male albino was mated with two normal females, W79♀, Nos. 3 and 4, of *Rana nigromaculata*, 141 wild-type

TABLE 12

Production of tadpoles homo- or heterozygous for an albino gene in the YM II stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of hatched tadpoles (St. 21) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1979 | YmII. Alb. 78, Nos. 1～3 | YmII. Alb. 78, No. 1 | 531 | 515 (97.0%) | 301 (56.7%) | 0 | 301 |
| | Wild 79, Nos. 3, 4 | YmII. Alb. 78, No. 1 | 170 | 145 (85.3%) | 141 (82.9%) | 141 | 0 |
| 1980 | YmII. Het. 79, Nos. 1～3 | YmII. Het. 79, No. 1 | 464 | 392 (84.5%) | 315 (67.9%) | 239 | 76 |

tadpoles were produced. As these heterozygous tadpoles attained sexual maturity in the breeding season of 1980, brother and sister matings between three females and one male, YM II. Het. 79♀, Nos. 1～3×YM II. Het. 79♂, No. 1, were made. The results showed that 239 of 315 hatching tadpoles obtained from these matings were of the wild-type and 76 were albinos (Table 12).

It is evident from these findings that the albinos are mutants due to a single recessive gene.

## 13. Hiro stock (HR)

In May of 1979, 102 albinic tadpoles of *Rana nigromaculata* were collected from rice fields in Hiro-machi, Kure City. As males attained sexual maturity in the

TABLE 13
Production of tadpoles homo- or heterozygous for an albino gene in the HR stock

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of hatched tadpoles (St. 21) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1980 | Wild 80, No. 1 | HR. Alb. 79, No. 1 | 219 | 210 (95.9%) | 190 (86.8%) | 190 | 0 |
| 1981 | HR. Alb. 79, Nos. 1～4 | HR. Alb. 79, No. 6 | 279 | 269 (96.4%) | 145 (52.0%) | 0 | 145 |
| 1982 | HR. Alb. 79, No. 5 | HR. Het. 80, No. 1 | 173 | 166 (96.0%) | 131 (75.7%) | 66 | 65 |

breeding season of 1980, one of them, HR. Alb. 79♂, No. 1, was mated with a normal female, W80♀, No. 1, of *Rana nigromaculata* collected from the field. From this mating, 190 wild-type tadpoles were produced. In 1981, matings between four female albinos and a male albino collected from the field in 1979, HR. Alb. 79♀, Nos. 1～4×HR. Alb. 79♂, No. 6, were made. All 145 hatched tadpoles (stage 21) produced from these matings were albinos. In 1982, a mating between a female albino and a heterozygous male, HR. Alb. 79♀, No. 5×HR. Het. 80♂, No. 1, was made. It was found that 66 of 131 hatched tadpoles (stage 21) produced from this mating were of the wild-type and 65 were albinos (Table 13).

It is evident from these findings that the albinos are mutants due to a single recessive gene.

## *II. Crossing experiments among 13 albino stocks*

### 1. Females of the Ex stock and males of the other 12 stocks

During the years from 1974 to 1980, mating experiments were made between females homozygous or heterozygous for the albino gene of the Ex stock and males homozygous or heterozygous for the albino genes of the other 12 albino stocks (Table 14). When a female albino of the Ex stock was mated with a male albino of the YM II stock, four male albinos of the HR stock and a male albino of the TJ stock, 164, 70 and 181 feeding tadpoles (stage III), 415 in total, were produced, respectively. All these tadpoles were albinos. When a female albino was mated with a male albino of the TY stock, a male albino of the Ns stock and a heterozygous male of the KM stock, 131, 107 and 125 feeding tadpoles (stage III), 363 in total, were produced, respectively. These tadpoles were all of the wild-type. When two heterozygous females were mated with two heterozygous males of the SN I stock, two heterozygous males of the SN II stock and a heterozygous male of the Fc stock, 155, 212 and 215 feeding tadpoles (stage III), 582 in total, were produced, respectively. Of these tadpoles, 113, 158 and 162, 433 in total, were of the wild-type and 42, 54 and 53, 149 in total, were albinos, respectively. Thus, the wild-type and albinic tadpoles obtained from the matings between heterozygous females and males were produced in an approximate 3: 1 ratio.

TABLE 14
Results of mating experiments between females of the Ex stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1974 | Ex. Het. 72, Nos. 1, 2 | SnI. Het. 72, Nos. 1, 2 | 191 | 159 (83.2%) | 155 (81.2%) | 113 | 42 |
| | | SnII. Het. 72, Nos. 1, 2 | 250 | 231 (92.4%) | 212 (84.8%) | 158 | 54 |
| | | YmI. Alb. 73, Nos. 1, 2 | 162 | 120 (74.1%) | 112 (69.1%) | 112 | 0 |
| | | Br. Alb. 73, Nos. 1, 2 | 165 | 106 (64.2%) | 91 (55.2%) | 45 | 46 |
| 1976 | Ex. Alb. 74, No. 1 | Km. Het. 75, No. 4 | 246 | 184 (74.8%) | 125 (50.8%) | 125 | 0 |
| | | Tj. Alb. 74, No. 4 | 214 | 193 (90.2%) | 181 (84.6%) | 0 | 181 |
| 1978 | Ex. Het. 72, Nos. 3, 4 | Go. Alb. 76, No. 3 | 294 | 274 (93.2%) | 259 (88.1%) | 259 | 0 |
| | | Fc. Het. 76, No. 2 | 371 | 350 (94.3%) | 215 (58.0%) | 162 | 53 |
| 1980 | Ex. Alb. 78, No. 3 | YmII. Alb. 78, No. 2 | 194 | 190 (97.9%) | 164 (84.5%) | 0 | 164 |
| | | Hr. Alb. 79, Nos. 2~5 | 204 | 73 (35.8%) | 70 (34.3%) | 0 | 70 |
| | | Ty. Alb. 79, No. 7 | 142 | 141 (99.3%) | 131 (92.3%) | 131 | 0 |
| | | Ns. Alb. 79, No. 4 | 142 | 142 (100 %) | 107 (75.4%) | 107 | 0 |
| 1980 | Ex. Het. 78, No. 5 | YmII. Alb. 78, No. 2 | 177 | 175 (98.9%) | 166 (93.8%) | 82 | 84 |
| | | Hr. Alb. 79, Nos. 2~5 | 186 | 146 (78.5%) | 145 (78.0%) | 71 | 74 |
| | | Ty. Alb. 79, No. 7 | 124 | 91 (73.4%) | 78 (62.9%) | 78 | 0 |
| | | Ns. Alb. 79, No. 1 | 145 | 123 (84.8%) | 117 (80.7%) | 117 | 0 |

When matings were made between one or two heterozygous females and two male albinos of the Br stock, a male albino of the Ym II stock and four male albinos of the Hr stock, 91, 166 and 145 feeding tadpoles, 402 in total, were produced, respectively. Of these tadpoles, 45, 82 and 71, 198 in total, were of the wild-type, respectively, while 46, 84 and 74, 204 in total, were albinos, respectively. Thus, the wild-type and albinic tadpoles were produced in a ratio of approximately 1 : 1. When one or two heterozygous females were mated with two male albinos of the Ym I stock and a male albino of each of the Go, Ty and Ns stocks, 112, 259, 78 and 117 feeding tadpoles (stage III), 566 in total, were produced, respectively. All these tadpoles were of the wild-type.

## 2. Females of the Sn I stock and males of the other 12 stocks

During the years from 1974 to 1980, females homozygous or heterozygous for the albino gene of the Sn I stock were mated with males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 15). When a female albino was mated with a male albino of the Ym II stock and four male albinos of the Hr stock, 136 and 46 feeding tadpoles, 182 in total, were produced. These tadpoles were all albinos. When a female albino was mated with a male

TABLE 15

Results of mating experiments between females of the SnI stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1974 | SnI. Het. 72, Nos. 1, 2 | SnII. Het. 72, Nos. 1, 2 | 260 | 223 (85.8%) | 174 (66.9%) | 130 | 44 |
| | | Ex. Het. 72, Nos. 1, 2 | 277 | 240 (86.6%) | 208 (75.1%) | 161 | 47 |
| | | YmI. Alb. 73, Nos. 1, 2 | 325 | 171 (52.6%) | 158 (48.6%) | 158 | 0 |
| | | Br. Alb. 73, Nos. 1, 2 | 311 | 214 (68.8%) | 139 (44.7%) | 73 | 66 |
| 1975 | SnI. Het. 72, No. 3 | SnII. Alb. 72, No. 1 | 311 | 300 (96.5%) | 245 (78.8%) | 116 | 129 |
| | | Tj. Alb. 74, No. 1 | 308 | 294 (95.5%) | 266 (86.4%) | 132 | 134 |
| 1976 | SnI. Alb. 74, No. 1 | Go. Het. 75, Nos. 1~5 | 1645 | 789 (48.0%) | 500 (30.4%) | 500 | 0 |
| | | Km. Het. 75, No. 4 | 258 | 217 (84.1%) | 139 (53.9%) | 139 | 0 |
| 1977 | SnI. Alb. 74, No. 2 | Fc. Het. 76, No. 1 | 512 | 400 (78.1%) | 344 (67.2%) | 179 | 165 |
| 1978 | SnI. Alb. 74, No. 3 | Ns. Alb. 77, No. 1 | 463 | 203 (43.8%) | 142 (30.7%) | 142 | 0 |
| | | Ty. Alb. 77, No. 1 | 500 | 319 (63.8%) | 211 (42.2%) | 211 | 0 |
| 1980 | SnI. Alb. 77, No. 4 | YmII. Alb. 78, No. 2 | 186 | 183 (98.4%) | 136 (73.1%) | 0 | 136 |
| | | Hr. Alb. 79, Nos. 2~5 | 247 | 61 (24.7%) | 46 (18.6%) | 0 | 46 |
| 1980 | SnI. Het. 77, No. 6 | YmII. Alb. 78, No. 2 | 159 | 154 (96.9%) | 135 (84.9%) | 66 | 69 |
| | | Hr. Alb. 79, Nos. 2~5 | 236 | 113 (47.9%) | 94 (39.8%) | 45 | 49 |

albino of the Ns stock, a male albino of the Ty stock, five heterozygous males of the Go stock and a heterozygous male of the Km stock, 142, 211, 500 and 139 feeding tadpoles (stage III), 992 in total, were produced, respectively. These tadpoles were all of the wild-type. When a female albino was mated with a heterozygous male of the Fc stock, 344 feeding tadpoles were produced. Of these tadpoles, 179 were of the wild-type and 165 were albinos.

When one or two heterozygous females were mated with a male albino of the Sn II stock, a male albino of the Ym II stock, four male albinos of the Hr stock, two male albinos of the Br stock and a male albino of the Tj stock, 245, 135, 94, 139 and 266 feeding tadpoles (stage III), 879 in total, were produced, respectively. Of these tadpoles, 116, 66, 45, 73 and 132, 432 in total, were of the wild-type and 129, 69, 49, 66 and 134, 447 in total, were albinos, respectively. Thus, the wild-type and albinic tadpoles were produced in a ratio of approximately 1: 1. When two heterozygous females were mated with two heterozygous males of the Sn II stock and two heterozygous males of the Ex stock, 174 and 208 feeding tadpoles, 382 in total, were produced, respectively. Of these tadpoles, 130 and 161, 291 in total, were of the wild-type, and 44 and 47, 91 in total, were albinos. Thus, the wild-type and albinic tadpoles were produced in a ratio of approximately 3: 1. When two heterozygous females were mated with two male albinos of the

YM I stock, 158 feeding tadpoles were produced. These tadpoles were all of the wild-type.

### 3. Females of the SN II stock and males of the other 12 stocks

During the years from 1974 to 1980, mating experiments were made between females homozygous or heterozygous for the albino gene of the SN II stock and males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 16). When one or two female albinos were mated with a male albino of

TABLE 16

Results of mating experiments between females of the SNII stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albnio |
| 1974 | SNII. Het. 72, Nos. 1, 2 | SNI. Het. 72, Nos. 1, 2 | 184 | 158 (85.9%) | 152 (82.6%) | 114 | 38 |
| | | EX. Het. 72, Nos. 1, 2 | 226 | 220 (97.3%) | 200 (88.5%) | 152 | 48 |
| | | YMI. Alb. 73, Nos. 1, 2 | 157 | 139 (88.5%) | 133 (84.7%) | 133 | 0 |
| | | BR. Alb. 73, Nos. 1, 2 | 182 | 144 (79.1%) | 108 (59.3%) | 57 | 51 |
| 1975 | SNII. Alb. 72, No. 1 | TJ. Alb. 74, No. 1 | 163 | 162 (99.4%) | 120 (73.6%) | 0 | 120 |
| 1977 | SNII. Alb. 74, Nos. 2, 3 | GO. Alb. 76, No. 1 | 448 | 339 (75.7%) | 189 (42.2%) | 189 | 0 |
| | | KM. Het. 75, No. 5 | 475 | 456 (96.0%) | 250 (52.6%) | 250 | 0 |
| | | FC. Het. 76, No. 1 | 210 | 152 (72.4%) | 120 (57.1%) | 59 | 61 |
| 1978 | SNII. Alb. 74, Nos. 4, 5 | NS. Alb. 77, No. 2 | 419 | 23 ( 5.5%) | 11 ( 2.6%) | 11 | 0 |
| | | TY. Alb. 77, No. 1 | 574 | 218 (38.0%) | 176 (30.7%) | 176 | 0 |
| 1979 | SNII. Alb. 74, Nos. 6, 7 | YMII. Alb. 78, No. 1 | 330 | 159 (48.2%) | 112 (33.9%) | 0 | 112 |
| | SNII. Het. 75, No. 3 | YMII. Alb. 78, No. 1 | 189 | 173 (91.5%) | 166 (87.8%) | 85 | 81 |
| 1980 | SNII. Het. 75, No. 6 | HR. Alb. 79, Nos. 2~5 | 185 | 77 (41.6%) | 74 (40.0%) | 35 | 39 |

the YM II stock and a male albino of the TJ stock, 112 and 120 feeding tadpoles were produced, respectively. These tadpoles were all albinos. When two female albinos were mated with a male albino of each of the GO, TY and NS stocks and a heterozygous male of the KM stock, 189, 176, 11 and 250 feeding tadpoles were produced, respectively. These tadpoles were all of the wild-type. When two female albinos were mated with a heterozygous male of the FC stock, 120 feeding tadpoles were produced. Of these tadpoles, 59 were of the wild-type and 61 were albinos.

When one or two heterozygous females were mated with a male albino of the YM II stock, four male albinos of the HR stock and two male albinos of the BR stock, 166, 74 and 108 feeding tadpoles, 348 in total, were produced, respectively. Of these tadpoles, 85, 35 and 57, 177 in total, were of the wild-type, and 81, 39 and 51, 171 in total, were albinos, respectively. Thus, the wild-type and albinic

tadpoles were produced in a ratio of approximately 1 : 1. When two heterozygous females were mated with two heterozygous males of the SN I stock and two heterozygous males of the Ex stock, 152 and 200 feeding tadpoles, 352 in total, were produced, respectively. Of these tadpoles, 114 and 152, 266 in total, were of the wild-type, and 38 and 48, 86 in total, were albinos, respectively. Thus, the wild-type and albinic tadpoles were produced in a ratio of approximately 3 : 1. When two heterozygous females were mated with two male albinos of the YM I stock, 133 feeding tadpoles were produced. These tadpoles were all of the wild-type.

4. Females of the BR stock and males of the other 12 stocks

During the years from 1975 to 1984, mating experiments were made between female *Rana brevipoda* homozygous or heterozygous for the albino gene of the BR stock and male *Rana nigromaculata* homozygous or heterozygous for the albino genes of the other 12 stocks (Table 17). When one or three female albinos were mated with a male albino of each of the Ex, SN I, SN II, HR and TJ stocks, 99, 96, 105, 148 and 100 feeding tadpoles, 548 in total, were produced, respectively. These tadpoles were all albinos. When the same four female albinos were mated with a male albino of each of the YM I, TY and Ns stocks, 104, 92 and 93 feeding tadpoles, 289 in total, were produced, respectively. These tadpoles were all of

TABLE 17

Results of mating experiments between females of the BR stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1975 | BR. Alb. 73, Nos. 1~3 | EX. Alb. 72, No. 1 | 292 | 111 (38.0%) | 99 (33.9%) | 0 | 99 |
| | | SNI. Alb. 72, No. 1 | 350 | 210 (60.0%) | 96 (27.4%) | 0 | 96 |
| | | SNII. Alb. 72, No. 1 | 326 | 173 (53.1%) | 105 (32.2%) | 0 | 105 |
| | | YMI. Alb. 73, No. 4 | 224 | 120 (53.6%) | 104 (46.4%) | 104 | 0 |
| | | TJ. Alb. 74, No. 2 | 210 | 101 (48.1%) | 100 (47.6%) | 0 | 100 |
| 1978 | BR. Het. 75, Nos. 1, 2 | KM. Alb. 77, No. 1 | 473 | 325 (68.7%) | 203 (42.9%) | 203 | 0 |
| | | GO. Alb. 76, No. 4 | 421 | 393 (93.3%) | 220 (52.3%) | 220 | 0 |
| | | FC. Het. 76, No. 2 | 519 | 177 (34.1%) | 139 (26.8%) | 106 | 33 |
| 1979 | BR. Het. 75, No. 3 | YMII. Alb. 78, No. 1 | 199 | 152 (76.4%) | 103 (51.8%) | 53 | 50 |
| 1984 | BR. Alb. 79, No. 4 | HR. Alb. 82, No. 7 | 169 | 164 (97.0%) | 148 (87.6%) | 0 | 148 |
| | | TY. Alb. 79, No. 9 | 143 | 116 (81.1%) | 92 (64.3%) | 92 | 0 |
| | | NS. Alb. 79, No. 6 | 115 | 102 (88.7%) | 93 (80.9%) | 93 | 0 |
| | BR. Het. 79, Nos. 4, 5 | HR. Alb. 82, No. 7 | 209 | 193 (92.3%) | 183 (87.6%) | 91 | 92 |
| | | TY. Alb. 79, No. 9 | 305 | 263 (86.2%) | 203 (66.6%) | 203 | 0 |
| | | NS. Alb. 79, No. 6 | 235 | 205 (87.2%) | 175 (74.5%) | 175 | 0 |

the wild-type.

When one or two heterozygous females were mated with a male albino of each of the Km, Go, Ty and Ns stocks, 203, 220, 203 and 175 feeding tadpoles, 801 in total, were produced, respectively. These tadpoles were all of the wild-type. When two heterozygous females were mated with a heterozygous male of the Fc stock, 139 feeding tadpoles were produced. Of these tadpoles, 106 were of the wild-type, and 33 were albinos. When one or two heterozygous females were mated with a male albino of the Ym II stock and a male albino of the Hr stock, 103 and 183 feeding tadpoles were produced, respectively. Of these tadpoles, 53 and 91 were of the wild-type, and 50 and 92 were albinos, respectively.

### 5. Females of the Ym I stock and males of the other 12 stocks

During the years from 1975 to 1980, mating experiments were made between females homozygous or heterozygous for the albino gene of the Ym I stock and males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 18). When one, two, three or four female albinos were mated with a

TABLE 18

Results of mating experiments between females of the YmI stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1975 | YmI. Alb. 73, Nos. 1, 2 | SnI. Alb. 72, No. 1 | 316 | 295 (93.4%) | 217 (68.7%) | 217 | 0 |
| | | Tj. Alb. 74, Nos. 1, 2 | 828 | 741 (89.5%) | 612 (73.9%) | 612 | 0 |
| 1976 | YmI. Alb. 73, No. 3 | SnII. Alb. 74, No. 2 | 214 | 189 (88.3%) | 128 (59.8%) | 128 | 0 |
| | | Km. Het. 75, No. 3 | 155 | 135 (87.1%) | 98 (63.2%) | 98 | 0 |
| | | Go. Het. 75, Nos. 1~5 | 953 | 724 (76.0%) | 563 (59.1%) | 563 | 0 |
| | | Tj. Alb. 74, No. 4 | 223 | 200 (89.7%) | 147 (65.9%) | 147 | 0 |
| 1977 | YmI. Alb. 73, No. 4 | Ex. Alb. 74, No. 2 | 233 | 228 (97.9%) | 199 (85.4%) | 199 | 0 |
| | | Br. Alb. 75, No. 4 | 179 | 15 ( 8.4%) | 8 ( 4.5%) | 8 | 0 |
| | | Km. Het. 75, No. 5 | 230 | 225 (97.8%) | 194 (84.3%) | 194 | 0 |
| | YmI. Het. 75, Nos. 1, 2 | Fc. Het. 76, No. 1 | 457 | 84 (18.4%) | 67 (14.7%) | 67 | 0 |
| 1978 | YmI. Alb. 75, Nos. 5, 6 | Go. Alb. 76, No. 3 | 489 | 480 (98.2%) | 404 (82.6%) | 404 | 0 |
| | | Ns. Alb. 77, No. 2 | 331 | 41 (12.4%) | 25 ( 7.6%) | 25 | 0 |
| | | Ty. Alb. 77, No. 3 | 394 | 252 (64.0%) | 221 (56.1%) | 221 | 0 |
| 1979 | YmI. Alb. 77, Nos. 7, 8 | Go. Alb. 76, No. 5 | 274 | 155 (56.6%) | 142 (51.8%) | 142 | 0 |
| | | Ty. Alb. 77, No. 5 | 303 | 298 (98.3%) | 270 (89.1%) | 270 | 0 |
| | | YmII. Alb. 78, No. 1 | 331 | 306 (92.4%) | 271 (81.9%) | 271 | 0 |
| 1980 | YmI. Het. 77, Nos. 4, 5 | Hr. Alb. 79, Nos. 2~5 | 545 | 267 (49.0%) | 261 (47.9%) | 261 | 0 |

male albino of each of six albino stocks, Ex, Sn I, Sn II, Ym II, Br and Ns, two male albinos of each of the Go and Ty stocks and three male albinos of the Tj stock, 199, 217, 128, 271, 8, 25, 546, 491 and 759 feeding tadpoles, 2644 in total, were produced, respectively. These tadpoles were all of the wild-type. When one or two female albinos were mated with five heterozygous males of the Go stock and two heterozygous males of the Km stock, 563 and 292 feeding tadpoles, 855 in total, were produced, respectively. These tadpoles were all of the wild-type.

When two heterozygous females were mated with four male albinos of the Hr stock and a heterozygous male of the Fc stock, 261 and 67 feeding tadpoles, 328 in total, were produced, respectively. All these tadpoles were of the wild-type.

### 6. Females of the Km stock and males of the other 12 stocks

In 1975, 1977, 1978, 1979 and 1980, mating experiments were made between females homozygous or heterozygous for the albino gene of the Km stock and

TABLE 19

Results of mating experiments between females of the Km stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1975 | Km. Alb. 74, No. 1 | Ex. Alb. 72, No. 1 | 98 | 94 (95.9%) | 91 (92.9%) | 91 | 0 |
| | | SnI. Alb. 72, No. 1 | 125 | 125 (100 %) | 125 (100 %) | 125 | 0 |
| | | SnII, Alb. 72, No. 1 | 103 | 101 (98.1%) | 94 (91.3%) | 94 | 0 |
| | | YmI. Alb. 73, No. 4 | 117 | 117 (100 %) | 105 (89.7%) | 105 | 0 |
| | | YmI. Het. 74, No. 1 | 91 | 90 (98.9%) | 86 (94.5%) | 86 | 0 |
| | | Tj. Alb. 74, No. 1 | 97 | 92 (94.8%) | 90 (92.8%) | 90 | 0 |
| | | Br. Alb. 73, No. 3 | 139 | 137 (98.6%) | 132 (95.0%) | 132 | 0 |
| 1977 | Km. Het. 75, Nos. 1, 2 | Ex. Alb. 74, No. 2 | 425 | 334 (78.6%) | 143 (33.6%) | 143 | 0 |
| | | SnI. Alb. 74, No. 3 | 313 | 164 (52.4%) | 105 (33.5%) | 105 | 0 |
| | | Br. Alb. 75, No. 4 | 665 | 84 (12.6%) | 65 ( 9.8%) | 65 | 0 |
| | | Go. Alb. 76, No. 1 | 354 | 56 (15.8%) | 32 ( 9.0%) | 32 | 0 |
| | | Fc. Het. 76, No. 1 | 522 | 108 (20.7%) | 86 (16.5%) | 86 | 0 |
| 1978 | Km. Alb. 77, Nos. 2, 3 | Go. Alb. 76, No. 3 | 480 | 453 (94.4%) | 425 (88.5%) | 425 | 0 |
| | | Ty. Alb. 77, No. 3 | 614 | 438 (71.3%) | 409 (66.6%) | 409 | 0 |
| 1979 | Km. Alb. 77, Nos. 4, 5 | Ns. Alb. 77, No. 3 | 385 | 78 (20.3%) | 48 (12.5%) | 48 | 0 |
| | | YmII. Alb. 78, No. 1 | 329 | 258 (78.4%) | 145 (44.1%) | 145 | 0 |
| 1980 | Km. Alb. 77, No. 8 | Hr. Alb. 79, Nos. 2~5 | 228 | 65 (28.5%) | 45 (19.7%) | 45 | 0 |
| | Km. Het. 77, No. 4 | Hr. Alb. 79, Nos. 2~5 | 192 | 64 (33.3%) | 62 (32.3%) | 62 | 0 |

males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 19). When one or two female albinos were mated with a male albino of each of 10 albino stocks, Ex, Sn I, Sn II, Ym I, Tj, Br, Go, Ty, Ns and Ym II, and four male albinos of the Hr stock, 91, 125, 94, 105, 90, 132, 425, 409, 48, 145 and 45 feeding tadpoles (stage III), 1709 in total, were produced, respectively. These tadpoles were all of the wild-type. When a female albino was mated with a heterozygous male of the Ym I stock, 86 feeding tadpoles (stage III) were produced. These tadpoles were all of the wild-type.

When one or two heterozygous females were mated with a male albino of each of the Ex, Sn I, Br and Go stocks, four male albinos of the Hr stock and a heterozygous male of the Fc stock, 143, 105, 65, 32, 62 and 86 feeding tadpoles, 493 in total, were produced. These tadpoles were all of the wild-type.

### 7. Females of the Go stock and males of the other 12 stocks

During the years from 1975 to 1980, mating experiments were made between females homozygous or heterozygous for the albino gene of the Go stock and males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 20). When one or three female albinos were mated with a male albino of

TABLE 20

Results of mating experiments between females of the Go stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1975 | Go. Het. 74, No. 1 | Ex. Alb. 72, No. 1 | 159 | 130 (81.8%) | 94 (59.1%) | 94 | 0 |
| | | SnI. Alb. 72, No. 1 | 163 | 141 (86.5%) | 77 (47.2%) | 77 | 0 |
| | | Br. Alb. 73, No. 3 | 142 | 127 (89.4%) | 84 (59.2%) | 84 | 0 |
| 1976 | Go. Het. 74, No. 1 | SnII. Alb. 74, No. 2 | 94 | 79 (84.0%) | 75 (79.8%) | 75 | 0 |
| | | YmI. Alb. 73, No. 5 | 101 | 72 (71.3%) | 69 (68.3%) | 69 | 0 |
| | | Km. Het. 75, No. 3 | 84 | 76 (90.5%) | 74 (88.1%) | 74 | 0 |
| | | Tj. Alb. 74, No. 3 | 97 | 74 (76.3%) | 70 (72.2%) | 70 | 0 |
| 1978 | Go. Alb. 76, Nos. 1～3 | SnII. Alb. 74, No. 4 | 360 | 352 (97.8%) | 146 (40.6%) | 146 | 0 |
| | | YmI. Alb. 75, No. 7 | 327 | 91 (27.8%) | 73 (22.3%) | 73 | 0 |
| | | Fc. Het. 76, No. 3 | 455 | 405 (89.0%) | 310 (68.1%) | 310 | 0 |
| | | Ns. Alb. 77, No. 1 | 337 | 267 (79.2%) | 158 (46.9%) | 158 | 0 |
| | | Ty. Alb. 77, No. 2 | 577 | 300 (52.0%) | 114 (19.8%) | 114 | 0 |
| 1978 | Go. Het. 74, No. 1 | YmI. Alb. 75, No. 7 | 137 | 111 (81.0%) | 99 (72.3%) | 99 | 0 |
| 1979 | Go. Alb. 76, Nos. 4～6 | YmII. Alb. 78, No. 1 | 341 | 241 (70.7%) | 110 (32.3%) | 110 | 0 |
| 1980 | Go. Alb. 78, No. 7 | Hr. Alb. 79, Nos. 2～5 | 156 | 122 (78.2%) | 99 (63.5%) | 99 | 0 |
| | Go. Het. 78, No. 2 | Hr. Alb. 79, Nos. 2～5 | 215 | 75 (34.9%) | 73 (34.0%) | 73 | 0 |

each of five stocks, Sn II, Ym I, Ns, Ty and Ym II, and four male albinos of the Hr stock, 146, 73 158, 114, 110 and 99 feeding tadpoles, 700 in total, were produced, respectively. These tadpoles were all of the wild-type. When three female albinos were mated with a heterozygous male of the Fc stock, 310 feeding tadpoles were produced. These tadpoles were all of the wild-type.

When a heterozygous female was mated with a male albino of each of five stocks, Ex, Sn I, Br, Sn II and Tj, and two male albinos of the Ym I stock and four male albinos of the Hr stock, 94, 77, 84, 75, 70, 168 and 73 feeding tadpoles, 641 in total, were produced, respectively. These tadpoles were all of the wild-type. When a heterozygous female was mated with a heterozygous male of the Km stock, 74 feeding tadpoles were produced. All these tadpoles were also of the wild-type.

### 8. Females of the Fc stock and males of the other 12 stocks

During the years from 1976 to 1984, mating experiments were made between females homozygous or heterozygous for the albino gene of the Fc stock and males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 21).

When one or two female albinos were mated with a male albino of each of four stocks, Sn II, Tj, Sn I and Ym II, and four male albinos of the Hr stock, 46, 55, 94, 275 and 81 feeding tadpoles (stage III), 551 in total, were produced, respectively. These tadpoles were all albinos. In contrast, 42 feeding tadpoles produced from a mating between a female albino of the Fc stock and a male albino of the Ym I stock were all of the wild-type. When the same female was mated with a heterozygous male of each of the Km and Go stocks, 72 and 56 feeding tadpoles (stage III), 128 in total, were produced. These tadpoles were all of the wild-type.

One or two heterozygous females of the Fc stock were mated with a male albino of each of three stocks, Ex, Sn II and Br, and four male albinos of the Hr stock, 152, 191, 155 and 76 feeding tadpoles, 574 in total, were produced, respectively. Of these tadpoles, 77, 97, 72 and 39, 285 in total, were of the wild-type, and 75, 94, 83 and 37, 289 in total, were albinos. Thus, the wild-type and albinic tadpoles were produced in a ratio of approximately 1 : 1. When two heterozygous females were mated with a male albino of each of four stocks, Ym I, Go, Ns and Ty, 17, 93, 61 and 86 feeding tadpoles, 257 in total, were produced, respectively. All these tadpoles were of the wild-type.

In the breeding season of 1981, twenty 3-year-old female albinos of the Fc stock which were produced in 1978 from matings, Fc. Het. 76♀, Nos. 1 and 2 × Fc. Het. 76♂, Nos. 2 and 3, were injected with bullfrog pituitaries. The results showed that ovulation occurred in all the 20 females and 819～1363 eggs, 1038.6 eggs on the average, were laid by each female. Of a total of 20772 eggs laid by these 20 female albinos, only four (0.02%) were normally colored, while the others were light-colored eggs. However, these four normal-colored eggs (0.11%) were found in a total of 3611 eggs laid by three females. More specifically, one, one and two normal-colored eggs were found in 1214 eggs of female Fc. Alb. 78♀, No. 6, 1113 eggs of female Fc. Alb. 78♀, No. 7 and 1284 eggs of

TABLE 21

Results of mating experiments between females of the Fc stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1976 | Fc. Alb. 74, No. 1 | SnII. Alb. 74, No. 2 | 69 | 66 (95.7%) | 46 (66.7%) | 0 | 46 |
| | | YmI. Alb. 73, No. 5 | 67 | 49 (73.1%) | 42 (62.7%) | 42 | 0 |
| | | Km. Het. 75, No. 1 | 81 | 76 (93.8%) | 72 (88.9%) | 72 | 0 |
| | | Go. Het. 75, Nos. 1～4 | 135 | 77 (57.0%) | 56 (41.5%) | 56 | 0 |
| | | Tj. Alb. 74, No. 3 | 81 | 64 (79.0%) | 55 (67.9%) | 0 | 55 |
| 1977 | Fc. Alb. 74, No. 1 | SnI. Alb. 74, No. 4 | 133 | 127 (95.5%) | 94 (70.7%) | 0 | 94 |
| 1978 | Fc. Het. 76, Nos. 1, 2 | Ex. Alb. 74, No. 4 | 176 | 164 (93.2%) | 152 (86.4%) | 77 | 75 |
| | | SnII. Alb. 74, No. 4 | 236 | 227 (96.2%) | 191 (80.9%) | 97 | 94 |
| | | Br. Alb. 75, No. 5 | 218 | 178 (81.7%) | 155 (71.1%) | 72 | 83 |
| | | YmI. Alb. 75, No. 7 | 91 | 19 (20.9%) | 17 (18.7%) | 17 | 0 |
| | | Go. Alb. 76, No. 3 | 112 | 109 (97.3%) | 93 (83.0%) | 93 | 0 |
| | | Ns. Alb. 77, No. 1 | 124 | 61 (49.2%) | 61 (49.2%) | 61 | 0 |
| | | Ty. Alb. 77, No. 4 | 138 | 116 (84.1%) | 86 (62.3%) | 86 | 0 |
| 1930 | Fc. Alb. 78, Nos. 2, 3 | YmII. Alb. 78, No. 2 | 350 | 322 (92.0%) | 275 (78.6%) | 0 | 275 |
| | | Hr. Alb. 79, Nos. 2～5 | 236 | 88 (37.3%) | 81 (34.3%) | 0 | 81 |
| | Fc. Het. 77, No. 9 | Hr. Alb. 79, Nos. 2～5 | 213 | 82 (38.5%) | 76 (35.7%) | 39 | 37 |
| 1981 | Fc. Alb. 78, Nos. 6～10 | SnI. Alb. 77, No. 7 | 254 [4] | 250 [4] (98.4%) | 178 [4] (70.1%) | 0 | 178 [4] |
| | | | 4* | 4* | 3* | 3* | 0 |
| 1984 | (Fc)/Sn I. W 81, No. 1 | (Fc)/Sn I. W 81, No. 1 | 291 | 266 (91.4%) | 164 (56.4%) | 130 | 34 |
| | | Fc. Alb. 80, No. 4 | 347 | 321 (92.5%) | 188 (54.2%) | 110 | 78 |
| | | Fc. Het. 78, No. 5 | 212 | 164 (77.4%) | 110 (51.9%) | 89 | 21 |
| | Fc. Alb. 80, No. 11 | (Fc)/Sn I. W 81, No. 1 | 351 | 332 (94.6%) | 210 (59.8%) | 107 | 103 |
| | Fc. Het. 78, No. 10 | (Fc)/Sn I. W 81, No. 1 | 661 | 637 (96.4%) | 504 (76.2%) | 362 | 142 |

[ ] Numbers of grayish yellow-colored eggs and tadpoles raised from these eggs

* Numbers of normal-colored eggs and tadpoles raised from these eggs

(Fc)/Sn I. W, wild-type frogs raised from normal-colored eggs in 1981

female Fc. Alb. 78♀, No. 10, respectively (Plate V, 39, 40). The four normal-colored eggs were inseminated with sperm of a male albino of the Sn I stock, Sn I. Alb. 77♂, No. 7, together with 254 light-colored eggs laid by five females. It was found that three of the four normal-colored eggs developed into normally feeding tadpoles. All of them were of the wild-type in coloration, normally metamorphosed and became wild-type frogs (Table 21).

It was also noteworthy that four of the light-colored eggs of the four female

albinos were grayish yellow, while the others were light yellow. One of the four grayish-yellow eggs was laid by the above female, Fc. Alb. 78♀, No. 7. The other three were found in 965 eggs of female Fc. Alb. 78♀, No. 8, 819 eggs of female Fc. Alb. 78♀, No. 9 and 1284 eggs of female Fc. Alb. 78♀, No. 10. When these four grayish-yellow eggs were inseminated with sperm of a male albino of the Sn I stock together with 250 light-yellow eggs, they developed into albinic feeding tadpoles, all of which became edematous and died without attaining the metamorphosing stage. Of the light-yellow eggs, 178 became albinic feeding tadpoles and 124 became albinic frogs after normal metamorphosis (Table 21).

Two of the three wild-type frogs raised from normal-colored eggs attained sexual maturity in the breeding season of 1984. One of the two frogs was a female, (Fc)/Sn I. W 81♀, No. 1, and the other was a male, (Fc)/Sn I. W 81♂, No. 1. A wild-type female laid only normal-colored eggs. From a mating of these female and male, 164 feeding tadpoles at stage III were obtained. Of these tadpoles, 130 were of the wild-type, while 34 were Sn I-type albinos. From the same female mated with a male Fc-type albino and a male heterozygous for gene $a^f$ of the Fc-type albino, 110 and 89 tadpoles at stage III were of the wild-type and 78 and 21 were Fc/Sn I-type albinos, respectively.

Secondly, the above wild-type male, (Fc)/Sn I. W 81♂, No. 1, raised from a normal-colored egg was mated with a female Fc-type albino and a female heterozygous for gene $a^f$ of the Fc-type albino. It was found that 210 and 504 feeding tadpoles were obtained from these two matings. Of these tadpoles, 107 and 362 were of the wild-type and 103 and 142 were Fc/Sn I-type albinos, respectively.

It was observed that no Fc-type albinos were produced from any of the above matings. Thus, it seems evident that reverse mutation of the albino allele to the wild-type allele occurred in a frequency of about 0.11% in the eggs of the three female Fc-type albinos.

### 9. Females of the Tj stock and males of the other 12 stocks

During the years from 1976 to 1980, mating experiments were conducted between females homozygous or heterozygous for the albino gene of the Tj stock and males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 22). When one, two or three female albinos were mated with a male albino of each of five stocks, Sn II, Ex, Sn I, Br and Ym II, and four male albinos of the Hr stock, 89, 133, 54, 21, 170 and 73 feeding tadpoles, 540 in total, were produced, respectively. These tadpoles were all albinos. When one or two female albinos were mated with a male albino of each of the Ym I and Go stocks, 88 and 113, 201 in total, became feeding tadpoles, respectively. These tadpoles were all of the wild-type. When a female albino was mated with two heterozygous males of the Km stock and five heterozygous males of the Go stock, 210 and 301 feeding tadpoles, 511 in total, were produced, respectively. All these tadpoles were also of the wild-type.

When two heterozygous females of the Tj stock were mated with a heterozygous

TABLE 22

Results of mating experiments between females of the Tj stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1976 | Tj. Alb. 74, No. 1 | SnII. Alb. 74, No. 2 | 208 | 156 (75.0%) | 89 (42.8%) | 0 | 89 |
| | | YmI. Alb. 73, No. 5 | 249 | 157 (63.1%) | 88 (35.3%) | 88 | 0 |
| | | Km. Het. 75, Nos. 2, 3 | 543 | 348 (64.1%) | 210 (38.7%) | 210 | 0 |
| | | Go. Het. 75, Nos. 1~5 | 1059 | 439 (41.5%) | 301 (28.4%) | 301 | 0 |
| 1977 | Tj. Alb. 74, Nos. 2, 3 | Ex. Alb. 74, No. 2 | 534 | 215 (40.3%) | 133 (24.9%) | 0 | 133 |
| | | SnI. Alb. 74, No. 4 | 270 | 91 (33.7%) | 54 (20.0%) | 0 | 54 |
| | | Br. Alb. 75, No. 4 | 289 | 28 ( 9.7%) | 21 ( 7.3%) | 0 | 21 |
| | | Go. Alb. 76, No. 1 | 646 | 139 (21.5%) | 113 (17.5%) | 113 | 0 |
| 1978 | Tj. Het. 75, Nos. 3, 4 | Fc. Het. 76, No. 3 | 522 | 246 (47.1%) | 128 (24.5%) | 100 | 28 |
| | | Ns. Alb. 77, No. 2 | 538 | 216 (40.1%) | 149 (27.7%) | 149 | 0 |
| | | Ty. Alb. 77, No. 3 | 495 | 302 (61.0%) | 211 (42.6%) | 211 | 0 |
| 1979 | Tj. Alb. 77, Nos. 4~6 | YmII. Alb. 78, No. 1 | 481 | 287 (59.7%) | 170 (35.3%) | 0 | 170 |
| 1980 | Tj. Alb. 77, No. 8 | Hr. Alb. 79, Nos. 2~5 | 191 | 75 (39.3%) | 73 (38.2%) | 0 | 73 |
| | Tj. Het. 77, No. 5 | Hr. Alb. 79, Nos. 2~5 | 184 | 70 (38.0%) | 65 (35.3%) | 33 | 32 |

male of the Fc stock, 128 feeding tadpoles were produced. Of these tadpoles, 100 were of the wild-type and 28 were albinos. When these two females were mated with a male albino of each of the Ns and Ty stocks, 149 and 211 feeding tadpoles (stage III), 360 in total, produced from these matings were all of the wild-type, respectively. When a heterozygous female was mated with four male albinos of the Hr stock, 65 feeding tadpoles were produced. Of these tadpoles, 33 were of the wild-type and 32 were albinos.

