From: Department of Anatomy,
The Royal Dental College,
Aarhus, Denmark.

THE INFRASEPTAL NASAL GLANDS IN MOUSE EMBRYOS WITH DENTAL MALFORMATIONS AND EXENCEPHALY INDUCED BY HYPERVITAMINOSIS A.

by

P. A. KNUDSEN

INTRODUCTION

Infraseptal nasal glands of the mouse are situated in mesenchyme below the septal cartilage of the nose, in front of and between the sockets of developing upper incisor tooth germs (Fig. 1). There is an anterior and a posterior gland on each side of the mid-line opening into the nasal cavity, below and behind the orifice of the nasal lacrimal duct. The glands found in normal mice have been described in detail by *Broman* (1916 a, b, 1921).

During an investigation of fused upper incisors in exencephalic mouse embryos (*Knudsen,* 1965) it was observed that the infraseptal glands exhibited variations in volume and position. Frequently, there seemed to be a considerable reduction of glandular tissue. Furthermore, the shape and size of acini varied. An examination of the glands was undertaken to elucidate some of these changes. The present paper describes the quantitative and morphologic variations of the infraseptal glands in exencephalic mouse embryos with fusion or agenesia of the upper incisors.

TABLE I

The normal and exencephalic embryos studied have been divided into 5 groups.

Number of embryos	Groups	
18	I	normal embryos
18	II	exencephalic embryos with two separate upper incisors
21	III	exencephalic embryos with fusio partialis or fusio subtotalis of the upper incisors
42	IV	exencephalic embryos with fusio totalis of the upper incisors
12	V	exencephalic embryos with bilateral agenesia of the upper incisors

MATERIAL AND METHODS

Exencephalic (93) and normal mouse embryos (18), all included in previous studies (*Knudsen,* 1965 a, b) formed the material of the present investigation. Selection and grouping of the exencephalic embryos was determined on the basis of the incisor anomalies (Table I). Glandular volume was calculated using the measure- and weigh method in the following manner: frontally cut histologic serial sections were projected on the screen of a Reichert Visopan microscope and the outline of the acini drawn on tracing paper (x 70 linear magnification). When section thickness was 7 μ, every 7th section was drawn, and when it was 10 μ, every 5th. From each sheet of paper 100 sq. cm. were cut for control weighing. All acini belonging to the same embryo were drawn on paper from the same sheet, which was cut into smaller pieces, so that each piece contained only acini drawn from one section. The weight of each piece of paper was determined before and after having cut out the acini, and the results recorded on a special form. The glands were divided into a predental and an interdental part and the volume was calculated for each of them. The t-test was used to determine the significance of differences between pairs of mean volumes.

In embryos with fusio dentium, the size of the space between the nasal capsule and the incisor germ was evaluated by measuring the distance in millimetres between the two structures in the mid-line, using the same sections as in volume measurement.

By plotting the values in a system of co-ordinates, a "triangle" was constructed, the area of which was calculated by planimetry.

RESULTS

GROUP I

Few variations in location or extent of the infraseptal glands were seen. Generally, the number of acini increased with increasing glandular volume whilst the size of the individual acini remained unaltered. The interdental part of the glands was enclosed by the alveolar bones of the incisor germs and the nasal capsule (Fig. 1). These structures limited the vertical and transversal extent of the glandular tissue.

The total volume*) of the glands was 57.3 (x 10^{-4}) cu. mm. The volume of the predental part was 6.7 (x 10^{-4}) cu. mm. (Table II). Variations in total volume were mainly due to variations in the interdental part of the glands (Text Fig. I).

GROUP II

This group comprised exencephalic embryos where the distance between the upper incisor germs was the same as in normal

