Tooth morphogenesis computational models

Tooth development constitutes a particularly interesting model for studying such epithelio-mesenchymal interactions (38, 39) and is an excellent subject for evolutionary studies (40) (9-11). The different induction stages that precede the morphogenesis and differentiation of the teeth result from reciprocal interactions among stomodeal epithelial cells and mesenchymal cells derived from neural crests (ectomesenchyme) (12, 13). The identification of numerous growth differentiation factors (14-17), transcription factors (18-21) and molecules of adhesion (22-24) expressed in the course of the development of the tooth have revealed associations of multiple genes with tooth morphogenesis. Studies on the functions of signals and tissue interactions in cultured tissue explants and in mutant mouse embryos have revealed inductive signaling and hierarchies in downstream transcription factors (25). Two computational models of tooth morphogenesis are proposed (41, 42). The models differ in the accuracy of their predictions and the detail and realism of the cellular behaviors and gene networks involved in tooth formation (43).

First, the Reaction-diffusion model which includes four cell behaviors: cells can secrete signaling molecules; cells can receive signaling molecules (and change their behaviors in consequence); cells can also divide and differentiate. The model includes a network of gene products that regulates these behaviors and interact between them (43). The model starts with four epithelial cells distributed in a regular rectangular grid. Three layers of esenchymal cells lie under these epithelial cells. All epithelial cells secrete activator at an intrinsic rate (k3) and also in response to the local activator concentration. Over time, in areas where the local activator concentration

exceeds a set threshold, the epithelial cells differentiate irreversibly into nondividing knot cells. These knot cells also secrete inhibitor at a rate equal to the local activator concentration. This inhibitor counteracts activator secretion and, in addition, enhances growth of the mesenchyme. The mesenchyme is mainly a three-dimensional space where diffusion and growth take place. (For more insight of the model please see (44, 45).

Another morphodynamic model included three more genes, BMP2, ectodin, and FGF-4. Ectodin is an extracellular sequesterer of several BMPs and in the model it acts by decreasing the concentration of free diffusible Bmp2 and 4. Fgf4 is also included. As in mouse teeth (46), it is secreted from the knots and it promotes proliferation of the underlying mesenchyme. Also as in mouse teeth (47), Bmp2 is secreted from the knots and enhances differentiation. In the model, Bmp4 is the activator and Shh is again assumed to be the inhibitor. These genetic differences, however, have a rather mild effect on model dynamics. The more significant changes have been made at the level of growth dynamics, cell biomechanics and proliferation. In contrast to Reaction-diffusion model, this model does not constrain cell position and displacement to a rectangular discrete grid. Instead, the cells in the epithelium form a grid that deforms and grows due to cell growth and division. The model only considers tooth development from the later moments of the bud stage. The model’s initial conditions consist of an initially flat epithelium. This epithelium represents the tip of the invagination before the first knot forms. The model starts with 19 hexagonal epithelial cells arranged in a hexagon with 20 layers of mesenchymal cells under them. Each cell has six neighbors and is situated at an arbitrary distance of 1 from

them. Cells in the borders have only 3 or 4 neighbors. Each cell is a three-dimensional volume that includes the cell itself and its immediate extracellular space. Molecular diffusion between two cells is proportional to the area of contact between those cells and their surroundings (finite volume method). This method is used because it allows accurate calculations even when cells change their shapes. The contour conditions for diffusion are the same than in Reaction-diffusion model.

Molecular studies have revealed that the instructive and permissive tissue interactions during mouse tooth development are mainly mediated by growth factor signaling. Development from initiation to eruption is governed by a sequential and reciprocal signaling process rather than simple one-way messages. The signaling involves all major signaling pathways, including TGF, FGF, Shh and Wnt as well as Eda, Notch, and EGF signaling, and studies with mouse mutants have shown that they are needed simultaneously during critical stages of development. Expression of signals is often redundant: several FGFs are expressed in the initiation stage epithelium, in the enamel knot and in the dental mesenchyme and they signal to receptors expressed differentially by mesenchymal and epithelial cells (48). Similar co-expressions are evident for BMP and Wnt signals. According to an in situ hybridization Turecková et al (49) investigated the expression patterns of the msx-1, msx-2, BMP-2 and BMP-4 genes, supposed to regulate early tooth development, in day 10-14 mouse embryonic upper diastema and molar regions, using 49 series of frontal sections. BMP-2 and BMP-4 expression was down regulated in the diastemal dental primordia during their regression starting at day 13. The temporo-spatial pattern of BMPs expression may be

associated with the disappearance of diastemal rudiments. Contrary to the molar anlagen, msx-2 gene expression did not detected in the diastemal dental rudiments after the stage of epithelial thickening. The deficiency of the msx-2 gene products may play a role in the growth retardation of diastemal dental primordia resulting in their subsequent involution.

Molar teeth in mice have attributes which make them more suitable for dental investigation than are rat molars. The albino mouse has a monophyodont dentition with a dental formula of incisors 1/1; cuspids O/O; premolars O/O; and molars 3/3; for a total of 16 teeth. The three molars arise successively in different periods of development with the first molars initiated first, followed by the second and third respectively. The first molars are the largest, and the third are the smallest .(22) Mice have only two tooth families, one incisor in the front and three molars in the back of each half of the jaws. The incisors and molars are separated by an area with no teeth, the diastema. However incipient abortive formation of dental germs is distinguishable in that space (33). It has been shown that mouse tooth germs in the diastema region degenerate at the early bud stage (50). Humans have extra tooth types in comparison to mouse. Human dentition consists of canines and premolars, which are formed by processes similar to tooth development in mouse but with higher levels of patterning complexity (51).

In the mandible the roots of the first and second molars are approximately of equal size, one mesial and one distal. The lower third molar has a single, short, stout root with very little tapering between neck of crown and tip of root. In the maxilla the three roots of the first molar are of unequal size: the

mesial is the longest and largest and has a distinct mesial inclination; the distal and palatine roots are about equal in length and size. The second upper molar has a fused palatine root that is broadest mesio-distally and contains two or three root canals; the remaining two roots of this tooth are placed mesially and distally with the latter being the longest of all three roots. The third upper molar has three very short roots: a mesial, a palatine and a distal of which the last is the longest. The cuspal pattern of the lower second molar of a newly erupted tooth appears to be a miniature of the human second lower molar. Roots of mature molars are similar in most respects to those of human teeth in the arrangement arid appearance of the tissues. There is one obvious difference the crown is being worn clown with relative rapidity bemuse of the enamel-free areas on the cusps