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GRADUATE STUDIES

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THE EFFECTS OF ELECTRIC CURRENT ON CONDYLAR CARTILAGE AND BONE GROWTH

by

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Department of Orthodontics

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Abstract

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Recent investigations have shown a correlation between the synovial fluid pressure relationships generated by mandibular postural appliances and the growth of the condyle. It has also been shown that when bone is deformed, an electric potential is generated. In turn, stimulation of bone growth by electric current has been demonstrated. It is thus possible that, in the absence of mandibular postural appliances, delivering an electric charge directly to the condylar cartilage through the synovial fluid may provide for increased growth of the condyle.

This investigation was designed to study the effects of a constant direct 20 uA current on condylar cartilage and bone growth in a young, growing animal. Platinum-irridium electrodes were placed near the lateral junction of condylar cartilage and bone. Because of the nature of the temporomandibular joint (TMJ), when the electrodes were in place they were surrounded by the synovial fluid of the TMJ. Initial and final radiographs, as well as serial weights, were obtained. Voltage across the electrodes

was measured every other day. Vital dyes were given at the beginning, midpoint, and end of the 30 day experimental period. After the animals were killed, histologic and ground bone sections were prepared and analyzed along with the rest of the data.

The findings of this investigation can be summarized as follows:

1. Histologically, the current caused a thickening of the prechondroblastic and matrix cartilage forming zones, thickened and more distinct layers between zones, and increased cellular size with more eosinophillic material present. Chondroid bone formation was occurring near the site of stimulation. These results taken together, indicate an increase in the rate and amount of cartilage and bone growth when compared to nonstimulated controls.

2. The histologic appearance caused by the electric stimulation is similar to that caused by a mandibular protrusive appliance.

3. There were grossly observed directional growth changes in the gonial angles and coronoid processes of the stimulated animals.

4. There were marked changes in the incisal wear patterns of stimulated vs. control animals. Incisor wear was indicative of a mandibular protrusion.

5. There was an observable increase in the anterior posterior thickness of the glenoid fossa of the stimulated condyles.

6. The stimulated animals showed an equal or greater amount of change in the distance from lower molar to lower incisor tooth positions.

7. Microfluorescent study of ground bone sections indicated an increase in the vertical heights of the stimulated condyles. There was also a reduction in the trabeculation patterns with dishing and indentations of the condylar surfaces.

8. The stimulated condyles showed an increase in the projected paper weight measurements. This was also obvious on visual examination as the stimulated condyles were grossly larger.

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DEDICATED

TO

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INTRODUCTION

The control of mandibular growth has been a perplexing problem for the orthodontist. It is difficult to predict, difficult to augment, and difficult to restrict. Since mandibular growth is such an integral part of classical orthodontics, it is important to be able to better understand and control this process.

Some of the factors thought to be involved in the control of the growth process include genetic control, differential cellular responses, membrane control, hormonal factors, neurotrophic control, oxygen tension, pH levels, temperature effects, vasomotor control, cyclic AMP and GMP, enzymatic factors and bioelectric potentials. The question is, which are the specific trigger factors and which are the subordinate or secondary ones (Enlow, 1973).

In orthodontics, only mechanical force is used to stimulate or restrict mandibular growth. On the other hand, within the last two decades, exogenous electric currents have been employed, both clinically and experimentally, in successful attempts to initiate osteogenesis in intact bones or to enhance bone apposition in the healing of fractures (Brighton et al., 1975). Further, there is evidence that supports the possibility that electric currents are generated within stressed tissues (Bassett and

Becker, 1962). These currents may charge macromolecules that interact with specific sites in cell membranes or mobilize ions across cell membranes (Zengo et al., 1974). Since mechanical force is a stress, it is possible that an electric current could replace this force and be able to stimulate cells to perform specific functions.

Until now, no research has been done on direct electric current stimulation of temporomandibular joint (TMJ) condylar cartilage and bone. This seems to be an ideal location for current application since the condyle has a naturally enclosed fluid space surrounding it which could help in charge distribution. Davidovitch et al. (1980a,b) have observed that electric currents can activate a large number of cells in a small, well delineated area. Since the TMJ capsular area represents a small enclosed space containing essentially a homogeneous charged solution, this area appears ideal for current application. This investigation attempted to cause stimulation of condylar cartilage and bone through the application of a direct constant current.

LITERATURE REVIEW

Discovery of Bioelectric Potentials

Bone healing by the use of electricity was used as early as the 19th century by Lente (1850). Its application was emperical, and although success was reported, there was little interst in pursuing this phenomenon by the scientific community.

Real interest in the effect electricity has on bone was caused by Fukada and Yasuda (1957). They reported that electric potentials are generated by the application of force to bone. The potentials were proportional to the amount and character of the applied loadings. They were negative when the tissue was under compression and positive when the tissue was under tension. They attributed this effect to the piezoelectric nature of the crystalline structure of bone. The concept of piezoelectricity is, in brief, that bioelectric charges are produced when the crystalline structure of bone is deformed due to a mechanical stress.

Working independently, Bassett and Becker (1962) also showed that bone produced electrical potentials when deformed. They found that the amplitude of the electrical potentials created in stressed bones was dependent on both the rate and magnitude of the deformation. Polarity was dependent on the direction of bending. Compressive areas were found to be electrically negative

in relation to other areas. They proposed that the stress induced bioelectric potentials were the signals in the operation of Wolff's law, and that these signals control both bone and cell activity. It should be noted that Wolff's law simply states that bone becomes adapted during its growth to the functional forces acting on it.

Friedenberg and Brighton (1966) and Friedenberg et al. (1971) measured spontaneous bioelectric potentials that exist in bone independent of any stress. They found that a potential does exist in vivo and will persist for several hours in freshly removed bone. It was also found that the diaphysis carries a positive charge in relation to the epiphysis or metaphysis. With the creation of a fracture, Friedenberg and Smith (1969) found that the normal electrical pattern is changed such that the entire bone becomes more negatively charged. The greatest increase in negativity occurred at the fracture site.

Friedenberg et al. (1971) speculated that the increase in negativity associated with compressed bone could be caused by many factors including:

1. An increase in vascularity with the red blood cells and endothelium negatively charged.
2. A higher protein to mineral ratio.
3. An increase in the overall cellular content.
4. pH changes in the healing of bone.
5. A different ionic concentration caused by the altered tissue metabolism.

Electrically Induced Bone Formation

Yasuda et al. (1955) were the first to report induced formation of a bony callus on a femur. This result occurred after a 100 microampere (uA) current was delivered over a three week period. If the current was raised above 1 milliampere (mA), bone destuction occurred.

Bassett and co-workers (1964) induced significant bone formation around surgically implanted cathodes in the femurs of adult beagle dogs. There were three current levels delivered: 1, 10 and 100 uA. The 1 uA current produced no differences in bone formation in comparison to controls; however, the 10 uA and 100 uA currents produced new bone after fourteen days. The tissue response around the anodes was similar to non-active electrodes in the contralateral control femurs.

O'Connor et al. (1969), repeated these experiments and found a similar, though not identical response to that reported by Bassett. In 7 out of 12 cases, more bone was formed around the cathode. In 2 cases however, less bone was found near the cathode. They cautioned that although the response appeared real, considerable species variation could be present.

Other researchers had difficulty duplicating the success of Bassett and O'Connor. Minkin et al. (1968), found that a 70 uA current may induce bone growth, but the increased deposition was not significantly different from that caused by sham electrodes.

Crelin and Deuker (1970) stimulated mice femurs with a 100 uA current. The amount of bone formation around the electrodes was never more than, and often less than, the bone formed around the sham electrodes.

Because of the wide variation in bone response to electric current, Lavine et al. (1969) and Lavine (1971) designed experiments to test what the response of bone was when it was located in the path of an electric current. In these experiments, any foreign body response to the electrodes would not be confused with the response caused by the current. An experimental defect, simulating a fracture, was created in rabbit femurs, and electrodes delivering 2 to 4 uA were implanted on each side of the defect. Results indicated a two-fold increase in the rate of healing when current was used. Lavine concluded that the electric current increased the rate of the calcification process.

In previous studies, it was observed that as resistance increased, the amount of current delivered to the bone decreased. This is in accordance with Ohm's law (Voltage = Current X Resistance). In an attempt to get around this problem, Friedenberg and associates (1970) studied bone response to a constant current source. This source was designed to deliver a constant amount of current despite an increase in resistance. Using this device to stimulate the femur of rabbits, they observed bone and cartilage formation, fibrous tissue proliferation, and tissue destruction at both the anode and the cathode. The reaction intensity appeared

to be related to the current density. Bone production was most frequently observed at the cathode at a current level between 5 and 20 uA. Above 20 uA, tissue destruction occurred, especially at the anode.

Friedenberg et al. (1971a) used this same constant current device to increase the healing rate of a fractured rat fibula. With these findings in mind, Friedenberg et al. (1971b) applied this treatment regimine to a non-union fracture in a 51 year old women after a conventional attempt of immobilization had not produced bony union. Using a 10 uA current with the cathode implanted in the fracture site, union was achieved in nine weeks.

Lavine et al. (1972) used approximately the same treatment to achieve bony union in a patient with congential pseudoarthrosis. This patient had not responded to conventional treatment of surgery and stabilization, and was a candidate for amputation. The 3.9 uA current produced, after a period of 4 months, osteogenesis and collagen formation.

As a further attempt at minimizing electrode polarization, and consequently increasing the currents effectiveness, Levy (1971) and Levy and Rubin (1972) used a pulsed monophasic direct current source to induce osteogenesis in femurs and ribs of dogs. The average increase in bone area for all the pulsed stimulation was 43%, while the average increase for the direct current stimulation was only 17%. Sham electrodes produced a 3% increase in bone area. The range for the pulsed current was 0.2 to 2.5 mA and

all direct current stimulators were set at 0.5 mA (500 uA), far above the optimal range reported by Friedenberg and associates (1970). This may have been the reason for the poor response of the bone to the direct current. With the pulsed current, the most active region of bone formation was between the electrodes with no preference for either electrode.

Stefan (1976) exposed fractured rabbit tibias and femurs to continuous and oscillating direct currents. He found that a 10 uA oscillating current reduced the period of healing from 40 to 15-20 days. Continuous current was less effective.

Martin and Gutman (1978) exposed immobilized rat femurs twice a day to an oscillating 30 Hz current, 200 V peak to peak. After 28 days, disuse osteoporosis had not developed. Instead, there was increased cortical area, bone mass, density and percent ash weight.

All previous studies involved the application of some type of current directly to the area to be stimulated. Davidovitch et al. (1980) noninvasively stimulated periosteal bone growth by the application of direct current to adjacent gingiva. He found increased staining intensities for cAMP and cGMP as well as a changed distribution pattern, indicating an increased level of cellular activity in the periosteum. It was therefore possible to noninvasively stimulate periosteal bone growth through the adjacent gingival tissue.

Pulsed Electromagnetic Fields

To use a direct current, either constant or pulsed, surgical procedures are often required for the implantation and removal of the electrodes. Because of the possible risks and complications involved, investigators sought a means of electrical stimulation that could be applied externally, without tissue contact, and cause bone growth. Such a device currently being studied involves a pulsed electromagnetic field (PEMF). The current is pulsed through tissue via external electromagnets and creates a field of stimulation between the electromagnets.

To investigate this phenomenon, Norton (1982) perturbed a mixed chondroblastic tissue with a PEMF. Some of the cultures were purposely retarded in growth to resemble an atrophic union, while others were grown to confluence in about half the normal time as a model for hypertrophic non-union. The PEMF appeared to promote normal healing. It was suggested that healing was accomplished by altering the cartilagenous lysosome activity in the matrix and possibly the sequence of events leading to calcification.

Bassett et al. (1979) exposed isolated chondrocytes from embryonic chick tibia to a PEMF. From this investigation they suggested it was possible to modulate the calcium handling ability of chondrocytes with PEMF's and that this effect could be responsible for the initiation of healing of non-unions. In addition, it appeared that the effects of the PEMF's are different from the

increased osteogenesis produced more directly by implanted electrodes delivering a direct current.

Possible Mechanisms of Biolectric Control

Functional forces have an important role in the shaping of the human face (Enlow, 1982). In order for this shaping to occur, bone must have the ability to respond to a change in mechanical stress. Since the response to a mechanical stress is similar to that from an electrical current (Lee et al., 1982), electricity should have a similar effect on bone as a mechanical stress.

The search for the ultimate signal that elicits the response of bone to a mechanical stress or electrical current has been long and intense. Many investigators have proposed various signals and theories, but to date no one is entirely sure what one, or combination of several, signals must occur before a response is elicited.

When an electric current is applied to a biologic system, Bassett (1971) found that the pH of the area changes. At the anode, acidic conditions are present and lysosomal enzymes and acid phosphates are more abundant in this area. This is consistent with the finding of bone resorption occurring at the anode. In alkaline conditions, he found insoluble calcium salts, osteoblasts and alkaline phosphatase. Since alkaline conditions are created near the cathode, this agrees with data showing osteogenesis occurring at the alkaline cathode. The changed pH of

area, caused by an electric current, could be a factor in the control of osteogenesis.

Brighton (1966) found that the growth apparatus of the epiphyseal plate was extremely sensitive to prevailing oxygen tension. He found that the growth rate was greater in low oxygen tension and that the oxygen tension on the cartilage side of the epiphyseal plate was different from that on the bone side of the epiphyseal plate. He also found (Brighton, 1971) that a gradation of oxygen tension exists in the epiphyseal plate that corresponds to the observed morphologic zones. He speculated that oxygen tension may be a physiological control mechanism governing growth of the epiphyseal plate. It was also shown that the reaction at the cathode results in oxygen consumption and hydroxyl radical production (Brighton et al., 1975). The formula $2H_2O + 4e^- + O_2 \rightarrow 4OH^-$ was proposed to substantiate this finding. This formula, along with the finding that the reaction at the cathode consumes oxygen, supports the idea that oxygen tension is lowered and local pH elevated at the cathode where bony production occurs.