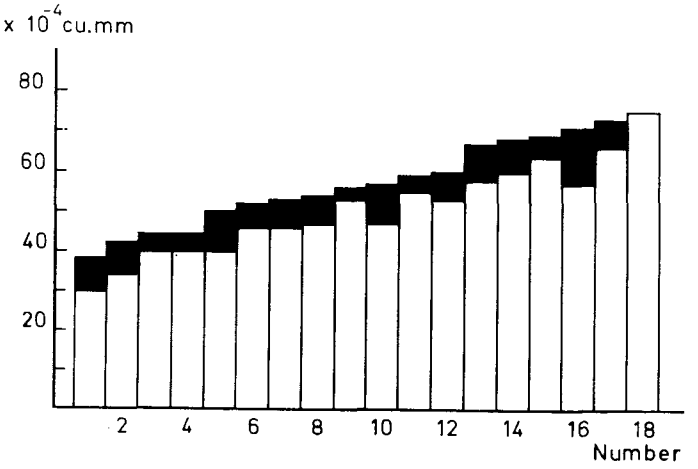


Fig. I. Volume of the infraseptal nasal glands in 18 normal mouse embryos (18 days). Black: predental glands. Unshaded: interdental glands.

*) Mean volume is everywhere implied in the text.

TABLE II

Mean volume (x 10 -4) in cu. mm of the infraseptal nasal glands in mouse embryos.
n = number, sd = standard deviation, m = mean.

Groups Embryos	n	Predental part m±sd	Interdental part m±sd	Total m±sd
I normal	18	6.7± 3.2	50.6±11.7	57.3±11.3
II exencephalic with separate incisors	18	6.4± 4.1	32.9±12.2	39.3±12.6
III exencephalic with fusio subtotalis and fusio partialis	21	6.4± 5.3	24.7±15.3	31.2±13.7
IV exencephalic with fusio totalis	42	11.1± 9.2	15.5±12.0	27.1±11.4
V exencephalic with agenesia	12	25.5±10.7		25.5±10.7

embryos. The glands had a relation to the nasal capsule and the incisor germs similar to group I (Fig. 2). The total volume was 39.3 (x 10-4) cu. mm, which was significantly less than in normal embryos ($P<0.001$). The predental glandular volume was almost unchanged, whereas the volume of the interdental part was strongly reduced (Text Fig. II). The number, shape and size of the acini were similar to those of normal embryos with similar glandular volume.

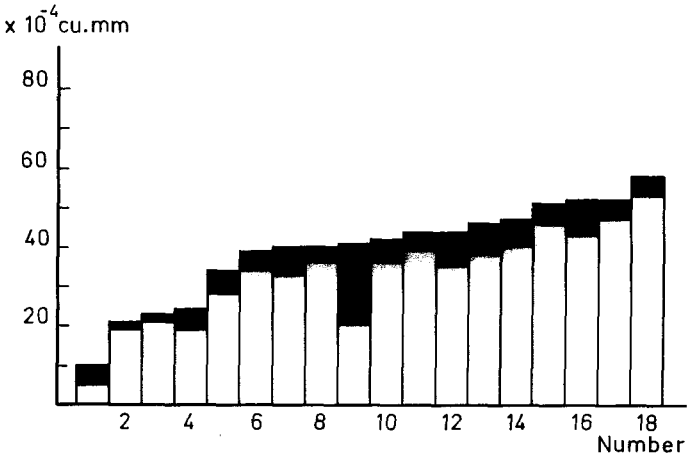


Fig. II. Volume of the infraseptal nasal glands in 18 exencephalic mouse embryos (18 days) with separate upper incisor germs. Black: predental glands. Unshaded: interdental glands.

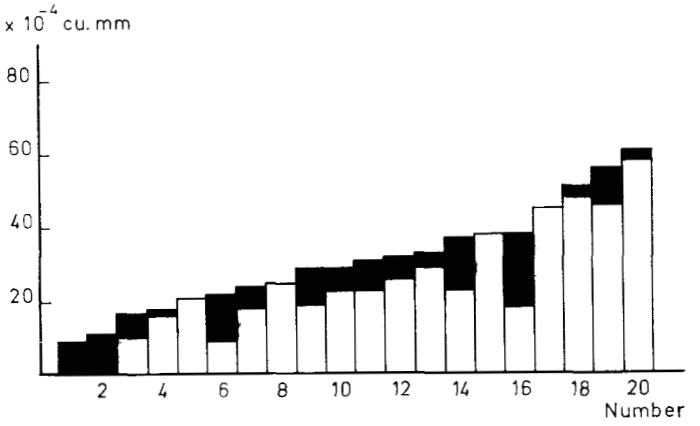


Fig. III. Volume of the infraseptal nasal glands in 20 exencephalic mouse embryos (18 days) with fusio subtotalis and partialis of the upper incisor germs. Black: predental glands. Unshaded: interdental glands.