### 10. Females of the Ty stock and males of the other 12 stocks

In 1979, 1980 and 1984, mating experiments were made between females homozygous or heterozygous for the albino gene of the Ty stock and males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 23). When one or four female albinos were mated with a male albino of each of eight other stocks, Sn II, Km, Go, Tj, Ym II, Ex, Sn I and Fc, and four male albinos of the Hr stock, 49, 42, 28, 38, 49, 138, 127, 105 and 55 feeding tadpoles, 631 in total, were produced, respectively. These tadpoles were all of the wild-type. When four female albinos of the Ty stock were mated with a heterozygous male of the Ym I stock, 36 feeding tadpoles were produced. All these tadpoles were also of the wild-type. When the same female albinos were mated with a male albino of the Ns stock, 19 feeding tadpoles were produced. All these tadpoles were albinos.

TABLE 23

Results of mating experiments between females of the Ty stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1979 | Ty. Alb. 77, Nos. 1～4 | SnII. Alb. 74, No. 5 | 145 | 86 (59.3%) | 49 (33.8%) | 49 | 0 |
| | | YmI. Het. 75, No. 5 | 115 | 68 (59.1%) | 36 (31.3%) | 36 | 0 |
| | | Km. Alb. 77, No. 2 | 161 | 96 (59.6%) | 42 (26.1%) | 42 | 0 |
| | | Go. Alb. 76, No. 5 | 139 | 68 (48.9%) | 28 (20.1%) | 28 | 0 |
| | | Tj. Alb. 77, No.6 | 90 | 60 (66.7%) | 38 (42.2%) | 38 | 0 |
| | | Ns. Alb. 77, No. 3 | 176 | 58 (33.0%) | 19 (10.8%) | 0 | 19 |
| | | YmII. Alb. 78, No. 1 | 124 | 87 (70.2%) | 49 (39.5%) | 49 | 0 |
| | Ty. Het. 78, Nos. 1～4 | Ns. Alb. 77, No. 4 | 281 | 189 (67.3%) | 155 (55.2%) | 77 | 78 |
| | | Ns. Het. 78, No. 1 | 263 | 261 (99.2%) | 241 (91.6%) | 182 | 59 |
| 1980 | Ty. Alb. 77, No. 5 | Ex. Alb. 78, No. 6 | 141 | 141 (100 %) | 138 (97.9%) | 138 | 0 |
| | | SnI. Alb. 75, No. 6 | 133 | 132 (99.2%) | 127 (95.5%) | 127 | 0 |
| | | Fc. Alb. 78, No. 3 | 123 | 122 (99.2%) | 105 (85.4%) | 105 | 0 |
| | | Hr. Alb. 79, Nos. 2～5 | 131 | 59 (45.0%) | 55 (42.0%) | 55 | 0 |
| | Ty. Het. 78, No. 5 | Ex. Alb. 78, No. 5 | 127 | 126 (99.2%) | 124 (97.6%) | 124 | 0 |
| | | SnI. Alb. 75, No. 5 | 140 | 138 (98.6%) | 130 (92.9%) | 130 | 0 |
| | | Fc. Alb. 78, No. 3 | 147 | 145 (98.6%) | 139 (94.6%) | 139 | 0 |
| | | Hr. Alb. 79, Nos. 2～5 | 167 | 127 (76.0%) | 123 (73.7%) | 123 | 0 |
| 1984 | Ty. Het. 79, Nos. 6, 7 | Br. Alb. 79, No. 8 | 286 | 278 (97.2%) | 251 (87.8%) | 251 | 0 |

When one or two heterozygous females were mated with a male albino of each of four other stocks, Ex, Sn I, Br and Fc, and four male albinos of the Hr stock, 124, 130, 251, 139 and 123 feeding tadpoles, 767 in total, were produced, respectively. These tadpoles were all of the wild-type. When four heterozygous females were mated with a male albino of the Ns stock, 155 feeding tadpoles were produced. Of these tadpoles, 77 were of the wild-type and 78 were albinos. When the same females were mated with a heterozygous male of the Ns stock, 241 feeding tadpoles were produced. Of these tadpoles, 182 were of the wild-type and 59 were albinos.

### 11. Females of the Ns stock and males of the other 12 stocks

During the years from 1979 to 1984, mating experiments were made between females heterozygous for the albino gene of the Ns stock and males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 24). When two or four heterozygous females of the Ns stock were mated with a male albino of each of eight stocks, Ex, Sn I, Sn II, Ym II, Br, Fc, Km and Tj, and four male

TABLE 24

Results of mating experiments between females of the Ns stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Alibno |
| 1979 | Ns. Het. 78, Nos. 1～4 | YmI. Het. 77, No. 6 | 290 | 283 (97.6%) | 263 (90.7%) | 263 | 0 |
| | | Km. Alb. 77, No. 3 | 261 | 241 (92.3%) | 236 (90.4%) | 236 | 0 |
| | | Go. Het. 78, No. 6 | 343 | 330 (96.2%) | 310 (90.4%) | 310 | 0 |
| | | Tj. Alb. 77, No. 7 | 410 | 405 (98.8%) | 387 (94.4%) | 387 | 0 |
| | | Ty. Het. 78, No. 1 | 417 | 414 (99.3%) | 358 (85.9%) | 268 | 90 |
| | | Ty. Alb. 77, No. 6 | 397 | 395 (99.5%) | 335 (84.4%) | 173 | 162 |
| 1980 | Ns. Het. 78, Nos. 5, 6 | Ex. Alb. 78, No.6 | 222 | 215 (96.8%) | 206 (92.8%) | 206 | 0 |
| | | SnI. Alb. 75, No. 6 | 227 | 216 (95.2%) | 207 (91.2%) | 207 | 0 |
| | | SnII. Alb. 74, No. 8 | 214 | 210 (98.1%) | 193 (90.2%) | 193 | 0 |
| | | YmII. Alb. 78, No. 2 | 261 | 255 (97.7%) | 241 (92.3%) | 241 | 0 |
| | | Fc. Alb. 78, No. 3 | 278 | 276 (99.3%) | 244 (87.8%) | 244 | 0 |
| | | Hr. Alb. 79, Nos. 2～5 | 289 | 106 (36.7%) | 103 (35.6%) | 103 | 0 |
| 1984 | Ns. Het. 79, Nos. 7, 8 | Br. Alb. 79, No. 8 | 247 | 211 (85.4%) | 177 (71.7%) | 177 | 0 |

albinos of the Hr stock, 206, 207, 193, 241, 177, 244, 236, 387 and 103 feeding tadpoles, 1994 in total, were produced, respectively. These tadpoles were all of the wild-type. When four heterozygous females were mated with a heterozygous male of each of the Ym I and Go stocks, 263 and 310 feeding tadpoles, 573 in total, were produced, respectively. All these tadpoles were also of the wild-type.

When four heterozygous females were mated with a male albino of the Ty stock, 335 feeding tadpoles were produced. Of these tadpoles, 173 were of the wild-type and 162 were albinos. On the other hand, when the same heterozygous females were mated with a heterozygous male of the Ty stock, 358 feeding tadpoles were produced. Of these tadpoles, 268 were of the wild-type and 90 were albinos.

### 12. Females of the Ym II stock and males of the other 12 stocks

In 1979 and 1980, mating experiments were made between female albinos of the Ym II stock and males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 25). When one or three female albinos were mated with a male albino of each of six other stocks, Sn II, Tj, Ex, Sn I, Br and Fc, and four male albinos of the Hr stock, 478, 453, 131, 150, 119, 70 and 82 feeding tadpoles, 1483 in total, were produced. All these tadpoles were albinos. When the same one or three female albinos were mated with a male albino of each of the Km and Go stocks and two male albinos of each of the Ns and Ty stocks, 321, 315, 211 and 547 feeding tadpoles, 1394 in total, were produced. These tadpoles were all of

TABLE 25

Results of mating experiments between females of the YM II stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1979 | YmII. Alb. 78, Nos. 1～3 | SnII. Alb. 74, No. 5 | 558 | 556 (99.6%) | 478 (85.7%) | 0 | 478 |
| | | YmI. Het. 75, No. 5 | 365 | 358 (98.1%) | 306 (83.8%) | 306 | 0 |
| | | Km. Alb. 77, No. 2 | 435 | 424 (97.5%) | 321 (73.8%) | 321 | 0 |
| | | Go. Alb. 76, No. 5 | 388 | 380 (97.9%) | 315 (81.2%) | 315 | 0 |
| | | Tj. Alb. 77, No. 6 | 538 | 529 (98.3%) | 453 (84.2%) | 0 | 453 |
| | | Ns. Alb. 77, No. 3 | 432 | 226 (52.3%) | 192 (44.4%) | 192 | 0 |
| | | Ty. Alb. 77, No. 5 | 467 | 444 (95.1%) | 418 (89.5%) | 418 | 0 |
| 1980 | YmII. Alb. 78, No. 4 | Ex. Alb. 78, No. 5 | 155 | 151 (97.4%) | 131 (84.5%) | 0 | 131 |
| | | SnI. Alb. 75, No. 6 | 172 | 161 (93.6%) | 150 (87.2%) | 0 | 150 |
| | | Br. Alb. 78, No. 7 | 128 | 122 (95.3%) | 119 (93.0%) | 0 | 119 |
| | | Fc. Alb. 78, No. 3 | 159 | 105 (66.0%) | 70 (44.0%) | 0 | 70 |
| | | Ty. Alb. 79, No. 7 | 150 | 142 (94.7%) | 129 (86.0%) | 129 | 0 |
| | | Ns. Alb. 79, No. 4 | 206 | 20 ( 9.7%) | 19 ( 9.2%) | 19 | 0 |
| | | Hr. Alb. 79, Nos. 2～5 | 233 | 89 (38.2%) | 82 (35.2%) | 0 | 82 |

the wild-type in contrast to the above tadpoles. When three female albinos were mated with a heterozygous male of the YM I stock, 306 feeding tadpoles produced from these matings were all of the wild-type.

### 13. Females of the HR stock and males of the other 12 stocks

In the breeding season of 1981, mating experiments were made between female albinos of the HR stock and males homozygous or heterozygous for the albino genes of the other 12 stocks (Table 26). When two female albinos were mated with a male albino of each of three stocks, SN I, SN II and TJ, 60, 67 and 71 feeding tadpoles, 198 in total, were produced, respectively. All these tadpoles were albinos. When two female albinos were mated with a heterozygous male of each of four stocks, Ex, YM II, BR and Fc, 60, 109, 37 and 51 feeding tadpoles, 257 in total, were produced, respectively. Of these tadpoles, 33, 47, 20 and 21, 121 in total, were of the wild-type and 27, 62, 17 and 30, 136 in total, were albinos, respectively. Thus, the wild-type and albinic tadpoles were produced in a ratio of approximately 1: 1. On the other hand, when two female albinos were mated with a male albino of each of five stocks, KM, YM I, Go, Ns and TY, 76, 62, 58, 44 and 18, 258 in total, produced from these matings were all of the wild-type.

TABLE 26

Results of mating experiments between females of the HR stock and males of the other 12 stocks

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | |
|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino |
| 1981 | HR. Alb. 79, Nos. 1, 2 | EX. Het. 78, No. 3 | 133 | 129 (97.0%) | 60 (45.1%) | 33 | 27 |
| | | SNI. Alb. 75, No. 7 | 142 | 127 (89.4%) | 60 (42.3%) | 0 | 60 |
| | | SNII. Alb. 79, No. 9 | 144 | 138 (95.8%) | 67 (46.5%) | 0 | 67 |
| | | YMII. Het. 79, No. 2 | 221 | 197 (89.1%) | 109 (49.3%) | 47 | 62 |
| | | BR. Het. 79, No. 1 | 124 | 105 (84.7%) | 37 (29.8%) | 20 | 17 |
| | | FC. Het. 77, No. 4 | 114 | 105 (92.1%) | 51 (44.7%) | 21 | 30 |
| | HR. Alb. 79, Nos. 3, 4 | TJ. Alb. 77, No. 9 | 173 | 165 (95.4%) | 71 (41.0%) | 0 | 71 |
| | | KM. Alb. 79, No. 4 | 148 | 144 (97.3%) | 76 (51.4%) | 76 | 0 |
| | | YMI. Alb. 79, No. 9 | 183 | 176 (96.2%) | 62 (33.9%) | 62 | 0 |
| | | Go. Alb. 79, No. 7 | 156 | 152 (97.4%) | 58 (37.2%) | 58 | 0 |
| | | Ns. Alb. 79, No. 5 | 166 | 161 (97.0%) | 44 (26.5%) | 44 | 0 |
| | | TY. Alb. 79, No. 8 | 156 | 162 (97.4%) | 18 (11.5%) | 18 | 0 |

## 14. Summary of the results from the mating experiments

The results of mating experiments among 13 albino stocks (Tables 14~26) showed that these 13 albino stocks were divided into five groups. The first group consists of eight albino stocks, Ex, SN I, SN II, YM II, HR, BR, TJ and FC, while the second, third and fourth groups include the Go, YM I and KM stocks, respectively. The fifth group consists of two albino stocks, TY and Ns. The recessive albino genes of the five groups are named *a*, *b*, *c*, *d* and *e*, respectively.

TABLE 27

Five groups of the 13 albino stocks and their phenotypes and genotypes in *Rana nigromaculata* and *Rana brevipoda*

| Group | Stock | Abbreviation of stock | Phenotype | Genotype |
|---|---|---|---|---|
| 1 | Stock from an egg irradiated with 145 rads of X-rays | EX | RA | $a^e a^e$ |
| | Stock from a spermatozoon irradiated with 50 rads of neutrons | SN I | | |
| | Stock from a spermatozoon irradiated with 130 rads of neutrons | SN II | | |
| | Yamaguchi II | YM II | | |
| | Hiro | HR | | |
| | *R. brevipoda* from Konko | BR | | |
| | Tojo | TJ | TJ | $a^t a^t$ |
| | Fuchu | FC | FC | $a^f a^f$ |
| 2 | Gion | Go | Go | *bb* |
| 3 | Yamaguchi I | YM I | YM I | *cc* |
| 4 | Kamogata | KM | KM | *dd* |
| 5 | Toyomatsu | TY | TY | *ee* |
| | Nishinomiya | Ns | | |

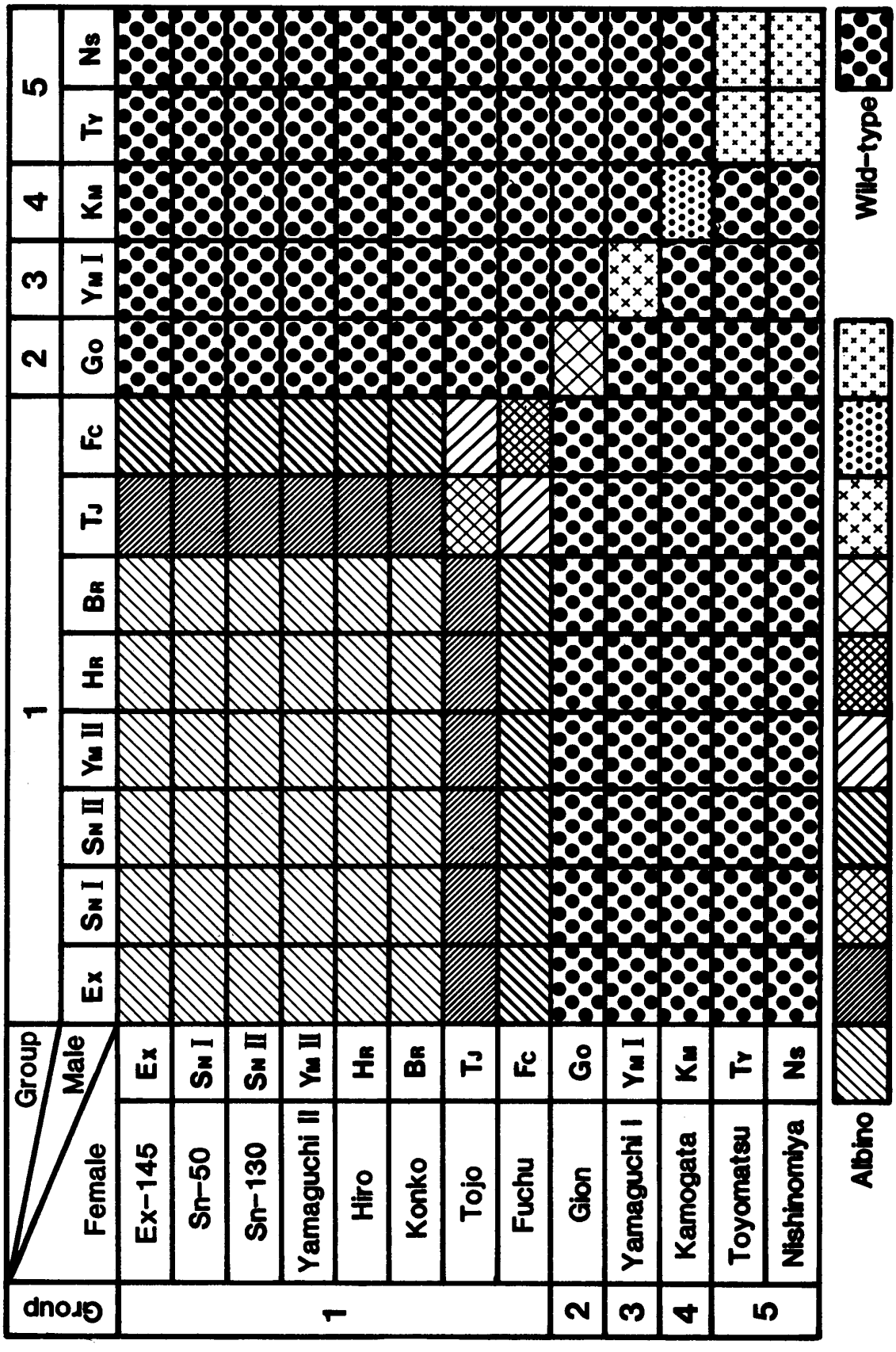


Fig. 2. Results of crossing experiments among 13 albino stocks.

While matings between albinos of the same group produce albinos only, those between albinos of different groups produce individuals of the wild-type alone. It is evident that albinos of different groups have different loci on chromosomes.

The eight albino stocks of the first group were definitely divided into three strains on the basis of differences in color of the dorsal surfaces and the pupils. Strain 1 consists of six albino stocks, Ex, Sn I, Sn II, Ym II, Hr and Br, in which the dorsal surfaces are light reddish-yellow and the pupils are bright red. Strain 2 includes the Tj stock, in which the dorsal surfaces are dull reddish-yellow and the pupils are deep red, while strain 3 includes the Fc stock, in which the dorsal surfaces are dark yellow and the pupils are dark grayish-red. Thus, the albinos of these three strains are due to three multiple alleles, $a^e$, $a^t$ and $a^f$ (Table 27; Fig. 2). The albinos of strain 1 are $a^e a^e$ in genotype and their phenotype is named Ra-type. The albinos of strain 2 are $a^t a^t$ in genotype and their phenotype is named Tj-type, while the albinos of strain 3 are $a^f a^f$ in genotype and their phenotype is named Fc-type.

When an albino of the Ra-type is mated with an albino of the Tj-type, the hybrid is an albino which is $a^e a^t$ in genotype and intermediate between the two types in phenotype. This phenotype is called Ra/Tj-type. When an albino of the Ra-type is mated with an albino of the Fc-type, the hybrid is $a^e a^f$ in genotype and intermediate between the two types in phenotype, being of Ra/Fc-type. When an albino of the Tj-type is mated with an albino of the Fc-type, the hybrid is $a^t a^f$ in genotype and intermediate between the two types in phenotype, being of Tj/Fc-type (Fig. 2).

When a mating is made between albinos of the first and second groups which differ from each other in albino gene locus, the hybrid is *AaBb* and of the wild-type in phenotype. The hybrids produced from matings between the first and third, the first and fourth, the first and fifth, the second and third, the second and fourth, the second and fifth, the third and fourth, the third and fifth, and the fourth and fifth groups are *AaCc*, *AaDd*, *AaEe*, *BbCc*, *BbDd*, *BbEe*, *CcDd*, *CcEe* and *DdEe* in genotype, respectively. All of these hybrids are of the wild-type in phenotype (Fig. 2).

Some of the female Tj-type ($a^t a^t$), Fc-type ($a^f a^f$) and Tj/Fc-type ($a^t a^f$) albinos laid a very few dark-colored eggs, together with abundant ordinary light-colored ones. From these unusual eggs, both albinic and wild-type tadpoles were produced by fertilization with sperm of male albinos of the first group (Tables 8, 9 and 28). Thereafter, it was confirmed that of a few unusual eggs laid by female Fc-type ($a^f a^f$) albinos, grayish-yellow and normal-colored eggs become Ra/Fc-type ($a^e a^f$) albinos and wild-type individuals, respectively, by fertilization with sperm of male albinos ($a^e a^e$) of the Sn I stock (Table 21). A wild-type female lays only normal-colored eggs. From a mating between a female and a male of these wild-type individuals, both wild-type individuals and Ra-type ($a^e a^e$) albinos are produced. From a mating between the above wild-type female and a male Fc-type ($a^f a^f$) albino or a male heterozygous for gene $a^f$, both wild-type individuals and Ra/Fc-type ($a^e a^f$) albinos are produced. From the above wild-

type male, both wild-type individuals and RA/FC-type ($a^e a^f$) albinos are also obtained by mating with a female FC-type ($a^f a^f$) albino or a female heterozygous for gene $a^f$. No FC-type ($a^f a^f$) albinos have been produced from any of these matings (Table 21). Thus, it is evident that the wild-type individuals raised from unusual normal-colored eggs is $Aa^e$ in genotype, and that gene $a^f$ has been lost in the normal-colored eggs.

## *III. Relationship between different albino genes*

### 1. Three strains of albinos in the first group

In order to clarify the characters of the three albino strains in the first group, the mating experiments presented in Table 28 were performed in 1979 by using the following individuals homozygous or heterozygous for the albino gene of each strain.

(1) Three female albinos, TJ/FC. Alb. 78♀, Nos. 1~3, produced in 1978 from a mating, TJ. Het. 75♀, No. 3×FC. Het. 76♂, No. 3 ($Aa^t$♀×$Aa^f$♂). These females are $a^t a^f$ in genotype and the TJ/FC-type in phenotype.

(2) A female albino, TJ. Alb. 77♀, No. 7, and a male albino of the TJ-type, TJ. Alb. 77♂, No. 7, produced in 1977 from a mating, TJ. Het. 75♀, No. 1× TJ. Alb. 74♂, No. 5 ($Aa^t$♀×$a^t a^t$♂). These female and male are $a^t a^t$ in genotype and the TJ-type in phenotype.

(3) Two female albinos, FC/SN II. Alb. 78♀, Nos. 1 and 2, and three male albinos, FC/SN II. Alb. 78♂, Nos. 1~3, produced in 1978 from a mating, FC. Het. 76♀, No. 1×SN II. Alb. 74♂, No. 4 ($Aa^f$♀×$a^e a^e$♂). These female and male albinos are $a^e a^f$ in genotype and the RA/FC-type in phenotype.

(4) Two female albinos, FC. Alb. 78♀, Nos. 2 and 3, two male albinos, FC. Alb. 78♂, Nos. 1 and 2, and six heterozygous females, FC. Het. 78♀, Nos. 3~8, produced in 1978 from a mating, FC. Het. 76♀, No. 1×FC. Het. 76♂, No. 2 ($Aa^f$♀×$Aa^f$♂). The female and male albinos are $a^f a^f$ in genotype and the FC-type in phenotype, while the heterozygous females are $Aa^f$ in genotype.

(5) A female albino, SN II. Alb. 74♀, No. 8, a male albino, SN II. Alb. 74♂, No. 6, and two heterozygous females, SN II. Het. 74♀, Nos. 4 and 5, produced in 1974 from a mating, SN II. Het. 72♀, No. 1×SN II. Het. 72♂, No. 1 ($Aa^e$♀× $Aa^e$♂). The female and male albinos are $a^e a^e$ in genotype and the RA-type in phenotype, while the heterozygous females are $Aa^e$ in genotype.

#### a. Production of $a^e a^f$ albinos

i) While 78 feeding tadpoles produced from a mating, SN II. Alb. 74♀, No. 8×SN II. Alb. 74♂, No. 6, were all RA-type ($a^e a^e$) albinos, 38 feeding tadpoles obtained from the same female by mating with a male FC-type ($a^f a^f$) albino, SN II. Alb. 74♀, No. 8×FC. Alb. 78♂, No. 1, were all RA/FC-type ($a^e a^f$) albinos. When the same female albino was mated with a male $a^e a^f$ albino, SN II. Alb. 74♀, No. 8×FC/SN II. Alb. 78♂, No. 1, 75 feeding tadpoles

were produced. Of these tadpoles, 40 were RA-type ($a^e a^e$) albinos and 35 were RA/FC-type ($a^e a^f$) albinos (Table 28).

ii) Of 307 feeding tadpoles produced from matings between two heterozygous $Aa^e$ females and a male $a^e a^e$ albino, SN II. Het. 74♀, Nos. 4 and 5 × SN II. Alb. 74♂, No. 6, 165 and 142 were wild-type ones and RA-type ($a^e a^e$) albinos, respectively. Similarly, 187 and 160 of 347 feeding tadpoles produced from matings between the same females and a male FC-type ($a^f a^f$) albino, SN II. Het. 74♀, Nos. 4 and 5 × FC. Alb. 78♂, No. 1, were wild-type ones and RA/FC-type ($a^e a^f$) albinos, respectively. When the same females were mated with a male RA/FC-type ($a^e a^f$) albino, SN II. Het. 74♀, Nos. 4 and 5 × FC/SN II. Alb. 78♂, No. 1, 228, 107 and 100 of 435 feeding tadpoles obtained were wild-type ones, RA-type ($a^e a^e$) albinos and RA/FC-type ($a^e a^f$) albinos, respectively (Table 28).

iii) Mating experiments were made between two females and one male of FC-type ($a^f a^f$) albinos, FC. Alb. 78♀, Nos. 2 and 3 × FC. Alb. 78♂, No. 1. One of the eggs laid by these females was grayish-yellow, while the others were light yellow. Feeding tadpoles were raised from 211 eggs including the grayish-yellow one. All these tadpoles were FC-type ($a^f a^f$) albinos. When mating experiments between the same females and a male RA-type ($a^e a^e$) albino, FC. Alb. 78♀, Nos. 2 and 3 × SN II. Alb. 74♂, No. 6, were made, one of 341 eggs was normal-colored, while the remaining 340 were light-yellow eggs. Of the light-yellow eggs, 265 grew into feeding tadpoles which were RA/FC-type ($a^e a^f$) albinos. The single normal-colored egg became a normal-colored feeding tadpole which completed metamorphosis and attained sexual maturity. This frog was of the wild-type. Mating experiments between the same two females and a male $a^e a^f$ albino, FC. Alb. 78♀, Nos. 2 and 3 × FC/SN II. Alb. 78♂, No. 2, were made. Of 344 eggs including a grayish-yellow egg, 253 became feeding tadpoles. Of these tadpoles, 133 including one raised from a grayish-yellow egg were RA/FC-type ($a^e a^f$) albinos, and 120 were FC-type ($a^f a^f$) albinos (Table 28).

iv) From matings between six heterozygous females and one male homozygous for the $a^f$ gene, FC. Het. 78♀, Nos. 3~8 × FC. Alb. 78♂, No. 2, 334 feeding tadpoles were produced. Of these tadpoles, 172 were of the wild-type and 162 were FC-type ($a^f a^f$) albinos. From matings between the same females and a male RA-type ($a^e a^e$) albino, FC. Het. 78♀, Nos. 3~8 × SN II. Alb. 74♂, No. 6, 370 feeding tadpoles were produced. Of these tadpoles, 192 were of the wild-type and 178 were RA/FC-type ($a^e a^f$) albinos. From matings between the same six females and a male RA/FC-type ($a^e a^f$) albino, FC. Het. 78♀, Nos. 3~8 × FC/SN II. Alb. 78♂, No. 2, 573 feeding tadpoles were produced. Of these tadpoles, 298 were of the wild-type, 134 were FC-type ($a^f a^f$) albinos and 141 were RA/FC-type ($a^e a^f$) albinos. Thus, these three kinds of tadpoles revealed a ratio of approximately 2: 1: 1 (Table 28).

TABLE 28
Results of mating experiments between females and males homo-

| Parents | | No. of eggs | No. of normal cleavages |
|---|---|---|---|
| Female | Male | | |
| SnII. Alb. 74, No. 8 | SnII. Alb. 74, No. 6 | 178 | 172 (96.6%) |
| | Fc. Alb. 78, No. 1 | 126 | 76 (60.3%) |
| | (Fc. Het. ♀ × SnII. Alb. ♂) Alb. 78, No. 1 | 172 | 165 (95.9%) |
| SnII. Het. 74, Nos. 4, 5 | SnII. Alb. 74, No. 6 | 330 | 326 (98.8%) |
| | Fc. Alb. 78, No. 1 | 381 | 354 (92.9%) |
| | (Fc. Het. ♀ × SnII. Alb. ♂) Alb. 78, No. 1 | 481 | 472 (98.1%) |
| Fc. Alb. 78, Nos. 2, 3 | Fc. Alb. 78, No. 1 | 321 | 287 (89.4%) |
| | SnII. Alb. 74, No. 6 | 341 | 324 (95.0%) |
| | (Fc. Het. ♀ × SnII. Alb. ♂) Alb. 78, No. 2 | 344 | 327 (95.1%) |
| Fc. Het. 78, Nos. 3~8 | Fc. Alb. 78, No. 2 | 767 | 432 (56.3%) |
| | SnII. Alb. 74, No. 6 | 642 | 453 (70.6%) |
| | (Fc. Het. ♀ × SnII. Alb. ♂) Alb. 78, No. 2 | 914 | 690 (75.5%) |
| (Fc. Het. ♀ × SnII. Alb. ♂) Alb. 78, Nos. 1, 2 | Fc. Alb. 78, No. 2 | 211 | 186 (88.2%) |
| | SnII. Alb. 74, No. 6 | 258 | 254 (98.4%) |
| | (Fc. Het. ♀ × SnII. Alb. ♂) Alb. 78, No. 3 | 326 | 303 (92.9%) |
| Tj. Alb. 77, No. 7 | Tj. Alb. 77, No. 7 | 169 | 141 (83.4%) |
| | (Fc. Het. ♀ × SnII. Alb. ♂) Alb. 78, No. 3 | 226 | 183 (81.0%) |
| (Tj. Het. ♀ × Fc. Het. ♂) Alb. 78, Nos. 1~3 | Tj. Alb. 77, No. 7 | 502 | 486 (96.8%) |
| | (Fc. Het. ♀ × SnII. Alb. ♂) Alb. 78, No. 3 | 432 | 400 (92.6%) |

[ ] Number of tadpoles raised from normal-colored eggs

v) Of 75 feeding tadpoles produced from matings between two female Ra/Fc-type ($a^e a^f$) albinos and a male Fc-type ($a^f a^f$) albino, Fc/Sn II. Alb. 78♀, Nos. 1 and 2 × Fc. Alb. 78♂, No. 2, 33 were Fc-type ($a^f a^f$) albinos and 42 were Ra/Fc-type ($a^e a^f$) albinos. When matings were made between the same females and a male Ra-type ($a^e a^e$) albino, Fc/Sn II. Alb. 78♀, Nos. 1 and 2 × Sn II. Alb. 74♂, No. 6, 180 feeding tadpoles were produced. Of these tadpoles, 92 were Ra-type ($a^e a^e$) albinos and 88 were Ra/Fc-type ($a^e a^f$) albinos. When the

or heterozygous for each of albino genes $a^e$, $a^t$ and $a^f$ (1979)

| No. of feeding tadpoles (St. III) | | | | | | | |
|---|---|---|---|---|---|---|---|
| Total | Wild | Albino | | | | | |
| | | RA | RA/TJ | TJ | RA/FC | TJ/FC | FC |
| 78 (43.8%) | 0 | 78 | 0 | 0 | 0 | 0 | 0 |
| 38 (30.2%) | 0 | 0 | 0 | 0 | 38 | 0 | 0 |
| 75 (43.6%) | 0 | 40 | 0 | 0 | 35 | 0 | 0 |
| 307 (93.0%) | 165 | 142 | 0 | 0 | 0 | 0 | 0 |
| 347 (91.1%) | 187 | 0 | 0 | 0 | 160 | 0 | 0 |
| 435 (90.4%) | 228 | 107 | 0 | 0 | 100 | 0 | 0 |
| 211 (65.7%) | 0 | 0 | 0 | 0 | 0 | 0 | 211 |
| 266 (78.0%) | 1 [1] | 0 | 0 | 0 | 265 | 0 | 0 |
| 253 (73.5%) | 0 | 0 | 0 | 0 | 133 | 0 | 120 |
| 334 (43.5%) | 172 | 0 | 0 | 0 | 0 | 0 | 162 |
| 370 (57.6%) | 192 | 0 | 0 | 0 | 178 | 0 | 0 |
| 573 (62.7%) | 298 | 0 | 0 | 0 | 141 | 0 | 134 |
| 75 (35.5%) | 0 | 0 | 0 | 0 | 42 | 0 | 33 |
| 180 (69.8%) | 0 | 92 | 0 | 0 | 88 | 0 | 0 |
| 261 (80.1%) | 0 | 53 | 0 | 0 | 141 | 0 | 67 |
| 127 (75.1%) | 0 | 0 | 0 | 127 | 0 | 0 | 0 |
| 176 (77.9%) | 0 | 0 | 82 | 0 | 0 | 94 | 0 |
| 425 (84.7%) | 0 | 0 | 0 | 203 | 0 | 222 | 0 |
| 365 (84.5%) | 1 [1] | 0 | 83 | 0 | 91 | 99 | 91 |

same females were mated with a male RA/FC-type ($a^e a^f$) albino, FC/SN II. Alb. 78♀, Nos. 1 and 2 × FC/SN II. Alb. 78♂, No. 3, 53 of 261 feeding tadpoles were RA-type ($a^e a^e$) albinos, 141 were RA/FC-type ($a^e a^f$) albinos and 67 were FC-type ($a^f a^f$) albinos (Table 28).

b. Production of $a^e a^t$ and $a^t a^f$ albinos

i) All 127 feeding tadpoles produced from a mating between a female and a

male of $a^t a^t$ albinos, TJ. Alb. 77♀, No. 7×TJ. Alb. 77♂, No. 7, were TJ-type ($a^t a^t$) albinos. In contrast, when the same female was mated with a male $a^e a^f$ albino, TJ. Alb. 77♀, No. 7×FC/SN II. Alb. 78♂, No. 3, 82 of 176 feeding tadpoles were RA/TJ-type ($a^e a^t$) albinos and 94 were TJ/FC-type ($a^t a^f$) albinos (Table 28).

ii) Of three female $a^t a^f$ albinos (Nos. 1~3), female No. 1 laid 1653 eggs including one normal-colored and female No. 2 laid 2019 eggs including two normal-colored. These three normal-colored eggs (0.08%) were found in a total of 3672 eggs of the two female albinos (Nos. 1 and 2). The other eggs of these two females were light yellow, except two eggs of female No. 1 which were grayish yellow. Female No. 3 laid 1227 light-colored eggs, of which one was grayish yellow. Of these eggs laid by the three females, 502 light-colored eggs including three grayish-yellow ones were inseminated with sperm of a male TJ-type ($a^t a^t$) albino (No. 7).

The results showed that 425 including the three grayish-yellow eggs became normally feeding tadpoles. Of these tadpoles, 203 including two raised from grayish-yellow eggs were TJ-type ($a^t a^t$) albinos and 222 including one raised from a grayish-yellow egg were TJ/FC-type ($a^t a^f$) albinos. When 431 light-colored eggs and one of the normal-colored eggs were inseminated with sperm of a male RA/FC-type ($a^e a^f$) albino, FC/SN II. Alb. 78♂, No. 3, 365 feeding tadpoles were produced. One of these tadpoles was raised from the normal-colored egg and was of the wild-type. This tadpole completed metamorphosis and became a frog which did not differ from the control *Rana nigromaculata*. Of the other 364 tadpoles, 83 were RA/TJ-type ($a^e a^t$) albinos, 91 were RA/FC-type ($a^e a^f$) albinos, 99 were TJ/FC-type ($a^t a^f$) albinos and the remaining 91 were FC-type ($a^f a^f$) albinos (Table 28). These four kinds of albinos should be produced in a ratio of 1: 1: 1: 1. It seems also evident that reverse mutation of the albino allele to the wild-type allele occurred in a frequency of about 0.08% in the eggs of the two female TJ/FC-type albinos.

## 2. Difference between albinos of the first and second groups

### a. Relationship of $a^e a^e BB$ and $AAbb$ albinos

A mating experiment was made in 1977 between a female $a^e a^e BB$ albino, SN II. Alb. 74♀, No. 2, and a male $AAbb$ albino, Go. Alb. 76♂, No. 1. It was found that all the offspring, $Aa^e Bb$, were of the wild-type. Of these offspring, three 2-year-old females, Nos. 1~3, were mated in 1979 with a 5-year-old male $a^e a^e BB$ albino, SN II. Alb. 74♂, No. 7, which was obtained in 1974 from a mating, SN II. Het. 72♀, No. 1×SN II. Het. 72♂, No. 1. The results showed that 158 of 314 feeding tadpoles soon after beginning to eat (stage 25) were of the wild-type and 156 were RA-type albinos.

As the offspring between a female $a^e a^e BB$ albino and a male $AAbb$ albino are all of the wild-type and $Aa^e Bb$ in genotype, their gametes should be divided into $Ab$ and $a^e B$, if $a^e$ and $b$ are situated on homologous chromosomes and no crossing-

TABLE 29

Results of mating experiments and gynogenesis in females heterozygous for albino genes $a^e$ and $b$ (1979)

| Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. 25) | | | |
|---|---|---|---|---|---|---|---|
| Female | Male | | | Total | Wild | Albino | |
| | | | | | | Go | Ra |
| (SnII. Alb. ♀ × Go. Alb. ♂) 77, Nos. 1~3 | SnII. Alb. 74, No. 7 | 515 | 508 (98.6%) | 314 (61.0%) | 158 | 0 | 156 |
| (SnII. Alb. ♀ × Go. Alb. ♂) 77, No. 4 | *R. clamitans* | 184 | 181 (98.4%) | 0 | 0 | 0 | 0 |
| | GH | 220 | 183 (83.2%) | 116 (52.7%) | 32 | 26 | 58 |
| | GD | 287 | 125 (43.6%) | 66 (23.0%) | 43 | 7 | 16 |
| (SnII. Alb. ♀ × Go. Alb. ♂) 77, No. 5 | GH | 225 | 201 (89.3%) | 137 (60.9%) | 34 | 38 | 65 |
| | GD | 386 | 261 (67.6%) | 118 (30.6%) | 79 | 15 | 24 |
| (SnII. Alb. ♀ × Go. Alb. ♂) 77, Nos. 4, 5 | GH | 445 | 384 (86.3%) | 253 (56.9%) | 66 | 64 | 123 |
| | GD | 673 | 386 (57.4%) | 184 (27.3%) | 122 | 22 | 40 |

GH, haploid gynogenesis GD, diploid gynogenesis

over occurs between the two genes. When the eggs of this female are gynogenetically developed, all of them should become albinos; no wild-type individuals will be produced. However, if $a^e$ and $b$ are situated on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%, their gametes should be divided into four kinds, $AB$, $Ab$, $a^eB$ and $a^eb$. In this case, gynogenetically developed haploid and diploid tadpoles should include wild-type and albinic ones in a ratio of 1: 3. These albinos also should include $Ab$ or $AAbb$, $a^eB$ or $a^ea^eBB$ and $a^eb$ or $a^ea^ebb$ individuals in a ratio of 1: 1: 1.

In 1979, 253 haploid tadpoles 13 days after hatching were obtained by pseudofertilization with UV-irradiated sperm of *Rana clamitans* from 445 eggs of two wild-type females which had been produced in 1977 from a mating between a female $a^ea^e$ albino and a male $bb$ albino, Sn II. Alb. 74♀, No. 2 × Go. Alb. 76♂, No. 1. Of these haploid tadpoles, 66 were of the wild-type, 64 were Go-type albinos and 123 were Ra-type albinos. The tadpoles of the wild-type were considered to be $AB$ in genotype, while the albinos of the Ra-type were considered to be a mixture of $a^eB$ and $a^eb$. Thus, three kinds of phenotypes of the haploid tadpoles, which are $AB$, $Ab$ and $a^eB+a^eb$ in genotype, seem to have been produced in a ratio of approximately 1: 1: 2.

