CASE WESTERN RESERVE UNIVERSITY

GRADUATE STUDIES

We hereby approve the thesis of

JARIYA THIMAPORN

candidate for the MASTER OF SCIENCE IN DENTISTRY degree.

Signed: Donald H. Enlow
(Chairman)
Stanley A. Strisik
Gary R Holt

Date 5-18-89

EFFECTS OF UNILATERAL PREMATURE FUSION OF THE ZYGOMAXILLARY SUTURE ON THE GROWTH OF NASOMAXILLARY COMPLEX:A HISTOLOGICAL ASSESSMENT

by

JARIYA THIMAPORN

Submitted in partial fulfillment of the requirements for the Degree of Master of Science

Department of Orthodontics

School of Dentistry

CASE WESTERN RESERVE UNIVERSITY

May 1989

EFFECT OF UNILATERAL PREMATURE FUSION OF THE ZYGOMAXILLARY SUTURE ON THE GROWTH OF NASOMAXILLARY COMPLEX: A HISTOLOGICAL ASSESSMENT

Abstract

by

Jariya Thimaporn, D.D.S.

The objectives of this study were to determine (1) the extent to which the process of bone remodeling can compensate during facial morphogenesis in response to the occurrence of experimental sutural synostosis, and (2) the locations and pattern of such remodeling adjustments.

The experiment was carried out on twenty two-week-old Hartley guinea pigs comprising four control and sixteen experimental animals. The left zygomaxillary suture of each experimental animal was immobilized with cyanoacrylate adhesive. In all animals, amalgam implants were placed on each side of the sutures on both sides of the face. Serial cephalometric radiographs were taken at the time of surgery and every two weeks thereafter. On the 51st day after the operation and on each successive 5th day thereafter, each animal was injected with contrastive vital bone dyes. Increments of implant

separation were recorded. Two histological procedures were use:paraffin embedded decalcified sections for the observation of fine bony structures and ground sections for ultraviolet visualization of vital dye staining of bone.

The results demonstrated that premature fusion of the left zygomaxillary suture, created by immobilization using cyanoacrylate adhesive, was achieved and resulted in constraining the bone growth at that suture as evidenced by a less than 0.5 mm. magnitude of implant separation in most animals. Adjustive compensation by adjacent sutures and by bone remodeling was seen as a consequence of premature sutural fusion. Adjustive compensation on the fused side differed from that on the non-fused side. Developmental asymmetry was found when comparing the fused side to the non-fused side with the distribution of depository and resorptive periosteal surfaces of the fused side proceeding differently from that on the non-fused side. No structural asymmetry, however, was found, since the right/left remodeling adjustment led to compensatory symmetry.

DEDICATED

TO

My parents, Tawee and Yupa Thimaporn, the most beautiful people in my life, for their unselfish love.

ACKNOWLEDGEMENTS

I would like to extend my sincere appreciation and gratitude to my advisory committee for their wise guidance, invaluable assistance, and endless patience; Dr. Donald Enlow, Dr. Jerald Goldberg, Dr. Stanley Hirsh ,and Dr. Gary Wolf.

Also, I would like to thank the following individuals for advice and technical assistance in carrying out the many tedious tasks involved in this study, as well as for their friendship; Dr. Michael Power, Dr. Robert Cederquist, Dr. James Simmelink, Dr. Lawrence Scaletta, Dr. Ordean Oyen, Dr. Ruth Nordlander, Charlotte Geleev, Adolf Vozka, Marcia Dunner, Andrew Slodov, Jing-Yi Shiau , Walter Schratz, and Bradley Kmentt.

And, most especially, I would like to thank Tadashi Nagura for always being with me through the rough road.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

INTRODUCTION

Mechanisms of craniofacial growth have been an interesting area of study for many decades. Numerous experiments have been conducted to better understand the control mechanism of craniofacial growth. At this point, much is known of what happens in craniofacial growth, but still little is understood of the precise control mechanisms involved.

In normal postnatal craniofacial skeletal growth process, two general morphogenic principles are involved; displacement and remodeling (13). Displacement type of growth occurs adjacent to sutures and other types of joints in response to the expansion of other tissues. Remodeling process occurs in the bone cortex and serves to adjust for necessary changes in the configuration of individual bone as a response to the physiological functioning of that particular bone (42). Although remodeling growth is distinct from sutural growth, these processes proceed in an organized manner and are coordinated.

Craniofacial anomalies result from disturbances in normal growth patterns. Although such anomalies may have several causes, premature fusion of craniofacial sutures directly inhibit normal displacement among involved bones, alter growth and produce an anomaly. Altered

growth of one structural unit may result in altered growth of adjacent structures, which in turn affect more distant structures (4). An anomaly becomes more obvious when an active or major growth site is disturbed or many growth sites become involved simultaneously. For instance, the Crouzon syndrome is a variable result of premature fusion of the coronal, sagittal, and lambdoidal sutures, premature fusion of the sutures and synchondroses of the basicranium, and premature fusion of the sutures of the orbit and nasomaxillary complex (33). The resultant anomalies are clinically obvious; brachycephaly or scaphocephaly (depending on the sutures involved), midface hypoplasia, a V-shaped maxillary arch, relative mandibular prognathism, etc. It has been shown that an overgrowth of bone will occur at remote suture sites as a compensation for a lack of bone growth at the site of fused sutures. This condition becomes more obvious in unilateral premature synostosis, i.e., plagiocephaly, caused by premature fusion of the coronal suture unilaterally. The basicranium is extremely asymmetric, the nasal bridge is deviated to the affected side, and the maxilla and mandible are asymmetric (32). In some instances, anomalies may be difficult to detect clinically because the affected growth site is not an active or major growth site, the affected area is not extensive, or because the

compensatory growth of other growth sites help mask the effects from the primarily involved growth sites.

The underlying role of compensatory growth of complementary growth sites now needs to be evaluated, in order to clarify the extent to which such adjustments can serve to correct a given anomaly. What happens to adjacent sutures and cortical bones might tell us how the morphological changes of involved bones are carried out to provide such compensation.

REVIEW OF THE LITERATURE

BIOLOGY OF SUTURE

A suture is one of the three forms of synarthroses (immovable joints). It is an articulation in which the margins of the bones are united by a thin layer of fibrous tissue.

Sutures are found only in the craniofacial complex among the facial and cranial flat bones. There are two main types of sutures, the cranial type and the facial type. Their embryonic development is different but eventually each yields a three-layered structure. The two layers adjacent to each bone are composed of collagen fibers directed at right angles to the internal sutural margin. A central zone is composed of cellular components, nerve fibers, blood vessels , and loose irregular directed collagen fibers. When this structure evolves, the suture becomes a site of continuous bony deposition and resorption throughout the growth of the craniofacial skeleton (28). With skeletal development and increasing age, the cellular contents and the concentration of collagen fibers tend to decrease, the fiber bundles become irregularly spaced and are associated with areas of bony deposition (29). However, the functional or enviromental stimulus for continued bone deposition at the facial sutural margin is not fully understood at this time.

Sutural configuration.

Three types of sutural configuration exist in both cranial and facial sutures:

1)The serrated type, with projections interdigitating from one bone to the other, 2)the butt-end type, in which type with the bone margin face each other as two relatively straight edges, and 3)the squamosal type, in which the bone margins slightly overlap each other.

The degree of serration relates to the magnitude of local functional stimuli and duration of activity. These projections increase in length with advancing age (27,29).

Moss has demonstrated, by transplanting sutural segment intracerebrally in the growing rat, that when the serrated configuration was freed from the functional stimuli it would change into a butt-end structure (39). This was supported by the experiment performed in rats by Smith and McKeown (50). They detached and replaced in situ segments of the coronal suture and found that either a butt-end configuration or a minimal overlapping structure resulted.

Sutures are capable of adapting and changing in configuration when subjected to the different functional demands in order to maintain regional integrity within the craniofacial complex. However, the serrated three-

dimensional interdigitation is better for accommodation to the shearing forces than the end-on-end type of suture (41).

Functions of sutures.

Many investigators have suggested two generally accepted functions for sutures. First, sutures provide a firm, although somewhat flexible, mechanical union between adjacent bones. Sutures prevent bone seperation under the external forces, resist and absorb shocks from the functional enviroment and allow slight movements between bones of the cranial and facial skeleton. Second, sutures provide a site for active bone growth which is stimulated by physical seperation of the bones resulting from increasing brain mass and other overlying tissues (1,2,6,42,29,39,47).

Persson, et al have suggested a third function, that sutures may partially modulate the effects of external forces(the increasing neural mass) on the sutures and thereby modify the rate of seperation of adjacent bones (46). Babler, et al have suggested that the suture tissue itself acts as a primary restraint during translatory growth, since a significant accelerated seperation rate between the frontal and parietal bones was found when the coronal suture was surgically removed (5).

Remodeling growth functions in two ways. First, it

serves as a mechanism adapted to the simultaneous adjustment and repositioning of each regional area in order to maintain the proportionate size of bone (relocation). Any given part of a bone is constantly moved away by combinations of resorption and deposition on selected inner and outer cortical surfaces. Secondly, it brings about a proportionate change in the size of all parts of each bone as the whole bone becomes elongated (11).

To better understand the mechanisms of craniofacial growth, researchers have tried to influence the development of the cranial and facial bones by altering bone deposition at the sutural margins by various means. Some experiments have intentionally fused or even removed the sutures to determine the effect on subsequent growth and development. In general, the results of those experiments indicate that sutures are sites of compensatory growth responding to the soft tissue enviroment and functional demands within which they normally exist.

Sutural responses to tensile and compressive force.

Several experiments were performed to accelerate or retard the growth of certain facial bones by applying tensile or compressive force to the interested sutures in order to better understand the adaptive nature of involved tissues.

Tuenge and Elder(1974) subjected Rhesus monkeys to extraoral high-pull forces of 700 grams to the maxilla for 69 to 72 days and evaluated the post-treament changes histologically and radiographically. Differential bone deposition at all sutural margins was found. The osteoclastic activity induced by treatment reversed quickly, while the osteoblastic activity seen as a result of treatment was very slowly reversed (54).

Jackson, et al (1979) evaluated the effect of an extraoral anterior-pull appliance on the dentofacial complex in Macaca nemestrina monkeys by means of cephalometric, histologic, and gross techniques. They found the compensatory deposition and resorption of bone not only at the sutural margins but also on external surfaces of bones. The amount of remodeling appeared to be proportional to a suture's distance from and orientation to the applied force system. The number of sinusoid vessels in the sutural ligament tended to increase in those sutures subjected to tensional force (26).

Linge (1972) observed sutural tissue reactions in maxillae of Rhesus monkeys subjected to an orthodontic expansion screw. He found that all collagen fiber bundles were pulled in the direction of mechanical traction, with pyknotic nuclei noted in areas where the direction of collagen bundles had been changed.

Accretion of cells in the middle zone of the suture were observed. Dilated blood vessels, capillaries and intercellular vacuoles were found in proportion to the degree of sutural widening. After a few days these tissue changes were characterized by remodeling. Eventually bone deposition and normalization of sutural width were resumed (35).

These findings were supported by a study, in rat, performed by Miyawaki and Forbes (1987) in which they found a correlation between the magnitude of tensile forces and osteogenic response (38) and also in a study of Ten Cate, et al in which sutures responded to rapid expansion by osteogenesis and fibrillogenesis and finally followed by remodeling (52).

Interestingly, when transverse expansion force was applied to the maxilla of young male hypocalcaemic Sprague-Dawley rats, the internasal sutures showed marked synostosis extending to one-fifth of the sutural area in the posterior part of the viscerocranium. This was suggested by Engstrom, et al (1985) to be due to the restriction of interosseous movements secondary to the expansion and the disturbance in bone remodeling induced by hypocalcaemia (7,8).

Sutural responses to altered enviroment.

Several experimental studies have altered the normal enviroment of various sutures in order to observe the

effect on sutural growth and subsequent craniofacial development.

Grigis and Pritchard (1958) studied the effect of skull damage by using glass cautery through the intact uterine wall on the development of the sutural pattern in 17 to 19 days old fetal rats. They found deviation of the sutural pattern in subsequent growth was due to an arrested development of the damaged bone but with overgrowth of undamaged bony margins (22).

Mabbutt and Kokich (1978) performed an extensive craniectomies on 10-14 days old rabbits and evaluated the subsequent regeneration of the calvariam grossly, radiographically, and histologically. They found a gradual regeneration of calvariam in which proper anatomic sutural articulations redeveloped although there was a tendency for synostosis to occur earlier than in normal sutures (37).

Watanabe, et al (1957) transplanted the zygomaxillary suture from one side of the face of a 3-week-old guinea pig to the subcutaneous tissue of the right abdomen, and then histologically examined the adaptation of the suture in the new enviroment. The result showed a gradual narrowing of the suture and eventual obliteration by the inward growth of new bone. This finding supports the view that tension transmitted to the suture from the growth of growing brain, muscle, or

other soft tissues has an important influence on the growth of sutures (56).

PREMATURE SYNOSTOSIS

All fetal premature synostoses have been found exclusively among the cranial sutures, not the facial sutures. Fetal head constraints (18,19,20) and intra-uterine compression (31) have been suggested as the causative factors even though such factors have failed to cause premature craniosynostosis after birth in experimental studies. Several possible reasons for this were proposed by Moss as a result of his study in which the periosteum was stripped from several cranial sutures to induce trauma,resulting in sutural fusion (41); as follows.

1. The maturational stage of the sutural area. Only presumptive, immature sutures respond by fusion since the sutural tissues were not yet differentiated and they did not yet exhibit the fibrous layers encapsulating the periosteum. Therefore the sutural tissue responses to trauma were able to spread across the entire suture area from one periosteal layer to the other.
2. The relative growth of the brain. Neo-natal fusion was uniform only in the sutural areas overlying non-expanding portions of the brain.

3. The differential functional demands. The sutural areas where morphology indicated relative non-function become fused consistently.

Premature synostosis among facial sutures never naturally occurs in the compressive condition. Two reasons were suggested.

1. Facial bones develop fibrous periosteal capsules around themselves, but the cranial primordia develop toward each other in a preformed continuous membrane known as the ectomininx. The periosteum of the facial suture would have a protective effect against fetal compression.

2. The neurocranium exposes its sutures to higher compessive forces in utero than the facial skeleton because of the more prominent shape, size, and the more rapid growth onset and rate of growth.

The evidence to date strongly suggests that pathogenesis of craniosynostosis is heterogeneous and that several mechanisms may be responsible for the same defect. There are several contributing factors to the craniosynostosis as listed below (6).

1. Genetic.
2. Metabolic: hyperthyroidism, hypercalcemia, vitamin D. deficiency, mucopolysaccharidoses, and mucolipidoses.
3. Hematologic: hyperplasia of the marrow, sickle

cell anemia, polycythemia vera, and congenital hemolytic icterus.

4. Drug teratogenic: diphenylhydantoin, valproic acid retinoic acid, oxymetazoline, aminopterin, and methrotrexate.
5. Physical: fetal head constraint, malformation, i.e.,microcephaly, encephalocele, shunted-hydrocephaly, and trigonocephaly.

Experimental manipulation on sutural synostosis

To simulate the clinical effect of premature craniosynostosis, many researchers have intentionally caused sutural fusion in experimental animals and have evaluated the subsequent growth of the skull cephalometrically, radiographically, and histologically. Various methods have been used to promote the earliy fusion of sutures. These methods included periosteal transplant, cyanoacrylate adhesive, periosteal excision, sutural destruction, and other methods.

Periosteal transplantation

Alhopuro (1973) used a free periosteal strip detached from the tibia of the same 14-day-old rabbit and transplanted immediately to the area of the unilateral extirpated and non-extirpated frontonasal sutures. He was able to induce bony fusion in both models within 3 weeks. The animals exhibited the deviation of their snouts to the operated side (1). The same procedure was

also performed in 14-day-old guinea pigs to achieve unilateral premature fusion of the premaxillo-maxillary suture. Bony bridges were formed in 4 weeks and resulted in deviation of the snout to the operated side, lengthening of zygomatic arch, and mesial wandering of molar teeth on the operated side (2).

Cyanoacrylate adhesive

Foley (1980) unilaterally immobilized the coronal suture in rabbits of various age using cyanoacrylate adhesive. He found the formation of a ectocranial periosteal bone bridging; not a synostosis within or across the internal portion of the sutural ligament, in rabbits of less than 8 weeks of age. He suggested the term " periosteal bone bridge " for this biological response (15).

Nappen proposed the important role of periosteum in inducing premature fusion of suture by the cyanoacrylate adhesive method. He found that the adhesive did not consistently induce synostosis in the growing rabbit if the periosteum was excised (44).

In order to successfully fuse a suture with adhesive the following conditions should be allowed.

1) Intact periosteum over the suture.
2) Direct contact of adhesive to the periosteum.
3) Roughened bone surfaces on both sides of the sutures (44,46).

Persson,et al (1979) successfully fused the coronal suture in 9-day-old rabbits by immobilization of the sutural area bilaterally with methyl-cyanoacrylate adhesive. Analysis of skull morphology by means of radiographic cephalometry revealed significant changes in angular dimensions of the vault with anteroposterior shortening. Craniectomy at 30 days after a skull deformity had been established resulted in a rapid separation of the bone at the sutural site which returned the skull to a normal configuration by 90 days of age (46).

This finding was in part supported by the study done by Babler and Persing (1982) who used the same method to create a bilateral premature fusion of the coronal suture in 9-day-old rabbits. Growth and morphometric changes were monitored in cephalometric radiographs by measuring changes in the location of amalgam implants. They found no compensatory (increased) growth at the other transverse sutures of the vault. Rather, growth at the frontonasal suture was markedly reduced, and it was not corrected after linear suturectomy of the fused suture. However, evaluation of anteroposterior dimensional change of the entire skull after suturectomy was not made (4).

Sutural destruction .

Sarnat (1958) surgically removed the median and

transverse palatal sutures in 8 months old Macaca rhesus monkeys in an attempt to create palatal defects. The defects were bridged by bone with regenerated sutures 34 months later. These results showed that suturectomy failed to induce synostosis (48).

Selman and Sarnat (1957) studied growth of the snout of 42 to 154 days old rabbits after bilateral and unilateral extirpation of frontonasal sutures. They found no significant difference in the total amount of increase of implant separation or in the size of the snout between the control and experimental groups or between the left and the right sides of animals in which there had been only a unilateral extirpation of the suture.

This finding was in contrast to the study by Babler, et al (1982) in which the coronal sutures of 6-8 weeks old rabbits were surgically removed. They found a significantly accelerated rate of separation of the freed frontal and parietal bones. Interestingly, analysis of growth at the adjacent sutures revealed that the accelerated separation of bones after suturectomy was compensated by reduced growth at these adjacent sutures (5).

Stenstrom, et al (1967) extirpated the premaxillo-maxillary suture unilaterally from of 1 month old guinea pigs with replacement by a bone graft in some animals

but with the defect left open in others. Three months after the operation, marked asymmetry of the snout due to deviation of the premaxilla toward the affected side was noted in the animals in which the defects were filled with bone graft. No asymmetry was found in those animals having the defects left open (51).

