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Growth pattern of the rabbit nasal bone region

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ABSTRACT

Two radiopaque implants were inserted into each left and right nasal bone in five female rabbits. Ventrodorsal cephalometric radiographs were taken at 6 and 16 weeks of age. From these radiographs, separate tracings were made on matte acetate paper of the left and right nasal bone regions including the radiopaque implants. The markers of the 16 week tracing were superposed on the markers of the 6 week tracing. The difference in the two established outlines represented the changes in size and shape in two dimensions that had occurred during the 10 week period. Our purpose was to determine the relative growth activity at several borders. The mean increase was about 6.79 mm at the proximal (posterior) border, 6.19 mm at the distal (anterior) border, 2.73 mm at the lateral border, 1.22 mm at the medial border. Thus, growth at the proximal and the distal borders was about the same and about twice that of the lateral border and about 5 times that of the medial border.

INTRODUCTION AND PURPOSE

Although bone is hard, semirigid, and supporting, it is a dynamic, sensitive, everchanging tissue. Any interference that affects the growth of bone(s) will alter the orderly progression of development and result in some type of deformity in any or all three planes: height, width and depth. The results of injury are determined by the site, severity, duration, and type of noxious agent and also significantly by the time of occurrence. The end result at any given time records the effects of all the vicissitudes.

Some principles of the biology of bone as they apply to the nasal region are central to this presentation. The three modes of postnatal growth of bone are (1) cartilaginous, (2) sutural, and (3) appositional and resorptive (remodeling). An appreciation of the differential responses and interrelationships of these processes is important to the recognition of both normal and abnormal conditions.

The following generalization may be made about skeletal growth. In the young, skeletal mass increases because cartilaginous (endochondral, nasal septum) and

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sutural growth are active and apposition is greater than resorption (positive growth). In the adult, skeletal mass is constant because apposition and resorption, although active, are in equilibrium, whereas cartilaginous and sutural growth have ceased (neutral growth). In old age, skeletal mass decreases because resorption is more active than apposition (negative growth).

Significant reports in regard to the growth of bone appeared in the literature two hundred years ago (Hunter, 1778). Many questions, however, are still unanswered. Any determination of growth of bone concerns itself with one or more of the following questions: What are the sites? What is the amount? What is the rate? Does it vary? When? What is the direction? What are the changes in proportion? What factors are influential? A number of different approaches have been employed to study the growth of bones (Sarnat, 1963). Each, however, has its limitations. One method may yield information about the sites of growth, another about the rate, while still another about direction. An informative procedure is the use of implants alone or in combination with serial radiography (Table 1) (Sarnat, 1968). By this method Gans and Sarnat (1951) and Selman and Sarnat (1955) studied sutural growth and Robinson and Sarnat (1955) determined the growth pattern (apposition and resorption as well as cartilaginous) of a single bone, the mandible.

Growth of the nasal bone region occurs in two principal ways, namely: on its various surfaces and at sutures (frontonasal and premaxillary-maxillary). The purpose of this study was to assay the growth pattern of the rabbit nasal bone region by determining relative amounts of growth at several borders. This was done by a combined method of serial gross and radiographic measurements between radiopaque implants inserted within a single bone, the nasal (Sarnat, 1978). There were several advantages: 1) it was a serial study, 2) there were permanent records, 3) the implants served as stable reference markers from which accurate information could be obtained as to sites, relative amounts and directions of growth, and 4) the radiographic changes that occurred from one period to another could be determined without killing or re-operating the animal. We have found no such report.

MATERIAL AND METHODS

Animals. Five growing female six-week-old New Zealand albino rabbits were used. These animals were selected primarily because of their rapid growth and a snout which lent itself to ready implantation and accurate serial radiography. A disadvantage, however, was the size and complexity of the pinna, which made difficult the insertion of ear posts for serial radiography. The animals were fed *ad libitum* on standard rabbit ration.