When 673 eggs of the above two wild-type females ($Aa^eBb$, Nos. 4 and 5) were refrigerated after insemination with UV-irradiated sperm of *Rana clamitans* in order to obtain gynogenetic diploids by suppressing extrusion of the second polar body, 184 of them became normally feeding diploid tadpoles (stage 25). Of these tadpoles, 122 were of the wild-type, 22 were Go-type albinos and 40 were Ra-type albinos. The finding that the albinos were much fewer than the wild-type tadpoles may be explained by the fact that post-reductional separation of a

chromosomal segment bearing the albino gene has occurred owing to crossing-over in some eggs and also by the fact that albinic eggs are weaker in resistibility to the cold-treatment than normal-colored ones. The existence of 40 $a^e a^e BB$ albinos in contrast to 22 *AAbb* ones is probably attributable to the fact that the $a^e a^e bb$ albinos are the RA-type in phenotype inasmuch as gene $a^e$ is epistatic when $a^e$ and *b* occur together. Thus, it is believed that genes $a^e$ and *b* are situated on non-homologous chromosomes or such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50% (Table 29).

b. Relationship of $a^t a^t BB$ and *AAbb* albinos

The offspring $Aa^tBb$ between female TJ-type ($a^t a^t BB$) albinos and male Go-type (*AAbb*) albinos are all of the wild-type. If genes $a^t$ and *b* are situated on homologous chromosomes and no crossing-over occurs, the $Aa^tBb$ females produce two kinds of eggs, *Ab* and $a^tB$. Gynogenetic haploids and gynogenetic diploids obtained from these eggs should be all albinos, as they are *Ab* and $a^tB$, and *AAbb* and $a^t a^t BB$, respectively. In this case, no wild-type tadpoles are produced. However, if crossing-over occurs between the two loci in a frequency of nearly 50% or the two genes are situated on non-homologous chromosomes, the $Aa^tBb$ females produce four kinds of eggs, *AB*, *Ab*, $a^tB$ and $a^tb$. In this case, four types of individuals must be produced from these eggs by haploid or diploid gynogenesis.

TABLE 30
Results of mating experiments and gynogenesis in females heterozygous for albino genes $a^t$ and *b* (1979)

| Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. 25) | | | |
|---|---|---|---|---|---|---|---|
| | | | | Total | Wild | Albino | |
| Female | Male | | | | | Go | TJ |
| (TJ. Alb. ♀ × Go. Alb. ♂) 77, Nos. 1, 2 | TJ. Alb. 77, No. 8 | 341 | 341 (100 %) | 328 (96.2%) | 161 | 0 | 167 |
| | *R. clamitans* | 142 | 133 (93.7%) | 0 | 0 | 0 | 0 |
| | GH | 150 | 107 (71.3%) | 59 (39.3%) | 15 | 10 | 34 |
| | GD | 250 | 26 (10.4%) | 21 ( 8.4%) | 15 | 2 | 4 |

GH, haploid gynogenesis GD, diploid gynogenesis

In 1979, matings were made between two heterozygous females (Nos. 1 and 2) obtained in 1977 from TJ. Alb. 74♀, No. 2 × Go. Alb. 76♂, No. 1 and a male albino (No. 8) obtained in 1977 from TJ. Alb. 74♀, No. 2 × TJ. Het. 75♂, No. 1. The results showed that 161 of 328 feeding tadpoles produced from these matings were of the wild-type and the other 167 were TJ-type ($a^t a^t$) albinos. When 150 eggs of the above two females were pseudofertilized with UV-irradiated sperm of *Rana clamitans*, 107 cleaved normally, 94 hatched and 59 became haploid tadpoles 13 days after hatching, whereas 133 of 142 eggs of the same two females inseminated with non-treated sperm of *Rana clamitans* cleaved normally and ceased development at the late blastula stage. Of the haploid tadpoles, 15 were

of the wild-type, 10 were Go-type albinos and 34 were TJ-type albinos. It is evident that the wild-type tadpoles were *AB*, the Go-type albinos were *Ab* and the TJ-type albinos were a mixture of $a^tB$ and $a^tb$ in genotype. When 250 eggs of the above two females were refrigerated after pseudofertilized with UV-irradiated sperm of *Rana clamitans*, 21 diploid feeding tadpoles (stage 25) were produced. Of these tadpoles, 15 were of the wild-type, two were Go-type albinos and four were TJ-type albinos. The last four probably included $a^ta^tBB$ and $a^ta^tbb$ albinos. The paucity of the albinos as compared with the wild-type tadpoles seems to be attributable to the fact that post-reductional separation of a chromosomal segment bearing the albino gene has occurred owing to crossing-over in some eggs and also to the fact that albinic eggs are weaker in resistibility to the cold-treatment than normal-colored ones. On the other hand, the albinos which are $a^tb$ or $a^ta^tbb$ in genotype are similar to those of $a^t$ or $a^ta^t$ in phenotype, as gene $a^t$ is epistatic as compared with gene *b* (Table 30).

These findings seem to indicate that gene $a^t$ and gene *b* are located on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs between the two loci in a frequency of nearly 50%.

### 3. Difference between albinos of the first and third groups

#### a. Relationship of $a^ea^eCC$ and *AAcc* albinos

The offspring between female albinos of the third group and male albinos of the first group, YM I. Alb. (*AAcc*)♀ × Ex. Alb. ($a^ea^eCC$)♂, are all of the wild-type. They are $Aa^eCc$ in genotype. Mature females of these offspring produce two kinds of gametes, if the two genes are situated on homologous chromosomes and no crossing-over occurs between them. However, if crossing-over occurs in a frequency of nearly 50% or the two genes are located on non-homologous chromosomes, four kinds of gametes, *AC*, *Ac*, $a^eC$ and $a^ec$ are produced in a ratio of 1 : 1 : 1 : 1.

TABLE 31
Results of mating experiments and gynogenesis in females heterozygous for albino genes $a^e$ and *c* (1979)

| Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. 25) | | | |
|---|---|---|---|---|---|---|---|
| Female | Male | | | Total | Wild | Albino | |
| | | | | | | YMI | RA |
| (YMI. Alb. ♀ × Ex. Alb. ♂) 77, Nos. 1, 2 | Ex. Alb. 74, No. 5 | 357 | 357 (100 %) | 309 (86.6%) | 156 | 0 | 153 |
| (YMI. Alb. ♀ × Ex. Alb. ♂) 77, No. 3 | *R. clamitans* | 164 | 104 (63.4%) | 0 | 0 | 0 | 0 |
| | GH | 145 | 103 (71.0%) | 95 (65.5%) | 21 | 25 | 49 |
| | GD | 254 | 78 (30.7%) | 33 (13.0%) | 20 | 4 | 9 |

GH, haploid gynogenesis GD, diploid gynogenesis

In 1979, mating experiments were made between two 2-year-old $Aa^eCc$ females, Nos. 1 and 2 obtained in 1977 from YM I. Alb. 73♀, No. 4 × Ex. Alb.

74♂, No. 2 and a male $a^ea^eCC$ albino, Ex. Alb. 74♂, No. 5, obtained in 1974. It was found that 309 of 357 eggs became feeding tadpoles (stage 25). Of these tadpoles, 156 were of the wild-type, being $Aa^eCC$ and $Aa^eCc$ in genotype, and 153 were albinos, being $a^ea^eCC$ and $a^ea^eCc$ in genotype (Table 31). From 145 eggs of the above two females, 95 haploid tadpoles seven days after hatching were obtained by pseudofertilization with UV-irradiated sperm of *Rana clamitans*. Of these haploid tadpoles, 21 were of the wild-type, 25 were albinos of the YM I-type and 49 were albinos of the RA-type. The RA-type albinos seemed to include $a^eC$ and $a^ec$ in genotype. The $a^ec$ albinos are similar to the $a^eC$ in phenotype, as gene $a^e$ is epistatic. Thus, it is believed that genes $a^e$ and $c$ are located on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%. From 254 eggs of the above two females, 33 diploid feeding tadpoles (stage 25) were obtained by refrigeration after pseudofertilization with UV-irradiated sperm of *Rana clamitans*. Of these tadpoles, 20 were of the wild-type, four were albinos of the YM I-type and nine were albinos of the RA-type. The paucity of the albinos seems to be attributable to the fact that in some eggs, post-reductional separation of a chromosomal segment bearing the albino gene has occurred owing to crossing-over and also to the fact that albinic eggs are weaker in resistibility to the cold-treatment than normal-colored ones (Table 31).

b. Relationship of $a^ta^tCC$ and *AAcc* albinos

i) In 1977, 342 feeding tadpoles (stage I) were produced from matings between four females and one male of the wild-type individuals obtained in 1975 from a mating, YM I. Alb. 73♀, No. 1 × TJ. Alb. 74♂, No. 1. These parents were all $Aa^tCc$ in genotype. Of the offspring, 193 were of the wild-type, including one *AACC*, two *AACc*, two $Aa^tCC$ and four $Aa^tCc$ in kind and ratio of genotypes, 50 were albinos of the YM I-type, including one *AAcc*, and two $Aa^tcc$, and 99 were albinos of the TJ-type, including one $a^ta^tCC$, two $a^ta^tCc$ and one $a^ta^tcc$. These numbers can be expected when genes $a^t$ and $c$ are located on non-homologous chromosomes and gene $a^t$ is dominant over gene $c$. When the four females were mated with a male YM I-type (*AAcc*) albino, YM I. Alb. 75♂, No. 6, 188 feeding tadpoles (stage I) were produced. Of these tadpoles, 88 were of the wild-type, including *AACc* and $Aa^tCc$, and 100 were albinos of the YM I-type, including *AAcc* and $Aa^tcc$ in genotype. When the same four females were mated with a male TJ-type ($a^ta^tCC$) albino, TJ. Alb. 74♂, No. 5, 265 feeding tadpoles (stage I) were produced. Of these tadpoles, 132 were of the wild-type, including $Aa^tCC$ and $Aa^tCc$, and 133 were albinos of the TJ-type, including $a^ta^tCC$ and $a^ta^tCc$ in genotype. When 86 eggs of the above four females were pseudofertilized with UV-irradiated sperm of *Rana clamitans*, five haploid feeding tadpoles (stage I) were produced. Of these tadpoles, one was of the wild-type, three were TJ-type albinos and one was a YM I-type albino. When 1500 eggs of the same four females were refrigerated after inseminated with UV-irradiated sperm of *Rana clamitans* in order to obtain gynogenetic diploids, 214 feeding tadpoles (stage I)

were produced. Of these tadpoles, 155 were of the wild-type, being *AACC* in genotype, 19 were YM I-type (*AAcc*) albinos and 40 were TJ-type albinos, including $a^ta^tCC$ and $a^ta^tcc$ in genotype. The paucity of the albinos as compared with the wild-type tadpoles is probably attributable to the fact that post-reductional separation of a chromosomal segment bearing the albino gene has occurred owing to crossing-over in some eggs and also to the fact that albinic eggs are weaker in resistibility to the cold-treatment than normal-colored ones (Table 32).

TABLE 32

Results of mating experiments and gynogenesis in females homo- or heterozygous for albino genes $a^t$ and $c$

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. I) | | | |
|---|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino | |
| | | | | | | | TJ | YMI |
| 1977 | (YMI. Alb. ♀ × TJ. Alb. ♂) Wild 75, Nos. 1~4 | YMI. Alb. 75, No. 6 | 397 | 276 (69.5%) | 188 (47.4%) | 88 | 0 | 100 |
| | | (YMI. Alb. ♀ × TJ. Alb. ♂) Wild 75, No. 1 | 537 | 515 (95.9%) | 342 (63.7%) | 193 | 99 | 50 |
| | | TJ. Alb. 74, No. 5 | 389 | 384 (98.7%) | 265 (68.1%) | 132 | 133 | 0 |
| | | *R. clamitans* | 433 | 413 (95.4%) | 0 | 0 | 0 | 0 |
| | | GH | 86 | 85 (98.8%) | 5 ( 5.8%) | 1 | 3 | 1 |
| | | GD | 1500 | 668 (44.5%) | 214 (14.3%) | 155 | 40 | 19 |
| 1979 | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Wild 77, No. 1 | YMI. Alb. 75, No. 8 | 180 | 180 (100 %) | 162 (90.0%) | 162 | 0 | 0 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Wild 77, No. 2 | YMI. Alb. 75, No. 8 | 245 | 244 (99.6%) | 231 (94.3%) | 231 | 0 | 0 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Wild 77, No. 3 | YMI. Alb. 75, No. 8 | 199 | 181 (91.0%) | 173 (86.9%) | 82 | 0 | 91 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Wild 77, No. 4 | YMI. Alb. 75, No. 8 | 184 | 183 (99.5%) | 172 (93.5%) | 172 | 0 | 0 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Wild 77, No. 5 | YMI. Alb. 75, No. 8 | 168 | 166 (98.8%) | 162 (96.4%) | 78 | 0 | 84 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Wild 77, No. 6 | YMI. Alb. 75, No. 8 | 180 | 180 (100 %) | 157 (87.2%) | 157 | 0 | 0 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Wild 77, No. 7 | YMI. Alb. 75, No. 8 | 183 | 181 (98.9%) | 119 (65.0%) | 119 | 0 | 0 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Wild 77, No. 8 | YMI. Alb. 75, No. 8 | 163 | 162 (99.4%) | 131 (80.4%) | 65 | 0 | 66 |
| 1979 | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Alb. 77, No. 9 | YMI. Alb. 75, No. 8 | 216 | 203 (94.0%) | 196 (90.7%) | 99 | 0 | 97 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Alb. 77, No. 10 | YMI. Alb. 75, No. 8 | 225 | 214 (95.1%) | 202 (89.8%) | 202 | 0 | 0 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Alb. 77, No. 11 | YMI. Alb. 75, No. 8 | 197 | 176 (89.3%) | 163 (82.7%) | 81 | 0 | 82 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Alb. 77, No. 12 | YMI. Alb. 75, No. 8 | 231 | 217 (93.9%) | 215 (93.1%) | 105 | 0 | 110 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Alb. 77, No. 13 | YMI. Alb. 75, No. 8 | 220 | 208 (94.5%) | 197 (89.5%) | 197 | 0 | 0 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Alb. 77, No. 14 | YMI. Alb. 75, No. 8 | 254 | 230 (90.6%) | 214 (84.3%) | 108 | 0 | 106 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Alb. 77, No. 15 | YMI. Alb. 75, No. 8 | 199 | 191 (96.0%) | 170 (85.4%) | 170 | 0 | 0 |
| | (YMI. Alb. ♀ × TJ. Alb. ♂) ♀ × TJ. Alb. ♂, Alb. 77, No. 16 | YMI. Alb. 75, No. 8 | 183 | 180 (98.4%) | 176 (96.2%) | 176 | 0 | 0 |

GH, haploid gynogenesis GD, diploid gynogenesis

ii) In 1979, mating experiments were made between eight females (Nos. 1～8) of the wild-type and eight female albinos (Nos. 9～16) of the TJ-type obtained in 1977 from (YM I. Alb. ♀ × TJ. Alb. ♂)♀ × TJ. Alb. ♂ and a male YM I-type (*AAcc*) albino, YM I. Alb. 75♂, No. 8. The results showed that 162, 231, 172, 157 and 119 feeding tadpoles (stage I) produced from five (Nos. 1, 2, 4, 6 and 7) of the eight wild-type females were all of the wild-type. On the other hand, 91 of 173, 84 of 162 and 66 of 131 feeding tadpoles (stage I) produced from the other three females (Nos. 3, 5 and 8) were albinos of the YM I-type. Thus, it is considered that the five wild-type females, Nos. 1, 2, 4, 6 and 7, were *Aa$^t$CC* in genotype and the other three females, Nos. 3, 5 and 8, were *Aa$^t$Cc*. While 202, 197, 170 and 176 feeding tadpoles (stage I) produced from four (Nos. 10, 13, 15 and 16) of the eight female YM I-type albinos, were all of the wild-type, 97 of 196, 82 of 163, 110 of 215 and 106 of 214 feeding tadpoles (stage I) produced from the remaining four female albinos (Nos. 9, 11, 12 and 14) were albinos of the YM I-type. Thus, it seems evident that the four female albinos, Nos. 10, 13, 15 and 16, were *a$^t$a$^t$CC* in genotype and that the other four female albinos, Nos. 9, 11, 12 and 14, were *a$^t$a$^t$Cc* in genotype (Table 32).

These findings seem to indicate that genes *a$^t$* and *c* are located on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%.

### 4. Difference between albinos of the first and fourth groups

The hybrids between *a$^e$a$^e$DD* albinos of the first group and *AAdd* albinos of the fourth group are all of the wild-type. They are *Aa$^e$Dd* in genotype. If genes *a$^e$* and *d* are situated on homologous chromosomes and no crossing-over occurs between the two loci, these hybrids produce two kinds of gametes which are *Ad* and *a$^e$D* in genotype. When brother and sister matings are made among the hybrids, KM-type (*AAdd*) albinos, wild-type (*Aa$^e$Dd*) individuals and RA-type (*a$^e$a$^e$DD*) albinos should be produced in a ratio of 1: 2: 1. If these two albino genes are situated on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%, or they are situated on non-homologous chromosomes, the *Aa$^e$Dd* hybrids produce four kinds of gametes, *AD*, *a$^e$D*, *Ad* and *a$^e$d*. By brother and sister matings, four types of individuals, (1) wild-type individuals, including one *AADD*, two *AADd*, two *Aa$^e$DD*, and four *Aa$^e$Dd* in kind and ratio of genotypes, (2) KM-type albinos, including one *AAdd* and two *Aa$^e$dd*, (3) RA-type albinos, including one *a$^e$a$^e$DD* and two *a$^e$a$^e$Dd* and (4) RA·KM-type albinos, being *a$^e$a$^e$dd*, should be produced in a ratio of 9: 3: 3: 1.

In 1977, mating experiments were made between three females (Nos. 1～3) and one male (No. 1) obtained in 1975 from a mating, KM. Alb. 74♀, No. 1 × SN I. Alb. 72♂, No. 1. The results showed that 722 feeding tadpoles (stage III) were obtained from 960 eggs. Of these tadpoles, 406 were of the wild-type, 131 were KM-type albinos, 143 were RA-type albinos and 42 were RA·KM-type albinos. The last type was of the RA-type in coloration, that is, lighter than the KM-type, while this was semitransparent like the KM-type. These numbers of the four

kinds of phenotypic tadpoles revealed a ratio of approximately 9: 3: 3: 1 (Table 33). When the above three females (Nos. 1～3) were mated with a male RA-type albino, SN I. Alb. 75♂, No. 5, 861 feeding tadpoles (stage III) were produced from 1050 eggs. Of these tadpoles, 433 were of the wild-type, including *Aa$^e$Dd* and *Aa$^e$DD* in genotype, and 428 were of the RA-type, including *a$^e$a$^e$DD* and *a$^e$a$^e$Dd*. The ratio of these numbers approximately corresponded to the theoretical ratio of 1: 1 (Table 33).

TABLE 33

Results of mating experiments between females heterozygous for albino genes *a$^e$* and *d* and males homozygous for gene *a$^e$* or heterozygous for *a$^e$* and *d* (1977)

| Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | | | |
|---|---|---|---|---|---|---|---|---|
| Female | Male | | | Total | Wild | Albino | | |
| | | | | | | KM | RA | RA·KM |
| (KM. Alb. ♀ × SNI. Alb. ♂) 75, No. 1 | SNI. Alb. 75, No. 5 | 380 | 372 (97.9%) | 370 (97.4%) | 187 | 0 | 183 | 0 |
| | (KM. Alb. ♀ × SNI. Alb. ♂) 75, No. 1 | 358 | 336 (93.9%) | 331 (92.5%) | 190 | 63 | 57 | 21 |
| (KM. Alb. ♀ × SNI. Alb. ♂) 75, No. 2 | SNI. Alb. 75, No. 5 | 244 | 227 (93.0%) | 183 (75.0%) | 93 | 0 | 90 | 0 |
| | (KM. Alb. ♀ × SNI. Alb. ♂) 75, No. 1 | 207 | 197 (95.2%) | 180 (87.0%) | 105 | 29 | 39 | 7 |
| (KM. Alb. ♀ × SNI. Alb. ♂) 75, No. 3 | SNI. Alb. 75, No. 5 | 426 | 424 (99.5%) | 308 (72.3%) | 153 | 0 | 155 | 0 |
| | (KM. Alb. ♀ × SNI. Alb. ♂) 75, No. 1 | 395 | 374 (94.7%) | 211 (53.4%) | 111 | 39 | 47 | 14 |
| (KM. Alb. ♀ × SNI. Alb. ♂) 75, Nos. 1～3 | SNI. Alb. 75, No. 5 | 1050 | 1023 (97.4%) | 861 (82.0%) | 433 | 0 | 428 | 0 |
| | (KM. Alb. ♀ × SNI. Alb. ♂) 75, No. 1 | 960 | 807 (84.1%) | 722 (75.2%) | 406 | 131 | 143 | 42 |

These findings seem to indicate that genes *a* and *d* are located on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%.

## 5. Difference between albinos of the first and fifth groups

The offspring obtained in 1978 from a mating between a heterozygous *Aa$^f$EE* female and a male *AAee* albino, Fc. Het. 76♀, No. 1 × TY. Alb. 77♂, No. 4, were all of the wild-type. These offspring were *AAEe* and *Aa$^f$Ee* in genotype. In 1979, ovulation occurred in four females (Nos. 1～4) of them, and 239, 303, 169 and 155 eggs, 866 in total, obtained from these females were inseminated with UV-irradiated sperm of *Rana clamitans*. It was found that 115, 153, 49 and 20, 337 in total, of these eggs, respectively, hatched normally and became haploid tadpoles. Of these tadpoles, 53 of 115 produced from female No. 1, 90 of 153 from female No. 2 and 10 of 20 from female No. 4, 153 of 288 in total, were albinos of the FC-type. The other 135 were of the wild-type. On the other hand, 49 haploid tadpoles raised from eggs of female No. 3 were all of the wild-type. As TY-type albinos are indistinguishable from wild-type tadpoles until 14 days after beginning to eat, the haploid tadpoles were reared as long as possible. However, most of the haploid tadpoles died before exhibiting the characters of

Ty-type albinos. Only five of 63 wild-type tadpoles and two of 90 Fc-type albinos obtained from female No. 2 were living 20 days after beginning to eat. While four of the five wild-type tadpoles were still of the wild-type, the remainder became a Ty-type albino. All the haploid tadpoles produced from females Nos. 1, 3 and 4 died at the early feeding stage.

When 164, 363, 213 and 162 eggs, 902 in total, obtained from the above four females, Nos. 1～4, were refrigerated after insemination with UV-irradiated sperm of *Rana clamitans* in order to produce gynogenetic diploids, 56, 204, 43 and 7, 310 in total, respectively, hatched normally and became diploid tadpoles. Of these tadpoles, 21 of 56 produced from female No. 1, 92 of 204 from female No. 2 and two of seven from female No. 4, 115 of 267 in total, were Fc-type albinos and the remaining 152 were of the wild-type. All 43 tadpoles produced from female No. 3 were of the wild-type. When these tadpoles were continuously reared, 82 of the 152 wild-type tadpoles were living after 20 days. Of these tadpoles, 59 were of the wild-type and 23 became Ty-type albinos. Of the 115 Fc-type albinos in total, 59 were living after 20 days (stage V) and still of the Fc-type. Of the 43 wild-type tadpoles produced from female No. 3, 39 were living after 20 days (stage V). Of these tadpoles, 31 were of the wild-type and eight became Ty-type albinos (Table 34). These findings seem to indicate that females Nos. 1, 2 and 4 were $Aa^fEe$ and female No. 3 was *AAEe* in genotype.

Mating experiments were made between the above three $Aa^fEe$ females, Nos. 1, 2 and 4 and a heterozygous *AAEe* male, Ty. Het. 78♂, No. 2. The results showed that 491 of 536 eggs hatched normally and all became wild-type tadpoles. When these tadpoles were continuously reared, 458 were living after 20 days (stage V). It was found that 351 of them were of the wild-type, including one *AAEE*, two

TABLE 34

Results of mating experiments and gynogenesis in females heterozygous for albino genes $a^f$ and *e* (1979)

| Parents | | No. of eggs | No. of normal cleavages | No. of hatched tadpoles | | | No. of feeding tadpoles (St. V) | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Total | Wild | Albino | Total | Wild | Albino | |
| Female | Male | | | | | | | | Ty | Fc |
| (Fc. Het. ♀ × Ty. Alb. ♂) 78, Nos. 1, 2, 4 | Ty. Het. 78, No. 2 | 536 | 521 (97.2%) | 491 (91.6%) | 491 | 0 | 458 (85.4%) | 351 | 107 | 0 |
| | *R. clamitans* | 293 | 277 (94.5%) | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | GH | 697 | 525 (75.3%) | 288 (41.3%) | 135 | 153 | 7 ( 1.0%) | 4 | 1 | 2 |
| | GD | 689 | 367 (53.3%) | 267 (38.8%) | 152 | 115 | 141 (20.5%) | 59 | 23 | 59 |
| (Fc. Het. ♀ × Ty. Alb. ♂) 78, No. 3 | Ty. Het. 78, No. 2 | 179 | 168 (93.9%) | 152 (84.9%) | 152 | 0 | 150 (83.8%) | 113 | 37 | 0 |
| | *R. clamitans* | 114 | 101 (88.6%) | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| | GH | 169 | 137 (81.1%) | 49 (29.0%) | 49 | 0 | 0 | 0 | 0 | 0 |
| | GD | 213 | 80 (37.6%) | 43 (20.2%) | 43 | 0 | 39 (18.3%) | 31 | 8 | 0 |

GH, haploid gynogenesis GD, diploid gynogenesis

*AAEe*, two *Aa$^f$Ee* and one *Aa$^f$EE* in kind and ratio of genotypes, and 107 were Ty-type albinos, including *AAee* and *Aa$^f$ee*. Of 179 eggs of female No. 3 (*AAEe*) inseminated with sperm of the above *AAEe* male, 152 hatched normally and became tadpoles which were all of the wild-type. After 20 days (stage V), 150 of them were living and it was found that 113 were still of the wild-type, including one *AAEE* and two *AAEe* in kind and ratio of genotypes, and 37 were Ty-type (*AAee*) albinos (Table 34).

These findings seem to indicate that genes *a$^f$* and *e* are situated on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%.

### 6. Difference between albinos of the second and third groups

The offspring between a female *BBcc* albino, YmI. Alb. 77♀, No. 7, and a male *bbCC* albino, Go. Alb. 76♂, No. 5, obtained in 1979 were all of the wild-type and *BbCc* in genotype. In 1981, brother and sister matings were made between five females (Nos. 1～5) and one male (No. 1) of these offspring. As presented in Table 35, 916 of 1078 eggs obtained from these females hatched normally and became tadpoles (stage 21). Of these tadpoles, 522 were of the wild-type and 394 were albinos. If genes *b* and *c* are situated on homologous chromosomes and no crossing-over occurs between the two loci, wild-type individuals and albinos will be produced in a ratio of 1: 1. If both genes are situated on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%, wild-type and albinic tadpoles should be produced in a ratio of 9: 7. The results of the above experiments actually showed that the numbers of wild-type and albinic tadpoles were 522 and 394, respectively. These numbers are very similar to the theoretical ratio of 9: 7. When 1607 eggs of the above five females were pseudofertilized with UV-irradiated sperm of *Rana clamitans*, 719 of them hatched normally and became haploid tadpoles. Of these tadpoles, 188 were of the wild-type and 531 were albinos. These numbers of wild-type and albinic tadpoles reveal a ratio of approximately 1: 3. When 1214 eggs of the above five females were refrigerated after pseudofertilized with UV-irradiated sperm of *Rana clamitans* in order to obtain gynogenetic diploids,

TABLE 35

Results of mating experiments and gynogenesis in females heterozygous for albino genes *b* and *c* (1981)

| Parents | | No. of eggs | No. of normal cleavages | No. of hatched tadpoles (St. 21) | | |
|---|---|---|---|---|---|---|
| Female | Male | | | Total | Wild | Albino |
| (YmI. Alb. ♀ × Go. Alb. ♂) 79, Nos. 1～5 | (YmI. Alb. ♀ × Go. Alb. ♂) 79, No. 1 | 1078 | 1013 (94.0%) | 916 (85.0%) | 522 | 394 |
| | GH | 1607 | 1493 (92.9%) | 719 (44.7%) | 188 | 531 |
| | GD | 1214 | 622 (51.2%) | 174 (14.3%) | 120 | 54 |

GH, haploid gynogenesis GD, diploid gynogenesis

174 hatched normally and became diploid tadpoles. Of these tadpoles, 120 were of the wild-type and 54 were albinos. The paucity of albinos as compared with the wild-type tadpoles seems to be attributable to the fact that post-reductional separation of a chromosomal segment bearing the albino gene has occurred owing to crossing-over in some eggs, and also to the fact that albinic eggs are weaker than normal-colored ones in resistibility to refrigeration (Table 35).

These findings seem to indicate that genes *b* and *c* are located on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%.

### 7. Difference between albinos of the second and fourth groups

a. Mating experiments performed in 1979

The offspring obtained in 1977 from a mating between a heterozygous *BBDd* female, Km. Het. 75♀, No. 1, and a male *bbDD* albino, Go. Alb. 76♂, No. 1, were all of the wild-type, being *BbDd* or *BbDD* in genotype. In 1979, brother and sister matings were made between four females (Nos. 1～4) and four males (Nos. 1～4) of these offspring. As presented in Table 36, Km-type (*dd*) albinos were produced from the four females by mating with three (Nos. 2～4) of the four males, while no such albinos were obtained from these females by mating with male No. 1. Thus, it is believed that females Nos. 1～4 and males Nos. 2～4 were *BbDd* and male No. 1 was *BbDD* in genotype.

i) In 1979, brother and sister matings between four females (Nos. 1～4) and three males (Nos. 2～4) of the offspring obtained from a mating, Km. Het. ♀ × Go. Alb. ♂, produced 1355 feeding tadpoles 14 days after beginning to eat (stage III) from among 1980 eggs. Of these tadpoles, 766 were of the wild-type, 236 were Km-type albinos, 265 were Go-type albinos and the remaining 88 were Go·Km-type albinos which were of the Go-type in appearance, being lighter than the Km-type albinos and were semitransparent like the latter. If genes *b* and *d* are situated on homologous chromosomes and there is no crossing-over between both loci, wild-type (*BbDd*) tadpoles, Km-type (*BBdd*) albinos and Go-type (*bbDD*) albinos should be produced in a ratio of 2: 1: 1, and no Go·Km-type (*bbdd*) albinos will be obtained. If crossing-over occurs in a frequency of nearly 50% or the two genes are situated on non-homologous chromosomes, wild-type tadpoles, including one *BBDD*, two *BbDD*, two *BBDd* and four *BbDd* in kind and ratio of genotypes, Km-type (*dd*) albinos, including one *BBdd* and two *Bbdd*, Go-type (*bb*) albinos, including one *bbDD* and two *bbDd*, and one Go·Km-type (*bbdd*) albinos will be produced in a ratio of 9: 3: 3: 1. The results of mating experiments actually showed that the numbers of wild-type tadpoles and Km-type, Go-type and Go·Km-type albinos were 766, 236, 265 and 80, respectively. These numbers revealed a ratio of approximately 9: 3: 3: 1 (Table 36).

ii) In 1979, when the above four *BbDd* females, (Km, Het♀ × Go. Alb♂) 77♀, Nos. 1～4, were mated with a *BbDD* male, (Km. Het♀ × Go. Alb♂) 77♀,

TABLE 36

Results of brother and sister matings and gynogenesis in females heterozygous for albino genes *b* and *d* (1979)

| Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | | | |
|---|---|---|---|---|---|---|---|---|
| Female | Male | | | Total | Wild | Albino | | |
| | | | | | | Km | Go | Go·Km |
| (Km. Het. ♀ × Go. Alb. ♂)77, No. 1 | (Km. Het. ♀ × Go. Alb. ♂)77, No. 1 | 165 | 165 (100 %) | 114 (69.1%) | 91 | 0 | 23 | 0 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 2 | 160 | 160 (100 %) | 101 (63.1%) | 55 | 19 | 19 | 8 |
| | (Km. Het. ♀ × Go.Alb. ♂)77, No. 3 | 164 | 163 (99.4%) | 135 (82.3%) | 74 | 29 | 24 | 8 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 4 | 162 | 162 (100 %) | 90 (55.6%) | 51 | 15 | 18 | 6 |
| (Km. Het. ♀ × Go. Alb. ♂)77, No. 2 | (Km. Het. ♀ × Go. Alb. ♂)77, No. 1 | 166 | 165 (99.4%) | 148 (89.2%) | 113 | 0 | 35 | 0 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 2 | 167 | 167 (100 %) | 161 (96.4%) | 92 | 30 | 30 | 9 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 3 | 169 | 167 (98.8%) | 35 (20.7%) | 17 | 4 | 10 | 4 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 4 | 166 | 165 (99.4%) | 152 (91.6%) | 77 | 31 | 34 | 10 |
| (Km. Het. ♀ × Go. Alb. ♂)77, No. 3 | (Km. Het. ♀ × Go. Alb. ♂)77, No. 1 | 193 | 193 (100 %) | 184 (95.3%) | 129 | 0 | 55 | 0 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 2 | 172 | 171 (99.4%) | 164 (95.3%) | 97 | 29 | 29 | 9 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 3 | 171 | 171 (100 %) | 148 (86.5%) | 90 | 26 | 25 | 7 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 4 | 167 | 166 (99.4%) | 114 (68.3%) | 72 | 9 | 24 | 9 |
| (Km. Het. ♀ × Go. Alb. ♂)77, No. 4 | (Km. Het. ♀ × Go. Alb. ♂)77, No. 1 | 169 | 168 (99.4%) | 159 (94.1%) | 116 | 0 | 43 | 0 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 2 | 167 | 165 (89.8%) | 69 (41.3%) | 38 | 13 | 14 | 4 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 3 | 150 | 150 (100 %) | 93 (62.0%) | 53 | 16 | 17 | 7 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, No. 4 | 165 | 165 (100 %) | 93 (56.4%) | 50 | 15 | 21 | 7 |
| (Km. Het. ♀ × Go. Alb. ♂)77, Nos. 1～4 | (Km. Het. ♀ × Go. Alb. ♂)77, No. 1 | 693 | 691 (99.7%) | 605 (87.3%) | 449 | 0 | 156 | 0 |
| | (Km. Het. ♀ × Go. Alb. ♂)77, Nos. 2～4 | 1980 | 1972 (99.6%) | 1355 (68.4%) | 766 | 236 | 265 | 88 |
| (Km. Alb. ♀ × Go. Alb. ♂)78, Nos. 1～4 | *R. clamitans* | 470 | 421 (89.6%) | 0 | 0 | 0 | 0 | 0 |
| | GH | 709 | 640 (90.3%) | 368* (51.9%) | 95* | 84* | 189* | 0 |
| | GD | 1031 | 628 (60.9%) | 245 (23.8%) | 129 | 39 | 43 | 34 |

* haploid hatched tadpoles GH, haploid gynogenesis GD, diploid gynogenesis

No. 1, 605 feeding tadpoles 14 days after beginning to eat were produced from among 693 eggs by the brother and sister matings. Of these tadpoles, 449 were of the wild-type and 156 were Go-type (*bb*) albinos. From these matings, wild-type tadpoles which theoretically consist of one *BBDD*, two *BbDD*, two *BbDd* and one *BBDd* in kind and ratio of genotypes and Go-type (*bb*) albinos which consist of one *bbDD* and one *bbDd* should be produced in a ratio of 3: 1. The results of the above mating experiments actually showed that 449 wild-type tadpoles and 156 Go-type (*bb*) albinos were produced. These numbers approximately agree with the expected ratio of 3: 1.

b. Gynogenesis performed in 1979

The offspring obtained in 1978 from a mating between a female *dd* albino and a male *bb* albino, Km. Alb. 77♀, No. 2×Go. Alb. 76♂, No. 3, were all of the wild-type, being *BbDd* in genotype. In 1979, 709 eggs of four females (Nos. 1～4) of the offspring were pseudofertilized with UV-irradiated sperm of *Rana clamitans* and 368 haploid tadpoles three days (stage I) after hatching were produced. Of these tadpoles, 95 were of the wild-type (*BD*), 84 were Km-type (*Bd*) albinos and 189 were Go-type albinos, being *bD* and *bd* in genotype. As the haploid tadpoles were retarded in growth owing to their poor appetite, Go-type (*bD*) albinos were indistinguishable from Go·Km-type (*bd*) albinos. Thus, the ratio of these 95 *BD*, 84 *Bd* and, 189 *bD* and *bd* tadpoles approximately agrees with the expected ratio of 1: 1: 2, as the *BD*, *Bd*, *bD* and *bd* tadpoles should be produced in a ratio of 1: 1: 1: 1 in the case where *bD* and *bd* are distinguishable (Table 36). When 1031 eggs of the above four females were refrigerated after insemination with UV-irradiated sperm of *Rana clamitans* in order to obtain gynogenetic diploids, 245 diploid feeding tadpoles (stage III) 14 days after beginning to eat were produced. Of these tadpoles, 129 were of the wild-type, 39 were Km-type (*dd*) albinos, 43 were Go-type (*bb*) albinos and 34 were Go·Km-type (*bbdd*) albinos. The paucity of albinos in gynogenetic diploids seems to be attributable to the fact that post-reductional separation of a chromosomal segment bearing the albino gene has occurred owing to crossing-over in some eggs, and also to the fact that albinic eggs are weaker than normal-colored ones in resistibility to refrigeration (Table 36).

These findings seem to indicate that genes *b* and *d* are situated on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%.

8. Difference between albinos of the third and fourth groups

a. Mating experiments performed in 1977

The offspring obtained in 1975 from a mating between a female *CCdd* albino and a male *ccDD* albino, Km. Alb. 74♀, No. 1×Ym I. Alb. 73♂, No. 4, were all of the wild-type, being *CcDd* in genotype. If genes *c* and *d* are situated on homologous chromosomes and no crossing-over occurs between them, these offspring produce two kinds of gametes, *Cd* and *cD*, in an equal number. By brother and sister matings of the *CcDd* offspring, wild-type (*CcDd*) individuals, Km-type (*CCdd*) albinos and Ym I-type (*ccDD*) albinos should be produced in a ratio of 2: 1: 1. If genes *c* and *d* are located on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%, they produce four kinds of gametes, *CD*, *Cd*, *cD* and *cd*, in a ratio of 1: 1: 1: 1. By brother and sister matings of *CcDd* offspring, wild-type individuals, including one *CCDD*, two *CCDd*, two *CcDD* and four *CcDd* in kind and ratio of genotypes, Km-type albinos, including one *CCdd* and two *Ccdd*, Ym I-type albinos, including one *ccDD* and two *ccDd* and Ym I·Km-type (*ccdd*) albinos should be

produced in a ratio of 9: 3: 3: 1. While the Ym I·Km-type (*ccdd*) albinos are indistinguishable from the Ym I-type (*cc*) albinos four days after beginning to eat (stage I), both kinds of albinos become distinguishable from each other seven days after beginning to eat (stage II). The difference between them is completely evident 14 days after beginning to eat (stage III).

i) In 1977, brother and sister matings were made between three *CcDd* females (Nos. 1～3) and one *CcDd* male (No. 1) obtained in 1975 from the above mating, Km. Alb. 74♀, No. 1×Ym I. Alb. 73♂, No. 4. As presented in Table 37, 669 feeding tadpoles 14 days after beginning to eat (stage III) were produced from among 996 eggs. Of these tadpoles, 390 were of the wild-type, 113 were Km-type (*dd*) albinos, 125 were Ym I-type (*cc*) albinos, and 41 were Ym I·Km-type (*ccdd*) albinos. These numbers of the four kinds of tadpoles, reveal a close approximation to the ratio of 9: 3: 3: 1.

