A Brief Review of Cartilage and Controlling Factors in Chondrocranial Morphogenesis

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INTRODUCTION

This paper deals with the cellular and major morphogenic properties of cartilage as applied to the ontogenesis of the chondrocranium in craniofacial morphogenesis (CFM). A brief review of cartilage and its growth and development intracacies necessarily come first, in a hierarchical sense, before the regulation of the chondrocranium. Despite the apparent instability of chondrocyte determination, chondrocranial growth during the early fetal period occurs in an ordered manner indicative of predominant genetic control.

CARTILAGE

Cartilage serves as a supporting structure, and through its proliferation allows increase in size of the organism. Ultimate ossification of the cartilage model provides a rigid support structure of even greater competence. The importance of cartilage and its structural and genetic properties is therefore crucial to an understanding of CFM: prenatally the chondrocranium and postnataally the synchondroses of the cranial base and cartilage of the nasal capsule which are primary growth determinants.

Cartilage is primarily composed of intercellular matrix, fibers and various cell types. What is interesting in terms of gene evolution is that there are alternative differentiation pathways open to the germinal cells (mesenchyme). And even beyond this seemingly labile system is evidence that there is some intrinsic instability of chondrocyte determination. In this regard, it is not uncommon for many cell types to differentiate as the result of environmental factors and not the result of any intrinsic programs. The harmony so characteristic of chondrogenesis may therefore be due to tissue interaction and chondrogenic modulation.

The primary matrix components of cartilage are chondroitin sulphates which
are mucopolysaccharides. When they are combined with a protein they are called protoglycans. The protein core appears to be specific to cartilage although it is similar to other cell types. The second major structural component of cartilage is collagen made up of units called tropocollagen which are several polypeptide chains arranged in a triple helix. These collagen molecules are arranged in fibers and constitute the fibrillar component as opposed to the ground substance mentioned above. The mechanisms by which the chondrocytes export these materials to the intercellular spaces is still unknown.

The differentiation sequence of cartilage begins with the germinal cell to the chondroblast. The chondroblasts are the so-called cartilage forming cells and their accumulation comprises the inner vascular layer of the perichondrium. From these cells differentiate the chondrocytes and then, significantly, hypertrophy of the chondrocyte, and finally the production of matrix by these chondrocytes. Hall (1970) has shown that normal chondroitin sulphate synthesis is necessary for chondrocyte hypertrophy and that "suppression of chondroitin synthesis may cause the germinal cells to remain in the osteogenic pathway" (Hall, 1970:390). It appears as though the chondrogenic pathway is determined by the accumulation of chondroitin precursors which stimulate the differentiation of the germinal cells through the sequence. Germinal cells which produce bone are apparently in a place where chondroitin sulphate synthesis is not favored, hence bone is produced.

Despite the synthesis of cartilage specific proteins by cartilage cells during their normal in vivo life span, there is an apparent instability of chondrocyte determination. In vitro experiments (Maclean, 1976) have shown that after prolonged periods of rapid division, chondrocytes seem to "forget" their commitment as the specificity of the genetic program is lost. "Examples of committed cells which lose or change their commitment are scarce in biology and they clearly are a potential source of information on how a genetic programme is normally fixed" (Maclean, 1976:157). In response to problems of cartilage tissue culture in vitro and the regulation of the differentiated state, Watanabe (1971) has been concerned with possible extrinsic factors which could account for this instability. Chondrocyte proliferation and differentiation usually regress (as discussed above), but if treated with a "conditioned medium" when cells are still young, those destined to regress are vitalized. And so there are indeed active factors responsible for abetting the gene control and allowing growth promotion. What these extrinsic substances are, however, we still do not know.

Hayflick (1980) has experimental evidence that normal human fibroblasts have a limited number of divisions. Furthermore, this limitation is controlled from within the nucleus. This points to a genetic-innate-phenomenon likely present in all cell types except cancer cells. In this respect, the supposed loss of chondrocyte specificity, may be a normal cellular aging process.

It may be then that chondrogenesis is strongly affected by the whole interactive system. "Interactions are both phylogenetically and ontogenetically older than early workers had fully appreciated" (Lash, 1974:209). In light of
this, chondrocyte determination may be an example of modulation; i.e.
significant control or effects on differentiation due to environmental factors and
not necessarily a change in genetic potential. This is quite common among
morphologic cell differentiation in embryogenesis. Germ cell differentiation
provides an excellent example. "Regardless of their genetic constitution
(-potential-) the early germ cells are bipotential and do not begin their
differentiation as eggs or sperms until after a certain residence within the
gonadal primordium" (Turner, 1969: 228, brackets mine). This is analogous to
the bipotentiality of germinal connective tissue cells discussed earlier.

CHONDROCRANIUM
The current census of opinion is that the primordeal cartilagenous cranial
skeleton (chondrocranium) grows in an autonomous manner and is under strict
genetic control. This is observationally apparent from embryological studies
indicating that the developmental events of all higher vertebrate embryos are
very similar (see De Beer, 1971, who described in great detail the morphological
similarity of vertebrate skulls in embryogenesis). The mechanisms of cartilage
and chondrocranial growth and development are not completely understood,
but one thing is clear, it occurs in an orderly, harmonious fashion. The
developmental integration from lower levels in the hierarchical organization of
vertebrates predispose the stability of the chondrocranium in evolution. G. L.
Stebbins (1969) coined the principle of Conservation of Organization reflecting
this view.

