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# Topographical variations of the incisive canal and nasopalatine duct in human fetuses

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**Abstract:** The incisive canal for nerves and vessels is generally thought to run along a suture between the incisive bone (IN) and maxilla. In contrast, there was a report saying the canal passes through the IN or primary palate in human fetuses. Examination of sagittal and frontal sections from 69 fetuses (31 of gestational age [GA] 9–15 weeks and 38 of GA 26–34 weeks) showed that the canal often penetrated the IN at the nasal half of its course and that, in other fetuses, the canal penetrated the IN along its entire course, irrespective of involvement of the nasopalatine duct. Canals developing in and corresponding to parts of the suture resulted in partial enlargement of the thin and tight sutures, which contained loose tissue, vessels, nerves and even a duct. Small processes of the IN were identified as upper irregular parts continuous with inferior main masses of bone in frontal sections but as bone fragments in sagittal sections. In some sections, a thin layer of the maxilla along the canal covered the medial or inferior aspect of the IN. Therefore, the incisive canal with or without duct exhibited a spectrum of variations in topographical relation to the IN-maxillary border. Because the primitive oronasal communication passes through the suture, the nasopalatine duct may have originated from the secondary developed elongation of the nasal epithelium at midterm. A large incisive fossa along the midline on the oral surface of the palate might make a macroscopic finding of variants difficult even in adults.

**Key words:** Incisive canal, Nasopalatine duct, Nasopalatine nerve, Maxilla, Human fetus

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## Introduction

In adults, the incisive canal is a narrow bony canal connecting the nasal cavity and incisive fossa, allowing passage of the nasopalatine nerve, a branch of the greater palatine artery and, in some individuals, a remnant of the nasopalatine duct [1]. The incisive canal is generally regarded as passing through the border between the primary and secondary palates, i.e., along the incisive-maxillary or transverse suture of the bony palate [2-4]. Gray's Anatomy [1] also described the course along the bony suture. The bony canal in adults consistently opens to an incisive fossa at a cross point of the transverse and longitudinal sutures of the bony palate. Moreover, examination of sagittal sections showed the presence of the duct, nerves and vessels through a canal along the suture [3, 5-7]. Therefore, the canal is usually considered to pass through a border between the primary and secondary palates, while the duct is commonly known as a derivative from a primary oronasal communication. However, Vacher et al. [3] demonstrated that, in sagittal sections of a single fetus at 14 weeks, the canal passes through the incisive bone (IN) or premaxilla. Moreover, Radlanski et al. [8] postulated a new concept that the canal develops within the embryonic IN or primary palate according to a relatively small number of spec-

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imens after palatal closure (six fetuses at approximately 10–27 weeks). Could such a limited observation change one of essential knowledges of embryology? Otherwise, are there individual and/or age-related variations present in topographical relation between the incisive canal and suture?

Using frontal sections of human midterm fetuses, our group recently demonstrated that the bony canal penetrates the IN, not along the IN-maxillary suture, in fetuses "with" a persistent nasopalatine duct [9]. It is unclear, however, whether the intra-IN course of the duct corresponds to a bony variation or anomaly. Is the intra-IN canal likely when the fetuses had no epithelial duct? In contrast to our previous study using frontal sections [9], comprehensive studies of the bony palate (e.g., Wood and Kraus [10]) were almost always based on observations of sagittal sections. However, since a single sectional plane chosen is likely to make an identification of thin bony structures difficult, we need to compare results from multiple sectional planes. Consequently, in spite of the long history of research, we should emphasize that anatomy of the incisive canal and duct is still debatable. This study therefore assessed the topographical anatomy of the incisive canal and the IN-maxillary suture using a large number of midterm and late-stage fetuses with or without a persistent nasopalatine duct.

## Materials and Methods

This study was performed in accordance with the principles of the Declaration of Helsinki 1995 (as revised in 2013). Serial or semiserial sections were obtained from 69 fetuses, including 31 specimens at gestational age (GA) 9–15 weeks and crown rump length (CRL) 36–110 mm and 38 of GA 26–34 weeks and CRL 210–290 mm. The sectional planes were sagittal in 32 specimens (nine midterm and 23 late-stage) and frontal in 37 specimens (22 midterm and 15 late-stage). In the world, to our knowledge, there is no collection of histological sections from abundant fetuses at 20-25 weeks or CRL 140–200 mm.

The 31 specimens of midterm fetuses were part of the large collection kept at the Embryology Institute of the Universidad Complutense, Madrid, and originated from miscarriages and ectopic pregnancies at the Department of Obstetrics of the University. All specimens were sectioned serially (frontal or sagittal) (Table 1). Most sections were stained with hematoxylin and eosin, with others stained with Masson trichrome, azan, orange G, or silver stain. The study protocol was approved by the ethics committee of the Universidad Complutense (B08/374). All midterm specimens were of Spanish white origin.

**Table 1.** Sites of incisive canals in frontal and sagittal sections of mid-term and late-term fetuses

| | No. | Frontal | | Sagittal | | Total | |
|---|---|---|---|---|---|---|---|
| | | IM suture[a] | Intra-incisive[b] | IM suture[a] | Intra-incisive[b] | IM suture[a] | Intra-incisive[b] |
| Midterm | 31 | 9 | 13 | 4 | 5 | 13 | 18 |
| Late stage | 38 | 4 | 11 | 6 | 17 | 10 | 28 |
| Total | 69 | 13 | 24 | 10 | 22 | 23 | 46 |

IM, incisive-maxillary suture. [a]Canal along the incisive-maxillary suture. [b]Intra-incisive canal (partly or entirely).

