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# Vertebral Bodies Ossification

## Osificación de Cuerpos Vertebrales

**Josefa Alarcón[1,2]; Laura García-Orozco[1,3]; José Villanueva[1]; Jhonatan Duque-Colorado[1,3]; Ignacio Roa[4] & Mariana Rojas[5]**

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**SUMMARY:** The vertebral column is the skeletal axis of the body, consisting of 33 vertebrae. Embryologically, vertebral development begins at the end of the third week of gestation and is completed around 25 years of age, when the ossification process concludes. The vertebral body originates from the sclerotome, which derives from the somitic or paraxial mesoderm. Ossification of the vertebral body progresses through three stages: mesenchymal, cartilaginous, and osseous. The mesenchymal stage spans from the fourth to the sixth week. During this stage, the sclerotomal portion of each somite undergoes a process known as resegmentation. The densely packed caudal half of one sclerotome fuses with the loosely arranged cranial half of the adjacent sclerotome segment, forming an intersegmental structure that will eventually develop into the vertebral column. The cartilaginous stage occurs from the sixth to the eighth week, marked by the appearance of six chondrification centers at the level of the vertebral body, costal processes, and neural arches. The osseous stage extends from the tenth intrauterine week to approximately 25 years of age. During this stage, primary and secondary ossification centers develop. The objective of the present work is to present instructional material based on chick embryos and human fetuses at various developmental stages as a contribution to the teaching of Embryology.

**KEY WORDS: Ossification; Vertebral body; Spine.**

## INTRODUCTION

Skeletal tissues are present throughout most of the body, exhibiting varied components from both morphological and tissue perspectives. However, despite this diversity, there are some fundamental common embryonic features; for example, all skeletal tissues arise from mesenchymal cells. In the trunk, the mesenchyme that gives rise to the segmented axial skeleton (vertebral column, ribs, and sternum) originates from the sclerotome of the mesodermal somites (Carlson, 2014).

The vertebral column extends along the midline from the base of the skull cranially to the pelvis caudally, and then beyond as the rudimentary tail (Kerr & Rosenberg, 2018). It is composed of 33 individual components called vertebrae, which articulate superiorly and inferiorly with each other, forming a segmented structure (Scheuer & Black, 2000; Ortiz-Maldonado, 2016; Kerr & Rosenberg, 2018).

The vertebral column serves as a bony axis with three main functions: it provides structural support, protects the nervous system by forming a secure canal for neural elements, and allows movement in all planes of space (Vargas Sanabria, 2012; Olivares & Rojas, 2013). The vertebral column is divided into general regions: (1) an occipital region, which becomes part of the skull base; (2) a cervical region, including the atlas and axis specialized vertebrae connecting the spine to the skull; (3) the thoracic region, where the ribs articulate; (4) the lumbar region; (5) a sacral region, where vertebrae fuse to form the sacrum; and (6) a caudal region, forming the tail in most mammals and the rudimentary coccyx in humans (Vargas Sanabria, 2012; Carlson, 2014).

Vertebrae are the basic units of the vertebral column. They vary in size and in certain features from one spinal region to another, and to a lesser extent within regions,

[1] Programa de Doctorado en Ciencias Morfológicas, Facultad de Medicina, Universidad de La Frontera, Temuco, Chile.
[2] Centro de Investigación en Ciencias Odontológicas (CICO-UFRO), Facultad de Odontología, Universidad de La Frontera, Temuco, Chile.
[3] Centro de Excelencia en Estudios Morfológicos y Quirúrgicos (CEMyQ), Facultad de Medicina, Universidad de La Frontera, Temuco, Chile.
[4] Departamento Ciencias Básicas Biomédicas, Facultad de Ciencias de la Salud, Universidad de Talca, Talca, Chile.
[5] Laboratorio de Embriología Comparada, Núcleo Integrativo de Biología y Genética, ICBM, Facultad de Medicina, Universidad de Chile, Chile.

