*J. Physiol.* (1963), **165**, *pp.* 508–518
*With 3 text-figures*
*Printed in Great Britain*

# THE RELATION BETWEEN STRUCTURE AND THE CONCENTRATION OF IODIDE BY THE SUBMANDIBULAR GLANDS OF MICE AND HAMSTERS

BY K. BROWN-GRANT * AND W. TAYLOR
*From the Department of Physiology, University of Birmingham*

(*Received* 16 *July* 1962)

The submandibular salivary glands of adult mice, rats, and hamsters are larger, in proportion to body weight, in the male than in the female, the difference being due to the presence of a greater number of larger convoluted granular tubules (serous tubules) (Lacassagne, 1940*a*; Burgen & Emmelin, 1961). This portion of the duct system develops after birth (Jacoby & Leeson, 1959) and appears to be under some hormonal control. Degranulation and shrinkage of the tubules follows castration or hypophysectomy in the male and is affected to a lesser extent by thyroidectomy or treatment with antithyroid drugs (for references, see Burgen & Emmelin, 1961). Conversely, growth of the tubules ('masculinization') occurs in the female during pregnancy and lactation (Desclin, 1958) or can be induced by testosterone (Lacassagne, 1940*b*). The function of this portion of the duct system is not clearly established, but it appears that it is this section that is responsible for the high concentration of iodide, relative to the blood, in the gland itself and in submandibular or mixed saliva in the mouse and the hamster (Logothetopoulos & Myant, 1956).

In the present study an attempt has been made to relate changes in the structure of the convoluted granular tubules (C.G.T.) to the ability of the gland to concentrate iodide *in vivo* and *in vitro*.

## METHODS

The animals used were albino mice of the Parkes strain bred in the laboratory, and golden hamsters (*Mesocricetus auratus*) obtained originally from two different commercial sources or bred in the laboratory. Both species were fed on Thompson's rat cubes containing 0·000047 % by weight of iodine, and drank tap water. In addition, the hamsters were given a small quantity of oats and fresh cabbage daily. The animals were kept in a light- and temperature-controlled animal room (18–20° C) for at least 3 weeks before experiments were begun. Castration was carried out under ether or Avertin (tri-brom-ethanol; Bayer Products) anaesthesia; hypothyroidism was produced by replacing the usual drinking water by a saturated solution of 4-methyl-2-thiouracil (MTU) in tap water.

* Present address: Department of Human Anatomy, South Parks Road, Oxford.

Organs for histological examination or whose weights or $^{131}I$ content were required were removed, trimmed, blotted, and weighed to 0·1 mg on a torsion balance. Tissues were fixed in 5% neutral formol-saline, embedded in paraffin and sectioned at 5–7 $\mu$. Usually only haematoxylin- and eosin-stained sections were prepared, but some glands were also stained by the periodic acid-Schiff method and counterstained with aniline blue and Orange G. Measurements of C.G.T. diameters and cell heights were made microscopically, by means of an eyepiece calibrated in arbitrary units which were subsequently converted to microns. The mean value for each gland was determined from measurements of twenty-five tubules selected at random from the whole gland. Only tubules were measured which possessed a lumen and which were cut in either transverse or oblique planes. The recorded tubular diameter was the minimum for that tubule, and cell heights were measured along the same axis. Mean values for each gland were pooled to obtain the group mean.

