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A NEW CONCEPT FOR ORTHODONTIC THERAPY; PULSED ELECTROMAGNETIC FIELDS TO MODIFY CELL BEHAVIOR IN BONE GROWTH

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— 국 문 초 록 —

지난 수 십년간 기계적인 측면에서의 교정장치나 치료술식은 급속한 발전을 하여왔으나 생물학적 관점에서의 현대 교정학은 아직도 낙후된 수준에 머물러 있다.

이러한 관점에서 기초의학을 기반으로 세포의 생물학적 잠재력을 극대화시켜 임상적 효율성을 증진시키기 위한 일련의 연구들이 진행되고 있다. 그 중에서 전기적 자극에 의한 골성장기전의 개념을 이용하는 분야는 가장 활발히 연구되고 있는 분야 중의 하나이다.

전기자극 중 가장 유용한 수단인 전자기장 이용법의 발달과정에 대해 알아보고 전기적 자극에 의한 골세포반응의 작용기전에 대한 많은 자료를 종합, 분석하여 그 가설적인 모형을 세우고 아울러 교정 임상에서 이러한 개념의 적용가능성과 그 범위에 대해 조명해 본다.

앞으로 이 분야에 대한 계속적인 연구로 머지않아 실제 임상에 활용되는 단계에 이르게 되길 기대한다.

The essence of orthodontic treatment is the movement of teeth through bone to obtain a individual dental occlusion and to be in harmony with surrounding stomatognathic system. Although there has been considerable refinement in appliances and techniques over the years, biologically related problems such as incleasing rates of orthodontic tooth movement, shortening periods of orthodontic treatment and accelerating bone growth have been little progress. At present, in orthopedic fields, remarkable advances in the clinic have been achieved, using electrical stimulation, by enhancement of biologic responces.[25-30,60-63] On the other hand, increase in rates of tooth movement in animal experiment by the use of locally applied direct current (DC) [50,146,157] and pulsed electromagnetic fields (PEMF)[158] have been reported in recent article. Mor

recently, under the experimental studies, enhancement of condylar growth in rat mandible was reported histologically by the application of Galvanic current[159] and PEMF[160] Pulsed electromagentic fields, more improved forms of electrical stimuli, will find broad application in dentistry, where they already have been shown to exert a major benificial effects. Biologicaly progressed orthodontic treatment such as shrotening the retention period after tooth movement and/or accelerating the healing process after rapid maxillary expansion, I think, could be also achieved by PEMF. The use of PEMF will ultimately bring economical, social, and psychological benefits to patients and equip orthodontists with the advanced technique. As an improved understanding into the nature of the biologic response generated by electrical stimulation should lead to advanced therapeutic procedures in clinical orthodontics, the author derive this concept from extensive investigations and a synthesis of a great quantity of basis biologic data. The purpose of this paper is to arouse orthodontists' interest in this new concept presented here and to stimulate discussions and further researches.

DEVELOPMENT OF ELECTRICALLY MEDIATED GROWTH CONCEPT

In most or all biological systems, mechanically induced electric charge separation is a fundamental phenomenon.[1,2,3] The first indication of this effect in biologic tissue perhaps was reported in 1941 by Martin[4], who observed an electric polarization in bundles of wool and hair when subjected to stress. The generic term "piezoelectricity" has been used to describe the behavior of long-chain biopolymers since the investigation was set about in earnest by Fukada and Yasuda,[5] 1957. Various theories about the origin of electric charge had been suggested.[5-10] In 1962, Bassett hypothesize that mechanically induced charge separation might control the activity of osseous cells and their biopolymeric byproducts. Thus the concept of electrically mediated growth mechanism, which involves biological growth and bone remodeling by any means, in living system was established[1,3,7] Japanese investigators had reported in the 1950s that bone formation could be stimulated electrically[11]. The new series of studies, undertaken in 1964 by Becker and Bassett[10], utilized miniaturized implantable battery packs with platinum-iridium electrodes. When implanted in the medullary canal of dogs, increased osteogenesis was observed around the cathode when a current of 3-4μA was established for 21 days. After these experiments were reported, many similar investigations were made. These included a study of constant[12-20] direct current, pulsed direct current,[21,22] and of AC singals.[23] From these several sources, these following observations seem to be established. First[13,14], osteoblastic activity characteristically is observed near the electrode that is held cathodic under DC conditions, while osteoclastic activity may be present under mild anodic condition. Second[14,15] there is a threshold current of 3-5μA necessary to stimulate osteogenesis in these systems. An optimal stimulus exists in the range of 5-20μA. Third[14], deleterious effects appear when currents larger than 50μA are employed. Fourth[21,23], in most cases under pulsed or AC conditions, a polarity-dependent cellular response (osteoblastic vs osteoclastic) is not observed.