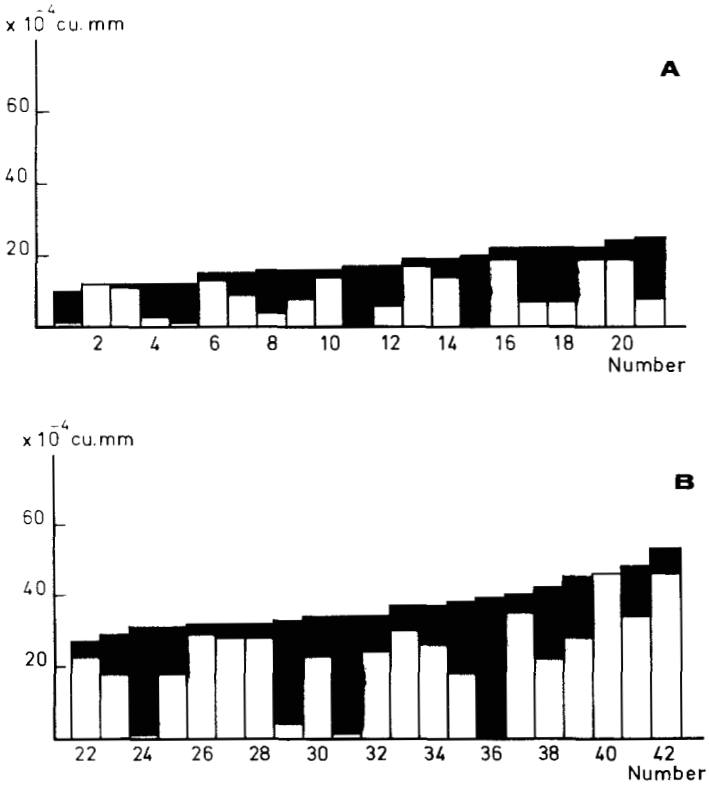


Fig. IV. In parts A + B the volume of the infraseptal nasal glands in 42 exencephalic mouse embryos (18 days) with fusio totalis of the upper incisor germs. Black: predental glands. White: interdental glands.

GROUP III

In this group of embryos, the fused incisor germs crossed the mid-line, thereby occupying, together with the alveolar bone, part of the space below the nasal capsule (Fig. 4). The total volume of the glands was 31.2 (x 10^{-4}) cu. mm (Text Fig. III); it was not significantly different from the total volume in exencephalic embryos with two incisors ($0.05<P<0.1$). In groups III and IV there was an occasional occurrence of glandular tissue behind the incisors. As the amount was inconspicuous it was included in the interdental portion.

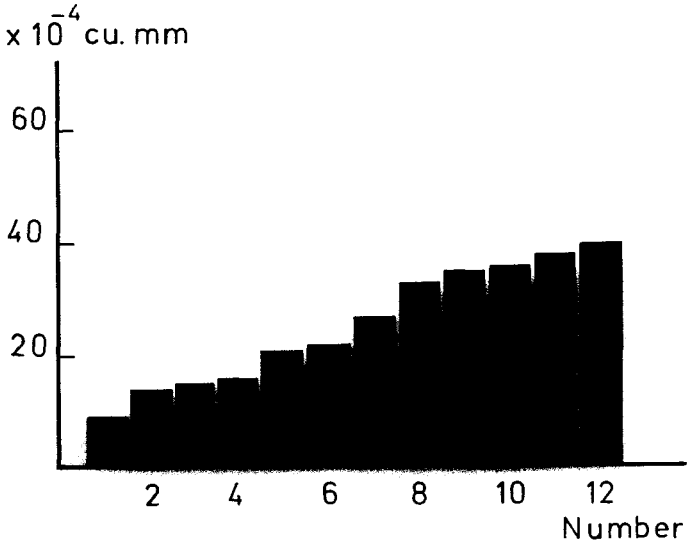


Fig. V. Volume of the infraseptal nasal glands in 12 exencephalic mouse embryos with agenesia of the upper incisor germs. The glands are supposed to be placed in the "predental" region.