The radioactive isotope used was $^{131}I$, obtained as a carrier-free solution of NaI in NaCl solution, 0·9 g/100 ml., from the Radiochemical Centre, Amersham. Samples of tissues and fluids were placed in polythene containers and counted in a well-type NaI crystal scintillation counter to a statistical accuracy of $\pm 1{\cdot}5\%$ or better. Experiments were of two kinds. Animals were injected intraperitoneally with 0·5–5·0 $\mu$c of $^{131}I$; 2 or 2·5 hr later they were anaesthetized with ether and killed by bleeding. When thiocyanate was given, the dose was 10 mg/100 g body weight intraperitoneally, either 20 min before or 1·5 hr after the injection of $^{131}I$. The $^{131}I$ content of salivary gland and blood samples was determined and the gland/blood (G/B) ratio (counts/sec/100 mg tissue divided by counts/sec/0·1 ml. blood) calculated. The tissue/medium (T/M) ratio for salivary gland slices incubated *in vitro* was determined by a modification of the method described by Fletcher, Honour & Rowlands (1956). The animals were stunned, killed by bleeding and the glands dissected out rapidly. Where a series of determinations on similar tissues were required, glands were cut into six or eight pieces, pooled and weighed before being placed in the incubation flasks; where a series of animals were compared, one or one and a half glands from each animal were weighed and sliced, the remainder being kept for histological examination. When the effect of various compounds added to the incubation medium was studied, one gland from each animal was used as control and the other as the experimental tissue. The incubation medium (Fletcher *et al.* 1956) was a buffered saline solution (adjusted to pH 7·4 at room temperature by the addition of M-$H_3PO_4$) of the following composition: water, purified by passage through an ion exchange resin, 100 ml., NaCl 850 mg, KCl 42 mg, $CaCl_2$ 24 mg, glucose 100 mg, $Na_2HPO_4$ 60 mg. All chemicals were of A.R. grade. Seventy to 120 mg of tissue was added to 3 ml. of medium in an unstoppered 25 ml. conical flask and shaken mechanically during incubation at 38° C in a water-bath. All preparations were carried out at 0° C; after adding the tissue slices to the flask, they were incubated at 38° C for 5 min before adding a further 0·5 ml. of medium containing 0·1 $\mu$c of $^{131}I$-iodide, and incubation was then continued for a further 50 min. Preliminary experiments showed that maximal T/M ratios were established after 40 min incubation and did not increase further up to 90 min of incubation. At the end of the period of incubation the slices were removed, washed, blotted, and counted and a sample of the incubation medium from each flask was also counted. The tissue/medium concentration ratio (T/M ratio) (counts/sec/100 mg tissue divided by counts/sec/0·1 ml. medium) was calculated.

All values given in text and tables are means $\pm$ standard error of the mean (S.E.M.). The significance of differences between means was estimated by the $t$ test and probability ($P$) values are quoted. A value for $P$ of $>0{\cdot}05$ is reported as not significant (N.S.). Where glands from the same animal were compared in the experiments *in vitro*, the mean control value and the mean difference ($\Delta$T/M) are quoted; $P$ values in such experiments refer to the results of a paired $t$ test.

## RESULTS

### *Gland/blood ratios for $^{131}I$ in normal and abnormal mouse glands*

The high concentration of radio-iodide in the submandibular glands of normal adult mice reported by previous workers (Logothetopoulos & Myant, 1956; Taurog, Potter & Chaikoff, 1959) was confirmed. Six adult male mice gave a G/B ratio of $4{\cdot}58 \pm 0{\cdot}69$; the value for the sublingual glands in these animals was $0{\cdot}45 \pm 0{\cdot}10$. The effect of KSCN given 20 min before or 1·5 hr after the radio-iodide was determined. The G/B ratios for the submandibular glands were $0{\cdot}82 \pm 0{\cdot}24$ and $0{\cdot}81 \pm 0{\cdot}10$ respectively ($P < 0{\cdot}001$). Sublingual gland G/B ratios were unchanged ($0{\cdot}37 \pm 0{\cdot}02$ and $0{\cdot}40 \pm 0{\cdot}01$). A possible difference between the G/B ratios for the submandibular glands of adult male and female mice was investigated. Seven male mice gave a value of $6{\cdot}80 \pm 1{\cdot}12$ and seven female mice $4{\cdot}74 \pm 0{\cdot}70$; this difference was, however, not statistically significant. Histologically, the convoluted granular tubules (C.G.T.) were much more prominent in sections from the glands of male mice. The mean tubule diameter was $23{\cdot}2 \pm 0{\cdot}6$ in males and $15{\cdot}3 \pm 0{\cdot}5$ in females ($P < 0{\cdot}01$) and the mean tubule cell heights were $9{\cdot}1 \pm 0{\cdot}2$ and $6{\cdot}5 \pm 0{\cdot}2$ ($P < 0{\cdot}01$).

