

sure. Posteriorly, entering in the tentacles, the nervous fibres form subepithelial plexuses (Figure 1) interconnected by bipolar cells (Figure 2). The plexuses give rise to several neuro-epithelial endings (Figure 3) and cells (Figure 4). This technique was also used with much success on annelids (Figure 5).

It would seem, then, that MAILLET's technique could have some use in histological works about peripheral nervous structures on invertebrates. We hope soon to obtain other results concerning this hypothesis¹⁵.

Résumé. La technique de fixation-coloration de MAILLET avait fourni des résultats intéressants pour la mise en évidence des fibres nerveuses amyéliniques chez les Vertébrés. Nous avons appliqué cette technique à des Invertébrés, notamment les Mollusques; elle s'est révélée efficace.

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- ¹ M. MAILLET, C. r. Séanc. Soc. Biol. 153, 939 (1959).
- ² M. MAILLET, Trab. Inst. Cajal Invest. biol. 54, 1 (1962).
- ³ V. JABONERO, L. FABRA, J. Y. MOYA and R. M. JABONERO, Z. mikrosk.-anat. Forsch. 43, 123 (1961).
- ⁴ V. JABONERO, M. J. GENIS and L. SANTOS, Z. mikrosk.-anat. Forsch. 69, 167 (1962).
- ⁵ V. JABONERO, R. MARTINEZ, F. MARIN-GIRON and R. M. JABONERO, Z. mikrosk.-anat. Forsch. 73, 96 (1965).
- ⁶ V. JABONERO, R. MARTINEZ and M. R. JABONERO, Z. mikrosk.-anat. Forsch. 73, 100 (1965).
- ⁷ V. JABONERO, L. R. PRIETO, A. P. CASAS and M. E. BENGOCHEA, Z. mikrosk.-anat. Forsch. 67, 1 (1961).
- ⁸ W. STACH, Acta histochem. 18, 377 (1964).
- ⁹ J. GOTHE, Z. mikrosk.-anat. Forsch. 72, 383 (1965).
- ¹⁰ M. MAILLET, Z. mikrosk.-anat. Forsch. 70, 397 (1963).
- ¹¹ A. R. CRUZ, Acta anat. 49, 232 (1962).
- ¹² L. STOCKINGER and J. GRAF, Mikroskopie 20, 16 (1965).
- ¹³ R. KOLB, A. FISCHINGER and L. STOCKINGER, Z. mikrosk.-anat. Forsch. 76, 184 (1967).
- ¹⁴ H. G. WIENKER, Z. mikrosk.-anat. Forsch. 76, 70 (1967).
- ¹⁵ We would like to express our sincere gratitude to Prof. DEMAL for his interest and encouragement in this work which has been made possible by the Fonds National des Etudes.

The Cloacal Gland Complex of the Japanese Quail

In the sexually active male Japanese quail (*Coturnix coturnix japonica*) the outer dorsal portion of the cloaca is greatly swollen and red in color, and when the area is pinched a white foamy mass is extruded¹. The foam is evacuated during copulation and defecation. This foam may be associated with the mechanics of fertilization^{1,2} but there is no direct evidence. Only a slight amount of foam can be expressed from the cloaca of laying quail. When adult males are removed from a photoperiod stimulatory for sexual activity and subjected to a non-stimulatory lighting regimen, the amount of cloacal foam produced and the histological complexity of the gland are depressed^{3,4}. Furthermore, no cloacal gland activity was observed when the average weight of the testes was less than 0.75 g⁴.

The cloacal gland complex consists of a series of tubular glands located in the dorsal lip of the cloaca, and empty into the proctodeum^{3,5}. They begin as solid epithelial buds in the dorsal proctodeum between the eleventh and twelfth day of incubation and establish a lumen by the fifteenth day⁵.

Whether these glands are truly cloacal derivative, or members of the anal gland series is questioned⁶.