GROUP IV

Exencephalic embryos with fusio totalis comprised the largest of the groups examined. The shape of the tooth germs varied, but

all germs crossed the mid-line, occupying, together with the alveolar bone, a considerably greater portion of the infraseptal space than in the preceding group (Figs. 3 and 5). The total volume of the glands is 27.1 (x 10^{-4}) cu. mm; it was significantly less than the total volume in group II ($0.001<P<0.01$). The average volume of the predental part, which was of the same order of magnitude in the preceding groups, was now almost doubled, being 11.1 (x 10^{-4}) cu. mm, i. e. on average 41 % of the glandular volume was located in the predental region (Text Fig. IV).

The predental part of the glands increased with decreasing infraseptal space as shown in Text Fig. VII.

Besides the considerable change in the relative size of the two parts of the gland there was also a change in the appearance of the acini, in particular of the predental part, where many of the acini were larger and more irregular than in the other groups (Text Fig. VI, middle). The large acini extended into the interdental part, where they were gradually replaced by small and more uniform acini; further back, only one or two were seen in each section.

GROUP V

This group comprised exencephalic embryos with bilateral agenesia of the upper incisors, whilst the number of molar germs was normal. The total glandular volume was 25.5 (x 10^{-4}) cu. mm (Text Fig. V), and not significantly different from the total volume in embryos with fusio totalis ($P>0.1$). Most of the acini were large and thus resembled those in the predental part of the glands in embryos with fusio totalis (Text Fig. VI (right).

In normal embryos, as well as in exencephalic embryos with separate incisors and fusio totalis, the incisor germs started opposite that part of the nasal capsule where the floor plate (anterior transversal lamina) loosed its connection with the lateral walls. In bilateral agenesia of the upper incisors, the entire glandular mass lied in front of this spot and thus corresponded in position to the predental part of the glands in embryos with normal or fused incisor germs. Immediately behind the last acini the nasal capsule was covered by a bone mantle, almost filling the space where the glands were anticipated (Fig. 6).

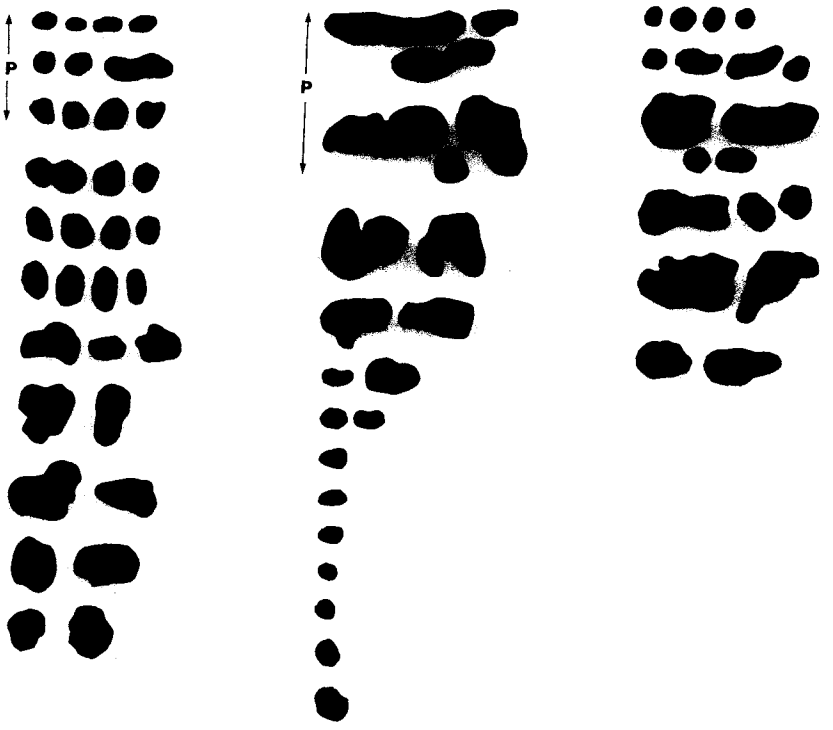


Fig. VI. The outline and relative size of representative acini of the infraseptal nasal glands in a normal mouse embryo (left), in an exencephalic embryo with fusio totalis of the upper incisor germs (middle) and in an exencephalic embryo with agenesia of the upper incisor germs (right).
P = predental part of the glands.