The 38 specimens of late-stage fetuses were parts of the collection of the Department of Anatomy, Akita University, Akita, Japan. These specimens had been donated by their families to the Department from 1975–1985 and preserved in 10% (w/w) neutral formalin solution for more than 30 years. The available data were limited to the date of donation and GA, but there was no information on family name, the name of the obstetrician or hospital and the reason for abortion. All late-stage specimens were of Far-East yellow origin, most likely of Japanese. The use of these specimens for research was approved by the ethics committee of Akita University (No. 1428). After removal of the palate in combination with its surrounding tissues including lower parts of the nose and the pterygoid of the sphenoid, the specimens were decalcified by incubating them at room temperature in Plank-Rychlo solution ($AlCl_2/6H_2O$, 7.0 w/v%; HCl, 3.6; HCOOH, 4.6) for 1–2 weeks. After routine procedure for paraffin embedded histology, frontal or sagittal semiserial sections (numbers of specimen) (Table 1), 5-μm-thick at 50-μm intervals, were prepared from each block and stained with hematoxylin and eosin. All observations and photographs were made with a Nikon Eclipse 80 camera (Nikon, Tokyo, Japan).

## Results

Figs. 1–4 display bony palates from midterm fetuses, while Figs. 5–7 show bony palates from late-stage fetuses, with frontal sections shown in Figs. 1–3 and 5 and sagittal sections in Figs. 4, 6, and 7. Frontal sections, of which posterior site is always shown in the top panel, were better at showing the superomedial processes of the IN. Likewise, frontal sections were able to show a fact that an essential left/right difference was absent in the present fetuses for them. Although Figs. 3E and 5F might make an impression of the left-right difference,


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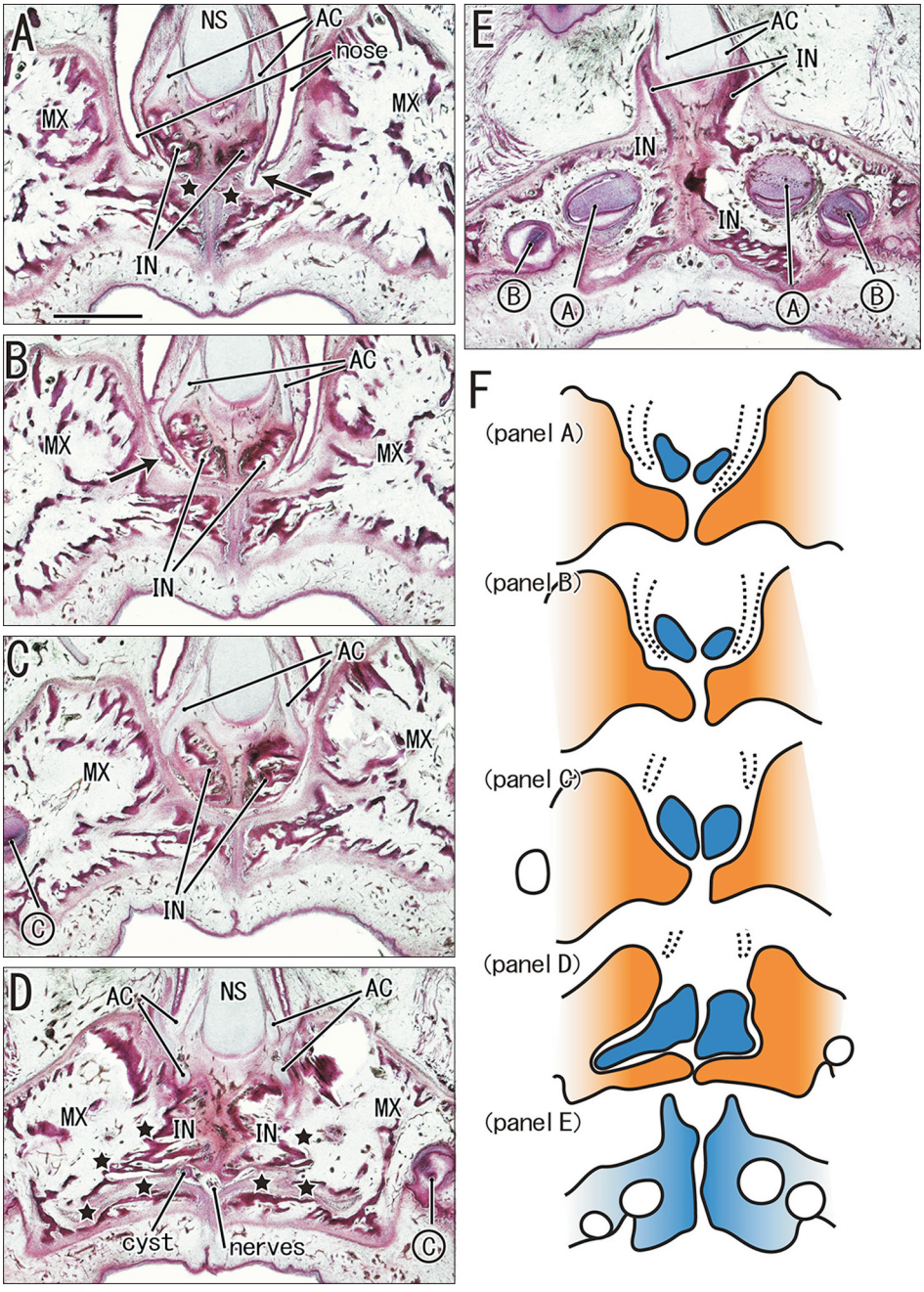


**Fig. 1.** An incisive canal passing along the suture between the incisive bone and the maxilla in a midterm fetus. Hematoxylin and eosin staining. Frontal sections of a specimen of crown rump length 84 mm. Panels A–E were prepared at the same magnification. Panel A shows the most posterior site in the figure. Panel F schematically demonstrates the topographical relationship between the incisive bone (blue) and the maxilla (orange), with the dotted lines showing a paired inferior elongation of the nasal epithelium. The arrows in panels A and B indicate the nasal epithelial elongation. A duct was absent. Stars indicate the incisive-maxillary suture distant from the canal. Panels B and C show dilation of the suture corresponding to the canal. Scale bar=1 mm. A, B, or C in circles indicate a tooth bud of the medial incisor, lateral incisor, or canine, respectively. AC, alar cartilages of the anterior nose; MX, maxilla; NS, nasal septum; IN, incisive bone.

a same bony structure was seen in different anteroposterior levels (e.g., Fig. 5D vs. Fig. 5F). Below, we will describe three types of the incisive canal or possible individual variations: a correspondence between a figure and the type is summarized in Table 2.