The effect on the G/B ratio for $^{131}I$ of procedures known to affect the size, appearance and activity of the C.G.T. was investigated. Treatment of male mice with MTU for 38 days caused the expected increase in thyroid gland weight and a decrease in submandibular gland weight. Seven controls gave values of $11{\cdot}6 \pm 1{\cdot}3$ mg/100 g body weight and $457 \pm 21$ mg/100 g body weight and seven MTU-treated animals $22{\cdot}5 \pm 2{\cdot}3$ ($P < 0{\cdot}01$) and $353 \pm 33$ ($P < 0{\cdot}05$). Histologically the glands showed a decrease in the proportion of tubules in relation to acinar tissue and some degranulation of tubule cells. The C.G.T. diameters and cell heights were reduced, but not significantly; control values were $23{\cdot}7 \pm 0{\cdot}3$ and $8{\cdot}9 \pm 0{\cdot}5$ as compared with $21{\cdot}8 \pm 0{\cdot}8$ and $8{\cdot}5 \pm 0{\cdot}3$ in MTU-treated animals. The G/B ratios were not reduced, but were in fact higher in MTU-treated animals, but the difference was not significant ($7{\cdot}40 \pm 1{\cdot}95$, compared with a control value of $5{\cdot}00 \pm 1{\cdot}5$). Twelve mice were castrated; six of these were injected with 500 $\mu$g of testosterone propionate in 0·1 ml. of oil subcutaneously twice weekly for 3 weeks. G/B ratios for these and for six control animals were determined 26 days after operation. Submandibular gland weights and seminal vesicle weights showed the expected changes; gland and vesicle weights (mg/100 g body weight) were $604 \pm 30$ and $411 \pm 47$ for controls; $343 \pm 21$ and $123 \pm 7$ for castrates and $519 \pm 14$ and $445 \pm 49$ for testosterone-injected castrates. Values for castrated animals were significantly different from control or injected animals ($P < 0{\cdot}001$ in all cases). Histologically the glands of castrate animals showed a marked decrease in the

proportion of tubules to acinar tissue with gross degranulation and shrinkage of tubule cells; these changes were not seen in the testosterone-treated animals. Histometric measurements gave values for intact, castrate, and castrate testosterone-treated animals of $30{\cdot}2 \pm 0{\cdot}6$, $19{\cdot}2 \pm 0{\cdot}6$ and $26{\cdot}9 \pm 0{\cdot}5$ ($P < 0{\cdot}01$) for tubule diameters and $10{\cdot}2 \pm 0{\cdot}3$, $7{\cdot}2 \pm 0{\cdot}2$ and $9{\cdot}2 \pm 0{\cdot}3$ ($P < 0{\cdot}01$) for cell heights. Despite these striking histological changes in the C.G.T., G/B ratios were not reduced, but were in fact significantly increased in the castrate group ($9{\cdot}91 \pm 2{\cdot}54$, compared with a control value of $3{\cdot}57 \pm 0{\cdot}58$; ($P < 0{\cdot}05$). The glands of castrate mice treated with testosterone, which were essentially normal in weight and in histological appearance, also gave a raised G/B ratio (of $6{\cdot}82 \pm 0{\cdot}92$; $P < 0{\cdot}02$). These changes in the direction of a raised G/B ratio for the atrophied glands of hypothyroid or castrated animals had not been expected; rather a decrease in G/B ratio had been expected. A similar, raised G/B ratio for $^{131}I$ has, however, been found for the atrophic glands of hypophysectomized mice by Taurog *et al.* (1959). These workers have suggested that the increase could be due to the accumulation of saliva of high $^{131}I$ content within the gland. Histologically there was no evidence of accumulation of saliva or of dilatation of the striated intra- or interlobular ducts or the collecting ducts in the glands of the mice used in the present experiments. An attempt was made to discover whether acute changes in the gland content of saliva influenced the G/B ratio. Ten male mice were anaesthetized with sodium pento-barbitone 1·5 hr after the injection of $^{131}I$. Five received saline intramuscularly and were killed 10 min later; five received pilocarpine nitrate (20 $\mu$g/10 g body weight intramuscularly) and were killed 10–15 min later, when they were salivating profusely. Gland/blood ratios were not significantly altered; controls $6{\cdot}49 \pm 1{\cdot}23$, pilocarpine-injected $5{\cdot}54 \pm 0{\cdot}83$.