DISCUSSION

When the incisor germs are fused or lacking, the amount and distribution of mesenchyme, bone and glands in the incisor region are largely affected. It is difficult, however, to evaluate the changes of mesenchyme and bone by exact measurement, but due to the fact that the identification of the infraseptal glands is easy and the volume determination rather simple, it is possible to recognize even small changes in distribution and volume of the glandular tissue, as well as in the size and shape of the acini.

It was anticipated that the total glandular volume would be reduced in fusio dentium, as the usual position of the glands about the mid-line is infringed upon by the tooth germs. The

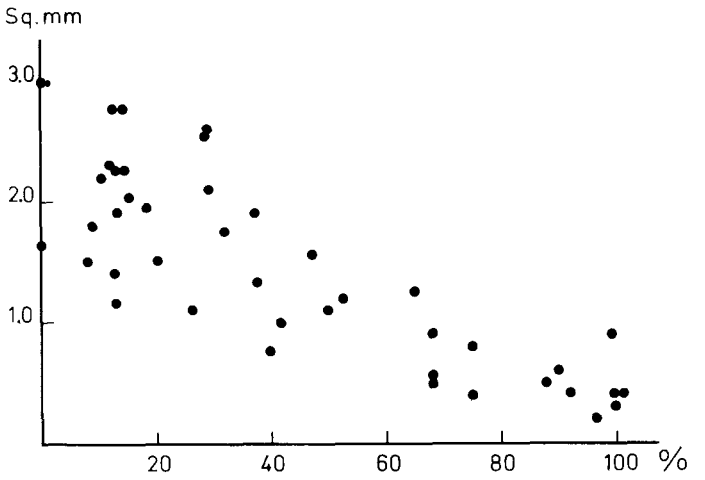


Fig. VII. The size of the mesenchyme space between the nasal septum and the upper incisor germ in case of fusio totalis is expressed by the area of a triangle constructed in the mid-line (ordinate). The amount (in per cent) of the infraseptal nasal glands located in the predental region (abscissa).

present investigation has clearly demonstrated, however, that the total volume of the glands is *always* reduced in exencephaly. It appears too, that there is no significant difference in the total volume of the glands in exencephalic embryos with normal and in those with partly fused incisor germs. The difference is only significant when fusion of the germs is complete. The cause of the glandular reduction is unknown, and connection between the volume of the glands and vitamin A dosage could not be found. It is possible that the reduction is part of a general restrictive action on embryonic development due to the severe brain malformation.

The infraseptal space is strongly reduced in embryos with fusio totalis, owing to the location and shape of the tooth germ. In embryos lacking both incisor germs the space is reduced, on account of a thick layer of bone beneath the nasal capsule. In both cases the glandular tissue has developed in the ample predental mesenchyme where the growth of the glandular tissue is supposed to be less restrained by mechanical factors, than in the narrow infraseptal space, partly bounded by rigid walls. It has not been possible to determine whether the development of large

acini in the predental region is also favoured by mechanical reasons or whether the presence of these particular structures is due to entirely different causes.

SUMMARY

In exencephalic mouse embryos the volume of the infraseptal glands is significantly reduced even when the incisor germs are normal. The greatest reduction is found in embryos with fusio totalis and agenesia. The predental part of the glands dominates in fusio totalis, where the room left for the interdental part is strongly reduced. In embryos with agenesia the glands are located in the anterior part of the infraseptal mesenchyme, whereas the posterior part contains bone tissue. The largest acini are found in the predental area in embryos with fusio totalis.

Acknowledgement:

The author wishes to thank Mr. Kaj Josephsen, stud. odont., for his painstaking and enthusiastic assistance during all the stages of this investigation.

