Development of Teeth and their Supporting Tissues
Some pictures may be disturbing or offensive to some. They are presented with the intent to educate.
TOOTH & RELATED TISSUES: Developmental goal

- PULP
- DENTINE
- ENAMEL
- CEMENTUM
- PERIODONTAL LIGAMENT/ PDL
- ALVEOLAR BONE
- GINGIVA
Tooth Development

A. Bud Stage
B. Cap Stage
C. Bell Stage
D. Dentino- and Amelogenesis
E. Dentino- and Amelogenesis
F. Crown formation
G. Root formation and Eruption
H. Function

Essentials of Oral Histology and Embryology,
Initiation of Tooth Development

The initiation of tooth development begins at 37 days of development with formation of a continuous horseshoe-band of thickened epithelium in the location of upper and lower jaws – **Primary Epithelial Band**

Each band of epithelium will give rise to 2 sub divisions:
1. **Dental lamina and**
2. **Vestibular lamina**
Vestibular Lamina

Figure from Ten Cate’s Oral Histology, Ed., Antonio Nanci, 6th edition
Morphogenesis

• The genesis of a morphe (of a shape; verb: to morph = to shape; change from one image to another)

• Molecular pathways
  – Bone morphogenetic protein (BMP)
    • Inhibitory signals
  – Fibroblast growth factor (FGF)
    • Stimulatory signals
  – Sonic Hedgehog (Shh)
    • Initiation of tooth formation; cap stage development
  – Wingless–related intergretion site (Wnt)
    • Stimulatory signals
    • Overexpression of Wnt inhibitor Dkk1 → NO DENTAL PLACODE
  – Notch
    • Lateral inhibition or inductive signaling
  – Ectodysplasin (Eda)
    • Formation of the placode
    • Inhibition → Oligodontia
    • Overactivation → more misshaped teeth
An inductive signalling network regulates mammalian tooth morphogenesis with implications for tooth regeneration
$Shh$ overexpression
Some Genes Regulating Tooth Development

• *MSX1* **MUT** tooth agenesis
• *Pax9* **MUT** tooth agenesis; bud to cap stage transition
• *Dkk1* **OVEREXPR** tooth agenesis
• *WNT10 A*: Most common gene in tooth agenesis
• *AXIN2*: hypodontia; colorectal cancer
• *LRP6, GREM2, SPRY2&4*
• *EDA*: Ectodermal dysplasia
• **SEE ALSO TABLE 5-2, Chapter 5**
Stomatodeum

Maxillary Process

Dental lamina

Dental placode
Regulated by Notch, Eda, Wnt, Shh

http://www.usc.edu/hsc/dental/ohisto/
Dental Lamina

- Dental lamina appears as a thickening of the oral epithelium adjacent to condensation of ectomesenchyme

- 20 areas of enlargement or knobs appear, which will form tooth buds for the 20 primary teeth

- Not all will appear at the same time. The first to develop are those of the anterior mandible region

- At this early stage the tooth buds have already determined their crown morphology

- Successional lamina: lamina from which permanent teeth develop

- The dental lamina begins to function at 6th prenatal week and continues to 15th year of birth (3rd molar)

Figures from: http://www.usc.edu/hsc/dental/ohisto/
HOW DO THINGS START?
Some intriguing experiments

• Ectoderm carrying dental morphogenetic information
  – Combined with skin mesenchyme $\rightarrow$ no teeth
  – Ectoderm carrying dental info combined with eye eectomesenchymeme $\rightarrow$ teeth

• Ectomesenchymeme responding to ectoderm
  – Ectomesenchymeme can elicit dental development when combined with non-dental ectoderm; BUT WHAT KIND OF ECTOMESENCHYME?
    • Late first arch ectomesenchymeme
An inductive signalling network regulates mammalian tooth morphogenesis with implications for tooth regeneration

FIGURE 5-1 Molecular signaling during tooth crown development. Expression sites of transcription factors (italic) and signaling molecules (bold) are listed.
Tooth development is a continuous process, however can be divided into 3 stages:

1. Bud Stage
2. Cap Stage
3. Bell Stage

No clear separation between stages

Sections may depict different stages, i.e. cap vs. bell

Histodifferentiation: End of cap stage and beginning of bell stage
1. Bud Stage

Initiation
Bud stage is characterized by a rounded, localized growth of epithelium surrounded by proliferating ectomesenchymal cells, which are packed closely beneath and around the epithelial bud (condensation).

http://www.usc.edu/hsc/dental/ohisto/
1. Bud Stage

In the bud stage, the enamel organ consists of peripherally located low columnar cells and centrally located polygonal cells.

http://www.usc.edu/hsc/dental/ohisto/
2. Cap Stage (Morphogenesis)

Condensation of the ectomesenchyme immediately subjacent to the tooth bud is caused by lack of extracellular matrix secretion by the cells thus preventing separation. **Histodifferentiation** begins at the end of cap stage.

Epithelial outgrowth called **Enamel Organ** because it will eventually form the Enamel. The term Dental Organ is not considered appropriate by some.

**Dental Papilla**: Ball of condensed ectomesenchymal cells (it will form dentin and pulp). The peripheral cells adjacent to the inner dental epithelium will enlarge and later differentiate into odontoblasts.
Dental follicle or dental sac is the condensed ectomesenchymal tissue surrounding the enamel organ and dental papilla. This gives rise to cementum and the periodontal ligament (support structures for tooth).
Lateral Lamina: extension from the dental lamina that is connected to the enamel organ

Enamel niche: It is an artifact produced during sectioning of the tissue. It occurs because the enamel organ is a 3D sheet of proliferating cells rather than a single strand and contains a concavity filled with ectomesenchyme

We can also see that the inner and the outer dental epithelium are being organized
**Enamel Knot**: Densely packed accumulation of cells projecting from the inner enamel epithelium into dental papilla. Organizational center for cusp development. FGFs, BMPs, MSX2; shares similarities with ridges of developing limbs. **Enamel Cord**: Extension of the enamel knot.

**Dental organ or tooth germ** is a term used to constitute the structure that has enamel organ, dental papilla and dental follicle
3. Bell Stage

- Continued growth leads to bell stage, where the enamel organ resembles a bell with deepening of the epithelium over the dental papilla.

- Continuation of histodifferentiation (ameloblasts and odontoblasts are defined) and beginning of morphodifferentiation (tooth crown assumes its final shape).

[Diagram of tooth development with labels: Dental lamina, Outer enamel epithelium, Inner enamel epithelium, Dental papilla, Dental follicle, Cervical loop.]

[Link to source: http://www.usc.edu/hsc/dental/ohisto/]
3. Bell Stage (Early)

**Inner enamel epithelium:** Short columnar cells bordering the dental papilla. These will eventually become ameloblasts that will form the enamel of the tooth crown by differentiating into tall columnar cells. The cells of inner dental epithelium exert an organizing influence on the underlying mesenchymal cells in the dental papilla, which later differentiate into odontoblasts.

**Outer enamel epithelium:** Cuboidal cells that cover the enamel organ. Their function is to organize a network of capillaries that will bring nutrition to the ameloblasts. In preparation to formation of enamel, at the end of bell stage, the formerly smooth surface of the outer dental epithelium is laid in folds. Between the folds, adjacent mesenchyme of the dental sac forms papillae that contain capillary loops to provide nutritional supply for the intense metabolic activity of the avascular enamel organ.

http://www.usc.edu/hsc/dental/ohisto/
3. Bell Stage (Early)

Stellate reticulum: Star-shaped cells with processes, present between the outer and the inner dental epithelium. These cells secrete glycosaminoglycans, which attract water, thereby swelling the cells and pushing them apart. However, they still maintain contact with each other, thus becoming star-shaped. They have a cushion-like consistency that may support and protect the delicate enamel organ. It is absent in the portion that outlines the root portions.