Stephans (1969) found that pH changes and calcium ion levels alter the permiability of the cellular membrane. Lowenstein (1967) found that intercellular membrane connections were sensitive to calcium ion changes. When a low calcium ion concentration exists, these "tight" junctions" are normally in close apposition. When the calcium ion concentration is high, they tend to separate. It was theorized that junctional control could act

as a means of communication in a colony of cells and may explain why an undifferentiated cell can become more specialized (Becker and Murray, 1967). From these findings it is possibe that electric current causes changes in both the pH and calcium ion levels present, leading to changed cellular communications, specialization or growth alterations.

Lavine et al. (1974) reported that cells in bone fracture sites treated by direct currents presented an increased number of membrane bound vesicles, a morphologic transition to increased levels of rough endoplasmic reticulum with ribosomal lined cisternae and vacuolization of mitochondria. These responses are indicative of increased cellular activity. It was also reported that mitochondria are sensitive to electrostatic charges and changes in calcium ion concentration (Lehringer, 1969). Since electric current does cause an increase in bone deposition, and mitochondria are essential for this process, electric current could be somehow directly or indirectly effecting the mitochondria.

Ionic migration is also known to occur in the presence of electric or magnetic fields. Because of this, Bassett (1971) suggested that since ions are important in enzyme synthesis in a cell, the electric field could alter the rate of enzyme synthesis and enzyme substrate reactions by changing the relative ionic concentrations.

Bourret (1976) found that pressure caused uptake of Ca^{++} into the cells which appears to have a depressive effect on the

enzyme that causes in cyclic AMP (cAMP) reduction. By depressing this enzyme, more cAMP is present. Since an electric current has an effect similar to pressure application, a current may cause increased uptake of Ca^{++}, with a corresponding increase in the amount of cAMP available for cellular use.

If electricity does indeed activate the cAMP system, then the electron or charge could act similar to a hormone in being the first messenger to the bone or cartilage cells. Norton (1977) found this kind of evidence when he saw an increase in cAMP upon electrical stimulation of epiphyseal cartilage. He speculated that cAMP change is the early cellular response to an electrical or mechanical perturbation which leads to bone remodeling. Cellular cAMP increases have been considered to be an intracellular signal for DNA synthesis and cell division (Pastar and Perlman, 1971).

Rodan and Johnson (1981) hypothesized that continuous direct currents do not actually penetrate cellular membranes, but cause electrochemical changes in the cellular membrane. These changes range from an ionic redistribution to a translocation of charged molecules on the cell surface. This response is similar to changes in membrane permiability before hormonal binding by surface receptors. Since current does not appear to penetrate cellular membranes, Rodan (1982) believes that a second messenger such as cAMP may actually cause the elicitation of the response.

Norton et al. (1977) showed significant alterations in the

cAMP content of cartilage cells in vitro in less than 15 minutes, also suggesting that cAMP may play a role in the initial response of cartilage cells to electrical stimulation. They also found elevated levels of cAMP at the anode which perhaps signals the early extrusion of substances involved in the degradation of mineralized soft connective tissue. Increased cAMP levels found later may be related to the continued synthesis of enzymes and acids that participate in the resorptive process.

Davidovitch (1980) found that a constant current could reach bone periosteal osteoblasts non-invasively and that exposure of gingiva to this current resulted in an increase in the number of cells stained for cAMP and cGMP. He to believes that these substances, cAMP and cGMP, are a type of second messenger, delivering the electric signal to the bone cells to increase osteogenesis.

Present knowledge seems to indicate that cellular activation occurs in or through the cellular membrane. Stimulation of cellular activation need not be invasive. Stimulation involves the influx of ions such as Ca^{++}, Mg^{++}, Na^{++}, K^{+}, Cl^{-} and inorganic phosphate as well as activation of the membrane bound enzymes adenylate cyclase and guanulate cyclase. Within the membrane, the enzymes act upon their respective substrates ATP and GTP, to produce cAMP and cGMP. These later substances, along with Ca^{++}, are considered to be an intracellular "second messenger" system which promotes the effects of external stimuli on specific target

cells. The increased levels of cyclic nucleotides and Ca^{++} can lead toward heightened enzymatic phosphorylation reactions, increased synthetic and secretory substances, and an enhanced rate of tissue formation.

STATEMENT OF PURPOSE

The use of direct electric current to stimulate bone growth has had much success. Brighton et al. (1982), has reported healing in 81% of the fractures of long bones stimulated with direct current that had previously gone nine months with non-union. Although the amount of successful healing varies from bone to bone, and with the amount of immobilization present, direct current stimulation is nonetheless a viable means of eliciting increased osteogenesis.

Since there is a preponderance of evidence that a constant direct electric current will elicit increased intramembraneous long bone growth, it is tempting to stimulate an area of endochondral bone, never before stimulated in the literature, that is in a rapid phase of growth. The purpose of this investigation is to determine if a constant direct electric current will cause increased condylar bone and cartilage formation in a growing animal.

METHODS AND MATERIALS

General Description

Ten newly weaned white female New Zealand rabbits (orycto-lagus cuniculus), 4-5 weeks old, were the experimental animals. Rabbits were chosen due to:

1. the ease of access to the condylar area,
2. morphology of the condylar area,
3. rapid phase of growth from 5 weeks to approximately 12 weeks,
4. ease of handling, and
5. cost.

Platinum-iriduim electrodes were implanted in the TMJ. The electrode leads exited the nape of the neck where they were connected to a battery pack which was immobilized on the back of the animal by a leather harness. The current (Frierdenberg et al., 1970) was adjusted to deliver 20 uA constantly. Three different vital dyes were given at the beginning, midpoint and end of the experiment. Animal weight was periodically recorded. Radiographs were taken after implantation and at death. At the end of the 30 day experimental period, histologic studies were accomplished.

Experimental Design

This study tested the effect of a 20 uA constant direct electric current on either the right or left condylar area of a young rabbit.

The total experimental group consisted of ten rabbits. Six of the rabbits had identical electrodes implanted near the lateral side of both condyles. The tip of the electrode was placed at a lateral junction of condylar cartilage and bone. The electrode tip did not actually touch the bone but lay in close proximity to it being bathed in synovial fluid. One condyle received stimulation while the other served as a sham control. Two additional rabbits had surgery only on one side of the head. Electrodes were placed as above and electric current was delivered to both animals. An additional two rabbits had no surgery and served as normal growth controls.

Electrodes

Teflon coated 30 gauge platinum iridium (90:10) electrodes (Grass Instruments, Quincy, Massachusetts) were used for both anode and cathode. Before implantation, two sutures were placed through the rubber sheaths surrounding the electrode lead so that the electrodes could be firmly secured in position. Three millimeters of teflon at the tip of the electrode was removed before implantation.

Interface Fabrication

The electrode leads extended under the skin from the

condyles to the back of the head. They exited the skin through an interface made of dacron mesh and Silastic. Each interface was made by folding a piece of sterile proline (Ethicon Inc., Somerville, New Jersey) in half and placing metal rods, with the same diameter as the electrode lead wire, through the mesh. Silastic 382 (Dow Corning, Midland, Michigan) was applied to the mesh to form a cylinder at the fold. After the metal rods were removed, holes were present to securely hold the lead wires (Figure 1).

Surgical Implantation of Electrodes, Leads and Interface

The rabbits were anesthetized with an intramuscular injection of Rompun, 5 mg/kg body weight (Bayvet Division, Cutter Laboratories Inc., Shawnee, Kansas). After a period of 5 minutes, an injection of Ketamine HCL, 25 mg/kg body weight, was given (Ketaset, Veterinary Products, Bristol Laboratory, Syracuse, New York). This produced satisfactory anesthesia for general surgery for 30-40 minutes. If the procedure extended beyond this time, the anesthesia was augmented by additional Rompun (1 mg/kg body weight).

After routine shaving and cleansing, an incision was made from approximately mid-temporalis to mid-cheek. Immediately before the incision was made, 0.20 cc of Lidocaine was given subcutaneously at the surgical site to minimize bleeding. Care

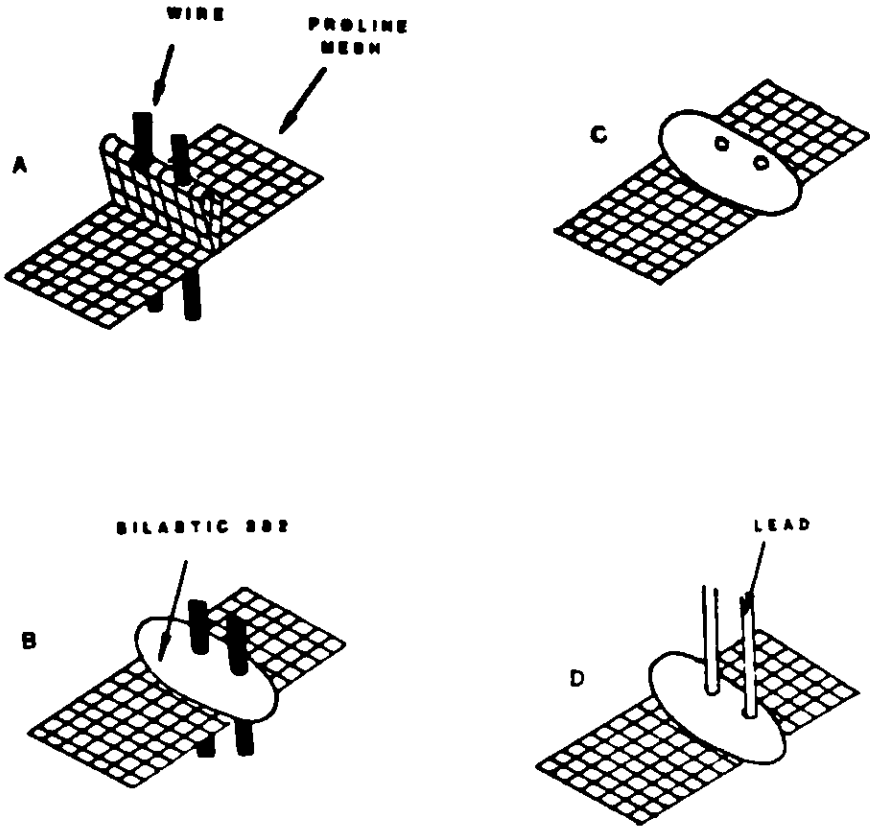


Figure 1. Interface fabrication

was taken such that the incision involved only the skin.

After reflection of the skin, an incision was made along the zygomatic arch for ease of periostium reflection. After reflection was complete, two holes were drilled through the zygomatic arch the same distance apart as the sutures in the rubber sheath of the electrode. Each suture was then passed through a hole in the zygomatic arch. Electrode placement was achieved. The electrode was secured in position by tieing the sutures which brought the electrode sheath securely against the zygomatic arch.

A second electrode was positioned in ear cartilage 1 cm from the tip of the condylar electrode and secured in place. This was accomplished by passing the sutures in the electrode sheath through the ear cartilage and securely tieing them (Plate I).

An incision was made on the back of the head and a blunt probe was inserted subcutaneously from this incision to the incision on the side of the face. Both leads were then threaded through this tunnel opening. Electrode color and position was noted. The incision on the side of the face was then washed with saline and closed with interrupted 4-0 silk sutures.

The same procedure was performed on the contralateral side of the head. The leads from the side selected for electrical stimulation were left long enough for attachment to the constant current device, while the leads from the opposite side were cut off and tucked under the skin at the nape of the neck.

PLATE I

Surgical procedure--electrodes in place.



The longer leads were then threaded through the interface which was inserted into the incision on the back of the head. The proline flaps were spread laterally under the dermis. Sutures were placed through the proline and secured to the adjacent skin. After healing was complete, the mesh produced an excellent mechanical lock (Gertter et al., 1971). The lead wires were secured to the interface with Silastic Medical Grade A Adhesive (Dow Corning, Midland, Michigan). The incision was washed with saline and closed with interrupted 4-0 silk sutures. Leads were connected to the power unit and initial measurements made 20 minutes post-incision closure (Plate II).

Power Unit

Each power unit contained a 9 volt mercury battery (Union Carbide Corporation, New York, New York) and was designed to deliver a constant 20 uA current (Figure 2). Once a variable resister was set for this amount of current, the circuit continued to deliver this level despite changes in resistance. Resistance could approach approximately 450,000 ohms before the circuit could no longer deliver this current level due to insufficient battery voltage. Each circuit was encased in a plastic box which was attached to a small leather animal harness and secured to the animal (Plate III).

PLATE II

Dorsoventral radiograph showing electrodes in place.