Periosteal stripping

Moss (1960) manually scraped periosteum overlying several sutural areas with an irridectomy knife in 4-day-old rats. Ectocranial fusion of sutures were found. Extension of the fusion through the depth of sutural ligament were found to be inconsistent (41).

Other methods .

Giblin and Alley (1944) tried to promote the premature fusion of the coronal suture in 3-week-old puppies by cutting a circular disc of bone within which was part of the coronal suture formed a diameter or major chord. This disc was rotated until the disc portion of the suture was aligned at 90 degree to the rest of the suture. However, 5 weeks later, only one-half of the circumference of the disc showed bony union with the other half remaining fibrous and unossified and serve to re-establish the continuity of the coronal suture (16).

PURPOSES OF THE STUDY

Although numerous experiments have been carried out to promote premature synostosis of sutures, with subsequent changes of involved bones and compensatory growth of adjacent sutures, the process of bone remodeling itself had not been eveluated in those experiments.

The purpose of this study was to determine the adaptive nature of compensatory remodeling of cortical bone throughout the nasomaxillary complex of animals subjected to unilateral premature fusion of the zygomaxillary suture by immobilization using methyl-2-cyanoacrylate adhesive.

The primary objective was to determine (1) the extent to which the process of bone remodeling can compensate during facial morphogenesis in response to the occurrence of experimental sutural synostosis, and (2) the locations and pattern of such remodeling adjustments.

RESEARCH QUESTIONS

1. Is there adjustive compensatory growth at adjacent sutures and within the facial bones responding to unilateral premature fusion of the zygomaxillary suture?.

2. What is the adaptive nature of compensation?.
3. Are there any differences in that compensation when
   a. The premature fusion side is compared to the non-fused side.
   b. The experimental group is compared to the control group.
4. If differences exist, what are the specific locations and amounts?
5. Is there developmental asymmetry when comparing the fused side to the non-fused side?
6. If there is developmental asymmetry, does it develop into a structural asymmetry?
7. Is the distribution of depository and resorptive fields in the involved bones altered?.
8. How does compensatory bone remodeling help explain morphological changes in the involved bones?.

HYPOTHESIS

H1: Lack of the displacement type of growth movement of one bone affects the remodeling growth pattern of that particular bone both qualitatively and quantitatively (changes in location of depository and resorptive areas and regional amounts of growth).

H2: Changes in the remodeling growth pattern of the fused side are different from those of the non-fused

side.

NULL HYPOTHESIS

NH1: Lack of the displacement type of growth movement of one bone does not affect the remodeling growth pattern of that particular bone both qualitatively and quantitatively (changes in location of depository and resorptive areas and amount of growth).

NH2: Changes in remodeling growth pattern of the fused side are not different from those of the non-fused side.

ASSUMPTIONS

1. All male and female, Hartley[a] strain guinea pigs of the same age possess the same morphologic skeletal pattern.
2. A linear growth pattern exists.
3. The surgical procedures do not affect growth potential except for the sutural fusion process itself.

---

a.(Hartley) - from Charles River, North Wilmington, Massachusetts.

MATERIALS AND METHODS

I) Laboratory methods.

Four two-week-old guinea pigs were used as a sham-operated control group.

Sixteen two-week-old guinea pigs were used as the experimental group.

Guinea pigs were chosen for this study because they are fast growing, have a relative large head size compared with rats, and extensive use in previous sutural growth studies make it one of the best known and reliable animals for this kind of experiment.

1) Experimental group:operative procedures.

a) All animals were anesthetized by the intraperitoneal injection of 30 mg./kg. pentobarbital.

b) On both sides of the face, skin shaving and local injection of 2% xylocaine at the site of surgery were performed. Incisions were made through the skin and underlying fascia to expose the zygomaxillary sutures. The periosteum adjacent to the sutures, not the periosteum across the sutures, was removed (Figure 1). The exposed bone surfaces of the left side of the face were roughened with the point of a needle to increase adhesive retention.

c) On left side, the zygomaxillary suture was immobilized by the application of three thin layers of

methyl-2-cyanoacrylate adhesive (Permabond 910)[b]. No adhesive was applied to the suture on the right side.

d) Amalgam implants were placed in the cortical bone on each side of the sutures on both left and right sides of the face (Figure 2). These cavities for amalgam were prepared by the use of a #35 inverted cone bur to a diameter of 0.75 mm.

e) Flaps were approximated with silk sutures.

2) Control group:operative procedures.

The procedures described above were repeated except that exposed bone surfaces on the left side of the face were not roughened and no adhesive was applied.

3) Radiographic procedures.

Serial cephalometric radiographs were taken at the time of surgery and every two weeks thereafter.

For this procedure, animals were anesthetized with ketamine/acepromazine mixture,[1 cc. of acepromazine ,10 mg./ml., with 10 cc. of ketamine,100 mg./ml., with a dose of 0.4-0.5 cc. IM./400 g.body weight].

Each animal head was secured in a the head holder constructed particularly for the guinea pig (Figure 3). The x-ray image was projected from the dorsal aspect of the head with a 6 inch film to tube distance, 80 kVp.,

---

b. (Permabond 910) - manufactured by Eastman-Kodak, Kingsport, Tennessee.

and 15 mA.

Measurement of the distance between the two amalgam implants were made directly three times from each original radiograph and from radiographs enlarged by a factor of three. These data were processed for statistical analysis.

4) Vital staining procedures.

On the 51st day after the operation, and on each succesive 5th day thereafter, each animal was injected with contrastive dyes that allowed sufficient time for new bone to absorb vital dye . These dyes were, in sequence, alizarin complexon (3omg./kg.), fluorexan (20mg./kg.), xylenol orange (30mg./kg.), and oxytetracycline (60mg./kg.). These marker dyes will allow analysis of time, magnitude, and locations of bone remodeling.

5) Specimen preparation.

On the 70th post-operative day, each animal was sacrificed by intraperitoneal and intercardiac injection of an overdose of pentobarbital. The thoracic cage was then opened. The vena cava was sectioned for drainage and 10% buffered formalin fixative was injected into the left ventricle until the formalin came out from the sectioned vena cava. The heads were skinned, decapitated and placed in 10% buffered formalin. The nasomaxillary complexes were removed from the entire skull, cut into

two halves, and identify as left or right. The midline suture was included in the left half.

One of control animals and three of experimental animals were prepared for ground section. All soft tissues were removed. Each half of nasomaxillary complex was embedded in bioplastic. The ground sections were cut by means of power-driven diamond waffered blade at 500 microns, and then polished to about 100 microns. The sections were covered using a routine medium and examined with the aid of an ultraviolet microscope filtered for light other than 450 nm. wavelength.

Two of control animals and nine of experimental animals were prepared for decalcified section. Formic acid-sodium citrate solution was used for tissue decalcification. Following complete decalcification, specimens were washed in running tap water overnight, dehydrated and infiltrated with paraffin. The specimens were then embedded in paraffin blocks. The sections were sliced with conventional microtome adjusted to 6 micron. The sections were stained with hematoxylin and eosin, mounted and examined with the aid of a light microscope. Locations of bone formation and resorption were recorded for each section.

II) Evaluation methods

1) Sutural fusion.

a) Dorsoventral cephalometric radiographs allowed 4

implants, 2 on the left and 2 on the right, to appear without superimposing among them (Figure 4). The first films taken at the time of operation were designated as T0. The following films taken every two weeks thereafter were designated as T2,T4,T6,T8,and T10 respectively.

The distance between the two implants on each side of the face was measured directly from the radiographs. Measurements were obtained from T0 through T10 for each animal.

b) Change in the distance between the two implants of the same side from one film when compared to the proceeding film allowed for the evaluation of sutural fusion. The successful fusion was considered obtained when the distance between the two implants was not increased or insignificantly increased (total increment was less than 0.5 mm.).

2) Comparison of growth.

a) Between left and right side of the same animal.

The total increment of the distance between the two implants of the left side was compared with that of the right side.

b) Between animals.

Since each animal may vary in growth rate, the total increment of the implant seperation of each side of the face was computed to obtained the ratio of left to right

(L/R). This ratio made it possible to compare one animal with another.

3) Changes of anatomical structures.

a) All radiographs were traced and the anatomical landmarks were identified to construct the geometric structure of the skull. The anatomical landmarks were defined as follow (Figure 6):

(1) Midpoint of the junction between central incisor and the alveolar process on the palatal aspect.

(2) The most anterior point of the incisomaxillary suture.

(3) The deepest point of the anterior curvature of the upper maxillary process.

(4) The deepest point of the curvature at the junction of the maxilla proper and the anterior surface of the lower maxillary process.

(5) The most convex point of the outer cortical surface of the anterior 1/2 of the zygomaxillary arch.

(6) The deepest point of the curvature at the junction of the maxilla proper and the posterior surface of the lower maxillary process.

(7) The most convex point of the outer cortical surface of the zygomaxillary arch.

(8) The most convex point of the outer cortical surface of the posterior 1/2 of the zygomaxillary

arch.

b) A line was drawn from a point of the landmark on the left side to the point representing the same landmark of the right side. Eight lines were obtained and designated respectively as the 1st line for the line connecting landmark No.1, the 2nd, 3rd,-----,and 8th line for the line connecting landmark No.8.

c) The midpoint of each line was located. A line was drawn to connect each midpoint except the midpoint of the 4th and 6th lines which were connected by a seperate line.

d) From landmark No.1, a line was drawn to the 2nd line of the same side. This line was parallel to the line connecting midpoint of the 1st and 2nd lines. The same procedures were repeated to create the line connecting landmark No.2 to the 3rd line, landmark No.3 to the 5th line, landmark No.5 to the 7th line, landmark No.7 to the 8th line, and landmark No.4 to the 6th line. Each new line was parallel to the midline of each section of the skull. These lines were designated respectively from anterior to posterior as a,b,-----, and f (Figure 7).

e) By comparing the linear anteroposterior(AP) dimension of each pair of a,b,-----, and f, it was possible to evaluate the changes of anatomical structures which reflected the compensatory growth of both adjacent sutures and cortical bones.

4) Ground section.

Vital dye lines represented the sites where new bone was formed. Since four different kinds of vital dye were injected in a sequence, the vital dye lines would appear in the same sequence as well. Color of each vital dye can be listed as follow:

Alizarin complexon - red

Fluorexane - green

Xylenol orange - orange

Oxytetracycline - yellow

Thus the yellow front would be the closest one to the cortical surface, followed respectively by the orange, green, and red front in the direction away from the cortical surface. The locations of vital dye lines were then recorded with corresponding colored pencils for futher interpretation.

5) Decalcified section.

The locations of bone formation and resorption were recorded by using the following citeria:

a) Area of bone formation.

- Lamellar bone pattern with primary longitudinal canal.
- Surface lined with osteoblasts.

b) Area of bone resorption.

- Surface characterized by convoluted type of compact bone.

- Scalloped, eroded, and serrated appearance of the surface with multinucleated osteoclasts.

Mapping of depository and resorptive areas of cortical bones of the entire nasomaxillary complex was determined by combinding the location of resorptive and depository areas obtained from ground and decalcified sections.

III) Statistical methods.

1) Error of measurement.

The measurement of the distance between the two implants on each film was made three times for both the original and enlarged films. Then the error of measurement was computed by utilizing Student t-test.

2) Evaluation of growth.

Since each measurement was made three times, the mean of the distance between the two implants was used for computation.

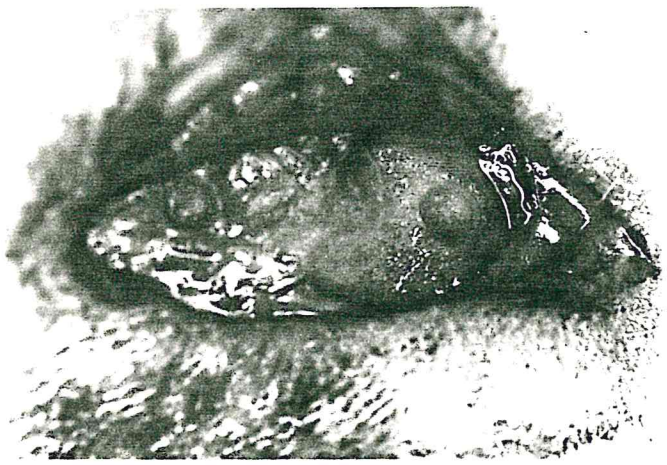
Multivariated Analysis of Variance (MANOVA) was used to test for the Significance of means for the following evaluations.

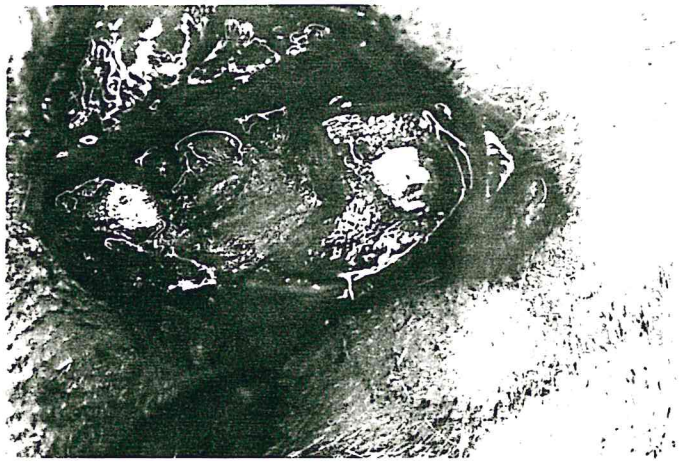
a) Effect of time on growth.(overall time)

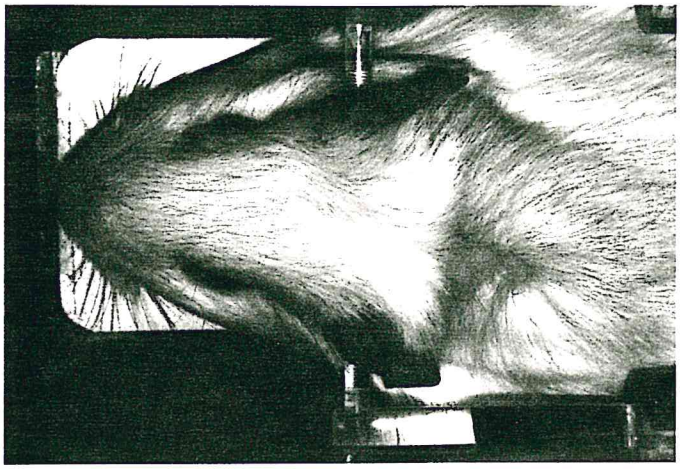
b) Effect of side on growth.(left,right,left & right)

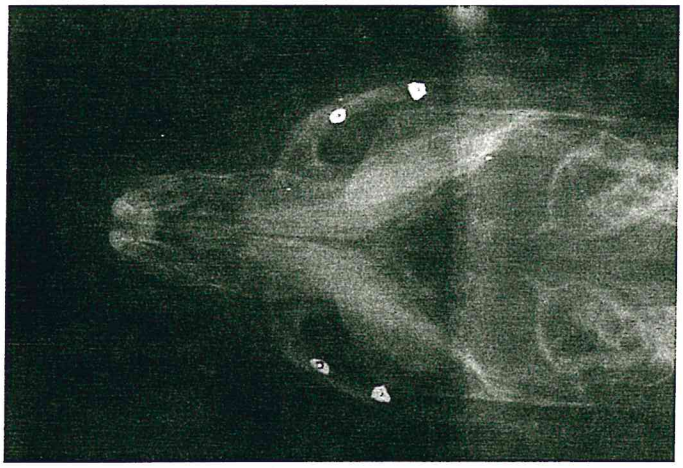
c) Time and side interaction.

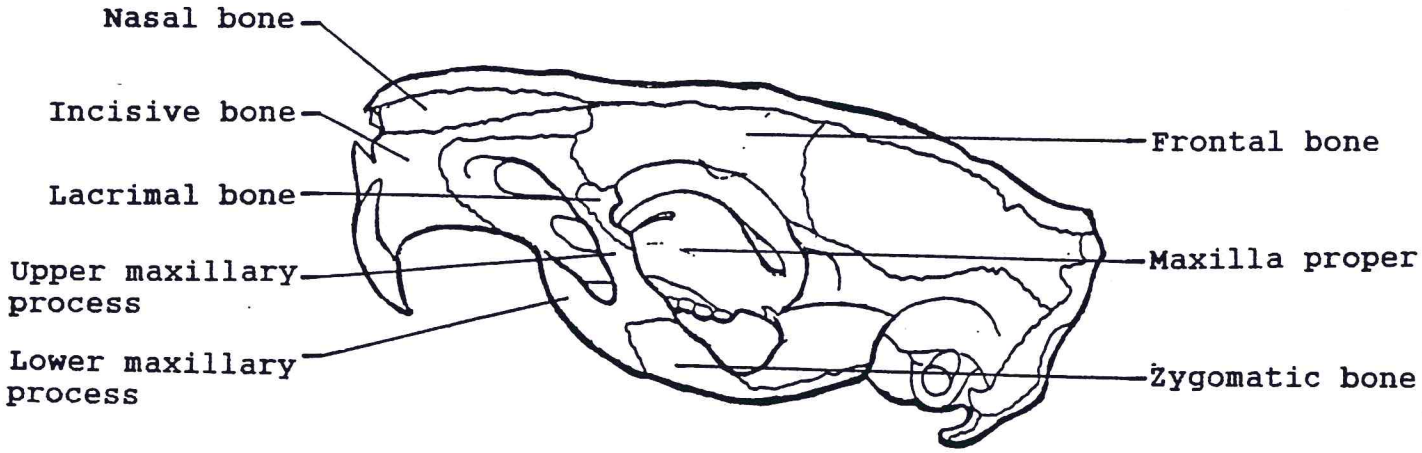
By using the Wilcoxon matched-pairs signed-ranks test, it was possible to evaluate and compare mean growth of left and right side at each time interval.