Table 1. A brief historical review of implant markers used in the longitudinal study of the growth of bones*

investigator	year	material used	bones studied	animal
gross (direct) studies				
Hales	1727	holes	tibia	chicken
Duhamel	1743	silver stylets	long bone	pigeon, dog
Hunter	1770	lead shot	tibia tarso-metatarsal	pig chicken
Humphry	1864	wires	mandible	pig
Gudden	1874	holes	parietal, frontal	rabbit
Wolff	1885	metal	frontal, nasal	rabbit
Giblin and Alley	1942	wax	parietal, frontal, etc.	dog
Roy and Sarnat	1956	stainless steel wire, black silk suture	rib	rabbit
gross (direct) and/or serial radiographic (indirect) studies				
Dubreuil	1913	metal	tibia	rabbit
Gatewood and Mullen	1927	shot	femur	rabbit
Troitzky	1932	silver wires	skull	dog
Levine	1948	dental silver amalgam	frontal, nasal	rabbit
Gans and Sarnat	1951	dental silver amalgam	various facial	monkey
Sissons	1953	metal	femur	rabbit
Selman and Sarnat	1953	dental silver amalgam	frontal, nasal	rabbit
Robinson and Sarnat	1955	dental silver amalgam	mandible	pig
Björk	1955	tantalum	various facial	human
Elgoyhen, et al.	1972	tantalum	various facial	monkey
Sarnat and Selman**	1978	dental silver amalgam	nasal	rabbit

* Modified after Sarnat.
** This report

Anesthesia. The rabbits were anesthetized by injection into the marginal ear vein of a 1% solution of pentobarbital sodium in distilled water (40 mg/kg body weight). To each dose 0.25 mg of atropine sulfate was added.

Metallic implants. The anesthetized animal was strapped prone on the operating board. The skin of the dorsum of the snout was clipped free of hair, cleansed with 70% ethyl alcohol, and isolated with sterile towels. An aseptic technique was observed throughout the surgical procedure. The skin and subcutaneous tissues were incised longitudinally in the midline. After the wound margins were retracted, the periosteum was incised, elevated, retracted, and the nasal bones were exposed.

A dental bur, mounted in a handpiece, was used to prepare two cavities, with an undercut at the base to facilitate retention in the cortical plate of each nasal bone. Into these cavities, dental amalgam was packed (Figure 1). When prepared, this material was pliable and could be readily handled and inserted into a newly created bone cavity. In addition, the amalgam expanded slightly after setting which aided retention. Two other important characteristics of this material were its tolerance by the local tissue and its radiopacity. An indentation was made in the center of each amalgam implant with the point of a caliper and for each pair of implants within the same bone the distance between these centers ($A_1 B_1$, $C_1 D_1$) was recorded to the nearest 0.1 mm. A direct measurement was also made between each implant and the frontonasal suture at the point S on the suture where a straight line crossed between the corresponding nasal implant (Figure 1). The soft tissues were then replaced and approximated with 4–0 black silk sutures. The animals were killed 70 days later by pentobarbital sodium injected intravenously and the measurements repeated. The heads were severed and the soft tissues dissected.

Serial cephalometric radiography. A special headholder had been designed and constructed to obtain comparable serial cephalometric radiographs (Selman and Sarnat, 1953). An ear post was placed in each external auditory meatus and an