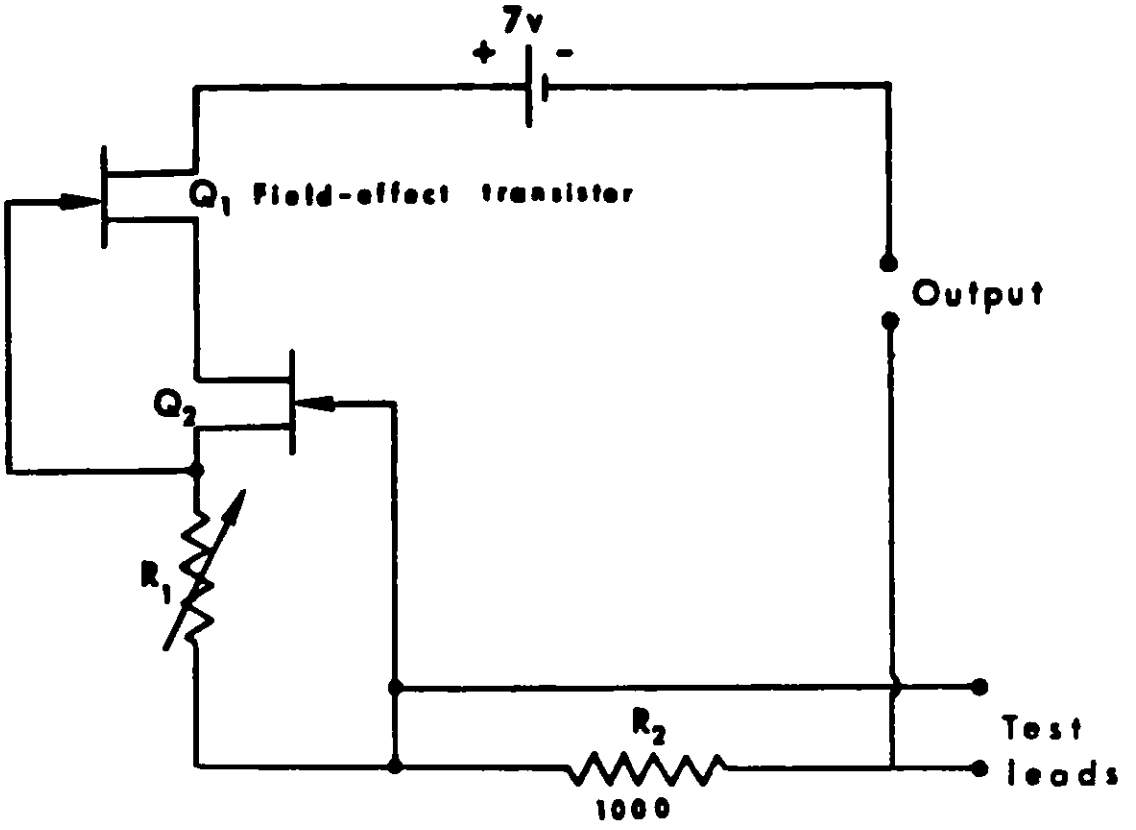


Figure 2. Diagram of circuit (from Friedenberg et al., 1970).

PLATE III

Power unit on animal



Each power unit was checked prior to and at the termination of the experiment. Battery voltages were also measured at this time. Measurements of voltage across the electrodes and total circuit resistance were made every other day during the experimental period and adjustments of the variable resistor were made in order to keep the current at 20 uA.

Cephalostat

A cephalostat was obtained for reliable positioning of the head for radiographic comparisons (Plate IV). The source film distance, object film, and source to object distance were kept constant for each animal.

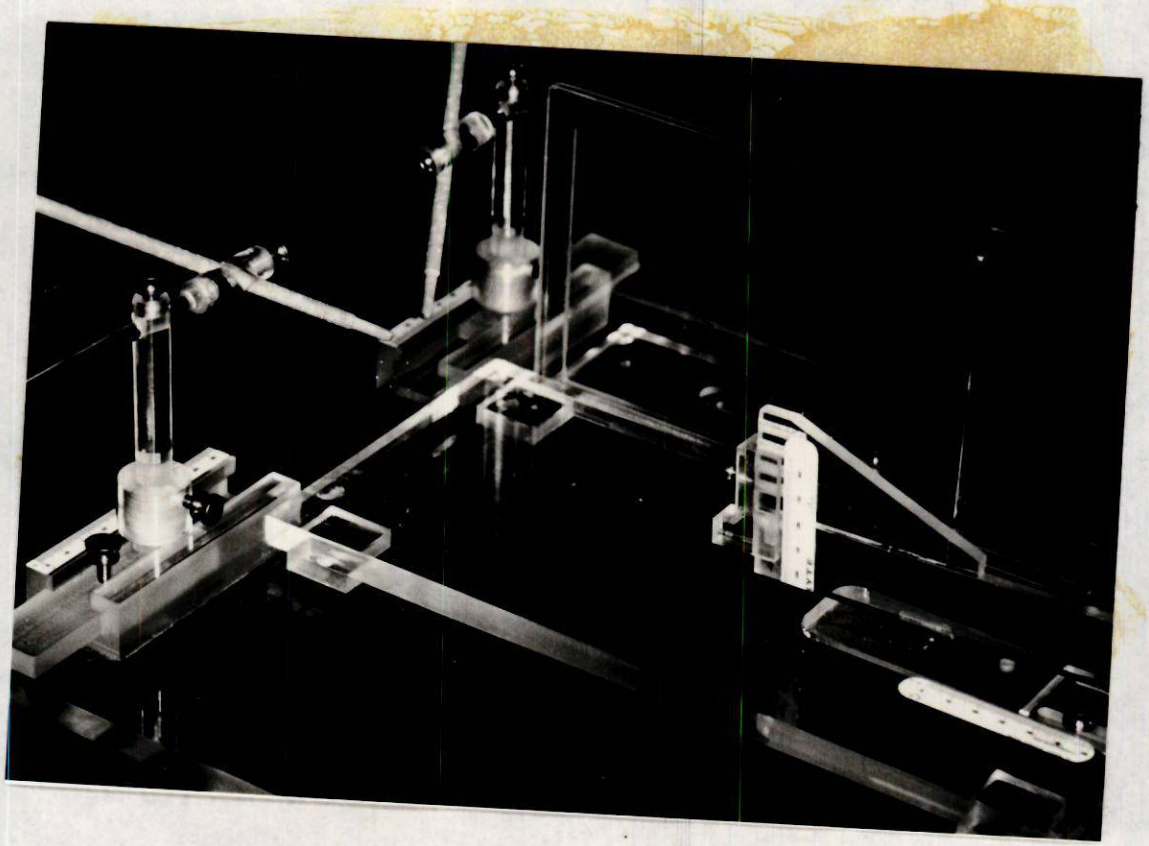
The head of the rabbit was held securely by two ear rods. A rubber band was placed around the snout to hold the mandible closed and the incisors were positioned on a small moveable metal bar. Both lateral and dorsoventral radiographs were taken post-surgery and again after the animals were killed. An exposure of 1.5 seconds at 90 KVP and 15 ma proved adequate.

Vital Dyes

Immediately after the surgical procedure intramuscular Oxytetracycline, 50 mg/kg body weight was administered (Pfizer Laboratories Division, Inc., Pfizer Inc., New York, New York). This was repeated again on day four. Oxytetracycline was chosen

PLATE IV

Cephalostat



as the initial vital dye because it has an antibiotic as well as a staining effect.

On the fifteenth experimental day, Alizarin complexon, 30 mg/kg body weight (Eastman Kodak Company, Rochester, New York) was mixed in 1 cc of a 2% $NaHCO_3$ solution and administered intraperitoneally. On the 28th experimental day DCAF, 15 mg/kg body weight (2,3 BIS) N, N^1DI (Carboxymethyl) (Aminomethyl Fluorescein) (Life Sciences Group of ICN Pharmaceuticals, Inc., Cleveland, Ohio) was mixed in 1 cc of a 2% $NaHCO_3$ solution and was administered intraperiotoneally.

Histologic Procedures

After the animals were killed, the mandibles were carefully dissected free from the cranium and placed in 10% buffered formalin for 5 days.

Six condyles, from 3 rabbits, were decalcified in a 4% solution of formic acid and then dehydrated in a series of alcohol baths. After paraffin embedding, they were sectioned at 6 u and stained with H & E and Trichromic, using routine histologic procedures.

The remaining specimens were cleaned of all soft tissue and embedded in Bio Plastic (Ward's, Rochester, New York). Serial ground sections, 60-65 u thick, were prepared and mounted for microfluorescent study.

Measurement of Data

The amount of condylar bone and cartilage growth was measured in two ways. One method involved measuring the ground bone sections, under a fluorescent microscope, for the amount of condylar growth that occurred after the initial oxytetracycline vital dye was given (Figure 3). This was a linear measurement of the two dimensional change in the growth that had occurred.

The second method involved was similar to that used by Wisotsky (1964). The ground bone sections were photographed under the fluorescent microscope at 6x magnification. Serial overlapping 35 mm slides were taken of each condyle. After the slides were developed, they were loaded into a projector set at a fixed distance from a screen. Each slide was then projected and the condylar area was traced, superimposing consistent structures from one slide to the next. In this way, the separate serial pictures could be assembled into one. After all of the condyles were outlined, they were cut out with scissors and the paper was weighed. Using this method, a numerical value could be assigned to the size of the condyles that would be comparable to a two dimensional surface area determination.

Facial growth change was also recorded using the lateral radiographs. Points were selected (Figure 4), and the initial and final radiographs were traced and measured.

Figure 3. Method of measurement of condylar growth from fluorescent microscopic slides. Point X is the coronal extent of the initial oxytetracycline vital due and also bisects the condylar neck width. Points 1 and 3 are the horizontal and Point 2 the vertical extent of coronal condylar growth as measured to the final DCAF vital dye line.

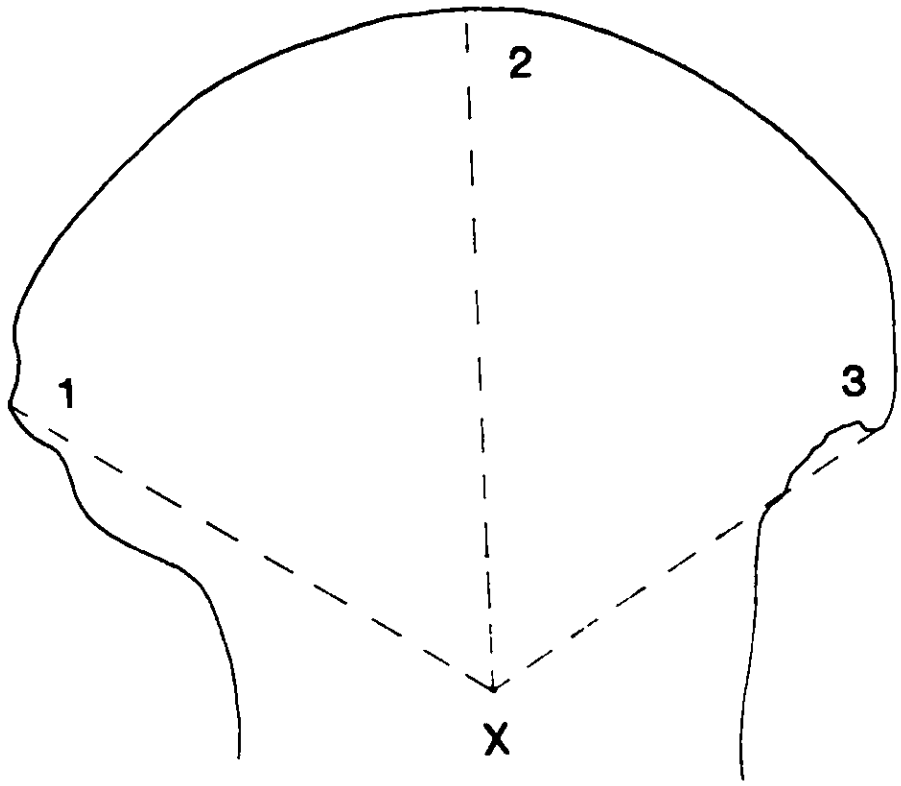
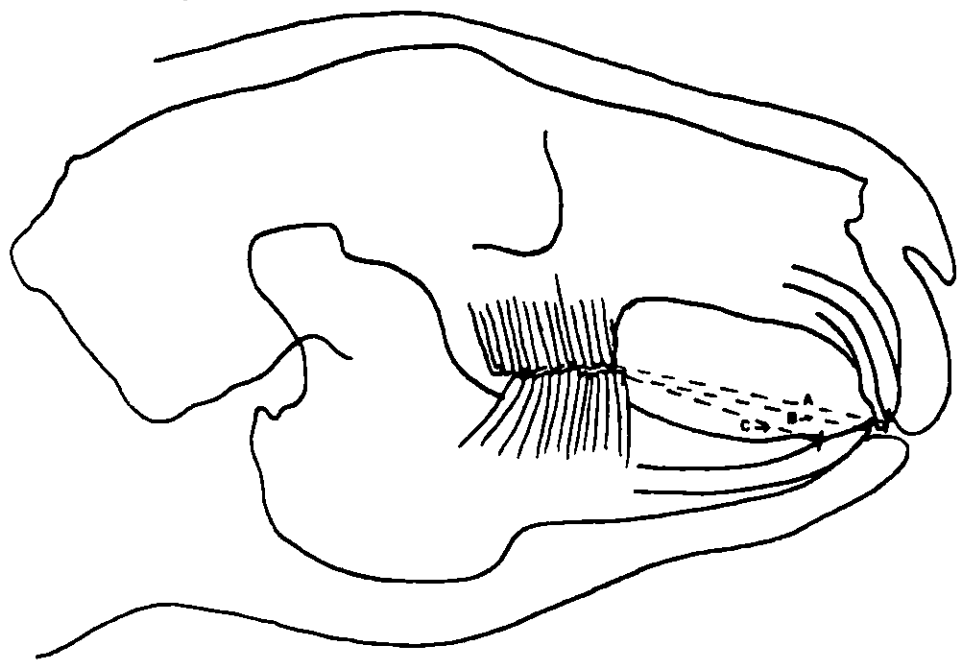

2
1
3
X

Figure 4. Point location in lateral radiographs. Distance A is from the anterior of the maxillary molar to the anterior of the maxillary incisor, B is from the anterior of the mandibular molar to the anterior of the mandibular incisor, and C is from the anterior of the mandibular molar to the anterior extent of bone distal to the lower incisor. All teeth were measured from the occlusal plane.