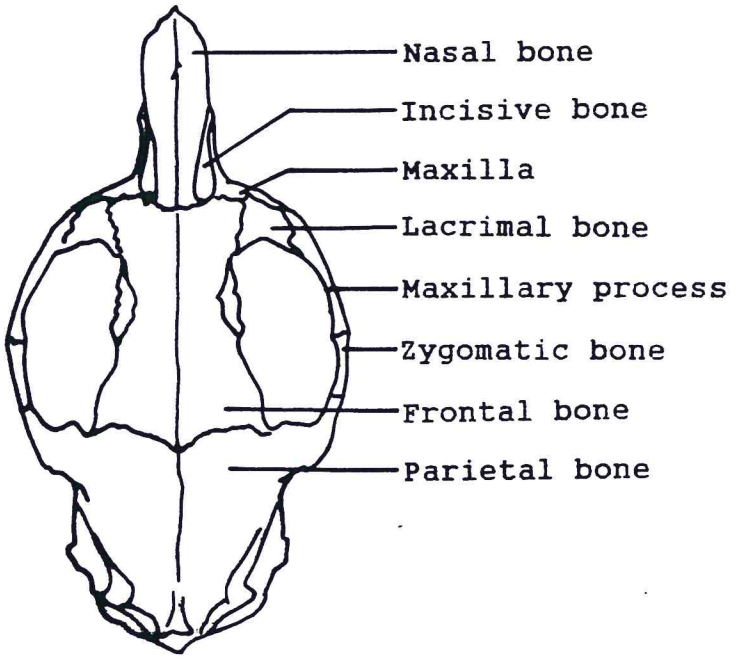


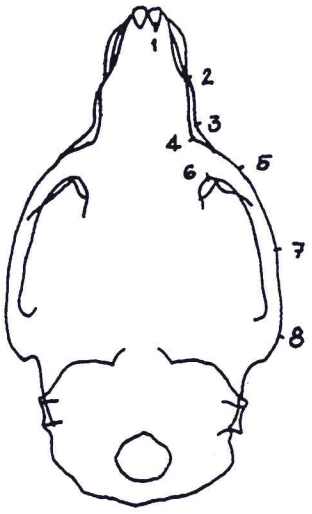


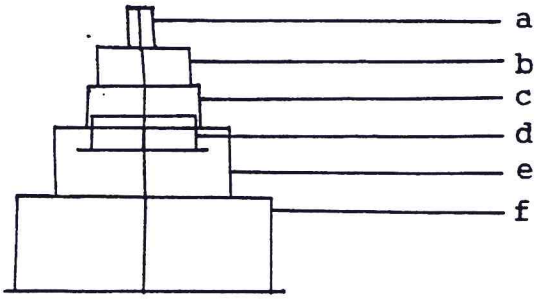





Nasal bone
Incisive bone
Lacrimal bone
Upper maxillary process
Lower maxillary process
Frontal bone
Maxilla proper
Zygomatic bone


Nasal bone
Incisive bone
Maxilla
Lacrimal bone
Maxillary process
Zygomatic bone
Frontal bone
Parietal bone


1
2
3
4
5
6
7
8


a
b
c
d
e
f

RESULTS

One control animal and three of experimental animals were died as the result of anesthetic complications during the operative procedures. Twelve animals remained; all of them tolerated the surgery very well. Silk sutures were removed on the 7th day after the surgery. All animals showed satisfactory weight gain post-operatively.

I. Evaluation of growth.

1) Control group.

Animals differed in the total amount of growth. However, comparison of the left and right sides of the same animals tended to show similarity in total growth even though at each time interval the left and right sides might have grown differently (Table I). Two animals exhibited almost one to one for the R/L ratio and one animal exhibited almost one to two for the R/L ratio (Table III).

2) Experimental group.

Each animal differed in the total amount of growth. In all animals except animals III9, III5, and I1, the right non-fused side grew significantly more than the left fused side. The average increment of the distance between the two implants on the fused side of each animal in this group was less than 0.5mm., indicated

that successful sutural fusion was obtained (Table II). In animals III9, III5, and I1, the average increment of the distance between the two implants on the fused side of each animal was more than 0.5 mm. (Table II). This indicated that poor sutural fusion was obtained in this group. R/L ratio of all experimental animals varied from 1.93 to 52.86 (Table III).

Sutural growth by time comparing fused and non-fused sides of each animal was determined by a series of graphs plotting the distance between the two implants at each time interval (Figures 8 - 20). These measurements showed that growth was greater on the non-fused sides in both the successful and poor sutural fusion groups.

The mean bone growth of control and experimental groups were computed and illustrated (Figures 21, 22). In the control group, the fused and non-fused sides showed a parallelism between each other which reflected a similar growth rate. Parallelism between the fused and non-fused sides was not found in the experimental group, since the growth rate of each non-fused side was greater than each fused side.

3) Statistical evaluation.

a) Error of measurement.

The error of measurement of the distance between the two amalgam implants was computed for both original and enlarged radiographs.

Mean error of the original radiographs was 0.000 with the SD. error of 0.087. Mean error of the enlarged radiographs was 0.000 with the SD. error of 0.009. However, the measurement obtained from the original radiographs was used for the further computation since it represented the actual amount of implant seperation.

b) Effect of time and side on growth(time by side)

Control group: No interaction between time and side(p = 0.09).

Experimental group: There was an interaction between time and side(p = 0.000).

c) Effect of time on growth(time)

Control group: Lt. side - significant effect(p = 0.007)
Rt. side - significant effect(p = 0.007)

Experimental group: Lt. side - no significant effect(p = 0.133)
Rt. side - significant effect(p = 0.004)

d) Effect of side on growth(side)

Control group: No effect at each interval(T0-T10).
No significant growth diference between Lt. and Rt. sides (p = 0.5930, 0.2850)

Experimental group: No effect at T0(p = 0.819)

Significant growth difference between left and right sides from T2 to T10(p = 0.045, 0.002, 0.001, 0.000, 0.000)

Premature fusion of the left zygomaxillary suture was successfully obtained and significantly constrained the growth of the left zygomaxillary suture as evidenced by implant separation.

II. Changes of anatomical structures.

1) Control group.

Comparing the left fused side to the right non-fused side, the geometric structure of the skull of all animals did not appear to show discernable change of anatomical structures. Only slight differences between each pair of constructed geometric structure, a Lt. and a Rt.,-----, and f Lt. and f Rt., was found. Similar geometric patterns were found from T0 to T10 (Figures 23 , 24, and 25).

2) Experimental group.

Comparing the left fused side to the right non-fused side, discernable differences between each pair of constructed geometric structure was found for a Lt. and a Rt.,-----,and f Lt. and f Rt., especially in the area of c, e, and f. which represented the anterior 1/2 of the maxilla proper, the upper maxillary process, and the zygomaxillary arch.

A similar geometric pattern for each animal was found at T0 and T10, but varied from T2 to T8 (Figures 26, 27, and 28). At T0, the left and right sides showed a similar pattern. From T2 to T8, appreciable changes of anatomical structures were noticed in the area of c, e, and f. For instance, f Lt., representing the posterior 1/2 of the zygomaxillary arch of the fused side, was shorter than f Rt.(non-fused side), but e Lt., representing the anterior 1/2 of the zygomaxillary arch of the fused side , was longer than e Rt.(non-fused side).By this combination, the linear AP dimension from the 5th line to the 8th line of left and right sides was equalized.

At T10, the left and right sides showed an appreciable pattern difference due to some combinations of compensatory growth from T2 to T8.

Adjustive compensation from adjacent sutures and cortical bones was noticed as a consequence of premature sutural fusion. The adjustive compensation of the fused side differed from that of the non-fused side.
Only developmental asymmetry was found when comparing the fused side to the non-fused side. No structural asymmetry developed.

III. Histological evaluation:

Each specimen was cut 15 mm. posteriorly to frontonasal suture. Each contained the anterior 3/4 of

frontal bones, the anterior 1/2 of zygomatic bones,nasal bones, maxillary bones, lacrimal bones, and incisive bones. Due to the complexity of nasoethmoidal complex and the irregularity of maxilla, only the periosteal side of outer cortical surfaces were reported and illustrated.

In the ground sections, the red and green vital dye fronts were present in all specimens (Figure 29). The orange front was found in only one animal and not in every part. No yellow front was noticed in any of the specimens. All present vital dye lines were in the predicted sequence; red, green, and orange toward the cortical surface. The location of vital dye lines matched with the location of bone formation observed in the decalcified sections. All areas that failed to show lines vital dye lines coincided with the areas of bone resorption observed in the decalcified sections. In general, when the periosteal side showed bone formation the endosteal side showed bone resorption and vice versa. This pattern was found in all specimens. By combinding and confirming each area of the ground sections to that of the decalcified sections, mapping of the depository and resorptive areas was achieved.

1) Control group.

Both left and right sides demonstrated the same pattern of depository and resorptive areas. Depository

areas were found on the entire outer surface of all bones except some areas of the maxilla and lacrimal bones. For better understanding, both areas were illustrated in Figures 34, 35, 36, and 37.

2) Experimental group.

a) The non-fused side.

Basically, the non-fused side demonstrated a similar pattern of bone deposition/resorption as the pattern seen in the control group except for the resorptive area of the maxilla in the area of the snout, just posterior to the incisomaxillary suture, and the alveolar process (areas B and C in Figure 38). In the snout part, the resorptive area (area B) was slightly smaller in the experimental group than in the control group. In the area of alveolar process, the resorptive area (area C) was slightly larger in the experimental group than in the control group (Figures 34 and 38).

b) The fused side.

Compared to the control group, the general pattern was similar except the resorptive area of the maxilla in the area of the snout, just posterior to the incisomaxillary suture, and the alveolar process (areas B and C in Figure 40). In the snout part, the resorptive area (area B) was slightly larger in the experimental group than in the control group. In the area of the alveolar process, the resorptive area (area C) was

slightly smaller in the experimental group than in the control group (Figures 36 and 40). A striking different pattern was found at the junction of the lower maxillary process and the maxilla proper (area A in Figure 41). Here, the experimental animals demonstrated a depository periosteal surface, whereas the control animals demonstrated a resorptive outer surface (Figures 31, 33, 37, and 41).

When comparing the fused side to the non-fused side of the experimental animals, the general pattern was also similar except for the resorptive areas B and C in Figures 38 and 40. In the snout part, the resorptive area (area B) was slightly larger on the fused side than on the non-fused side. In the area of the alveolar process, the resorptive area (area C) was slightly smaller on the fused side than on the non-fused side. The most striking different pattern was also found at the junction of the lower maxillary process and the maxilla proper (area A in Figures 39,41); the fused side demonstrated a depository periosteal surface, whereas the non-fused side demonstrated a resorptive surface similar to the control animals (Figures 30, 31, 32, 33, 35, 37, 39, and 41).

3) Comparison of resorptive areas.

The sizes of the resorptive areas could be ranked from the largest to the smallest as follow:

a) Area of the snout.

1st - The fused side of experimental animals.

2nd - The non-fused side of experimental animals.

3rd - Control animals.

b) Area of alveolar process.

1st - The non-fused side of experimental animals.

2nd - Conrol animals.

3rd - The fused side of experimental animals.

When comparing the bone areas that surrounded the tooth buds on the left fused and right non-fused sides in each animal, some pattern differences were occasionally encountered; i.e., the left outer cortical surface showed periosteal bone formation while the right outer cortical surface showed endosteal bone formation. Such differences varied between animals.

TABLE I

Average increment of implant separation of control group at each time interval(mm)

Animal	T2-T0		T4-T2		T6-T4		T8-T6		T10-T8	
	Lt.	Rt.	Lt.	Rt.	Lt.	Rt.	Lt.	Rt.	Lt.	Rt.
II1	0.27	0.07	0.06	0.10	0.14	0.10	0.16	0.10	0.14	0.43
II3	1.17	1.27	0.76	1.03	0.47	1.00	0.60	0.44	0.77	0.83
II2	0.36	1.14	0.64	0.70	0.16	0.73	0.37	0.43	0.43	0.57

TABLE II

Average increment of implant separation of experimental group at each time interval(mm)

Animal	T2-T0		T4-T2		T6-T4		T8-T6		T10-T8	
	Lt.	Rt.	Lt.	Rt.	Lt.	Rt.	Lt.	Rt.	Lt.	Rt.
III9	0.00	1.36	0.43	0.84	0.50	0.46	0.53	0.64	0.44	0.36
III5	0.30	0.96	0.00	0.97	0.73	1.40	0.97	0.10	0.00	0.80
I1	0.10	0.78	0.10	0.50	0.63	0.56	0.00	0.10	0.00	0.00
II8	0.14	0.30	0.03	0.20	0.00	0.03	0.00	0.20	0.00	0.44
II5	0,13	0,73	0.00	0.93	0.03	0.13	0.00	0.27	0.14	0.20
IV1	0.00	0.77	0.23	1.13	0.04	0.43	0.06	0.47	0.04	0.10
III6	0.60	1.53	0.00	1.40	0.00	0.90	0.00	0.60	0.00	0.64
II6	0.10	0.84	0.03	0.46	0.03	0.17	0.00	0.23	0.00	0.47
III4	0.03	0.97	0.07	1.47	0.00	0.33	0.00	0.73	0.13	0.57
III1	0.03	1.04	0.00	0.26	0.03	0.24	0.04	0.86	0.00	0.14
III8	0.13	5.00	0.00	1.20	0.07	0.83	0.00	0.47	0.03	0.73
II9	0.00	1.03	0.00	1.04	0.03	0.70	0.04	0.36	0.00	0.57

TABLE III

Animal	Total increment of implant seperation		R/L
	Left	Right	ratio
Control			group
II1	0.77	0.80	1.04
II3	3.77	4.57	1.21
II2	1.96	3.57	1.82
Experimental group			
III9	1.90	3.66	1.93
III5	2.00	4.23	2.12
I1	0.83	1.94	2.34
II8	0.17	1.17	6.88
II5	0.30	2.26	7.53
IV1	0.37	2.90	7.84
III6	0.60	5.07	8.45
II6	0.16	2.17	13.56
III4	0.23	4.07	17.70
III1	0.1	2.54	25.40
III8	0.23	8.23	35.78
II9	0.07	3.70	52.86

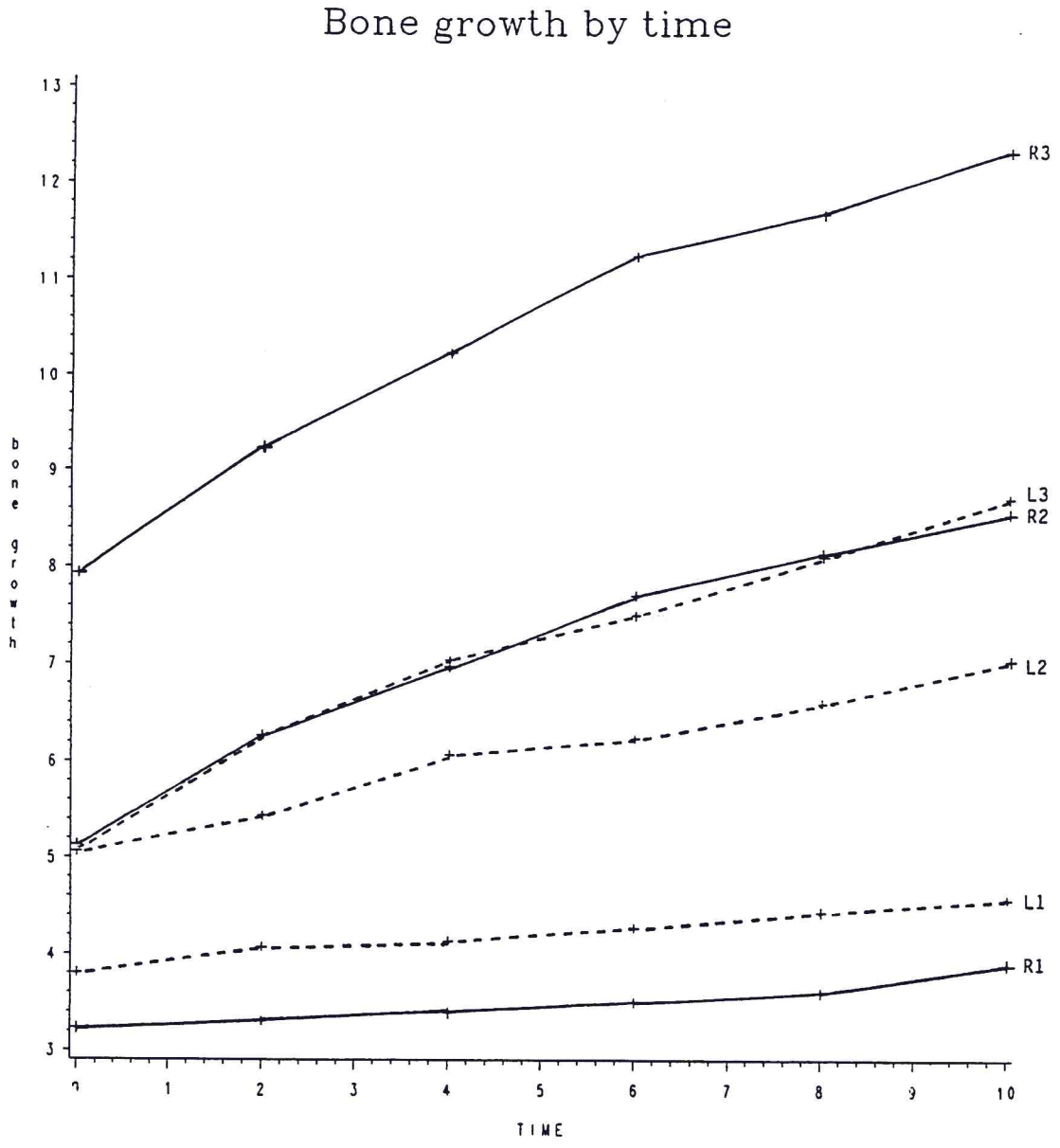


Figure 8. Graph plotting the distance between the two implants at each time interval for the control animals (II1, II2, and II3).

——— Right side

----- Left side

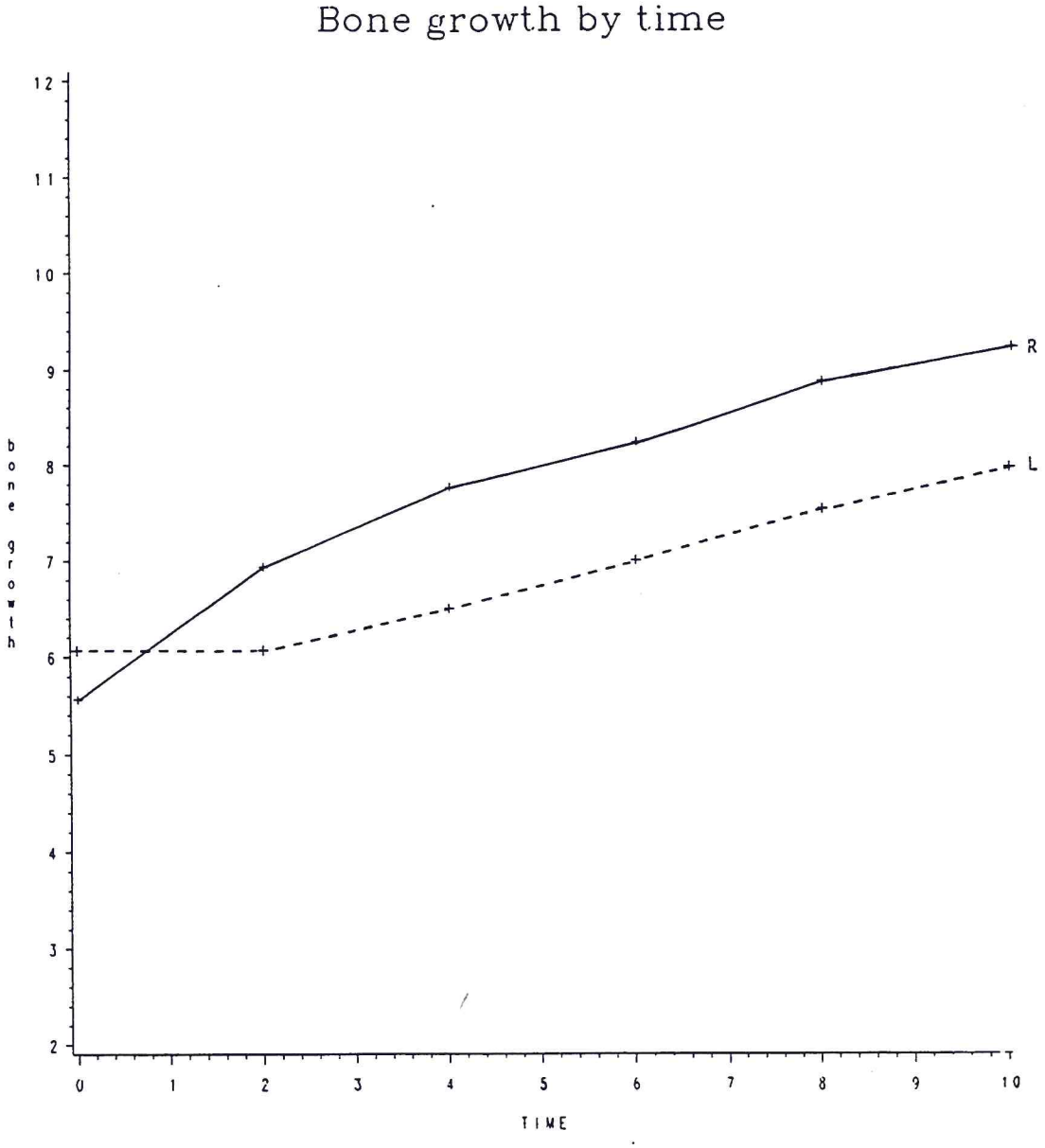


Figure 9. Graph plotting the distance between the two implants at each time interval for the experimental animal (III9).