### *Tissue/medium concentration ratios for $^{131}I$ in vitro*

The histological examination of the glands and the experiments with pilocarpine described above did not exclude the possibility that the raised G/B ratios observed were due to accumulation of saliva within the glands. It was considered, however, that the T/M concentration ratio for $^{131}I$ *in vitro* was less likely to be affected in this way, and that this might be a better index of the extent of development of the C.G.T. and of their iodide-concentrating capacity. In a preliminary experiment glands from nine male mice incubated for 50 min gave a T/M ratio of $2{\cdot}82 \pm 0{\cdot}28$; the second gland from each of these animals was incubated for 50 min in the presence of $KClO_4$ at a final concentration in the medium of $4 \times 10^{-5}$M. The mean change in T/M ratio ($\Delta$T/M) was $-1{\cdot}73$, $P$ value in the paired $t$ test $< 0{\cdot}01$.

The T/M ratios were determined for slices of glands from male and female

mice, from male mice treated with MTU, and from castrated male mice with and without testosterone injection. The T/M ratios obtained and other relevant results are given in Table 1. No measurements of tubule diameters or cell height were made on glands from these animals, but at least two glands from each group were sectioned and examined. The histological differences were essentially the same as those observed in the glands from the animals used in the experiments on G/B ratios. Glands from male animals gave a higher T/M ratio than those from females. Atrophied glands

TABLE 1. T/M ratios for $^{131}I$ *in vitro* for submandibular gland slices from male and female mice and from male mice given methyl-thiouracil or castrated. Values are means ± standard error of the mean and *P* values refer to the *t* test

| Animals | No. | T/M ratio | Submandibular gland (mg/100 g body wt.) | Thyroid (mg/100 g body wt.) | Seminal vesicles (mg/100 g body wt.) |
|---|---|---|---|---|---|
| Intact males | 7 | 2·32 ± 0·11 | 456 ± 20 | — | — |
| Intact females | 7 | 1·94 ± 0·18 | 300 ± 18 | — | — |
| *P* | — | N.S. | < 0·01 | — | — |
| Control males | 6 | 1·71 ± 0·60 | 502 ± 19 | 8·9 ± 0·8 | — |
| MTU-treated males | 6 | 2·20 ± 0·24 | 455 ± 35 | 29·8 ± 33 | — |
| *P* | — | N.S. | N.S. | < 0·001 | — |
| Control males | 10 | 3·04 ± 0·15 | 397 ± 18 | — | 335 ± 15 |
| Castrate males | 10 | 5·05 ± 0·25 | 291 ± 12 | — | 77 ± 5 |
| *P* | — | < 0·01 | < 0·001 | — | < 0·001 |
| Control males | 6 | 2·26 ± 0·23 | 555 ± 22 | — | 486 ± 37 |
| Castrate males | 6 | 3·14 ± 0·40 | 348 ± 20 | — | 70 ± 11 |
| Castrate testosterone-treated | 6 | 2·14 ± 0·11 | 492 ± 15 | — | 273 ± 19 |
| *P* Control v. castrate | — | N.S. | < 0·001 | — | < 0·001 |
| *P* Control v. treated | — | N.S. | N.S. | — | < 0·001 |
| *P* Treated v. castrate | — | < 0·05 | < 0·001 | — | < 0·001 |

showing shrinking and degranulation of the C.G.T. from hypothyroid and untreated castrate animals had higher T/M ratios than glands from control animals. It seemed, therefore, that the ability to concentrate iodide either *in vivo* or *in vitro* bore no positive relationship to the maintenance of the normal histological appearance of the convoluted granular tubules in adult male mice.