A
B
C

RESULTS

The results of this investigation are presented in six categories: (1) gross inspection, (2) weight gain, (3) fluorescent microscopic measurements, (4) projected paper weight measurements, (5) radiographic measurements, and (6) general histologic survey.

Gross Inspection

In general, surgical non-stimulated and nonsurgical control condyles had similar appearances. The size was relatively consistent and there was no change in shape. Stimulated condyles showed a wide variation in response in comparison to the nonstimulated condyles (Plates V, VI and VII). Some were grossly larger than the non-stimulated contralateral condyle. Others showed shape alterations in that they were more bulbous and less uniform in shape.

Two of the stimulated mandibles showed a noticeable change in gonial angle and coronoid process orientation (Plate VIII). The gonial angles were curved inward and the coronoid processes were enlarged and also turned inward.

It was also observed that the lower incisor teeth in 5 out of the 8 animals that received stimulation had unusual wear patterns (Plate IX). There were also changes in the occlusion. No

PLATE V

Normal 2 year old adult male



PLATE VI

Stimulated condyle on left, nonsurgical non-stimulated condyle on the right (animal #00)



PLATE VII

Stimulated condyle on the right, surgical nonstimulated condyle on the left (animal #4).



PLATE VIII

Gonial angle and coronoid process directional changes of a stimulated mandible (animal #4).







PLATE IX

Incisal wear patterns in a nonsurgical control animal (animal #7)

Incisal wear patterns in a stimulated animal (animal #4).





attempt was made at a correlation of incisal wear or change in occlusal pattern to the side of stimulation.

Weight Gain

Weight gain of the rabbits was rather inconsistent (Figure 5). In spite of this, the animals experienced no difficulty in eating due to the experimental procedures. The weight variations often seen before death were probably caused by an acute bacterial or mucoid enteritis. These infections were often so severe as to warrent premature sacrifice in 3 of the animals (animals #6, 7 and 8).

Fluorescent Microscopic Measurements

Five animals were used for condylar ground bone measurements using fluorescent microscopy. One of the five was a control. The original intent of the experiment was to use seven animals, but on two of the animals, the vital dyes did not stain the bones properly and they could not be used.

The amount of two-dimensional condylar growth along with age in days at the time of surgery and death are shown in Table I. Generally, three out of the four stimulated animals experienced an increase in condular growth in a vertical direction. Growth horizontally was not directional. The control animal had similar bilateral growth along all lines.

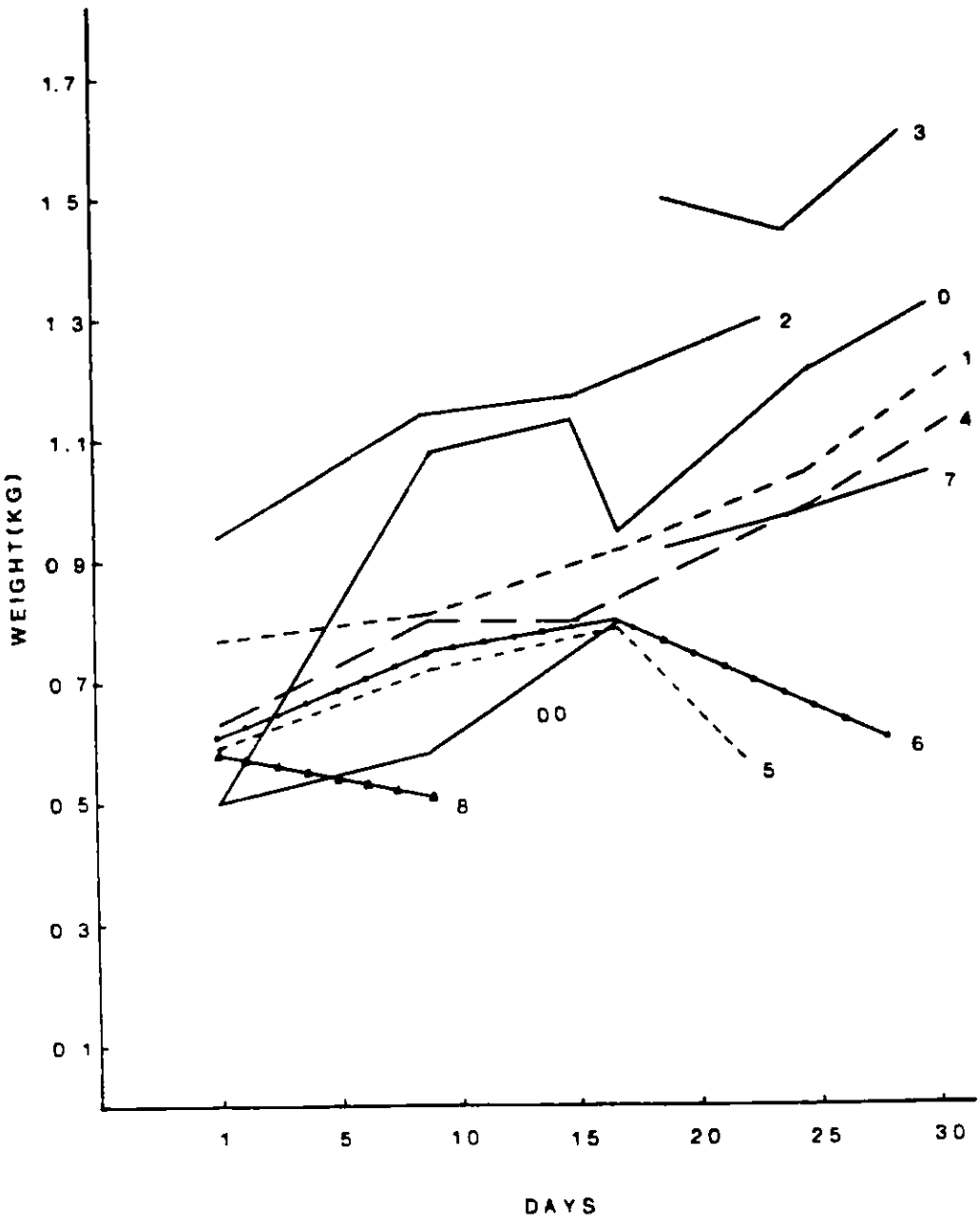


Figure 5. Weight gain.

TABLE I

TWO DIMENSIONAL CONDYLAR GROWTH CHANGES

Animal	Condyle	Measurement		
		1	2	3
1	Right*	70	85	75
	Left	59	73	72
2	Right*	91	113	92
	Left	71	102	92
4	Right*	57	120	90
	Left	61	70	99
6	Right	65	81	77
	Left*	64	68	77
7**	Right	68	66	74
	Left	73	67	73

* stimulated condyle
** nonsurgical control

Projected Paper Weight Measurements

The same animals used for the fluorescent microscopic measurements were also used for the projected paper weight measurements. The values of the paper weight measurements are shown in Table II along with age at death. The traced condyles are shown in Figures 6-10. All are equally magnified. Generally 3 out of the 4 experimental animals showed an increase in condylar paper weight of the stimulated condyle. One animal showed a decrease. Control animal condylar paper weights were very similar bilaterally and were greater than the experimental animals condylar paper weights despite a similar age at the time of death.

Radiographic Measurements

The amount of change in tooth position from initial to final radiographs, along with presurgical age, number of days between radiographs and age at death, are shown in Table III. Animal #00 is not shown because the initial radiograph was not taken. No general relationships were recognized between any of the measurements as too many variables exist for such a small sample size.

It should be noted that when initial and final dorsal ventral radiographs are compared, on 3 of the animals, there appears to be a anterior-posterior thickened articular surface in the glenoid fossa (Plate X).

TABLE II

PROJECTED PAPER WEIGHT MEASUREMENTS

Animal	Condyle	Weight (grams)	Age (in days) at Death
1	Right*	1.30	68
	Left	1.10	
2	Right*	2.18	66
	Left	1.97	
4	Right*	2.62	64
	Left	1.04	
6	Right	1.58	65
	Left *	1.47	
7**	Right	1.66	61
	Left	1.75	

* stimulated condyle
** nonsurgical control

Figure 6. Animal #1 condylar tracings. Dashed line is the surgical nonstimulated control condyle.

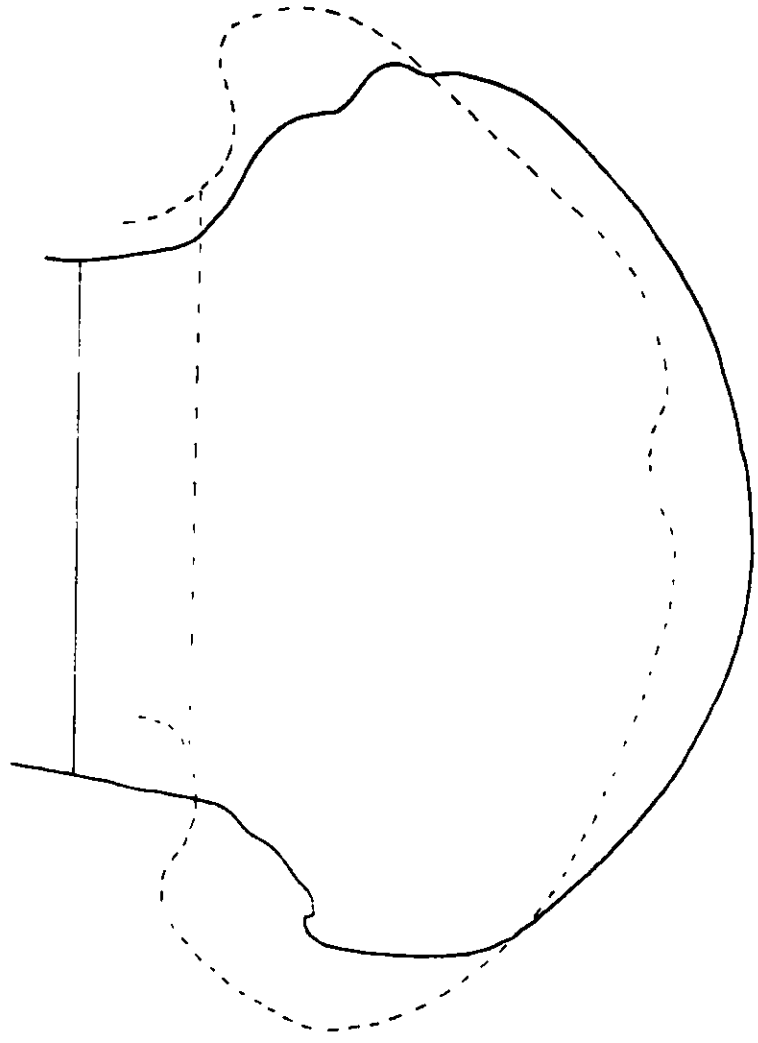


Figure 7. Animal #2 condylar tracings. Dashed line is the surgical nonstimulated control condyle.

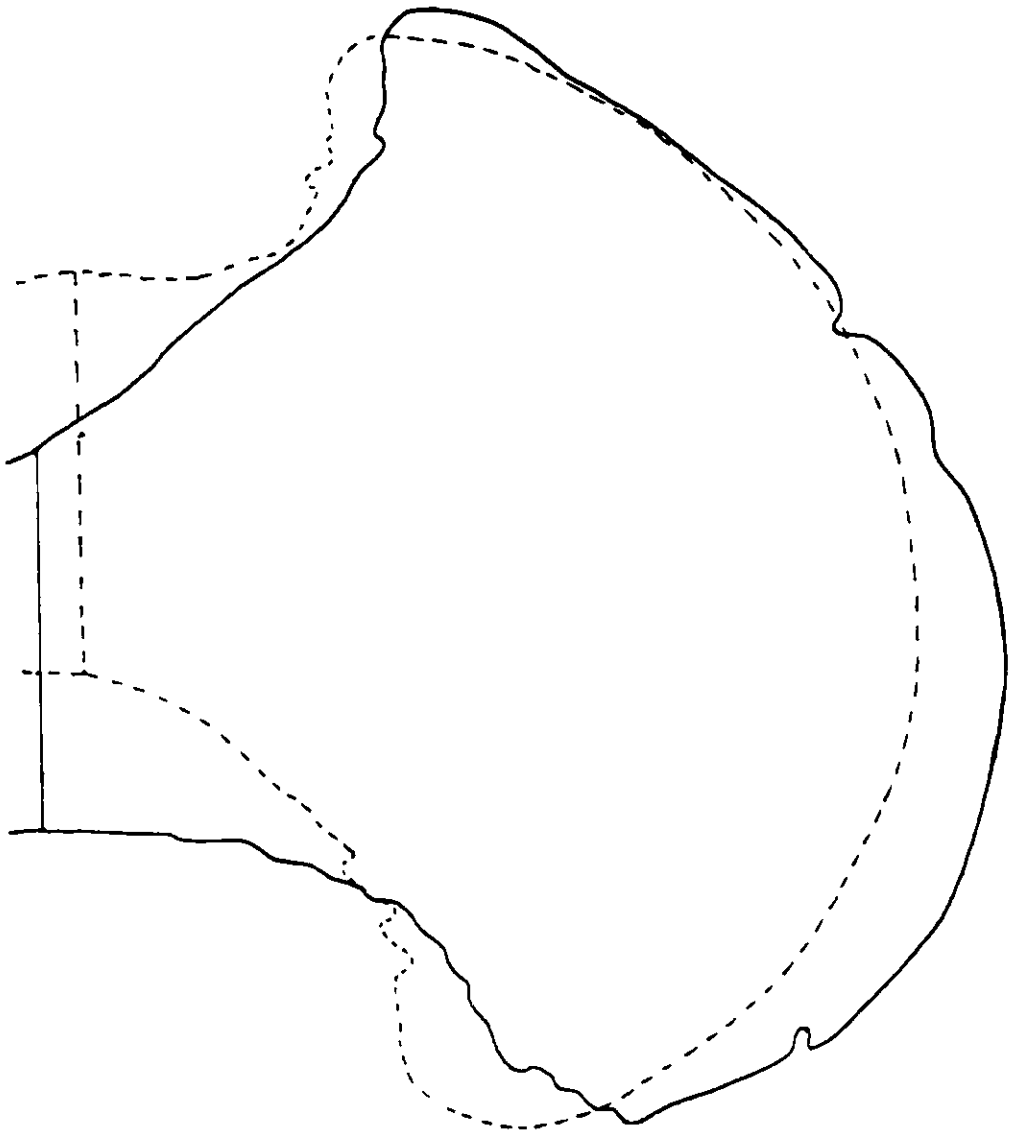


Figure 8. Animal #4 condylar tracings. Dashed line is the surgical nonstimulated control.