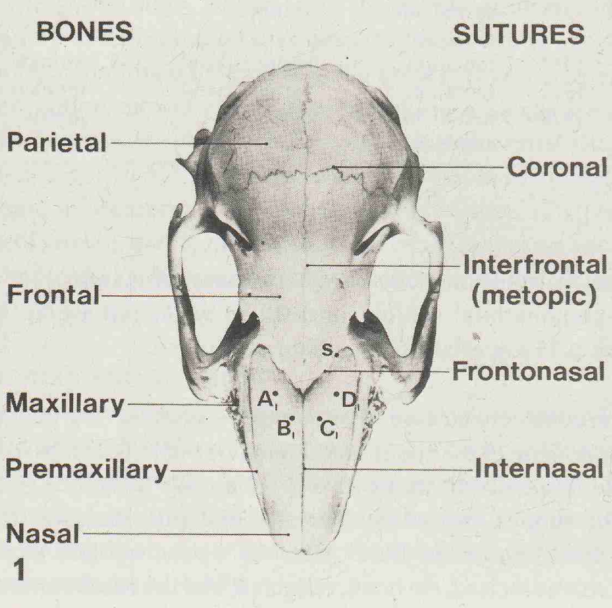
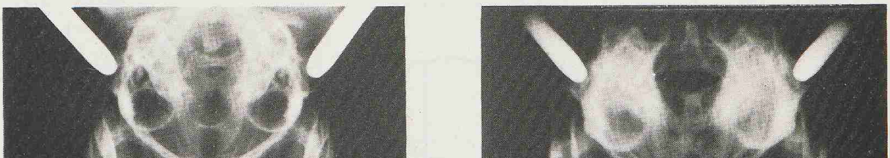


Figure 1.
Dorsal view of rabbit skull showing sites of implantation of dental amalgam in right (A_1, B_1) and left (C_1, D_1) nasal bones.
S, point on frontanasal suture

incisal pin placed between the maxillary incisors to orient the head in the same position each time the radiographs were taken. A ventrodorsal cephalometric radiograph was taken of the frontonasal region with the plane porion-interdentale (between the external auditory canals and the point of convergence of the upper incisors) oriented to the horizontal. This view had proved advantageous in the study of the rabbit snout (Selman and Sarnat, 1955).

Immediately upon completion of the surgical procedure a cephalometric radiograph was taken (Figure 2). This was repeated 10 weeks later at death (Figure 3). A separate tracing of each left and right nasal bone region was made on a matte acetate paper of the first and last radiograph with special attention given to the position of the implant images and the borders of the nasal bone region. On the radiograph a midline was established extending between the upper incisors and along the septovomeral midpalatal region. The lateral border was the lateral border of the premaxilla. The distal border was the most distal border of the nasal bone. The proximal border was determined by direct gross measurements of the



Figures 2, 3. Ventrodorsal cephalometric radiographs of rabbit at 6 (Figure 2) and 16 (Figure 3) weeks of age. Note increase in size and change in shape of the skull and particularly the snout. D_1, one of 2 implants in left nasal bone. The relationship between implants in the same bone did not change during the 10-week period. The tips of the ear posts are in the external auditory canals. The incisal pin in position.

distance of each nasal bone implant to the frontonasal suture at two points (such as D_1 to S) at 6 and 16 weeks of age (Figure 1). From this, the pattern and position of the frontonasal suture was estimated. A base for the serial radiographic tracings was obtained by placing the latter tracing over the initial one in a position where the two implant images (A_1 and B_1 or C_1 or D_1), recorded on each tracing, superposed. The difference in the two established outlines of the nasal bone region represented the changes in size and shape in two dimensions that had occurred during this period (Figures 4, 5). Selected measurements were made of the differences (Table 2).

Table 2. Approximate rabbit nasal bone region growth in mm at selected sites determined from tracings of ventrodorsal radiographs[1] and by direct measurements from implants to frontonasal suture[2], at 6 and 16 weeks of age (see Figures 4, 5).