### *Superomedial processes of the IN*

In frontal sections, the bony processes were consistently paired and attached to each other across a narrow midline suture below the vomer. Thus, midline sutures of bilateral INs were observed not only in the lower large mass near the oral cavity but also between the upper processes: both were likely to be included in a single frontal section (Figs. 2C, 3E, 5I). These processes were adjacent to anterior (alar) nasal cartilages at anterior sites (Figs. 1B, 2C, 3D) as well as to the vomers at posterior sites (Figs. 3A, B, 5C, F, I). At anterior sites, the INs had generated plate-like processes similar to the paired processes of the vomer sandwiching the nasal septum

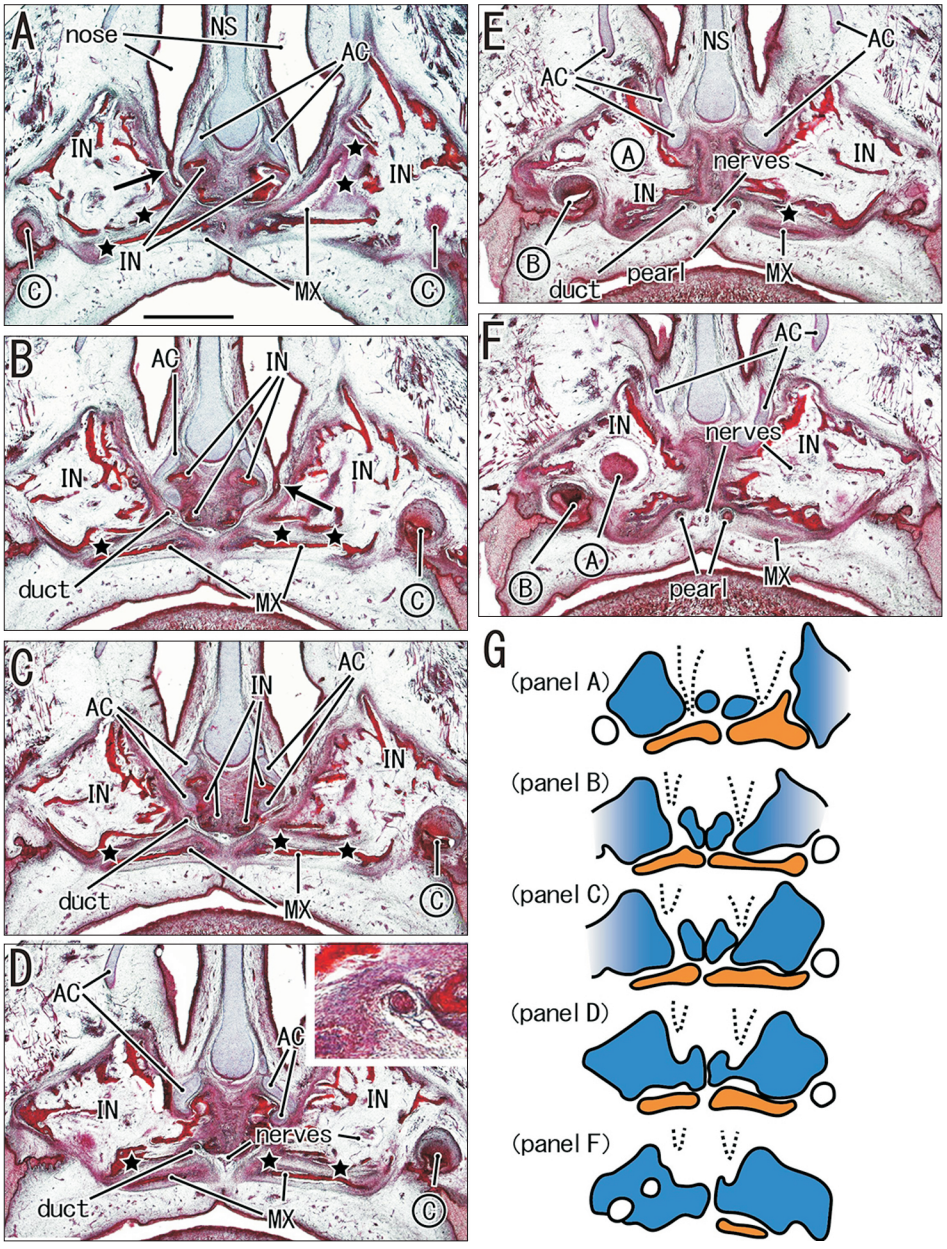


**Fig. 2.** Presence of an intra-incisive canal in the nasal half, with the oral half appearing to pass through the incisive-maxillary suture in a midterm fetus. Hematoxylin and eosin staining. Frontal sections of a specimen of crown rump length 77 mm. Panels A–F were prepared at the same magnification. Panel A shows the most posterior site in the figure. Panel G schematically demonstrates the topographical relationship between the incisive bone (blue) and the maxilla (orange), with the dotted lines showing a paired inferior elongation of the nasal epithelium. Panels A and B show that paired loose spaces or canals penetrate the incisive bone (IN), followed by inferior elongation of the nasal epithelium (arrows). However, the lower half of the incisive canal is sandwiched by the incisive bone and maxilla (MX). Stars indicate the incisive-maxillary suture distant from the canal. The epithelial elongation continues to a thin duct (inset at the right-upper in panel D) and connects with an epithelial pearl in the incisive fossa. Scale bar=1 mm. A, B, or C in circles indicate a tooth bud of the medial incisor, lateral incisor, or canine, respectively. AC, alar cartilages of the anterior nose; NS, nasal septum; pearl, palatal epithelial pearl.