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although their basic structure remains the same. A typical vertebra consists of a vertebral body, a vertebral arch, and seven processes (Gilbert, 2005; Ortiz-Maldonado, 2016). The vertebral body is the anterior, largest, roughly cylindrical part of the bone, providing strength and bearing body weight (Chin *et al*., 2011). Vertebral body size increases down the column, especially below T4, due to increasing weight-bearing demand. The vertebral arch is located posterior to the body and consists of pedicles and laminae (Ortiz-Maldonado, 2016). The arch and posterior body wall form the vertebral foramen. Seven processes extend from the arch: one midline spinous process projecting posteriorly, two transverse processes projecting posterolaterally, and four articular processes (two superiors, two inferiors), each with an articular surface (Chin *et al*., 2011).

## EMBRYONIC FORMATION OF THE VERTEBRAL BODY

In the third week, a flat trilaminar disc forms from the bilaminar disc, made of three germ layers: ectoderm, mesoderm, and endoderm. At this point, the basic body plan and axial orientation are established (Moore *et al*., 2013; Kaplan *et al*., 2005). The cranio-caudal axis is defined by the primitive streak. This polarization is first marked by the formation of the notochord, and later by the neural plate, induced by the notochord on the dorsal ectoderm (Carlson, 2014). A longitudinal neural groove appears in the neural plate, flanked by neural folds. Fusion of these folds forms the neural tube, the primordium of the central nervous system (Fig. 1) (Moore *et al*., 2013).

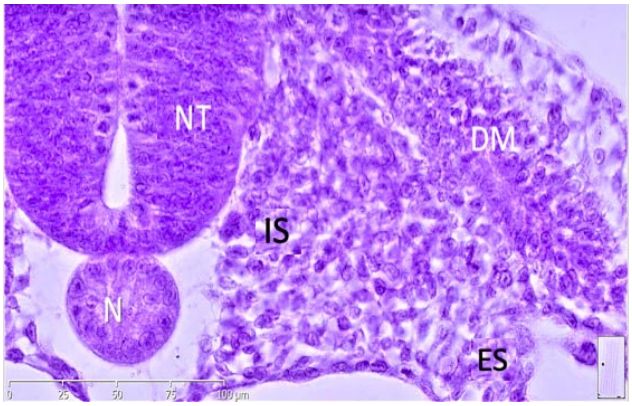



Fig. 1. Transverse section of a 48-hour chick embryo. The internal sclerotome (IS), external sclerotome (ES), notochord (N), neural tube (NT), and dermomyotome (DM) are observed. H&E staining, 400X magnification.

After passing through the primitive streak, mesodermal cells migrate laterally between ectoderm and endoderm, forming a continuous layer of mesenchymal cells. On either side of the notochord, the mesoderm differentiates into three regions: paraxial (somitic), intermediate, and lateral (Moore *et al*., 2013; Kaplan *et al*., 2005).

Closest to the neural tube lies the paraxial mesoderm, which by the end of the fifth week gives rise to 42–44 somites (Kerr & Rosenberg, 2018). The first significant event in somitogenesis is segmentation of the paraxial mesoderm. By the end of week three, the paraxial mesoderm is segmented into somitomeres in the head region and somites from occipital to caudal regions. Somites differentiate into a ventromedial part, the sclerotome, and a dorsolateral region, the dermomyotome (Fig. 1). The sclerotome will form the axial skeleton (Skórzewska *et al*., 2013; Carlson, 2014; Rojas & Smok, 2014).

### Somitic (Paraxial) Mesoderm

Each somite consists of mesodermal cells arranged in concentric spirals around the center. From occipital to caudal regions, the mesoderm organizes into somites. The first pair appears in the occipital region around day 20 (Kerr & Rosenberg, 2018). New somites form in a cranio-caudal sequence at a rate of about three pairs per day until the fifth week, reaching 42–44 pairs (Kerr & Rosenberg, 2018; Sadler, 2019). There are 4 occipital, 8 cervical, 12 thoracic, 5 lumbar, 5 sacral, and 8–10 coccygeal pairs. The first occipital and the last 5–7 coccygeal somites later disappear; the rest contribute to the axial skeleton (Sadler, 2019).