The indications that these glands are sex-dependent⁴ stimulated a series of experiments. Sexually mature males exposed at least 8 weeks to a photoperiod of 16 h light to 8 h dark (16L:8D), which is stimulatory for both testicular function and cloacal gland activity⁴, were implanted s.c. with a 3 mg pellet of diethylstilbesterol and then killed 10 days post-implantation. Actively laying females on 16L:8D were implanted with 5 mg of crystalline testosterone and killed 10 days later. In another experiment, groups of immature males and females were reared to 4 weeks of age under the limited light condition 8L:16D⁴. Half of each sex were implanted with 5 mg of crystalline testosterone and all were killed 10 days later.

The dorsal lip of the cloaca was removed and processed for histological examination.

Observations and discussion. The cloaca of the Japanese quail differed from that described for the chicken⁷. Columnar epithelium lined only the coprodeum and urodeum. The proctodeum was lined entirely by stratified squamous epithelium, including the entrance into the bursa of Fabricius. The glandular complex was located in the dorsal portion of the proctodeum between the bursa of Fabricius and the anal aperture or vent (Figure 1), and are not the anal glands reported in other birds⁶.

Macroscopically, the active cloacal gland complex measured approximately 10 × 12 × 2 mm. Microscopically, the complex comprised a series of branched tubular glands lined by a simple columnar epithelium. Each had a main central lumen that communicated with the proctodeum through a small pore (Figure 2) and gave rise to a series of tubules or sacs. The complex was divided into lobular units by connective tissue septae rich in elastic fibers.

A small, unreported, ventral glandular complex (Figure 3) was also found.

The active cloacal gland (Figure 4) had tall columnar epithelium with basal nuclei and abundant cytoplasm with variable sized vacuoles. The apices of many cells had numerous microprojections, some being pinched off to enter the lumen (Figure 5). Secretory material was abundant, and both cytoplasm and secretory material

- ¹ K. IKEDA and K. TAJI, Scient. Rep. Matsuyama agric. Coll. 13, 1 (1954).
- ² W. H. COIL and D. K. WETHERBEE, Ohio J. Sci. 59, 268 (1959).
- ³ B. D. SACHS, Science 157, 201 (1957).
- ⁴ W. O. WILSON, H. ABPLANALP and L. ARRINGTON, Poultry Sci. 41, 17 (1962).
- ⁵ C. L. NAGRA, R. K. MEYER and N. BILSTAD, Anat. Rec. 133, 415 (1959).
- ⁶ W. B. QUAY, Auk 84, 379 (1967).
- ⁷ M. L. CALHOUN, *Microscopic Anatomy of the Digestive System of the Chicken* (Iowa State College Press 1954).

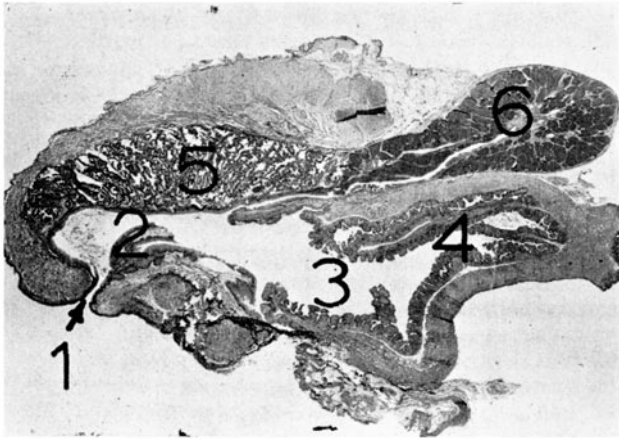


Fig. 1. Sagittal section of cloaca of a male Japanese quail. (1) anus; (2) proctodeum; (3) urodeum; (4) coprodeum; (5) cloacal gland complex; (6) bursa of Fabricius. H and E, $\times 5.8$.

stained intensely with the periodic acid-Schiff reaction, aldehyde fuchsin, mucicarmine and alcian blue, and metachromatically with toluidine blue. Thus indicating the presence of neutral and acidic mucins, glucose, and disulfide bonds; the secretory material was undoubtedly a glycomucoprotein.

The inactive gland had low columnar epithelium, sparse amounts of secretory material, and reduced histological complexity of the gland (Figure 6).

Diethylstilbesterol administration inhibited both cloacal gland and testicular activity. Castration also inhibited cloacal gland activity⁵. The cloacal glands of immature males or immature females on a non-stimulatory light treatment or laying females could be stimulated to activity by testosterone administration.