(Figs. 1E, 5I). Likewise, the maxilla was also likely to issue a plate-like anterior part lining the inferior or medial aspect of incisive canal (IC) (Figs. 2E, F, 5B, F, I). These process and plate provided a complicated anatomy of a border or suture between the IN and maxilla.

In sagittal sections, the aforementioned superomedial process of the IN was sometimes identified as a single plate (Fig. 4B) but often identified as fragments (Figs. 4F, 7A), making identification of the IN-maxillary suture more difficult than in frontal sections. Moreover, the apparent branching or joining of the suture and/or canal in sagittal sections (Figs. 4F, 7B) also made understanding difficult, inasmuch as the suture branches were consistently located on the posterior side of the incisive canal.

### *Incisive-maxillary suture and the incisive canal*

At sites distant from the incisive canal, the IN-maxillary sutures were narrow and tight and contained no or few vessels (stars in Figs. 2B, 3B, 4A, E, 5A, D, G, 6A, 7A). Sutures along the lateral aspect of the IN were difficult to identify because they consisted only of loose mesenchymal tissue without trabeculae (stars in Figs. 1D, 2A right, 3D right). In contrast,


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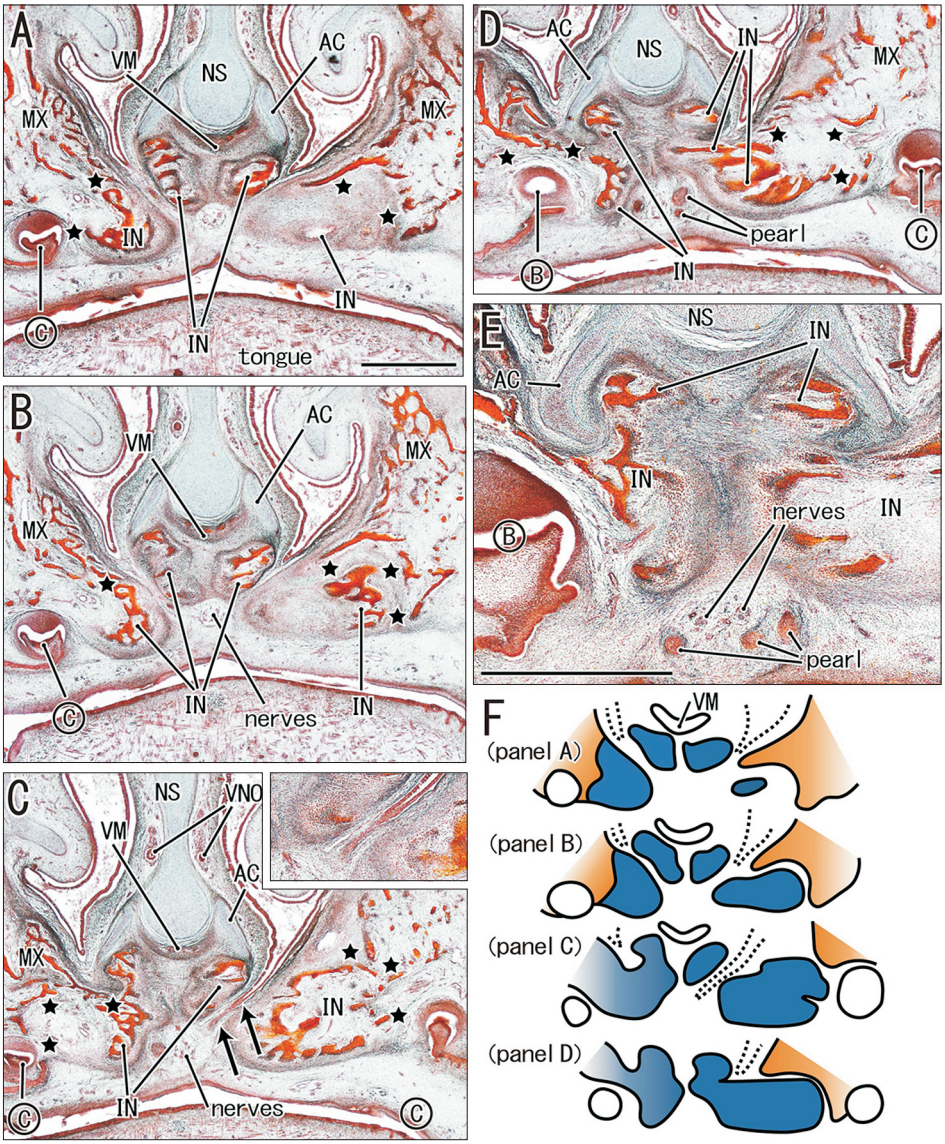


**Fig. 3.** A midterm fetus, in which almost the entire course of the canal takes an intra-incisive course. Hematoxylin and eosin staining. Frontal sections of a specimen of crown rump length 70 mm. Panels A–E were prepared at the same magnification. Panel A shows the most posterior site in the figure. Panel F schematically demonstrates the topographical relationship between the incisive bone (blue) and the maxilla (orange), with the dotted lines showing a paired inferior elongation of the nasal epithelium. Panel C displays that the epithelial elongation (arrows) is sandwiched by parts of the incisive bone (IN). Panel E exhibits candidate nasopalatine nerves (nerves) as well as a tight tissue in the midline suture. Stars indicate the incisive-maxillary suture distant from the canal, although the lateral part is unclear in panels A, C, and D. Scale bar=1 mm. B or C in circles indicate a tooth bud of the lateral incisor, or canine, respectively. AC, alar cartilages or the anterior nose; MX, maxilla; NS, nasal septum; pearl, palatal epithelial pearl; VM, vomer; VNO, vomeronasal organ.

the incisive canal was always thick and contained vessel-rich loose tissue continuous with a submucosal layer of the nose, a histological similarity easily identifying the incisive canal, irrespective of whether the section included an elongation of nasal epithelium (see below). Frontal sections, with a wide view of the nasal cavity, clearly demonstrated the continuation of tissue from the nose to the canal (Figs. 1C, 2A, 3B, 5B, F, H). In contrast, sagittal sections along and near the midline showed that the vomer and nasal septum cartilage occupied a large area above the upper opening of the incisive canal (Figs. 4D, 7A). Frontal sections of the course almost along the left-right axis also clearly showed the inferomedial elongation of the nasal epithelium entering the incisive canal (Figs. 1A, B, 2A, 3A, 5B, F, H). This epithelial elongation sometimes reached an epithelial pearl in the incisive fossa (Figs. 2E, 4G, 5C), a morphology defined as being a real nasopalatine duct [9, 11]. This morphology was observed in 22 fetuses of CRL >70 mm or GA 11 weeks.