RÉSUMÉ

LES GLANDES NASALES INFRA-SEPTALES CHEZ LES EMBRYONS DE SOURIS EXENCÉPHALES PAR SUITE D'HYPERVITAMINOSE A ET PRÉSENTANT DES MALFORMATIONS DENTAIRES

Chez les embryons de souris exencéphales, le volume des glandes infra-septales est réduit de manière significative, même lorsque les germes des incisives sont normaux. La réduction la plus importante a été trouvée chez les embryons présentant une fusion totale et une agénésie. La partie prédentaire des glandes domine dans la fusion totale, où l'espace restant pour la partie interdentaire est fortement réduit. Chez les embryons présentant une agénésie, les glandes sont localisées à la partie antérieure du mésenchyme infra-septal, tandis que la partie postérieure contient du tissu osseux. Les acinus les plus grands se trouvent dans la zone prédentaire chez les embryons présentant une fusion totale.

ZUSAMMENFASSUNG

DIE INFRASEPTALDRÜSEN BEI MÄUSEEMBRYONEN MIT ZAHNMISSBILDUNGEN UND EXENCEPHALIE DURCH ÜBERDOSIERUNG MIT A-VITAMIN VERURSACHT

Bei exencephalen Mäuseembryonen ist das Volumen der Infraseptaldrüsen (Glandulae nasales infraseptales) bedeutend reduziert, auch in den Fällen wo die Schneidezahnkeime normal sind. Die grösste Reduktion findet man bei Embryonen mit fusio totalis und Agenesie. Der prädentale Teil der Drüsen dominiert bei fusio totalis, wo der Raum des interdentalen Teiles bedeutend reduziert ist. Bei Embryonen mit Agenesie sind die Drüsen im anterioren Teil des infraseptalen Mesenchyms zu finden, während der posteriore Teil Knochengewebe enthält. Die grössten Endstücken befinden sich in dem prädentalen Gebiet bei Embryonen mit fusio totalis.

REFERENCES

Broman, I., 1916 a: Über eine bisher unbekannte infraseptale Nasenhöhlendrüse bei den Nagern. Vorläufige Mitteilung. Anat. Anz. *49:* 170.

Broman, I., 1916 b: Om gnagarnas Glandula nasalis infraseptalis och dess utveckling. Svenska Läk.-Sällsk. Handl. *42:* 717—736.

Broman, I., 1921: Über die Entwickelung der konstanten grösseren Nasenhöhlendrüsen der Nagetiere. Z. Anat. Entwickl.-Gesch. *60:* 439—586.

Knudsen, P. A., 1965 a: Congenital malformations of upper incisors in exencephalic mouse embryos induced by hypervitaminosis A. I. Types and frequency. Acta odont. scand. *23:* 71—89.

Knudsen, P. A., 1965 b: Congenital malformations of upper incisors in exencephalic mouse embryos induced by hypervitaminosis A. II. Morphology of fused upper incisors. Acta odont. scand. *23:* 391—409.

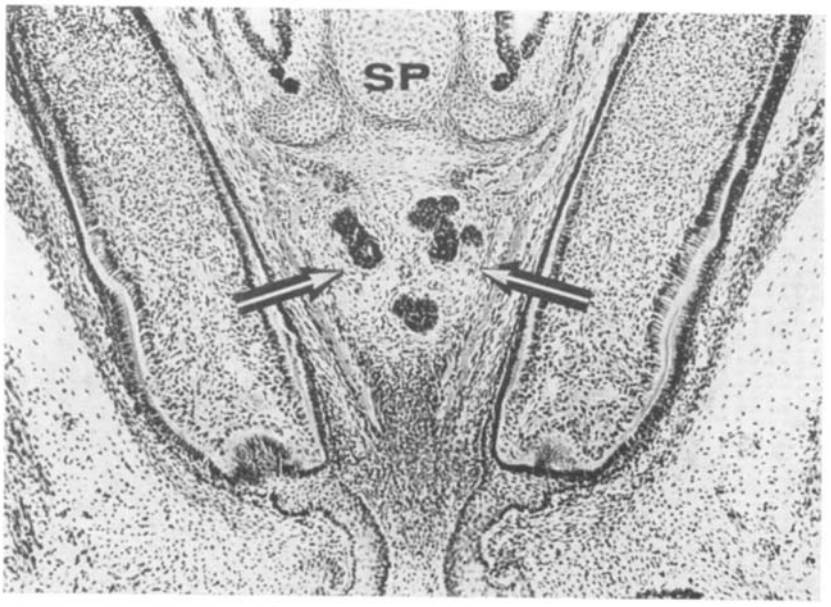
Address: *Aarhus Tandlægehøjskole,*
Vennelyst Boulevard,
Aarhus C, Denmark

PLATES

PLATE 1.