TABLE 37

Results of mating experiments and gynogenesis in females heterozygous for albino genes *c* and *d*

| Year | Parents | | No. of eggs | No. of normal cleavages | No. of feeding tadpoles (St. III) | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Female | Male | | | Total | Wild | Albino | | |
| | | | | | | | Km | YmI | YmI·Km |
| 1977 | (Km. Alb. ♀ × YmI. Alb. ♂)75, Nos. 1～3 | YmI. Alb. 75, No. 6 | 800 | 702 (87.8%) | 561 (70.1%) | 295 | 0 | 266 | 0 |
| | | (Km. Alb. ♀ × YmI. Alb. ♂)75, No. 1 | 996 | 979 (98.3%) | 669 (67.2%) | 390 | 113 | 125 | 41 |
| 1979 | (Km. Alb. ♀ × YmI. Alb. ♂)² Wild 77, No. 1 | (Km. Alb. ♀ × YmI. Alb. ♂)² Wild 77, No. 1 | 293 | 251 (85.7%) | 223 (76.1%) | 168 | 55 | 0 | 0 |
| | | GH | 282 | 239 (84.8%) | 63 (22.3%) | 35 | 28 | 0 | 0 |
| | | GD | 256 | 123 (48.0%) | 58 (22.7%) | 43 | 15 | 0 | 0 |
| | (Km. Alb. ♀ × YmI. Alb. ♂)² Wild 77, No. 2 | (Km. Alb. ♀ × YmI. Alb. ♂)² Wild 77, No. 1 | 332 | 310 (93.4%) | 252 (75.9%) | 149 | 46 | 42 | 15 |
| | | GH | 224 | 161 (71.9%) | 122 (54.5%) | 31 | 27 | 34 | 30 |
| | | GD | 529 | 466 (88.1%) | 130 (24.6%) | 83 | 12 | 15 | 20 |
| | (Km. Alb. ♀ × YmI. Alb. ♂)² Wild 77, No. 3 | (Km. Alb. ♀ × YmI. Alb. ♂)² Wild 77, No. 1 | 285 | 261 (91.6%) | 172 (60.4%) | 103 | 28 | 32 | 9 |
| | | GH | 250 | 243 (97.2%) | 97 (38.8%) | 25 | 25 | 24 | 23 |
| | | GD | 228 | 90 (39.5%) | 36 (15.8%) | 29 | 3 | 3 | 1 |
| | (Km. Alb. ♀ × YmI. Alb. ♂)² Wild 77, No. 4 | (Km. Alb. ♀ × YmI. Alb. ♂)² Wild 77, No. 1 | 192 | 29 (15.1%) | 21 (10.9%) | 16 | 5 | 0 | 0 |

(Km. Alb. ♀ × YmI. Alb. ♂)² Wild 77, No. 1 = {(Km. Alb. 74♀, No. 1 × YmI. Alb. 73♂, No. 4) 75♀, No. 1 × (Km. Alb. 74♀, No. 1 × YmI. Alb. 73♂, No. 4) 75♂, No. 1} Wild 77♂, No. 1

GH, haploid gynogenesis GD, diploid gynogenesis

ii) By matings between the above three females, (KM. Alb. ♀ × YM I. Alb♂) 75♀, Nos. 1~3, and a male *ccDD* albino, YM I. Alb. 75♂, No. 6, 561 feeding tadpoles 14 days after beginning to eat (stage III) were obtained from among 800 eggs. Of these tadpoles, 295 were of the wild-type and 266 were YM I-type (*cc*) albinos. These numbers reveal a ratio of approximately 1: 1.

b. Mating experiments and gynogenesis performed in 1979

Brother and sister matings were made in 1979 between four females and one male of the above 390 wild-type individuals obtained in 1977 from brother and sister matings, (KM. Alb♀ × YM I. Alb♂) 75♀, Nos. 1~3 × (KM. Alb♀ × YM I. Alb♂)75♂, No. 1. Besides, gynogenetic haploids and diploids were produced from eggs of three of the four females (Table 37).

Females Nos. 1 and 4 were assumed to be *CCDd* in genotype, as no YM I-type albinos were produced from these females. In contrast, females Nos. 2 and 3 were considered to be *CcDd* in genotype, as KM-type, YM I-type and YM I·KM-type albinos were produced from them.

i) From 485 eggs of the above two wild-type females (Nos. 1 and 4) which were mated in 1979 with a wild-type male (No. 1), 244 feeding tadpoles 14 days after beginning to eat (stage III) were produced. Of these tadpoles, 184 were of the wild-type, including one *CCDD*, two *CCDd*, two *CcDd* and one *CcDD* in kind and ratio of genotypes, and 60 were KM-type albinos, including *CCdd* and *Ccdd*. These numbers of the wild-type and albinic tadpoles reveal a ratio of approximately 3: 1.

ii) When 282 eggs of female No. 1 (*CCDd*) were pseudofertilized with UV-irradiated sperm of *Rana clamitans*, 63 haploid tadpoles 14 days after beginning to eat were produced. Although these tadpoles were somewhat retarded in growth, it was found that 35 of them were of the wild-type (*CD*) and 28 were KM-type (*Cd*) albinos. When 256 eggs of the same female were refrigerated after pseudofertilized with UV-irradiated sperm of *Rana clamitans* in order to obtain gynogenetic diploids, 58 diploid feeding tadpoles 14 days after beginning to eat (stage III) were produced. Of these tadpoles, 43 were of the wild-type and 15 were KM-type (*CCdd*) albinos. The paucity of albinos in gynogenetic diploids seems to be attributable to the fact that post-reductional separation of a chromosomal segment bearing the albino gene has occurred owing to crossing-over and also to the fact that albinic eggs are weaker than normal-colored ones in resistibility to the cold-treatment.

iii) Of 617 eggs of the above two wild-type females (Nos. 2 and 3) which were mated in 1979 with a wild-type male (No. 1), 424 became feeding tadpoles 14 days after beginning to eat (stage III). Of these tadpoles, 252 were of the wild-type, 74 were KM-type albinos, 74 were YM I-type albinos, and 24 were YM I·KM-type albinos. These numbers of the four kinds of phenotypes reveal a ratio

of approximately 9: 3: 3: 1.

iv) When 474 eggs of the above two wild-type females (Nos. 2 and 3) were pseudofertilized with UV-irradiated sperm of *Rana clamitans*, 219 haploid tadpoles of 14 days after beginning to eat (stage III) were produced. Of these tadpoles, 56 were of the wild-type (*CD*), 52 were Km-type (*Cd*) albinos, 58 were Ym I-type (*cD*) albinos and 53 were Ym I·Km-type (*cd*) albinos. When 757 eggs of the same two females were refrigerated after pseudofertilization with UV-irradiated sperm of *Rana clamitans* in order to obtain gynogenetic diploids, 166 diploid feeding tadpoles 14 days after beginning to eat were produced. Of these tadpoles, 112 were of the wild-type, 15 were Km-type albinos, 18 were Ym I-type albinos and 21 were Ym I·Km-type albinos. The paucity of the albino tadpoles in gynogenetic diploids seems to be attributable to the fact that post-reductional separation of a chromosomal segment bearing the albino gene has occurred owing to crossing-over and also to the fact that albinic eggs are weaker than normal-colored ones in resistibility to refrigeration.

These findings seem to indicate that genes *c* and *d* are situated on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%.

## 9. Difference between albinos of the fourth and fifth groups

### a. Mating experiments performed in 1978 and 1979

The offspring obtained in 1978 from a mating between a female *dd* albino and a male *ee* albino, Km. Alb. 77♀, No. 3 × Ty. Alb. 77♂, No. 3, were all of the wild-type, being *DdEe* in genotype. Brother and sister matings were made in 1979 between eight females (Nos. 1～8) and one male of these offspring. It was found that 808 feeding tadpoles 20 days after beginning to eat (stage V) were obtained from among 1307 eggs. Of these tadpoles, 449 were of the wild-type, 146 were Km-type albinos, 161 were Ty-type albinos and 52 were Km·Ty-type albinos. These numbers of the four kinds of individuals reveal a ratio of approximately 9: 3: 3: 1 ratio (Table 38).

TABLE 38

Results of brother and sister matings and gynogenesis in females heterozygous for albino genes *d* and *e* (1979)

| Parents | | No. of eggs | No. of normal cleavages | No. of normal hatched tadpoles | No. of feeding tadpoles (St. V) | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Female | Male | | | | Total | Wild | Albino | | |
| | | | | | | | Km | Ty | Km·Ty |
| (Km. Alb. ♀ × Ty. Alb. ♂) 78, Nos. 1～8 | (Km. Alb. ♀ × Ty. Alb. ♂)78, No. 1 | 1307 | 1259 (96.3%) | 1086 (83.1%) | 808 (61.8%) | 449 | 146 | 161 | 52 |
| | *R. clamitans* | 400 | 385 (96.3%) | 0 | 0 | 0 | 0 | 0 | 0 |
| | GH | 1290 | 1235 (95.7%) | 977 (75.7%) | 5 ( 0.4%) | 5 | 0 | 0 | 0 |
| | GD | 1663 | 1072 (64.5%) | 552 (33.2%) | 227 (13.7%) | 146 | 31 | 35 | 15 |

GH, haploid gynogenesis GD, diploid gynogenesis

b. Gynogenesis performed in 1979

When 1290 eggs obtained from the above eight females (Nos. 1～8) were pseudo-fertilized with UV-irradiated sperm of *Rana clamitans*, 977 of them hatched normally and became haploid tadpoles. These tadpoles were all of the wild-type. Most of them died of edema or some other abnormalities. Only five tadpoles were living 20 days after beginning to eat. Those were still of the wild-type. When 1663 eggs of the same eight females were refrigerated after pseudo-fertilized with UV-irradiated sperm of *Rana clamitans* in order to obtain gynogenetic diploids, 552 of them hatched normally. All of these tadpoles were of the wild-type. Twenty days after beginning to eat (stage V) 227 tadpoles were living. While 146 of them were of the wild-type, 31 were Km-type albinos, 35 were Ty-type albinos and 15 were Km·Ty-type albinos.

These findings seem to indicate that genes *d* and *e* are located on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%.

## *IV. Coloration of 13 albino stocks*

### 1. External appearance

a. Wild-type

Mature *Rana nigromaculata* distinctly reveal sexual dimorphism during the breeding season. In males, the dorsal surfaces are grayish yellow or grayish yellow-green and usually have no dark spots, while in females the dorsal surfaces are grayish white or brownish gray in ground color and have many brownish-black spots. These spots are more or less connected with one another and in some cases they almost completely cover the back of the trunk. Both males and females have a pale stripe on the median line and two pale stripes on the dorso-lateral folds. While these stripes are distinct in females during the breeding season, the dorso-median stripe becomes especially conspicuous. The sexual difference in color and pattern is also observable in the seasons others than the breeding, although it is not conspicous (Plate I, 1, 2).

In contrast to mature frogs, juvenile *Rana nigromaculata* do not reveal sexual dimorphism which occurs owing to effects of sexual hormones. In other words, the color and pattern proper to this species can be only observed in juvenile frogs. The dorsal colors of juvenile *Rana nigromaculata* are roughly divided into three types, green, brown and a mixture in which the anterior part of the body is green and the posterior part is brown. The green part of the mixture type has various dimensions. On the dorsal surfaces of juvenile frogs there are many small black spots. The pupils are black. The eggs of *Rana nigromaculata* are generally grayish brown or brownish black in color of the animal half.

The dorsal colors of mature and juvenile *Rana brevipoda* are also divided into three types, green, brown and a mixture of green and brown, as found in juvenile

*Rana nigromaculata*. Typical specimens of *Rana brevipoda* have no dorso-median stripe. *Rana brevipoda* differ from *Rana nigromaculata* in that they reveal no sexual dimorphism in color and pattern. Moreover, mature frogs have nearly the same coloration as that of juvenile ones. The brown color of *Rana brevipoda* somewhat differs from that of juvenile *Rana nigromaculata* in that the former is darker. *Rana brevipoda* have a few black spots which are roundish and isolated from one another on the back (Plate I, 3, 4). The eggs of *Rana brevipoda* are usually bluish gray in color of the animal half.

b. Albinos of the first group

i) RA-type ($a^e a^e$) albinos

The dorsal surfaces of mature females are light greenish-yellow or light reddish-yellow. There are several beige or light-beige spots on the back. These spots correspond to the brownish-black spots of wild-type females and reflect the color of muscles seen through the semitransparent skin of the spot places. The dorsal surfaces of mature males are bright greenish-yellow or bright yellow and have no beige spots. On the hind limbs of both males and females, there are several beige spots. The pupils are bright red in contrast to the black pupils of wild-type frogs. The pupils of the albinos of the BR stock are also bright red and do not differ from those of the other albinos of the RA-type, although this stock belongs to *Rana brevipoda*. No sexual difference is found in color of pupils (Plate I, 5~8; Plate II, 9~16).

The RA-type albinos can be evidently distinguished from wild-type individuals at stage 20 immediately after hatching. While black melanophores in the eye areas of the wild-type individuals appear at the time between stages 19 and 20, they do not appear in the RA-type albinos. At the late tadpole stage (stage X), the RA-type albinos are light yellowish-orange in dorsal color. Their pupils are deep yellowish-pink. The RA-type albinos shortly after completion of metamorphosis and beginning to eat are light reddish-yellow, light yellow or light greenish-yellow in dorsal color and there is no sexual difference. These juveniles very closely resemble the mature females in color and pattern. The eggs of mature females are light greenish-yellow or light yellow in color of the animal half (Plate V, 38a).

ii) TJ-type ($a^t a^t$) albinos

The dorsal surfaces of the TJ-type albinos are somewhat darker than those of the RA-type albinos. Mature females are dull reddish-yellow and have a dull-yellow spot around each of several dermal tubercles, while mature males are dull yellow and have no spots around dermal tubercles. The hind limbs of both males and females are the same as the dorsal surfaces of mature females in ground color and have dull-yellow spots. The dorsal surfaces of juvenile males and females are bright or dull reddish-yellow. The pupils of the TJ-type albinos are deep red (Plate III, 17, 18).

The TJ-type albinos can be completely distinguished from wild-type individuals

at stage 20 immediately after hatching. They have no melanophores containing black pigment in the eye areas at this stage, while such melanophores appear between stages 19 and 20 in the wild-type individuals.

The dorsal surfaces of the TJ-type albinos at the late tadpole stage (stage X) are light orange and darker in color tone than those of the RA-type albinos. At this developmental stage, the pupils are bright red and darker in color tone than those of the RA-type albinos. The irises are surrounded with grayish frames.

The eggs of mature females are light reddish-yellow or light yellow and slightly darker than those of the RA-type albinos (Plate V, 38b).

iii) FC-type ($a^f a^f$) albinos

The dorsal surfaces of mature females are light yellowish-brown or dark yellow and darker in color tone than those of the TJ-type albinos. The spots surrounding dermal tubercles as well as those on the dorso-lateral surfaces of the trunk and the dorsal surfaces of the hind limbs have various colors from yellowish-brown to dark brown. The dorsal surfaces of mature males are dull yellow and have no spots surrounding the dermal tubercles. The pupils are dark grayish-red (Plate III, 19, 20).

The FC-type albinos can not be distinguished from wild-type individuals at the stage 20 immediately after hatching. Although both wild-type and FC-type individuals reveal melanophores containing black pigment around the eye areas of the tadpoles between stages 19 and 20, this pigment gradually fades away in the FC-type albinos in contrast to the wild-type tadpoles. At the age of about four days after beginning to eat (stage I), the albinos of this stock can be completely distinguished from the wild-type tadpoles (Plate III, 24c). The FC-type albinos are dull yellowish-orange at the late tadpole stage (stage X) and somewhat darker in color tone than the TJ-type albinos. At this stage, the pupils are red.

The eggs of mature females are light yellow or light reddish-yellow and completely the same as those of the TJ-type albinos in this respect (Plate V, 38c).

iv) Hybrid-type albinos in the first group

External characters were observed in three kinds of hybrid-type albinos produced in the first group, RA/TJ-type ($a^e a^t$), RA/FC-type ($a^e a^f$) and TJ/FC-type ($a^t a^f$). The RA/TJ-type ($a^e a^t$) albinos obtained from matings between RA-type ($a^e a^e$) and TJ-type ($a^t a^t$) albinos are intermediate between the two types of albinos in color of the dorsal surfaces and the pupils. The RA/FC-type ($a^e a^f$) albinos obtained from matings between RA-type ($a^e a^e$) and FC-type ($a^f a^f$) albinos as well as the TJ/FC-type ($a^t a^f$) albinos obtained from matings between TJ-type ($a^t a^t$) and FC-type ($a^f a^f$) albinos are also intermediate between the two parental type albinos in color of the dorsal surfaces and the pupils and various other characters. Thus, the dorsal surfaces of the above six kinds of albinos are arranged from light yellow to dark yellow in the following order: RA-type>RA/TJ-type>TJ-type>RA/FC-type>TJ/FC-type>FC-type albinos. The color tones of the pupils of these six kinds of albinos are also arranged from bright red to dark

red nearly in the same order as found in those of the dorsal surfaces. Although the TJ-type ($a^t a^t$), RA/FC-type ($a^e a^f$) and TJ/FC-type ($a^t a^f$) albinos are very similar to one another in color tone and the differences among them are very slight, the differences among the six kinds of albinos are evidently observed at the tadpole and juvenile frog stages (Plate III, 21～24).

c. Albinos of the second group

The second group includes only the albinos of the GO stock at present. The albinos of this group are due to a single recessive gene (*b*) whose locus differs from those of the albinos of the first group. Among mature GO-type albinos, there is usually an equal number of males and females. The dorsal surfaces of mature females are bright yellow and clearer than those of albinos of the other groups. Several spots tinged with dull reddish-yellow or beige are found around dermal tubercles on the dorsal and dorso-lateral surfaces of the trunk and on the dorsal surfaces of the hind limbs. The color of these spots reflects that of muscles seen through the semitransparent skin of the places where brown-black spots exist in mature wild-type females.

The dorsal surfaces of mature males are bright greenish-yellow and have no spots. However, several spots tinged with dull reddish-yellow or beige are found on the lateral and posterior surfaces of the trunk and on the dorsal surfaces of the hind limbs. The color of pupils is deep red, as found in the TJ-type albinos, although it is slightly lighter than the latter (Plate IV, 25, 26).

The GO-type albinos can scarcely be distinguished from wild-type individuals at stage 20 immediately after hatching. Although both wild-type and GO-type individuals reveal melanophores containing a small amount of black pigment around the eye areas of the tadpoles between stages 19 and 20, this pigment gradually fades away in the GO-type albinos in contrast to the wild-type tadpoles, in which the pigment becomes darker. The GO-type albinos can be completely distinguished from the wild-type tadpoles at stage 25, soon after beginning to eat.

At the late tadpole stage (stage X), the GO-type albinos are light orange in body color and slightly lighter than the TJ-type ones, although they are very similar to the latter. The pupils of the GO-type albinos at this stage are bright red and slightly lighter than those of the TJ-type albinos.

The dorsal surfaces of male and female juvenile frogs are bright yellow, as those of mature females. The eggs of mature females are light yellow (Plate V, 38d).

d. Albinos of the third group

This group includes only the albinos of the YM I stock at present. These albinos are due to a single recessive gene (*c*) whose locus differs from those of the first and second groups. Among the albinos of this group, there is always nearly an equal number of males and females.

The dorsal surfaces of mature females are deep yellow. There are several spots tinged with yellowish or dark brown on the dorsal, lateral and posterior surfaces of the trunk and the dorsal surfaces of the hind limbs. The dorsal surfaces of

mature males are dull yellow. Somewhat obscure spots as compared with those of females are found on the dorsal and lateral surfaces of the trunk and the dorsal surfaces of the hind limbs. The pupils of males and females are dark red. The albinos of this group show a good growth and become larger than the control wild-type frogs (Plate IV, 27, 28).

The YM I-type albinos can be correctly distinguished from wild-type individuals at stage 20 immediately after hatching. While melanophores containing black pigment appear in the eye areas of wild-type individuals between stages 19 and 20, this pigment is very faint in the YM I-type albinos, although it appears. At the late tadpole stage (stage X), the YM I-type albinos are light yellow in dorsal color. Their pupils are red and somewhat darker than those of the TJ-type albinos belonging to the first group.

The dorsal surfaces of juvenile frogs are light reddish-yellow in both males and females. The eggs of mature females are pale beige (Plate V, 38e).

e. Albinos of the fourth group

The albinos of the KM stock are included in this group. These albinos are due to a single recessive gene (*d*) whose locus differs from those of the albinos of the first, second and third groups. In the KM-type albinos, there is nearly an equal number of males and females. The dorsal surfaces of mature albinos of this stock are semitransparent and light reddish-yellow in appearance. Both males and females have several spots tinged with dull reddish-yellow or various tones of beige on the dorsal and lateral surfaces of the trunk and on the dorsal surfaces of the hind limbs. The pupils are deep red. The irises are semitransparent and tinged with grayish yellow (Plate IV, 29, 30).

The KM-type albinos can not be distinguished from wild-type individuals at stage 20 immediately after hatching. When they begin to eat, the dark pigment around the eyes becomes paler and iridophores covering the body lose their luster. At the age of about seven days after beginning to eat (stage II), the albinos of this stock can be completely distinguished from the wild-type tadpoles. The whole bodies of the albinos are semitransparent and the dorsal surfaces are pinkish-beige or common beige. They are the darkest among the tadpoles of the 13 albino stocks. The pupils are bright red and slightly lighter than those of the TJ-type albinos.

The eggs of mature females are beige and the darkest among those of the 13 albino stocks except the eggs of the albinos of the following fifth group (Plate V, 38f).

f. Albinos of the fifth group

This group includes the albinos of the TY and Ns stocks. These albinos are due to a single recessive gene (*e*) whose locus differs from those of the albinos belonging to the other four groups. As there are no differences in color and pattern of both juvenile and mature frogs between these two stocks, the latter is named TY-type. In the albinos of each of the two stocks, nearly an equal number

of males and females is always found.

The dorsal surfaces of mature frogs are dull yellow-green or brownish olive, corresponding to the green-type or brown-type of normal-colored frogs. They have several spots tinged with green, dark brown or deep yellowish-brown on the dorsal and lateral surfaces of the trunk and the dorsal surfaces of the hind limbs. Pupils are reddish black (Plate IV, 32; Plate V, 33~35).

The Ty-type albinos can not be distinguished from wild-type individuals at stage 20 immediately after hatching. However, the pigment of melanophores in the areas around the eyes begins to become paler than that of the wild-type tadpoles at stage 22. At stage 25 when tadpoles begin to eat, the eyes of the Ty-type albinos are evidently paler, while the body color of the latter begins to fade away at the age of seven days after beginning to eat. At stage III about 14 days after beginning to eat, it is very easy to distinguish the albinos from the wild-type tadpoles.

The bodies of the Ty-type albinos at the late tadpole stage (stage X) are light yellowish-orange and their pupils are deep red or dark red. The dorsal surfaces of juvenile frogs immediately after metamorphosis are light yellowish-orange and do not remarkably differ from those of juvenile albinos of the other groups (Plate IV, 31). However, since the time when the frogs begin to take food, the body color gradually becomes darker and is dull yellow-green or brownish-olive at the stage before sexual maturity. No differences in color and pattern are found between mature males and females.

The eggs of mature females are slightly lighter in color tone than those of wild-type females (Plate V, 38g). There are larger and smaller eggs besides normal-sized ones. Many frogs have right and left eyeballs which are unequal in size. The Ty-type albinos are usually weak in viability since stage V as compared with those of the other groups.

g. Summary of color patterns in the seven phenotypes

The colors of the dorsal surfaces and pupils of albinos are arranged as follows from light to dark.

Ra-type ($a^e a^e$) > Go-type ($bb$) > Tj-type ($a^t a^t$) > Km-type ($dd$) >
Ym I-type ($cc$) > Fc-type ($a^f a^f$) > Ty-type ($ee$)

The colors of the animal halves of eggs laid by mature female albinos are arranged as follows from light to dark (Plate V, 38).

Ra-type ($a^e a^e$) > Go-type ($bb$) > Tj-type ($a^t a^t$) ≥ Fc-type ($a^f a^f$) >
Ym I-type ($cc$) > Km-type ($dd$) > Ty-type ($ee$)

The body colors of albinos at the late tadpole stage (stage X) are arranged as follows from light to dark.

Ra-type ($a^e a^e$) > Go-type ($bb$) > Tj-type ($a^t a^t$) > Ym I-type ($cc$) >
Ty-type ($ee$) > Fc-type ($a^f a^f$) > Km-type ($dd$)

## 2. Electron-microscopic structure

The ultramicroscopic structure of the dermal chromatophores in the dorsal

skin of the trunk and the spot areas of the hind limbs of albinos belonging to the 13 stocks was examined under an electron microscope. Of the three kinds of chromatophores, xanthophores, iridophores and melanophores, the melanophores always exist under the iridophores and above the collagenous layer of the dermis. While the melanophores of wild-type frogs are filled with completed melanosomes, those of albinos are abnormal and contain premelanosomes or immature melanosomes. No completed melanosomes are usually found in the melanophores of any kind of albinos. Albinos of different groups or strains differ from one another in development, size, number and structure of premelanosomes.

The xanthophores in various kinds of albinos do not differ from those of wild-type frogs except that those of the Go-type albinos of the second group contain very abundant carotenoid vesicles. The iridophores in various kinds of albinos are completely normal except that those of the KM-type albinos of the fourth group are incomplete in formation of reflecting platelets.

a. Developmental stages of melanosomes

The developmental stages of melanosomes from the most primitive premelanosomes to the most completed melanosomes found in the 13 stocks of albinos as well as in wild-type frogs are divided into the following six stages (Fig. 3).

Stage I. The premelanosomes are small vesicles which are spherical or spheroidal in shape and smaller than 0.3 $\mu$ in major axis. They are enveloped with a limiting double membrane. The inside is almost homogeneous and contains a vague structure similar to the cytoplasmic matrix or an indistinct fibrous structure (Fig. 3a).

Stage II. The most advanced premelanosomes at this stage are long-ellipsoidal. In the longitudinal section, each premelanosome is enveloped with a limiting double membrane and contains six to nine fibers which are about 80 Å in width and arranged parallel along the long axis. Each fiber appears to be a string of minute granules which are about 75 Å in diameter or to be a coiled thread. The minute granules or definite parts of each fiber are connected with those of the neighboring fibers in transversal, parallel lines. Thus, the fibers form several plates which are curved along the inner wall of the limiting membrane or form incomplete tubes. A transversal section of each premelanosome shows a kind of concentric structure (Fig. 3b).

Stage III. In the premelanosomes at this stage, the longitudinal fibers are distinctly thicker than those of stage II. The coiling structure of each fiber is distinctly observable. As found in premelanosomes of stage II, several longitudinal fibers form a plate by connecting with the neighboring fibers. In the most advanced premelanosomes, these plates are contracted in width and the concentric structure found in the transversal section of each premelanosome is considerably resolved. As the fibers are fairly electron-dense, the premelanosomes are easily distinguished from the other parts of cytoplasm in an unstained preparation in contrast to those of stages I and II (Fig. 3c).

Stage IV. The longitudinal fibers found in the longitudinal section of each

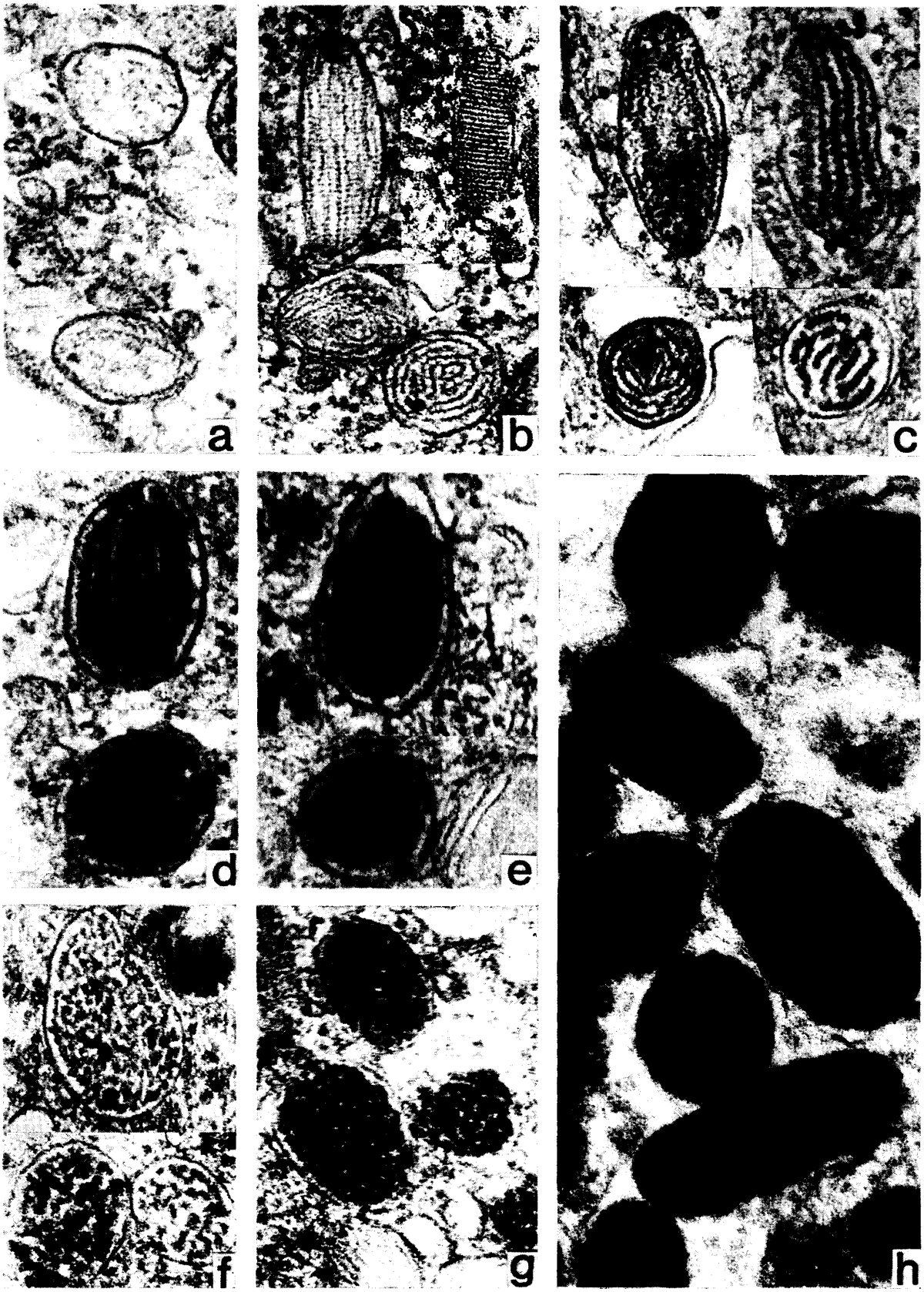


Fig. 3. Developmental stages of melanosomes. ×64000

a~g. Premelanosomes. h. Completed melanosomes.

a. Stage I. b. Stage II. c. Stage III. d. Stage IV.
e. Stage V. f. Stage IIIa. g. Stage IVa. h. Stage VI.

premelanosome are remarkably thick and almost fill up the inside of the premelanosome. The fibers are about 320 Å in thickness. In the transversal section, the concentric structure is scarcely recognizable. The inside of the limiting double membrane seems to be filled with granules which are about 320 Å in diameter and connected irregularly with the neighboring granules. As the longitudinal fibers are remarkably electron-dense, the premelanosomes are conspicuous from the other parts of cytoplasm in an unstained preparation. They seem to be light brown under an optical microscope, although their shape and structure are not observable (Fig. 3d).

Stage V. The longitudinal fibers are extremely thick and fused with one another. They become an almost homogeneous electron-dense mass. As compared with completed melanosomes in the melanophores of wild-type frogs, the premelanosomes of stage V are smaller and lower in electron density. They have a distinct limiting membrane in contrast to the completed melanosomes. Under an optical microscope, their shape and structure are obscure, although they seem to be almost black (Fig. 3e).

Stage VI. The melanosomes observed in melanophores of wild-type frogs have no limiting membrane in most cases. They are homogeneous and extremely electron-dense in an unstained preparation. They are ellipsoidal and usually about 0.5 $\mu$ in major axis (Fig. 3h).

Modification of stages III and IV. Whereas the premelanosomes and melanosomes found in albinos of the 13 stocks and wild-type frogs are divided into six developmental stages, there are two modifications of premelanosomes, which are named stages IIIa and IVa.

In stage IIIa, the longitudinal fibers found in premelanosomes of stage II are broken to pieces, and the inside of the limiting membrane of each premelanosome is coarsely filled with these pieces. Thus, only a granular structure is seen in both the longitudinal and transversal sections of the premelanosomes (Fig. 3f). In stage IVa, the inside of the premelanosomes is closely filled with granules which have increased in size and electron density. The premelanosomes of stages IIIa and IVa are almost spheres in most cases. They seem to have changed from an ellipsoidal to a spherical form (Fig. 3g).

b. Chromatophores of wild-type frogs

In the dorsal skin of wild-type frogs, there are three kinds of dermal chromatophores, xanthophores, iridophores and melanophores (Fig. 4a). The melanophores are arranged at the lowest position. When the number of melanosomes and premelanosomes was counted in 20 squares, 2.3 $\mu \times 2.3$ $\mu$, of melanophores

Fig. 4. Electron microphotographs of dermal chromatophores in the dorsal skin of a wild-type *Rana nigromaculata*.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore
b. A part of a normal melanophore. ×34000
ME, melanosomes MT, mitochondria

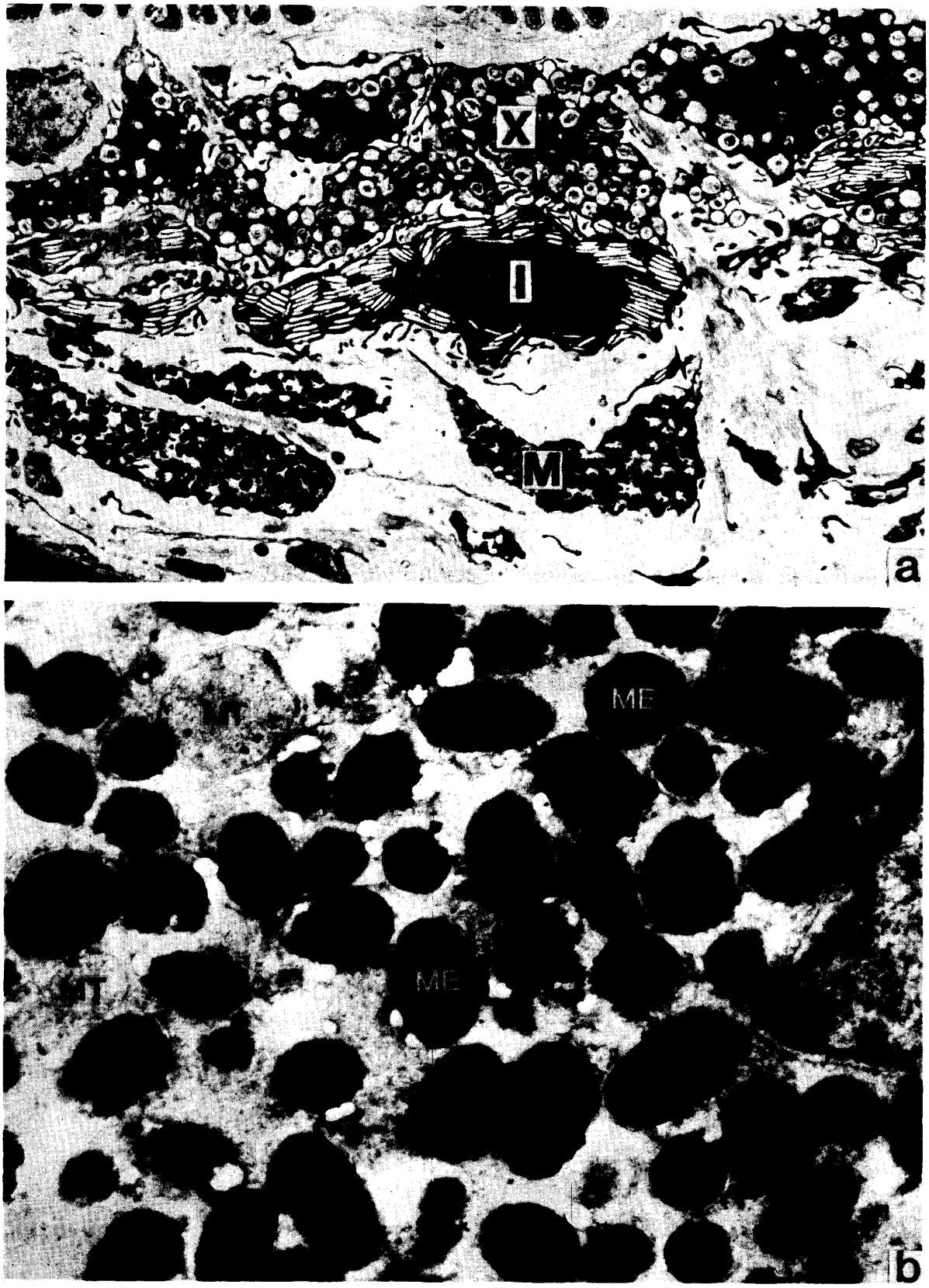


Fig. 4

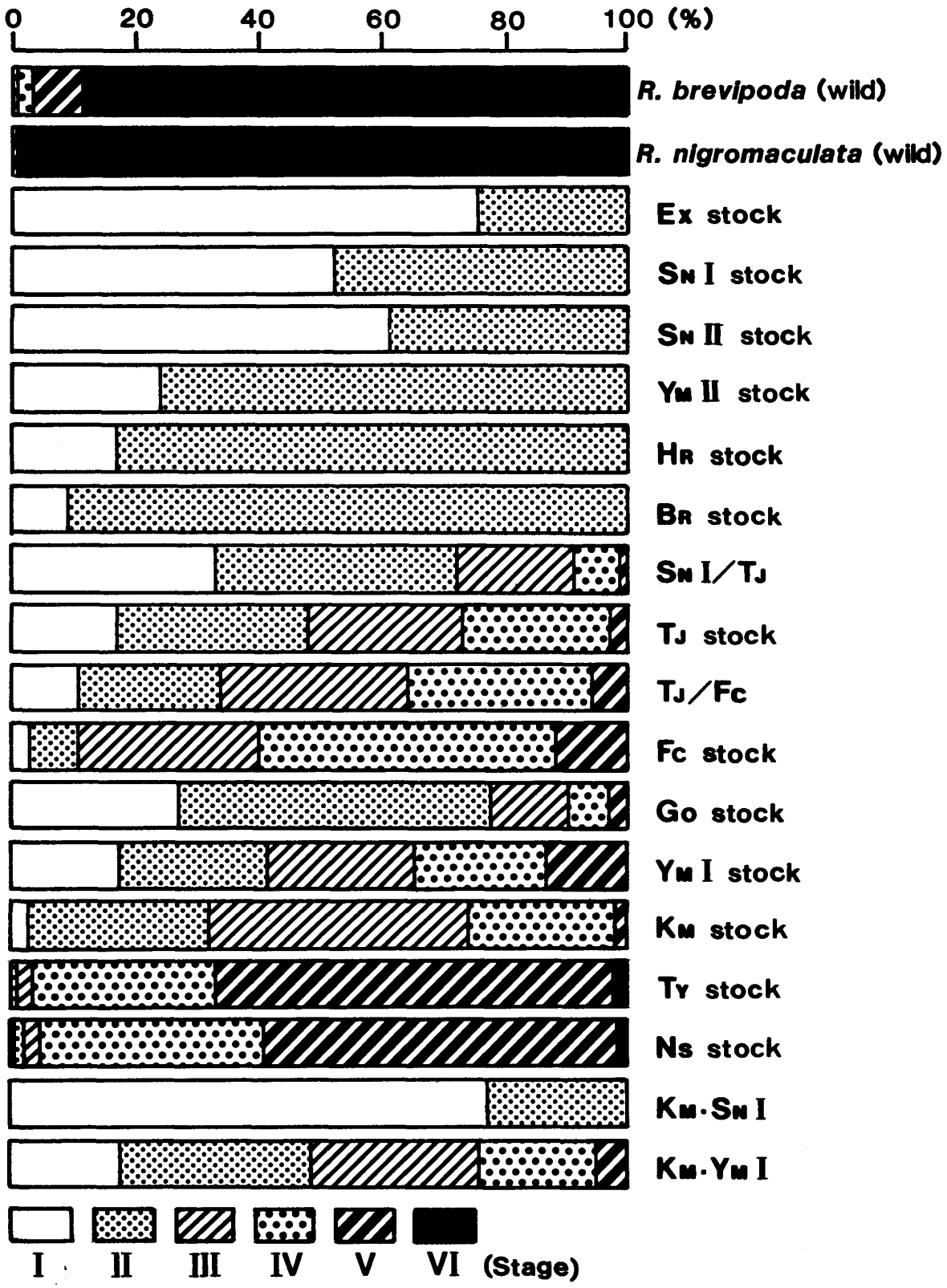


Fig. 5. Percentages of six developmental stages of melanosomes in 17 kinds of albinos and wild-type frogs of the two species.

in the dorsal skin of a green wild-type frog, one square contained 26~41, 32.3±1.0 on the average. When the numbers in 20 squares were totalled, they were 646, of which 99.2% were melanosomes of stage VI, while 0.6% and 0.2% were premelanosomes of stages V and IV, respectively (Fig. 5). When the size of 20 melanosomes was measured, they were 0.51±0.02 $\mu$ in major axis and 0.34±0.01 $\mu$ in minor axis (Fig. 4b).

c. Chromatophores of the first group

i) RA-type ($a^e a^e$) albino

Ex stock

While the xanthophores and iridophores of the Ex stock are normal and do not differ from those of the wild-type frogs, the melanophores are very abnormal (Fig. 6a). Their cytoplasm is meager even in the part surrounding the nucleus. The dendritic processes are remarkably slender. In an unstained preparation, no premelanosomes are distinguishable from the neighboring cytoplasmic matrix. There are only premelanosomes of stages I and II which are very small, being 0.25±0.01 $\mu$ in major axis and 0.12±0.05 $\mu$ in minor axis. They are extremely sparse in distribution. When the number of premelanosomes was counted in 20 squares, each of which was 2.3 $\mu$×2.3 $\mu$ in size, one square contained 0~18 premelanosomes, 4.4±1.1 on the average. The total 20 squares contained 87 premelanosomes, of which 75.0% and 25.0% were of stages I and II, respectively (Fig. 5). No more advanced stages of premelanosomes are found in this stock. In the cytoplasm, there are numerous mitochondria and abundant vacuoles of various sizes, while rough endoplasmic reticula are scarcely found (Fig. 6b).