### *Intra-incisive canal and the canal along the suture*

The development of the incisive canal, corresponding to the IN-maxillary suture, resulted in partial enlargement of the thin and tight sutures, which contained vessel-rich loose tissue in 23 of the 69 fetuses (33.3%) (Figs. 1B, 4C, F, 5B, 7B). Along the canal, a thin layer of the maxilla sometimes covered the medial aspect of the IN (Fig. 5B, C). However, other than the maxilla, the incisive canal in 46 fetuses (66.7%) was sandwiched partly or entirely by the upper process and the lower

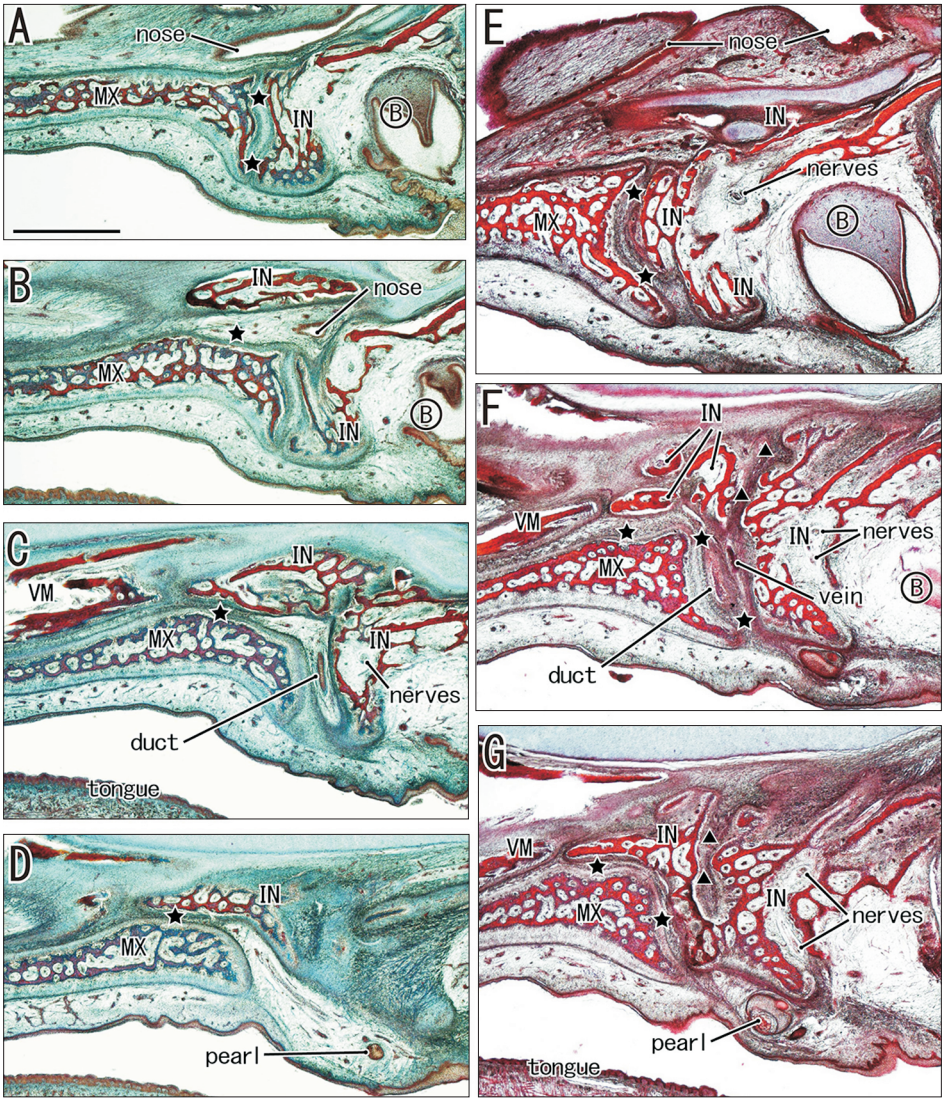


**Fig. 4.** Incisive canal and the incisive-maxillary suture in sagittal sections of two midterm fetuses with ducts. Hematoxylin and eosin staining. Panels A–D display a specimen of crown rump length (CRL) 82 mm, and panels E–G exhibit a specimen of CRL 100 mm. All panels were prepared at the same magnification. Panels A and E show the most lateral sites in each of the specimens. Panels D and G exhibit the presence of an epithelial pearl in the incisive fossa of both specimens. The smaller specimen showed a simple morphology, in which the suture (stars) was partly dilated to contain a duct (C). The larger specimen showed a complicated morphology, in which the suture (stars) appears to merge with an intra-incisive canal (triangles) in panel F. In panel G, however, the intra-incisive canal (anterior; triangles) and the suture (posterior; stars) are separated by a process of the incisive bone (IN). This morphology looks like a branching of the suture. Scale bar=1 mm. B in circles indicates a tooth bud of the lateral incisor. MX, maxilla; pearl, palatal epithelial pearl; VM, vomer.

mass of the IN. In 35 of these fetuses, the canal penetrated the IN at the nasal half of the course (Figs. 2B, C, 5E, F, 7A, B), whereas, in the other 11, the canal penetrated the IN along the entire course (Figs. 3C, 5I). The course of the canal was not connected with the presence of a persistent nasopalatine duct. Topographical variations between the canal and suture at midterm are summarized in Figs. 1F, 2G, and 3F. In addition, candidate nasopalatine nerves were observed in (1) the incisive canal and incisive fossa (Figs. 3B, E, 5B, E, H), (2) the midline suture of the bilateral INs (Figs. 1D, 2D), and/or (3) the center of the lower mass of the IN on the superoposterior side of the second tooth (Figs. 2D, 4C, E, 6C, D, 7A, B). These nerves differed from the branches of the major palatine nerve coming from the posterior site (Fig. 6A, B).