### Molecular Regulation of Somite Differentiation

Signals for somite differentiation come from surrounding structures, including the notochord and neural tube. Secreted proteins from the noggin and sonic hedgehog genes—produced by the notochord and basal plate of the neural tube—induce the ventromedial part of the somite to form the sclerotome (Skórzewska *et al*., 2013). These cells lose epithelial characteristics, becoming mesenchymal. Induced sclerotome cells express the transcription factor PAX1, initiating a gene cascade for cartilage and bone formation (Sadler, 2019).

By the end of week four, sclerotome cells form loose mesenchyme (embryonic connective tissue) capable of forming bone. Bones initially appear as condensations of mesenchymal cells forming bone models. Condensation marks the start of selective gene expression preceding differentiation (Sadler, 2019).

## Ossification of the Vertebral Body

Vertebral development begins at the end of the third week and is completed around age 25, ending the ossification process. Vertebrae undergo endochondral ossification (Breeland *et al*., 2025). Development is divided into three stages:

**Precartilaginous (mesenchymal) stage**: In week 4, mesenchymal cells from the sclerotomes gather around the notochord and neural tube. The sclerotome undergoes a process called resegmentation (Domínguez-Gasca *et al*., 2018; Sadler, 2019) (Fig. 2). Each sclerotome consists of a loosely packed cranial half and a densely packed caudal half (Fig. 3). The caudal half of one sclerotome fuses with the cranial half of the sclerotome below, forming an intersegmental structure that becomes the vertebral body (Sadler, 2019).

Notably, while vertebral bodies result from sclerotome recombination, neural arches, pedicles, and costal elements mainly develop from the dense caudal part of each segmental sclerotome (Christ & Wilting, 1992). This explains why pedicles, originating from the sclerotome's caudal part, usually connect to the superior pole of the adult vertebra (Cunningham *et al*., 2016).

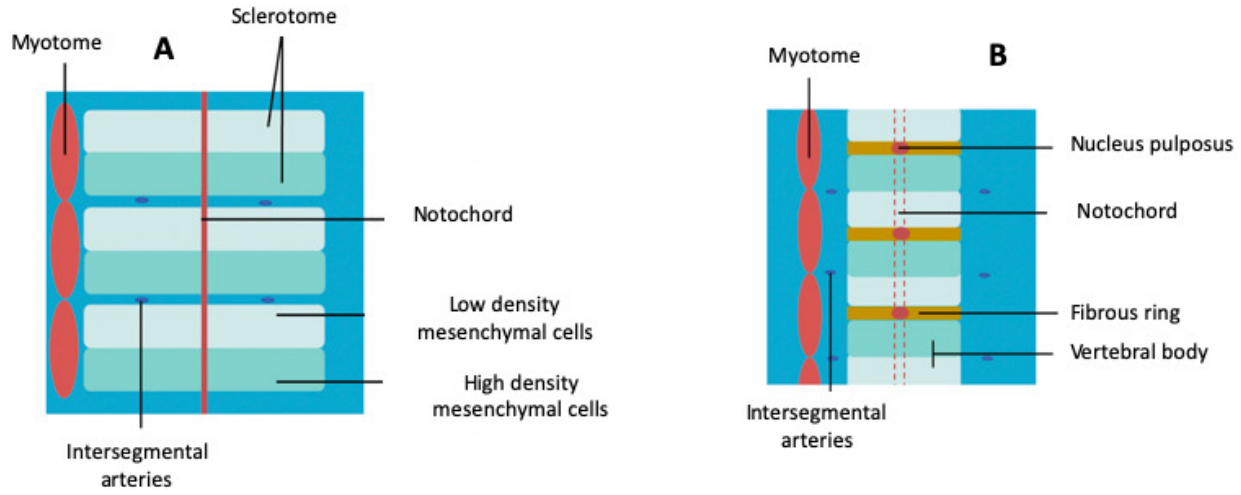



Fig. 2. A. Schematic frontal section of a 4-week-old embryo showing a condensation of sclerotome cells around the notochord. This condensation consists of a cranial zone with low cell density (light sclerotomite) and a caudal zone with high cell density (dark sclerotomite). B. Schematic frontal section showing that the vertebral body is formed from the cranial half and caudal half of two successive sclerotomes. At this stage, intersegmental arteries cross the vertebral bodies. The notochord degenerates except in the intervertebral disc region, where it forms the nucleus pulposus.