SN I stock

As in the Ex stock, xanthophores and iridophores of this stock are normal, while the melanophores are very abnormal and contain no melanosomes (Fig. 7a). When the number of premelanosomes was counted in 20 squares, each of which was 2.3 $\mu$×2.3 $\mu$ in size, one square contained 0~26, 3.7±0.8 on the average. The total 20 squares contained 74 premelanosomes, of which 52.0% and 48.0% were of stages I and II, respectively (Fig. 5). While mitochondria, minute granules, being about 23 m$\mu$ in diameter, and vacuoles of various sizes are abun-

Fig. 6. Electron microphotographs of dermal chromatophores in the dorsal skin of an albino of the Ex stock ($a^e a^e$) belonging to the first group.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore
b. A part of an abnormal melanophore. ×34000
PM, premelanosomes MT, mitochondria V, vacuoles

Fig. 7. Electron microphotographs of dermal chromatophores in the dorsal skin of an albino of the SN I stock ($a^e a^e$) belonging to the first group.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore
b. A part of an abnormal melanophore. ×34000
PM, premelanosomes G, GOLGI apparatus MT, mitochondria V, vacuoles

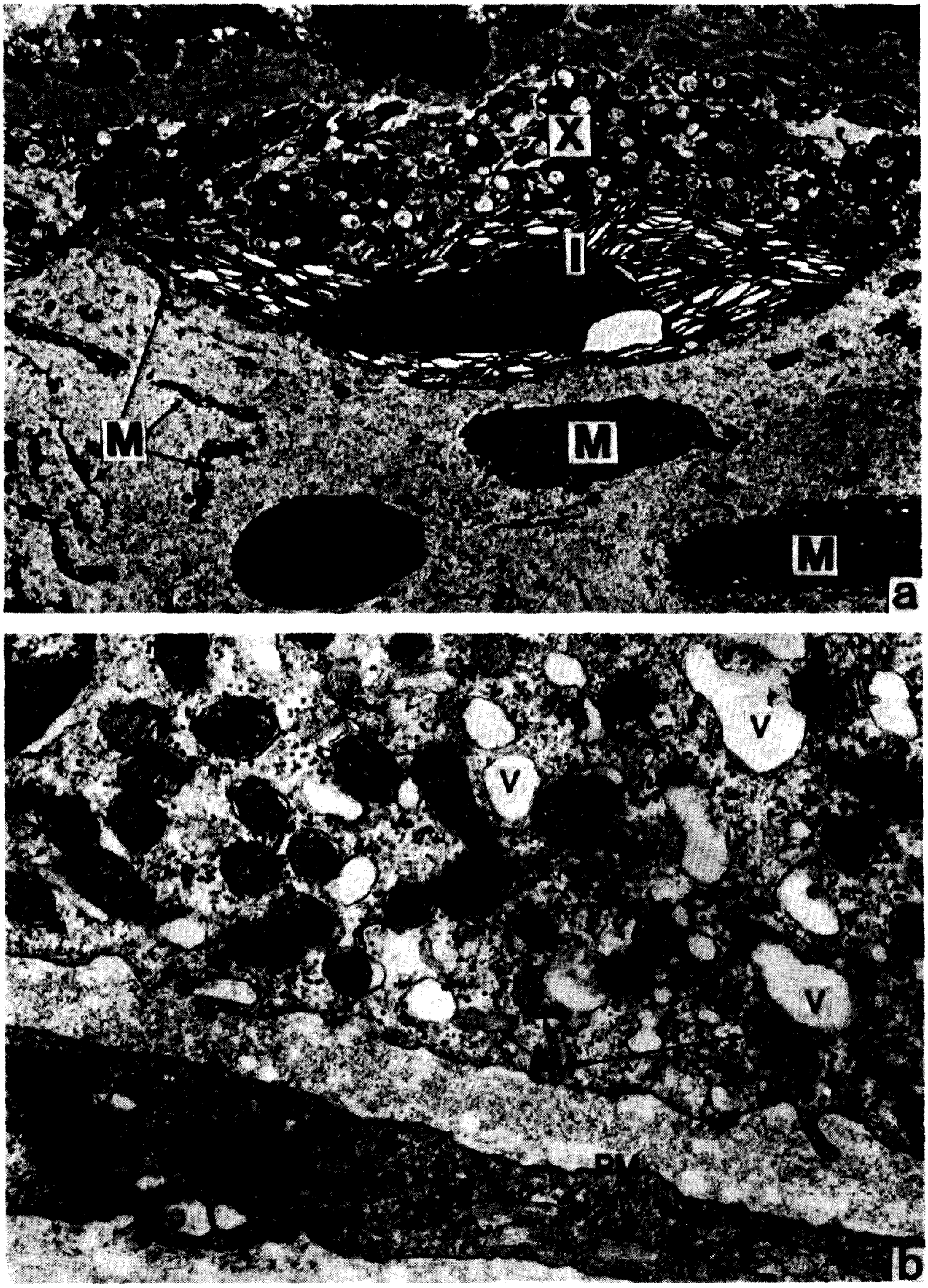


Fig. 6

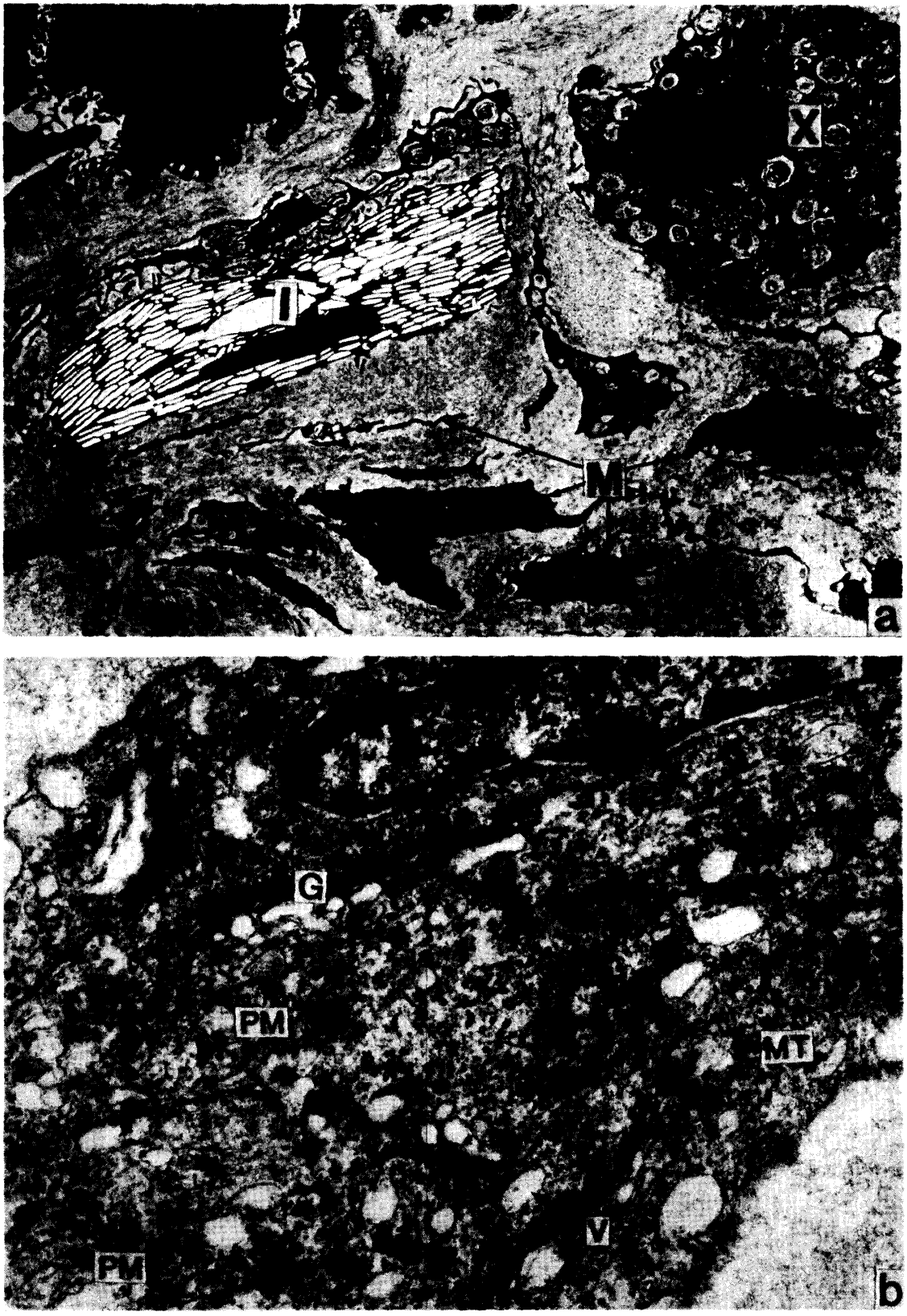


Fig. 7

dant in the cytoplasm, rough endoplasmic reticula are scarcely found. The dendritic processes of melanophores are slender and emaciated. The premelanosomes are remarkably smaller as compared with completed melanosomes. They were $0.23 \pm 0.02\ \mu$ and $0.11 \pm 0.01\ \mu$ in major and minor diameters, respectively (Fig. 7b).

Sn II stock

As in the above two albino stocks, the xanthophores and iridophores are normal, but the melanophores are very abnormal. The dendritic processes of melanophores are very slender. When the number of premelanosomes was counted in 20 squares, each of which was $2.3\ \mu \times 2.3\ \mu$ in size, one square contained $0 \sim 16$, $3.9 \pm 0.9$ on the average. The total 20 squares contained 78 premelanosomes, of which 61% and 39% were of stages I and II, respectively (Fig. 5). In the cytoplasm, mitochondria and small vacuoles were abundantly found. The premelanosomes were $0.25 \pm 0.01\ \mu$ and $0.12 \pm 0.01\ \mu$ in major and minor diameters, respectively (Fig. 8).

Ym II stock

The xanthophores and iridophores of this stock are completely normal. In contrast, the melanophores are very abnormal and contain no melanosomes. When the number of premelanosomes was counted in 20 squares, each of which

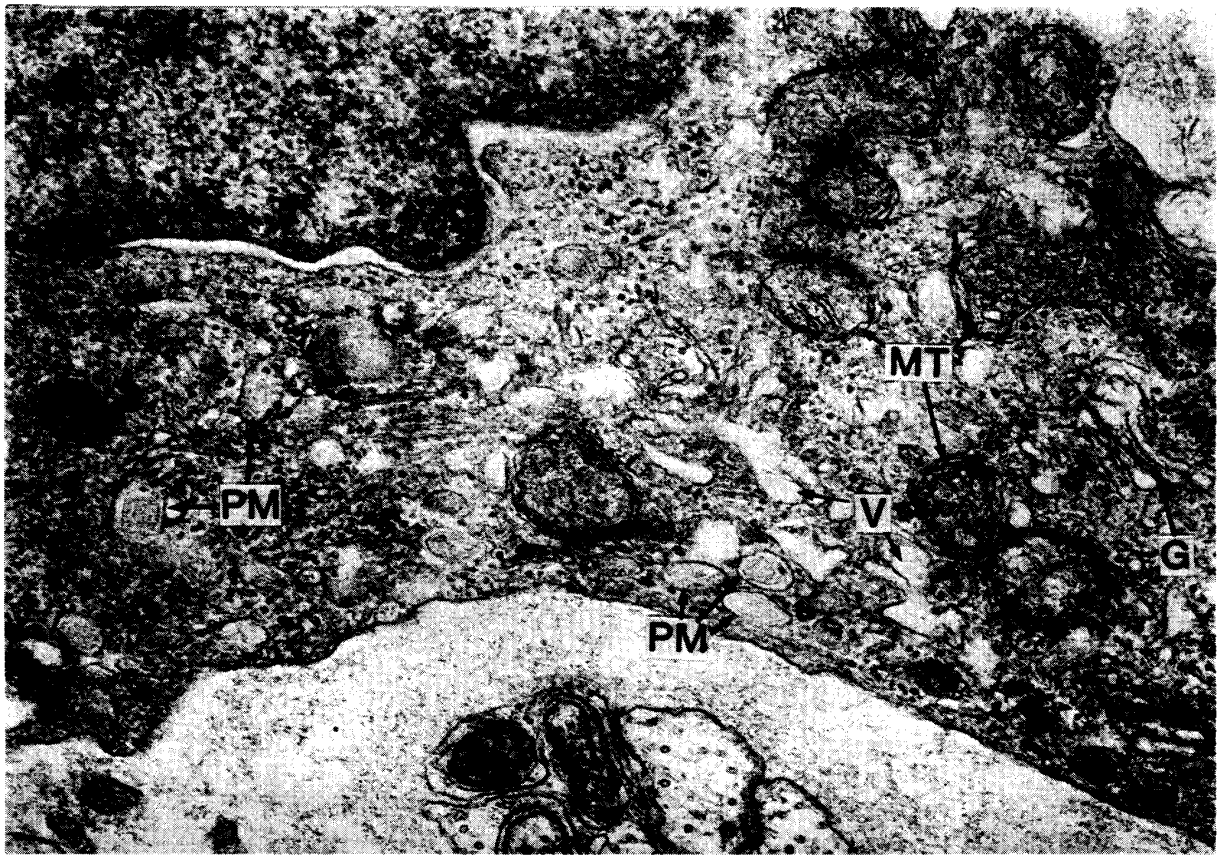


Fig. 8. Electron microphotograph of an abnormal melanophore in the dorsal skin of an albino of the Sn II stock ($a^e a^e$) belonging to the first group. ×34000

PM, premelanosomes MT, mitochondria V, vacuoles G, Golgi apparatus

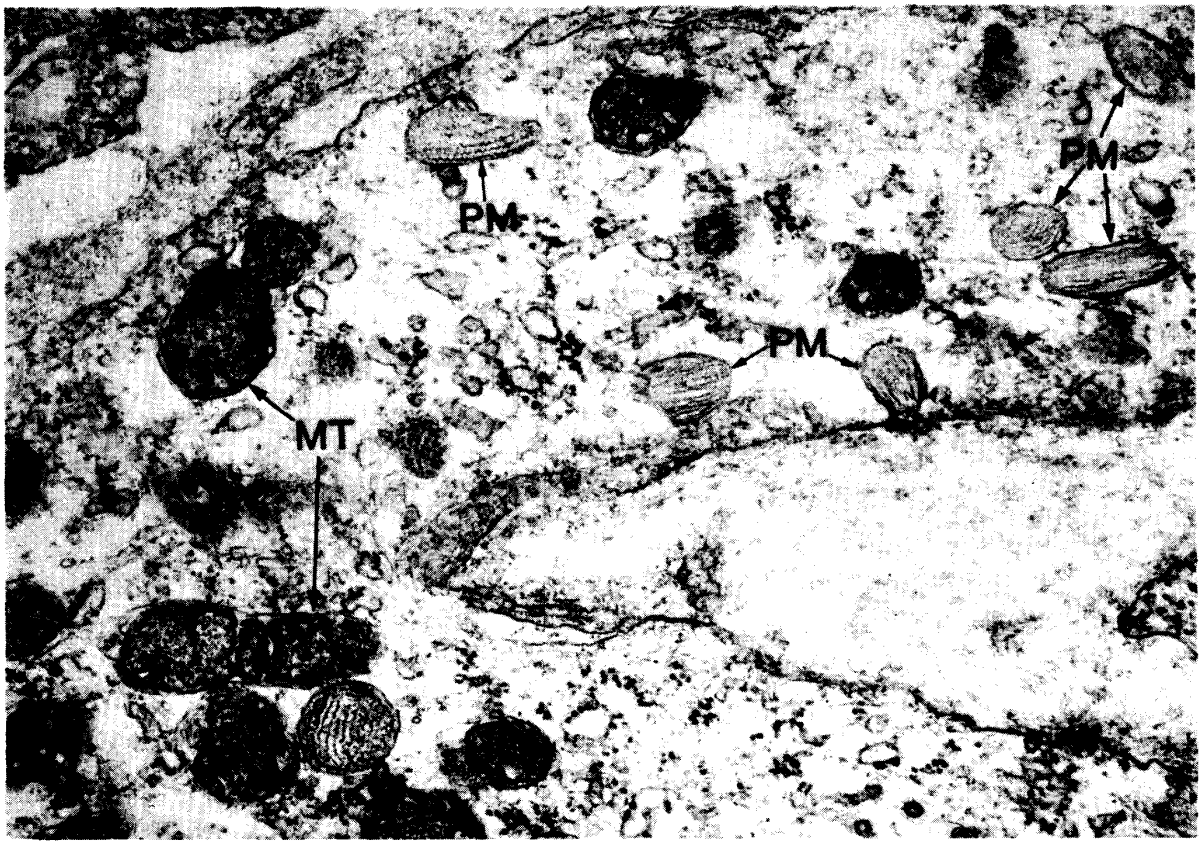


Fig. 9. Electron microphotograph of an abnormal melanophore in the dorsal skin of an albino of the YM II stock ($a^e a^e$) belonging to the first group. ×34000
PM, premelanosomes MT, mitochondria

was 2.3 $\mu \times 2.3$ $\mu$ in size, one square contained 0~22, $5.7 \pm 1.3$ on the average. The total 20 squares contained 113 premelanosomes, of which 24% and 76% were of stages I and II, respectively (Fig. 5). There were no premelanosomes which were observable in an unstained preparation. The premelanosomes were $0.30 \pm 0.01$ $\mu$ and $0.25 \pm 0.01$ $\mu$ in major and minor axes, respectively (Fig. 9). The melanophores of this stock are nearly the same in the other respects as those of the other RA-type stocks.

HR stock

The melanophores of this stock are very abnormal, while the xanthophores and iridophores are completely normal. They contain only premelanosomes of stages I and II, which are unobservable in an unstained preparation, as those of the above four stocks. However, they differ from the latter in that they contain abundant premelanosomes. In other words, their cytoplasm is usually filled up with premelanosomes, while that of the above four stocks is almost occupied with organelles other than premelanosomes. When the number of premelanosomes was counted in 20 squares, each of which was 2.3 $\mu \times 2.3$ $\mu$ in size, one square contained 6~42, $26.6 \pm 2.3$ on the average. The total 20 squares contained 531 premelanosomes, of which 17% and 83% were of stages I and II, respectively (Fig. 5).

In the melanophores lying just under iridophores, reflecting platelets and pterinosomes are found in a considerably high frequency among premelanosomes. Such melanophores contain well-developed lysosomes and numerous groups consisting of several premelanosomes surrounded with a limiting membrane. The melanophores have dendritic processes which are remarkably thicker than those of the melanophores of the above four stocks. The premelanosomes contained in the melanophores are somewhat larger than those contained in the latter melanophores, that is, $0.35 \pm 0.01\ \mu$ and $0.21 \pm 0.01\ \mu$ in major and minor diameters, respectively (Fig. 10).

Br stock

The dorsal skin of mature wild-type *Rana brevipoda* has three kinds of dermal chromatophores, xanthophores, iridophores and melanophores, as found in *Rana nigromaculata* (Fig. 11a). However, these chromatophores of *Rana brevipoda* somewhat differ in development from those of *Rana nigromaculata*. While the xanthophores and melanophores of *Rana brevipoda* are slightly inferior in development to those of *Rana nigromaculata*, the iridophores of the former are slightly superior to those of the latter. The melanosomes contained in the melanophores are mostly accomplished ones (stage VI). When the number of melanosomes or premelanosomes were counted in 20 squares, each of which was $2.3\ \mu \times 2.3\ \mu$ in size, one square contained $25 \sim 42$, $31.8 \pm 1.1$ on the average. The total 20 squares contain 636, of which 1.0%, 2.5%, 7.5% and 89.0% were of stages III, IV, V and VI, respectively (Fig. 5). The number of premelanosomes was slightly larger than that in *Rana nigromaculata*. When the size of 20 melanosomes was measured, they were $0.50 \pm 0.02\ \mu$ and $0.34 \pm 0.01\ \mu$ in major and minor axes, respectively (Fig. 11b).

The xanthophores and iridophores of albinic *Rana brevipoda* do not differ from those of wild-type specimens. In contrast, the melanophores are very abnormal and extremely ill-developed. They contain no melanosomes (Fig. 12a). When the number of premelanosomes was counted in 20 squares, each of which was $2.3\ \mu \times 2.3\ \mu$ in size, each square contains $0 \sim 11$, $3.2 \pm 0.6$ on the average. The total 20 squares contain 64 premelanosomes, of which 9% and 91% were of stages I and II, respectively (Fig. 5). When the size of 20 premelanosomes was meas-

Fig. 10. Electron microphotographs of abnormal melanophores in the dorsal skin of an albino of the Hr stock ($a^e a^e$) belonging to the first group. ×34000

a. An abnormal melanophore containing reflecting platelets, a pterinosome, well-developed lysosomes and premelanosomes.
RP, reflecting platelets PT, pterinosome L, lysosomes PM, premelanosomes

b. An abnormal melanophore containing abundant premelanosomes.

Fig. 11. Electron microphotographs of dermal chromatophores in the dorsal skin of a wild-type *Rana brevipoda*.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore

b. A part of a normal melanophore. ×34000
ME, melanosomes PM, premelanosomes

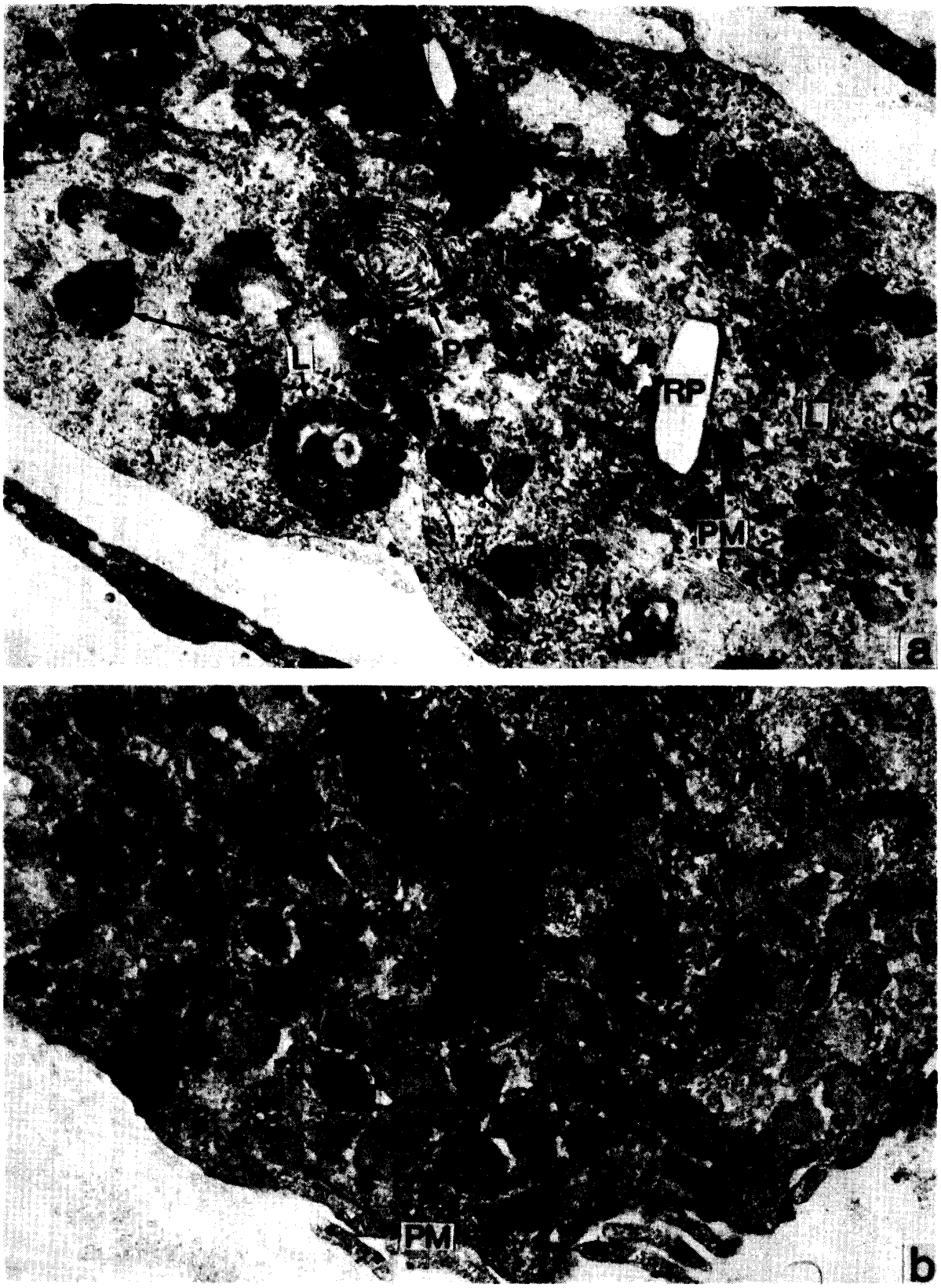


Fig. 10

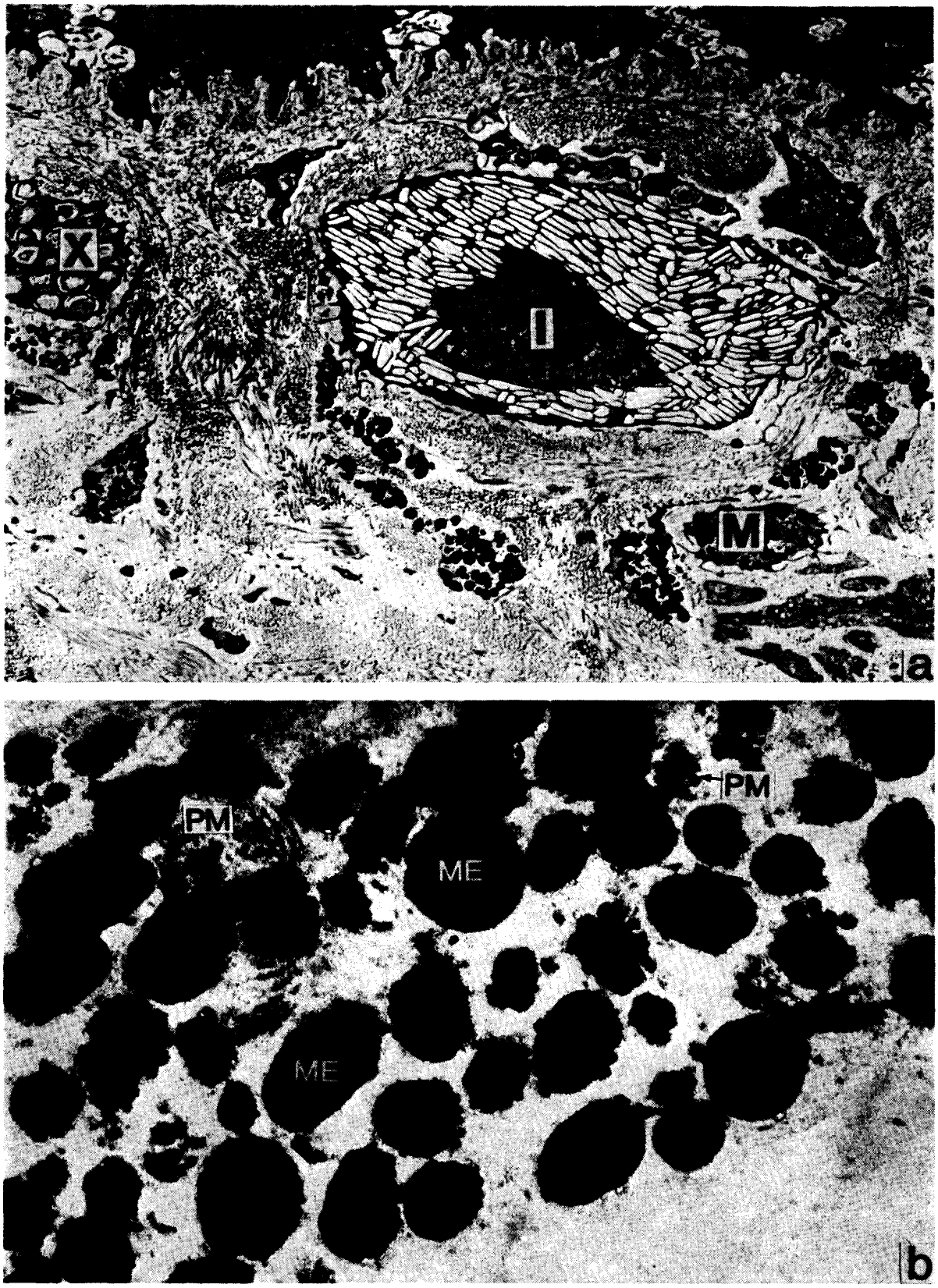


Fig. 11

ured, they were $0.32 \pm 0.04\ \mu$ and $0.13 \pm 0.01\ \mu$ in major and minor axes, respectively, and completely similar to those of the Ex, Sn I, Sn II and Ym II stocks in size (Fig. 12b).

ii) Tj-type ($a^t a^t$) albino

While the xanthophores and iridophores are completely normal, the melanophores are very abnormal (Fig. 13a). However, the melanophores of this stock remarkably differ in abnormality from those of the stocks stated above. The dendritic processes are thicker than those of melanophores of the Hr stock. When the number of premelanosomes was counted in 20 squares, each of which was $2.3\ \mu \times 2.3\ \mu$ in size, one square contained $15 \sim 30$, $21.8 \pm 1.1$ on the average. The total 20 squares had 436 premelanosomes, of which 16.5%, 31.0%, 25.0%, 24.0% and 3.5% were of stages I, II, III, IV and V, respectively (Fig. 5). In the cytoplasm of melanophores, considerably numerous mitochondria are observed. The premelanosomes were $0.36 \pm 0.01\ \mu$ and $0.25 \pm 0.01\ \mu$ in major and minor axes, respectively (Fig. 13b).

iii) Fc-type ($a^f a^f$) albino

The xanthophores and iridophores are normal. Although the melanophores are abnormal and contain no melanosomes, they are superior to those of the Tj stock in development. Their dendritic processes are thick and expanded in a wide area. When the number of premelanosomes was counted in 20 squares, each of which was $2.3\ \mu \times 2.3\ \mu$ in size, one square contained $20 \sim 42$, $33.0 \pm 1.6$ on the average. The total 20 squares had 660 premelanosomes, which were of various stages from I to V, although most of the premelanosomes of stage III and all the premelanosomes of stage IV were replaced with those of stages IIIa and IVa. In this stock, most of the premelanosomes of the later stage are nearly spherical. Of the 660 premelanosomes, 2.5%, 8.0%, 28.5%, 49.0% and 12.0% were of stages I, II, III+IIIa, IVa and V, respectively (Fig. 5). The cytoplasmic matrix of melanophores is somewhat dense and includes innumerous minute granules besides many mitochondria. The premelanosomes were $0.30 \pm 0.01\ \mu$ and $0.26 \pm 0.01\ \mu$ in major and minor axes, respectively (Fig. 14).

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Fig. 12. Electron microphotographs of dermal chromatophores in the dorsal skin of an albino of the Br stock ($a^e a^e$) belonging to the first group.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore
b. A part of an abnormal melanophore. ×34000
PM, premelanosomes V, vacuoles

Fig. 13. Electron microphotographs of dermal chromatophores in the dorsal skin of an albino of the Tj stock ($a^t a^t$) belonging to the first group.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore
b. A part of an abnormal melanophore. ×34000
PM, premelanosomes MT, mitochondria V, vacuoles

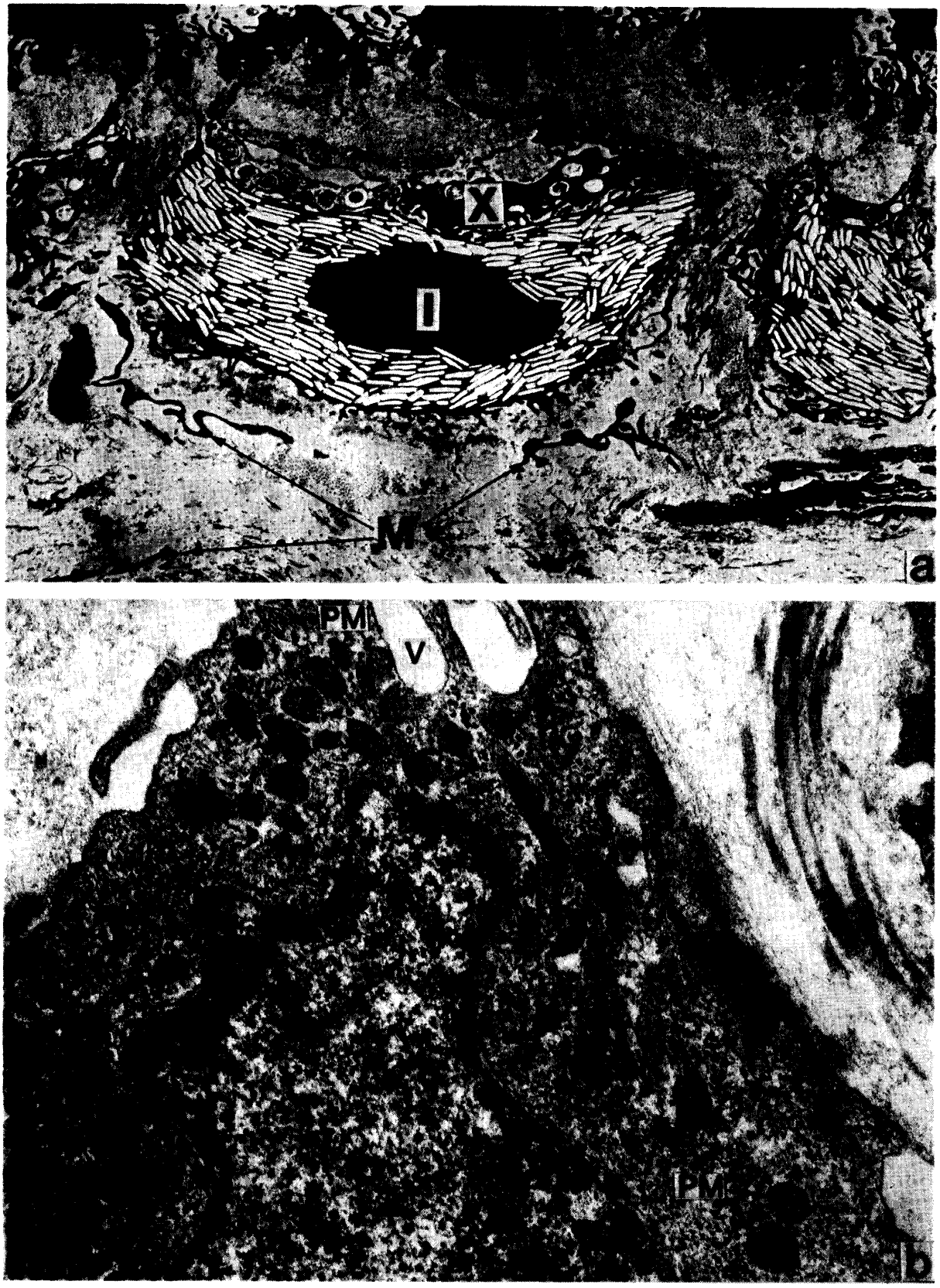


Fig. 12

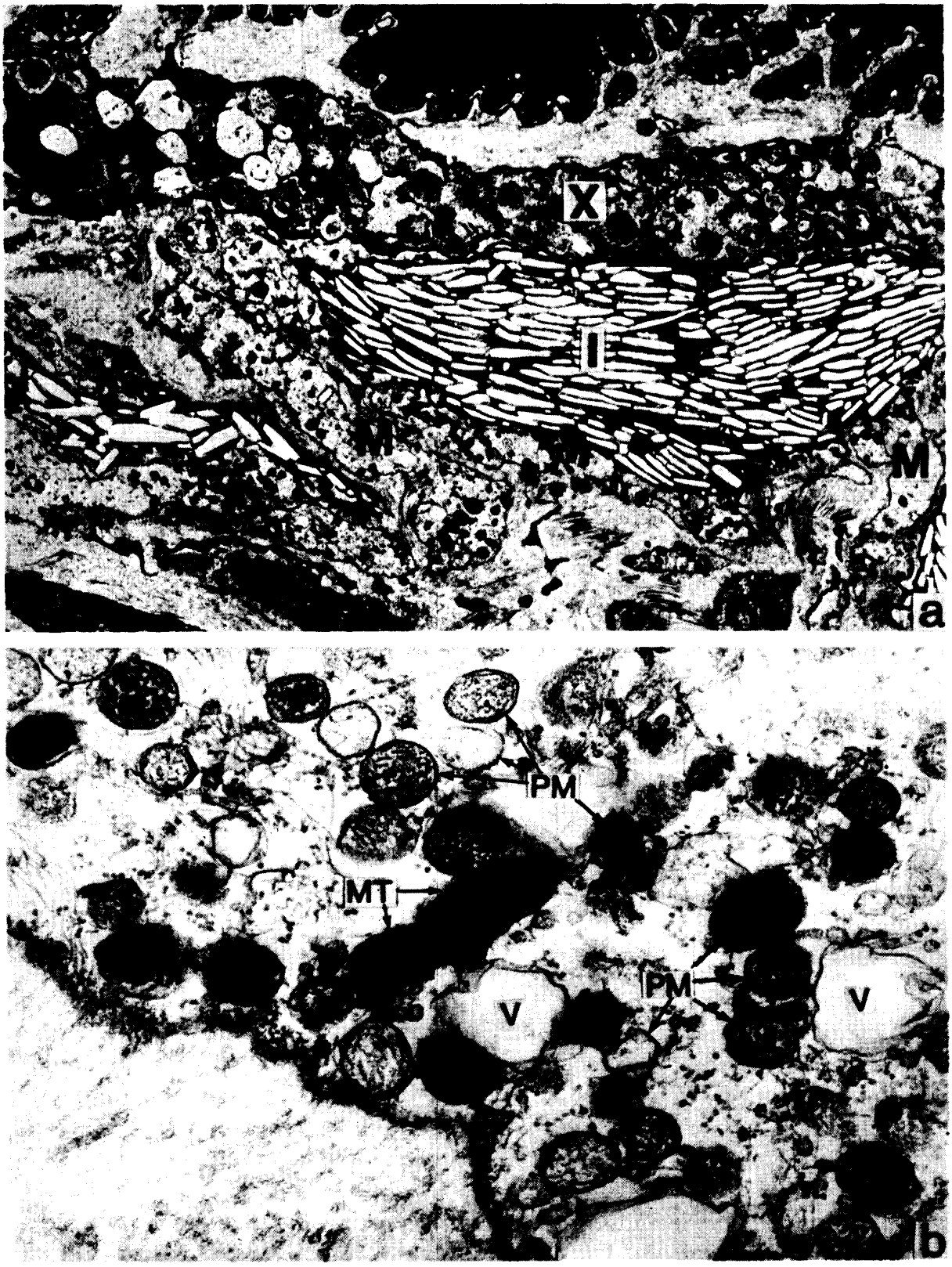


Fig. 13

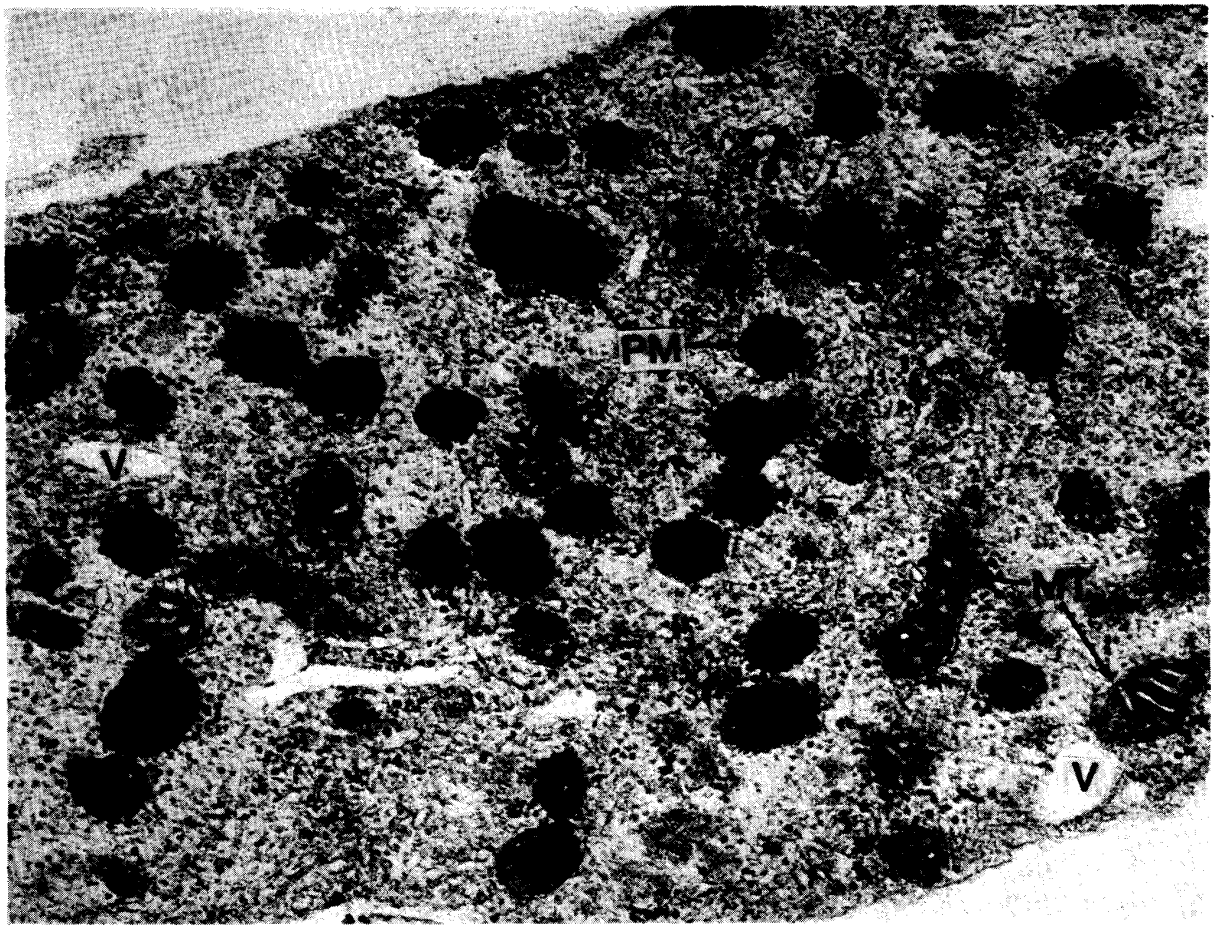


Fig. 14. Electron microphotograph of an abnormal melanophore in the dorsal skin of an albino of the Fc stock ($a^f a^f$) belonging to the first group. ×34000
PM, premelanosomes MT, mitochondria V, vacuoles

iv) Ra/Tj-type ($a^e a^t$) albino

Mature male and female albinos (Ra/Tj-type) produced in 1975 from a mating, Sn I. Het. 72♀, No. 3×Tj. Alb. 74♂, No. 1, were observed. These albinic hybrids ($a^e a^t$) are intermediate in coloration between the albinos of the Sn I and Tj stocks, as described above. An electron-microscopic observation of the dorsal skin shows that the xanthophores and iridophores are normal, while the melanophores are abnormal. The melanophores are situated under the iridophores as usual. Their dendritic processes are usually thicker than those of the Sn I stock. The cytoplasm surrounding the nucleus is somewhat voluminous. In this respect, the melanophores of these albinic hybrids resemble those of the Tj stock. They contain more numerous premelanosomes than those of the albinos of the Sn I stock and, moreover, include premelanosomes of stages III, IV and V besides I and II, in contrast to the Sn I stock whose albinos contain only premelanosomes of stages I and II in their melanophores.