### *Gland/blood ratios in mice and hamsters of different ages*

The C.G.T. develop post-natally from the striated intralobular ducts during the first four weeks of life (Jacoby & Leeson, 1959). Initially, it was intended to measure the G/B ratio *in vivo* of young male mice, in the expectation that a G/B ratio greater than unity might not be found until after the C.G.T. had developed. In fact a complex series of changes in G/B ratio with age was observed; these are illustrated in Fig. 1. Figure 2 shows the changes in submandibular gland weight in relation to body weight in these animals. Each point represents the mean of 5–7 male mice from one or two litters. High G/B ratios are found even in animals only a few days

old. The possibility that these values might be due to a delay in equilibration between tissues and blood was considered; however, in all cases the liver/blood concentration ratio for $^{131}I$ was determined and was usually 0·3–0·4 and never more than 0·5. In animals more than 15 days old the sublingual gland/blood ratio was also determined and was always less

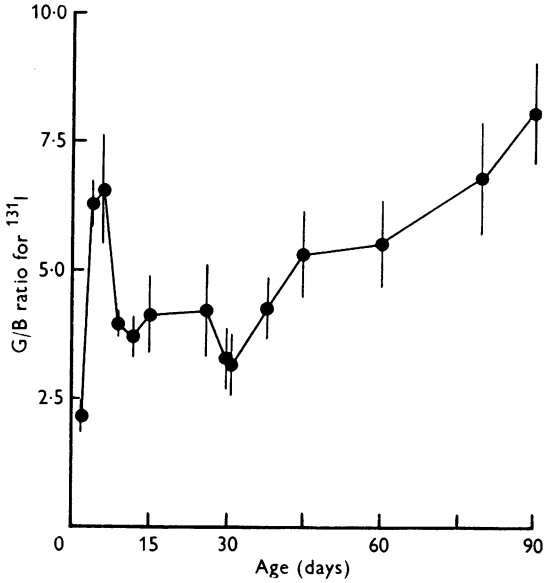


Fig. 1. The gland/blood ratio for $^{131}I$-iodide *in vivo* for the submandibular glands of male mice of different ages. Each point represents the mean of values from five to seven animals from 1 or 2 litters. Vertical bars indicate ± standard error of the mean.

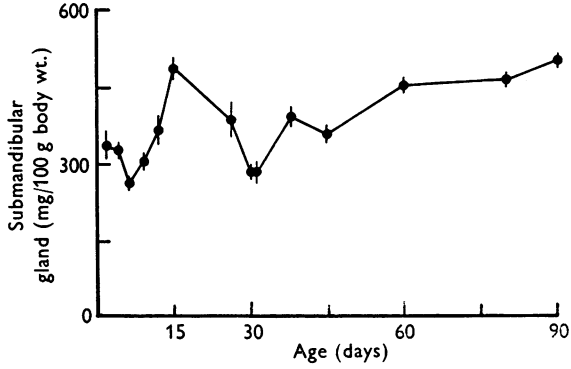


Fig. 2. The relative weight (mg/100 g body weight) of the submandibular gland in male mice of different ages. Each point represents the mean value for the groups of animals whose G/B ratios are shown in Fig. 1. Vertical bars indicate ± standard error of the mean.