Afterwards, the demonstration of increased osteogenesis by DC stimulation led orthopedic

surgeons to the clinically attractive idea that it might be possible to speed up fracture healing by electrical stimulation. Through the animal experiments[7,18,19,20,24] and fracture site of human long bone,[25,26,27,28,29,30,31,32,33], successful reports had appeared. Jorgen[34,35] observed that healing process on fractured human tibia was accelerated 30% by pulsating asymmetrical direct current. Brighton[36] et al analysed the relationship between charge, current density, and the amount of bone formed in the medullary canal of the intact rabbit tibia. At a given current density and a peak current, the amount of bone formed is linearly related to charge (constant current X time), indicating that constant direct current produces more new bone formation than does pulsed direct currents. This results are in agreement with those of Hassler and co-workers[24]. Based on these findings, further, a new cathode was designed with eight active ports evenly distributed along its length and providing two and one half times the amount of bone formed by a conventional cathode. More recently, electric firld or pulsed electromagnetic fields inducing electric current in bone tissue have been developed to promote osteogenesis in specific situation.[56,57,60-63]

On the other hand, Shandler et al[37] investigated the effect of direct current on the repair of osseous lesions in the canine mandible and proposed that the practical use of electrical stimulation in oral and maxillofacial surgery be possible. In 1977, Norton[38] showed endosteal new bone formation in periodontal defect area applied topically direct current. Kopczyk[39] also reported similar results. This observation may verify the usefullness of electrical stimuli in alveolar bone regeneration. Yasuda et al[40] recorded negative electrical potential at compressed cortex and positive potential at distracted side by applying stress on long bone. Steinberg[41], in vitro and Cochran[42], in vivo experimented with similar method respectively to observe piezoelectricity of bone. Also Gillooly et al[43] reported this charge separation in alveolar bone. Zengo in 1973[44] and 1974[45] demonstrated that areas in alveolar bone described as characterized by osteoblastic activity were electronegative and conversely areas of positivity (or electrical neutrality) were observed in regions characterized by osteoclasia deforming force on teeth in vivo and in vitro. These findings seem consistent with views of Epker and Frost[46], in that changes in the surface curvature of bone caused by loading correlate with specific cellular responses. Certainly these observation can interprete the known architectural responses of bone to mechanical factors, Wolff's law.[47] The importance to orthodontics of electric phenomena in bone has been reviewded by Gold[48]. Beeson et al[49] applied constant 10μA current to the mandible of cats via electrodes surgically implanted mesial and distal to premolar undergoing orthodontic treatment. However no significant difference in the rate of tooth movement was seen. Subsequently, experimental results that orthodontic tooth movement may be accelerated by the use of locally applied electric currents was reported by Davidovitch et al[50]. They suggested that direct current enhance cellular enzymatic phosphorylation activities in periodontal tissues. Under histologic observation, they found osteoblastic activity was increased near the cathode, but no typical osteoclastic lacunae at the alveolar socket wall near the anode was detected.

The clinical indication for the use of electrical stimulation to induce growth of bone include alveolar ridge augmentation grafting, osseous repair after removal of large bone-destroying lesions,