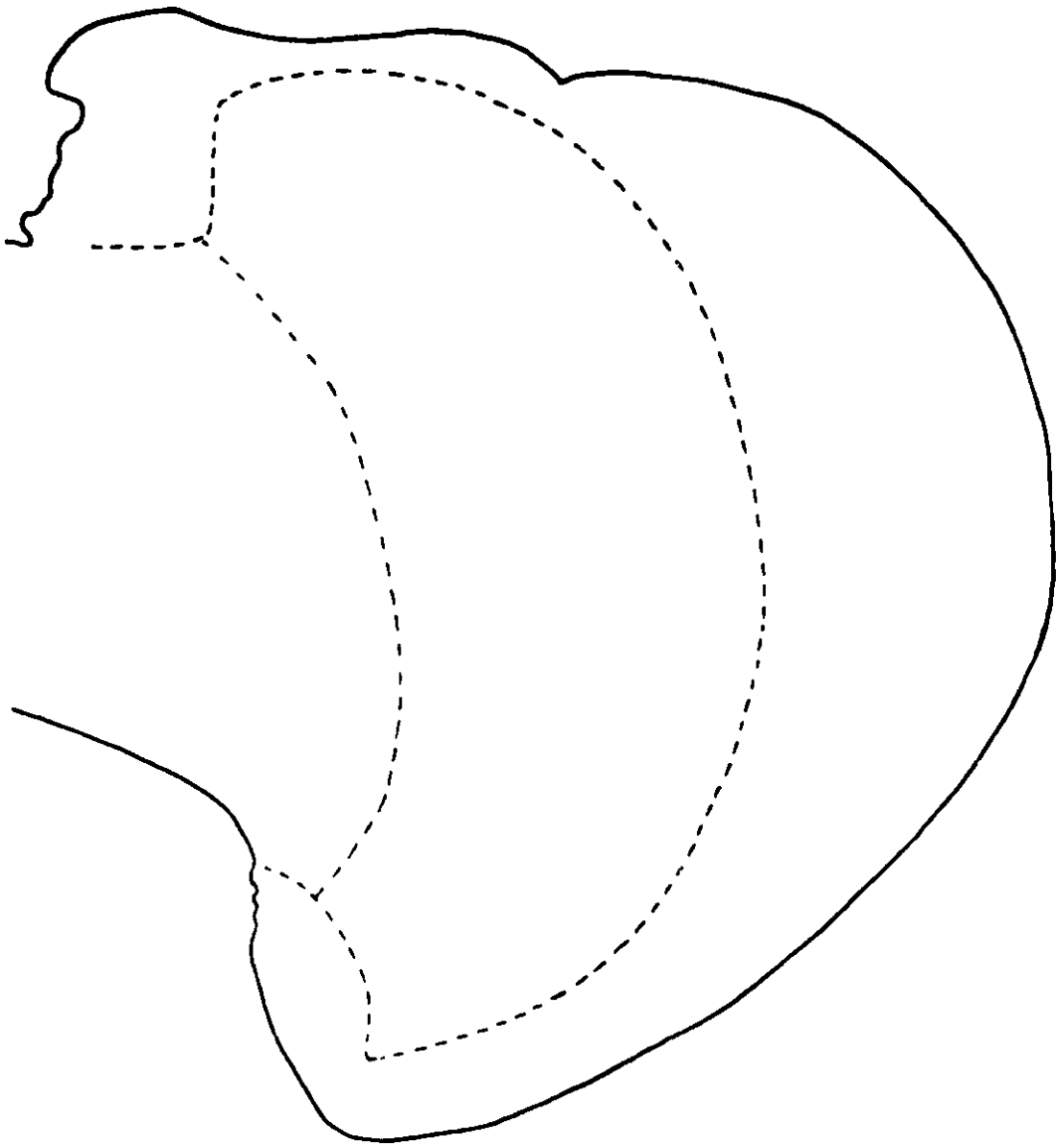


Figure 9. Animal #6 condylar tracings. Dashed line is the surgical nonstimulated control condyle.

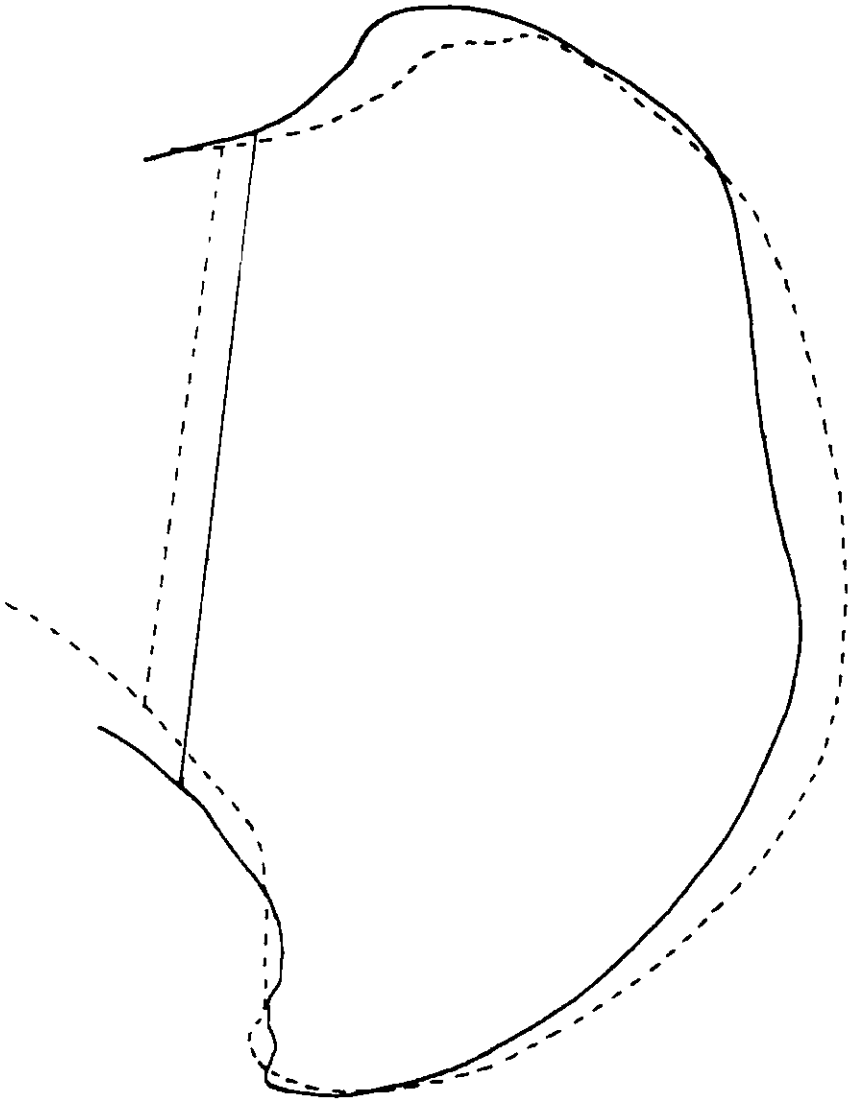


Figure 10. Animal #7 condyle tracings. Control. Dashed line is the right condyle.

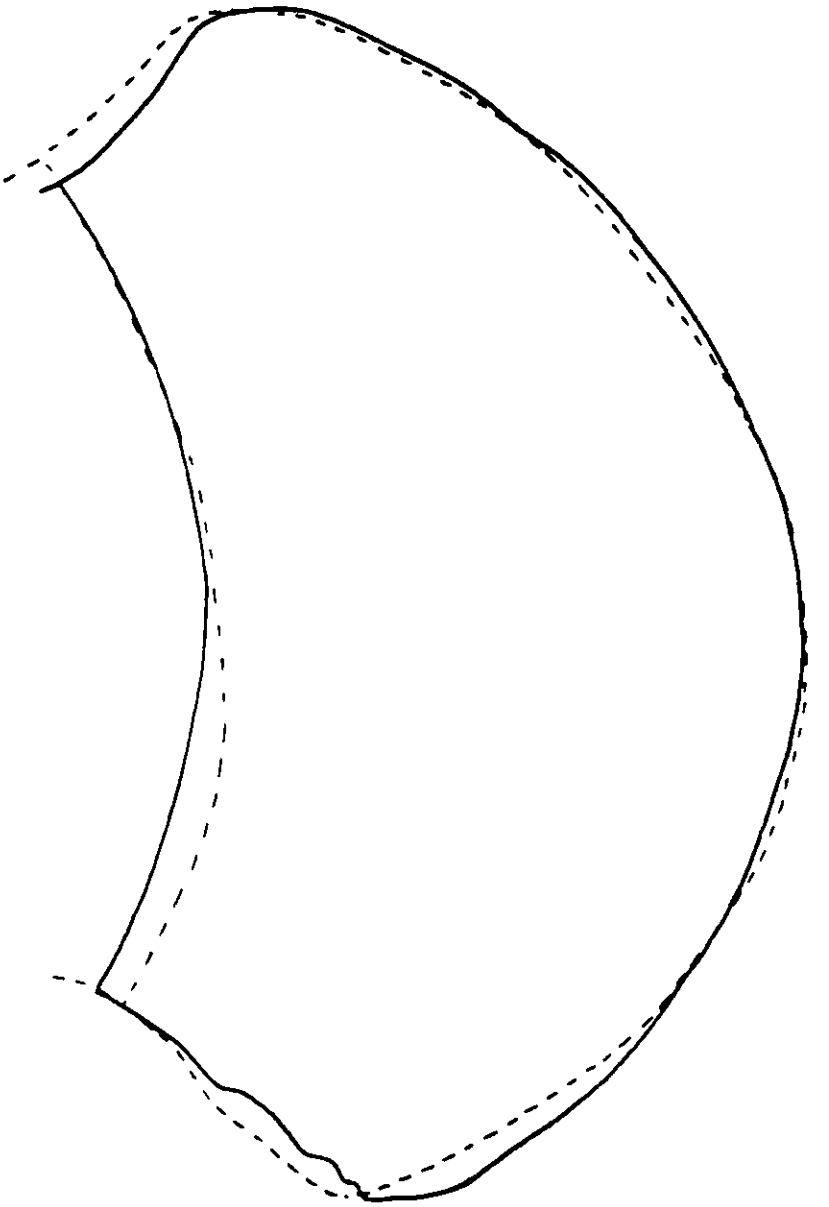


TABLE III

TOOTH POSITION CHANGES AND

AGES AT VARIOUS EVENTS

Animal	Tooth Position Change (mm)*			Age in Days			
	A	B	C	At Surgery	At Initial Radiograph	At Death	Of Stimulation
0**	4.5	3.0	5.0	35	44	65	21
1	-1.5	3.0	1.5	37	44	68	24
2	3.5	3.5	2.0	34	39	66	27
3***	3.0	2.5	3.0	--	33	66	33
4	4.0	5.5	3.0	34	39	64	25
5	2.5	2.5	1.5	36	43	57	14
6	0.0	1.5	0.0	38	43	65	22
7***	3.0	2.5	2.0	--	42	61	19
8	0.0	-1.0	-0.5	38	43	46	3

* rounded to nearest 0.5 mm
** surgical implantation on one side only
*** nonsurgical controls

PLATE X

Dorsoventral radiograph showing anterior posterior thickened articular surface of the glenoid fossa on the right hand side of the picture.



General Histologic Survey

Before histologic sections were obtained, each condyle was oriented so as to coronally bisect the condyle.