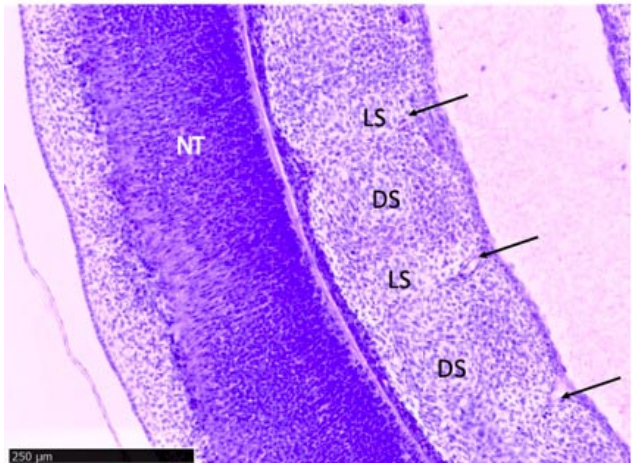



Fig. 3. Sagittal section of a chick embryo. Neural tube (NT), light sclerotomies (LS), and dark sclerotomies (DS) are observed. Arrows indicate segmental arteries. Magnification 200X, H&E.

The notochord degenerates within future vertebral bodies but expands between vertebrae, forming the nucleus pulposus of intervertebral discs. This nucleus becomes surrounded by concentric fibers forming the annulus fibrosus; together, these form the intervertebral disc (Figs. 4 and 5) (Kerr & Rosenberg, 2018; Sadler, 2019).

**Cartilaginous stage:** In week 6, chondrification centers appear in each mesenchymal vertebra—up to six per template. The two centers in the centrum fuse by the end of the embryonic period, forming the cartilaginous centrum (Trancho *et al*., 2011). Simultaneously, centers in the neural arches fuse with each other and the centrum (Fig. 6). Spinous and transverse processes develop from chondrification center extensions in the neural arch. The entire vertebral column becomes cartilaginous (Sadler, 2019).

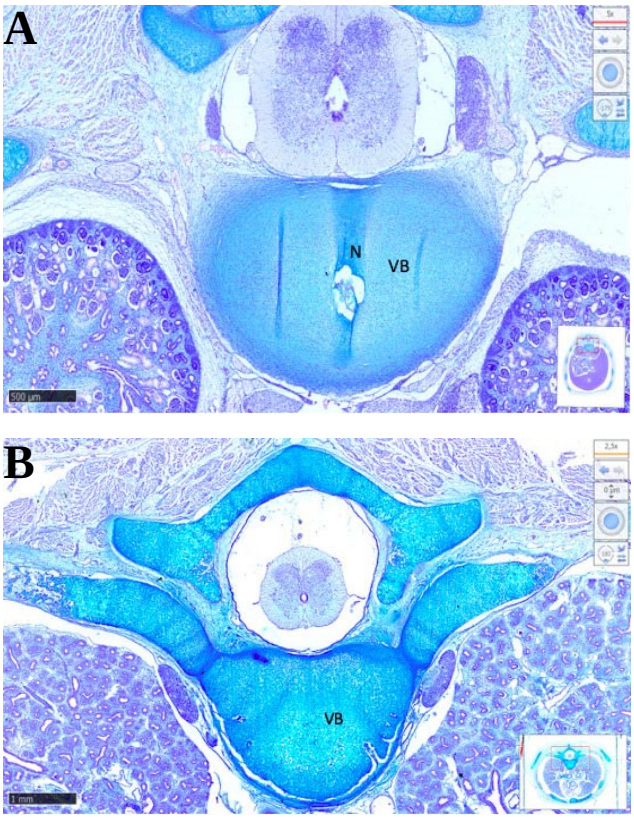



Fig. 4. (A) Cross-section through a 12-week human fetus. The cartilaginous vertebral body and notochordal remnant are visible. Magnification 5X, Alcian blue H&E staining. (B) Cross-section through a 14-week human fetus. The cartilaginous vertebral body is visible, but the notochord is no longer identifiable. Magnification 35X, Alcian blue H&E staining.