fractures, nonunions, bone grafting for discontinuity defects, pseudoarthrosis, chronic osteomyelitis, osteotomies, alloplastic implants, treatment of periodontal disease and orthodontic therapy. Until now, electrostimulation of osteogenesis has been attempted with various electric and magnetic signals. These approaches may be grouped according to; a) the methode of application-invasive, semi-invasive, and non-invasive; b) the form of energy used—electric, magnetic, and electromagnetic; and c) whether the applied signal is constant (static) or nonconstant (dynamic). At the time the direct current stimulation observation was first put on a sound foundation in 1964,[10] it appeared likely that fracture repair could be speeded by this technique. From the outset, however, several disadvantages of this approach were apparant. First, most fracture will heal spontaneously without open reduction, and the hazard attendant upon routine surgical use of electrodes did not seem justified. Second, these electrodes would require another surgical procedure for removal. Third, all electrodes are subjected to some electrolysis and have a deleterious effect on osteogenesis.[51] Fourth, the electrode-stimulated bone formation was spatially limited, whether D.C. or pulses were employed, because the electrode was small in size, compared to the repair site. This would require multiple electrodes for effectiveness in the long bones of man. For these reasons, alternate methods of influencing the electrical environment of an osseous repair site were sought. These included, non-electrochemical electric fields. Although effective in influencing alterations in cell function,[52,53] fracture repair,[54] and bone growth,[55,56] this method was rejected because it required large fields (500-1000 V/cm), potential hazard to patients and possible current leaks would result in major electrolysis effects.[57] The second approach was begun in the late 1960s and made use of the fact that time-varying (pulsing) electromagnetic fields induce current in conductors, Lenz's law. Although bones are relatively poor conductors ($5\text{-}10^5$ ohm)[58,59], the coupling efficiency between the bone and the field is such that safe driving voltages (10-30V) in the coil induce biologically-significant tissue voltages[61] (0.1-50mV) Pilla[60], Bassett et al[61] observed that fields with specific characteristics speed the repair of canine fibular osteotomies. Frank[62] sought to examine the effects of a specific osteoinductive field on a healing ligament in an animal model. Bassett and Pawluk[57] reported that fracture repair could be accelerated by specific pulse wave (circuit type 1-P1-Hz, circuit type 10-P65-Hz), with analysing mechanical, histologic and radiographic examinations. In 1977, for the first time, Bassett et al[63] documented the therapeutic use, in surgically resistant pseudarthroses, of low energy electromagnetic fields pulsing in the extremely low frequency range.

Fear has been expressed by some investigations in this area that electrical stimulation by fields and currents may result in cancer, possibly through a general enhancement of growth. No evidence of such effects was present in these experiments. Quite to the contrary, the reparative tissue in the stimulated specimens, generally, was less cellular than the controls.[57] None of the histologic material revealed any evidence of neoplastic changes. Hamphrey and Seale[64] reported in 1959 the use of direct currents to retard tumor growth in experimental animals. Becker[65] reported the use of electrodes in Carcinosarcoma regression. Recently, in the preliminary study of the electromagnetic fields effects on the behavior of Meth A sarcomas in experimental animals, marked inhibition of tumor growth was observed over the 10-day observation

period.[57] The hope that magnetic fields could induce regression or total inhibition of tumor cells have remained controversial and unsubstantiated.[66] The interest in possible biological effects of extremely low frequency magnetic fields (1-1000Hz) has increased in recent years due to the growing problem of technological contamination of the environment which has magnetic pollution from countless sources and the social awareness of the need to preserve our ecology. Some behavioral, neurophysiological and neurochemical effects have been reported in mammals,[67,68] but they require high intensities and present evidence indicates that no potential hazard exists at exposure levels of extremely low frequency magnetic fields (ELMF) with frequencies from 7 to 75 Hz and intensities of 3-10 Gauss or electrical fields from 1 to 29 v/m (rms).[69,70]

It would appear from these considerable clinical findings and basic researches that the electrical findings and basic researches that the electrical stimulation applied in various forms is reasonably safe when considered from "biologically harmful" standpoint, and they were found to be safe, practical and effective by the Federal Drug Administration in Number, 1979.

MECHANISM OF MECHANOELECTRICAL PHENOMENON

Various proposals have been made to account for the generation of strain-related potentials in bone. Very briefly, these can be summarized as follows.

a) Classical piezoelectric theory,[5] It is proposed that bone possess a distribution of fixed charge such that strain results in separation of the centers of positive and negative charge, which generates a net polarization and an external potential.

b) Semiconductor theory;[8] It is proposed that bone contain array of P-N junction associated with the collagen bone mineral (hydroxyapatite) interfaces and that these junctions, similar to those observed in other materials system, generate a potential difference when strained.

c) Streaming potential theory[7]; It is proposed that, in moist tissues, movement of electrolyte fluid (intracellular fluid) due to applied pressure past bound charges within bone generate streaming potentials that contribute to potentials observed when bone is strained.

d) Dipole orientation[71,72,73]; It is proposed that deformation of bone cause reorientation, specifically rotated, of dipoles in collagen associated with intermolecular and-fibril bonds and that such reorientation produce a net dipole moment and a resultant external potential.

Biopolymers such as bone, tendon, cartilage, muscle, dentin, cementum, etc have the properties of pyroelectric, ferroelectric, solid state and electret. Piezoelectricity is one of such behavior of long-chain biopolymers[72,74]. These include collagen, cellulose, keratin, protein polysaccharides, and nucleic acids, among others.[71] It was reported that there is considerable transduction of mechanical force to electric potential in whole teeth.[75] Braden et al[76] was unable to discern any piezoelectric effect with enamel. However, with dentin, results comparable with those obtained using bone were observed. The differences observed between enamel and dentin may possibly be attributed to the fact that enamel has to collagen.

