

# Effects of Gonadectomy on the Histological Structure of the Mandibular Glands in Male Mice

## III. Effects of Testosterone Administration

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### INTRODUCTION

The histological changes of the mice mandibular glands arising from gonadectomy were described in the previous papers<sup>1,2)</sup>, with the conclusion that there was a close connection between the testes and granular tubules of the mandibular glands.

However, it was left unconfirmed whether an atrophy of the granular tubule was caused by the disappearance of testosterone or not.

Since LACASSAGNE's (1940)<sup>3)</sup> report, there have been several literatures<sup>4,5,6,7,8,9)</sup> on the histological changes of the mandibular glands following after the administration of sexual hormone to the intact or gonadectomized animals. But in these literatures, there seemed to be some disarrangement concerning the age of animals, the periods after gonadectomy and the dosage and term of hormone-administration. Moreover, the literatures describing about the quantitative histological changes effected by hormone-administration during the long periods are scanty.

To examine the fact that disappearance of testosterone by the gonadectomy leads to the atrophy of the granular tubules in the gonadectomized mice, the changes of the histological features of the mandibular glands in the mice which were put under the testosterone-administration after gonadectomy were observed.

### MATERIALS AND METHODS

A total of 107 male ICR-JCL mice were divided evenly as possible by the weight and littermate into four groups.

Group 1 (testosterone administration). ... Animals were gonadectomized when 90 days old and have been injected subcutaneously with 0.1 ml testosterone solution (0.025 mg testosterone propionate/1 ml sesame oil) every day since the 20th day after gonadectomy. Then, animals were sacrificed to examine the mandibular glands on the 3rd, 5th, 10th and 20th day after the initial testosterone administration.

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Group II (oil administration). ... Animals have been injected subcutaneously with 0.1 ml sesame oil every day since the 20th day after the gonadectomy carried out when 90 days old, and were sacrificed on the 10th and 20th day after the initial administration.

Group III (castration). ... Animals were gonadectomized when 90 days old, and sacrificed on the 20th, 30th and 40th day after gonadectomy.

Group IV (control). ... As the intact control group, the mandibular glands in the intact mice 110, 120 and 130 days old were employed.

The rearing and the methods of observations were the same as those described in the previous paper<sup>1)</sup>. The body-weight of the animals used in this experiment is shown in figure 1.

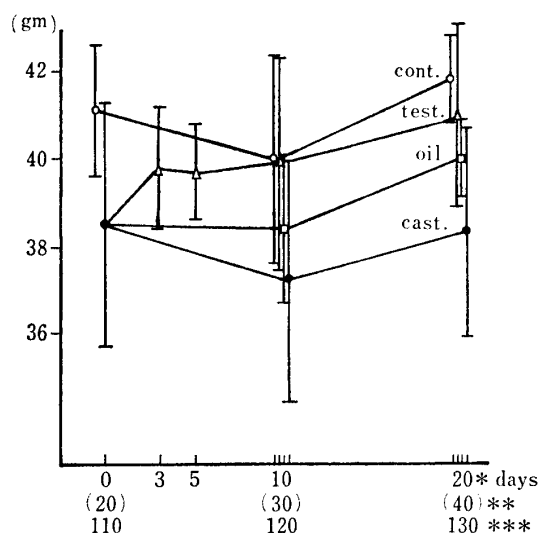


Fig. 1. Body-weight

\* days after the beginning of testosterone or sesame-oil administration.

\*\* days after gonadectomy.

\*\*\* days after birth.

△ test. group I (testosterone administration)

□ oil group II (oil administration)

● cast. group III (castration)

○ cont. group IV (control)

— | — means ± standard deviation.

## OBSERVATIONS

### 1) gland-weight (Fig. 2, 3)

The mandibular gland-weight and gland-weight-percentage per the body-weight in the group III (castration) were constant during the experimental period and distinctly smaller than those in the group IV (control). The gland-weight in the group II (oil administration) was similar to that in the group III (castration). On the 3rd day in the group I