Fig. 1. Normal mouse embryo (26 d III/4). Frontal section. The infraseptal nasal glands (arrows) in mesenchyme between upper incisor germs (x 63). SP = septal cartilage.

Fig. 2. Exencephalic mouse embryo with separate incisor germs (234 d I/3). Sagittal section between upper incisor germs. Mesenchyme with infraseptal nasal glands. Note the extent of the glands (x 63). (Labial side to the left). SP = septal cartilage.


SP

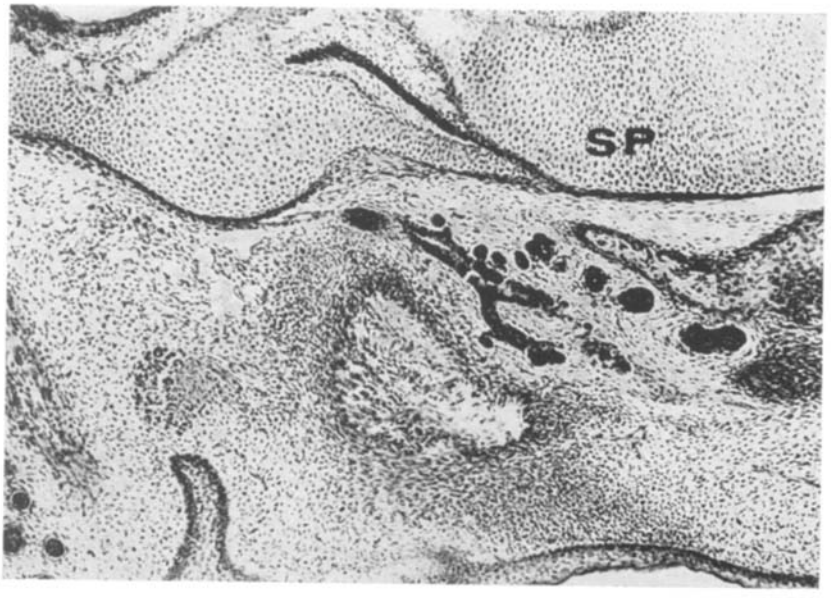

SP

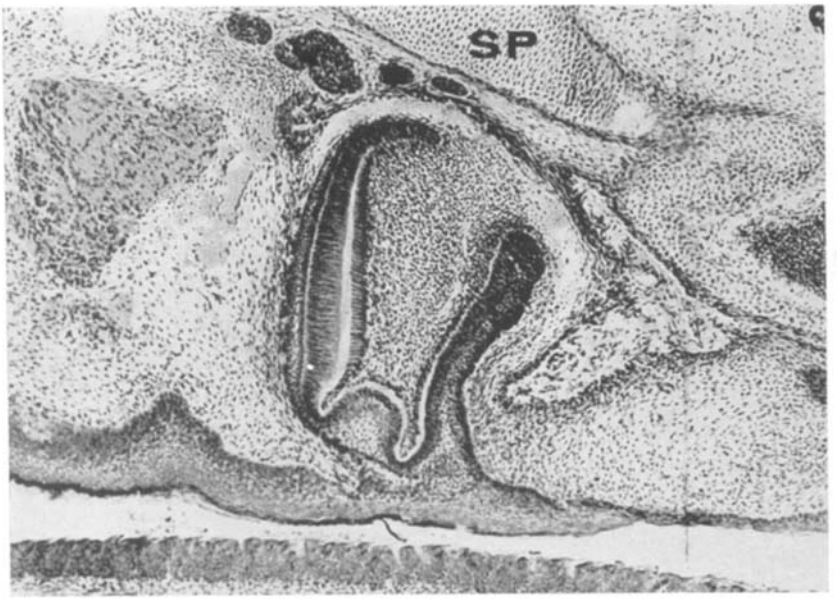
PLATE 2.

Fig. 3. Exencephalic mouse embryo (226 d I/3) with fusion of upper incisor germs. Sagittal section. Note the extent and size of the predental part of the infraseptal nasal glands and the limited space between the tooth germ and the septal cartilage of the nasal cavity (x 63).