Piezoelectricity in polymers according to dipole orientation theory is caused by a stress-induced rotation of dipoles around asymmetric carbon atoms, for example, CO and NH dipoles

for polypeptides. Because the hydrogen bond formed between the N-H group on one residue and the C=O group on an adjacent turn of the helix is nearly parallel with the helix axis, unidirect ional orientation of the helix dipoles is formed to produce a resultant external potential.

In general, at an electric double layer forming at a solid-liquid interface, a potential difference, the zeta potential occurs between the layer of ions along the wall and the more diffuse accumulation in the fluid because of preferential adsorption of either positive or negative ions from the electrolyte. When liquid is forced through the capillary system, the diffuse layer of ions is caused to move, thus producing a potential along the capillaries called the "streaming potential"[77]. Such potential can arise in the vascular channels and in the extravascular fluid spaces in bone as a result of pulsatile blood flow, muscle activity, and physiologic loading, the relationship is given by the Helmholtz-Smoluchowski equation $E = P\epsilon\zeta/4\pi\eta\lambda$, where E is the streaming potential, p, the driving pressure, ϵ, the dielectric constant, ζ, the zeta potential, η, the viscosity, and λ, the conductivity of the solution in the capillary system. Local streaming potentials that occur, for example, on the surfaces of mineralized tissue due to stress-induced streaming of the bone fluid will be directly dependent upon both the instantaneous pH of the local bone fluid and the calcium level in the local extracellular fluid[77]. The faster the movement of the liquid the greater is the magnitude of the streaming potential. This phenomenon may explain the observation by Dwyer[78] and Bassett that no measurable voltages occur when bone is slowly deformed a given amount but that voltages are detected when the same bone is rapidly deformed by the same amount. According to Eriksson[77], bending deformation of a long bone causes the liquid in the transverse channels to be displaced toward the convex surface, this liquid is electrically positive charged and therefore causes the convex side to become positive with respect to the concave side. After measuring streaming potential in human dentin, the other side, Mumford et al[80] commented that because fluid movement in dentinal tubules is considered important in causing dental pain, the development of these electric potentials may be significant in the excitation of sensory receptors.

It has been shown by the author[81] that although collagen is piezoelectric, wet collagen is not piezoelectric because of the bound water. When measuring the piezoelectric properties of collagen with increasing water content, the voltages will progressively decrease due to an electrical leakage through water and reach zero at approximately 300 mg H_2O/g dry collagen, specifically chemically bonded into the strucutre. With a water content increasing, voltages reappear that are heavily dependent upon pH.[77] This finding is consistent with the existence of freewater in the collagen and the production of stress induced streaming voltages. This is also supported by the data on decay rates of the signal. The decay time for the piezoelectric signal can be obtained from the relation.

$V(t) = SD\frac{\partial r(t)}{\partial t}$[82] where S is the applied stress, D is the piezoelectric constant, and r(t) is the dielectric response function. For bone saturated with 0.9% saline[83], it has been seen that r(t) reaches its long time value in less than 100μ sec. Thus, the voltages will decay to nearly zero

within 100μsec after the step force is applied. On the other hand, the observed decay rates for wet bone signal are a few tenths of a second or higher, suggesting some source for this signal other than the piezoelectric effect. The above results suggest that the electromechanical voltages measured in bone be due to streaming potentials. It can be assumed that some mechanism other than the piezoelectric effect must be responsible for the observed voltage in the wet state.

POSSIBLE MECHANISM OF ELECTRICAL STIMULATION

How do these electrical stimuli trigger a response in bone cells? The mechanism of action of electrical stimulation applied in various ways has not been fully elucidated, however, a great deal of data have been accumulated suggesting some fundamental action of it.

Electrical Stimuli On Cell Membrane

Cell recognition and growth are regulated in large part by epigenetic alteration in the structure of cell surface receptors as well as by changes in their mobility and distribution. Any such changes in the structure, pattern, or dynamic state of receptors at the cell surface is called surface modulation[84]. Grodsky[85] has modeled the plasma membrane as a sheet of dipoles under electric strain attributable to the local electric field generation by the presence of cations in the polyanionic structure of the outer membrane. Polyanionic proteinaceous material forming a sheet on cell membrane surfaces appears to be the site of detection of these weak electrical stimuli. Experimental observations concerning the effects of low frequency electromagnetic fields on efflux of Ca^{+2} from cell membrane led investigators like Adney,[86] Bull[87] and Kaczmarek[88] to postulate that more Ca^{+2} efflux may be produced at particular extremely low frequency fields. These findings support the hypothesis that a low frequency weak extracellular electric gradients may be transduced in a specific class of extracellular negative binding sites, normally occupied by calcium ions and susceptible to competitive hydrogen ion binding. In addition, Bawin[89] documented that a stimulus capable of modifying the efflux of an ion essential in excitation and regulation function by 15% would be associated with dramatic changes in perception and behavior.