Stratum intermedium: Cell layer between the inner dental epithelium and stellate reticulum which have high alkaline phosphatase activity. They assist inner dental epithelium (ameloblasts) to form enamel.

http://www.usc.edu/hsc/dental/ohisto/
Dental Papilla: Before the inner dental epithelium begins to produce enamel, the peripheral cells of the mesenchymal dental papilla differentiate into odontoblasts under the organizing influence of the epithelium. First, they assume a cuboidal shape and then a columnar form and acquire the specific potential to produce dentin. The basement membrane that separates the enamel organ and the dental papilla just prior to dentin formation is called the “membrana preformativa”
3. Bell Stage

Higher power view

http://www.usc.edu/hsc/dental/ohisto/
3. Bell Stage

Cervical loop: Area where the inner and the outer enamel epithelium meet at the rim of the enamel organ. This point is where the cells will continue to divide until the tooth crown attains its full size and which after crown formation will give rise to the epithelium for root formation. It is also called “Zone of Reflexion”.

http://www.usc.edu/hsc/dental/ohisto/
3. Bell Stage

http://www.usc.edu/hsc/dental/ohisto/
3. Bell Stage

Dental lamina (and the lateral lamina) will disintegrate and loose contact with oral epithelium. Sometimes, these epithelial cells will persist when they are called “epithelial pearls” or “cell rests of Serres”

Clinical significance: Cysts will develop (eruption cysts) and prevent eruption, or they may form odontomas (tumors) or may form supernumerary teeth

http://www.usc.edu/hsc/dental/ohisto/
TOOTH TYPE DETERMINATION

- Homodonts and heterodonts

You and me and our cats and dogs

From a biology lecture; U of Miami
TOOTH TYPE DETERMINATION

• Field model
  – Determining factors reside in the ectomesenchyme
  – Overlapping fields of expression
  – Major determining genes include
    • Barx1, Dlx1/2 → molars
    • Dlx1/2, MSX1 → canines and premolars
    • MSX1,2 Alx3 → incisors

• Clone model
Future crown patterning occurs in the bell stage, by folding of the inner dental epithelium. Cessation of mitotic activity within the inner dental epithelium determines the shape of a tooth.

Figure from Ten Cate’s Oral Histology, Ed., Antonio Nanci, 9th edition
Clinical Correlation. Several odontogenic cysts and tumors can arise from developing tooth structures. Two such conditions are:

1. Ameloblastoma – which are tumors of odontogenic epithelium that may arise from cell rests of enamel organ or from the developing enamel organ among other things
Histology resembles enamel organ epithelium with peripheral columnar ameloblast-like cells surrounding loosely arranged stellate-reticulum-like cells.
2. Odontogenic Myxoma: Tumor of the jaw that arise from odontogenic ectomesenchyme. Histologically, looks similar to mesenchymal portion of a developing tooth (dental papilla).
Formation of Permanent Dentition

Successional tooth bud

http://www.usc.edu/hsc/dental/ohisto/

The tooth germs that give rise to permanent incisors, canines and premolars form as a result of further proliferative activity within the dental lamina, lingual to the deciduous tooth germ.

The developing permanent molars have no deciduous predecessor and their tooth germs originate from the dental lamina that extends posteriorly beneath the oral epithelium after the jaws have grown.
A timetable to remember
Entire primary dentition initiated between 6 and 8 weeks of embryonic development.

Successional permanent teeth initiated between 20th week in utero and 10th month after birth

Permanent molars between 20th week in utero (first molar) and 5th year of life (third molar)
You must remember the following:

- Hard tissue formation starts at the late stages of the bell stage
- Differentiation of cells into odontoblasts and ameloblasts
- Dentin is formed before enamel
- Dentin initiates the formation of enamel
Bell Stage

Hard Tissue Formation

Deposition of dental hard tissues is called “apposition”

After the crown attains its final shape during cap to early bell stage, the inner dental epithelial cells stop to proliferate, except the cells at the cervical loop

First layer of dentin appears at the cusp tips and progresses cervically, and the columnar cells of the inner enamel epithelium become elongated and show reverse polarization, with the nuclei adjacent to stratum intermediate (ameloblasts)

The boundary between the odontoblasts and inner enamel epithelium defines the future dentino-enamel junction

http://www.usc.edu/hsc/dental/ohisto/
For dentinogenesis and amelogenesis to take place normally, the differentiating odontoblasts and ameloblasts will receive signals form each other – “reciprocal induction”