The secretion expressed directly from the cloacal gland of freshly killed, sexually active males was clear to opalescent and mucoid, and not the white froth seen evacuated by defecation, copulation or manually from the cloaca. The transformation from a viscous mucoid into a froth by the physical capture of air did not seem probable and another explanation was sought. Bacterial isolations from

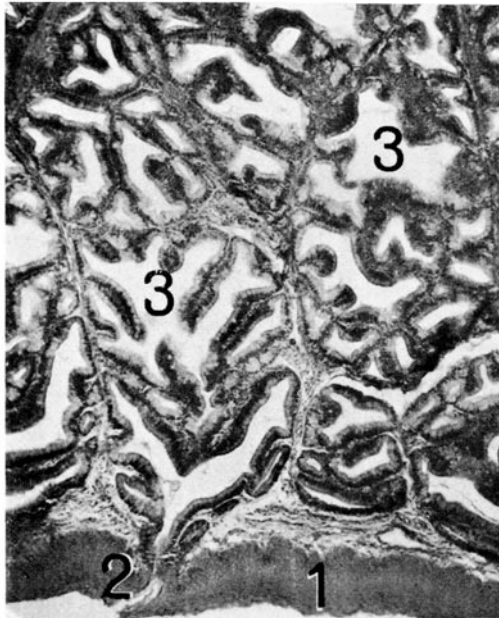


Fig. 2. Sagittal section through cloacal gland complex of a sexually active male quail. (1) stratified squamous epithelium of proctodeum; (2) excretory pore; (3) lobules. H and E, $\times 78$.

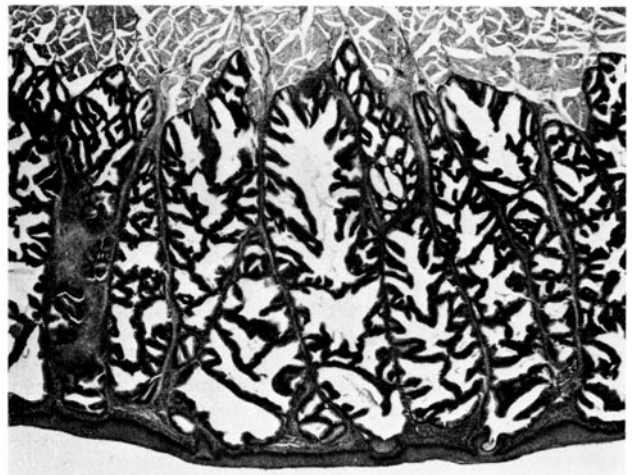


Fig. 4. Sagittal section of an actively secreting cloacal gland of a sexually active male quail. The cloacal gland is 2.0 mm thick. Contrast the histological complexity and size of this active gland to that of the inactive gland in Figure 6. H and E, $\times 29$.



Fig. 3. Sagittal section through ventral proctodeum of a sexually active male quail. (1) proctodeum; (2) stratified squamous epithelium of proctodeum; (3) ventral cloacal glands. Alcian blue-PAS, $\times 267$.

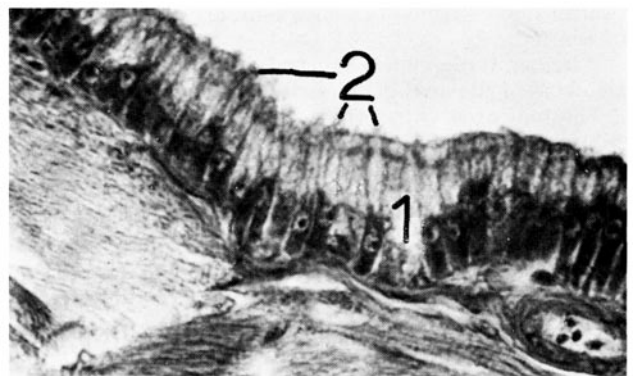


Fig. 5. Secretory epithelium of an active cloacal gland. (1) typical tall columnar cells with denser staining infranuclear regions and lighter staining supranuclear regions; (2) microprojections contributed to the glandular secretion. H and E, $\times 1000$.

the froth yielded equal numbers of *Escherichia coli* and *Proteus mirabilis*. Both organisms are well known for their activity to produce gas from glucose and other sugars. Furthermore *E. coli* may have an outer lipomucoprotein cell wall layer⁸ which would permit a strong attachment to other mucoproteins. Almost equal amounts of carbon dioxide and hydrogen gas are produced from glucose by these bacteria, and the cohesive force of the mucoprotein could prevent the gases from escaping, thus creating the froth.