Brighton et al[90,91] suggested that direct current produce changes in cell's electrochemical environment. The alterations in the microenvironment in the vicinity of a cathode that delivers 10-20μA at a potential of less than 1V are at least two, namely, the lowering of the local oxygen tension and a raising of the local pH. When the potential rises, oxygen consumption gives way to electrolysis, that is, hydrogen production occurs. A decrease in PO_2 is favorable to cartilage and bone formation[92,93,94,95] and that an alkaline environment is favorable to calcification is suggested by the rather high pH (7.70 ± 0.05) found in the zone of hypertrophic cells of the growth plate by Howell and coworkers[96]. The number of hydroxyl radicals produced is directly proportional to the current applied. At higher microamperages, the pH of the local tissue fluid may increase to toxic levels. This suggestions support a previous experiment[14] that coagulation necrosis, cell death, and no bone formation occurred in the vicinity of a cathode that delivered 50 or 100μA. Also the pH decreases; chlorine and later oxygen are evolved at the anode[97]. On

the other hand, it is known that a cathodal current excites cell membrane to reduce the net membrane potential whereas a anodal current actually makes cells more resistant to excitation than normal by enhancing the net potential.

Rodan et al[98] have demonstrated that in their experiment of oscillating electric fields, the rapidly increasing potential differences between the plate electrodes briefly perturb the membrane potential either directly (through field effects) or by way of transient currents generated in the electrolyte solution surrounding the cells.

It may be useful to consider the situation of a generalized glycoprotein receptor which can diffuse laterally in the plane of the cell surface membrane.[84] Specific glycoproteins including the cell surface receptors that control ion permeability and adenylate cyclase activation have molecular weights between 300,000–400,000 and containing three of four types of polypeptide of molecular weights between 40,000 and 65,000[99]

Calcium ions are essential in the regulation of the resting membrane potential and is the sequence of events in synaptic excitation. In the reasonable that calcium can move inward not only by diffusion and by moving through sodium channels, but also through potassium channels and N-Ca exchange.[100] Mullin[100] concluded from several data that for times of the order of an action potential, depolarization leads to an entry of calcium via both sodium and potassium channels. It is believed that closing and opening of the gates of the Na channel is caused by "gating potential" that occurs in the lipid matrix of the cell membrane adjacent to the channels. Any sudden change in membrane potential causes a simultaenous change in the gating potential of the sodium channel and increase the channel's permeability.[101] One theory is that membrane-bound calcium ions creates a positive field near the channels and blocks entry of sodium ions.[101] At the onset of the next depolarization, the first event is theoretically to remove these calcium ions to increase the permeability of the sodium channels to Na ions. It is becoming increasingly evident that gating activity in excitable membrane is dependent upon absorbed divalent ions.[102,103] Thus we can postulate that electrical stimuli may in such way, either release of membrane-bound Ca^{+2} or changes in the gating potential, alter the permeability of the bone cell to calcium. This theory is supported by Messer.[104] Cations[105,106]. charged macromolecules[107] and charge-charge interaction were shown to have pronounced effect on the activity of adenylate cyclase. One might anticipate, therefore, that certain conformational changes produced by polycations might stimulate cyclase without the participation of hormones[107].

In vivo effect of electromagnetic fields on healing of fibular osteotomies, Bassett et al[57] suggested that specific wave form elicit an orientational improvement of fibroosseous callus. In 1964, it was suggested that collagen fibrils be oriented by weak, direct, electric currents (1μA) as they were reconstituted from solution in laboratories.[108] Furthermore, the electric impulses may somehow realign the collagen molecules to play a role in initiating calcification[109]. Collectively, this evidence suggests that both cells and/or collagen itself may be affected by electrical stimuli (Fig. 1).