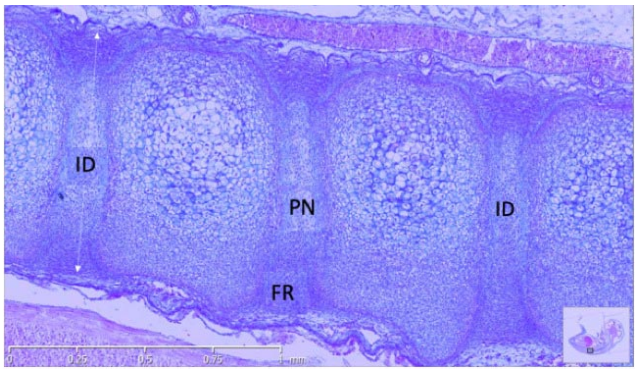



Fig. 5. Sagittal section of a 47 mm human fetus. The notochord has involuted but remains between the vertebrae as the nucleus pulposus (PN), which is surrounded by circular fibers that form the annulus fibrosus (FR). The nucleus pulposus and the annulus fibrosus together form the intervertebral disc (ID). Magnification 5X, H&E staining with Alcian blue.

**Osseous stage:** This begins in week 10 (crown-rump length 60 mm) and continues through week 11 (CRL 63 mm) until age 25. In the osseous stage, primary and secondary ossification centers appear (Skórzewska *et al*., 2013).

**Primary ossification:** histological data have shown that the spine begins to mineralize during the tenth week of gestation (Bareggi *et al*., 1994; Skórzewska *et al*., 2013). Each vertebra ossifies from three primary centers: one in the centrum itself and two in each half of the neural arch (Moore *et al*., 2013; Skórzewska *et al*., 2013) (Fig. 7 and 8). Ossification of the neural arches proceeds in a craniocaudal direction, while the central centers, which first

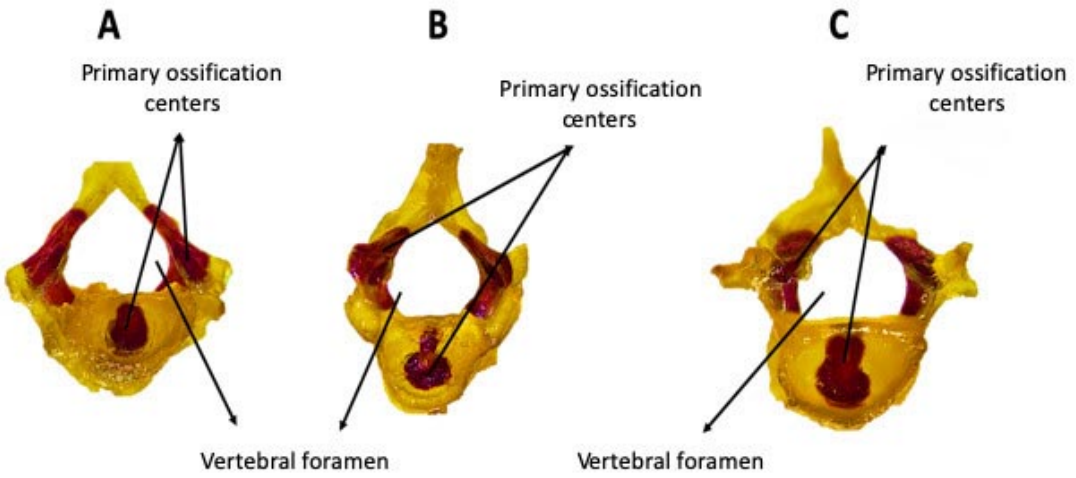



Fig. 6. Primary ossification of the vertebrae of a 20-week fetus. A. Cervical vertebra. B. Thoracic vertebra. C. Lumbar vertebra. The primary ossification centers are shown: one in the centrum itself and two in each half of the neural arch.