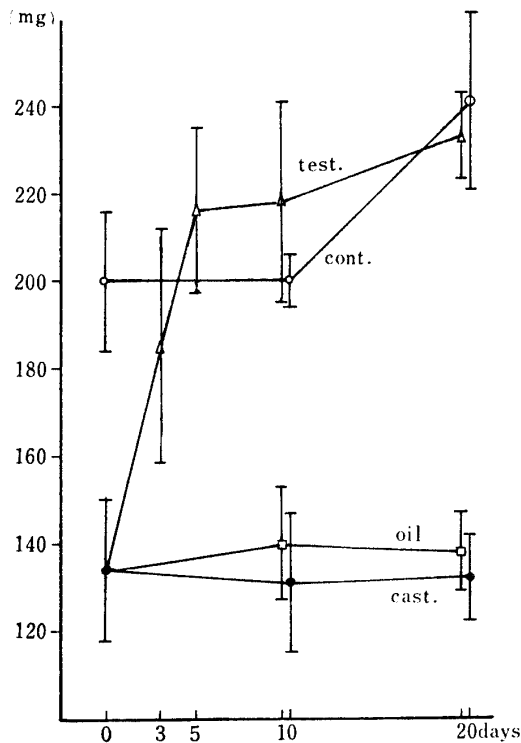


Fig. 2. Gland-weight

The form of the graphs in Fig. 2~Fig. 10 is the same as that in Fig. 1.

result of the histological observation in the group II (oil administration) was similar to that in the group III (castration). As mice grew older in the group IV (control), the content of granules in the granular tubules increased, the size of tubules as well as the height of their epithelial cells becoming larger.

On the 3rd day of experimental period in the group I (testosterone administration), the granular tubules were confirmed to be composed of certain epithelial cells containing numerous granules and the other epithelial cells not showing so much increase of granules. Generally the former cells were recognized near the intercalated ducts. On the 5th day, granules were observed in almost all the cells of the granular tubules, moreover, the size of tubules and the height of their epithelial cells became larger than those of the mice observed on the 3rd day. On the 10th and 20th day after testosterone-administration, the content of granules, the size of tubules and the height of their epithelial cells become more and more enlarged, the striated tubules becoming shorter.

These histological features of the mandibular glands were almost similar to those of the intact control mice 120 and 130 days old.

### 3) quantitative assessments

#### a) acini (Fig. 4, 5)

The number of the acini in 335 square microns in a lobulus in the group III (castration) was nearly constant during the twenty days of experimental period and distinctly

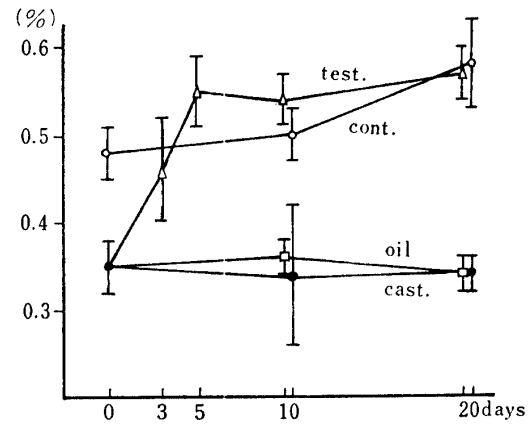


Fig. 3. Gland-weight-percentage per body-weight.

(testosterone administration) the gland-weight increased markedly, on the 5th day it increased quite suddenly, after that time its growth was slow.

### 2) histological observations

The changes of the histological feature of the mandibular glands in the group III (castration) were almost the same as those described in the previous paper<sup>1)</sup>. The re-

greater than that in the group IV (control) as shown in figure 4.

The changes of the acinar number in the group II (oil administration) showed no significant difference from those in the group III (castration). On the 5th day of experimental period in the group I (testosterone administration) the number of the acini decreased suddenly, on the 20th day being reduced to less than half of that in the group III (castration). Namely, the number of the acini was made to be returned to that of the intact control mice by the testosterone administration covering over five days.

The changes of the area of the acini in the definite field among the four groups were almost the same as those of the number of the acini as shown in figure 5.