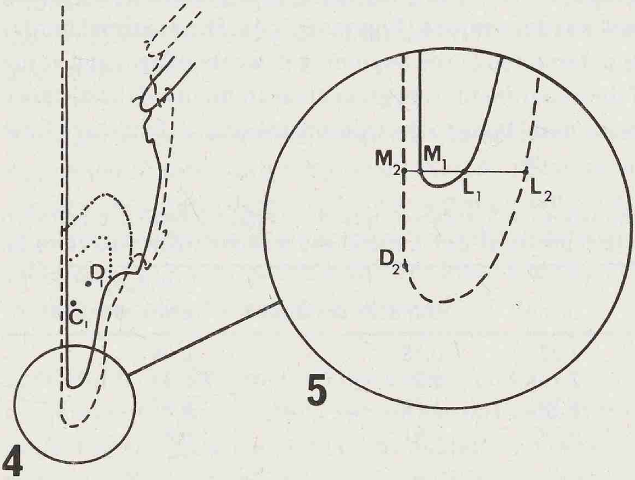
animal no.	medial border[1] (M_1-M_2)	lateral border[1] (L_1-L_2)	distal border[1] (M_1-D_2)	implant to FN suture[2]
7 left	1.4	2.5	5.3	D_1-S 7.9 C_1-S 7.0
right	0	2.8	4.2	B_1-S 5.7 A_1-S 8.3
21 left	1.4	2.8	7.8	D_1-S 8.9 C_1-S 8.7
right	1.4	2.9	7.6	B_1-S 7.4 A_1-S 8.9
26 left	1.0	2.4	6.6	D_1-S 6.9 C_1-S *
right	1.0	2.3	6.6	B_1-S * A_1-S 5.8
28 left	1.1	1.9	4.9	D_1-S 4.5 C_1-S 3.3
right	1.1	1.9	4.9	B_1-S 4.7 A_1-S 3.1
35 left	1.2	4.0	6.9	D_1-S 9.0 C_1-S 7.3
right	1.2	3.8	7.1	B_1-S 8.1 A_1-S 7.6

M_1 most distal point on medial border at 6 weeks of age;
M_2 represents M_1 on medial border at 16 weeks of age;
M_2-L_2 line at right angles to medial borders;
D_2 most distal point on medial border at 16 weeks of age;
L_2 point on lateral border at 16 weeks of age;
M_1-M_2 increase in medial dimension;
L_1-L_2 increase in lateral dimension;
M_2-D_2 increase in distal dimension.
* Implants B_1, and C_1, not seen on 16 weeks of age radiograph.

RESULTS

Gross. Of the implants inserted, two were missing (animal) No. 26). The remaining implants were well tolerated and invariably covered on the dorsum by a 0.1 to 0.3 mm layer of bone. Measurements taken between implants in each nasal bone were the same at the beginning and end of the experiment. Measurements were taken between each implant (in nasal bones) and a point on the frontonasal suture at the beginning and the end of the experiment (Figure 1). In this way the contributions of the nasal aspect of the suture to the increase in size of the nasal bone were determined at two points (Table 2).

Serial cephalometric radiographs and tracings. Measurements between the estimated centers of the images of the metallic implants in the same nasal bone showed no change at any time from the beginning to the end of the study (Figures 2, 3). Consequently these implants were used as sites of reference for superposing the tracings of the radiographs. In this way, not only sites but also, amount of change could be determined during the 10 week period that the radiographs were taken (Figure 4). The sites of growth were at all of the borders, and it was possible



Figures 4, 5. Tracings of left nasal bone region taken from ventrodorsal cephalometric radiographs in Figures 2, 3 and superposed on implant images in left nasal bone. Figure 4. C_1 and D_1, implant tracing, 16 weeks of age Estimated position and pattern of frontonasal suture at 6 weeks of age, ...; at 16 weeks of age, -.-.-. Note growth pattern of nasal bone region with greatest increase in size proximally and distally, less laterally, least medially. Figure 5. Enlarged area of Figure 4 to demonstrate reference points for measurements. M_1, most distal point on medial border at 6 weeks of age; M_2, represents M_1, on medial border at 16 weeks of age; M_2-L_2, line at right angles to medial borders; D_2, most distal point on medial border at 16 weeks of age; L_1, point on lateral border at 6 weeks of age; L_2, point on lateral border at 16 weeks of age; M_1-M_2, increase in medial dimension; L_1-L_2, increase in lateral dimension; M_2-D_2, increase in distal dimension.

to compare the relative amounts of growth (Table 2). The most prolific growth was at the proximal and distal borders, less at the lateral and least at the medial border (Figures 4, 5). The mean increase was about 6.79 mm at the proximal (posterior) border, 6.19 mm at the distal (anterior) border, 2.73 mm at the lateral border and 1.22 mm at the medial border (Table 3). Thus growth at the proximal and distal borders was about the same and about twice that of the lateral border and about 5 times that of the medial border.