During the germ layer stage in embryogenesis, the ectoderm, mesoderm and
endoderm are discernable as a plate of cells which ultimately folds and fuses to
form the tubular embryo. At the cephalic end of the embryo, neural crest cells
migrate underneath the surface ectoderm to form cell populations of mesen-
chyme (embryonic connective tissue cells), possibly from inductive influences
from the underlying mesoderm (Johnston, 1972). Cell migrations are ventral,
posterior and anterior to the eye. Proliferation of the mesenchymal condensa-
tions in these areas contribute to the major swellings in the facial region.
Hence "virtually all the skeletal and connective tissues of the face and anterior
pharyngeal regions are consequently of neural crest origin" (Johnston,

The development of the chondrocranium from mesenchyme is a process
poorly understood. Carlson (1973), however, summarizes evidence that the
notochordal epithelium produces its own connective tissue sheath which may
actively direct or promote specific ontogenetic processes. Apparently the sheath
acts as or produces an inducer to promote cartilage synthesis. Maclean's (1976)
in vitro experiments also provide evidence that an inducer molecule is secreted
by the notochord and ventral spinal cord which promotes cartilage formation
from the mesenchyme cells. Hoyte (1975) reports on a series of experiments
designed to show the inductive influence of the notochord, cervical neural tube,
and even brain on the basioccipital segment. Fetal rat observations of the
developing cranial base also substantiate the notochords' role in development.
It has been shown that in the development of the cells in the basioccipital segment of the primordial cranial base, "the cells closest to the notochord were the most differentiated ones" (Dorenbos, 1973:65): see Figure 1.

![Figure 1](image)

Fig. 1. Cross section through notochord in the basioccipital segment in a 14 day rat fetus. (from Dorenbos, 1973: 64, reproduced with permission from the publisher, Swets and Zeitlinger B.V., Amsterdam).

The question that presents itself is now that chondrogenesis is underway, what factors influence its growth? Functional demand theorists find exception to embryonic growth and conclude that it is controlled by regulating factors other than physiological demand. So if the sole function of the embryo is to develop, then such factors as differentiation and changes in tissue mass may well be "the predetermined goals toward which embryonic feedback systems are directed" (Goss, 1964:46). The most reasonable conclusion to make from this is that the chondrocranium is under predominant genetic control.

Van Limborgh (1972) has designed a convenient model for expressing controlling factors in CFM and explains that the high degree of autonomy of chondrocranial growth is assumed to be almost exclusively controlled by intrinsic genetic factors. His model, however, also includes epigenetic factors which are defined as "genes which exert their influences outside the cells or tissues to which they are inherent" (Van Limborgh, 1972:38); i.e. originating from the growing adjacent structures within the cranium and face. This
definition accords quite well with Waddington (1962). He defines epigenesis as, "whose final state is not determined by the initial conditions" (Waddington, 1962:47). This statement refers to Waddington's 'epigenetic action system' where gene action is separated by a numerous set of interactions from the gene level to the subsequent phenotype. Van Limborgh's utilization of this concept is in regard to genetic-programmed growth of a structure and its ultimate epigenetic affects (spatial, mechanical, etc.) on surrounding structures. Van Limborgh's model also includes local environmental factors; i.e. originating from forces such as produced by the muscles of mastication.

These factors lend some suspicion to the view that chondrocranial growth is under predominantly genetic control. Even Van Limborgh, in a discussion on periosteal and sutureal tissues, points out that these tissues are controlled by epigenetic and local environmental factors and that the growing cartilage and bone may play a key role as epigenetic influences. These influences are themselves directed back towards the structures that are producing them; note that this illustrates "functionalism" prenatally — to co-ordinate a whole integrated suite of growing structures in an orderly manner. From a biosynthetic and tissue level of discourse on differentiation (see above), further environmental influences might be hypothesised. The most amenable conclusions concerning prenatal growth and development at this time should reflect an integrated approach:

1: Predominantly genetic and tissue interaction influence on:
   a. mesenchymal migration and proliferation,
   b. notochordal induction of chondrogenesis, and
   c. formation of the primordial chondrocranium.

2: Increased epigenetic and local environmental influence on:
   a. the early-mid-fetal life period when various centres of ossification in the craniofacial region commence, and
   b. the genetic potentiality of cartilage, hence chondrocranial growth, and
   c. the various structural elements — responding to the forces exerted by the surrounding growing structures.

**SUMMARY**

It is evident that chondrocyte determination is an example of cellular aging. This possibly leads to the erroneous view that chondrocytes have a "weak" genetic control mechanism. If such was the case, chondrocranial growth and development would be a very "shaky" process indeed. It is recognized however that chondrocranial growth is under predominantly genetic and tissue interaction influence especially during the early phase of growth. These control factors plus other epigenetic and local environmental influences are responsible for the harmonious growth of the chondrocranium.
Key-Words: cartilage — chondrocranium — chondrocyte — determination — craniofacial morphogenesis.

REFERENCES


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