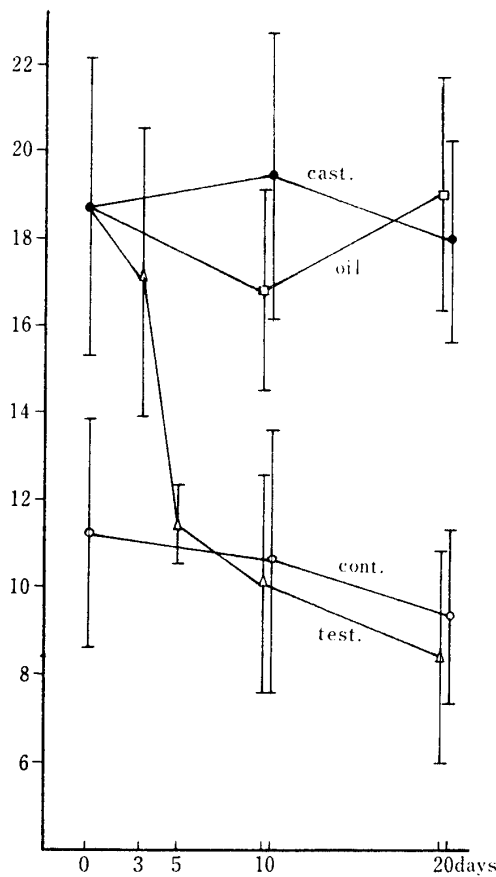


Fig. 4. Number of acini in 335 μ².

the above-stated definite field, however, some extreme changes were caused by the administration of testosterone. The areas of the granular tubules in the group II (oil administration) and the group III (castration) were constant respectively during the experimental period and far smaller than those in the group IV (control). As a result of testosterone-administration, on the 5th day after initial administration the area of the granular tubules increased suddenly, on and after the 10th day following up to the value of area observable in the intact control mice.

The size of a granular tubule in the histological sections increased also in consequence

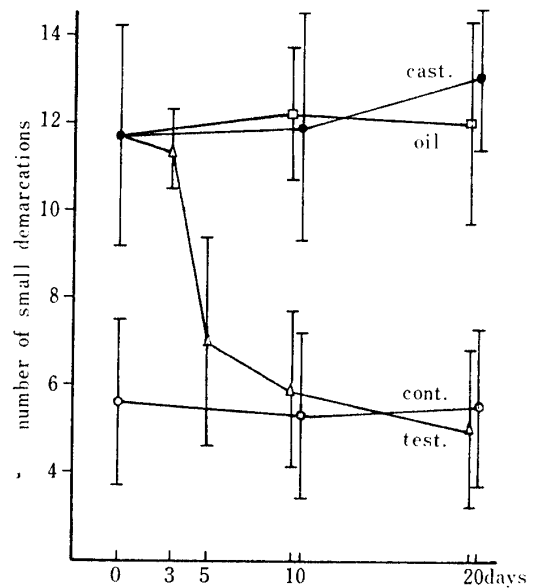


Fig. 5. Area of acini in 335 μ².

Therefore, the size of an acinus showed no difference among the four groups.

#### b) granular tubules (Fig. 6, 7, 8, 9, 10)

The number of the granular tubules in 335 square microns in the histological sections showed no significant difference among the four groups.

On the area of the granular tubules in

of the testosterone-administration as shown in figure 8.

The tubular/acinar ratio (T/A) was about 3.5 in the group IV (control), while in the group III (castration) and II (oil administration) respectively about 0.9. As shown in figure 9, on the 5th day in the group I (testosterone administration) it increased to 2.5; on the 20th day being in excess of the value in the intact controls.

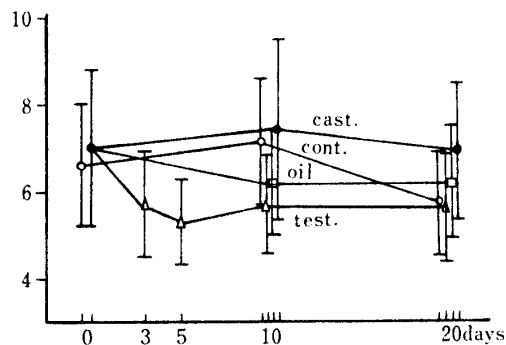


Fig. 6. Number of granular tubules in 335 μ².

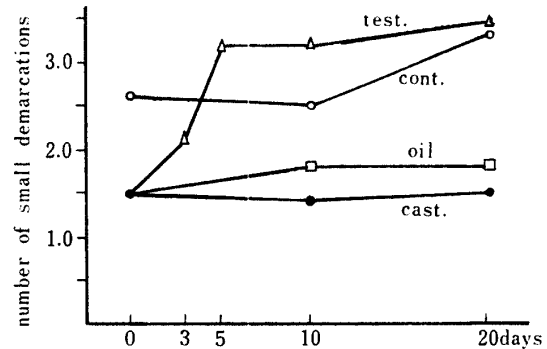


Fig. 8. Size of a granular tubule.