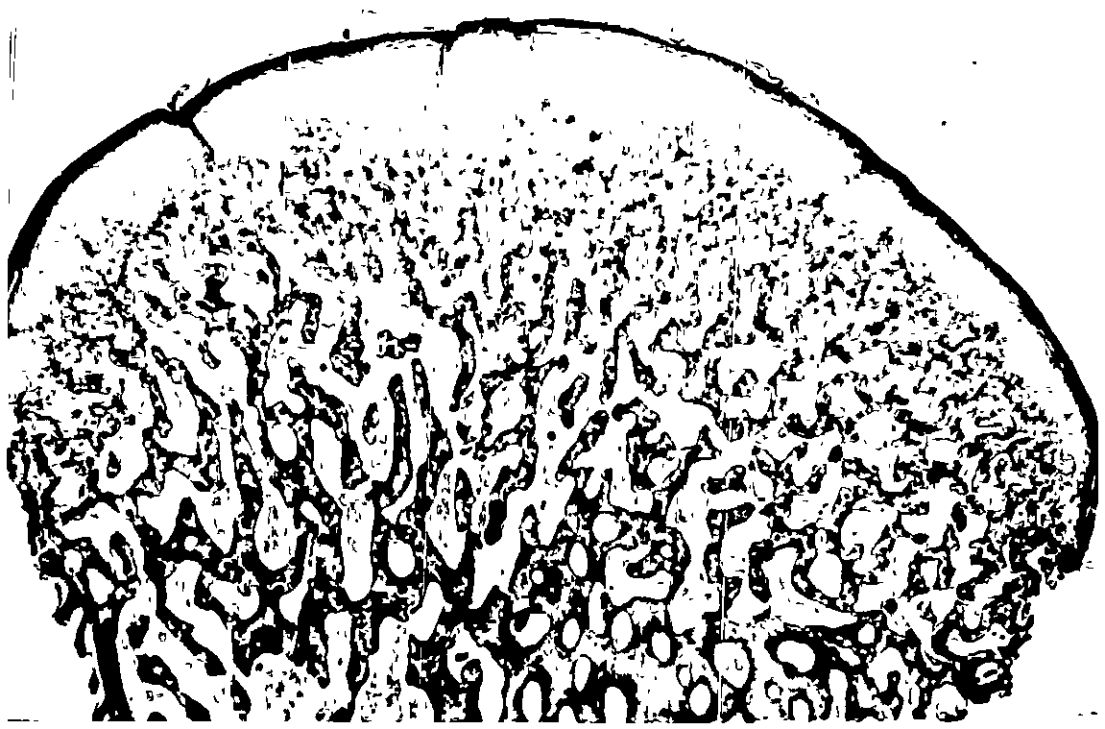
Nonsurgical and surgical nonstimulated control condyles showed routine zones of cartilage growth with normal zonal thicknesses and cell appearances. Bone growth showed normal trabeculation patterns. The major bone type present was fine cancellous wooven bone.

The stimulated condyle showed a normal capsular layer of fibrous tissue. The subcapsular or prechondroblastic layer appeared thicker and more distinct indicating a greater amount of mitotic cell divisions of the prechondroblasts. The chondroblastic layer had fewer cells and a more eosinophillic matrix. The matrix material was greater in amount and more distinct. The zone of cell hypertrophy was thicker and the cells were larger. Where cartilage was being removed and bone formation occurring, the zones were more distinct and also thicker (Plates XI, XII and XIII). Lamellar bone was being deposited on the endosteal surface of the coarse cancellous trabeculae of the nonlamellar bone. None of the above was observed in any of the control animals.

PLATE XI

Nonsurgical nonstimulated control condyle, 6x magnification (Animal #1).

Stimulated condyle, 6x magnification (animal #5). Note cartilage growth and chondroid bone formation on and down the right side of the condylar neck.





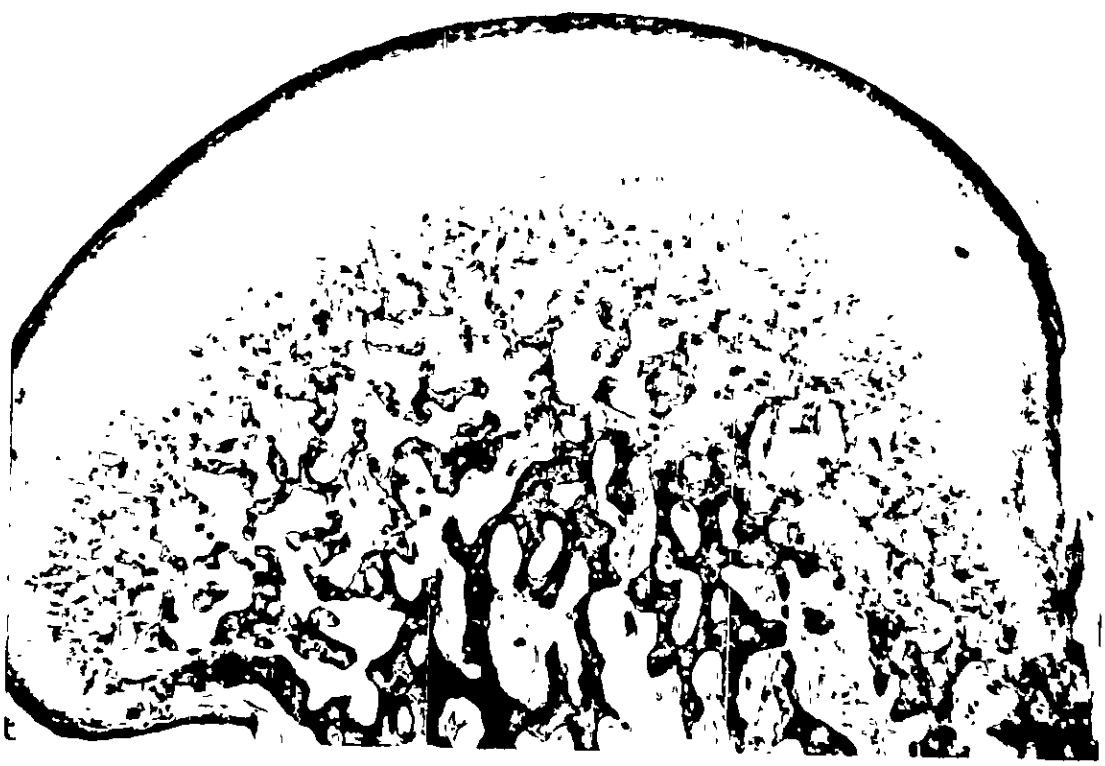
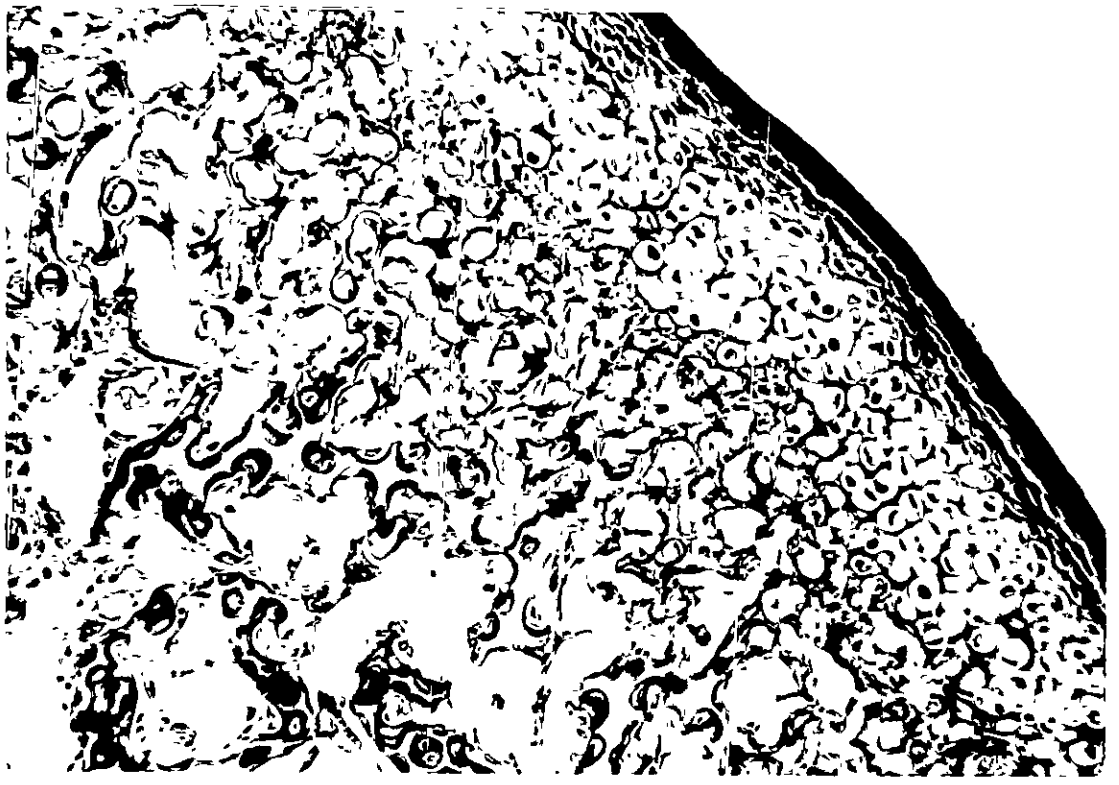


PLATE XII

Nonsurgical nonstimulated control condyle, 30x magnification (animal #1).

Stimulated condyle, 30x magnification (animal #5).



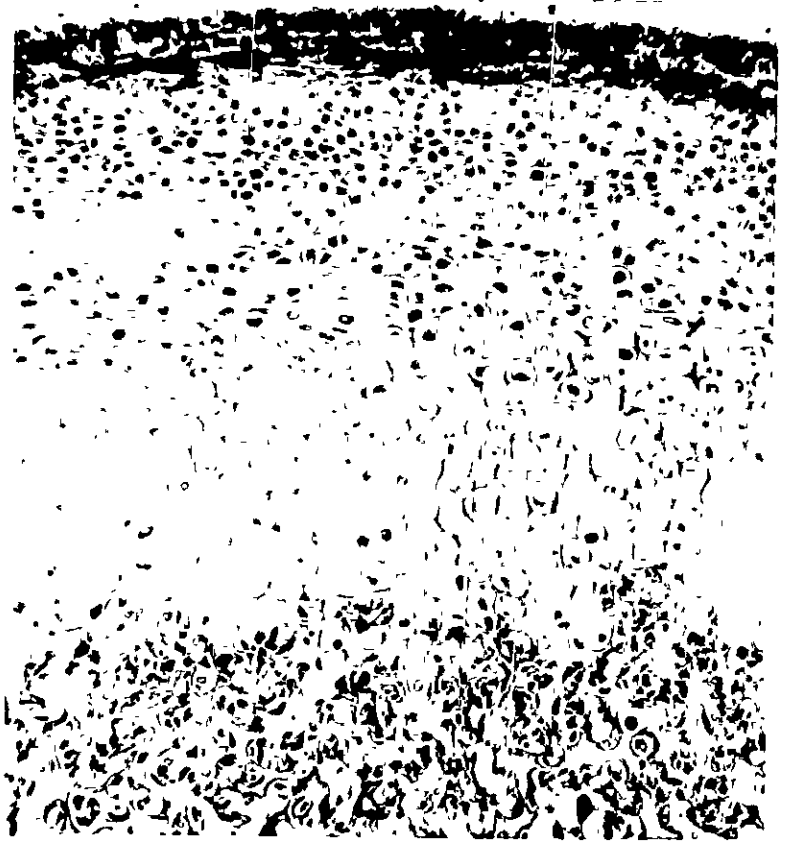
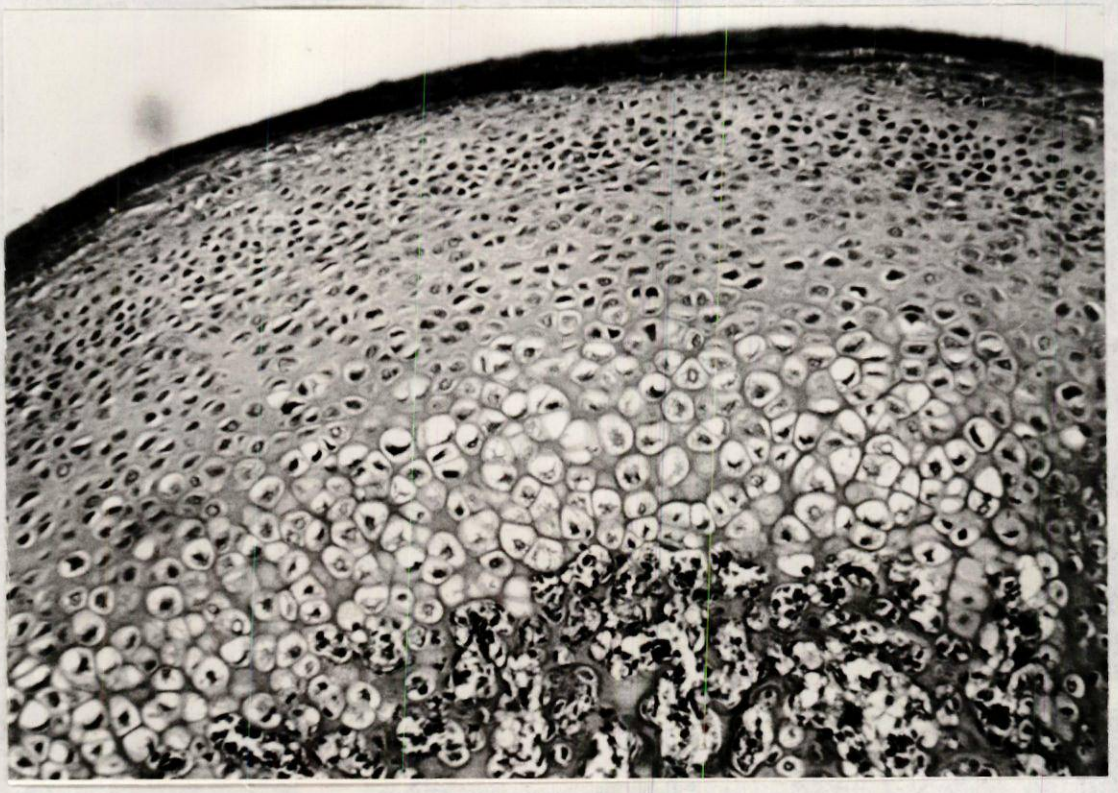
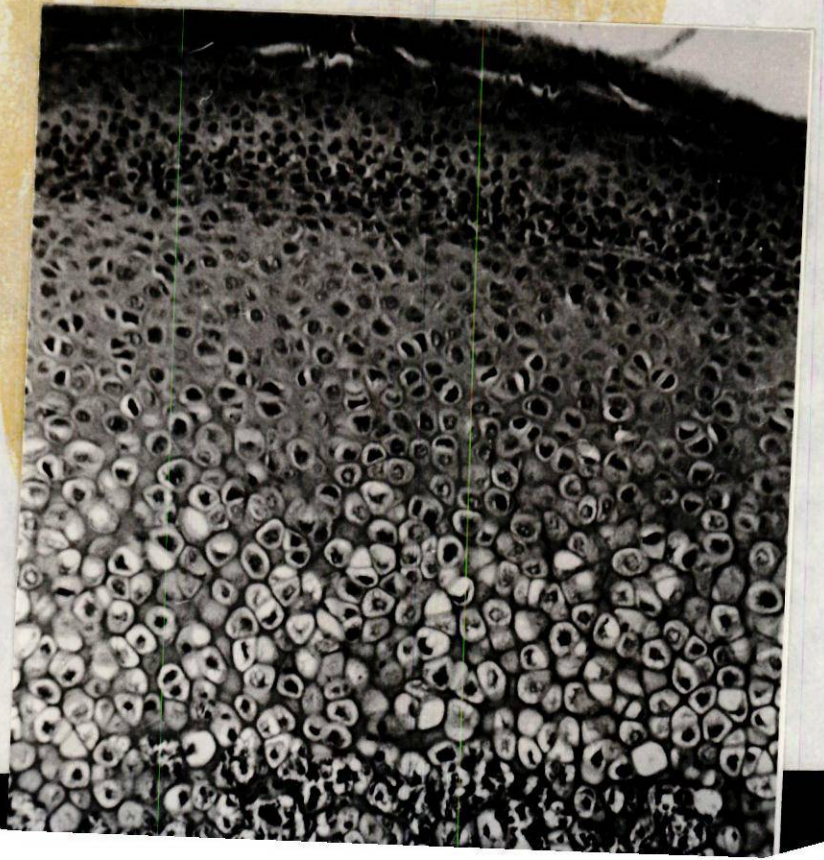