——— Right side

----- Left side

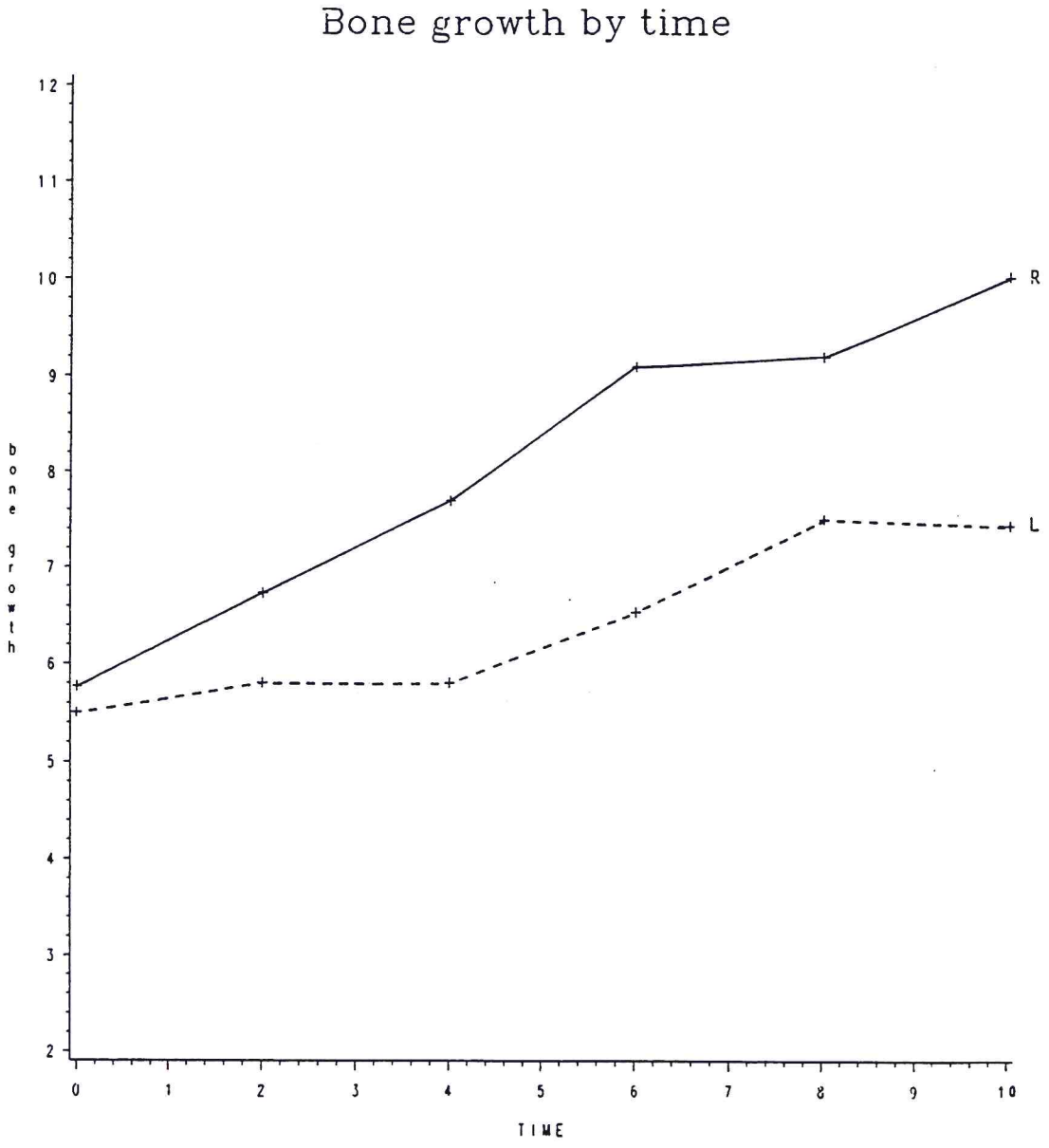


Figure 10. Graph plotting the distance between the two implants at each time interval for the experimental animal (III5).

——— Right side

----- Left side

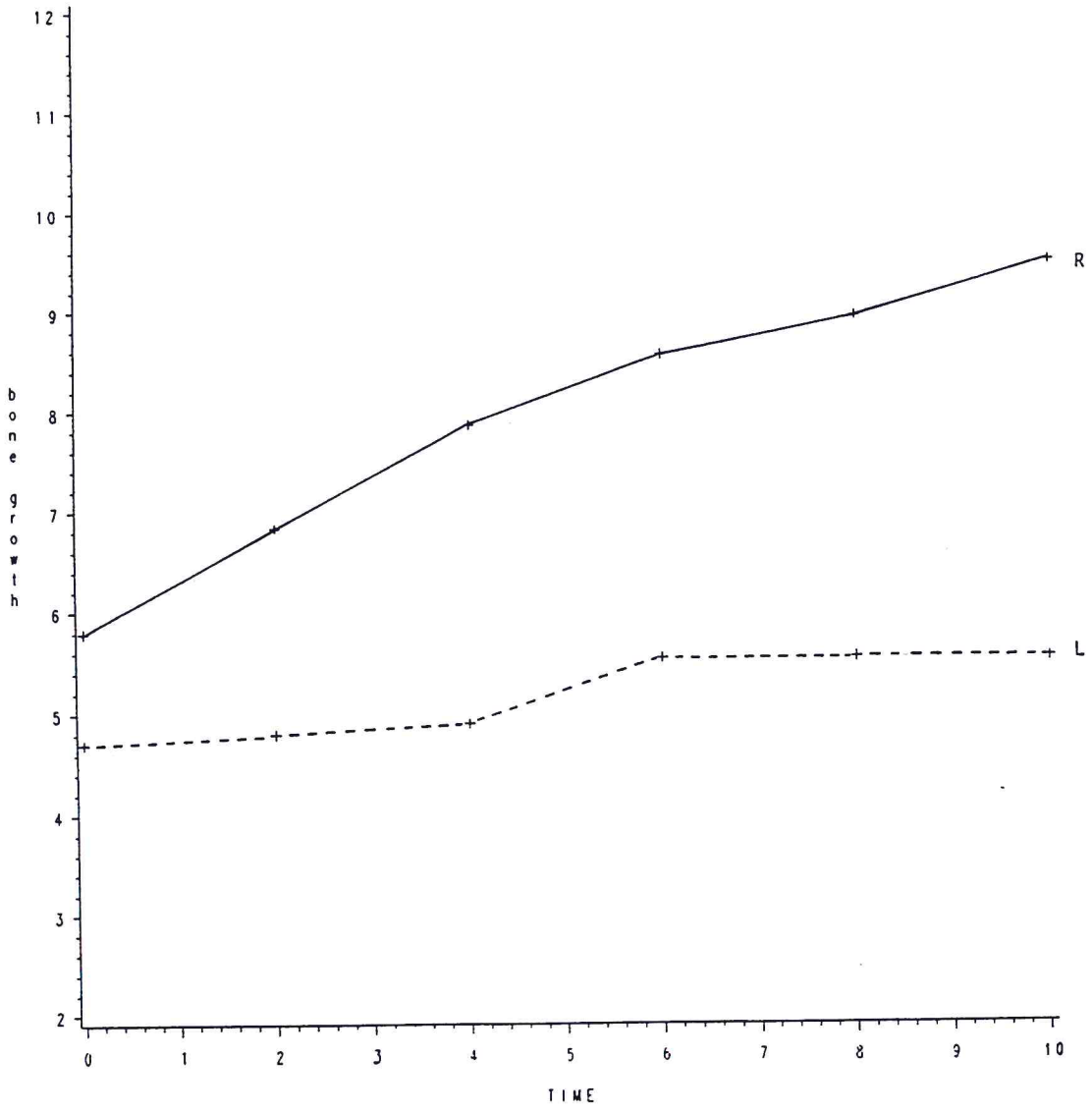


Figure 11. Graph plotting the distance between the two implants at each time interval for the experimental animal (I1).

——— Right side

----- Left side

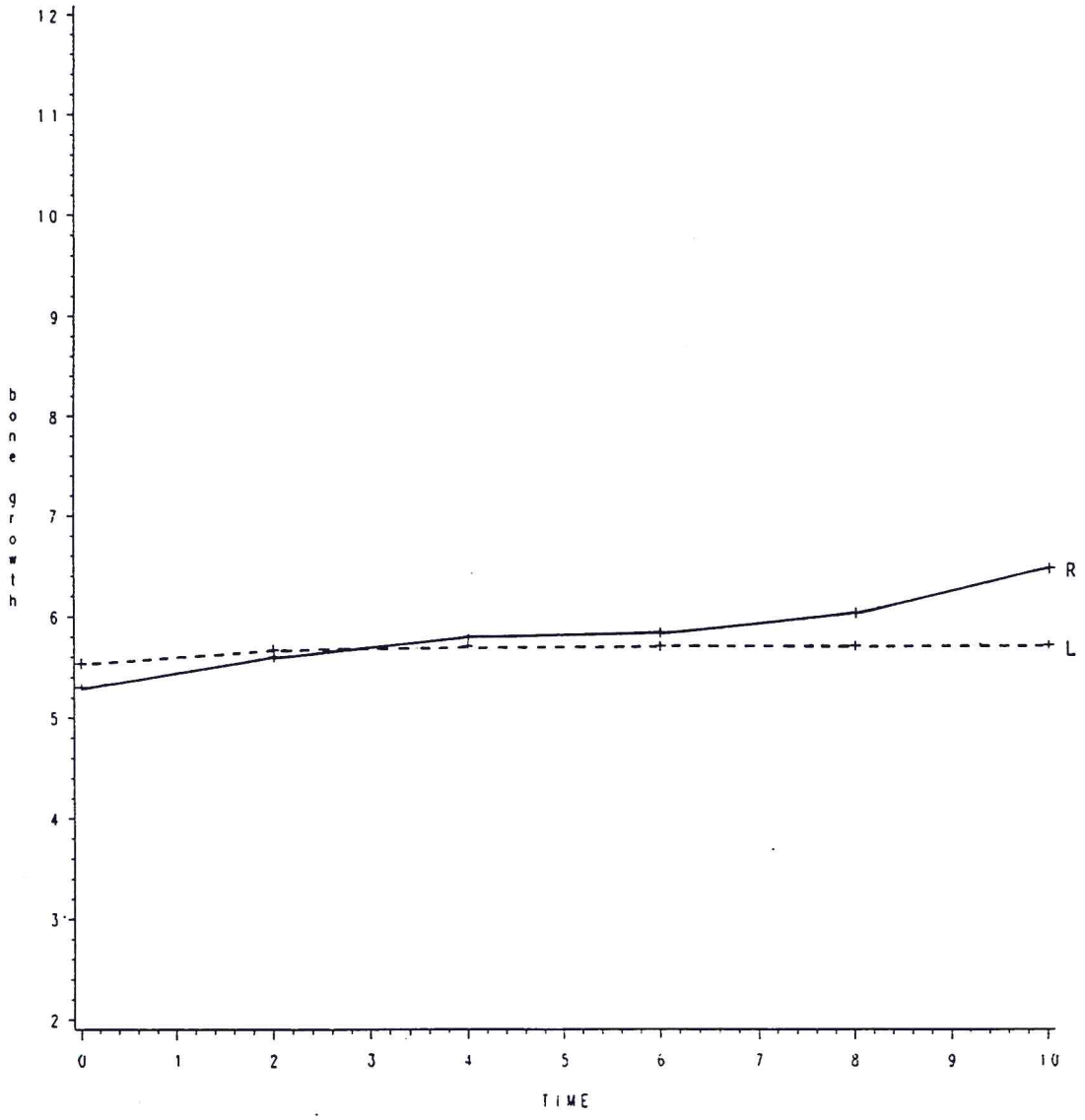


Figure 12. Graph plotting the distance between the two implants at each time interval for the experimental animal (II8).

——— Right side

----- Left side

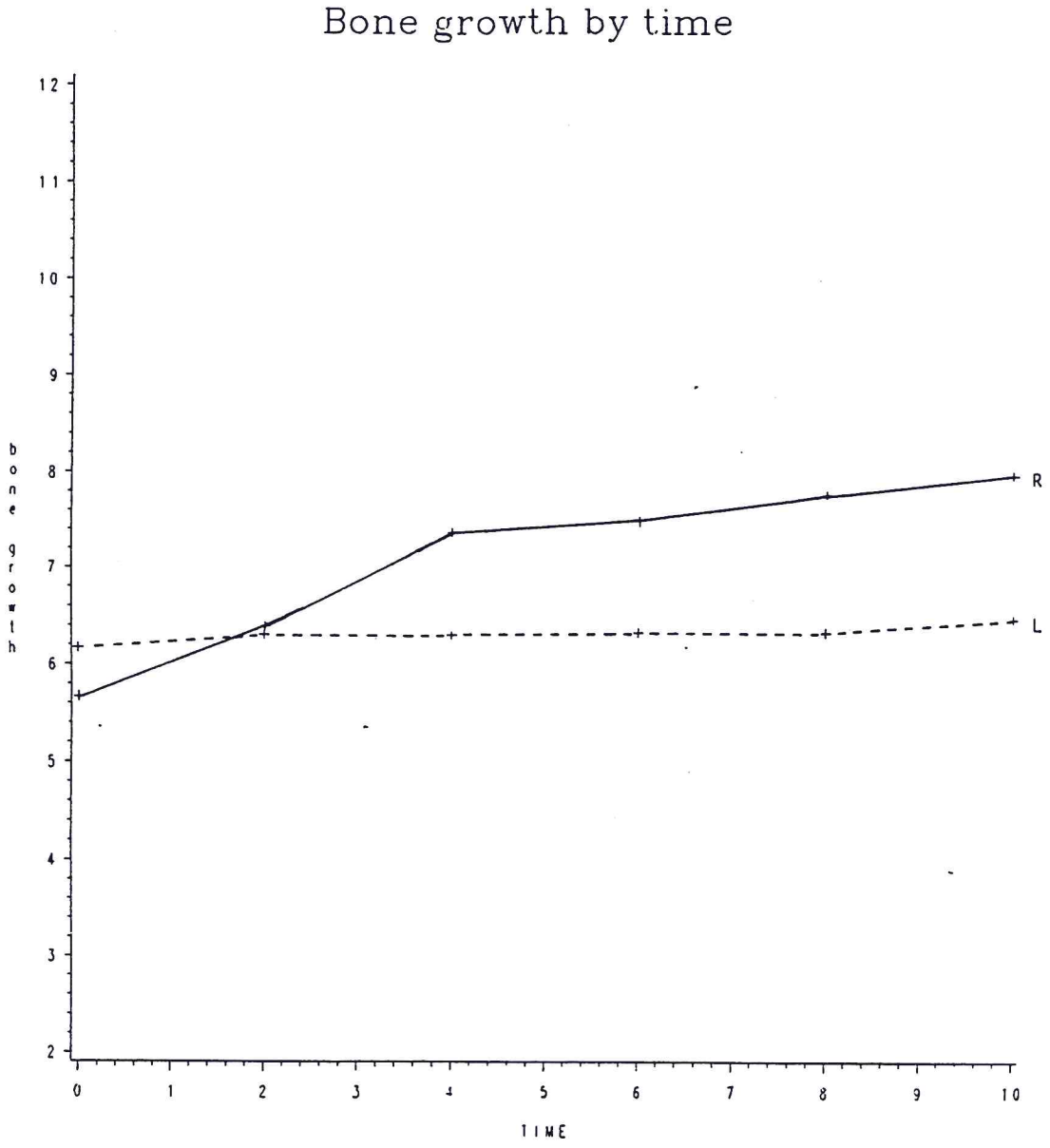


Figure 13. Graph plotting the distance between the two implants at each time interval for the experimental animal (II5).

——— Right side

----- Left side

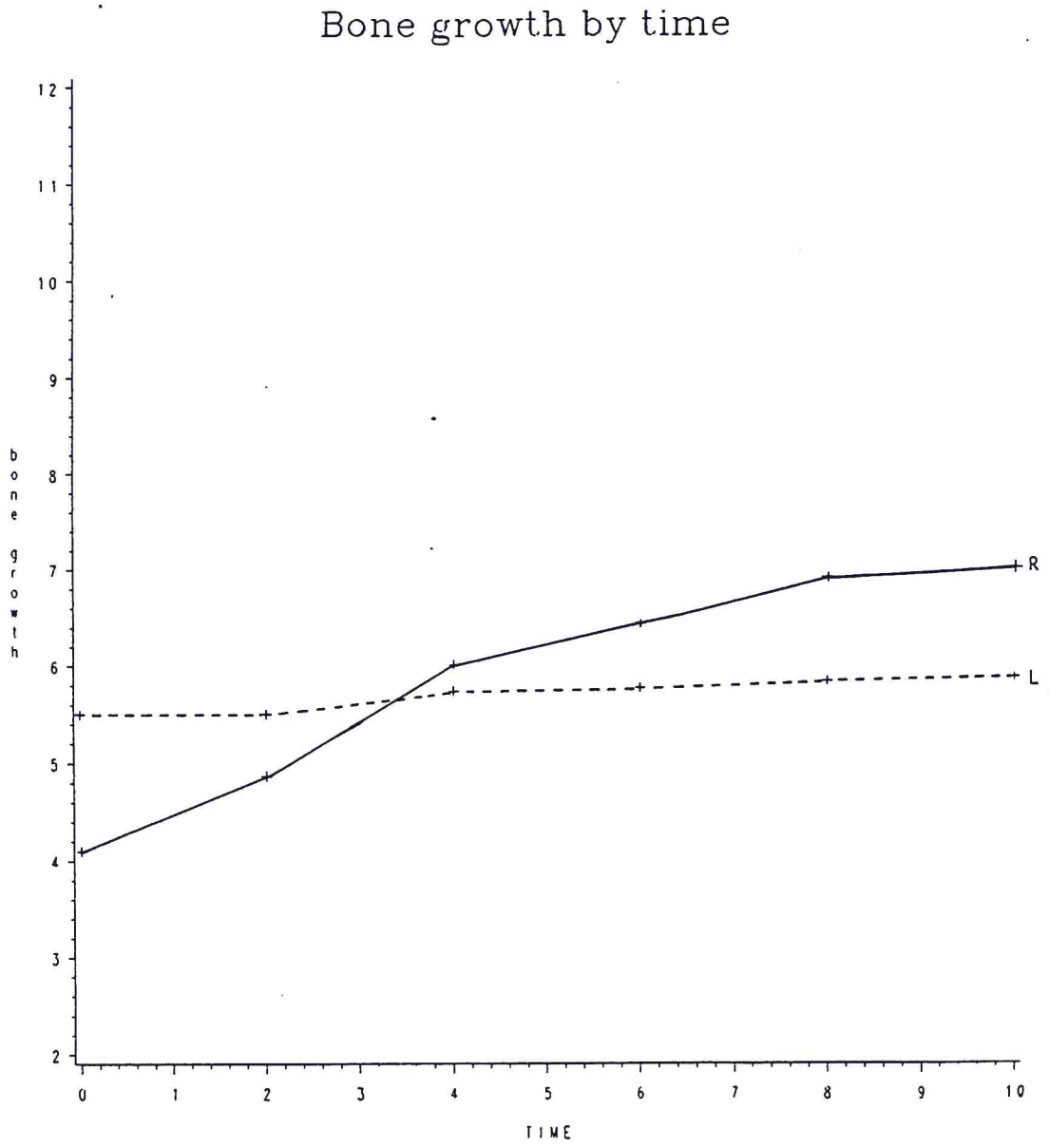


Figure 14. Graph plotting the distance between the two implants at each time interval for the experimental animal (IV1).