The pH of the foam varied from 6.3–6.6 with an average of 6.5¹. This pH could result from the generated carbon dioxide and subsequent carbonic acid. Thus, within the observed pH range of the froth, the bicarbonate-carbonic acid equilibrium could serve as an effective buffer⁹.



Fig. 6. Sagittal section of a typical inactive cloacal gland, as seen in sexually immature quail of both sexes, in the sexually mature female or in the sexually mature male kept on a non-stimulatory lighting regimen or implanted with estrogen. The cloacal gland is 0.65 mm thick. H and E, $\times 29$

Résumé. Chez les cailles japonaises, le complexe glandulaire du cloaque est en fait localisé dans la lèvre dorsale du cloaque et non de l'anus. Il existe un complexe glandulaire similaire et très petit du côté ventral. La glande active se colore intensément avec le réactif périodique «acide Schiff», avec la fuchsine aldéhyde, avec le bleu d'alciane et métachromatiquement avec le bleu de toluidine. Ceci indique la présence d'une sécrétion de glycomucoprotéines. Le liquide transparent sécrété est transformé en masse blanche mousseuse au contact des bactéries *E. coli* et *Proteus mirabilis*, présentes en quantités équivalentes dans le proctodeum. Les gaz émis consistent probablement en H₂ et CO₂, ce dernier peut agir comme tampon et stabiliser le pH de la sécrétion autour de la valeur 6,5.

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⁸ P. H. CLARKE and M. D. LILLY, *Nature* 195, 516 (1962).

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Deciduoma Induction in the Rat by X-Irradiation

It is well known that X-irradiation can interrupt pregnancy, since implantation is a radiosensitive response of the female reproductive tract¹.

In the course of experiments planned to study the sensitivity of the decidual response to X-irradiation given on different days after mating, a completely unexpected finding emerged: when a pseudopregnant female was given a sublethal dose of X-rays on the fifth day of pseudopregnancy, deciduomata were found 4 days later at autopsy.

A group of 16 female rats were made pseudopregnant by mating with vasectomized males (the day a vaginal plug was found was designated as day 1 of pseudopregnancy). Half of these animals served as controls, being sham-irradiated. On the fifth day, the other 8 rats were given a local irradiation in the lower left part of the abdomen. The right part of the abdomen and the remainder of the body were shielded with lead $\frac{1}{4}$ inch thick. In this way only 1 of the uterine horns was irradiated. The doses ranged from 500–1000 r, and they were equally effective in the limited number of animals so far studied. The source was a Picker X-ray unit operating at 260 KV. Autopsies were done on the ninth day after mating (4 days after irradiation). The presence of deciduomata was

evaluated macroscopically. In addition the uteri were processed for histology and serially sectioned.

At autopsy none of the control animals presented deciduomata. 4 of the tested animals presented macroscopically visible deciduomata on the left horn and 1 of these presented a deciduoma also on the right horn (not irradiated) near the cervix uteri.

The deciduomata appeared histologically to be composed of normal decidual cells, presenting both mesometrial and antimesometrial characteristics (Figure). The epithelium was degenerating all around the uterine lumen at the site of the deciduoma in the manner typical of decidualization and implantation². The orientation of the deciduoma was generally somewhat disordered, as is frequently observed in all experimentally induced deciduomata. In all the deciduomata obtained by the method described, only the ventral or the dorsal side of the endometrium reacted. The uterine lumen was pushed towards the other side.

¹ M. M. KETCHEL and U. K. BANIK, *Nature* 202, 1021 (1964).

² R. H. KREHEBIEL, *Physiol. Zool.* 10, 212 (1937).