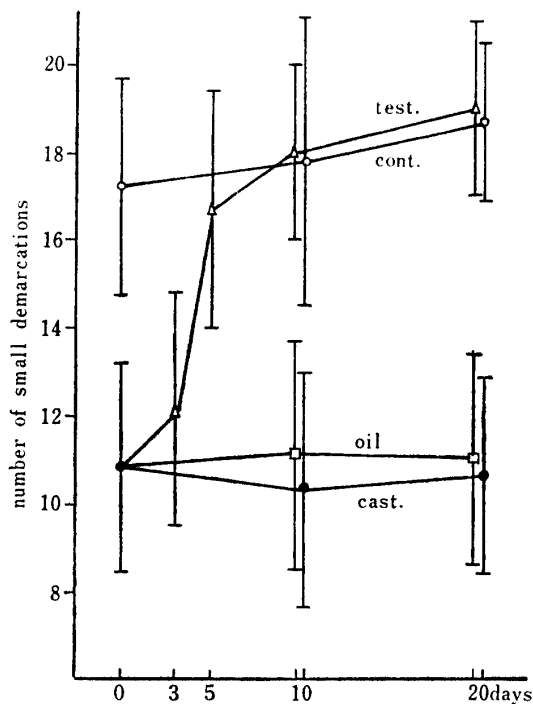


Fig. 7. Area of granular tubules in 335 μ².

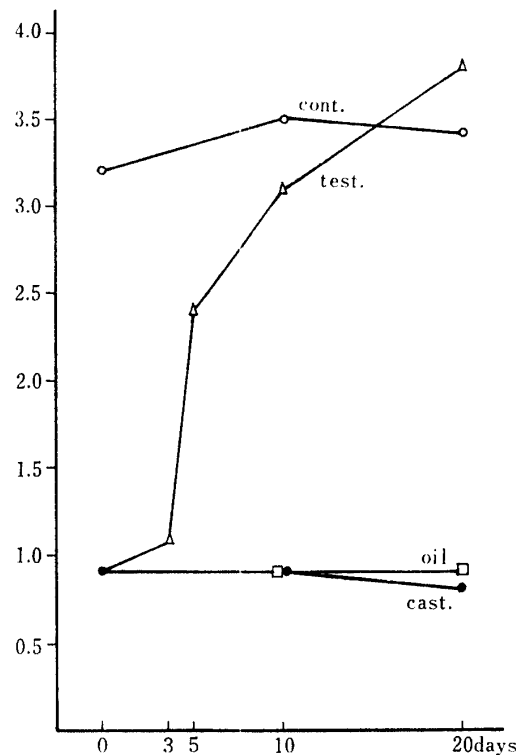


Fig. 9. Tubular/acinar ratio (T/A)

The changes of the diameters of the granular tubules and lumens and the height of their epithelial cells are shown in figure 10. These three measurement-values were found to be the same as those in the group IV (control) on and after the 10th day in

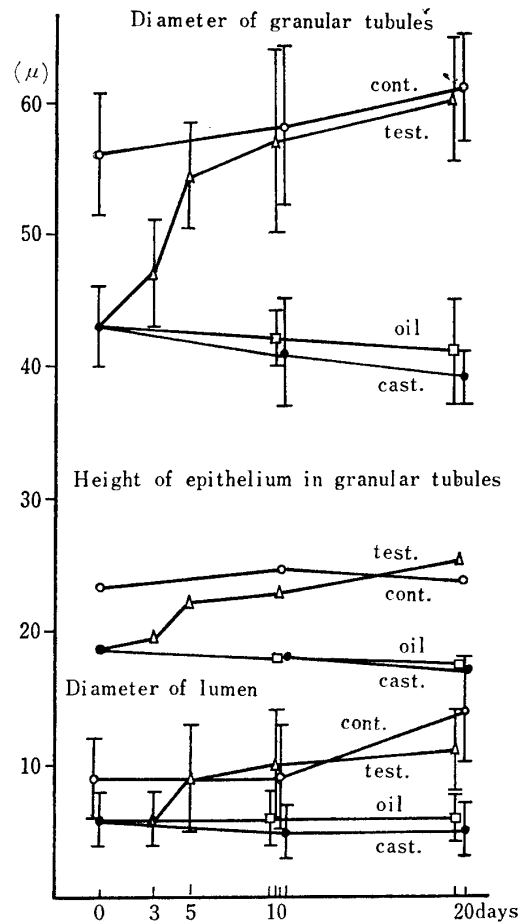


Fig. 10. Diameter of granular tubules.

\* Height of epithelium in granular tubules.

Diameter of lumen of granular tubules.