——— Right side

----- Left side

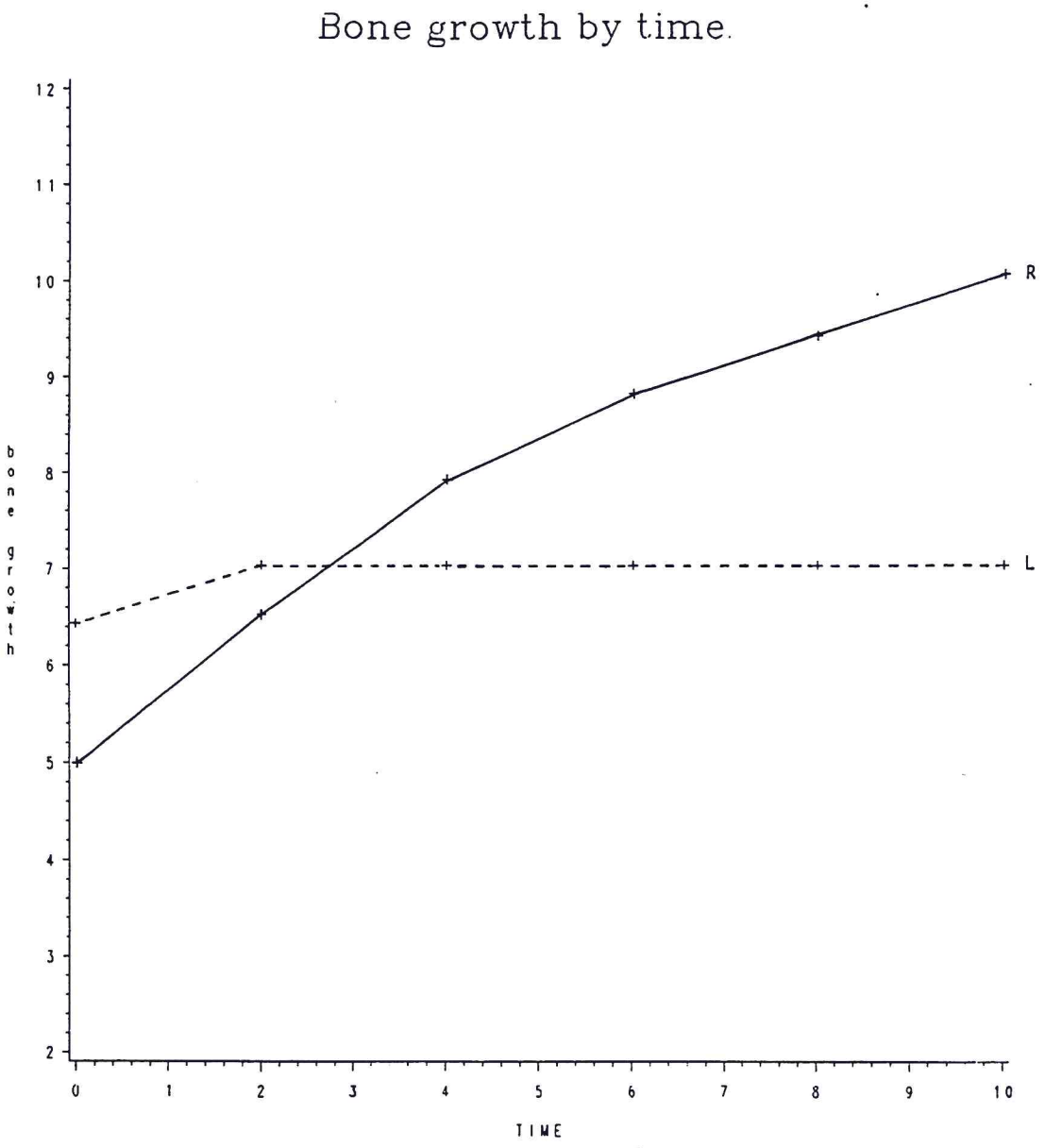


Figure 15. **Graph plotting the distance between the two implants at each time interval for the experimental animal (III6).**

——— Right side

----- Left side

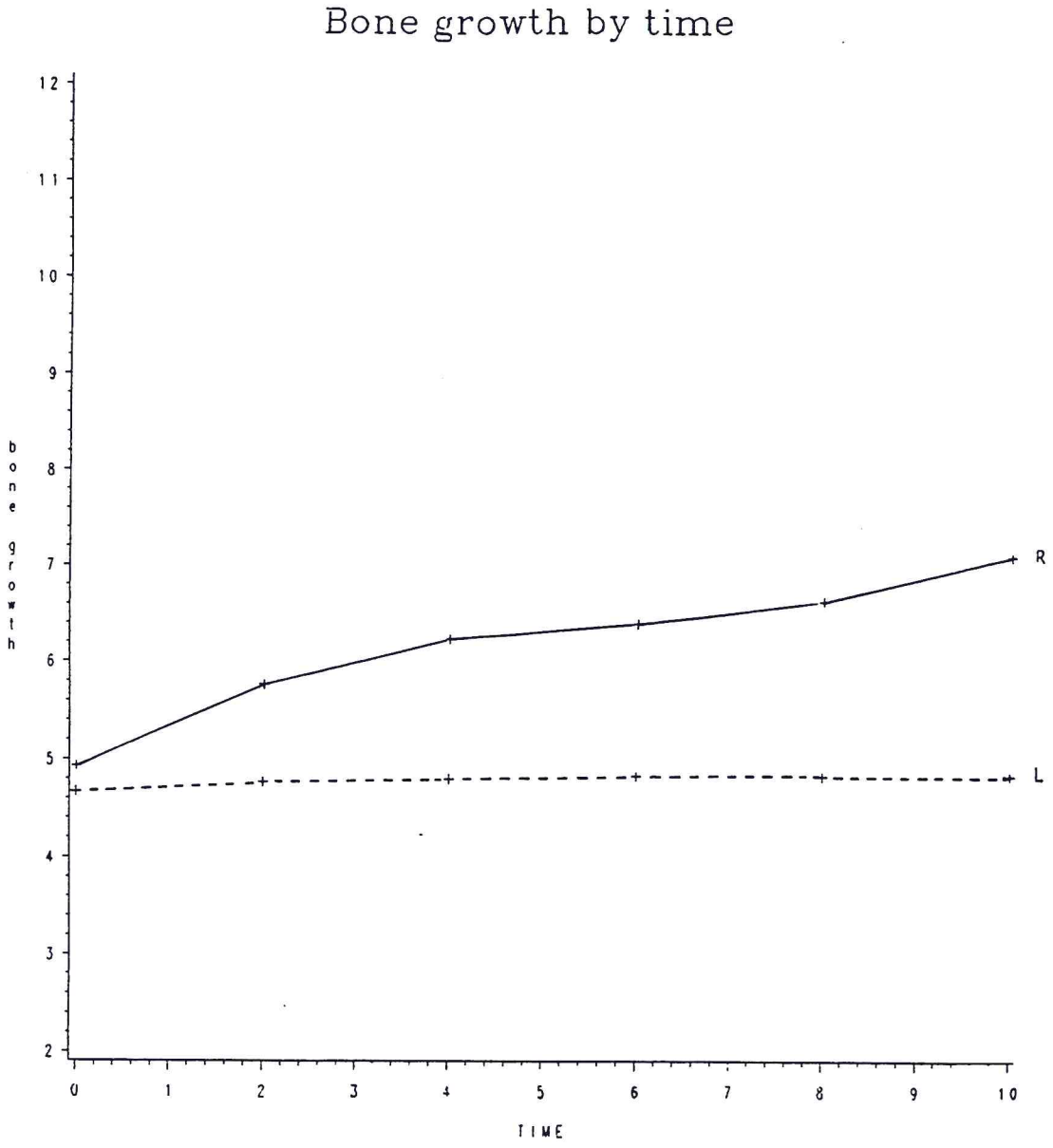


Figure 16. Graph plotting the distance between the two implants at each time interval for the experimental animal (II6).

——— Right side

----- Left side

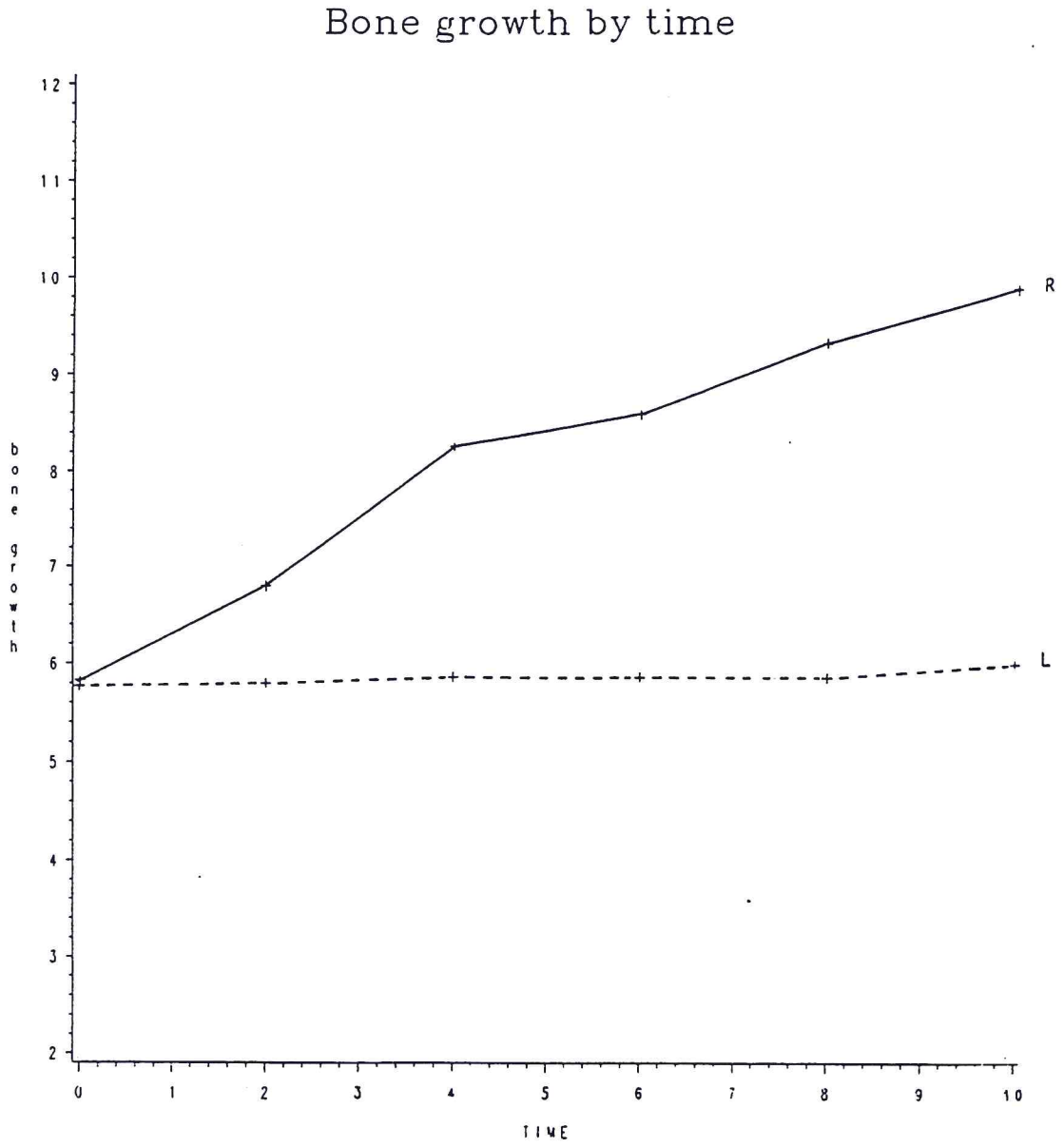


Figure 17. Graph plotting the distance between the two implants at each time interval for the experimental animal (III4).

——— Right side

----- Left side

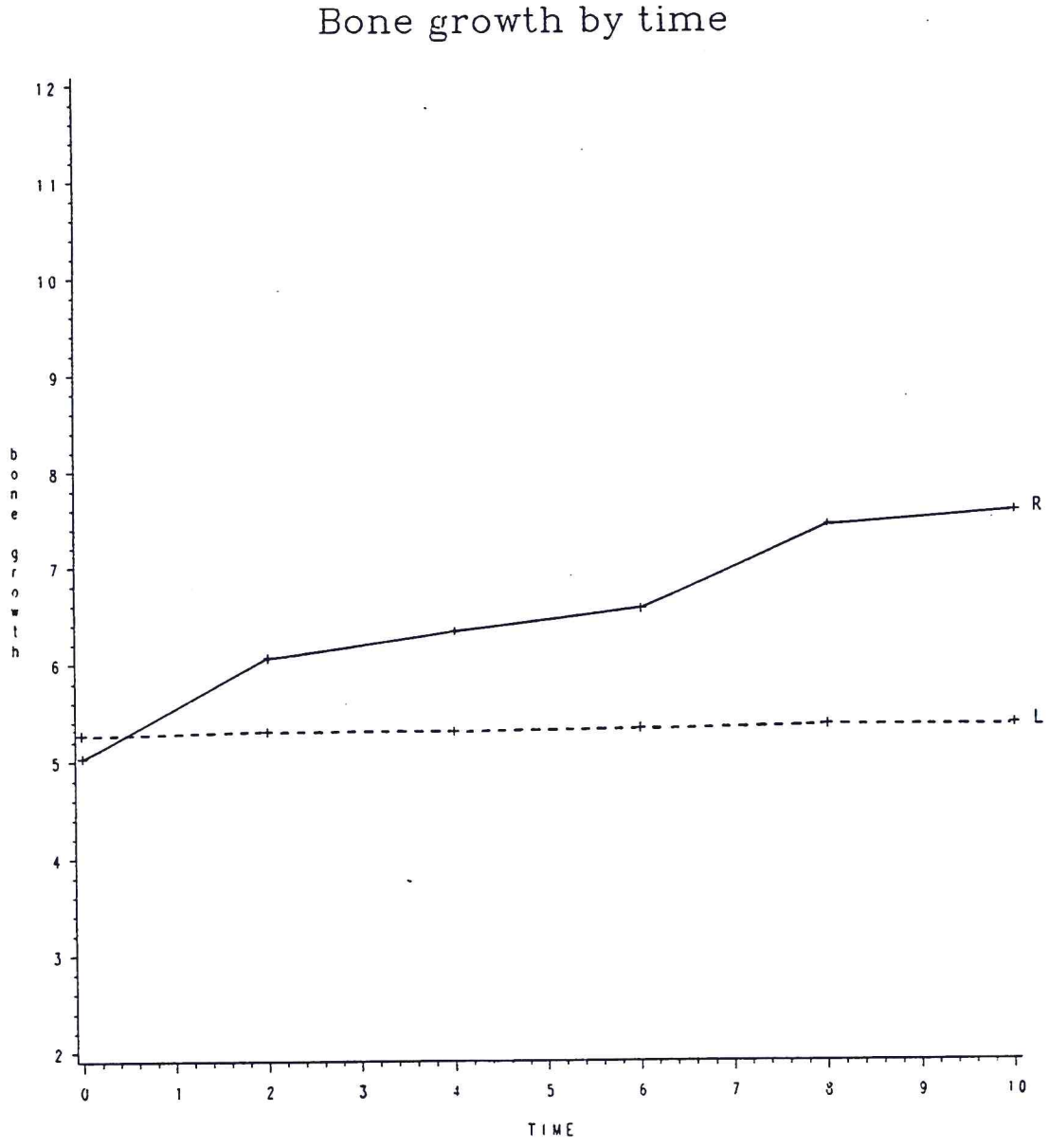


Figure 18. **Graph plotting the distance between the two implants at each time interval for the experimental animal (III1).**

——— Right side

----- Left side

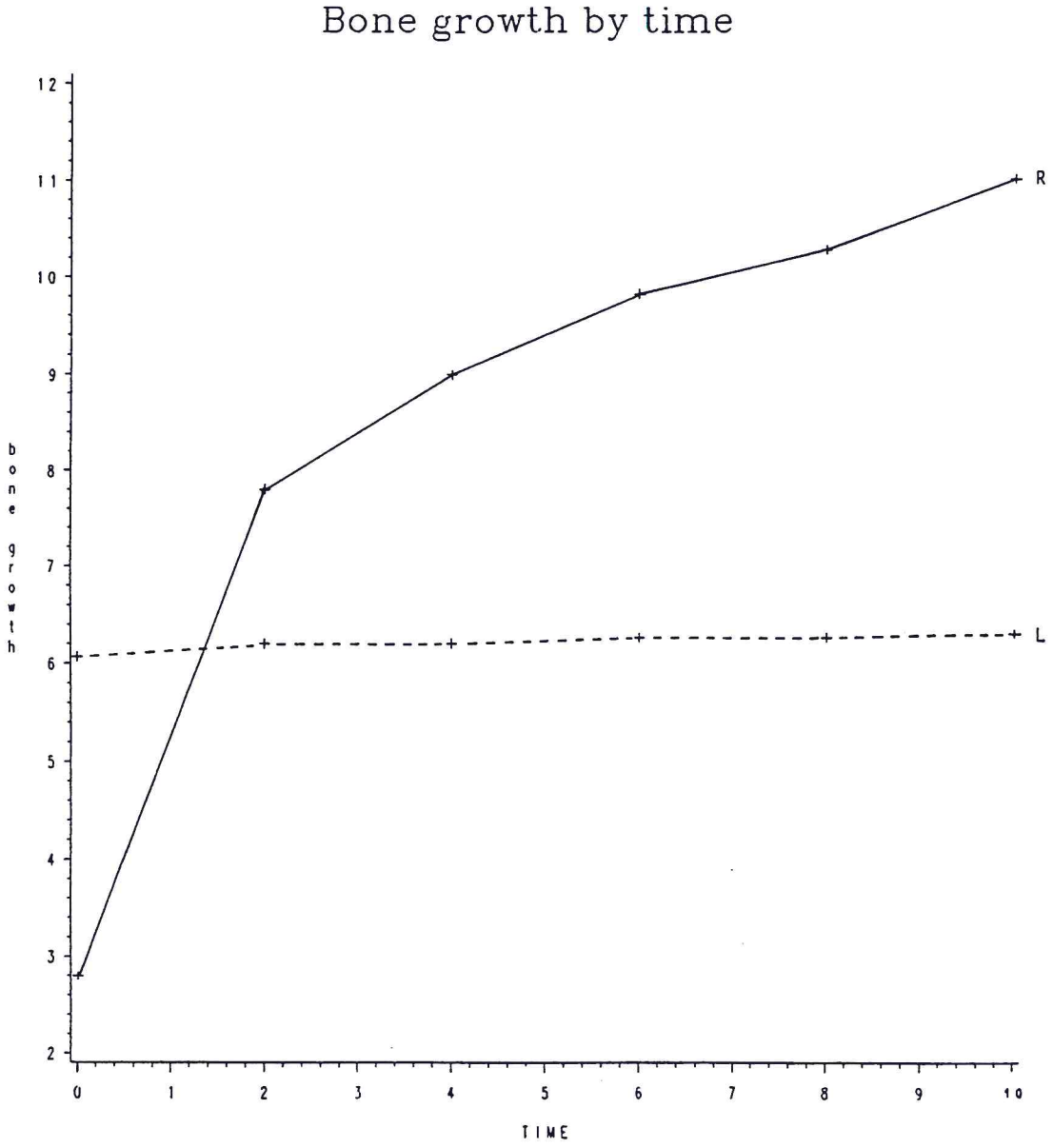


Figure 19. Graph plotting the distance between the two implants at each time interval for the experimental animal (III8).