PLATE XIII

Surgical nonstimulated control condyle, 30x magnification (animal #5).

Stimulated condyle, 30x magnification (animal #5).





The stimulated condyle also showed, at the area of stimulation, extensive formation of chondroid bone on the lateral border of the condyle which graded into the woven nonlamellar bone on the lateral neck of the condyle (Plate XIV). There was also cartilage formation from the condylar head along the lateral border of the condyle. None of this was observed in the control condyle.

Fluorescent Microscopy

The nonsurgical and surgical nonstimulated control condyles had similar growth patterns and appearances.

The stimulated condyles showed an increase in size in three out of the 4 animals (Table III). This is a reduction in the trabeculation patterns and an overall "fuzzy" appearance in the areas of trabeculation. The zone of staining between the condylar bony head and cartilage is reduced and less distinct. The outer condylar surface in 3 out of 4 stimulated specimens showed areas of shallow dishing and indentations (Plate XV and Figures 6-10).

PLATE XIV

Chondroid bone formation in a stimulated condyle, 30x magnification (animal #5).

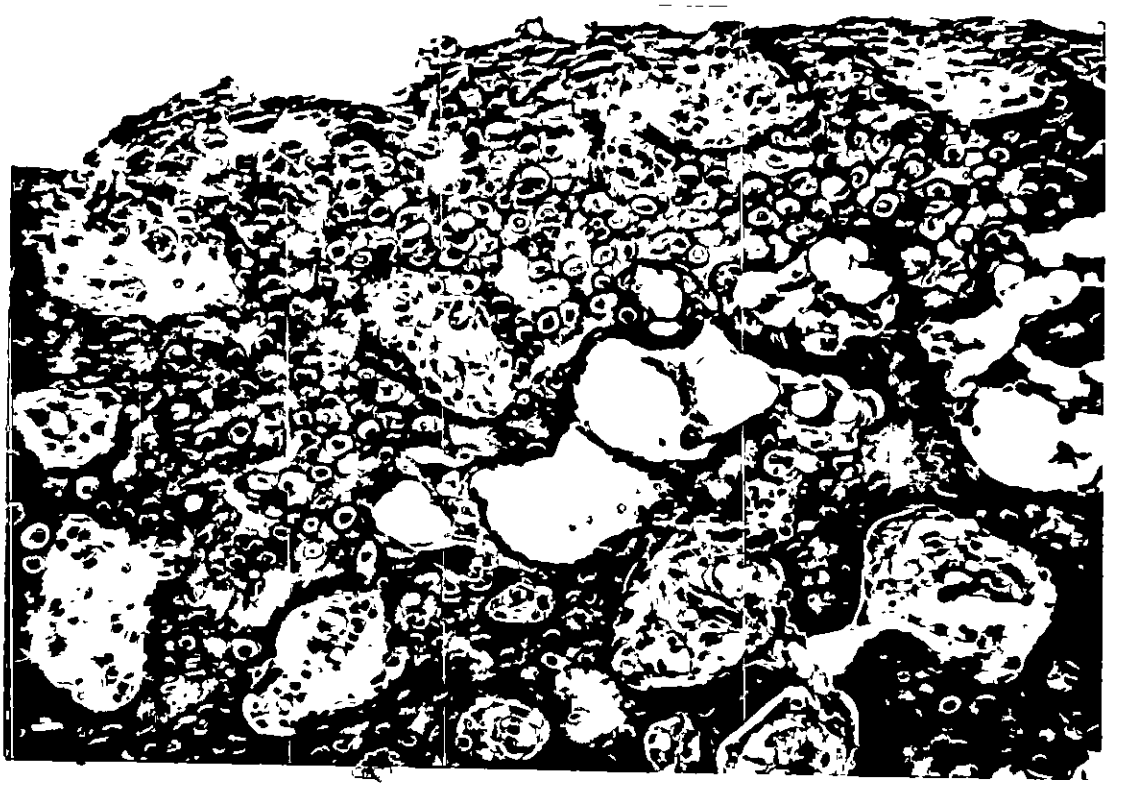
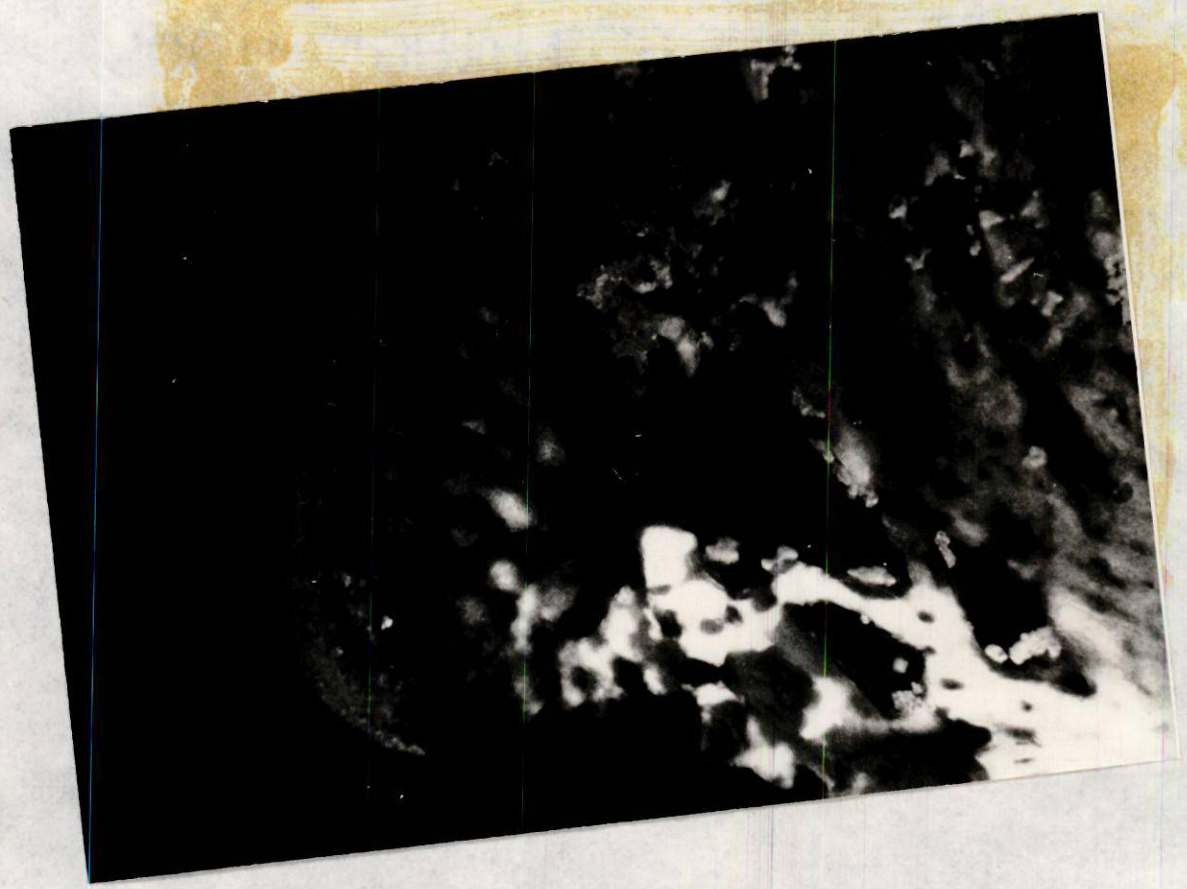


PLATE XV

Ground bone section of a stimulated condyle showing condylar surface dishing (animal #4). Fluorescent microscopy, 6x magnification.



DISCUSSION

Since the work of Fukada and Yasuda (1957) showing that electrical potentials are generated by the application of force to bone, many investigators have used external electricity to enhance osteogenesis. Various methods of delivering the electricity to the bone have been used including nonconstant direct current with invasive electrodes (Bassett et al., 1964), constant direct current with invasive electrodes (Friedenberg et al., 1970), constant direct current with non-invasive electrodes (Davidovitch et al., 1980 a, b), pulsed direct current with invasive electrodes (Levy and Rubin, 1972), and non-invasive electromagnetic field transduction (Bassett and Pawluk, 1975). As of yet, there is no real consensus on which technique is best in terms of the desired effect. However, most of the research using invasive constant direct current electrodes shows positive results. Brighton et al. (1977) showed that when electrodes are placed into bone and the current is between 5 and 20 uA, osteogenesis occurs around the cathode while resorption may occur around the anode. Davidovitch et al. (1980a,b) exposed gingiva to a constant direct current which resulted in an increase in the number of cells stained for cAMP in periosteal tissues. These nucleotides, along with C^{++}, are considered to be the intracellular "second messengers" which mediate the effects of external stimuli on their target cells. In

this case the external stimuli, the "first messenger", is the direct electric current. The increased levels of cyclic nucleotides lead toward heightened enzymatic phosphorzlation reactions, synthetic and secretory activity, and an enhanced rate of tissue growth.

This investigation attempted to stimulate condylar cartilage and bone growth by the invasive application of an electrode carrying a direct constant current. This type of current application has caused stimulation in bone growth (Brighton, 1977) but no attempt has been made at condylar stimulation.

Condylar Growth

The subject of why and how the condyle and mandible grow and if they are related has received much attention. The early conceptual view of mandibular growth was that the hyaline cartilage in the condyle represented the main growth center for mandibular growth. The growth of the condyle caused a downward and forward shift of the entire mandible. Ramal, corpal and alveolar growth were only secondary processes (Weinmann and Sicher, 1955).

Later, Moss and Rankow (1968) felt that the condylar cartilage was not a primary growth site in any way responsible for mandibular growth as a whole. They felt that the condylar cartilage represents a site of secondary and compensatory growth and that it grows to maintain articulation with the basicranium as the orofacial matrix expands.

Today, many theoreticians have turned away from the view that the condyle represents a kind of "horn of cornucopia" from which mandibular growth occurs (Enlow, 1982). Nonetheless, the condyle still represents a major field of growth and many are presently attempting to better understand it.

I believe that condylar growth cannot be regarded as the sole determinant of mandibular growth. Other growth sites are also involved, such as the alveolus, ramus and corpus. A change in the size of the condyles may precipitate changes in other areas of the mandible to compensate for the changed connective tissue capsular relationships. Whether a change in condylar size will be incorporated into the facial complex as growth occurs around the enlarged condyle is yet to be determined. But, it seems likely that a change in condylar size should have some expression as a change in the relationship of the mandible and its teeth to the rest of the facial skeleton.

Petrovic (1972) found that condylar cartilage, unlike the epiphyseal cartilage of long bones, reacts to mechanical stimulation produced by orthopedic forces. Headgear, producing a mandibular propulsion, caused an increased growth of condylar cartilage by stimulating cell proliferation in the prechondroblastic zone. He felt the prechondroblastic zone should instead be called the "orthopedists zone".

The present investigation also showed an increased thickness of the prechondroblastic zone of the stimulated condyle. There was also an increase in the thickness and eosinophillic staining of the matrix zone. Thickened and more distinct layers between the cartilage growth zones indicate greater mitotic cell division. The increased prechondroblastic cellular size and lightly stained nuclei indicate a cellular stimulation with the total number of cells increasing. The contralateral nonstimulated condyle showed none of these changes. Electric stimulation has apparently caused an increase in the rate and amount of cartilage growth.