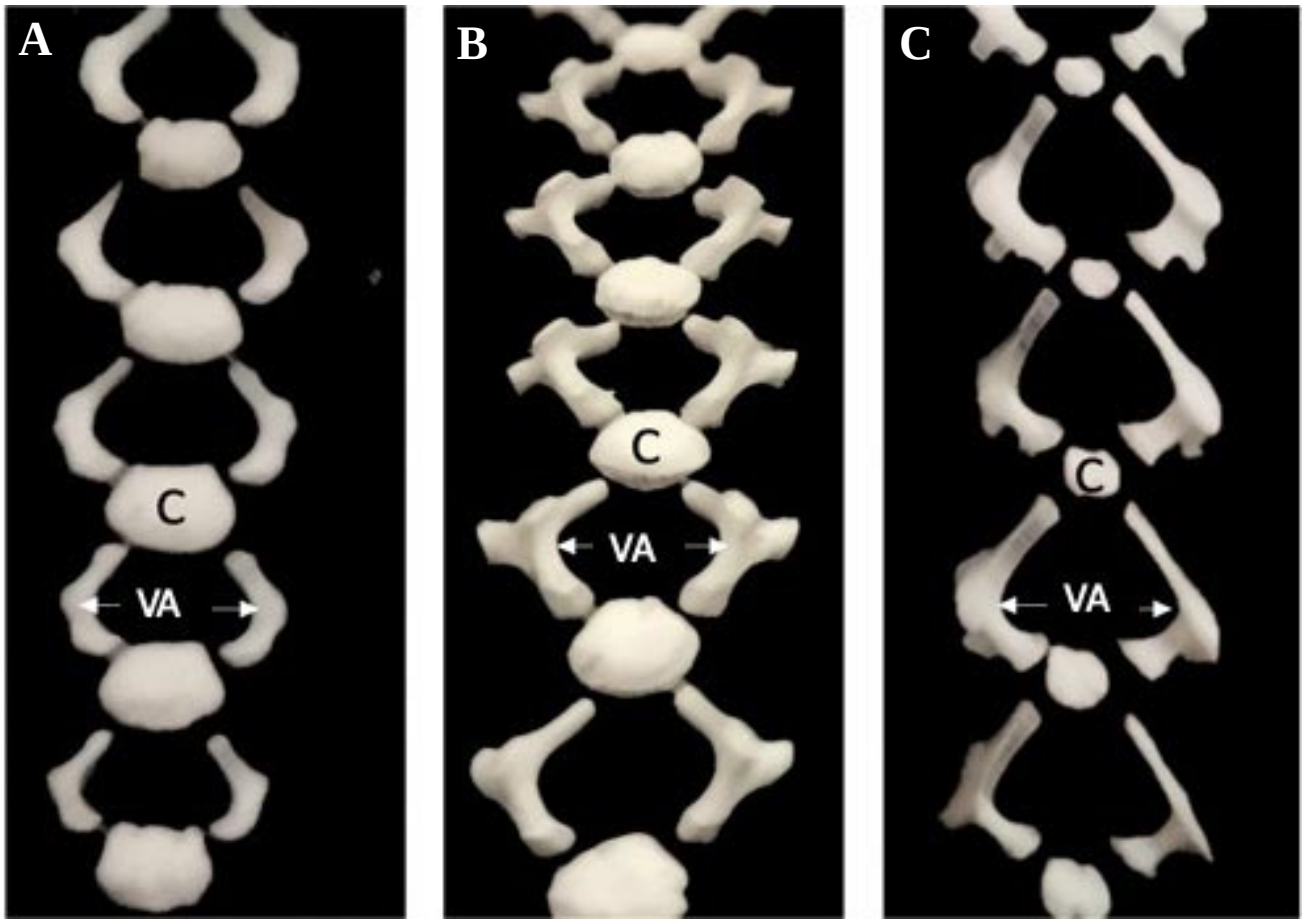



Fig. 7. Bony parts of vertebral bodies after that of a 26-28 week-old fetus. A. Cervical vertebra; B. Thoracic vertebra; C. Lumbar vertebra; Centrum (C); Halves of vertebral arch (VA).