When the number of premelanosomes was counted in 20 squares, each of which was 2.3 $\mu$×2.3 $\mu$ in size, one square contained 7~24, 16.6±1.0 on the average. The total 20 squares had 332 premelanosomes. As there were about 3.7 and 21.8


Fig. 15. Electron microphotograph of an abnormal melanophore in the dorsal skin of an albinic hybrid of the RA/TJ-type ($a^e a^t$). ×34000

PM, premelanosomes V, vacuoles

premelanosomes in the melanophores of the SN I and TJ stocks, respectively, the number, 16.6, was nearer to the TJ stock than the SN I, although it was intermediate between the two stocks. Of the 332 premelanosomes contained in the 20 squares, 33.2%, 39.3%, 19.2%, 6.6% and 1.7% were of stages I, II, III, IV and V, respectively (Fig. 5).

These findings indicate that albino genes $a^e$ and $a^t$ of the first group are codominant alleles. The $a^e a^t$ albinic hybrids produce premelanosomes which are intermediate in number and differentiation between the $a^e a^e$ and $a^t a^t$ albinos in their melanophores (Fig. 15).

v) TJ/FC-type ($a^t a^f$) albino

Mature male and female albinos (TJ/FC-type), produced in 1978 from a mating, TJ. Het. 75♀, No. 3 × FC. Het. 76♂, No. 3, were examined. These albinic hybrids are intermediate in coloration between the albinos of the TJ and FC stocks. While the xanthophores and iridophores are completely normal, melanophores show abnormality characteristic of albinos. When the number of premelanosomes was counted in 20 squares, each of which was 2.3 $\mu$ × 2.3 $\mu$ in size, one square contained 21 ~ 41, 30.7 ± 1.8 on the average. As there were about 21.8 and 33.0 premelanosomes in the albinos of the TJ and FC stocks, respectively, the number, 30.7, in the TJ/FC albinic hybrids was intermediate between those of the


Fig. 16. Electron microphotograph of an abnormal melanophore in the dorsal skin of an albinic hybrid of the TJ/Fc-type ($a^t a^f$). ×34000

G, GOLGI apparatus PM, premelanosomes V, vacuoles

two stocks. The total 20 squares had 614 premelanosomes, of which 11.4%, 23.4%, 4.5%, 25.9%, 3.0%, 25.9% and 6.0% were of stages I, II, III, IIIa, IV, IVa and V, respectively (Fig. 5). The premelanosomes of stages III and IIIa were in a ratio of 9: 52, while those of stages IV and IVa were in a ratio of 6: 52. The premelanosomes characteristic of the Fc albinos were overwhelmingly predominant in number as compared with those characteristic of the TJ albinos.

These findings evidently indicate that albino genes $a^t$ and $a^f$ of the first group are codominant alleles. The $a^t a^f$ albinic hybrids produce premelanosomes which are intermediate in number between the $a^t a^t$ and $a^f a^f$ albinos and a mixture of those characteristic of the albinos of these two stocks in melanization process (Fig. 16).

d. Chromatophores of the second group

In the albinos of the Go stock belonging to the second group, the xanthophores are especially well-developed and contain remarkably abundant carotenoid vesicles as compared with the albinos of the other stocks (Fig. 17a). This made the dorsal yellow color of mature albinos of the Go stock especially clear. Although the carotenoid vesicles are mostly gathered to form a mass surrounded with

pterinosomes in the xanthophores of normal brown-type frogs, they are somewhat evenly distributed together with pterinosomes in addition to forming one or more large masses in the xanthophores of the Go albinos. Some xanthophores extend under iridophores their thick processes which include masses of carotenoid vesicles. The iridophores of the albinos of this stock are completely normal.

The melanophores are extremely thin and almost consist of very slender dendritic processes. As it was difficult to set up a square of 2.3 $\mu \times 2.3$ $\mu$ in size, the number of premelanosomes was counted in 20 rectangles, each of which was 2.3 $\mu \times 1.15$ $\mu$ in size. The number of premelanosomes counted in such a rectangle was doubled in order to compare with that counted in a square which was 2.3 $\mu \times 2.3$ $\mu$ in size. The results showed that one square contained 2~26, $10.6 \pm 1.4$ on the average, and that the total 20 squares contained 212 premelanosomes. While this number of premelanosomes is larger than those counted in the Ex, Sn I, Sn II, Ym II and Br stocks of the first group, it was smaller than those in the Hr, Tj and Fc stocks of the same group.

Of the premelanosomes counted in the Go albinos, 27.0%, 50.5%, 12.5%, 6.5% and 3.5% were of stages I, II, III, IV and V, respectively (Fig. 5). Each premelanosome was $0.32 \pm 0.01$ $\mu$ and $0.21 \pm 0.01$ $\mu$ in major and minor axes, respectively (Fig. 17b).

e. Chromatophores of the third group

In the albinos of the Ym I stock belonging to the third group, the xanthophores and iridophores are completely normal and do not differ from those of wild-type frogs (Fig. 18a). While the melanophores are also similar to those of the wild-type frogs in external appearance of extending thick dendritic processes, they are abnormal in inner structure. Their cytoplasm is filled with premelanosomes of stages I~V. When the number of premelanosomes was counted in 20 squares, each of which was 2.3 $\mu \times 2.3$ $\mu$ in size, one square contained 22~56, $43.2 \pm 2.0$ on the average. The total 20 squares contained 863 premelanosomes, of which 17.5%, 24.0%, 24.0%, 20.5% and 14.0% were of stages I, II, III, IV and V, respectively (Fig. 5). Each premelanosomes was $0.37 \pm 0.01$ $\mu$ and $0.20 \pm 0.01$ $\mu$ in major and minor axes, respectively. Although some of the premelano-

Fig. 17. Electron microphotographs of dermal chromatophores in the dorsal skin of an albino of the Go stock (*bb*) belonging to the second group.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore
b. Parts of two abnormal melanophores. ×34000
PM, premelanosomes

Fig. 18. Electron microphotographs of dermal chromatophores in the dorsal skin of an albino of the Ym I stock (*cc*) belonging to the third group.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore
b. A part of an abnormal melanophore.
PM, premelanosomes V, vacuoles ×34000

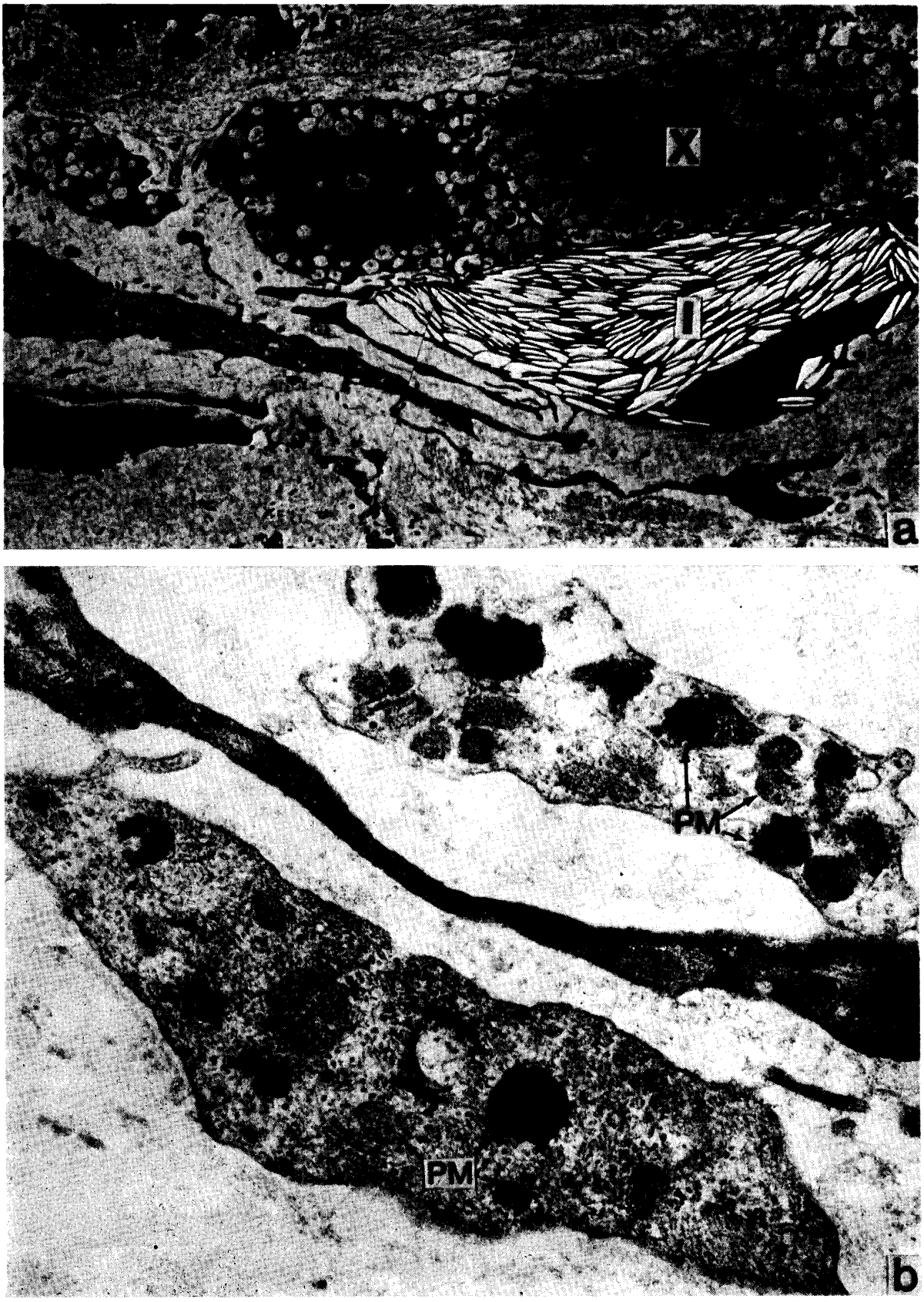


Fig. 17

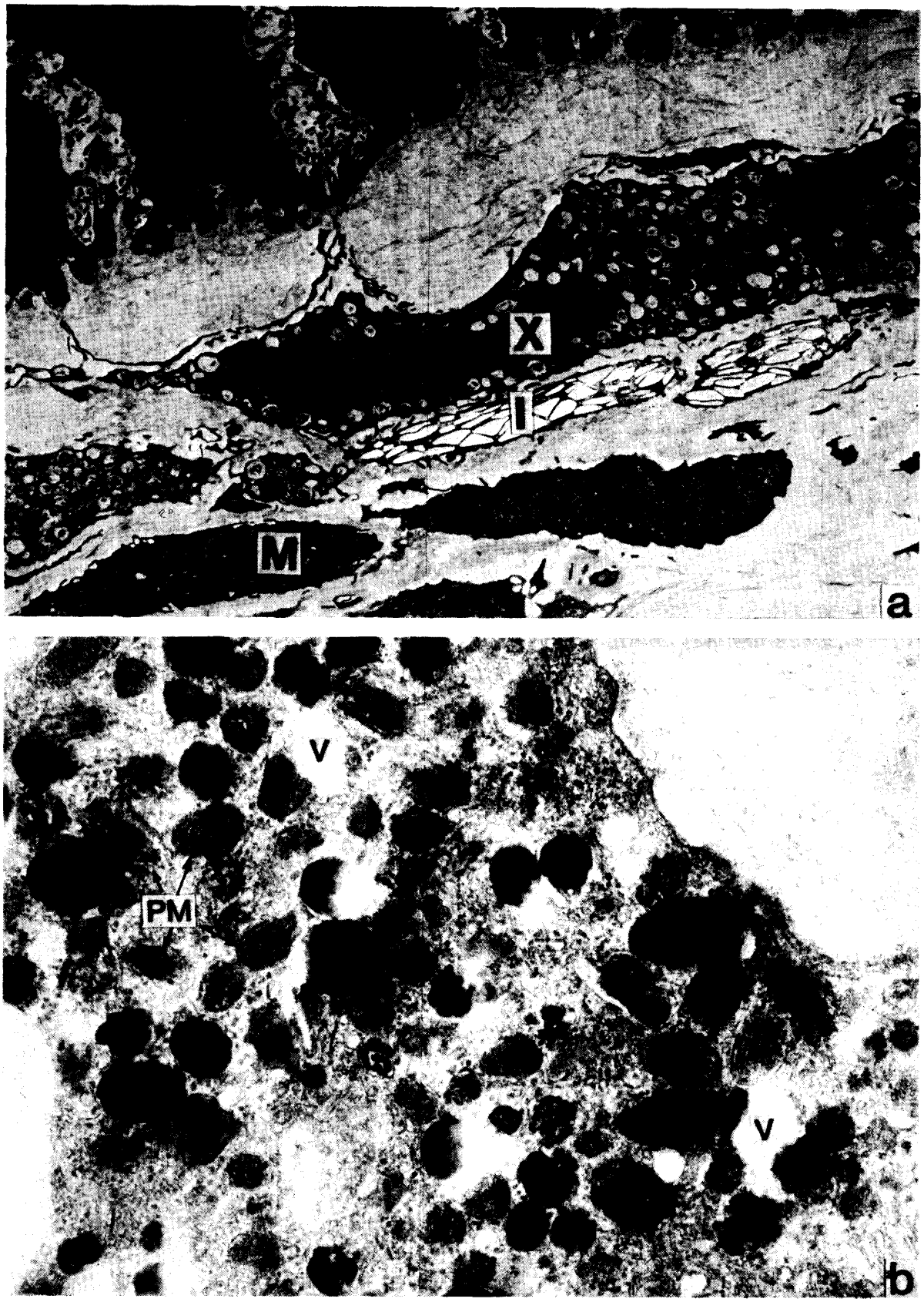


Fig. 18

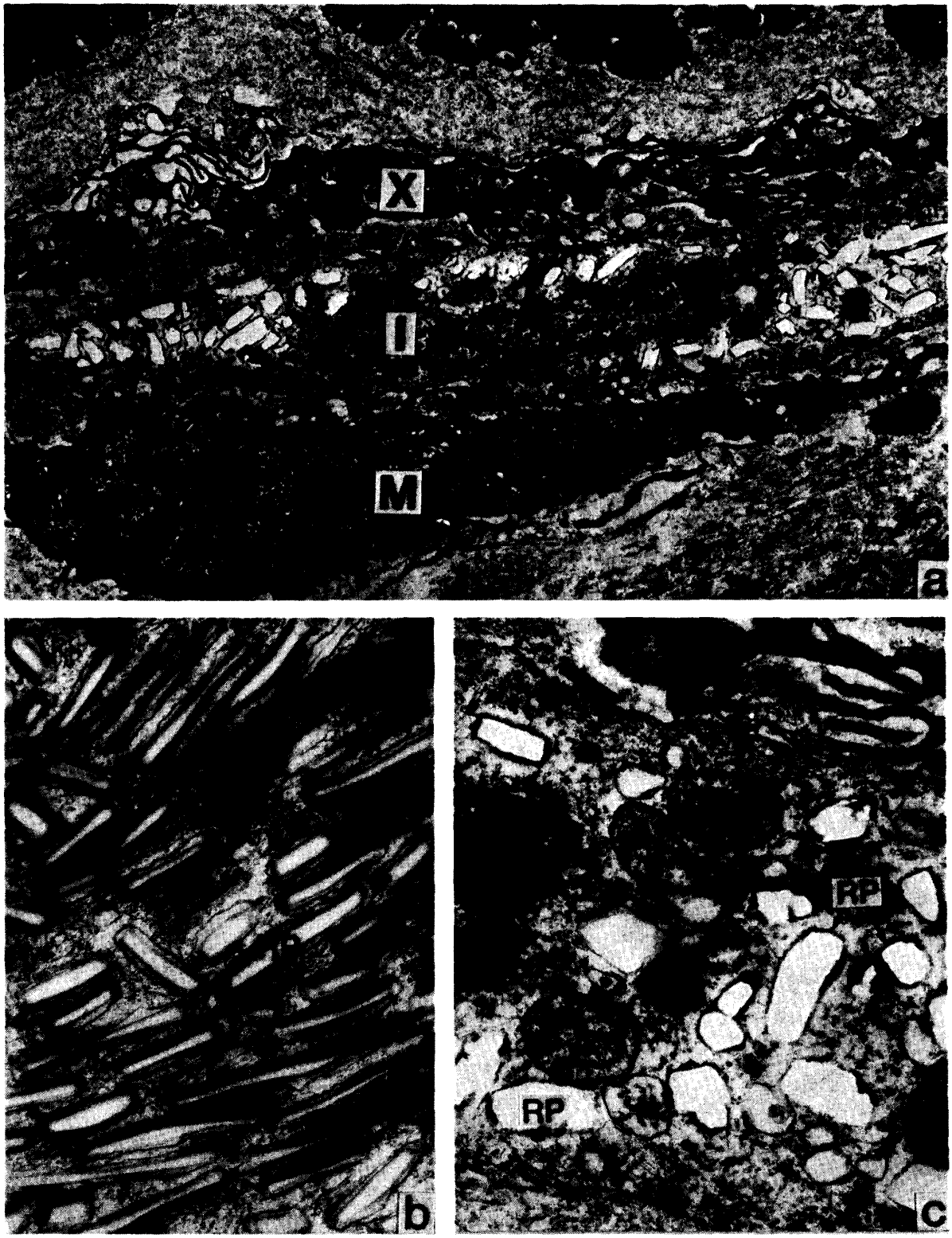


Fig. 19. Electron microphotographs of dermal chromatophores in the dorsal skin of an albino of the KM stock (*dd*) belonging to the fourth group.

a. Three kinds of chromatophores. ×6000
X, xanthophore I, iridophore M, melanophore
b. A part of a normal iridophore in the control frog. ×22000
c. A part of an abnormal iridophore. ×22000
RP, reflecting platelets MT, mitochondria

somes of stage V were near the completion of melanization, they were considerably smaller than the melanosomes of the wild-type frogs. However, the number of premelanosomes included in a square of 2.3 $\mu \times 2.3$ $\mu$ is larger than that of melanosomes in the wild-type frogs, as the latter is about 32.3. It is remarkable that the number of premelanosomes in the Ym I albinos is the largest among those of the 13 stocks of albinos (Fig. 18b).

f. Chromatophores of the fourth group

In the albinos of the Km stock belonging to the fourth group, the xanthophores are normal and similar to those of the brown wild-type frogs in structure. The carotenoid vesicles are almost gathered to form a mass surrounded with pterinosomes. In contrast to the xanthophores, the iridophores and melanophores are abnormal in the albinos of this stock (Fig. 19a).

Whereas the iridophores are nearly the same as those of brown wild-type frogs in width, being about 20 $\mu$, they are very thin, being about 3 $\mu$, less than one-third of those of the wild-type frogs in thickness. The reflecting platelets in each iridophore are remarkably fewer and smaller than those of the wild-type frogs and very irregular in shape. While the iridophores of the wild-type frogs are filled with slender reflecting platelets which are about 2 $\mu$ in length and 0.1 $\mu$ in thickness (Fig. 19b), those of the Km albinos are less than 1 $\mu$ in length and about 0.3 $\mu$ in thickness and sparsely distributed. Most of the reflecting platelets are of irregular shape and considered to be masses consisting of two or more of them. As the abnormal iridophores of the Km albinos do not reflect light, the skin of this albino stock is semitransparent (Fig. 19c).

The melanophores show an abnormality characteristic of this stock, although they have no accomplished melanosomes, as in the other albino stocks. When the number of premelanosomes was counted in 20 squares, each of which was 2.3 $\mu \times 2.3$ $\mu$ in size, one square contained $7 \sim 34$, $15.6 \pm 1.5$ on the average. The total 20 squares had 312 premelanosomes, of which 3.0%, 29.0%, 42.0%, 23.5% and 2.5% were of stages I, II, III, IV and V, respectively (Fig. 5). The premelanosomes were smaller than those of wild-type frogs. They were $0.40 \pm 0.01$ $\mu$ and $0.23 \pm 0.05$ $\mu$ in major and minor axes, respectively. In each

Fig. 20. Electron microphotographs of abnormal melanophores in the dorsal skin of an albino of the Km stock (*dd*) belonging to the fourth group.

a. An abnormal melanophore containing premelanosomes. ×34000
b. An abnormal melanophore containing masses of premelanosomes. ×34000
c. A mass of premelanosomes. ×64000
PM, premelanosomes V, vacuoles PMM, masses of premelanosomes

Fig. 21. Electron microphotographs of dermal chromatophores in the dorsal skin of an albino of the Ty stock (*ee*) belonging to the fifth group.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore
b. A part of an abnormal melanophore. ×34000
PM, premelanosomes V, vacuoles

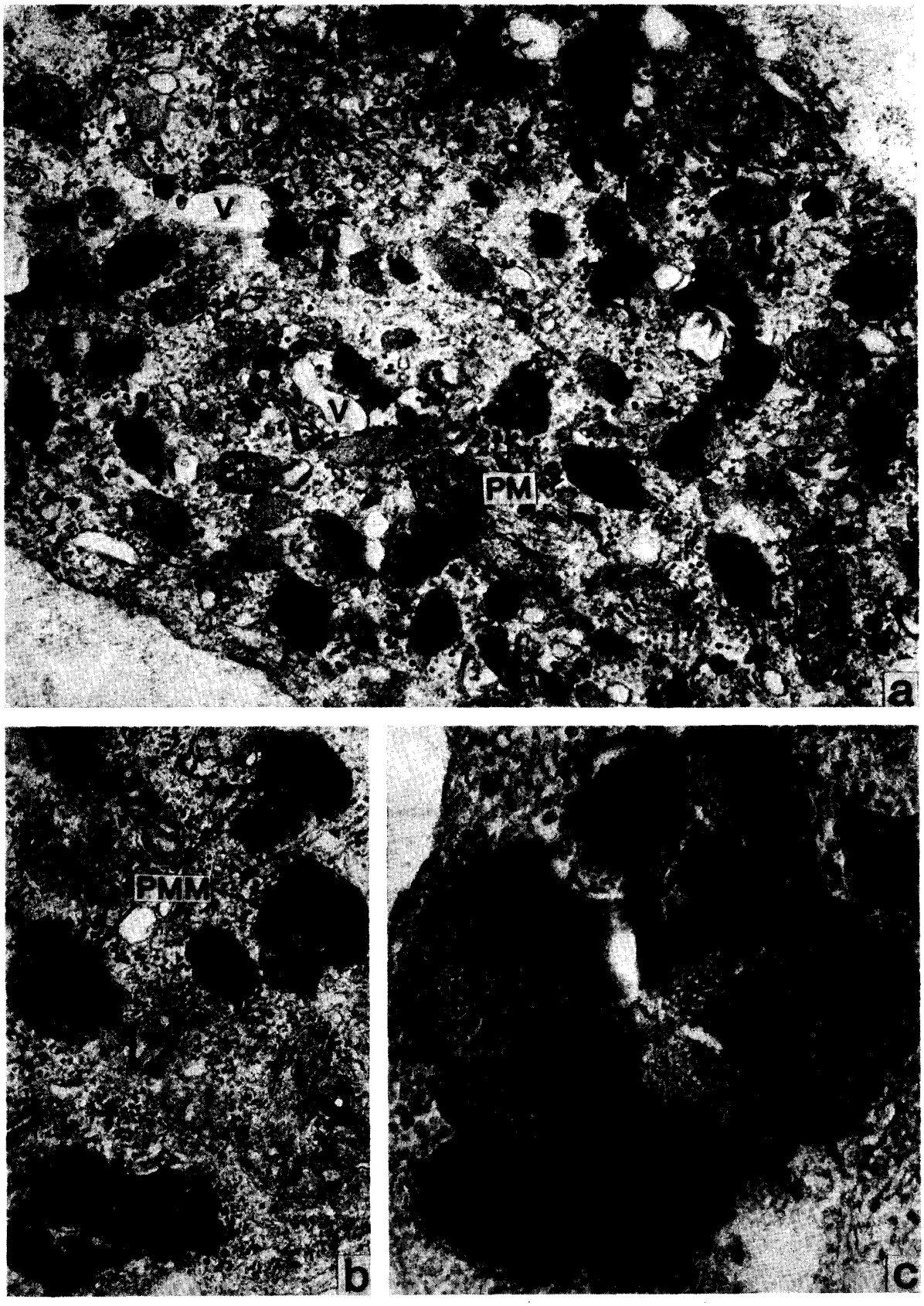


Fig. 20

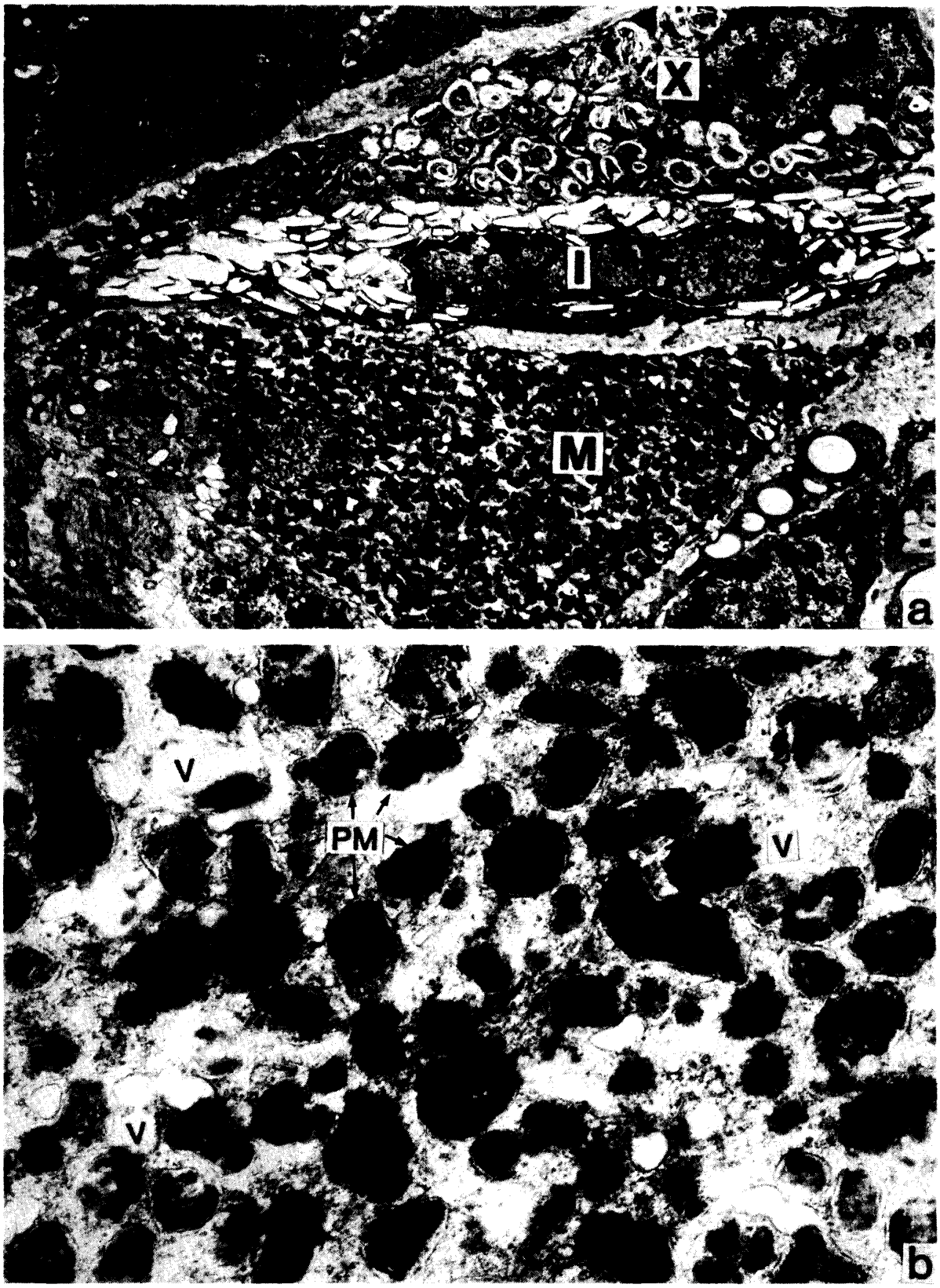


Fig. 21


Fig. 22. Electron microphotographs of dermal chromatophores in the dorsal skin of an albino of the Ns stock (*ee*) belonging to the fifth group.

a. Three kinds of chromatophores. ×4000
X, xanthophore I, iridophore M, melanophore
b. A part of an abnormal melanophore. ×34000
ME, melanosome PM, premelanosomes V, vacuoles

melanophore, some premelanosomes were gathered to form several masses. Such a mass consisted of two to 32 premelanosomes which were of a mixture of stages I~V. There were no melanophores not containing any masses of premelanosomes (Fig. 20). There were also no figures which showed degeneration of premelanosomes.

The origin of the masses of premelanosomes is not evident. These masses may have been produced by fusion of neighboring premelanosomes or by budding of an original premelanosome.

g. Chromatophores of the fifth group

Ty stock

In the albinos of this stock, the xanthophores and iridophores are completely normal. Although the melanophores are abnormal, they differ from the albinos of the other stocks in that they contain numerous premelanosomes which have advanced in melanization (Fig. 21a). Thus, an electron-microscopic photograph of them appears to be that of a wild-type frog at a glance.

When the number of premelanosomes and melanosomes was counted in 20 squares, each of which was 2.3 $\mu \times 2.3$ $\mu$ in size, one square contained 25~46, $36.7 \pm 1.2$ on the average. This number was larger than that in wild-type frogs. The total 20 squares had 734, of which 1.5%, 2.0%, 29.5%, 64.5% and 2.5% were stages II, III, IV, V and VI, respectively (Fig. 5). The premelanosomes and melanosomes were $0.36 \pm 0.01$ $\mu$ and $0.26 \pm 0.01$ $\mu$ in major and minor axes, respectively. They were somewhat smaller than the melanosomes in the wild-type frogs. There were some irregularly shaped premelanosomes among those of stage V. Such premelanosomes seemed to have been produced by fusion of two or more of them.

The melanophores of the Ty stock have many vacuoles of various sizes besides mitochondria in the cytoplasm (Fig. 21b).

Ns stock

While the xanthophores and iridophores are completely normal, the melanophores are abnormal and resemble those of the Ty stock (Fig. 22a). When the number of premelanosomes and melanosomes was counted in 20 squares, each of which was 2.3 $\mu \times 2.3$ $\mu$ in size, one square contained 25~41, $34.0 \pm 1.0$ on the average. The total 20 squares had 680, of which 0.5%, 2.0%, 2.5%, 36.0%, 57.0% and 2.0% were of stages I, II, III, IV, V and VI, respectively (Fig. 5). The premelanosomes and melanosomes were $0.39 \pm 0.01$ $\mu$ and $0.24 \pm 0.01$ $\mu$ in major and minor axes, respectively (Fig. 22b).

### *V. Dermal chromatophores of albinic hybrids between two different groups*

Only two of the various kinds of albinic hybrids between different groups were examined in terms of electron-microscopic structure of dermal chromatophores. One of them is two Km·Sn I albinos between the first and the fourth group, and the other is a Km·Ym I albino between the third and the fourth group.

### 1. Km·Sn I albinos (*a^e a^e dd*)

Dermal chromatophores in the dorsal skin were observed in two mature female Km·Sn I albinos (*a^e a^e dd*) which had been produced in 1977 from a mating between a female (No. 1) and a male (No. 1) obtained from Km. Alb. ♀ × Sn I. Alb. ♂ (Table 33). In these hybrid albinos, the irises are obscure and the pupils are bright red like those in the albinos of the Sn I stock. The dorsal surfaces are also similar to those of the Sn I albinos in coloration, although they appear to be slightly darker than the latter, owing to the semitransparent skin of the whole body. However, the dorsal surfaces of these Km·Sn I albinos are remarkably lighter than those of the albinos of the Km stock (Plate V, 36).

When the dermal chromatophores were observed under an electron microscope, it was found that the iridophores and melanophores are abnormal, while the xanthophores are completely normal. In the iridophores, the reflecting platelets are incomplete in formation; they are very few and small, and, moreover, some of them are irregularly shaped. These findings are similar to those of the albinos of the Km stock. While the area surrounding the nucleus is somewhat abundant in cytoplasm and includes endoplasmic reticula, Golgi apparatus and mitochondria, the other areas are scanty in cytoplasm.

The melanophores are also scanty in cytoplasm and their dendritic processes are generally slender like those of the albinos of the Sn I stock. The premelanosomes are very few, as those in the latter (Fig. 23a). When the number of premelanosomes was counted in 20 squares, each of which was 2.3 $\mu \times 2.3\ \mu$ in size, one square contained 0~11, $4.15 \pm 0.70$ on the average. The total 20 squares had 83 premelanosomes, of which 77% and 23% were of stages I and II, respectively (Fig. 5). Besides, the melanophores contain some masses of aggregated premelanosomes and granules which are about 0.3 $\mu$ in diameter and appear to be lysosomes including premelanosomes.

In the Km·Sn I albinos (*a^e a^e dd*), the abnormality of iridophores is due to albino gene *d* of the Km stock, while the abnormality of melanophores in size, number and melanization of premelanosomes is due to albino gene *a^e* of the Sn I stock. However, the aggregation of premelanosomes is evidently due to albino gene *d* of the Km stock (Fig. 23b).

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Fig. 23. Electron microphotographs of abnormal melanophores in the dorsal skin of an albinic hybrid of the Km·Sn I-type (*a^e a^e dd*). ×34000

a. An abnormal melanophore containing premelanosomes.

b. An abnormal melanophore containing a few premelanosomes and abundant lysosomes.

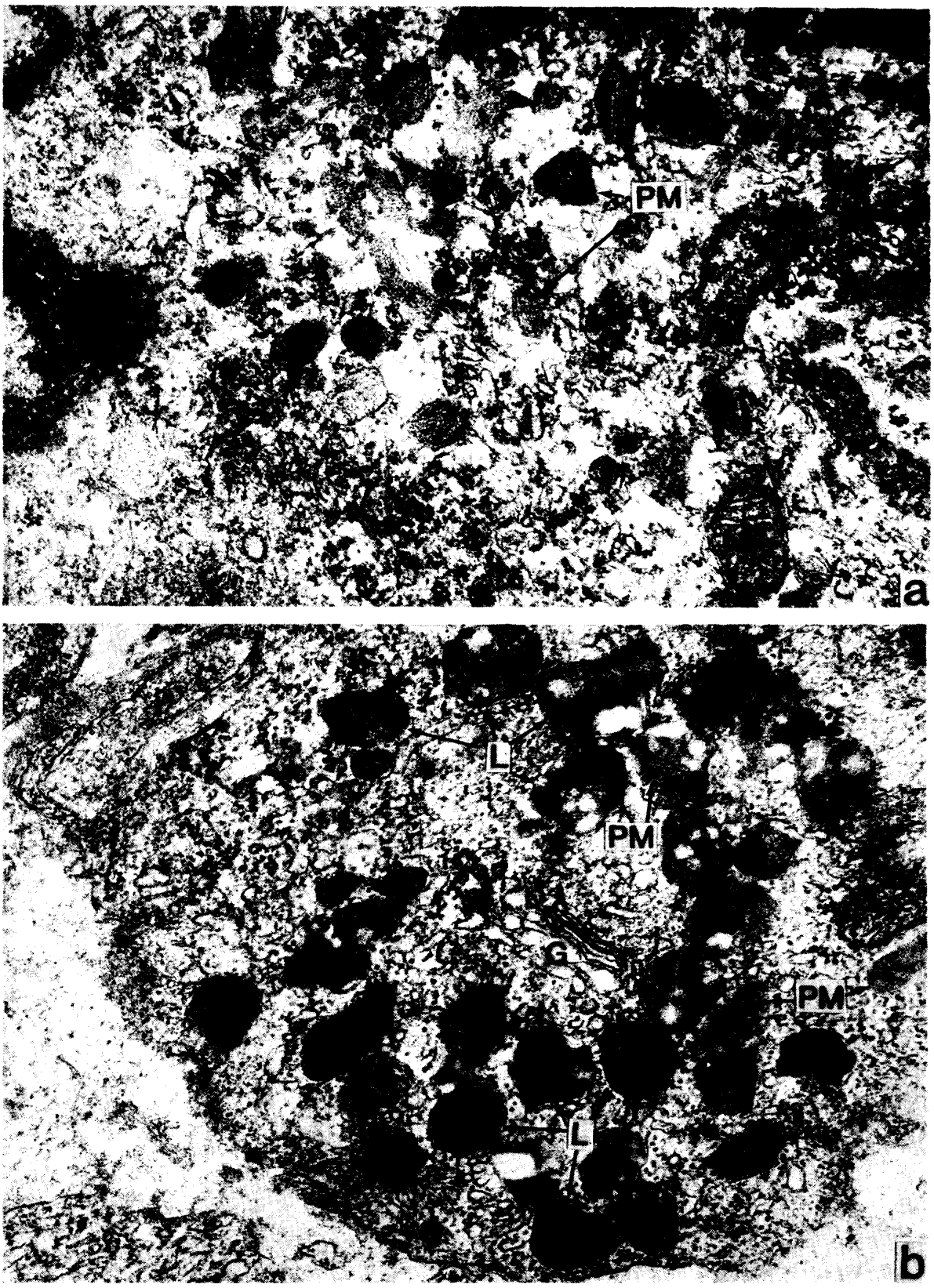
PM, premelanosomes MT, mitochondria G, Golgi apparatus L, lysosomes

Fig. 24. Electron microphotographs of abnormal melanophores in the dorsal skin of an albinic hybrid of the Km·Ym I-type (*ccdd*). ×34000

a. An abnormal melanophore containing abundant premelanosomes.

b. An abnormal melanophore containing masses of premelanosomes.