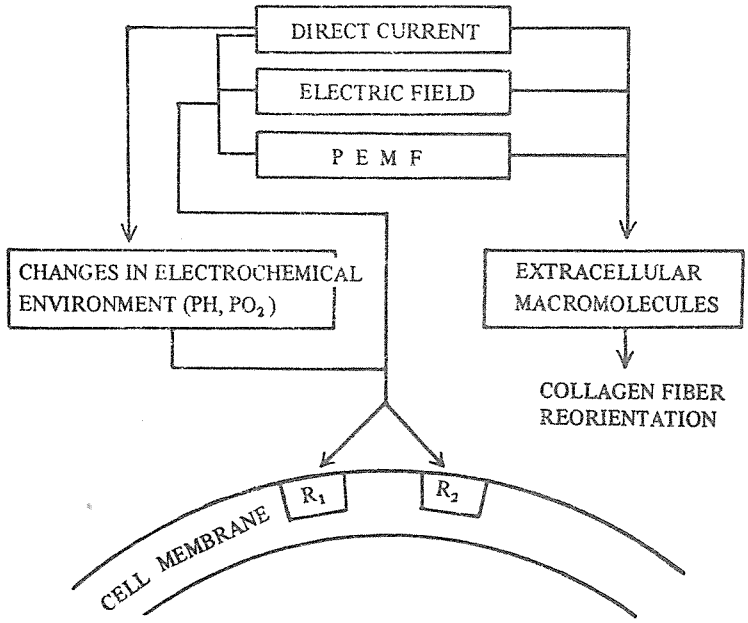

Fig. 1. Both cells and collagen fibrils may be affected by variously applied electrical stimuli. There are two kinds of receptors in plasma membrance: for ionic channel (R_2) and for adenylate cyclase (R_1). Two models can be possible; a) Electrical stimuli affect both receptors for adenylate cyclase and ionic channel; b) Electrical stimuli affect selectively R_1 or R_2.

Interrelationship of Calcium and Cyclic Nucleotides

It is postulated that cyclic nucleotides alter the intracellular metabolism of calcium and conversely that calcium alters the metabolism of cyclic nucleotides.

There is considerable eivdence that calcium serves as a second messenger[110-112]. The various reported effects of calcium on cellular function include enzyme inhibition or activation, activating of transport systems, regulation of mitosis[106,126,127] transcription[113] calcification[114], secretion[115]. The rise in cytosolic calcium ion concentration inhibits adenylate cyclase[116-118] and it activates or inhibits in some cases phosphodiesterase.[119-121] The increase in cyclic GMP concentration may be due to an activation of a soluble cytosolic guanylate cyclase by Ca^{+2} rather than a direct extracellular messenger activation of a membrane-bound guanylate cyclase.[122-124] The effects of cyclic AMP have been found to be exerted by activation of protein kinase which catalyzes the transfer of phosphate from ATP to various enzymes to activate or in some cases inactivate it. According to a recent report by Greenbard[125], these phosphorylation reactions are the central events occuring in the activation mechanism of cells by external stimuli and it may be mediated by either cAMP, cGMP or Ca^{+2}. It has been shown that while synthesis of protein is correlated with high concentration of cAMP[126], secretion of cellular product, for example, lysosomal enzymes from neutrophil, is associated with elevation of cellular cGMP contents by increase

Ca^{+2} uptake[127] Goldberg and his collegues[128] have attention to the apparant universality of the interrelationship by formulating the Yin-Yang hypothesis that cAMP and cGMP may impose contrasting or opposing regulatory influences. Ca^{+2}, cAMP, and cGMP all are involved at some point in normal proliferative responses in vivo [Figure 2]. Cell devision is generally initiated in appropriate cell lines in vivo or in vitro by some external or initiating signal. Many lines of normal cells grow logarithmically until the density or cell number becomes high enough to form a single confluent layer of cells. Once confluency is reached, the cells stop growing or grow at a very much reduced rate.[129] Rasmussen and Goodman[112] concluded from considerable evidence of the role of cAMP in cell division that in some cell lines cAMP is a regulator of growth rate and cell morphology and cAMP is not a universal regulator of cell dividision, cell growth, or contract inhibition. Induction of proliferation of normal fibroblasts is associated not only with a fall in [cAMP] but with a rise in [cGMP] [130-132]. Moreover, it has been suggested that the fall in[cGMP] be the important signal in density dependent growth inhibition and a rise in [cAMP] decrease the rate of cell proliferation at some point in G_2 [133]. Also, a subsequent rise in cAMP was reported to be a singal for cytodifferentiation[134,135]. These data are consistent with the concept that cyclic nucleotides are a major messenger in the phenomenon of contact inhibition. Pastan et al[136] measured the activities of adenylate cyclase and phophodiesterase as a function of increasing cell population density. They found that as cell density increased and the cell reached confluency, the cyclase activity per milligram cell protein continued to rise, but diesterase activity fell and