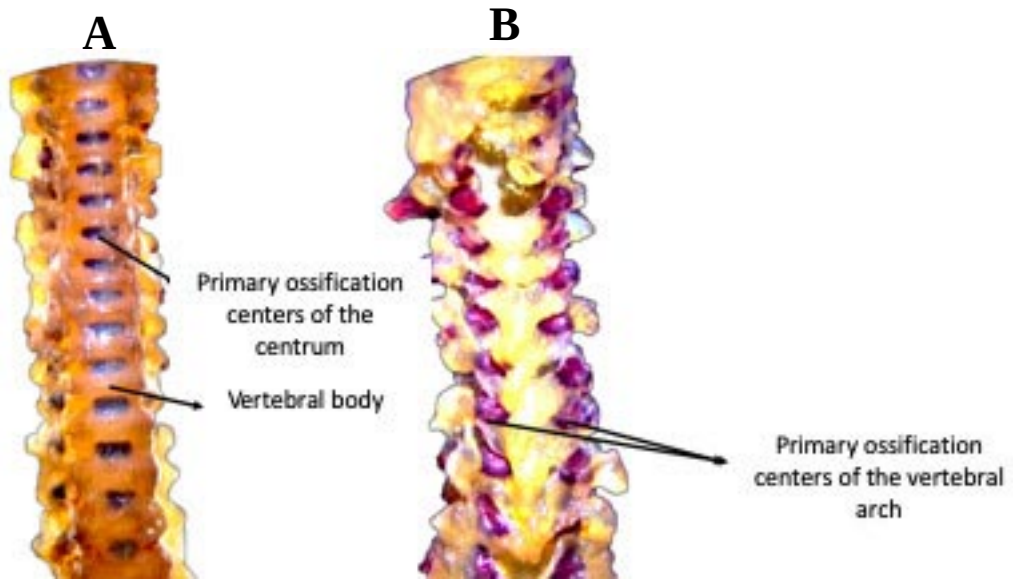



Fig. 8. Vertebral column of a 20-week-old fetus. A. Anterior view of the vertebral column. Primary ossification centers of the centrum of each vertebra are observed. B. Posterior view of the vertebral column. Primary ossification centers of the vertebral arch are observed.

appear in the lower thoracic region, subsequently appear both cranially and caudally from their initial site of appearance (Noback & Roberston, 1951; Bareggi *et al*., 1994; Skórzewska *et al*., 2013). The progressive onset of ossification centers is closely related to gestational age (Bareggi *et al*., 1994).

Each typical vertebra consists of three bony parts joined together by cartilage: a vertebral arch, a vertebral body, and the transverse processes. The bony halves of the vertebral arch usually fuse during the first 3–5 years (Trancho *et al*., 2011). The vertebral arch articulates with the centrum at the cartilaginous neurocentral joints (Fig. 8), which allow the vertebral arches to grow as the spinal cord increases in length. These joints disappear when the vertebral arches fuse with the centrum between the third and sixth year (Moore *et al*., 2013).

**Secondary ossification:** After puberty, five secondary centers appear per vertebra—one at the tip of the spinous process, one at each transverse process tip, and two annular epiphyses at the superior and inferior edges of the vertebral body.

The vertebral body is composed of a combination of the annular epiphyses and the bone located between them. The vertebral body includes the centrum, parts of the vertebral arch, and the articular facets for the heads of the ribs. All secondary centers fuse with the rest of the vertebrae by approximately 25 years of age (Trancho *et al*., 2011; Moore *et al*., 2013).