PMM, masses of premelanosomes




Fig, 23

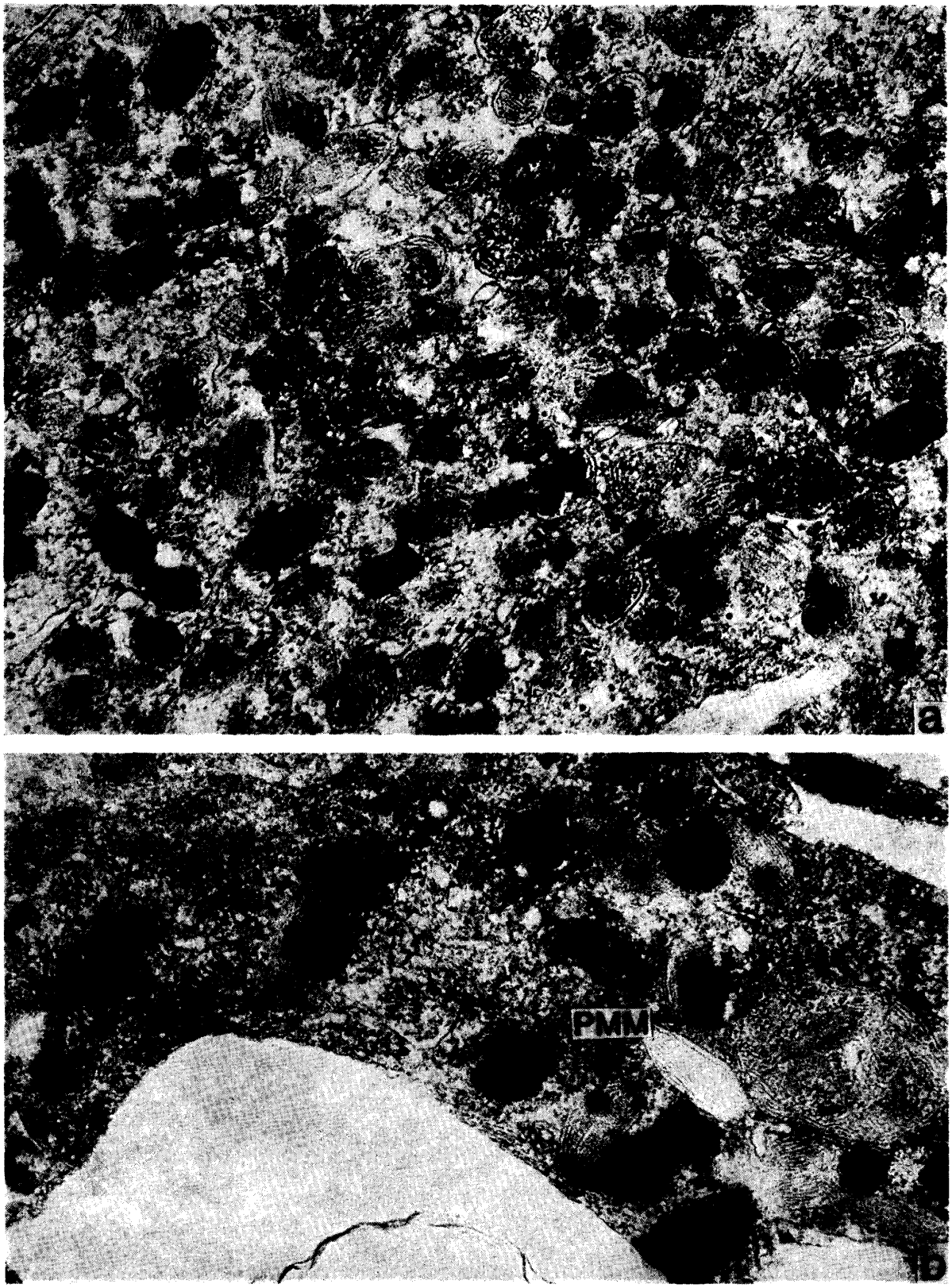



Fig. 24

### 2. Km·Ym I albinos (*ccdd*)

Dermal chromatophores in the dorsal skin were examined in a mature female Km·Ym I albino (*ccdd*) which had been produced in 1977 from a mating between a female (No. 1) and a male (No. 1) obtained from Km. Alb. ♀ × Ym I. Alb. ♂ (Table 37).

In this hybrid albino (Plate V, 37), the xanthophores are completely normal, while the iridophores and melanophores are abnormal. As found in the albinos of the Km stock, the formation of reflecting platelets is remarkably disturbed. In contrast, the melanophores resemble those of the albinos of the Ym I stock in distribution of premelanosomes (Fig. 24a). They have long dendritic processes which are considerably thick everywhere. When the number of premelanosomes was counted in 20 squares, each of which was 2.3 $\mu \times 2.3$ $\mu$ in size, one square contained $20 \sim 50$, $42.7 \pm 1.7$ on the average. This average number of premelanosomes in a square of 2.3 $\mu \times 2.3$ $\mu$ is distinctly larger than the 15.6 in the Km albinos, while it is similar to the 43.2 in the Ym I albinos. The total 20 squares had 853 premelanosomes, of which 17.7%, 30.5%, 27.3%, 19.1% and 5.5% were of stages I, II, III+IIIa, IV+IVa and V, respectively (Fig. 5).

Masses of aggregated premelanosomes are always found in all the melanophores, as in the Km·Sn I albinos. Moreover, the melanophores are divided into two kinds on the basis of included premelanosomes. In some melanophores, the advanced premelanosomes are mostly of stages III and IV, while those in the other melanophores are mostly of stages IIIa and IVa (Fig. 24b).

In the Km·Ym I albino (*ccdd*), it is found that the abnormality of iridophores is due to albino gene *d* derived from the Km stock, while the number and melanization of premelanosomes in the melanophores are predominatly due to albino gene *c*. However, the formation of premelanosome masses is considered to be due to albino gene *d* of the Km stock.

## *VI. Dopa reaction*

Of the 13 albino stocks, seven including the Sn I, Tj and Fc of the first group, the Go of the second group, the Ym I of the third group, the Km of the fourth group and the Ty of the fifth group, were used together with wild-type frogs to test dopa reaction in chromatophores of the dorsal skin of mature frogs. The cytochemical localization of tyrosinase activity by dopa reaction was observed under an optical and an electron microscope.

### 1. Wild-type

Dopa reaction in the dermal chromatophores of the dorsal skin of green wild-type frogs was observed under an electron microscope. As the melanophores were filled with completed melanosomes and Golgi apparatus was scarcely observable, the presence of dopa-reaction product was not determined. However, xanthophores became electron-dense as a whole, as the pterinosomes with a

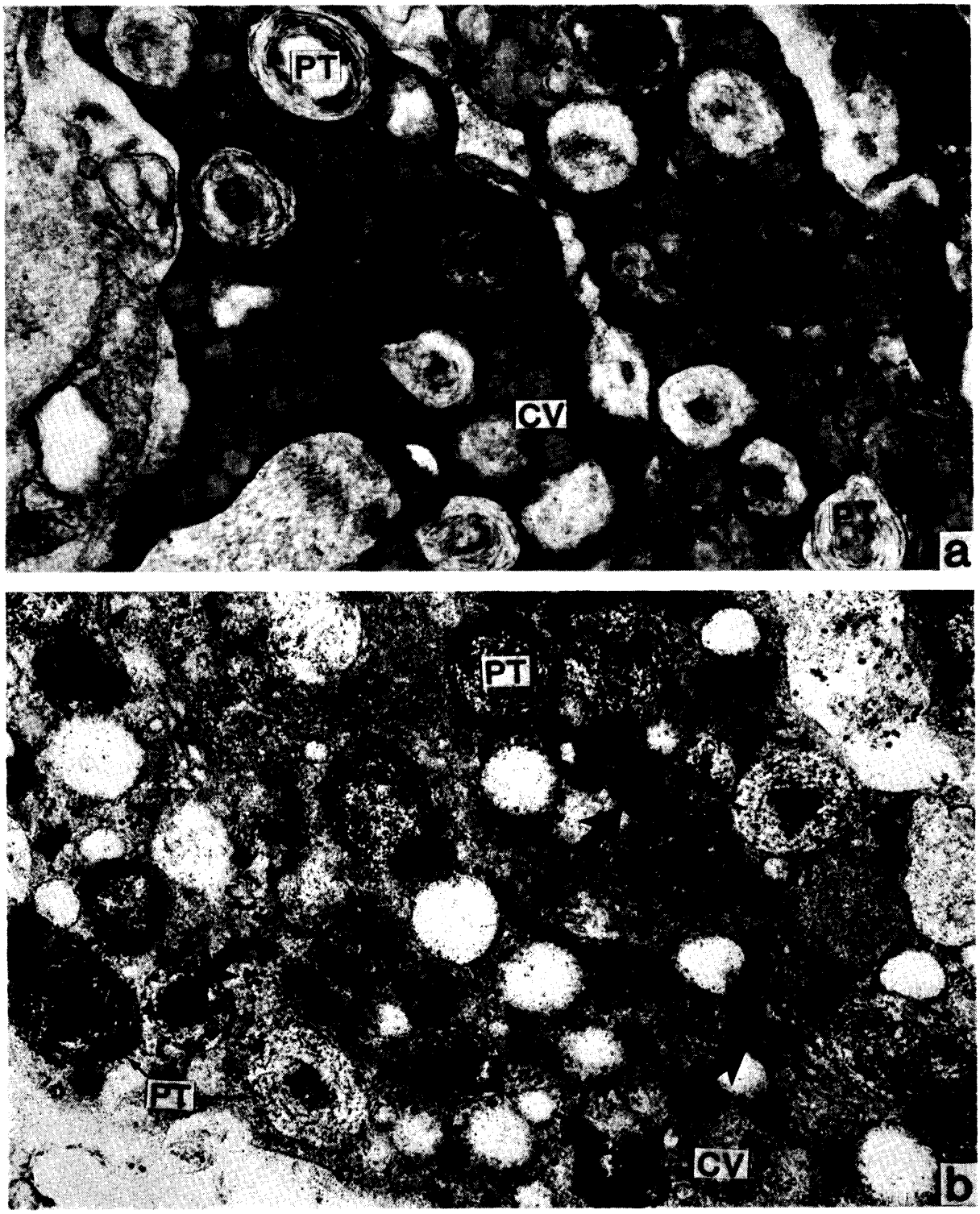



Fig. 25. Electron microphotographs of a dopa-treated xanthophore in the dorsal skin of a wild-type *Rana nigromaculata*. ×22000

a. Non-treated xanthophore.

b. Dopa-treated xanthophore.

PT, pterinosomes CV, carotenoid vesicles G, GOLGI apparatus

Arrows indicate dopa-reaction products.

lamella structure and minute glycogen granules were deposited with dopa-reaction products. Some pterinosomes were especially dopa positive and closely resembled melanosomes in electron density. Distinct dopa-reaction product was deposited in tubules adjacent to GOLGI apparatus (Fig. 25).

In iridophores, dopa-reaction product was deposited in the thin membrane of each reflecting platelet. Especially, the areas surrounding small reflecting platelets at the early stage of formation were remarkably electron-dense owing to deposition of abundant dopa-reaction product. In the neighborhood of GOLGI apparatus, no deposition of dopa-reaction product was found (Fig. 26).

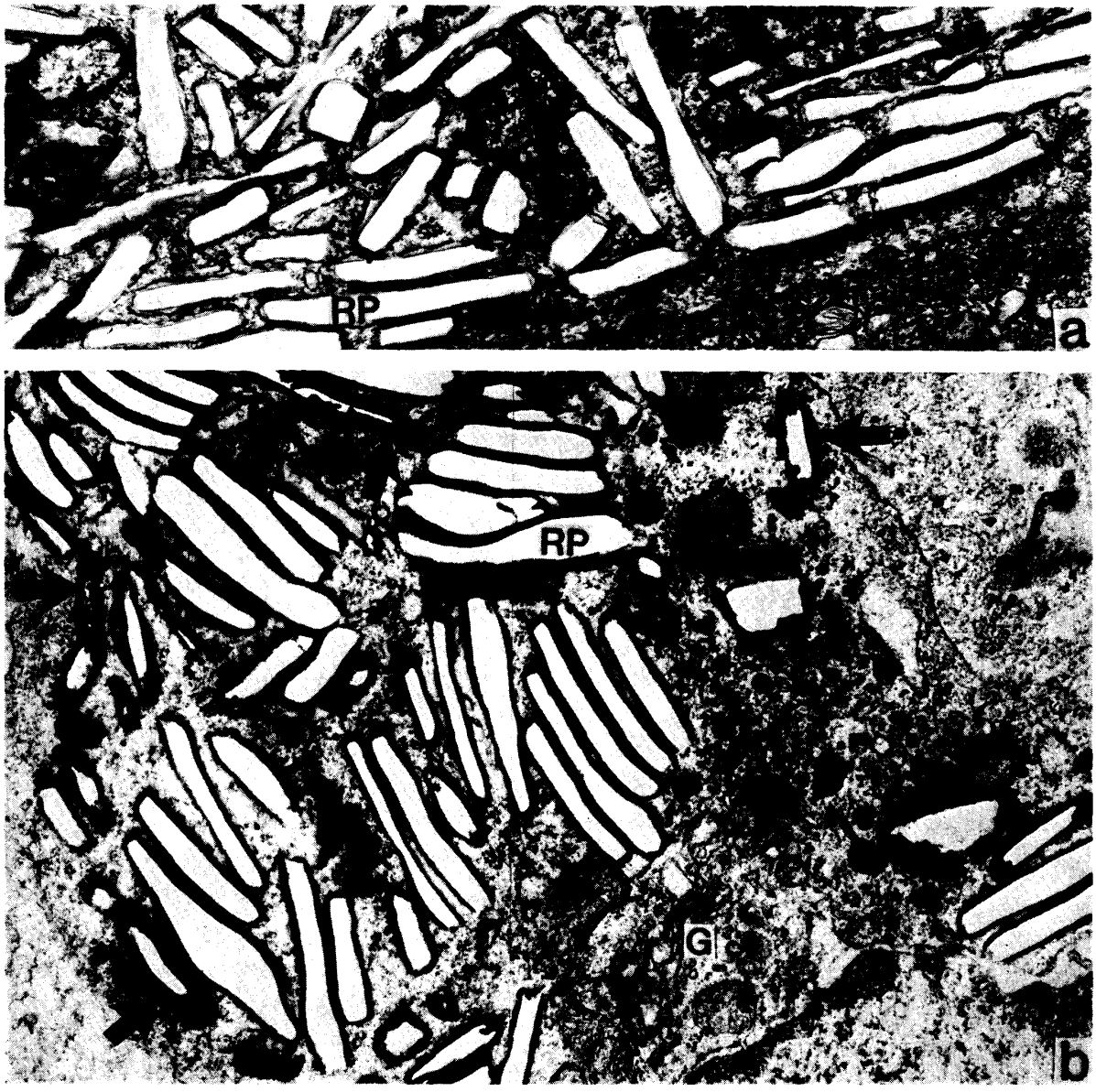



Fig. 26. Electron microphotographs of a dopa-treated iridophore in the dorsal skin of a wild-type *Rana nigromaculata*. ×22000

a. Non-treated iridophore.
b. Dopa-treated iridophore.
RP, reflecting platelets G, GOLGI apparatus Arrows indicate dopa-reaction products.

## 2. Albinos of the first group

### a. Sn I stock

When the dermal chromatophores of albinos of the Sn I stock are observed in an unstained frozen section by lighting with transmitted light, masses of carotenoid vesicles in xanthophores appear yellow, while iridophores appear dark gray as they do not transmit light. Melanophores are semitransparent and indistinguishable from connective tissue cells.

When the above section was incubated in dopa medium, no characteristic dopa-reaction occurred in the area where the dermal chromatophores were situated, although erythrocytes and some cells of mucous glands were deposited with dopa-reaction product. Even if a piece of skin incubated previously in dopa medium was sectioned on a freezing microtome and observed under an optical microscope, dermal chromatophores also did not show any dopa reaction, as in the above case. In this case, the iridophores became almost semitransparent owing to falling of reflecting platelets during the treatment.

Under an electron microscope, the melanophores incubated in dopa medium or in a mixture of dopa and PTU did not differ from non-treated ones. In the melanophores incubated in dopa medium, no dopa-reaction product was found anywhere in spite of existence of premelanosomes of stages I and II and of well-developed Golgi apparatus. Thus, it is considered that the albinos of the Sn I stock are completely lacking in tyrosinase activity (Fig. 27a).

### b. Tj stock

The dermal chromatophores in the dorsal skin of albinos of the Tj stock appear to be the same as those in the Sn I stock, when they are observed under an optical microscope, although they differ from the latter under an electron microscope. When incubated in dopa medium, the melanophores at the later differentiation stage were fairly deposited with dopa-reaction product. The dopa reaction occurred in premelanosomes of stages III and IV. The longitudinal fibers in the premelanosomes became obscure and evenly darkened by deposition of dopa-reaction product. This darkening differed from the regular melanization, where the minute granules constructing longitudinal fibers of each premelanosomes became thicker and gradually filled up the spaces between the fibers. No deposition of dopa-reaction product was found around the Golgi apparatus (Fig. 27b).

### c. Fc stock

The xanthophores and iridophores in the dorsal skin of albinos of the Fc stock did not show any dopa reaction, as found in those of wild-type frogs. In contrast, deposition of dopa-reaction product occurred in premelanosomes at the late differentiation stage. These darkened premelanosomes were somewhat similar in appearance to the completed melanosomes in the wild-type frogs, although they were smaller than the latter. They were the same in size as the premelanosomes of melanophores which were not incubated in dopa medium (Fig. 27c).

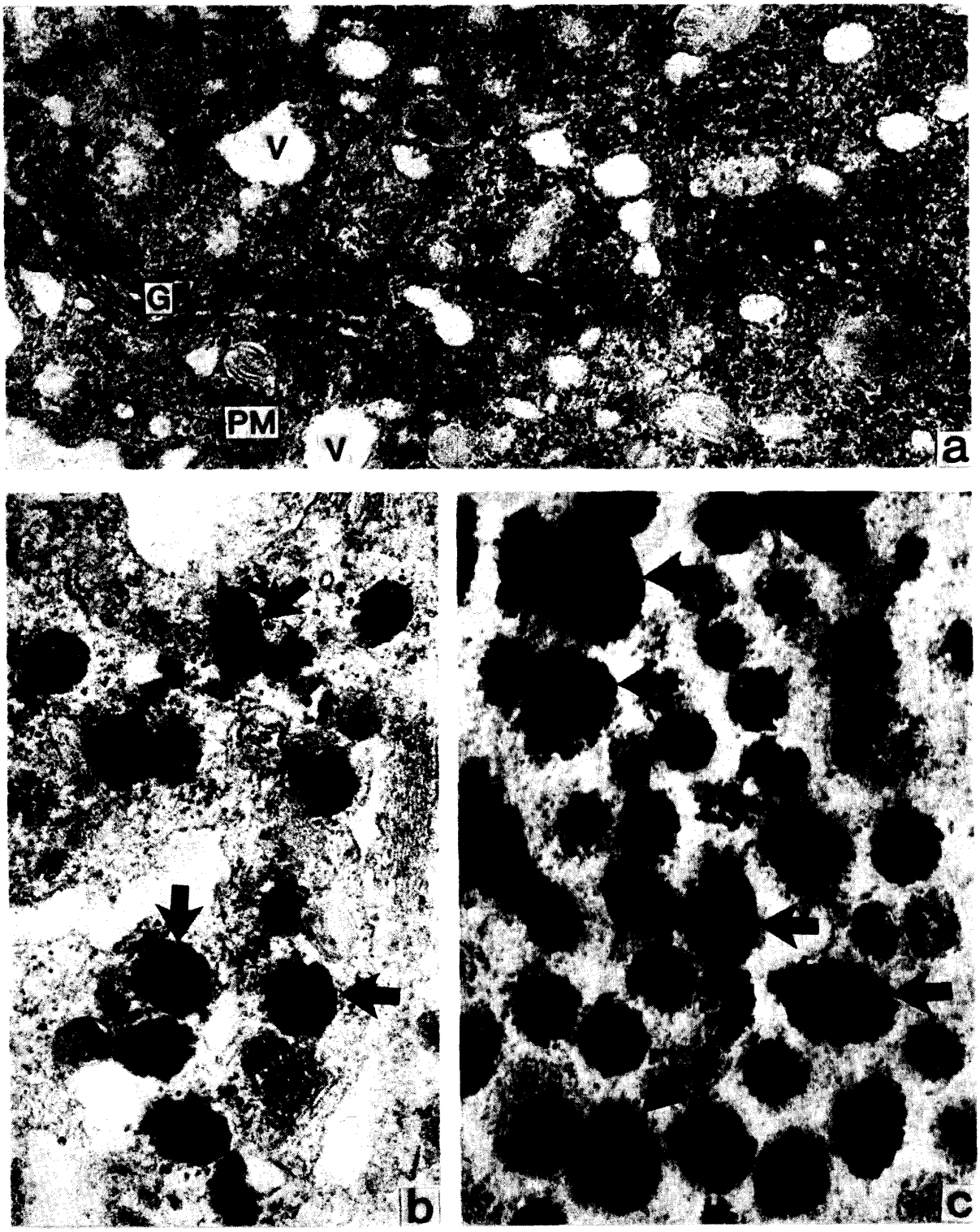



Fig. 27. Electron microphotographs of dopa-treated melanophores in the dorsal skins of albinos of the Sn I, Tj and Fc stocks belonging to the first group. ×34000

a. Sn I stock. b. Tj stock. c. Fc stock.
PM, premelanosomes G, Golgi apparatus V, vacuoles
Arrows indicate dopa-reaction products.

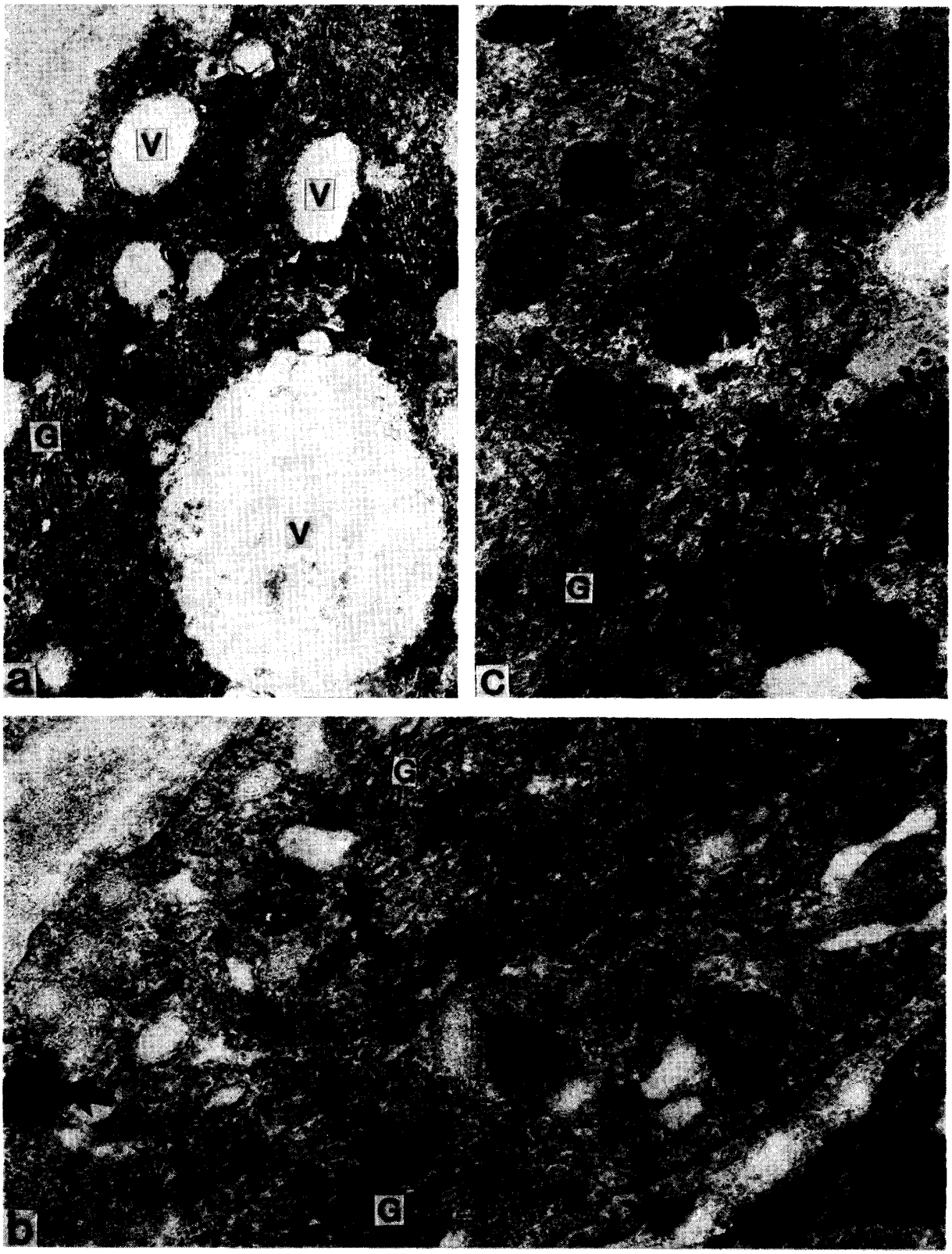



Fig. 28

### 3. Albinos of the second group

Dopa reaction was tested in albinos of the Go stock belonging to the second group. When a section of the dorsal skin was incubated in dopa medium, the melanophores became evidently dark and revealed their existence. However, in contrast to the cases of wild-type frogs as well as the YM I and TY stocks belonging to the third and fifth groups, respectively, the darkened parts in the skin of the Go stocks were usually isolated as a narrow spot and showed that the dopa reaction occurred in melanophores alone. Moreover, the darkening was not deep and appeared brown.

When the dorsal skin incubated in dopa medium was observed under an electron microscope, somewhat distinct deposition of dopa-reaction product was found in premelanosomes of stages III~V. This deposition usually occurred in one or more tubules situated in the neighborhood of GOLGI apparatus (Fig. 28a). Thus, the organization in producing and transporting tyrosinase to premelanosomes seems to have been established to some extent in the albinos of the Go stock.

### 4. Albinos of the third group

In an unstained section of the dorsal skin in albinos of the YM I stock belonging to the third group, melanophores can be recognized by existence of premelanosomes of stage V under an optical microscope. However, the outline of each melanophore is indistinct, owing to incomplete blackening and sparseness in distribution of the melanosomes.

When a section of the dorsal skin was incubated in dopa medium, it made a remarkable change and resembled that of the dorsal skin of a wild-type frog. In the range of melanophores lying between the basal lamina of the epidermis and the collagenous layer of the dermis, many darkened cells were continuously distributed. This finding shows that the albinos of the YM I stock have melanophores which are high in tyrosinase activity. On the other hand, a section of the dorsal skin incubated in a mixture of dopa and PTU was not deposited with dopa-reaction product.

When melanophores incubated in dopa medium were observed under an electron microscope, deposition of dopa-reaction product was distinctly found in the marginal area of GOLGI apparatus and in premelanosomes of stages III~V (stage 28b). Dopa reaction did not occur in melanophores incubated in a mixture of dopa and PTU. The production and activation of tyrosinase seem to be normal in the YM I albinos. It is also evident that there is no abnormality in the mechanism of producing premelanosomes, as the latter is rather more numerous

Fig. 28. Electron microphotographs of dopa-treated melanophores in the dorsal skins of albinos of the GO, YM I and TY stocks.

a. Go stock belonging to the second group. ×22000
b. YM I stock belonging to the third group. ×22000
c. TY stock belonging to the fifth group. ×34000

G, GOLGI apparatus V, vacuoles Arrows indicate dopa-reaction products.

than the melanosomes in the melanophores of wild-type frogs.

### 5. Albinos of the fourth group

When an unstained frozen section of the dorsal skin of albinos of the Km stock belonging to the fourth group is observed under an optical microscope, iridophores are difficult to be recognized owing to their faint color in contrast to the albinos of the other stocks. Melanophores are completely invisible. In a section incubated in dopa medium, deposition of dopa-reaction product was not detectable in chromatophores, as found in the Sn I and Tj stocks. However, erythrocytes and mucous gland cells were deposited with dopa-reaction product like those of albinos of all the other stocks.

When the dorsal skin of albinos of the Km stock were incubated in dopa medium, the premelanosomes of stages III~V in melanophores were distinctly

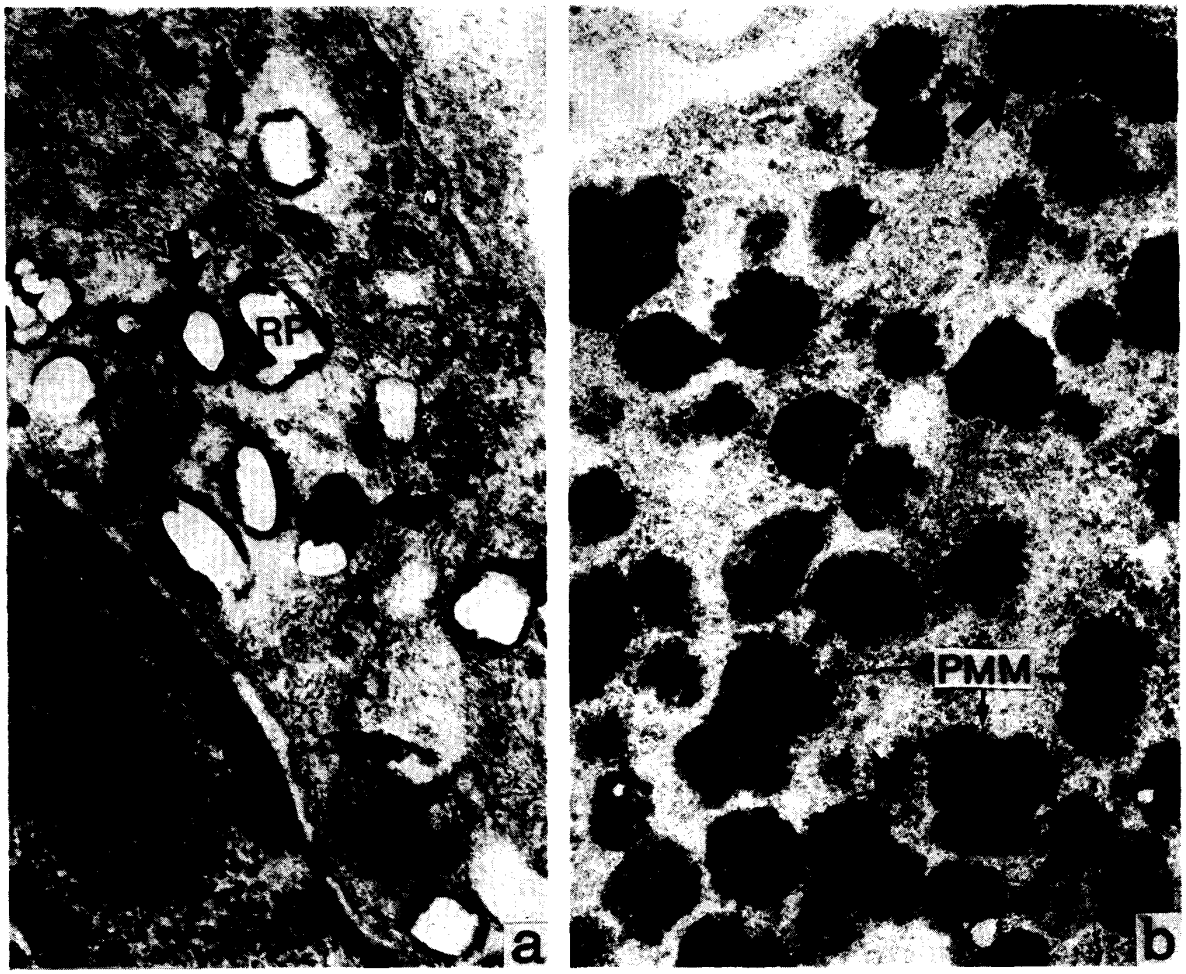



Fig. 29. Electron microphotographs of dopa-treated chromatophores in the dorsal skin of an albino of the Km stock belonging to the fourth group.

a. Iridophore. ×22000
b. Melanophore. ×34000
RP, reflecting platelets PMM, masses of premelanosomes
Thick arrows indicate dopa-reaction products.

deposited with dopa-reaction product. Dopa reaction was scarcely observable in the neighborhood of GOLGI apparatus, as it was very feeble. In aggregated masses of premelanosomes, those of stages III~V were also deposited with dopa-reaction product. However, the electron density of the dopa-reaction product in the KM stock was not so high as that in the YM I stock (Fig. 29a). On the other hand, the deposition of dopa-reaction product was found in the thin membrane surrounding each reflecting platelet (Fig. 29b).

The situation that the tyrosinase activity was not histochemically recognized after dopa reaction under an optical microscope is probably attributable to the fact that the premelanosomes were few in the melanophores of the albinos of the KM stock, as found in those of the Go stock.

### 6. Albinos of the fifth group

As the albinos of the TY and Ns stocks of the fifth group are very similar to each other, only the former was examined in terms of dopa reaction. The sections of the dorsal skin do not remarkably differ from those of wild-type frogs under an optical microscope. When the sections were incubated in dopa medium, they were also very similar to those of the wild-type frogs.

When melanophores incubated in dopa medium were examined under an electron microscope, the dopa reaction was comparatively difficult to be confirmed, as the melanophores were almost filled up with nearly completed melanosomes and the space of GOLGI apparatus was very narrow. However, in the area where premelanosomes were sparsely distributed, the deposition of dopa-reaction product was found in the neighborhood of GOLGI apparatus. This was also observed in premelanosomes of stages III~V (Fig. 28c). These premelanosomes were distinguished from completed melanosomes on the basis of their smaller size, although they fairly resembled melanosomes of stage VI in electron density.

## DISCUSSION

### 1. Kind of albinos

Albinism is the most popular among various mutations found in amphibians. When albinos appear in the field, they are easily discovered as they are conspicuous in color from the surroundings. At the tadpole stage, their discovery is especially easy, as a large number of them are usually produced at the same time. In this case, the situation that about one-fourth of a spawn is albinos indicates that the albinism is due to a recessive gene in the homozygous condition. The assumption that the albinism of amphibians is due to a single recessive gene has been made by many investigators, such as EALES (1933) and SMALLCOMBE (1949) in *Rana temporaria*, TOKUNAGA (1949) in parthenogenetically developed *Rana nigromaculata*, HUMPHREY (1967) and BENJAMIN (1970) in the axolotl, BROWDER (1972) and SMITH-GILL, RICHARDS and NACE (1970, 1972) in *Rana pipiens* and HOPERSKAYA (1975) and BLUEMINK and HOPERSKAYA (1975) in *Xenopus laevis*.

NISHIOKA (1977) has reported that albinos were produced together with eight other kinds of color variants by irradiating gametes with X-rays or neutrons in *Rana nigromaculata*, and that the albinos were due to a single recessive gene in the homozygous condition, like the gray-eyed and black-eyed variants.

The present authors (1977) have elucidated in *Hyla arborea japonica* that the inheritance of albinism is not so simple as generally believed up to that time. They performed crossing experiments among 10 albino stocks of this species collected from an area of about 6000 square kilometers surrounding Hiroshima. It was found that the 10 stocks are sorted into three groups, consisting of the first group with five stocks, the second group with two stocks and the third group with three stocks. These three groups differ from one another in the loci of their albino genes. The hybrids of albinos belonging to different groups are of the wild-type in color and pattern. The first, second and third groups of albinos slightly differ from one another in colors of the body and eyes. These different colors of albinos in the first, second and third groups are due to autosomal recessive genes, *f*, *s* and *t*, respectively. The dermal chromatophores of albinos have generally no completed melanosomes. They contain only colorless premelanosomes, except those of the colored albinos of one of the five stocks belonging to the first group. The colored albinos are due to a dominant gene (*C*) linked with the albino gene *f*. These albinos have a small number of incomplete melanosomes in addition to many premelanosomes in the dermal melanophores as well as in the pigment epithelial cells of the retina. There is another unusual stock among the five stocks of the first group. This stock consists of black-eyed albinos which are due to a dominant gene (*B*) linked with the albino gene *f*. These albinos are characteristic of the existence of completed melanosomes in the pigment epithelial cells of the retina. Of the four stocks other than the stock including colored albinos, three are very similar to one another in number and size of premelanosomes, while the remainder differs from the three in the scarcity and smallness of premelanosomes and thus seems to be controlled by a different allele of the albino gene *f*.

The two stocks of albinos belonging to the second group are similar to each other in number, size and minute structure of premelanosomes. In contrast, the three stocks of albinos belonging to the third group distinctly differ from one another in number, size, shape and minute structure of premelanosomes.

In the present study using the *Rana nigromaculata* group, 13 albino stocks including three obtained from irradiated gametes and 10 collected from the field were divided into five groups. The latter consist of the first group with eight stocks, the second group with one stock, the third group with one stock, the fourth group with one stock and the fifth group with two stocks. The albinos of each group are due to a single recessive gene in the homozygous condition and have a locus (*a*, *b*, *c*, *d* or *e*) differing from the loci of the other groups. The matings between albinos of two different groups produce wild-type individuals, while those of albinos belonging to the same group produce albinos only. The eight albino stocks of the first group are genetically divided into three strains, strain 1 with six stocks, strain

2 with one stock and strain 3 with one stock. These three albino strains of the first group are due to three multiple alleles, $a^e$, $a^t$ and $a^f$. It is very interesting that all the three albino stocks produced from irradiated gametes genetically belong to strain 1 of the first group, together with the other three albino stocks collected from the field. Although it is important to confirm if albinos belonging to the other groups can be produced by irradiating gametes, it seems evident to the present authors that the definite locus of the first group is apt to be changed from the wild-type gene into the albino by irradiation of the gamete.

Although the dorsal surfaces and the pupils of albinos are, generally speaking, light yellow and red, respectively, different groups or strains slightly differ from one another in color tone from light to dark, as found in those of *Hyla arborea japonica*. In contrast to the latter species, albino strains having a dominant gene, such as *B* and *C* linked with the albino gene, have not yet been found in the *Rana nigromaculata* group. The dermal chromatophores of all the albinos in the *Rana nigromaculata* group are abnormal and contain premelanosomes or immature melanosomes. The different groups or strains of albinos differ from one another in development, size, number and structure of premelanosomes as those of *Hyla arborea japonica*. However, in *Rana nigromaculata* there are two complicated albino strains, in which xanthophores or iridophores are abnormal in addition to abnormal melanophores. One is the single stock of the second group, in which the carotenoid vesicles contained in the xanthophores are exceptionally abundant. This makes the yellow dorsal color of mature albinos remarkably clear. The other complicated albino strain is the single stock of the fourth group, in which the reflecting platelets contained in the iridophores are remarkably few, small and very irregular in shape. Owing to the abnormality of iridophores, the skin of albinos of this stock is semitransparent.

It has not yet been determined whether the abnormality of xanthophores or iridophores in the albinos of the second and fourth groups is due to pleiotropy of the single recessive albino gene or to another mutant gene linked very closely with the albino gene. In contrast to the black-eyed and colored albinos in *Hyla arborea japonica*, in which a dominant gene (*B* or *C*) is linked with a recessive albino gene (*f*), no segregation of the imaginable recessive gene controlling the abnormality of xanthophores or iridophores from the recessive albino gene has been found up to the present, although many mating experiments have been performed by using frogs heterozygous for the albino gene.

## 2. Color of albinos

The five groups of albinos as well as the three strains of the first group considerably differ from one another in color tone of the dorsal surfaces and the pupils, although these are, generally speaking, yellow and red, respectively. The seven kinds of phenotypes in total are definitely arranged from light to dark as follows on the basis of differences in the color tones of the dorsal surfaces and pupils.

RA-type ($a^e a^e$)>GO-type ($bb$)>TJ-type ($a^t a^t$)>KM-type ($dd$)>
YM I-type ($cc$)>FC-type ($a^f a^f$)>TY-type ($ee$)

The color tones of albinos seem to be almost brought about by the premelanosomes contained in dermal melanophores. The RA-type albinos including six stocks have only premelanosomes of stages I and II in differentiation. These albinos are the lightest in color tones of the dorsal surfaces and the pupils. The albinos of the three stocks produced from irradiated gametes are characteristic of a comparatively small number of premelanosomes in the dermal melanophores, although the BR stock of *Rana brevipoda* collected from the field contains also a small number of premelanosomes.

All the albinos other than the RA-type ($a^e a^e$) have premelanosomes of stages III~V in their melanophores. As the color tones of these albinos are supposed to be brought about by the density of premelanosomes of stages III~V, the product of the total number of premelanosomes in a definite square by a percentage of premelanosomes of these stages was calculated (Table 39). The results show that the order in the density of premelanosomes of stages III~V agrees with that in the color tones of the dorsal surfaces and the pupils, except the KM-type (*dd*) albinos which is replaced with the TJ-type ($a^t a^t$). This replacement seems to be attributable to the fact that the skin of the KM-type albinos is semitransparent owing to abnormality of dermal iridophores.