than 0·7. The glands of at least two mice from each group were sectioned and examined. The changes in G/B ratio and in relative weight are best considered together with the histological findings. The terminology used is that suggested by Jacoby & Leeson (1959); the histological findings are in agreement with those reported briefly by Jacoby (1959). At 2 days the gland is composed mainly of the major interlobular ducts; the lobules are barely recognizable and contain simple ducts and terminal tubules only, in loose connective tissue; the G/B ratio is low, but above 1·0. At 4 and 6 days lobules are better formed; the interlobular ducts, intercalated ducts and terminal tubules are easily distinguished; they form the major mass of the gland at this stage: a few epithelial cells budded out of the terminal tubules can be seen; the relative weight of the gland is about the same; the G/B ratio is high. At 9, 12, and 15 days, the most striking change is the rapid progressive development of acinar cells. By 22 days the proportion of acinar to tubule tissue has reversed from the state at 6 days of age. This rapid growth of acinar tissue is probably related to the abrupt increase in relative weight and the striking fall in the G/B ratio (Figs. 1 and 2). Changes in the duct system are also occurring during this period. Basal striations are distinguishable in some of the more distal portions of the intralobular duct system and tubules cut in longitudinal section appear convoluted. Between 22 and 35 days the relative proportions of tubules and acini remain more or less the same, as does the relative weight and the G/B ratio. The nature of the ducts present is changing, however, the primary intercalated ducts of the early stages are becoming much less common, and the major proportion of the intralobular duct system is now composed of striated ducts with increasingly obvious granulation of the apical portion of more and more of the duct cells. Whole sections of convoluted duct with the typical adult appearance of the cells are seen by 30–31 days; the G/B ratio may be a little lower at this stage and also the relative gland weight. From this age on there is a progressive increase in the proportion of typical C.G.T. within the lobules at the expense of the acinar cells; only short sections of striated intralobular ducts are seen. The relative weight of the gland increases over this period, as does the G/B ratio.

The submandibular gland of the adult hamster also shows a high concentration of iodide relative to the blood. Logothetopoulos & Myant (1956) reported G/B ratios of 4–5; in the animals used in these experiments values of 1·5–2·5 were more common. A strain difference such as has been reported in mice (Taurog *et al.* 1959) seems the most likely explanation for this difference. The G/B ratios obtained from a series of forty-eight hamsters are shown in Fig. 3; all glands were examined histologically. Values for males and females did not differ significantly and results from both were combined. The G/B ratios at all ages are much lower than those for mice,

but even in animals 5 days of age the G/B ratio for the submandibular gland is higher than that for the sublingual. At this age the lobules of the gland contain a very few striated ducts, but are composed mainly of terminal tubules and primary intercalated ducts. At 10–15 days of age the G/B ratio is a little higher; developing acini are present; the major intralobular ducts are more obvious, but basal striations are still only seen in a very few of these duct cells. At 20–30 days of age the majority of the

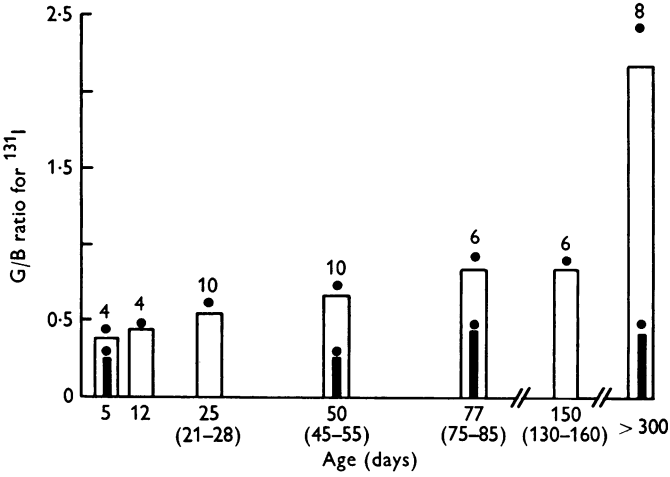


Fig. 3. The gland/blood ratios for radio-iodide *in vivo* for the submandibular glands of male and female hamsters of different ages. Figures above the columns give the number of animals in each group and figures below give the mean age in days and the range of ages. Solid columns indicate G/B ratios for the sublingual glands. The dots above the columns indicate plus one standard error of the mean in all cases.