——— Right side

----- Left side

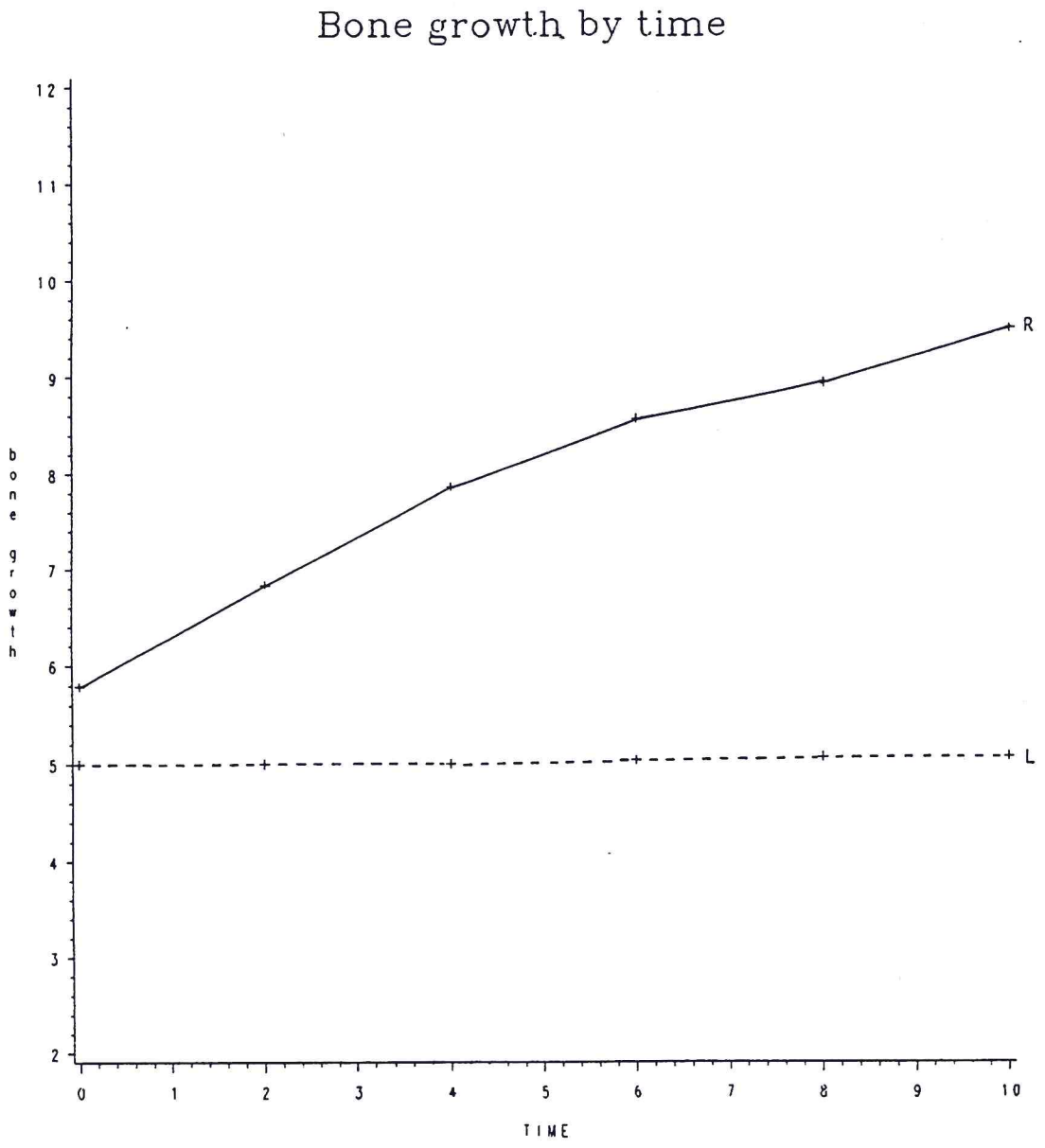


Figure 20. Graph plotting the distance between the two implants at each time interval for the experimental animal (II9).

——— Right side

----- Left side

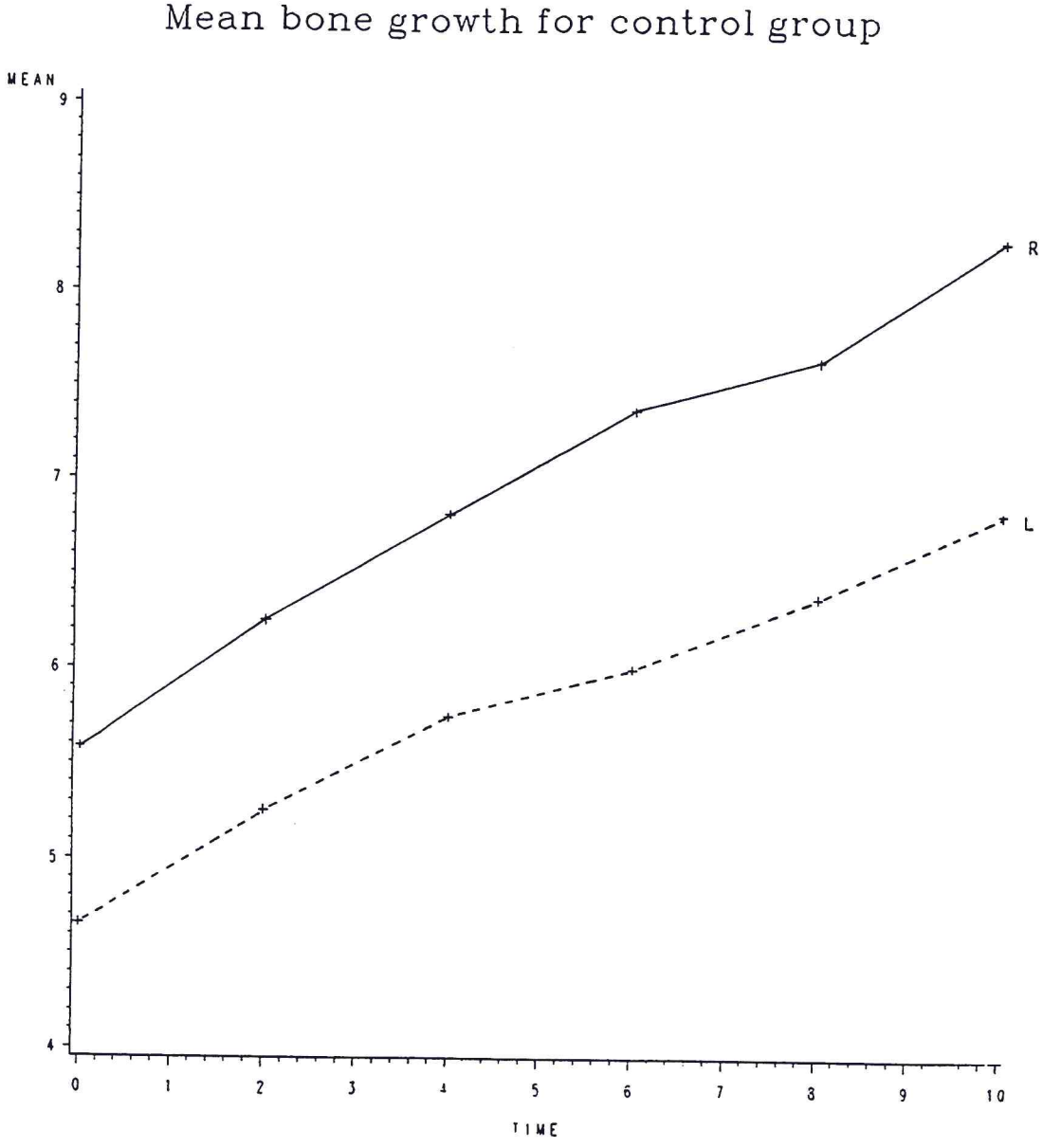


Figure 21. Graph plotting the mean distance between the two implants at each time interval for the control group.

——— Right side

----- Left side

Mean bone growth for the experimental group

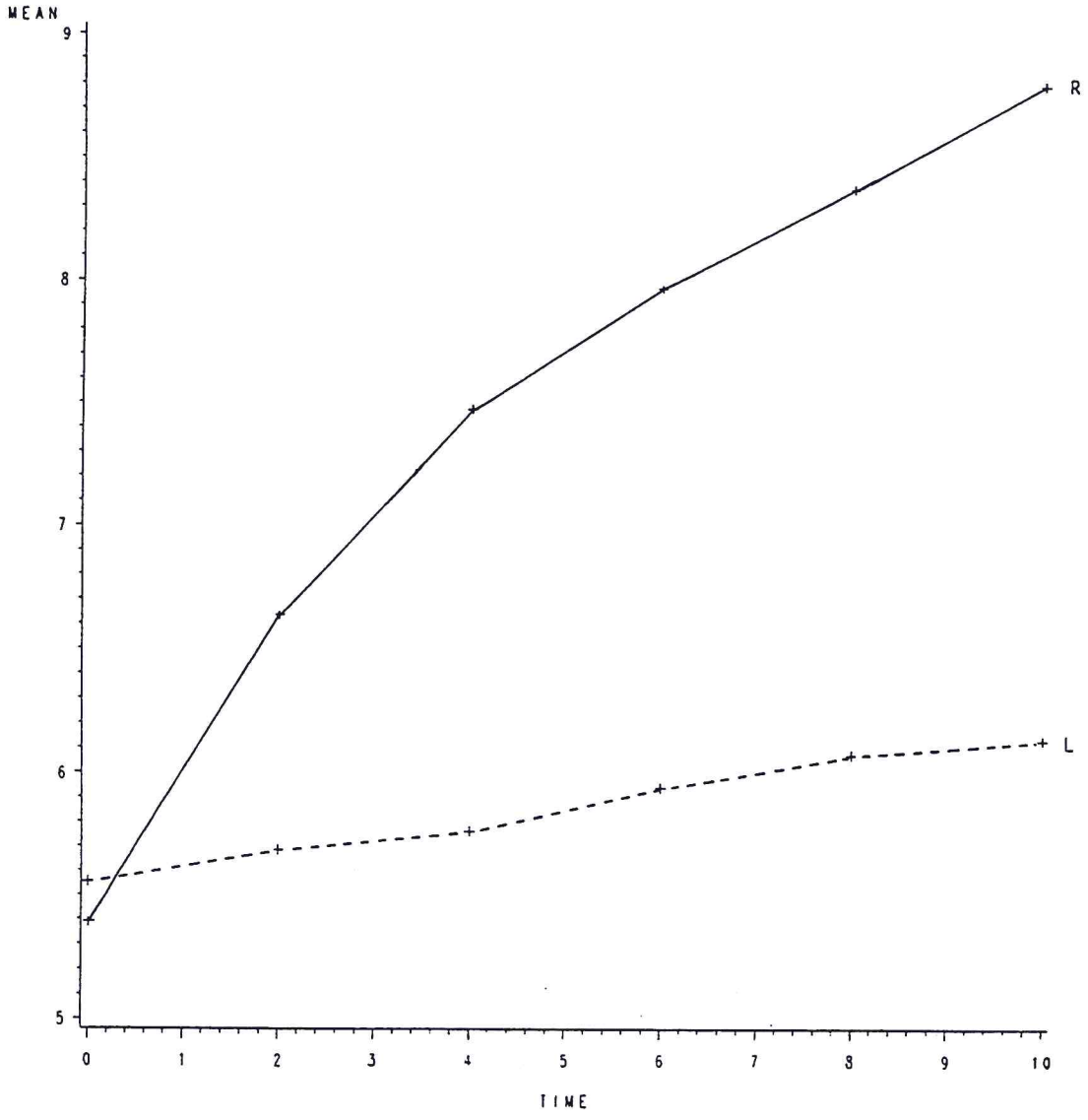
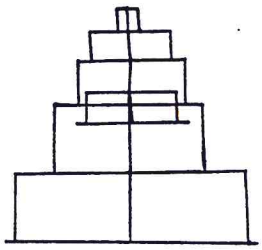
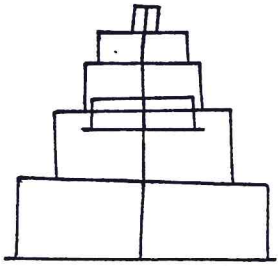


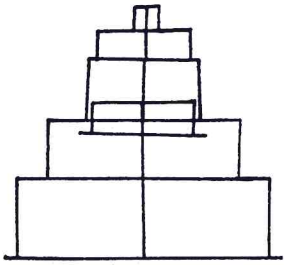
Figure 22. Graph plotting the mean distance between the two implants at each time interval for the experimental group.