SP = septal cartilage.

Fig. 4. Exencephalic mouse embryo with fusio partialis of upper incisor germs (106 d I/2). Frontal section. Note the narrow space available for the infraseptal nasal glands; compare with Fig. 1 (x 63).

SP = septal cartilage.


SP

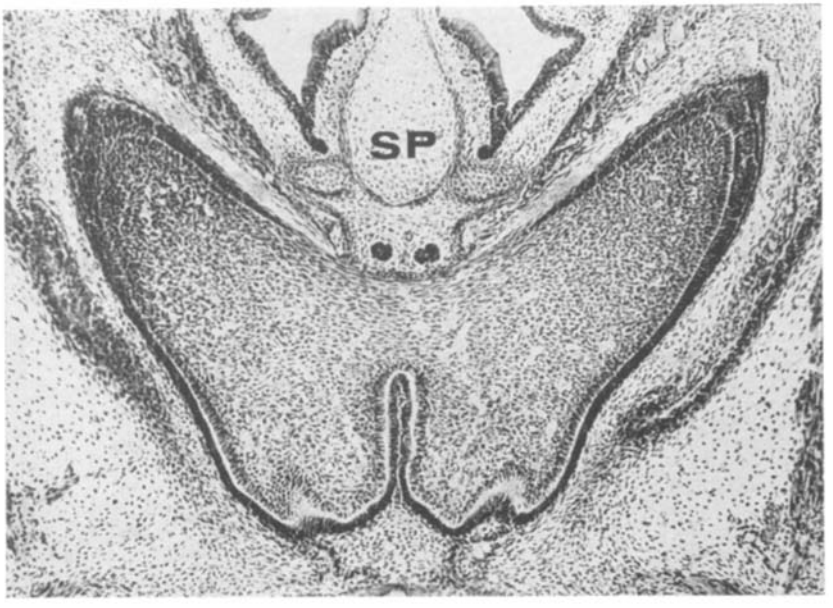

SP

PLATE 3.

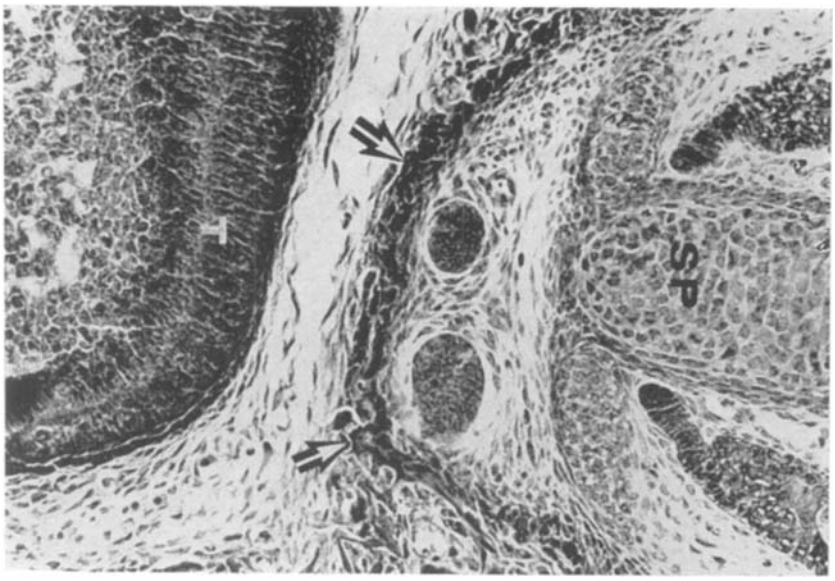
Fig. 5. Exencephalic mouse embryo with fusio totalis of upper incisor germs (70 d I/3). Frontal section of the labial part of the tooth germ. The infraseptal nasal glands and mesenchyme are separated from the incisor germ by bone (arrows). Note the narrow space available for the glands (x 160).

T = tooth germ.

SP = septal cartilage.

Fig. 6. Exencephalic mouse embryo with agenesia (agenesis) of both upper incisor germs (211 d I/2). Frontal section. A bone mantle (arrows) occupies the area where the infraseptal nasal glands are anticipated (x 63).

E = oral epithelium.


SP