Since both this study and Petrovic's (1972) show similar findings, it appears that electric stimulation has a similar effect as mandibular propulsive headgear. The reason for this may be the change in the synovial fluid pressure. Roth (1982) found that during function, fluctuations in the synovial fluid pressure occur. Ward (1983), functionally positioned a mandible forward and found a marked increase in synovial fluid pressure with a gradual stabilization back to the base line before propulsion. When a mandible was retrusively positioned, the pressure increased and did not return to baseline. He hypothesized that synovial fluid may be the medium by which pressure changes, due to altered mandibular positions, are translated to the joint surfaces. It appears that extraoral mandibular appliances cause an increase in

synovial fluid pressure which in turn exerts an effect on condylar growth. Since electricity has a similar effect, maybe the increased synovial fluid pressure, like electricity, cause a change in the charge or density of the charge of the synovial fluid, effecting condylar growth.

Another possible explanation relating extraoral force application and electric current to similar changes in condylar growth, is the oxygen tension level present in the area associated with the condyle. Brighton (1969) found that the growth apparatus at the epiphyseal plate was extremely sensitive to oxygen tension and that the rate of growth was greater in low oxygen tension. Since there is a marked increase in synovial fluid pressure with extraoral force application, there is probably also a decrease in oxygen tension since there is an inverse relationship (Richman et al., 1980) between oxygen tension and synovial fluid volume, (assuming an increase in pressure causes an increase in volume). Also, Brighton et al. (1975) found that the reaction at the cathode results in oxygen consumption. Since in this investigation oxygen consumption is occurring at the cathode near the condyle, oxygen tension becomes less, and the growth rate increased. In sum, the electric stimulation could have the same net effect as an extraoral force; lower oxygen tension, causing increased condylar growth.

When a current is applied to a biological system, the inherent conductive nature of electricity will not permit its own confinement to a specific area. All of the tissues surrounding the synovial fluid were effected by the current applied to the synovial fluid. Some of the effects may have been increased muscle tonicity with hypertrophy, increased levels of neurotrophic substances released from stimulated nerves, and increased cellular divisions and activity levels of the surrounding enthothelium. Since a mandibular propulsive appliance can cause similar findings, such changes may have been part of the physiologic basis for the increased levels of condylar growth observed in this investigation.

The cartilage growth seen on and down the lateral side of the condyle (Plate XI) may be the result of the changed environment caused by the electrical stimulation. When endochondral bone is subjected to high pressure, extreme levels of hypoxia occur due to the decreased blood supply from the crushed periosteal membrane. This hypoxia causes the formation of chondroblasts versus osteoblasts from undifferentiated connective tissue and an endochondral mode of osteogenesis occurs. Electrical stimulation may mimic low oxygen tension hypoxia and cause differentiation of chondroblasts vs osteoblasts from undifferentiated connective tissue. This could cause cartilage growth in any area of the condyle where electric stimulation occurs due to the fact that the

entire condyle is composed of endosteal bone and has the ability to form bone either endosteally or intramembranously.

The chondroid bone seen next to the cartilage growth on the lateral side of the condyle (Plates XI and XIV) represents very active bone growth. Histologically, chondroid bone resembles cartilage. Because it can undergo internal metaplasia into other bone tissue types, it may be the only bone tissue that has an interstitial mode of growth (Enlow, 1982). Since chondroid bone is not seen on the contralateral nonstimulated condyle, it has to be assumed that the electrical stimulation has caused the formation of this rapid growing, multipotential pathway bone tissue.

It should be noted that the stimulated condyles studied microfluorescently showed shape and size alterations as well as surface dishing and indentations when compared to nonsurgical controls and surgical nonstimulated controls (Figures 6-10). The #2 surgical nonstimulated control condyle also showed these alterations (Figure 7). This may have been caused by the surgical intervention, or it could be due to the altered function caused by the shape change of the stimulated contralateral condyle. The explanation for the indentations as well as size and shape alterations could be that the electric has caused cartilage and bone formation at different rates at different sites of the condyle, depending on what amount of current density reached a specific region of the condyle and how susceptible it was to growth.

In the microfluorescent study, 3 out of the 4 stimulated animals showed an increase in the vertical height of condylar growth when compared to surgical nonstimulated controls and non-surgical controls. There was also an increase in the projected two dimensional paper weights of these stimulated condyles compared again to controls. There has obviously been some effect on the growth rate of the condyles caused by the electric stimulation.

Hard Tissue Changes

In spite of there being no unusual histologic alterations of the stimulated animals in the gonial angle or coronoid process regions when compared to controls, two of the stimulated animals showed size and directional changes in these regions (Plate VIII). These bone areas appeared to be "bending" and elongating toward the current. It may be that the current has some type of an orientation effect. It also could be that the changes observed in the gonial angles and coronoid processes could be from a change in function, an application of Wolff's law. According to Wolff's law, the increased size of the condylar area could be causing an altered muscle function which in turn could cause a change in form. Enlow (1982) states that the connective tissue capsule, muscle, nerves, etc. cause the external shape of the human skull. The altered muscle function caused by the increase in condylar

size, could also according to this theory, change the bony architecture.

There was also an increase in the anterior posterior width of the glenoid fossa noted on three of the stimulated animals (Plate X). This may indicate that the effect of the current was not localized to the condyle. Since the synovial fluid contacts many tissues, and since the electrode was in the fluid, there is the possibility that all of the tissues coming in contact with the synovial fluid were stimulated to accelerate growth.

Although only condylar bone changes were investigated in this experiment, further research should determine what effect current has on the other tissue types present in this area. It may be that the rate of turnover or level of differentiation of the cells effects the response to a current. Davidovitch (1980) has used direct current to stimulate periosteal bone growth through the gingival tissues. Often, gingival necrosis occurs. This may be because gingiva, having a more rapid turnover rate than bone, may have an acceleration of growth upon stimulation, to the point where cell death occurs. Bone, having a less rapid turnover rate, accelerates in growth but not to the point of necrosis. It may be that stimulation should only be directed at condylar bone to prevent necrotic changes.

Even though no neoplastic or aberrant growth was observed, there is this possibility due to the platinum electrodes used in

this experiment. When electric current is passed through a platinum-iridium electrode, the platinum is liberated as cis-platinum. Cis-platinum is used in the treatment of certain carcinomas. Because of its destructive ability, the liberation of cis-platinum over a length of time may cause bone destruction or change.

Although in general the lateral radiographic tooth position measurements have no pattern, the lower molar to lower incisor measurement may have meaning. It appears that 5 out of the 6 stimulated animals (animal #8 not included due to early death, #00 had no post-surgical radiographs taken), had equal or greater growth than the controls. Either the lower molar was inhibited in growth or the lower incisor was accelerated in anterior growth. On 5 of the 6 stimulated animals there was also an altered wear pattern of the incisors (Plate IX). This could have been a deliberate shift of the jaw forward by the animals to stay away from the traumatized condylar area or it could have been a changed growth pattern due to the electrical stimulation.

Polarization

The circuit used in this investigation was designed to maintain a constant current even if the resistance changed due to polarization of the electrodes. Although it is tempting to believe that polarization can be avoided, it is inevitable that when an electrode is placed in a biological system, and an electric

current passes through it, polarization will occur due to the inherent nature of the system. The amount of resistance that occurs due to this polarization can be divided into three categories (Feder, 1963):

1. polarization due to an ionic gradient build up near the electrode-solution interface,
2. polarization at the electrode surface, and
3. polarization in the electrode itself caused by temperature change, impurities and other variables.

The amount of polarization that eventually occurs determines what the resistance becomes and in turn how much voltage is required to deliver a set amount of current at this resistance. As the polarization increases, more voltage is needed to deliver a given amount of current. The voltage measured across the electrodes gave an indication of the change in the amount of resistance during the experiment (Tables IV and V). Resistance, either electrode or tissue, reached a peak of approximately 445,000 ohms at 20 uA of current due to the limitation of the 9 V battery. Since a voltage above 1.2 V is reducing water, this may be needed for a growth change to occur because as water is reduced the pH becomes more alkaline. Bassett (1971) found alkaline conditions near the cathode. He also found insoluble calcium salts, osteoblasts and alkaline phosphatase in the alkaline conditions at the cathodes where osteogenesis was occurring, all indicative of bone growth.

TABLE IV

VOLTAGE CHARGES

Days	Voltage (Ohms) Animal #00	#0	#1	#2	#4	#5	#6	#8
1	.002	.002	1.20	1.05	0.44	1.89	0.44	0.52
3	--	0.20	1.50	1.04	0.94	1.16	1.07	1.26
5	--	1.06	1.75	0.61	0.95	1.12	1.28	1.08
7	1.30	1.06	3.15	1.55	1.33	1.12	1.30	1.16
9	2.64	1.08	3.11	0.73	1.46	1.04	1.08	
11	2.50	1.09	3.12	0.55	1.95	1.10	1.02	
13	2.25	1.12	3.05	0.72	2.11	1.12	1.05	
15	0.23	3.15	3.03	0.69	1.95	1.10	1.09	
17		8.87	8.89	0.63	2.27	1.10	1.09	
19		8.97	8.88		2.40	1.18	1.30	
21			8.87		8.11	1.21	1.09	
23			8.87		8.86	1.25	1.04	
25			8.87		0.70			
27			8.85		8.11			
29			2.38		8.10			
30					8.10			
31			8.85					
32			2.26					

TABLE V

ELECTRODE RESISTANCE AT THE BEGINNING, MIDPOINT, AND END OF THE EXPERIMENTAL PERIOD

Animal	Resistance (Ohms)		
	Beginning of Experiment	Midpoint of Experiment	End of Experiment
00	100	132,000	11,500
0	100	54,000	448,500
1	60,000	444,000	113,000
2	52,500	36,500	31,500
4	22,000	97,500	405,000
5	94,500	56,000	62,500
6	22,000	52,500	52,000
8	50,000	54,000	58,000

Note: Resistance was calculated using Ohm's law, Voltage = Current x Resistance, voltage was known (Table IV) and the current level delivered was 20 uA.

Although it is not known exactly what level of voltage is optimal for electric stimulation, it seems that it should be somewhere near the range of 0.95-1.35 V. In this range, the pH of the stimulated tissue will be altered, and most research seems to indicate the importance of pH change for growth to be stimulated. Even though pH levels in this voltage range are not the only changes occurring, they may nonetheless be an important contributing factor for growth alteration.

Above 5.0 V, H^+ ions are liberated from solutions. Increased amounts of H^+ ions cause a shift to a more acidic condition, a response reported by many to occur at the anodes, where tissue destruction takes place. Voltage at or above this level can also cause cell lyses, specifically mast cell lyses, with the release of tissue degrading lysosymal enzymes, further potentiating the possibility of bone degeneration. It therefore seems appropriate to indicate that voltages above 5.0 V may cause bone destruction and be inherently noxious to a biological system.

Significance

This investigation seems to demonstrate that condylar growth can be stimulated. It appears that the synovial fluid helps to distribute the electric current since the electrode was placed on the lateral border of the condyle and the total condyle

was stimulated. Presently, it is not known if this growth will regress or become reduced in the long term due to the functional forces acting upon it. Assuming that this growth change occurs while the rest of the face grows along with it, it seems reasonable that this change could be permanent and observable.

Although more research is needed to quantify the effects and study the long term consequences, the findings of this preliminary study appear promising. A combination of a mechanical force and electrical stimulation may decrease treatment time. With the development of an electrical stimulating device, possibly intraauricular, elastics may become obsolete. Patients who receive condylectomies may be able to regenerate new articular cartilage.

Future research should be directed at reproducible placement of electrodes and determination of the total area response to the current. It would also be of interest to place an anode near the condyle to see if growth inhibition occurs.

Bioelectric stimulation is only one facet in the control process of bone growth. Bone growth involves many regulatory steps with different combinations of triggering agents for the variety of results observed. The study and use of bioelectricity has become a major tool for probing the interrelationship among the various mechanical, chemical and electrical properties of tissues.

SUMMARY AND CONCLUSIONS

Recent investigations have shown a correlation between the synovial fluid pressure relationships generated by mandibular postural appliances and the growth of the condyle. It has also been shown that when bone is deformed, an electric potential is generated. In turn, stimulation of bone growth by electric current has been demonstrated. It is thus possible that, in the absence of mandibular postural appliances, delivering an electric charge directly to the condylar cartilage through the synovial fluid may provide for increased growth of the condyle.

This investigation was designed to study the effects of a constant direct 20 uA current on condylar cartilage and bone growth in a young, growing animal.

The findings of this investigation can be summarized as follows:

1. Histologically, the current caused a thickening of the prechondroblastic matrix and cartilage forming zones, thickened and more distinct layers between zones, and increased cellular size with more eosinophillic material present. Chondroid bone formation was occurring near the site of stimulation. These results taken together, indicate an increase in the rate and amount of cartilage and bone growth when compared to non-stimulated controls.

2. The histologic appearance caused by electric stimulation is similar to that caused by a mandibular protrusive appliance.

3. There were grossly observed directional growth changes in the gonial angles and coronoid processes of the stimulated animals.

4. There were marked changes in the incisal wear patterns of stimulated vs control animals. Incisor wear was indicative of a mandibular protrusion.

5. There was an observable increase in the anterior posterior thickness of the glenoid fossa of the stimulated condyles.

6. The stimulated animals showed an equal or greater amount of change in the distance from lower molar to lower incisor tooth positions.

7. Microfluorescent study of ground bone sections indicated an increase in the vertical heights of the stimulated condyles. There was also a reduction in the trabeculation patterns with dishing and indentations of the condylar surfaces.

8. The stimulated condyles showed an increase in the projected paper weight measurements. This was also obvious on visual examination as the stimulated condyles were grossly larger.

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