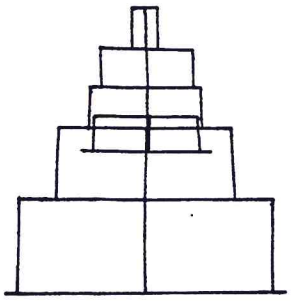
——— Right side

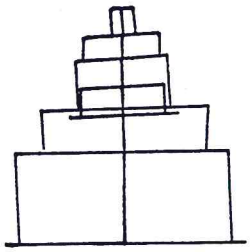
----- Left side

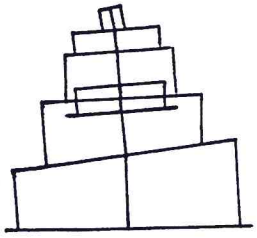


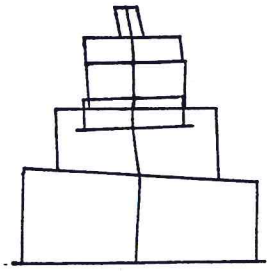


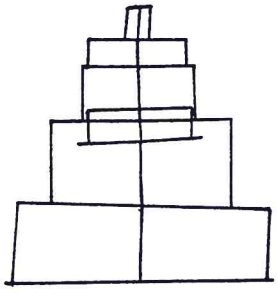


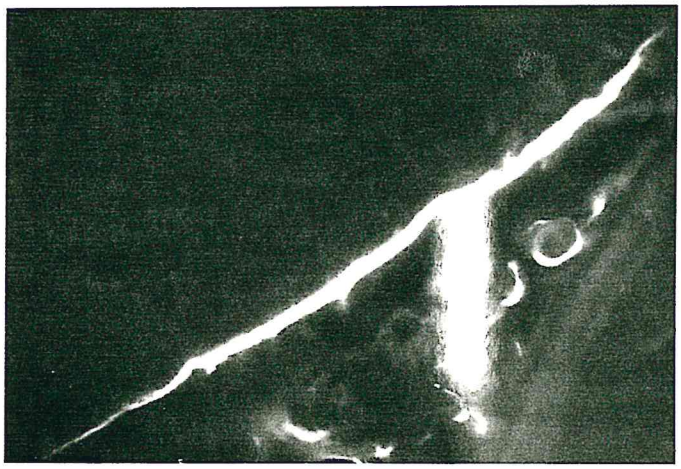


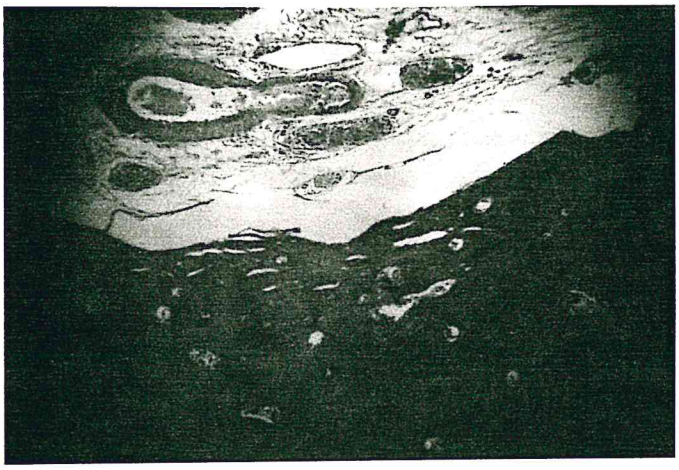


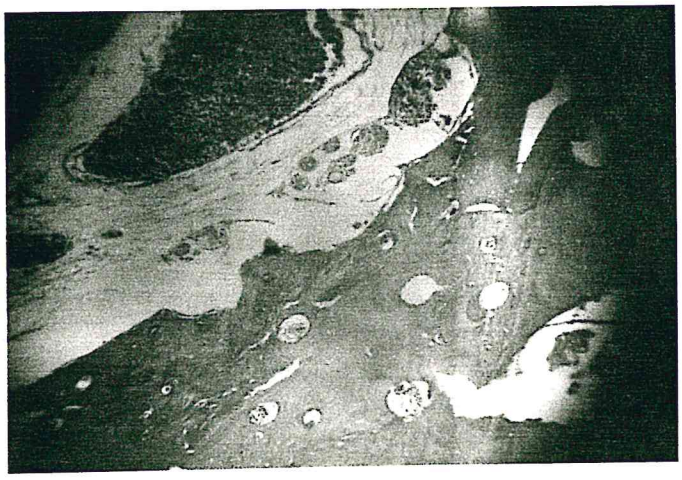


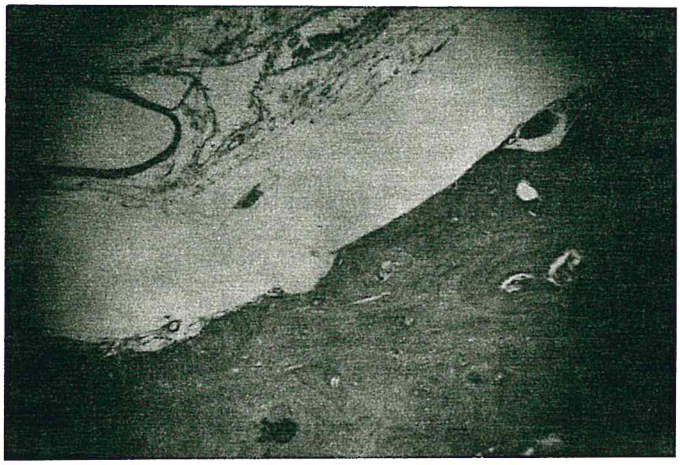


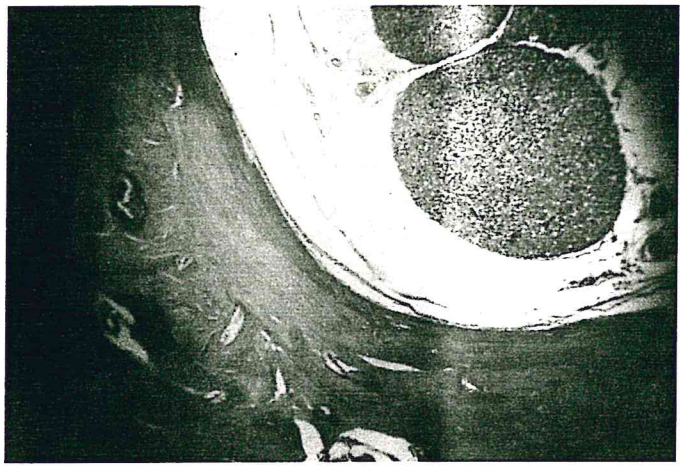


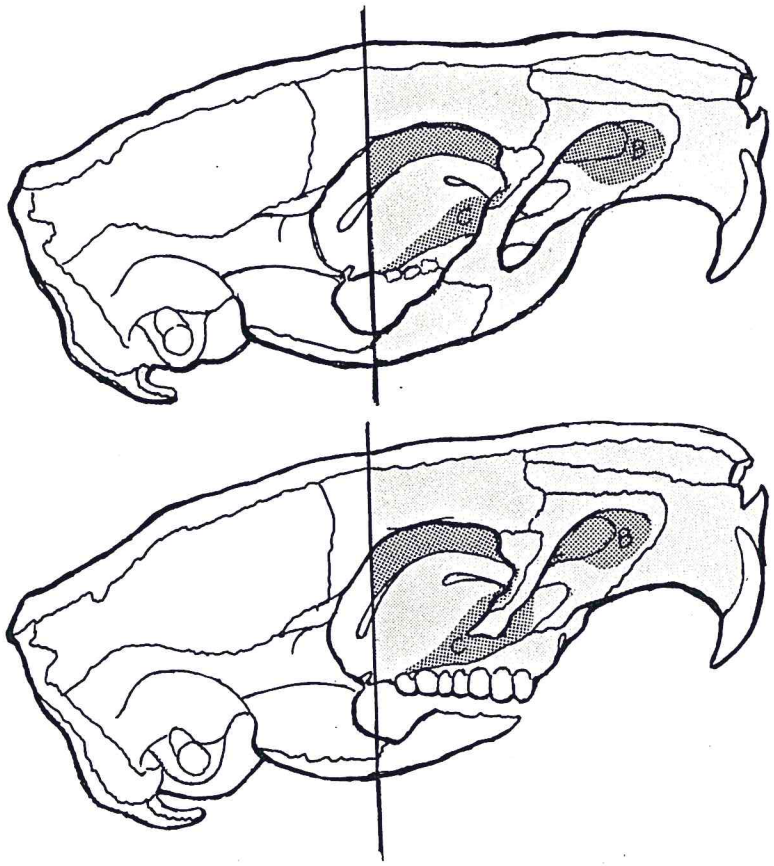


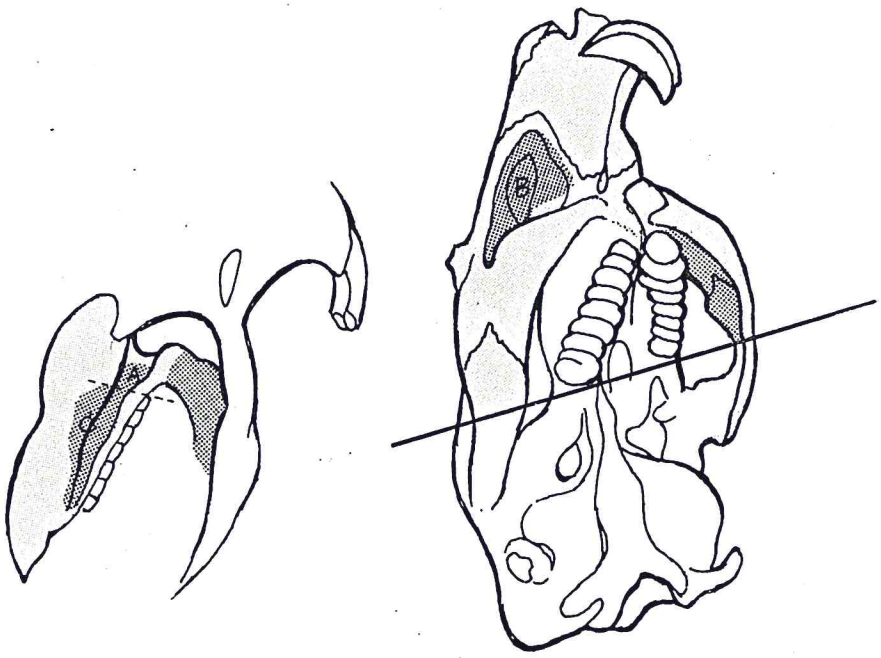


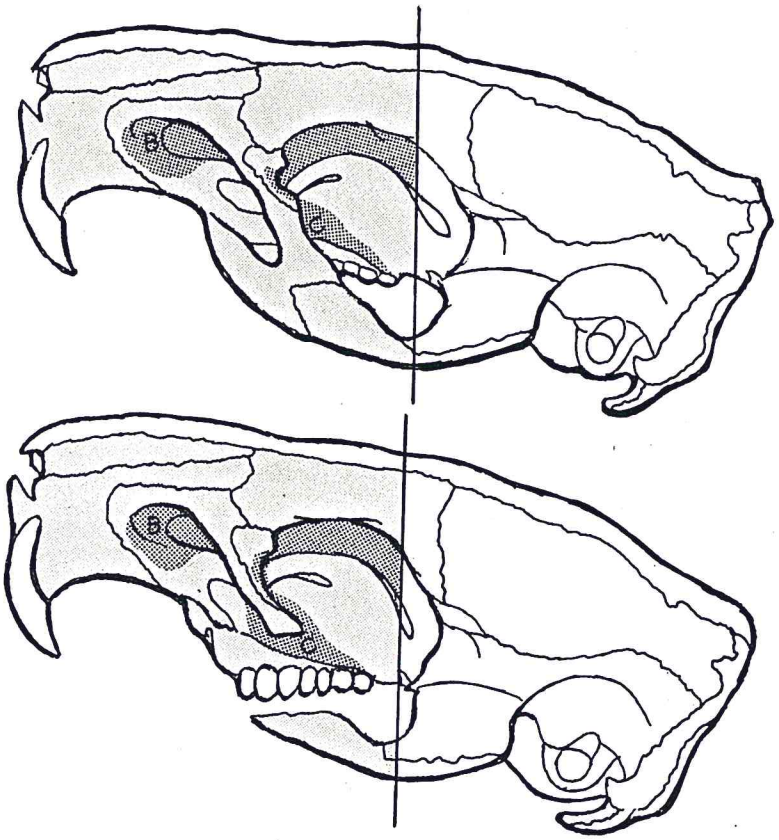


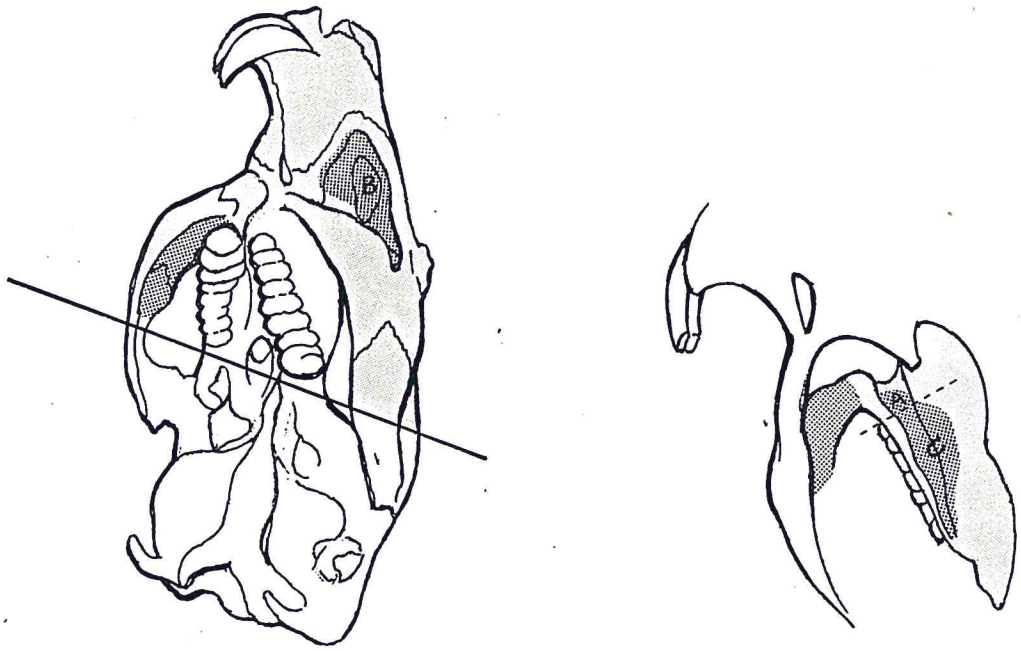


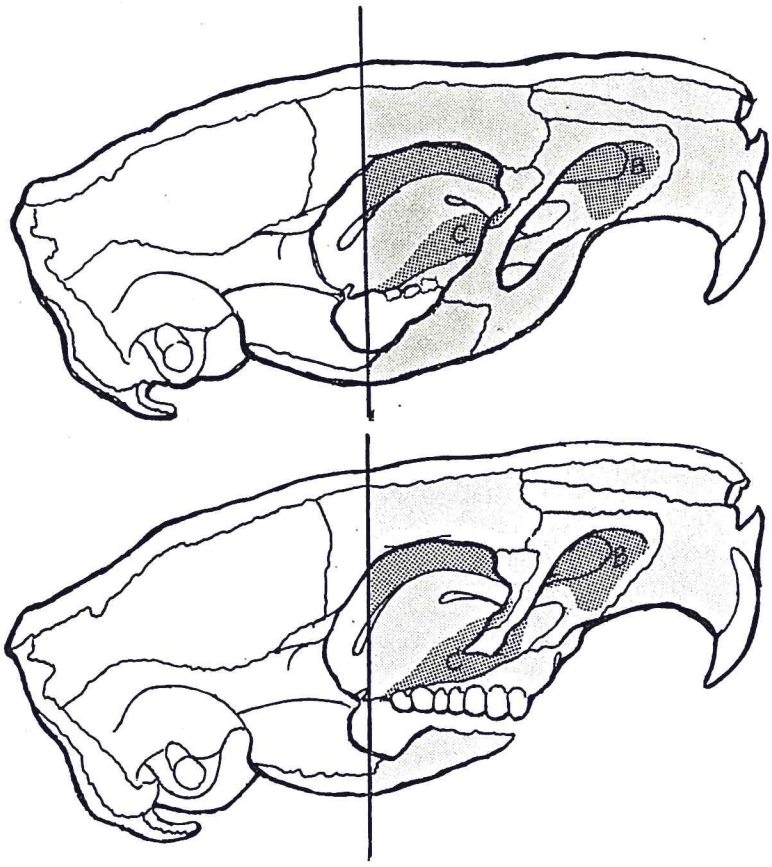


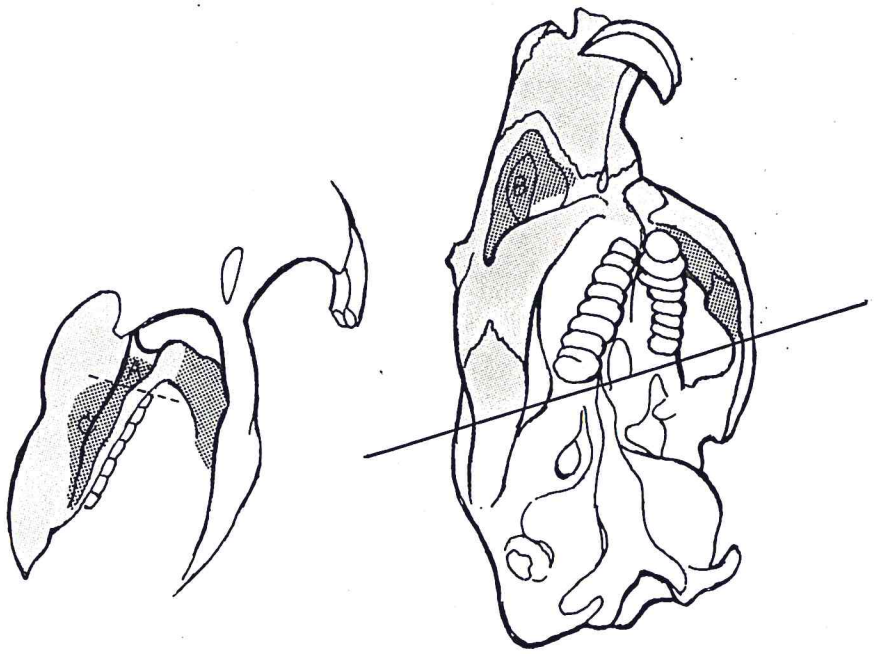

B
B

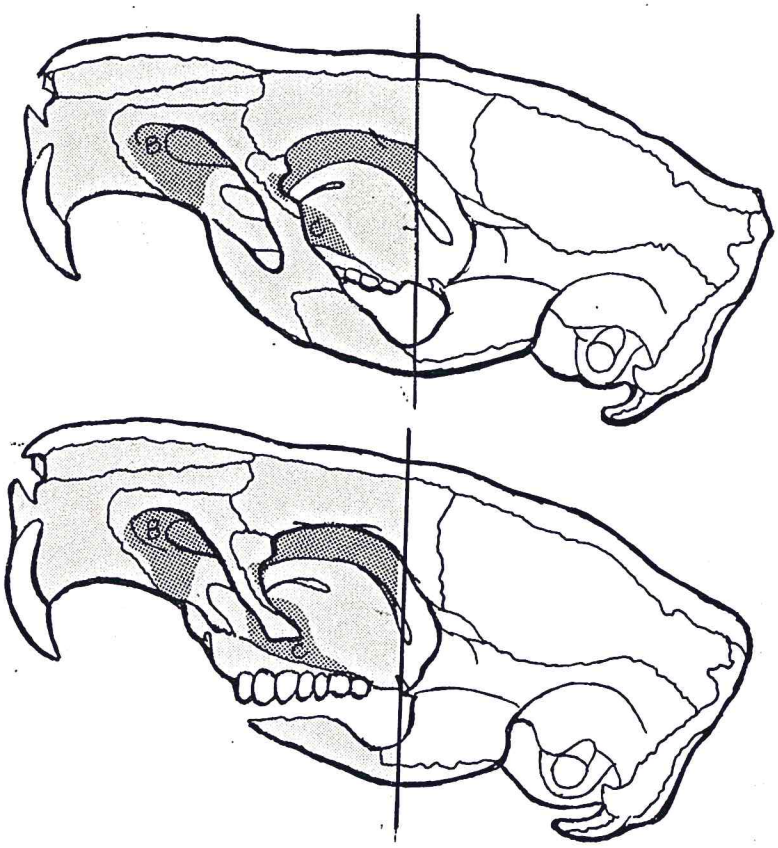





B'
B






BC
B
BC

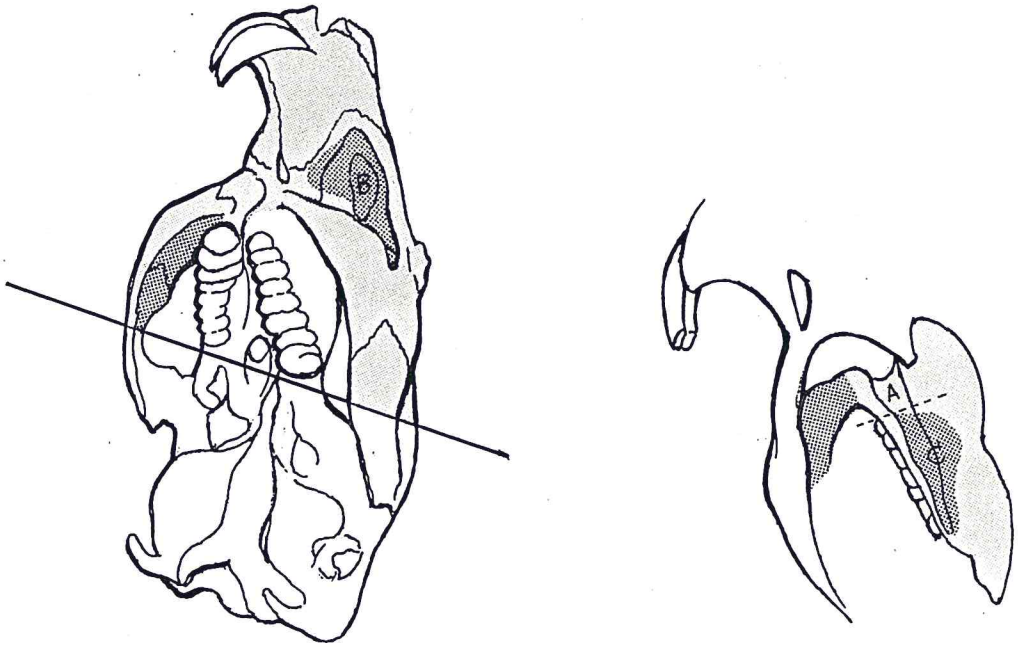

B
A
B

TABLE IV

Summary of location and nature of compensatory remodeling of cortical bone

Location	Control gr.	Experimental gr.	
	Both sides	Fused side	Non-fused side
Area of the snout	-	-	-
Area of the alv.process	-	-	-
Junction of the lower max. process and the max. proper	-	+	-

(+) = Bone formation.

(-) = Bone resorption.

DISCUSSION

Sexual dimorphism was not considered as a parameter for this study. Animals were chosen at random by litter and no attempt was made to select or avoid one sex over the other. The evaluation of sutural growth versus sutural fusion was made by comparing total implant separation of the fused to the non-fused side in the same animal. The R/L ratio was used when comparing among animals in order to avoid differences in individual growth rates.