DISCUSSION

The purpose of this study was to establish the growth pattern of the rabbit nasal bone region in terms of sites, relative amounts and directions of growth. This is based on the use of fixed reference areas within the nasal bone in relation to the nonfixed borders. The fixed reference areas were the two images on the serial ventrodorsal cephalometric radiographs of silver amalgam implants inserted in each nasal bone (A_1 and B_1, C_1 and D_1). These images, as well as the outline of the nasal bone regions, were transferred to translucent matte acetate paper. The tracings of the 6 and 16 week-old rabbit radiographs were superposed on the implant images in each nasal bone. In this way the growth pattern of the distal, lateral and medial borders was determined (Figures 2, 3, 4). The proximal border (frontonasal suture) was determined at the beginning (6 weeks of age) and at the end (16 weeks of age) of the experiment from gross measurements of the distance between the implants in the nasal bone to the frontonasal suture. Thus each time

Table 3. Statistical determinations in 5 rabbits of nasal bone region growth in mm from 6 to 16 weeks of age at proximal, distal, lateral and medial borders (see Table 2).

	means	standard deviation	standard error of mean
medial border	1.22	0.18	0.08
lateral border	2.73	0.75	0.33
distal border	6.19	1.31	0.58
proximal border (implants to FN suture)	6.79	1.81	0.81

paired T-test	probability
lateral border – medial border	0.009*
distal border – lateral border	0.002*
proximal border – lateral border	0.002*
distal border – medial border	0.001*
proximal border – medial border	0.002*
proximal border – distal border	0.367**

* Significant at 1% level
** Not siginificant

two points were determined on the frontonasal suture. The rest of the suture was estimated from gross studies.

The design of this experiment included measurements between implants within the same bone. Since evidence showed a constant relationship between these implants, we concluded that there was no interstitial growth of bone. If interstitial growth did occur, with consequent change in distance between implants within the same nasal bone, this experiment would be invalid. Thus, an essential start is a fixed stable reference site from which dependable measurements of growth may be made.

The accuracy of the results was limited by a variety of factors such as similar repositioning of the head for serial radiographs, a true ventrodorsal view, duplicability of tracing of radiographs, and superposing of tracings. In this radiographic study changes were determined in two planes of space without consideration of the increasing curvature of bone with growth of this region. Of all the determinations those at the frontonasal suture were the least accurate. Although no two animals exhibited identical quantitative growth, the general growth pattern was similar (Table 2). The length of the rabbit nasal bone is highly variable (Latimer and Sawin, 1962).

The basic pattern of a bone is inherent. In addition, prenatal and postnatal environmental factors influence growth, thereby affecting the external form and internal architecture of part of a bone to a complex of bones. Growth of the nasal region occurred by the addition of bone at the internasal, premaxillary-maxillary and frontonasal sutures and at the free distal and lateral borders. Dorsal, but not ventral, surface changes were observed.

John Hunter (1778) proposed that resorption was as charactristic of bone growth as apposition.* In this study apposition was determined for a given period. In evaluating the growth pattern of the mandible, however, it was possible to determine not only the total amount of apposition along the posterior border but also the total amount of resorption along the anterior border of the ramus (Robinson and Sarnat, 1955).

Contiguous bones which are identical, or mirror images, are joined by a suture with identical bone growth activity on both sides as for example the sagittal or internasal sutures. The essentially straight internasal suture of the growing rabbit

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* Another Englishman, Lewis Carroll, in *Alice in Wonderland* described some 12 decreases and increases in size, "... resorption and apposition"(?).

joins symmetrical nasal bones which taper. Constancy of the equal amount of bone growth on either side of the suture maintains the symmetrical bone and straight suture form. The particular anteroposterior shape of the snout, i.e. narrower anteriorly and wider posteriorly, raises a question as to the growth gradient all along the internasal and premaxillary-maxillary sutures as well as the lateral borders of the premaxilla. Progressive decrease anteriorly in the rate of sutural bone growth accentuates this tapering. Anterior sutural growth of the snout is arrested earlier than posterior sutural growth (Massler and Schour, 1951).