\* The value is  $\frac{\text{Diameter of granular tubule} - \text{Diameter of lumen of granular tubule}}{2}$

the group I (testosterone administration). The number of the nuclei in the granular tubules showing the round-shape in the histological sections presented no significant difference among the four groups.

## DISCUSSION

LACASSAGNE (1940)<sup>7,10)</sup> reported that when the gonadectomized adult mice were administered 5 mg testosterone-propionate beginning on the 23rd day after operation, the diameter of the granular tubules regained its normal value only through one-day-administration of the drug, the gland-weight returning on the 3rd day.

JUNQUEIRA (1949)<sup>4)</sup> described that when 14 mg testosterone was injected to the gonadectomized mice beginning on the 25th to 39th day after operation, the granular tubules were filled with granules and hypertrophied more than those in the normal control.

Moreover, SHAFER and MUHLER (1953)<sup>9)</sup> indicated that the implantation of 102 mg testosterone pellet into the gonadectomized mice induced more increase of the size and

number of the granular tubules than those in the control mice.

In these studies the dosage of testosterone was far larger than that in the present experiment, so it was conjectured that the excess dosage should be the cause of the changes of the histological feature.

As the start of the present experiment, the granular tubules in the gonadectomized mice showed distinguished atrophy resembling the result of the previous experiment. The injection of small dose of testosterone (0.0025 mg per day) produced slight increase in the size of granular tubules, in the height of their epithelial cells and in the tubular/acinar ratio on the 3rd day after initial administration. Those values increased suddenly on the 5th day, reaching approximately the values in the intact control mice on and after the 10th day. On the other hand, no change of the granular tubules was observed by the administration of sesame oil only.

These observations clearly show that the development of the granular tubules in mice mandibular glands is accelerated by the action of testosterone.

Furthermore, these findings support the view described in the previous two papers<sup>1,2)</sup> that the disappearance of testosterone from the testes leads to the atrophy of the granular tubules.

#### SUMMARY

The male mice gonadectomized when 90 days of age were injected with 0.1 ml testosterone propionate solution (0.025 mg testosterone-propionate/1 ml sesame oil) subcutaneously every day beginning on the 20th day after gonadectomy. Then the mandibular glands were examined on the 3rd, 5th, 10th and 20th day after the initial testosterone-administration.

1) By the testosterone-administration covering over five days, the mandibular gland-weight and the gland-weight-percentage per the body-weight reached those of the intact control mice.

2) The number and area of the acini in a definite field of the histological sections decreased suddenly on the 5th day after the initial testosterone-administration, reducing slowly after that. On the contrary, in consequence of testosterone-administration covering over ten days, the area of the granular tubules in a definite field enlarged almost equal to that of the intact control mice.

3) The diameter of the granular tubules and the height of their epithelial cells increased suddenly on the 5th day after the initial administration, increasing slowly after that.

4) An increase in the content of granules in the epithelial cells was observed in a part of the granular tubules near the intercalated ducts on the 3rd day after the initial administration, the content of granules becoming similar to that in the intact control mice on and after the 10th day.

5) These observations clearly show that the administration of testosterone to the gonadectomized mice accelerates quantitatively the development of the granular tubules of the mandibular glands.

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## Explanation of Plates

## Abbreviations

a ..... acinus

g ..... granular tubule

- Plate 1. Mandibular gland of the mouse sacrificed on the 20th day after gonadectomy, showing the decrease of granules in the granular tubules. (110 days of age, Group III) H·E staining,  $\times 100$ .
- Plate 2. Mandibular gland of the 110 days old intact control mouse. (Group IV) H·E staining,  $\times 100$ .
- Plate 3. Mandibular gland of the mouse sacrificed on the 10th day after initial sesame oil-administration. The feature is almost similar to that in Plate 1. (120 days of age, Group II) H·E staining,  $\times 100$ .
- Plate 4. Mandibular gland of the mouse sacrificed on the 10th day after initial testosterone-administration, showing increase of granules in the granular tubules. (120 days of age, Group I) H·E staining,  $\times 100$ .
- Plate 5. Mandibular gland of the 130 days old intact control mouse, showing a numerous granules in the granular tubules. (Group IV) Azan staining,  $\times 100$ .
- Plate 6. Mandibular gland of the mouse sacrificed on the 10th day after initial testosterone-administration. The feature is almost similar to that in Plate 5. (120 days of age, Group I) Azan staining,  $\times 100$ .