intralobular ducts show basal striations, and the relative proportion of ducts as opposed to acinar tissue has risen a little. At 50 days of age the relative proportion of tubules remains about the same, but some cells now contain a small number of rather poorly staining eosinophilic granules, while at 70 days the appearance of the tubules is much the same, though their mass, relative to that of the acinar tissue, has increased slightly. In older animals granulation is more marked and at the same time the number of tubules is strikingly increased. The development of the typical adult granular tubule is much slower in the hamster than in the mouse; the G/B ratio appears to be more closely related to the relative proportion of duct tissue of any type in the gland than to the development of the typical adult histological appearance.

## DISCUSSION

The sexual dimorphism of the submandibular glands of adult mice provided the basis for a test of the hypothesis that differences in the extent of the post-natal development of the convoluted granular tubules might be reflected in differences in the gland/blood ratio for radio-iodide *in vivo* and the tissue/medium ratio for gland slices incubated *in vitro*. Higher values were observed in males than in females, as predicted. The differences were not statistically significant, but since these experiments were carried out, G/B ratios for $^{131}I$ determined by a similar method have been reported to be significantly higher in male than in female mice (Llach, Tramezzani & Cordero Funes, 1960).

Hypothyroidism and castration result in atrophy of the submandibular glands of male mice; the weight change is due to a decrease in size of the C.G.T. with shrinking and degranulation of the duct cells. The histological changes are associated with a decrease in the protease and amylase content of the gland (Shafer & Muhler, 1960). The G/B ratio and the T/M ratio are raised, however, in such glands, despite the occurrence of typical changes in C.G.T. Hypophysectomy, which also causes tubular atrophy, has been reported by Taurog *et al.* (1959) to result in a raised G/B ratio *in vivo*. These workers have suggested that this could be due to the retention of saliva of high iodide content within the gland. No histological or experimental evidence to support this view was obtained in the present experiments.

Studies on the G/B ratio of the submandibular gland of hamsters and mice of different ages supported the view that iodide concentration is not dependent on the presence of tubules with the typical adult histological appearance. Concentration of iodide above the level observed in the sublingual gland occurs in hamsters of 5 days of age, in which the glands are composed mainly of undifferentiated tubules and the G/B ratio increases, as striated intralobular ducts develop and form an increasing proportion of the gland mass, long before convoluted granular tubules begin to form at 50–70 days of age. In the adult the ratio seems to be related more to the proportion of intralobular ducts of all types than to the presence of typical convoluted granular tubules. The mouse was studied in greater detail; glands of young animals show a high G/B ratio before convoluted granular tubules or even striated intralobular ducts are present. The ratio falls between 6 and 10 days of age, owing perhaps to a 'dilution' of the concentrating tubules by the rapid development of acinar tissue over this period. From 12 to 40 days of age the ratio remains fairly constant, despite the fact that the appearance of the intralobular duct system changes dramatically as the adult C.G.T. develop at about 30 days

of age. Subsequently there is a steady rise in G/B ratios; the appearance of the tubules is not changing over this period but the relative proportion of tubule to acinar tissue in the gland is increasing. In both species the G/B ratio appears to be related more to the relative mass of duct tissue within the lobules than to the presence of ducts of the typical adult appearance.

The G/B and T/M ratios may not seem to be very direct indices of the total iodide-concentrating capacity of the gland. The values obtained for glands from male and female mice differ, however, the values for males being higher, as would be expected from the greater proportion of tubular tissue in these glands. The concentration of $^{131}$I in mixed or submandibular gland saliva relative to plasma might be thought to be a better measure of iodide concentrating capacity, but Cohen & Myant (1959) found no difference in saliva/plasma ratios for $^{131}$I in male and female mice. Further, the concentration of $^{131}$I-iodide in saliva is known to be influenced by the rate of flow of saliva (for references, see Brown-Grant, 1961) and procedures resulting in atrophy of the C.G.T. may also cause a diminished rate of saliva production (Shafer & Muhler, 1960), which would complicate the interpretation of the results.