Increments of implant separation of the fused and non-fused sides of each control animal at each time interval was not equal. This indicated different growth rates between the fused and non-fused sides at each time interval. Also, total implant separation of both sides among different animals were not equal. This can be explained that animals had not yet fully expressed total growth capacity at the time of sacrifice.

In this study, the zygomaxillary suture of the left side was immobilized with cyanoacrylate adhesive that produced a mechanical lock which in turn disturbed the inherent magnitude of tissue proliferation, reduced or eliminated interosseous movement, and eventually caused sutural fusion. These results were also found by Engstrom,et al (1985) in which they note that the

restriction of interosseous movement seemed to be a primary condition for sutural fusion.

Measurement of distances between amalgam implants of the fused sides indicated less sutural growth than the non-fused sides in most animals. Early in the experiment (before the onset of fusion), this resulted from the reduction of osteogenic function at the sutural interface, while the non-fused sides continued growing at a less constrained rate. Later in the experiment when sutural fusion had been obtained, the distance between amalgam implants of the fused sides somehow slightly increased in some time intervals. This could be because of the possible residual growth of the zygomaxillary suture on the medial aspect of the zygomaxillary arch, since only periosteal bone bridge was formed across the suture. The internal portion of the sutural ligament was not synostosed by cyanoacrylate adhesive as stated in the study of Foley (15). Also invloved is the fact that the very young zygomaxillary arch was yet very thin when the surgery was performed. Thus, amalgam implants occupied almost the whole thickness of the bone. Therefore, if residual sutural growth existed, it would be possible for implants to separate after the periosteal bone bridge was formed.

However, compared to control animals, the R/L ratio indicated the same degree of sutural fusion occurred in

all of the experimental animals, eventhough three of them displayed a lesser magnitude of fusion.

Babler,et al (1982) found no effect of periostectomy on bone growth at sutural sites when only the periosteum on both sides of the suture, not across the top, was excised. A similar procedure was performed for removal of the periosteum in this study. Hence, any pattern of subsequent growth in all animals would not be affected by periostectomy but rather due to sutural fusion.

Sutural fusion successfully constrained the growth of the left zygomaxillary suture as demonstrated by a less than 0.5 mm. magnitude of implant separation in most animals. Adjustive compensation from other sutures and regional remodeling was found as a consequence of sutural fusion. The adjustive compensatory growth of the fused side was different from that of the non-fused side. Developmental asymmetry was noticed when comparison between both sides was made. No anatomical asymmetry, however, developed since the developmental asymmetry offset the structural asymmetry. Many possible reasons may also be involved.

1) The zygomaxillary suture may not play a major role in the facial growth of guinea pigs. It does not represent an active growth site. This possibility seems untenable since significant sutural growth was observed in the control animals.

This possibility, however, was supported by the study of Watanabe,et al (1957) in which they transplanted the zygomaxillary suture from one side of the face of the 3-week-old guinea pigs to the subcutaneous tissue of the right of the abdomen of the same animal. Analysis of the skull showed no significant asymmetry or any deformity.

2) There may have been compensatory growth of bone in response to altered muscle function. Sutural and cortical bone remodeling have been shown to be altered by a changed jaw muscle activity (Koskinen, 1977; Engstrom, et al, 1985).

Adaptation of jaw muscle function following sutural fusion might have occured in some areas in such a way that periosteal bone remodeling would compensate for a lack of growth at the fused suture. Therefore symmetric skulls resulted. Evaluation of jaw muscle function was not made in this study.

3) Brain growth

Brain growth plays major role in thrusting the nasomaxillary complex forward through the growth of cranial base and cranial vault. The frontonasal sutures have an elongate transverse orientation. The primary and secondary displacement of the frontal bones might greatly affect the displacement of the nasomaxillary complex. Thus, it might mask the effect

of the lack of primary displacement of the left maxilla caused by premature fusion of the zygomaxillary suture.

Normally, facial sutures fuse later than cranial sutures. If the facial suture is intentionally fused after the fusion of cranial sutures, there may be a greater tendency for a resultant deformity. However, compensation at other facial sutures might occur as well.

4) Increase in growth of adjacent sutures on the fused side might compensate for the lack of growth at the fused suture and result in symmetric skulls. This was not evaluated in the present study since no implants were utilized in other sutures.

The geometric pattern from T0 to T10 in the control group seemed to be nearly symmetrical even though slight bilateral variations are seen. It is concluded that right/left side growth was essentially uniform.

In contrast to the control group, the experimental animals demonstrated active compensatory growth by showing distinct right/left differences in geometric pattern from T0 to T10. Interestingly, the geometric pattern began changing structurally from T2 to T8 but then , later, returned to virtual symmetric pattern in T10. The details of the patterns varied somewhat among animals. However, they all indicated compensatory growth

from both growth sites: other (non-fused) sutures and regional bone remodeling.

In the present study , it was not possible to locate precisely the compensatory growth sites during each specific time interval. With the aid of implants in additional sutural areas, we would have been able to locate whatever compensatory sutural sites were involved. Serial histological sections obtained from the animals at each time interval, however, did help to identify some areas of compensatory remodeling. This showed that compensatory responses occurred within areas c, d, e, and f, which represented the anterior 1/2 of the maxilla proper, the upper and lower maxillary processes, and the zygomaxillary arch, although in variable combinations among the different animals.

From the histological evaluation, the distribution of depository and resorptive areas in ground sections was found to coincide with that seen in the decalcified sections, even though not all vital dye lines appeared.

Engstrom,et al (1985) proposed that a compensatory periosteal growth occured in the region of deficient sutural growth. In the present study, a significant area of compensatory remodeling of cortical bone was found in the maxilla proper of the fused side close to the junction of the lower process and the maxilla proper (area A in Figure 41). This area was not very far from

the fused suture. The nature of the compensation of the fused side was by periosteal bone formation. In contrast to the fused side, the non-fused side demonstrated periosteal bone resorption in the corresponding area (area A in Figure 39), which was also the same pattern as seen on both the fused and non-fused sides in the control animals. Some degree of response was also found in the maxillae in the area of the snout and the alveolar process (area B and C), which were more distant from the fused suture. Different sizes of resorptive areas were seen when comparing among the left sides of experimental group, the right sides of the experimental group, and the control group.

Virchow (1852) found that normal growth was inhibited in a direction perpendicular to a prematurely fused suture, and that compensatory growth occured in a direction parallel to the prematurely fused suture. The zygomaxillary suture is V-shaped with the apex of V pointing anteriorly. If the compensatory growth occured parallel to the fused suture, the V-shape of the suture might complicate the direction and area of compensation.

Variation in the pattern of bone remodeling in the areas surrounding tooth buds related to different stages of development and eruption of individual teeth. The extent to which this might or might not be associated with responses to sutural fusion is not known.

Although possible compensatory responses by adjacent sutures were not evaluated in this study, nevertheless, symmetric skulls resulted from some combinations of compensatory remodeling growth which likely indicated sutures as well as other regional areas in the maxillary complex. The resultant gross symmetry of the skull indicates that a compensatory mechanism had fully expressed its adjustive capability to overcome the anatomic effect of an experimental sutural fusion. However, in normal growth and development asymmetries often ocurr. Therefore, why does not the process of compensation help adjust the affected skull in such cases during growth and development?. A possibility is that a condition that can affect the skull (premature fusion of a suture, injury, etc.) may also disturb the process of compensation to some degree and thus lead to an impairment of compensatory mechanism.

The actual magnitude of bone formation and resorption in each animal could not be determined with precision in this study. Different planes of serial sections for each region throughout the nasomaxillary complex would be needed to obtain such information in conjunction with vital dyes and increased distributions of radiographic markers.

In the present study, the adjustive compensation of other facial sutures was found subsequent to

experimental fusion of a facial suture. It is suggested that a sequel study now be carried out in which the nature of response of facial sutures to premature fusion of basicranial sutures be determined.

CONCLUSIONS

Unilateral premature fusion of the zygomaxillary suture promoted by means of cyanoacrylate adhesive successfully inhibited the primary displacement of the maxilla and the zygomatic bone at the fused site. Compensatory responsive remodeling at adjacent sutures and in certain regions of the nasomaxillary complex was observed. Developmental asymmetry was found when the fused side was compared to the non-fused side. No resultant anatomical asymmetry was detected. Adaptive compensation involving periosteal bone deposition occurred at the junction of the lower maxillary process and the maxilla proper on the fused side (area A), and periosteal bone resorption in corresponding areas on the non-fused side served to offset the developmental asymmetry thus leading to a compensatory lack of gross anatomical asymmetry.

In the region of the snout, the resorptive area (area B) on the fused side was slightly larger than that on the non-fused side and on both sides of the control group. In the area of the alveolar process, the resorptive area (area C) on the fused side was slightly smaller than that on the non-fused side and on both sides of the control group.

These patterns of compensatory remodeling and

adaptations by adjacent sutures adjusted the anatomical form of the skull during the subsequent development with eventual craniofacial symmetry resulting.

BIBLIOGRAPHY

Alhopouro,S., Ranta,R., and Ritsila,V.(1973): Growth of the rabbit snout after bony fusion of the frontonasal suture achieved by means of a free periosteal transplant. Proc. Fin. Dent. Soc., 69:166-167.

Alhopouro,S., Ranta,R., and Ritsila,V.(1973): Growth of the facial skeleton in guinea pigs after experimental fusion of the premaxillo-maxillary suture. Proc. Fin. Dent. Soc., 69:263-265.

Babler,W.J., and Persing,J.A.(1981): Alteration in cranial suture growth associated with premature closure of the coronal suture in rabbits. Anat. Rec., 199:14A.

Babler,W.J., and Persing,J.A.(1982):Experimental alteration of the cranial suture growth: Effects on the neurocranium,basicranium,and midface. The international conference on factors and mechanisms influencing bone growth,edited by A. Dixon and B. Sarnat. Alan Liss, New York, 333-345.

Babler,W.J., Persing,J.A., Persson,K.M., Winn,H.R., Jane,J.A., and Roeheaver,G.T.(1982): Skull growth after coronal suturectomy, periostectomy,and dural transection. J.Neurosurg., 56:529-535.

Cohen,M.Jr.,(1986): The etiology of craniosynostosis. From:Craniosynostosis:diagnosis, evaluation, and management. Edited by Cohen,M.Jr., Ravan Press, New York., 59-79.

Engstrom,C., Kiliaridis,S., and Thilander,B.(1985): Facial suture synostosis related to alter craniofacial bone remodeling by biomechanical forces and metabolic factors. Normal and abnormal bone growth: basic and clinical research. Edited by A. Dixon and B. Sarnat. Alan R. Liss, New York, 379-391.

Engstrom,B., and Thilander,B.(1985): Premature facial synostosis:The influence of biomechanical factors in normal and hypocalcaemic rats. Eur. J. of Orthod., 7(85):35-47.

Enlow,D.H.(1962): A study of the post-natal growth and remodeling of bone. Am.J.Anat., 110:79-101.

Enlow,D.H., and Bang,S.(1965):Growth and remodeling of the human maxilla. Am.J.Orthod., 51(6):446-464.

Enlow,D.H., and Hunter,W.S.(1966): A differential analysis of sutural and remodeling growth in the human face. Am.J.Orthod., 52(11):823-830.

Enlow,D.H.(1966): A morphogenetic analysis of facial growth. Am.J.Orthod., 52(4):283-299.

Enlow,D.H.(1982): Handbook of Facial Growth. W.B. Saunders, Philadelphia.

Enlow,D.H.(1986): Normal craniofacial growth. From: Craniosynostosis:diagnosis, evaluation, and management. Edited by M.Cohen Jr., Ravan Press, New York, 131-156.

Foley,W., and Kokich,V.G.(1980): The effects of the mechanical immobilization on sutural gevelopment in the growing rabbits. J. Neurosurg., 53:794-801.

Giblin,N., and Alley,A.(1944):Studies in skull growth. Coronal suture fixation. Anat.Rec., 88:143-153.

Goldberg,G., Enlow,D.H., Whitaker,L.A., Zin,J.E., and Kurihara,S.(1981): Some characteristics in several craniofacial syndromes. J.Oral Surg., 39:489-498.

Graham,J.M., Badura,R.J., and Smith,D.W.(1979): Metopic craniosynostosis as a consequence of fetal head constraint:two interesting experiments of nature. Pediatrics, 65(5):1000-1002.

Graham,J.M., Badura,R.J., and Smith,D.W.(1979): Sagittal craniosynostosis:Fetal head constraint as one possible cause. J.of Pediatrics, 95(5):747-750.

Graham,J.M., Badura,R.J., and Smith,D.W.(1980): Coronal craniosynostosis:Fetal head constraint as one possible cause. Pediatrics, 65:995-999.

Grigis,F.G.and Pritchard,J.J.(1955): Morphologic status of cranial sutures. J.Anat., 89:577.

Grigis,F.G.and Pritchard,J.J.(1958): Effect of skull damage on the development of sutural patterns in the rat. J.Anat., 92:39.

Herring,S.W.(1972): Sutures- a tool in functional cranial analysis. Acta.Anat., 83:222-247.

Hellquist,R.(1972): Facial skeletal growth after periosteal resection. An osteometric, roentgenographic, and histologic study in the rabbit and guinea pig., Scand.J.Plast.Reconstr.Surg., 10(suppl 10):1-98.

Isotupa,K., Koski,K., and Makinen,L.(1965):Changing in architecture of growing cranial bones at sutures as revealed by vital staining with alizarin red in the rabbits. Am.J.Phy.Anthropol., 23:19-22.

Jackson,G., Kokich,V.G., and Shapiro,P.A.(1979): Experimental and post-experimental response to anteriorly directed extraoral force in young Macaca nemestrina. Am.J.Ortho., 75(3):318-333.

Kokich,V.G.(1976): Age changes in human frontozygomatic suture from 20 to 95 years. Am.J.Orthod. , 69:411-430.

Kokich,V.G.(1986): Biology of sutures. From: Craniosynostosis:diagnosis, evaluation, and management Edited by Cohen,M.Jr., Ravan Press, New York., 81-103.

Koskinen,L., Isotupa,K., and Koski,K.(1976): A note on craniofacial sutural growth. Am.J.Phys.Antropol., 45:511-516.

Koskinen,L.(1977): Adaptive sutures. Changes after unilateral masticatory muscle resection in rats. A microscopic study. Proc.Fin.Dent.Soc., 73(suppl 10-11): 1-79.

Koskinen-Moffett,L.K., Moffett,B.C.Jr., and Graham,J.M.Jr.(1982): Cranial synostosis and intra-uterine compression. A developmental study of human sutures. The international conference on factors and mechanisms influencing bone growth.Edited by A. Dixon and B. Sarnat. Alan Liss New York, 365-378.

Kreiborg,S., and Bjork,A.(1981): Craniofacial asymmetry of a dry skull with plagiocephaly. Eur.J.Orthod., 3:195-203.

Kreiborg,S.(1986): Postnatal growth and development of the craniofacial complex in premature Craniosynostosis. From:Craniosynostosis:diagnosis, evaluation, and management. Edited by Cohen,M.Jr., Ravan Press, New York., 157-189.

Latham,R.A., and Burstone,W.R.(1966): The postnatal pattern of growth at the sutures of the human skull. Dent.Practit., 17:61-67.

Linge,L.(1972): Tissue reaction incedent to widening of facial sutures. Trans.Europ.Orthod.Soc., 487-497.

Linge,L.(1976): Tissue reactions in facial sutures subsequent to external mechanical influences. In:Factors

affecting the growth of the midface. Edited by J.A. McNamara Jr., University of Michigan, Center for Human Growth and Development., 251-275.

Mabbutt,L.W., and Kokich,V.G.(1979): Calvarial and sutural re-development following craniectimy in neonatal rabbits. J. Anat., 129:413-422.

Miyawaki,S., and Forbes,D.P.(1987): The morphologic and biomechanical effects of tensile force application to the interparietal suture of the Sprague-Dawley rat. Am.J. of orthod. Dentofac. Orthoped., 92(2): 123-133.

Moss,M.L.(1957): Experimental alteration of the sutural area morphology. Anat.Rec., 127:569-590.

Moss,M.L.(1959): The pathogenesis of premature cranial synostosis in man. Acta.Anat., 37: 351-370.

Moss,M.L.(1960) inhibition and stimulation of sutural fusion in the rat calvaria. Anat.Rec., 136:457-468.

Moss,M.L., and Salentijn,L.(1969): The primary role of functional matrices in facial growth. Am. J. Orthod., 55(6):556-577.

Moffett,B.C.(1971): Remodeling of craniofacial articulations by various orthodontic appliances in rhesus monkey. Trans.Eur.Orthod.Soc, 207-216.

Nappen,D.L., and Kokich,V.G.(1983): Experimental craniosynostosis in growing rabbits. The role of the periosteum. J.Neurosurg., 58:101-108.

Persson,M.(1973): Structure and growth of facial sutures: Histologic, microangiographic, and autoradiographic studies in rat and a histologic study in man. Odot.Rev., 24, Suppl.26.

Persson,K.M., Roy,W.A., Persing,J.A., Rodeheaver, G.T., and Winn,H.R.(1979): Craniofacial growth following experimental craniosynostosis and craniectomy in rabbits J.Neurosung., 50:187-197.

Pritchard,J.J., Scott,J.H., and Grigis,F.G.(1956): Structure and development of cranial and facial sutures. J.Anat., 90:73-86.

Sarnat,B.G.(1958): Palatal and facial growth in Macaca rhesus monkeys with surgically produced palatal clefts. Plast.& Reconstruct.Surg., 22:29-41.

Selman,A.J., and Sarnat,B.G.(1957): Growth of the rabbit snout after extirpation of the frontonasal suture:A gross and serial roentgenographic study by means of metallic implants. Am.J.Anat., 101(2):273-293.

Smith ,H.G., and McKeown,M.(1974): Experimental alteration of the coronal suture area: A histological and quantitative microscopic assessment. J.Anat., 118(3):543-559.

Stenstrom,S.J., and Thilander,B.L.(1967): Facial skeleton growth after bone grafting to surgically created premaxillo-maxillary suture defects:An experimental study on guinea pig. Plast.& Reconstruct. Surg., 40(1):1-12.

Ten Cate,A.R., Freeman,E., and Dickinson,J.B.(1977): Sutural development:structure an its response to rapid expansion. Am.J. Orthod., 71:622-636.

Thor,O.(1964): Compression of facial sutures by external pressure (Miwaukee Brace) demonstrated by the implant method. Trans.Eur.Orthod.Soc., pages 221-232.

Tuenge,R.H., and Elder,J.R.(1974): Post-treatment changes following extraoral high-pull traction to the maxilla of Macaca mulatta. Am.J.Orthod., 66(6):618-644.

Van Limborgh,J.(1982): Factors controlling skeletal morphogenesis. In:Factors and mechanisms influencing bone growth. Edited by A. Dixon and B. Sarnat., Alan Liss Press New York., pages 1-17.

Watanabe,M., Laskin,D.M., and Brodie,A.G.(1957): The effects of autotransplantation on growth of zygomaxillary suture. Am.J.Anat., 100:319-335.