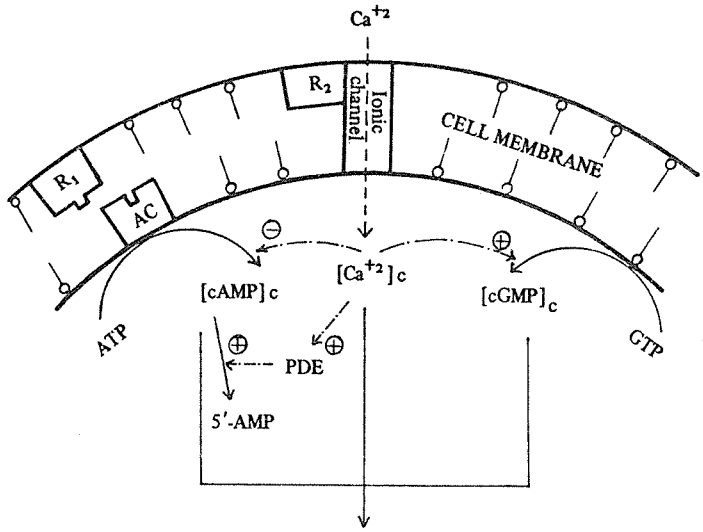

Fig. 2. A schematic representation of the interrelationship of Ca^{+2} and cyclic nucleotides in control of cell proliferation by electrical stimulation.

R_1 : Receptor for Adenylate cyclase, R_2 : Receptor for Ionic channel,
AC : Adenylate cyclase, PDE: phosphodiesterase
⊕ ; ACTIVATION ⊖ ; INHIBITION

the concentration of cAMP rose.

The adenylate cyclase loses its sensitivity to calcium during maturation from the proliferative to the hypetrophying state[117]. There is evidence that membrane changes with the altered cartilage phospholipid composition during the maturation of chick epiphysial cartilage cells elicit differences in membrane enzymes, an increase in 5′-nucleotidase and a decrease in adenosine inhibition of adenylate cyclase. This differences lead to higher cAMP level in the hypertrophying cells[137,138,139].

Experimental observations that mitochondrial calcium can play a role in biological calcification have been reported by several investigators.[140,141] The mechanism of this calcium release from mitochondria and cell membranes in the bottom of the zone of hypertrophic cells may be regulated to the local tissue oxygen tension[114]. The interrelated nature of event at the plasma and intracellular membranes is further emphasized by the recent evidence indicating that a rise in intracellular (cytosolic) Na^{+} may regulate the release of Ca^{+2} from mitochondria[142]. This means that depolarization may produce calcification mediated by Ca^{+2} release from mitochondria.

Direct Current Effects On Cellular Reponse

Bassett[143] demonstrated increase of DNA and collagen synthesis by fibroblasts grown in both continuous and extremely low frequency interrupted (1Hz) direct current field. There was a report [144] that the application of constant direct current had no effect on either the initiation or rate of mineral deposition in the osteotomy. However, once the normal content of the callus had been achieved, the application of the current did result in a continuation of mineral deposition in the callus. In recent investigation[50] examining the effects of electric current ($15\mu A$) on periodontal ligament and alveolar bone, it was found that while near the cathode, osteoblasts, osteoprogenitor cells and periodontal ligament cells were activated to produce increased cellular enzymatic phosphorylation, leading to synthetic and secretory processes, near the anode many mononucleated cells and a mixture of fibroblasts were crowded. As previously reported[14], at above $50\mu A$ of anode, especially $100\mu A$, electrocoagulation or fibrinous degeneration was found. These evidence make us assume that anodal current may cause both inflammatory change in tissue followed by aggregating migratory cells, and activation of osteocytes, eliciting a osteolysis. The osteocyte may be activated by changes in pH or PO_2 [145] accompanied with electrochemical modification near the anode, thus releasing collagenase[84] and prostaglandin.[146] Also it is postulated to play a key role in demineralization.[144] Degraded bone matrix by collagenase and collagen product by demineralization were indeed shown to be chemotactic to monocytes, presumed to be precursors of osteoclast.[147] Prostaglandins produced by osteocytes and/or macrophages are able to not only induce bone resorption which was hypothesized by Rodan[148] that a shape change in osteoblast is caused by resorbing agent to expose it to osteoclasts or osteoclastic projection, but also act as a chemical mediator for inflammatory response. In the experiment of Davidovitch et al.,[50] they suggested that orthodontic tooth movement be accelerated by the use of locally applied optimal direct current. However, at least two problems are insoluble. First, since the