Stages of Apposition

1. Elongation of inner enamel epithelium
2. Differentiation of odontoblasts
3. Formation of dentin
4. Formation of enamel
At the same time or soon after the first layer of dentin (mantle dentin) is formed, the inner enamel epithelial cells differentiate into ameloblasts and secrete enamel proteins. These proteins further will help in the terminal differentiation of odontoblasts. The ameloblasts will then start laying down organic matrix of enamel against the newly formed dentinal surface. The enamel matrix will mineralize immediately and form the first layer of enamel. The formation of enamel is called amelogenesis.

http://www.usc.edu/hsc/dental/ohisto/
Apposition

At the same time when the inner enamel epithelium is differentiating, the undifferentiated ectomesenchymal cells increase rapidly in size and ultimately differentiate into odontoblasts.

The increase in size of the papillary cells leads to elimination of the acellular zone between dental papilla and inner enamel epithelium.

Differentiation of odontoblasts from ectomesenchymal cells are induced by influence from the inner enamel epithelium.

Experiments have shown that if there is no inner enamel epithelium, there is no dentin formed.

http://www.usc.edu/hsc/dental/ohisto/
Dentinogenesis

Dentin is formed by odontoblasts that differentiate from ectomesenchymal cells of dental papilla with influence from the inner enamel epithelium.

Differentiation of odontoblasts is mediated by expression of signaling molecules and growth factors in the inner enamel epithelial cells.

http://www.usc.edu/hsc/dental/ohisto/
## Time Line of Human Tooth Development

*(Table 5-2 in Text book)*

<table>
<thead>
<tr>
<th>Age</th>
<th>Developmental Characteristics</th>
</tr>
</thead>
<tbody>
<tr>
<td>42 to 48 days</td>
<td>Dental lamina formation</td>
</tr>
<tr>
<td>55 to 56 days</td>
<td>Bud stage; deciduous incisors; canines and molars</td>
</tr>
<tr>
<td>14 weeks</td>
<td>Bell stage for deciduous teeth; bud stage for permanent teeth</td>
</tr>
<tr>
<td>18 weeks</td>
<td>Dentin and functional ameloblasts in deciduous teeth</td>
</tr>
<tr>
<td>32 weeks</td>
<td>Dentin and functional ameloblasts in permanent first molars</td>
</tr>
</tbody>
</table>
Incremental pattern of dentin and enamel formation from initiation to completion

Growth areas of developing crown. Growth at cusp tip, intercuspal region, and cervical region

Development of root begins after the enamel and dentin formation has reached the future cementoenamel junction.

Epithelial cells of the inner and outer enamel epithelium proliferate from the cervical loop of the enamel organ to form the **Hertwig’s epithelial root sheath**. The root sheath determines if a tooth has single or multiple roots, is short or long, or is curved or straight.

[http://www.usc.edu/hsc/dental/ohisto/](http://www.usc.edu/hsc/dental/ohisto/)
Eventually the root sheath will fragment to form several discrete clusters of epithelial cells known as *epithelial cell rests of malassez*. These will persist in adults within the periodontal ligament.

http://www.usc.edu/hsc/dental/ohisto/
The epithelial rests appear as small clusters of epithelial cells which are located in the periodontal ligament adjacent to the surface of cementum. They are cellular residues of the embryonic structure known as Hertwig's epithelial root sheath.

http://www.usc.edu/hsc/dental/ohisto/
Primary apical formen

Epithelial diaphragm: the proliferating end of the root sheath bends at a near 45-degree angle. The epithelial diaphragm will encircle the apical opening of the dental pulp during root development.
Secondary apical foramen form as a result of two or three tongues of epithelium growing inward toward each other resulting in multirooted teeth.
Direction of root growth versus eruptive movement of tooth
Embryogenesis of the Periodontium

The PDL forms from the dental follicle shortly after root development begins.
Gingival sulcus

Free gingiva

Enamel space

Free gingival groove

Junctional epithelium

Attached gingiva
Figure from Ten Cate’s Oral Histology, Ed., Antonio Nanci, 7th and 9th editions