### *Bias suggested by the sectional plane and other factors*

Although sagittal sections were advantageous for understanding the anteroposterior arrangement of the IN, maxilla, palatal bone and upper teeth, the vomer and nasal septum cartilage of late-stage specimens tended to be injured during histological preparation of sagittal sections (Fig. 7). Because of difficulties identifying irregularly shaped upper processes of the IN and because of the course of the incisive canal along the left-right axis near the nose, the intra-IN canal tended to be more frequently observed in frontal than in sagittal sections (Table 1). Sagittal sections sometimes provided a complicated morphology, in that the IN-maxillary suture appeared to be a "branch" of the incisive canal (Figs. 4F, 7B, C). Our observations of late-stage specimens were not based on serial sections but on semiserial sagittal sections at 50 micron


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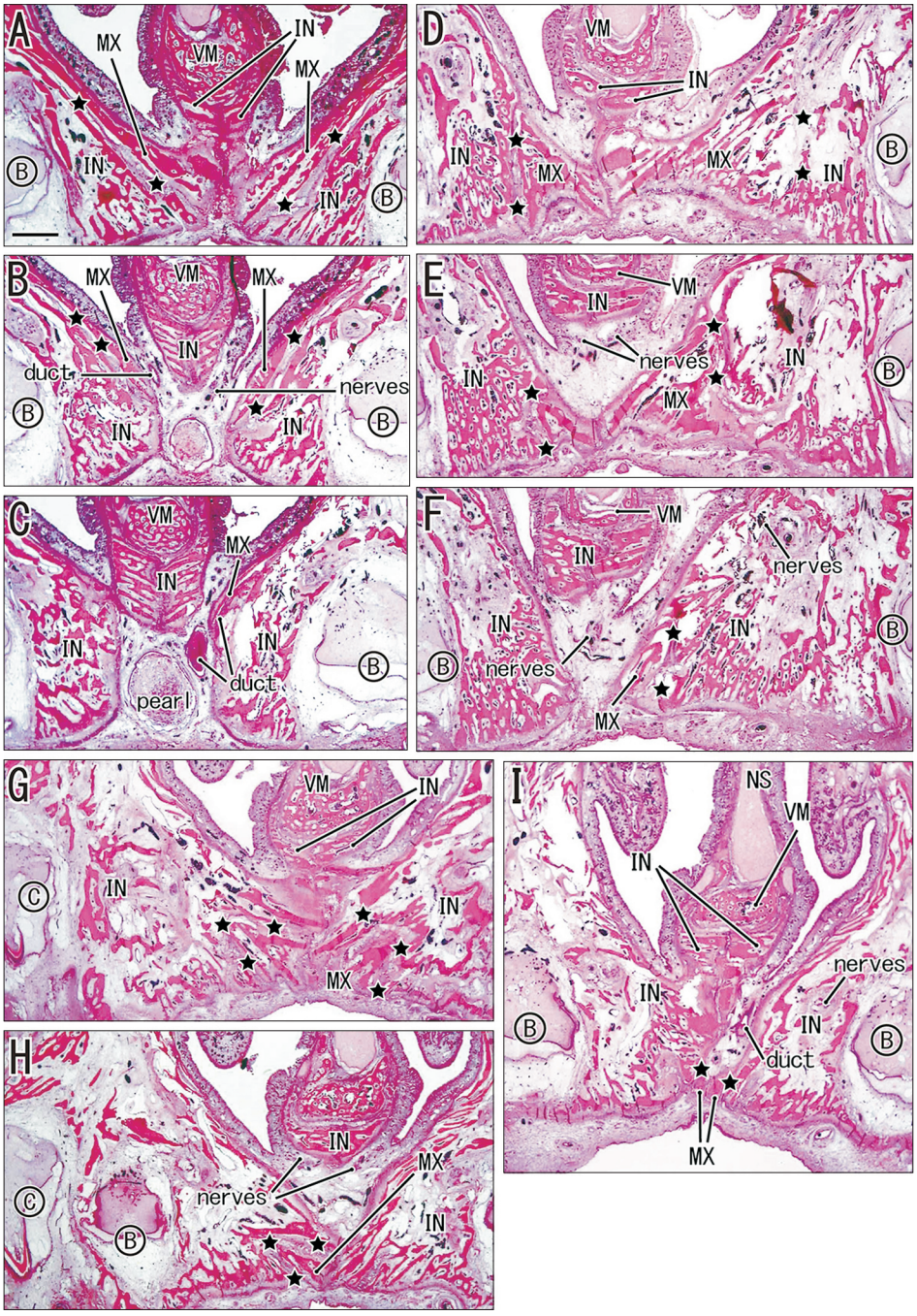