TABLE 39

Number and developmental stage of melanosomes and premelanosomes in 20 squares, each of which is 2.3 $\mu \times 2.3$ $\mu$ in size

| Group | Strain | Stock | Phenotype | Genotype | Number (1) | Percentages of stages III~VI (2) | (1) × (2) |
|---|---|---|---|---|---|---|---|
| | | *R. brev.* | Wild | | 636 | 100 | 636 |
| | | *R. nigr.* | Wild | | 646 | 100 | 646 |
| I | 1 | EX | RA | $a^e a^e$ | 87 | 0 | 0 |
| | | SN I | | | 74 | 0 | 0 |
| | | SN II | | | 78 | 0 | 0 |
| | | YM II | | | 113 | 0 | 0 |
| | | HR | | | 531 | 0 | 0 |
| | | BR | | | 64 | 0 | 0 |
| | 2 | TJ | TJ | $a^t a^t$ | 436 | 52.5 | 228.9 |
| | 3 | FC | FC | $a^f a^f$ | 660 | 89.5 | 590.7 |
| II | | GO | GO | *bb* | 212 | 22.5 | 47.5 |
| III | | YM I | YM I | *cc* | 863 | 58.5 | 504.9 |
| IV | | KM | KM | *dd* | 312 | 68.0 | 212.2 |
| V | | TY | TY | *ee* | 734 | 98.5 | 723.0 |
| | | NS | | | 680 | 97.5 | 663.0 |

In the FC-type ($a^f a^f$) and TY-type (*ee*) albinos, the existence of abundant premelanosomes of stage IV or V in the dermal melanophores is probably more effective in darkening the dorsal skin, as compared with the condition in the other types of albinos. Especially, the TY-type albinos consisting of the TY and NS stocks contain a few completed melanosomes (stage VI) in dermal melanophores together with numerous premelanosomes of various stages, in contrast to the other types of albinos. There is no doubt that the existence of completed melanosomes

makes the dorsal skin of albinos darker, even if they are of a very small amount.

HOPERSKAYA (1975) has reported a mutant strain causing periodic albinism in *Xenopus laevis*. While the albinic females lay milk-white eggs, melanin granules appear in the pigment epithelium of the eyes and a little in skin melanophores of embryos. However, they almost disappear in the tadpole stage. BLUEMINK and HOPERSKAYA (1975) have confirmed that mature melanosomes as well as premelanosomes are lacking in the eggs of these periodic albinos.

In contrast to the milk-white eggs of albinos in *Xenopus laevis*, those of albinos in *Rana nigromaculata* are generally light yellow or beige in color of the animal halves and differ in color tone from light to dark with the groups and strains of albinos. When the eggs of the seven kinds of albinos with different phenotypes are arranged from light to dark in color tone of the animal halves, the order of the eggs is in accord with that of the mature albinos, except that the KM-type (*dd*) albinos are replaced with the FC-type ($a^f a^f$) albinos. The eggs of the TY-type (*ee*) albinos are the darkest and slightly lighter in color tone than those of wild-type females. Although the eggs of albinos belonging to the seven kinds of phenotypes have not yet been examined under an electron microscope, it is very probable that the color tones of the animal halves are brought by the density of premelanosomes and melanosomes contained there.

The albinic tadpoles of the seven kinds of phenotypes are generally light orange or light yellowish-orange at the late stage. In the RA-type ($a^e a^e$), TJ-type ($a^t a^t$) and YM I-type (*cc*) albinos, their tadpoles can be completely distinguished from the wild-type individuals at stage 20 immediately after hatching, as the black melanophores found in the eye areas of the wild-type tadpoles are lacking or very faint. Although the FC-type ($a^f a^f$), GO-type (*bb*), KM-type (*dd*) and TY-type (*ee*) albinos can scarcely be distinguished from the wild-type tadpoles immediately after hatching, as they have black melanophores, they become completely distinguishable soon or four to fourteen days after beginning to eat, as their melanophores lose color after hatching.

The tadpoles at the late stage of the seven kinds of phenotypes are arranged from light to dark in the color tone of the dorsal surfaces. The order of the RA-type ($a^e a^e$), GO-type (*bb*) and TJ-type ($a^t a^t$) tadpoles is the same as that of the mature albinos. The tadpoles of the KM-type (*dd*) albinos are the darkest, while those of the TY-type (*ee*) albinos which are the darkest at the mature frog stage are paler than those of the FC-type ($a^f a^f$) albinos. It is probable that the color tones of tadpoles are attributable to the density of premelanosomes and melanosomes in the melanophores of the dorsal skin.

It seems interesting that the order in color tone of the albinos of the different groups completely overlaps that of the albinos of the different strains belonging to the first group. This phenomenon seems to indicate that the differentiation and number of premelanosomes in dermal melanophores have no direct relation to the difference of loci in the five groups. At present it is unknown how the albinos having different loci differ from one another in inability to produce completed melanosomes. It is very probable that the different kinds of albinism in

the five albino groups is due to different barriers in the complicated biochemical process of melanin production, except that only the albinism of the RA-strain belonging to the first group seems to be largely attributable to deficiency in differentiation of premelanosomes.

Another interesting problem is production of a very few normal-colored eggs mingled with abundant ordinary light-colored ones by female TJ-type ($a^t a^t$), Fc-type ($a^f a^f$) and TJ/Fc-type ($a^t a^f$) albinos. Four (0.1%) of 3949 eggs obtained from three female $a^t a^t$ albinos and three (0.08%) of 3672 eggs obtained from two female $a^t a^f$ albinos in 1979 were normal-colored. Two (0.11%) of 1795 eggs obtained in 1980 from two female $a^f a^f$ albinos and four (0.11%) of 3611 eggs obtained in 1981 from three female $a^f a^f$ albinos were also normal-colored. These normal-colored eggs always developed into wild-type individuals, when they were fertilized with sperm of male albinos of the same group. Two of the four normal-colored eggs of the female $a^f a^f$ albinos developed normally by fertilization with sperm of a male $a^e a^e$ albino and became a female and a male wild-type frogs $(a^f)a^e$. This female laid only normal-colored eggs. When these female and male were mated with frogs homozygous or heterozygous for gene $a^f$, no $a^f a^f$ albinos were produced, while wild-type frogs $(a^f)a^f$ and $a^e a^f$ albinos were obtained.

These findings seem to show that reverse mutation of the albino allele to the wild-type allele occurred in a frequency of about 0.1% in the eggs of the above female $a^t a^t$, $a^f a^f$ and $a^t a^f$ albinos and that genes $a^t$ and $a^f$ are somewhat labile in nucleotide sequence in contrast to the other five genes, $a^e$, $b$, $c$, $d$ and $e$.

## SUMMARY

1. Genetic and morphologic studies were made on 13 albino stocks in the *Rana nigromaculata* group. Of these albino stocks, three, EX–145 stock (EX), SN–50 stock (SN I) and SN–130 stock (SN II), were induced by irradiation of gametes, while the remaining 10, *Rana brevipoda* stock (BR), Yamaguchi I stock (YM I), Kamogata stock (KM), Gion stock (GO), Tojo stock (TJ), Fuchu stock (FC), Toyomatsu stock (TY), Nishinomiya stock (NS), Yamaguchi II stock (YM II) and Hiro stock (HR), were collected from the field. In order to obtain albinic offspring in each of the 13 albino stocks, matings were made between female and male albinos, between albinos and heterozygous individuals or between heterozygous females and males. Albinos were also produced from heterozygous individuals by the method of diploid gynogenesis.

2. From the results of the mating experiments and diploid gynogenesis, it was evident that the albinos of each stock are mutants due to a single recessive gene in the homozygous condition.

3. Females of each of the 13 albino stocks were mated with males of the other 12 stocks. The results of these mating experiments showed that the 13 albino stocks were divided into five groups. The first group consists of eight albino stocks, EX, SN I, SN II, YM II, HR, BR, TJ and FC, while the second, third and

fourth groups include the Go, Ym I and Km stocks, respectively. The fifth group consists of the Ty and Ns stocks.

While matings between different albino stocks of the same group produce albinos only, those between albinos of different groups produce wild-type individuals alone. Thus, it is evident that the albinos of each group have a locus (*a*, *b*, *c*, *d* or *e*) differing from the loci of the other groups. The hybrids among albinos of the five different groups are *AaBb*, *AaCc*, *AaDd*, *AaEe*, *BbCc*, *BbDd*, *BbEe*, *CcDd*, *CcEe* or *DdEe* in genotype.

4. The eight albino stocks of the first group are genetically divided into three strains. Strain 1 consists of six albino stocks, Ex, Sn I, Sn II, Ym II, Hr and Br, while strains 2 and 3 include the Tj and Fc stocks, respectively. These three albino strains are due to three multiple alleles, $a^e$, $a^t$ and $a^f$. The albinos of strains 1, 2 and 3 are $a^ea^e$, $a^ta^t$ and $a^fa^f$ in genotype, respectively. The phenotypes of the albinos of strains 1, 2 and 3 are named Ra-type, Tj-type and Fc-type, respectively. The hybrids among albinos of the three strains are albinos which are $a^ea^t$, $a^ea^f$ and $a^ta^f$ in genotype and intermediate between the parental albinos in phenotype. These phenotypes are named Ra/Tj-type, Ra/Fc-type or Tj/Fc-type.

5. Voluminous mating experiments were performed to clarify the differences between the first and second groups, the first and third groups, the first and fourth groups, the first and fifth groups, the second and third groups, the second and fourth groups, the third and fourth groups, and the fourth and fifth groups. Besides, gynogenetic haploids and diploids were produced from eggs of female hybrids between the second and fourth groups, the third and fourth groups, and the fourth and fifth groups. The results of these experiments indicated that genes *a* and *b*, *a* and *c*, *a* and *d*, *a* and *e*, *b* and *c*, *b* and *d*, *c* and *d*, and *d* and *e* are located on non-homologous chromosomes or on such positions of homologous chromosomes as crossing-over occurs in a frequency of nearly 50%.

6. The dorsal surfaces of mature male *Rana nigromaculata* are grayish yellow or grayish yellow-green and usually have no dark spots, while those of mature females are grayish white or brownish gray in ground color and have many brownish-black spots. Both males and females have a pale stripe on the median line and two pale stripes on the dorso-lateral folds. Juvenile *Rana nigromaculata* do not reveal sexual dimorphism. Their dorsal colors are roughly divided into three types, green, brown and a mixture of green and brown. On the dorsal surfaces of juvenile frogs, there are many small black spots. The dorsal colors of mature and juvenile *Rana brevipoda* are also divided into three types, green, brown and a mixture of green and brown.

7. Although the dorsal surfaces and the pupils of albinos are, generally speaking, light yellow and red, respectively, these colors in the seven types of albinos including the five groups and the three strains of the first group are arranged as follows from light to dark.

Ra-type ($a^ea^e$) > Go-type ($bb$) > Tj-type ($a^ta^t$) > Km-type ($dd$) >
Ym I-type ($cc$) > Fc-type ($a^fa^f$) > Ty-type ($ee$)

While the eggs of albinos are light yellow or beige in color of the animal half, those of the above seven types of albinos are arranged as follows from light to dark.

RA-type > GO-type > TJ-type ≥ FC-type > YM I-type > KM-type > TY-type

The body colors of albinos at the late tadpole stage (stage X) in the seven types are arranged as follows from light to dark.

RA-type > GO-type > TJ-type > YM I-type > TY-type > FC-type > KM-type

8. About 0.1% of the eggs laid by some of the female TJ-type ($a^t a^t$), FC-type ($a^f a^f$) and TJ/FC-type ($a^t a^f$) albinos are normal-colored. These normal-colored eggs become wild-type frogs by fertilization with sperm of male albinos belonging to the same group. When wild-type female and male frogs obtained from matings between female $a^f a^f$ albinos and a male $a^e a^e$ albino were mated with frogs homozygous or heterozygous for gene $a^f$, no $a^f a^f$ albinos were produced. These findings seem to show that reverse mutation of the albino allele to the wild-type allele occurs in a frequency of about 0.1% in the eggs of the above female albinos.

9. The dermal chromatophores in the dorsal skins of albinos belonging to the 13 stocks were examined under an electron microscope. The melanophores of these albinos are abnormal and contain premelanosomes or immature melanosomes, although those of the TY-type albinos contain a very few completed melanosomes in addition. The albinos of different groups or strains differ from one another in development, size, number and structure of premelanosomes.

10. The xanthophores in various kinds of albinos do not differ from those of wild-type frogs except that those of the GO-type albinos of the second group contain very abundant carotenoid vesicles.

The iridophores in various kinds of albinos are completely normal except that those of the KM-type albinos of the fourth group are incomplete in formation of reflecting platelets.

11. Albinic hybrids, SN I/TJ ($a^e a^t$) and TJ/FC ($a^t a^f$), in the first group are intermediate between the albinos of the parental stocks in coloration of the body and electron-microscopic structure of dermal chromatophores. Albino genes $a^t$ and $a^e$ or $a^f$ of the first group are codominant alleles.

12. Albinic hybrids, KM·SN I ($a^e a^e dd$) and KM·YM I ($ccdd$), between albinos of different groups are similar to the KM stock in abnormality of iridophores, owing to the existence of albino gene $d$ of the KM stock. The number and melanization of premelanosomes in melanophores are almost due to albino gene $a^e$ of the SN I stock or $c$ of the YM I stock, while the formation of premelanosome masses is due to albino gene $d$ of the KM stock.

13. The cytochemical localization of tyrosinase activity by dopa reaction was observed in albinos of seven stocks including the SN I, TJ and FC of the first group, the GO of the second group, the YM I of the third group, the KM of the fourth group and the TY of the fifth group. While dopa reaction is completely lacking in the dermal melanophores of the dorsal skin taken from albinos of the SN I stock of the first group, it occurs in albinos of all the other stocks. The deposition of

dopa-reaction product is distinctly found in premelanosomes at the late differentiation stage when the premelanosomes are easily distinguished from the cytoplasmic matrix owing to their high electron-density.

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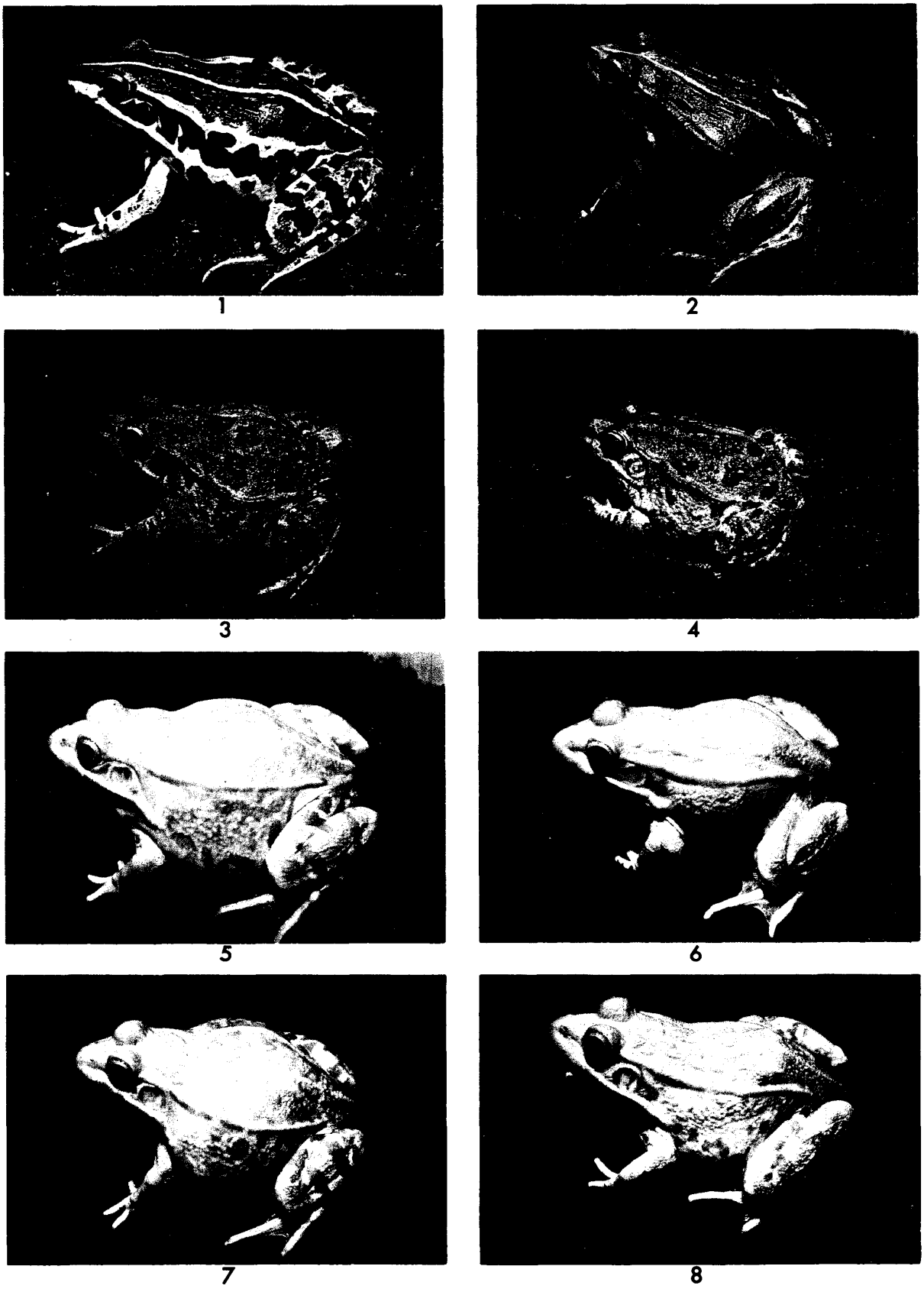
# EXPLANATION OF PLATES

## PLATE I

Albinos of two stocks belonging to the first group and wild-type frogs in the *Rana nigromaculata* group. ×0.6

1. Wild-type *Rana nigromaculata* (♀).
2. Wild-type *Rana nigromaculata* (♂).
3. Wild-type *Rana brevipoda* (♀).
4. Wild-type *Rana brevipoda* (♂).
5. Albino (♀) of the Ex stock ($a^e a^e$) in *Rana nigromaculata*.
6. Albino (♂) of the Ex stock ($a^e a^e$) in *Rana nigromaculata*.
7. Albino (♀) of the Sn I stock ($a^e a^e$) in *Rana nigromaculata*.
8. Albino (♂) of the Sn I stock ($a^e a^e$) in *Rana nigromaculata*.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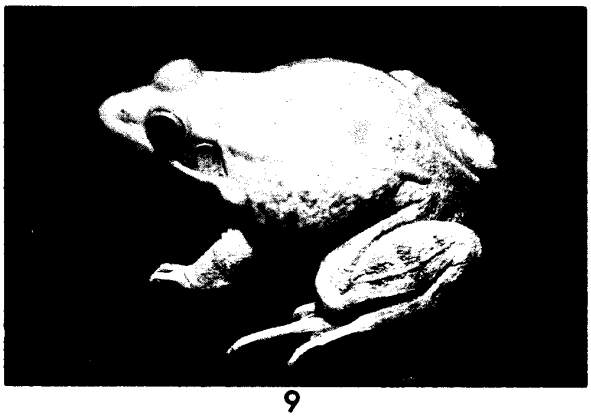
## PLATE II

Albinos of four stocks belonging to the first group in the *Rana nigromaculata* group.
×0.6

9. Albino (♀) of the SN II stock ($a^e a^e$) in *Rana nigromaculata*.
10. Albino (♂) of the SN II stock ($a^e a^e$) in *Rana nigromaculata*.
11. Albino (♀) of the YM II stock ($a^e a^e$) in *Rana nigromaculata*.
12. Albino (♂) of the YM II stock ($a^e a^e$) in *Rana nigromaculata*.
13. Albino (♀) of the BR stock ($a^e a^e$) in *Rana brevipoda*.
14. Albino (♂) of the BR stock ($a^e a^e$) in *Rana brevipoda*.
15. Albino (♀) of the HR stock ($a^e a^e$) in *Rana nigromaculata*.
16. Albino (♂) of the HR stock ($a^e a^e$) in *Rana nigromaculata*.



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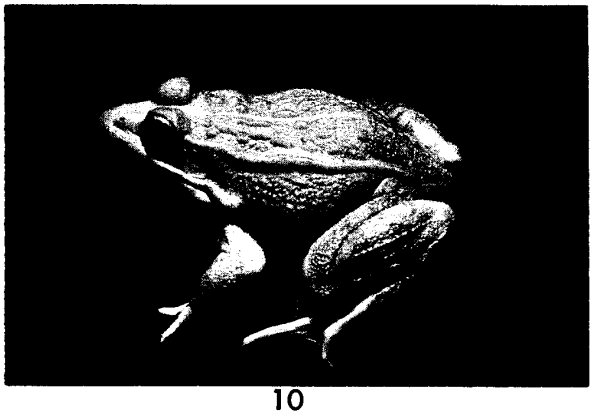

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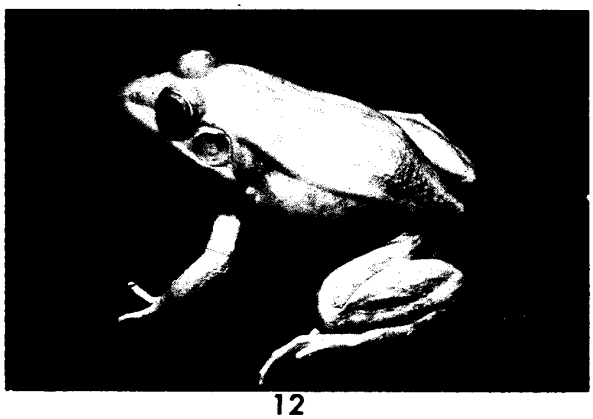

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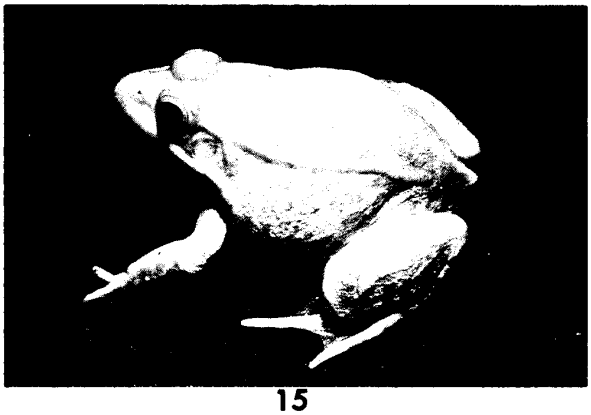

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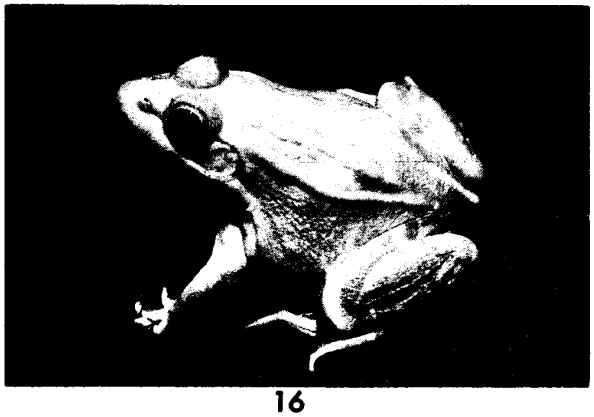

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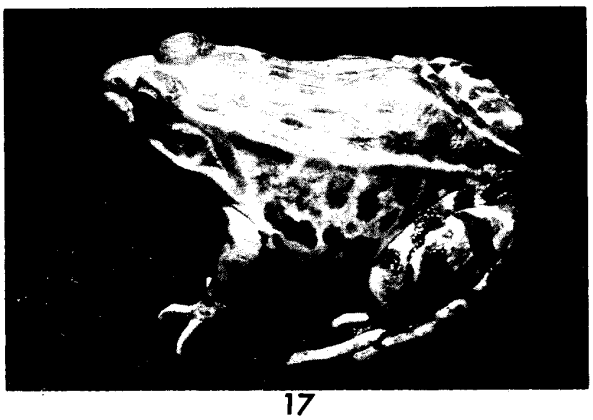
## PLATE III

Albinos of three stocks belonging tǫ the first group and their hybrids in *Rana nigromaculata*.

17. Albino (♀) of the TJ stock ($a^t a^t$). ×0.6
18. Albino (♂) of the TJ stock ($a^t a^t$). ×0.6
19. Albino (♀) of the FC stock ($a^f a^f$). ×0.6
20. Albino (♂) of the FC stock ($a^f a^f$). ×0.6
21. Albinic hybrid (♀) of the TJ/SN I ($a^e a^t$). ×0.6
22. Albinic hybrid (♀) of the FC/SN II ($a^e a^f$). ×0.6
23. Albinic hybrid (♀) of the TJ/FC ($a^t a^f$). ×0.6
24. Albinic tadpoles of the SN II stock (a), the SN II/FC (b) and the FC stock (c), and wild-type tadpoles (d). ×1.0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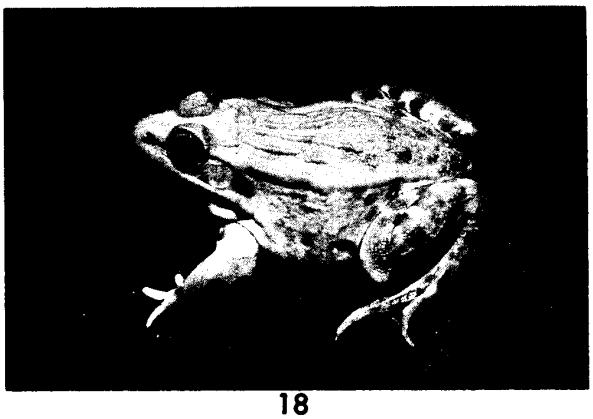
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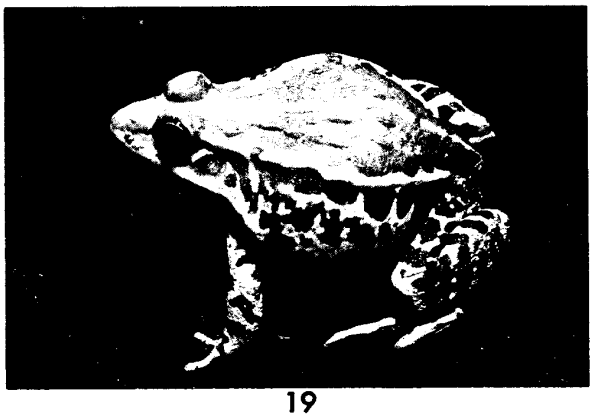
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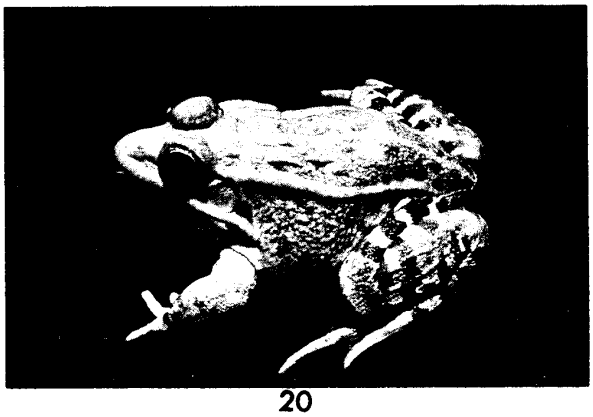
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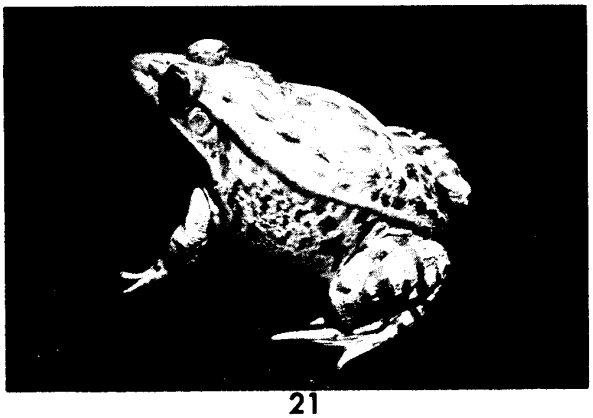
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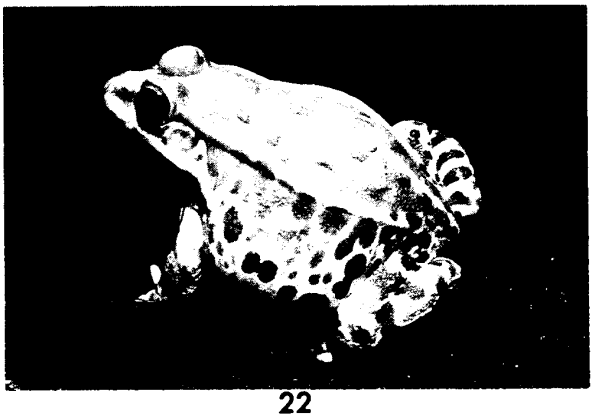
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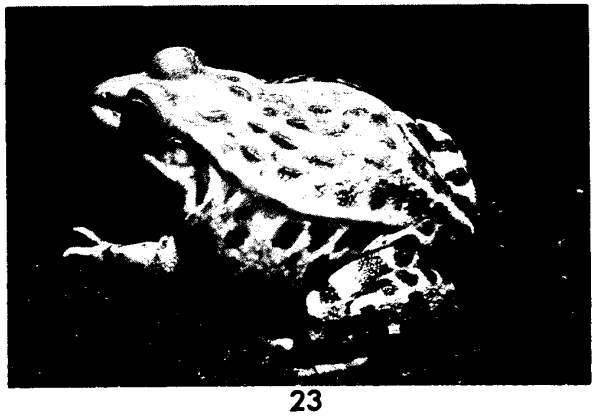
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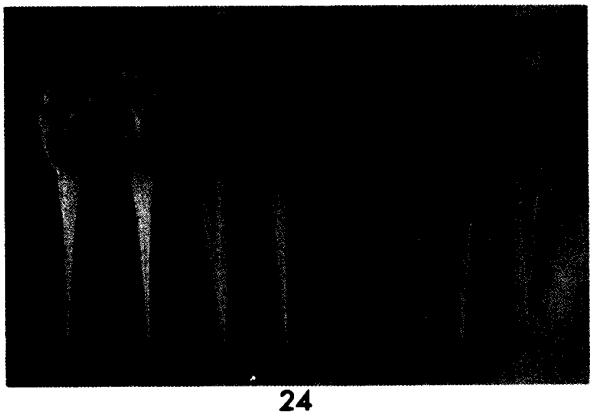
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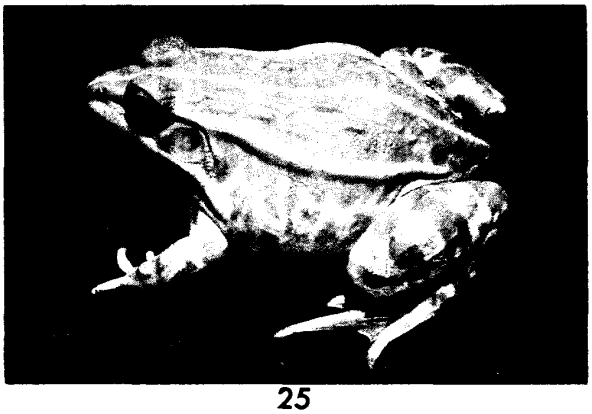
## PLATE IV

Albinos of four stocks belonging to the second, third, fourth and fifth groups in *Rana nigromaculata*.

25. Albino (♀) of the Go stock (*bb*) belonging to the second group. ×0.6
26. Albino (♂) of the Go stock (*bb*) belonging to the second group. ×0.6
27. Albino (♀) of the Ym I stock (*cc*) belonging to the third group. ×0.6
28. Albino (♂) of the Ym I stock (*cc*) belonging to the third group. ×0.6
29. Albino (♀) of the Km stock (*dd*) belonging to the fourth group. ×0.6
30. Albino (♂) of the Km stock (*dd*) belonging to the fourth group. ×0.6
31. Albinos at the juvenile stage of the Ty stock (*ee*) belonging to the fifth group. ×0.8
32. Albino (♀) at the mature stage of the Ty stock (*ee*) belonging to the fifth group. ×0.6



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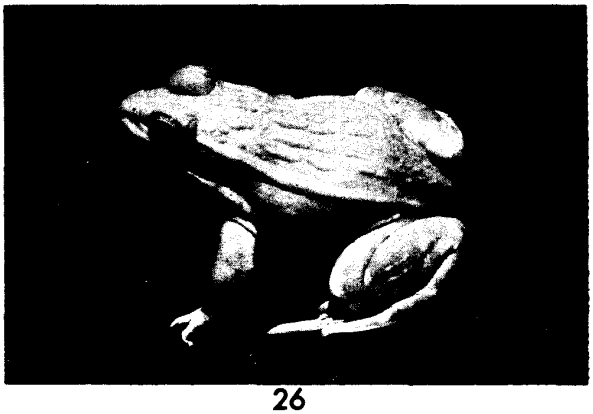

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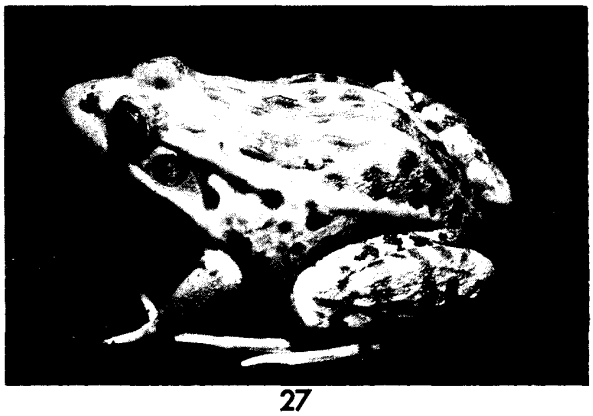

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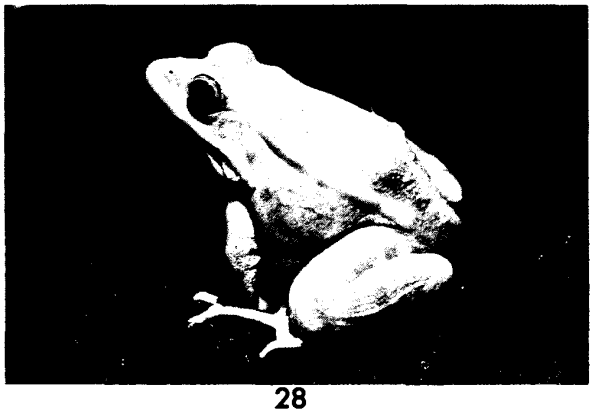

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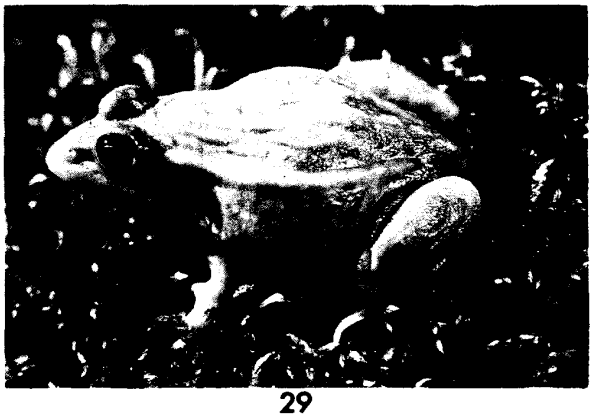

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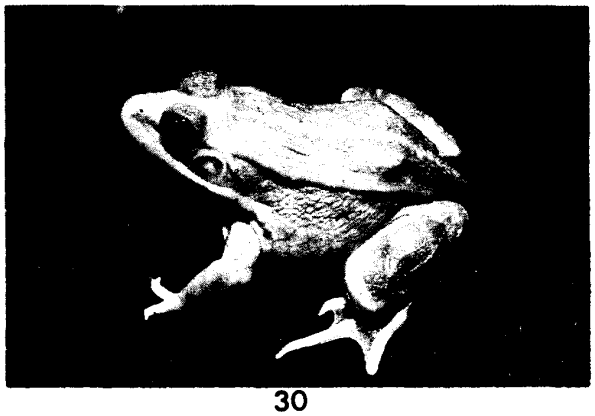

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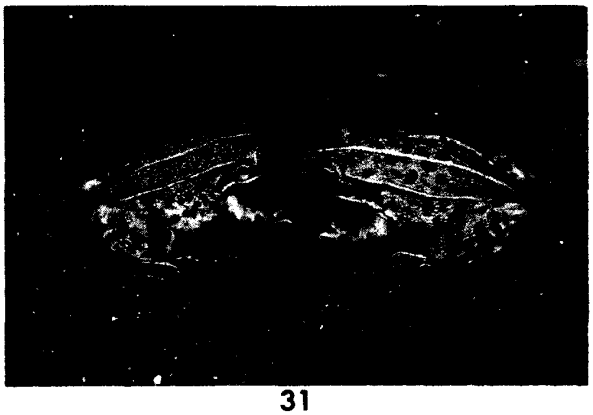

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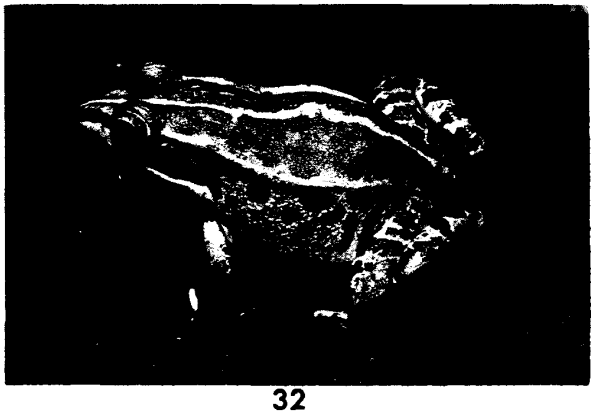

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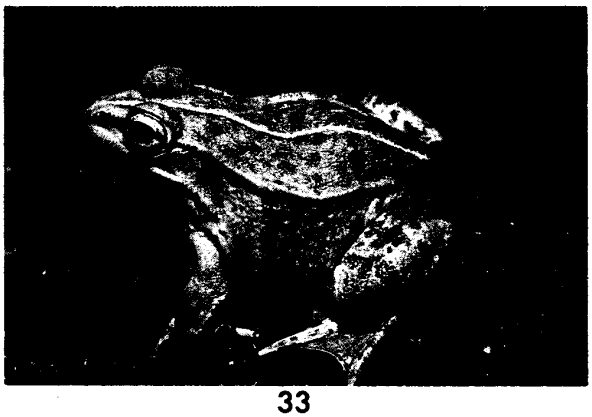
## PLATE V

Albinos of two stocks belonging to the fifth group, two kinds of albinic hybrids and eggs of female albinos.

33. Albino (♂) of the Ty stock (*ee*) belonging to the fifth group. ×0.6
34. Albino (♀) of the Ns stock (*ee*) belonging to the fifth group. ×0.6
35. Albino (♂) of the Ns stock (*ee*) belonging to the fifth group. ×0.6
36. Albinic hybrid of the Km·Sn I ($a^e a^e dd$). ×0.6
37. Albinic hybrid of the Km·Ym I (*ccdd*). ×0.6
38. Eggs of female albinos of the Sn I (a), Tj (b), Fc (c), Go (d), Ym I (e), Km (f) and Ty (g) stocks and a wild-type female (h). ×0.6
39. Ovary of a female albino of the Fc stock. A normal-colored egg is indicated by an arrow. ×1.6
40. Eggs laid by a female albino of the Fc stock after ovulation was accelerated by pituitary injection. Two normal-colored eggs are found among light-colored ones. ×2.0



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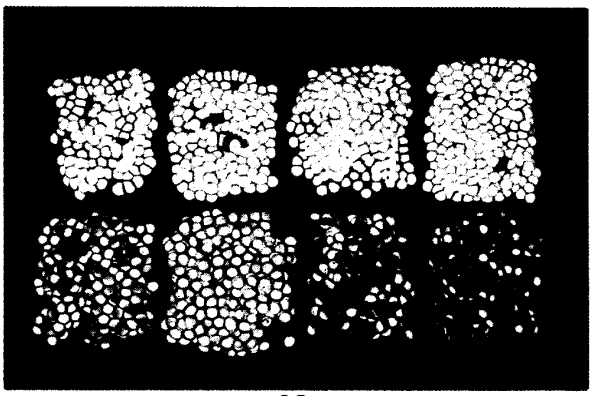

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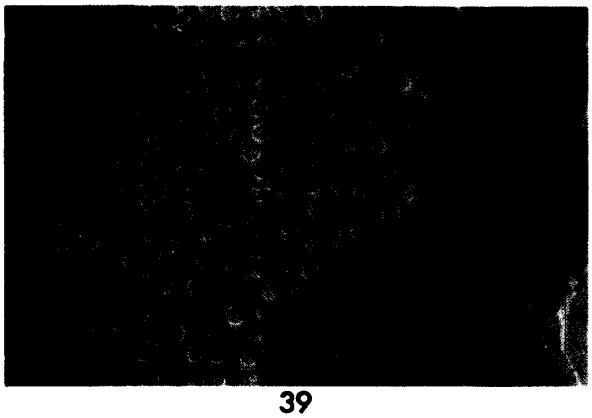

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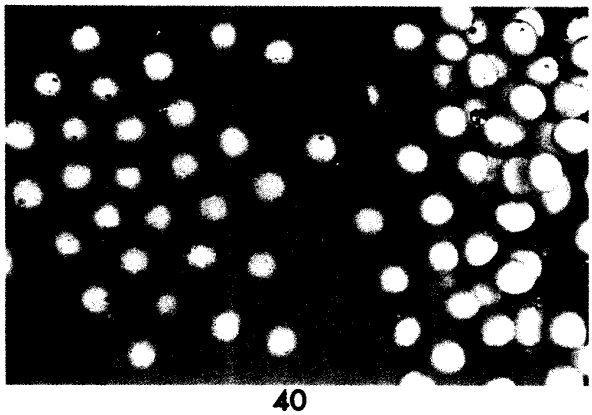

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