alveolar bone resorption in compressed side is not due to the physiologic osteoclasis by osteoclastic resorption but due to the extensive pathologic osteolysis by osteocyte and inflammatory response, bone regeneration in this area may not happen normally and increased pathologic tooth mobility accompanied with fibrous tissue and macrophage may not replaced reversibly. Second, alteration of electrochemical environment is affected at regions only within 2 or 3mm from the anodal electrode. Actually, however, the effective distance from gingiva to alveolar bone is often too far to be effective for osteocytic activation.

With this view, the possible use of electric current in tooth movement is doubtful and more disucssion should be made.

Effects of EF and PEMF on Cellular Response

Electric field stimulate DNA synthesis in cartilage cells.[98] In this experiment, they support the hypothesis that Na^{+} and Ca^{+2} fluxes generated by the electrical perturbation trigger DNA synthesis in cartilage cells.

Variation in pulse characteristics could produce different responses, depending on the type of cell or tissue understudy and on its functional status. Recent evidence revealed that DNA synthesis was increased in bone cell by PEMF and was dependent upon wave form and cell culture techniques.[149] PEMF with specific pulse wave increased the rate of synthesis of proteoglycan and collagen and diminished the rate of bone resorption utilizing biochemical measurement.[150] Two pulses in clinical uses, the repetitive single pulse and the repetitive pulse train, produced different results from each other and from controls when the specific activity of m-RNA or cellular transcription was monitored.[151] In the case of the PEMF treated non-union, this pulses enhance the preliminary step of endochondral ossification, namely calcification of fibrocartilage in the gap between the ununited bone fragments. This soft tissue, which is an impediment to vascular invasion and osteogenesis, becomes calcified under PEMF treatment both in animal model[152] and in biopsy material from human[58]. On the other hand, in recent study, the investigation[153] was found that cartilaginous lyzsozyme which involve proteoglycan degradation is activated by PEMF, thus it may promote normal healing. It is well known that in vivo calcification of cartilage is associated with disaggregation of proteoglycans possibly by lysozyme.[154] In the basis of present information, it seems clear that specific waveform characteristics may be required to trigger selective biologic responses for bone growth.[155]

CLINICAL IMPLICATION OF PEMF

More recently, reports all the more[149,156] have appeared that bone cell was influenced by PEMF. These data suggest that electrical stimulation (direct current or PEMF) may be a potent tool in accelerating bone turnover. Moreover, these evidences lead us to postulate that intramembraneous ossification as well as endochondral ossification may be enhanced by electrical stimuli, such as sutural growth and/or alveolar remodeling. This concepts of electrically mediated bone growth can be applied to clinical orthodontics; first, using PEMF, specific pulse pattern

applied, during retention period after tooth movement, to alveolar bone may promote calcification of newly deposited bundle bone followed by enhanced rigidity of alveolus and stabilized new functional entity, avoiding relapse effectively and subsequently shortening retention period; secondly, healing process of palatal expanded tissue, for example, rapid maxillary expansion, can be accelerated by electrical stimulation. It is becoming clear that specific wave form characteristics may be required to trigger selective biological responses. In programming a series of pulses that affect each stage of repair of rapid expanded maxilla, these will entail a pulse to trigger DNA synthesis to increase the population of messenchymal cells followed by pulses to maximize collagen and proteoglycan synthesis by activated osteoblasts and finally a pulse similar to those in use for non-union to increase rates of calcification and maturation. As a result, the high quality of bone tissue and rapid formation of maturated midpalatal suture may bring a satisfactory result and thus treatment period may be shortened. The application of the direct current, on the other hand, to expanded suture will continue mineral deposition[50] and become too rigid to be susceptibel to relapse. In addition, it may be possible clinically to use a functional appliance combined with Direct current or PEMF so as to produce greater amount of mandibular growth than might be expected by the application of either system as a result of additional or synergistical effect on the condylar growth.

This concept is based on a great deal of basic data on hand. The degree of precision in programming cellular events is not a vain hope and probably can be achieved within the foreseeable future. Obviously, there are many technical and scientific barriers still to be crossed. Future advances of this important new discipline in the clinic must be made only with the support of basic science and animal experiments. The future of electrotherapeutics in clinical orthodontics, using PEMF or direct current, is bright if it rests on precise communications and a fundation of hard data.

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