Development of the squamosomandibular articulation in the Mongolian gerbil (Meriones unguiculatus).
II. Succinate dehydrogenase activity

LESLIE P. GARTNER, JAMES L. HIATT, M. A. KHAN and D. VINCENT PROVENZA

Department of Anatomy, Baltimore College of Dental Surgery,
Dental School, University of Maryland at Baltimore, USA

Synopsis. Succinate dehydrogenase activity has been studied, according to the method of Nachlas et al. (1957), in the developing tissues forming the squamosomandibular articulation in the Mongolian gerbil from its inception through the sixth postnatal day. Increased activity was observed in the chondroblasts, osteoblasts and mesenchymal tissues of the developing articulation. The chondroclasts of the developing mandibular condyle displayed intense reaction as did the osteoclasts of the developing bony articulation. Succinate dehydrogenase activity appeared to be related to the functional maturity of the cellular elements of the developing joint.

Introduction

Succinate dehydrogenase, a mitochondrial enzyme, catalyses the formation of fumarate from succinate in the tricarboxylic acid cycle (White et al., 1972). It is one of the principal enzymes concerned with oxidative phosphorylation, for it connects the glycolytic pathway via the tricarboxylic acid cycle to the electron transport system. Consequently its presence, and more specifically its degree of activity, is directly related to cellular metabolic levels. Biochemical investigations of dehydrogenase levels in the chick epiphysis (Lee & Shapiro, 1974) indicate that succinate dehydrogenase is the most active of all mitochondrial dehydrogenases. Histochemically, dehydrogenases have been localized in various hard tissues, such as the odontogenic organ (Burstone, 1960a; Provenza et al., 1972) and bone and cartilage (Tonna, 1958; Burstone, 1960a, b; Eng & Esterly, 1972; Fischer, 1973, 1974, 1975). However, relatively few studies concerning the activity of succinate dehydrogenase in bone and cartilage have involved oral tissues (Burstone,
Gartner, Hiatt, Khan and Provenza

1960a, b; Fullmer, 1964), and none were performed on the developing mandible. Yet, this is an especially interesting osteogenic region, for the formation of the mandible and its ramus, condyle and coronoid process involve an intimate association between intramembraneous bone formation, secondary cartilage formation and the subsequent resorption of Meckel's cartilage (Bhaskar et al., 1953; Frommer & Margolies, 1971; Hall, 1970; Raitova, 1971; Savostin-Asling & Asling, 1973; Gartner et al., 1976; Meikle, 1976; Trevisan & Scapino, 1976).

The purpose of the present investigation is to examine the succinate dehydrogenase activities of the tissues involved in these three contiguous phenomena in the late prenatal and early post-natal Mongolian gerbil.

Materials and methods

Animals selected for this study ranged in age from 22 days of gestation to six days post-natal. All foetuses (22) and neonates (24) were sacrificed by chloroform inhalation followed by decapitation. The severed heads were immediately frozen on the rapid freeze bar of a Lipshaw cryostat and were mounted on a chuck using Cryofoam. Consecutive serial sections, 14 μm thick, were recovered in the frontal plane, while some heads were sectioned in the horizontal plane.

Demonstration of succinate dehydrogenase activity was accomplished by incubation of the air-dried sections in a Nitro BT medium (Nachlas et al., 1957) for a period of 13 min at 37°C. Blue diformazan deposits, resulting from the reduction of Nitro BT as the final electron acceptor, were observed as bluing of the tissue sections.

Subsequent to incubation, the tissues were counterstained with Safranin O, fixed in 10% neutral buffered formalin, dehydrated in xylene and mounted in Permount. Controls were prepared in the identical fashion, except that the substrate was deleted from the incubation medium.

Results

Based on subjective, visually-quantified increases in reaction deposits within the cells, the following terminology was used to describe the observed succinate dehydrogenase activity: negative, slight, moderate and high. These terms were also numerically coded from 0–3 for tabular summarization (Table 1).

Prior to the 18th day of gestation, no evidence of the gerbil's mandibular joint could be detected (Gartner et al., 1976). While on that day the mesenchymal condensation, presaging the future squamomandibular joint, evidenced slight to moderate succinate dehydrogenase activity (Fig. 1). The presumptive lateral pterygoid muscle also displayed slight to moderate activity (Fig. 1). By the 22nd day of gestation the condylar region of the squamomandibular joint possessed a cartilaginous core (Gartner et al., 1976) and the chondrogenic cells exhibited slight to moderate enzyme activity. The chon-