### Ossification Exceptions

Exceptions to the typical ossification pattern occur in the C1, C2, and C7 vertebrae, as well as in the sacrum and coccyx (Fig. 9). The axis and atlas vertebrae have a special morphology and a characteristic origin. The atlas lacks a body, which allows space for the penetration of the odontoid process, which arises from the axis. This process consists of three fused bodies, which are believed to correspond to: 1) the middle segment of the vertebral body of a transitional bone (the proatlas), absent in humans, 2) the body that would have belonged to the atlas, and 3) the normal body of the axis. This configuration allows for greater rotation of the head over the cervical spine. When the ubiquitously expressed transgene Hoxa7 was introduced into the mouse germline, the cranial portion of the vertebral column shifted posteriorly. The base of the occipital bone was transformed into an occipital vertebra (the proatlas), and the atlas merged with its body, resulting in an axis without the odontoid process (Moore *et al*., 2013).

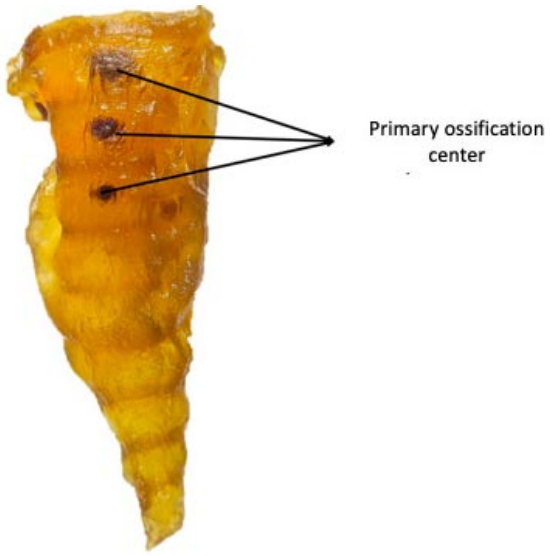



Fig. 9. The development of the sacrum of a 20-week-old fetus is shown.

## CONCLUSION

Proper vertebral column development relies on a coordinated sequence of molecular and morphological signals. Vertebral development is a complex process beginning at the end of the third week of gestation and ending by age 25. The mesoderm differentiates into the sclerotome, which gives rise to the vertebral body. Vertebral development includes a mesenchymal stage forming the vertebral model, a cartilaginous stage (weeks 6–10) forming six chondrification centers, and an ossification stage from week 8 to age 25 with secondary ossification.

Understanding fetal ossification timing and sequence helps assess skeletal maturity. Among fetal bones, the spine is particularly useful for ultrasound evaluation of fetal development.

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**RESUMEN:** La columna vertebral es el eje óseo del organismo constituido por 33 vértebras. Embriológicamente, el desarrollo de las vértebras comienza durante la tercera semana de gestación y culmina hacia los 25 años de vida, completándose el proceso de osificación. El cuerpo vertebral se origina del esclerotoma, que proviene del mesodermo somítico o paraxial. La osificación del cuerpo vertebral pasa por tres etapas: Mesenquimática, cartilaginosa y ósea. La etapa mesenquimática se extiende desde la cuarta a la sexta semana. En esta etapa, la porción del esclerotoma de cada somita experimenta un proceso denominado resegmentación. La parte caudal de densidad elevada

de un esclerotomo se fusiona con la parte craneal laxa del segmento del esclerotomo adyacente, para formar una estructura intersegmentaria, que continuará hasta convertirse en la columna vertebral. La etapa cartilaginosa se extiende desde la sexta a la octava semana. Aparecen seis centros de condrificación a nivel del cuerpo vertebral, procesos costales y arcos neurales. La fase ósea se extiende desde la décima semana intrauterina hasta aproximadamente los 25 años de edad. En la etapa ósea aparecen centros de osificación primarios y secundarios. El objetivo del presente trabajo es presentar material instruccional realizado en embriones de pollo y de fetos humanos en distintas fases del desarrollo como un aporte a la enseñanza de la Embriología.

**PALABRAS CLAVE: Osificación; Cuerpo vertebral; Columna.**

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Corresponding author:
Dra. Mariana Rojas R.
Laboratorio de Embriología Comparada
Programa de Anatomía y Biología del Desarrollo
Facultad de Medicina, ICBM
Universidad de Chile
CHILE

E-mail: mrojasr@u.uchile.cl
dramrojas@hotmail.com