**Fig. 5.** Frontal sections of three late-stage fetuses with and without ducts. Hematoxylin and eosin staining. Panels A–C displays a specimen of crown rump length (CRL) 230 mm with a duct; panels D–F exhibits a specimen of CRL 250 mm without a duct, and panels G–I show a specimen of CRL 270 mm with a duct. All panels were prepared at the same magnification. Panels A, D and G exhibit the most posterior site in each specimen. In all three specimens, an epithelial elongation of the nose extended into loose tissue of the incisive canal. Stars indicate the incisive-maxillary suture. Panels B and C display an incisive canal passing along the suture between the incisive bone (IN) and the maxilla (MX). Panels E and F show an intra-incisive canal on the left-hand side, rather than a canal between the incisive bone and maxilla on the right-hand side. Panel H and I show bilateral intra-incisive canals. Nerves are seen in the canal as well as in the incisive bone (panel F). Scale bar=1 mm. B or C in circles indicate a tooth bud of the lateral incisor, or canine, respectively. pearl, palatal epithelial pearl; VM, vomer.

intervals. We were concerned that this method may be associated with a failure to identify the small upper processes of the IN. However, possibly due to easy identification of thick loose tissue continuous with the submucosa of the nose, the intra-IN canal tended to be found more frequently in semiserial sections of late-stage fetuses than in serial sections of midterm fetuses (Table 1). There was no evidence of rearrangement or reconstruction of the upper parts of the IN during intrauterine growth after 15 weeks.

Consequently, in both of midterm and late-stage fetuses, we found not only the intra-IN canal but also the "classical" canal along the IN-maxillary suture (Figs. 1, 4, 6). There were variations between both of the typical morphologies ("mixed" in Table 2). Nevertheless, we should pay much attention to avoid bias from sectional planes. Sagittal sections were better for the demonstration of the classical canal because, converse-

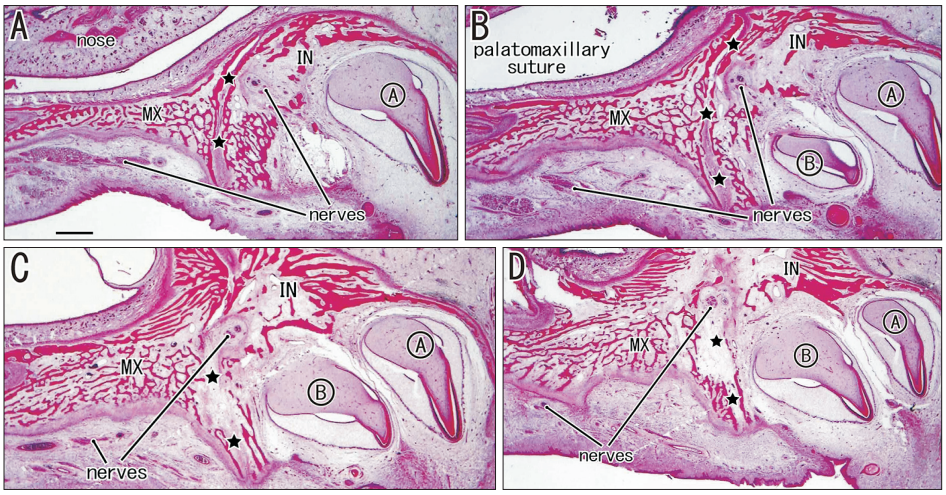


**Fig. 6.** Sagittal sections of a late-stage fetus of crown rump length 255 mm with a canal along the suture. Hematoxylin and eosin staining. Panel A displays the most lateral site in the specimens. This specimen exhibits a simple morphology, in which an incisive canal containing nerves and loose tissue (C, D) corresponds to parts of a suture (stars) between the incisive bone (IN) and maxilla (MX). A duct is absent. Stars indicate the suture, which is narrow and tight in panels A and B. All panels were prepared at the same magnification. Scale bar=1 mm. A or B in circles indicate a tooth bud of the medical incisor, or lateral incisor, respectively.

ly, frontal sections emphasized upper details of the IC as well as a plate-like anterior part of the maxilla.

## Discussion

A study history of the incisive canal is very long and, still now, the structure is generally considered as a product according to one of basic rules of embryology, i.e., a gap of the border between the primary and secondary palates (see the Introduction). However, the present observation demonstrated that the incisive canal with or without nasopalatine duct exhibited a spectrum of variations in topographical relation to the IN-maxillary suture. In contrast to the recent new concept [8], some fetuses carried the intra-IN canal opening to the incisive fossa, while the others had the incisive canal along the IN-maxillary suture.

We identified an intra-IN canal, with or without a nasopalatine duct, as a normal morphology. The upper half of each incisive canal was characterized by both the inferomedial elongation of the nasal epithelium and a continuation of the thick nasal submucosal tissue. The canal seemed to be formed during nasal epithelial development at midterm. Conversely, the development of the intra-IN canal did not require a persistent nasopalatine duct reaching the incisive fossa or connecting with the epithelial pearl. Thus, this study did not emphasize our previous definition of the nasopalatine duct as a duct connecting the nasal epithelium and the paramedian epithelial pearl [9]. This duct apparently did not originate from the primitive oronasal communication prior to the midline palatal fusion. Although we previously reported the presence of this duct in specimens of GA 14 weeks [9], in the present study we identified this duct in specimens of GA 11 weeks. This lower limitation of age or stage was difficult to explain if the duct originated from the primitive oronasal communication during early phases of palatal fusion.

Some cells packed in the midline epithelial seam after fusion may later form a duct that connects to the differentiated nasal epithelium. Actually, the midline seam connects to the epithelial pearls in the incisive fossa [11-13]. However, the nasopalatine duct in the incisive canal often penetrated the IN, not the IN-maxillary suture. The primitive oronasal communication passes through the suture, as the latter was apparently the most anterior site in the loose midline. Therefore, conversely, most or all nasopalatine ducts likely originate from the "secondary" elongation of the nasal epithelium after midterm. The irregularly shaped upper parts of the IN with several processes seemed also to be a product of invasion of the nasal epithelium and submucosal tissue.