Secondly, it might be argued that the initial assumption on which this work was based, that it is the C.G.T. that are responsible for the high concentration of iodide in the submandibular glands of adult animals, is a mistaken one. The autoradiographic evidence obtained by Logothetopoulos & Myant (1956) appears, however, to be unequivocal. Granted that these are the elements responsible in the adult, it would appear from the results presented here that the ability to concentrate iodide cannot be related to the histological appearance of the tubule cells and that concentrating ability is normal, or even enhanced, in tubules that would be described as atrophic on the basis of their histological appearance or their digestive-enzyme content.

## SUMMARY

1. The gland/blood ratio *in vivo* and the tissue/medium ratio *in vitro* for $^{131}$I-iodide is higher for the submandibular glands of male than those of female mice.

2. Glands from hypothyroid or castrate male mice show higher ratios than those from normal controls, although the convoluted granular tubules are histologically atrophic.

3. The glands of young mice and hamsters concentrate iodide *in vivo* before the convoluted granular tubules develop.

4. It is suggested that these ratios measure the iodide-concentrating capacity of the glands and that this capacity is retained or even enhanced in the histologically atrophic tubule.

Our thanks are due to Mr Melvin Sherwood for his valuable technical assistance and to Mr David Varley who prepared the sections for us. A grant from the Faculty of Medicine Research Fund to K. B-G., towards the cost of purchase of the scintillation counter used, is gratefully acknowledged.

## REFERENCES

BROWN-GRANT, K. (1961). Extrathyroidal iodide concentrating mechanisms. *Physiol. Rev.* **41**, 189–213.

BURGEN, A. S. V. & EMMELIN, N. G. (1961). *Physiology of the Salivary Glands.* London: Arnold.

COHEN, B. & MYANT, N. B. (1959). Concentration of salivary iodide: a comparative study. *J. Physiol.* **145**, 595–610.

DESCLIN, J. (1958). Masculinization des glandes sous-maxillaires chez la souris femelle pendant la gestation et la lactation. *C.R. Soc. Biol., Paris*, **152**, 1844–1848.

FLETCHER, K., HONOUR, A. J. & ROWLANDS, E. N. (1956). Studies on the concentration of radioiodide and thiocyanate by slices of the salivary gland. *Biochem. J.* **63**, 194–199.

JACOBY, F. (1959). Observations on the post-natal development of the mouse submaxillary gland. *J. Anat., Lond.*, **93**, 579 (Abstract).

JACOBY, F. & LEESON, C. R. (1959). The post-natal development of the rat submaxillary gland. *J. Anat., Lond.*, **93**, 201–216.

LACASSAGNE, A. (1940*a*). Dimorphisme sexuel de la glande sous-maxillaire chez la souris. *C.R. Soc. Biol., Paris*, **133**, 180–181.

LACASSAGNE, A. (1940*b*). Reactions de la glande sous-maxillaire à l'hormone male chez la souris et le rat. *C.R. Soc. Biol., Paris*, **133**, 539–540.

LLACH, J. F., TRAMEZZANI, J. H. & CORDERO FUNES, J. R. (1960). A sexual difference in the concentration of iodine-131 by the submaxillary gland of mice. *Nature, Lond.*, **188**, 1204–1205.

LOGOTHETOPOULOS, J. H. & MYANT, N. B. (1956). Concentration of radio-iodide and $^{35}$S-thiocyanate by the salivary glands. *J. Physiol.* **134**, 189–194.

SHAFER, W. G. & MUHLER, J. C. (1960). Endocrine influences upon salivary glands. *Ann. N.Y. Acad. Sci.* **85**, 215–227.

TAUROG, A., POTTER, G. D. & CHAIKOFF, I. L. (1959). The effect of hypophysectomy and of TSH on the mouse submaxillary iodide pump. *Endocrinology*, **64**, 1038–1051.