What factors play a role in growth of the snout? Selman and Sarnat (1957) and Sarnat (1958) noted no gross facial skeletal deformity after extirpation of the frontonasal or midpalatal sutures respectively. They concluded that the suture was a secondary or accommodating site of growth. Four months after resection of a large amount of nasal septum in young growing rabbits, when contrasted with the controls, the snout was not as long and not as large, the nasal and premaxillary bones were not as large nor were the nasal cavity and piriform aperture (Sarnat and Wexler, 1966; Sarnat, 1970; Verwoerd et al., 1979). At the posterior border of the septal defect, there was a strong downward deflection of the nasal bones in an anterior direction. This was in contrast to the smoothly curved dorsum of the control animals.

Questions arose regarding the role of the nasal septum and its sites of activity as one of the factors related to growth and form of the snout. Was the snout deformity after resection of nasal septum a result of either lack of growth or lack of support? In a different experiment large amounts of nasal septum were resected in adult rabbits (Sarnat and Wexler, 1967). After a postoperative survival of 16 weeks, study of the dissected skulls showed a large septal defect, but no deformity of the snout. These experiments suggested that the deformity of the snout, including less large nasal bones, after resection of nasal septum in growing rabbits could be the result of a lack of growth rather than a lack of support by the nasal septum.

Does growth of the nasal septum drive the snout forward, with sutural accommodation of the related bones? Did resection of a large part of the septum trigger closure or cessation of activity of the suture complex in the growing nasal bone region? Does normal interaction of the various growth patterns in a bone complex require maintenance of normal spatial relations of the individual units in the complex? Further investigation is indicated such as a study of nasal growth as in this report after septal resection.

Clinical comment. Precise analogies cannot and should not be made between rabbits and human beings. The amount of increase in separation of the implants on either side of the frontonasal suture indicated that this was a site of considerable growth. Since growth at this region was not affected after resection of the frontonasal suture, it was considered to be a secondary growth site. The proximal, frontal end of the nasal bone in rabbits is probably the most prolific of the four growth sites studied. Since severe trauma to this growth site did not result in a clinical deformity, this information may be relevant in regard to an osteotomy as part of a rhinoplastic procedure in a child or adolescent.

In contrast, resection of cartilaginous nasal septum in young rabbits produced a severe and striking deformity with a less large snout. From these findings, one interpretation to consider is that the growing cartilaginous nasal septum in rabbits is a primary and important growth site of the snout and the upper portion of the face. Thus, one might assume that, in children, trauma to the nasal bones providing they are repositioned, will not result in as severe a deformity, if any, as trauma to the nasal septum. It would be advisable that children who have sustained injuries to the cartilaginous septum and nose be treated and observed not only for immediate but also for late septal and nasal deformities. Furthermore, sufficient trauma to the septal region, in surgical procedures might have an untoward effect upon growth.

RÉSUMÉ

Deux implants radiopaques ont été introduits dans les os nasaux gauche et droit chez cinq lapins femelles. Des radiographies céphalométriques ventrodorsales ont été prises à l'âge de 6 semaines et à l'âge de 16 semaines. On a fait de ces radiographies des calques séparés, sur du papier acétate mat, des régions de l'os nasal gauche et de l'os nasal droit, y compris les implants radiopaques. Les marques du calque de la 16ème semaine ont été posées sur les marques du calque de la 6ème semaine. La différence dans les deux contours établis représentait en deux dimensions, les changements en grandeur et en forme qui se sont produits pendant la période de la 10ème semaine. Notre objectif a été de déterminer l'activité de croissance relative aux différents bords. La croissance moyenne a été d'environ 6,79 mm au bord proximal (posterior), de 6,19 mm au bord distal (anterior), de 2,73 mm au bord latéral et de 1,22 mm au bord médial. Or, les croissances aux bords proximal et distal étaient à peu près égales; elles se montaient à environ deux fois celle du bord latéral et à environ cinq fois celle du bord médial.

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