Previous studies showed "sagittal sections" that included the incisive canal along the IN-maxillary suture [2, 5-7, 14], but these studies did not mention the possibility of an intra-IN canal, which for a long time had not been a focus of study. Thus, at the beginning of this study, we considered that a midterm fetus with an intra-IN canal would likely die before


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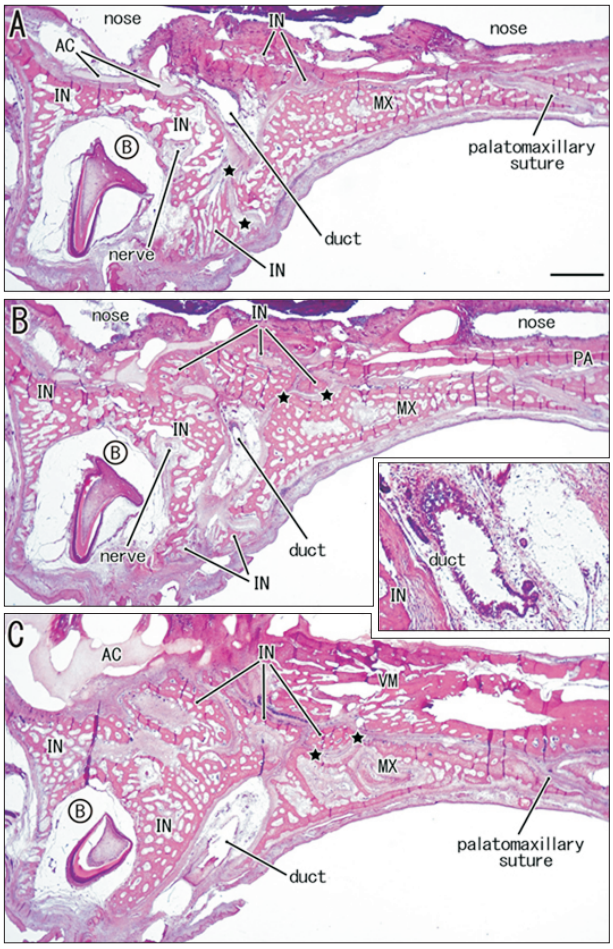


**Fig. 7.** Sagittal sections of a late-stage fetus of crown rump length (CRL) 290 mm with an intra-incisive canal and duct. Hematoxylin and eosin staining. Panel A displays the most lateral site in the specimens. The course of the intra-incisive canal with a duct is intra-incisive laterally (A) and along a suture between the incisive bone (IN) and maxilla (MX) medially (B, C). The suture (stars) turns posteriorly in upper sites (B), but the posterior part appears to be a branch of suture. On the lateral side of the canal and duct between the IN and MX, the suture is narrow and closed (stars in panel A). An inset in panel B shows a higher magnification view of the duct. Panels A–C were prepared at the same magnification. Scale bar=1 mm. B in circles indicates a tooth bud of the lateral incisor. AC, alar cartilages of the anterior nose; PA, palatine bone; VM, vomer.

reaching late-stage or full term. Indeed, this study proofed that fetuses with the intra-IN canal could be alive until late-stage or even full term. However, sagittal sections themselves had disadvantage for the demonstration. First, it might be difficult to define the margins of the vomer and anterior nasal cartilages adjacent to the IN during fetal development [4, 15-18]. Second, the upper processes of the IN were difficult to identify on sagittal sections. In most samples, the intra-IN canal was observed in the nasal half of the course, lying between the upper process and the lower large mass of the bone. Third, an inferomedial elongation of the nasal epithelium, a simple marker for identifying the incisive canal, was found to run along the left-right axis rather than the supero-inferior axis. Therefore, the nasal epithelium entering the canal or suture was often not seen on sagittal sections. In contrast, frontal sections clearly demonstrated that the vomer was sandwiched between the nasal septum cartilage and the upper process of the IN.

**Table 2.** Orientation of figures: ages, sectional planes, and sites of incisive canals

| | Midterm | Late-stage |
|---|---|---|
| Canal along an incisive-maxillary border | Fig. 1 (Fr), Fig. 4A–D (Sag) | Fig. 5A–C (Fr), Fig. 6 (Sag) |
| Canal through the incisive bone | Fig. 3 (Fr) | Fig. 5G–I (Fr) |
| Mixed course depending on the sites | Fig. 2 (Fr), Fig. 4E–G (Sag) | Fig. 5D–F (Fr), Fig. 7 (Sag) |

Fr, frontal sections; Sag, sagittal sections.

Because the present specimens included large fetuses near term, newborns and children are also likely to carry the intra-IN canal. Advances in computed tomography have revealed various abnormalities of the incisive canal and nasopalatine duct in adults [19-24]. These abnormalities were apparently caused by a remnant of a nasopalatine duct or its fragment, with or without upper migration, as these variants were connected to the nasal epithelium. However, a large incisive fossa along the midline on the oral surface of the palate might make a macroscopic finding of the intra-IN canal difficult. Detailed examinations of adult morphology may confirm the high incidence of intra-IN ducts. However, the topographical relationship between the IN and maxilla (i.e., the anatomy of the suture) may change during postnatal growth. In this context, we should remind a plate-like anterior part of the maxilla that lined the inferior or medial aspect of IN and that disturbed our clear identification of the IN-maxillary suture (Figs. 2E, F, 5B, F, I). We did not deny a possibility that, depending on eruption and growth of the face, these thin bony plates could be absorbed because they were located closely to anterior teeth and nose.

### *Study limitations*

The present specimens were of two different origins: Spanish for the midterm and Far-East origin for the late-stage. Although this racial difference was likely to influence incidence of variations (Table 1), we were able to demonstrate

a real presence of variations in one of the basic and famous structures in the human head. Due to the large size, we used semiserial sections (50-μm interval) for observations of the late-stage specimens. We believe that, because of more than 100-μm thicknesses of the canal and duct, even semiserial sections provided a precise information as to whether these structures were present or absent.

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## Author Contributions

Conceptualization: JHK, GM. Data acquisition: JHK, GM. Data analysis or interpretation: JFRV. Drafting of the manuscript: JHK, GM. Critical revision of the manuscript: SS, HA, JFRV. Approval of the final version of the manuscript: all authors.

## Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

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