Craniofacial Development in Rats with Early Resection of the Zygomatic Arch

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Eighty-two-day-old male Wistar rats were selected to study the pattern of craniofacial growth following resection of the zygomatic arches. Rats were divided into three groups: group I (n = 14), the control group; group II (n = 15), with unilateral resection of the zygomatic arch; and group III (n = 8), with bilateral resection. Direct day skull and cephalometric measurements show increased facial projection and decreased transverse facial width on the side of the resected arch. If the results are extrapolated to the growth pattern of patients with the Treacher Collins syndrome, we can conclude that the zygomatic arch acts as a "moderator" in the morphologic development of the face. (Plast. Reconstr. Surg. 95: 486, 1995.)

In Mexico, 2 percent of all live births have one or more malformations, 40 percent of which involve the face and calvarium.1 Approximately 40 percent of patients who come to our department have some type of craniofacial malformation, which in 1991 resulted in 739 operations. Due to the complexity of their conditions, a multidisciplinary approach is required, with long periods of disability and high cost of care.

The syndromes that involve the first and second branchial arches especially are relevant because of the complex treatment they demand. Treacher Collins syndrome (mandibulofacial dysostosis) is a prime example. Alterations occur in the ears, eyelids, orbit, nose, cheeks, mouth, and palate as a result of bilateral Tessier facial clefts 6, 7, and 8.24 In severe cases, the zygoma is absent, and the facial bones display an excessive convex profile and also a decrease of their transverse dimension (Fig. 1). The maxilla is projected forward and upward. The mandible is small, retruded, and vertically oriented with an anterior open bite.

According to Munro and Kay,2 Enlow,6 and McNamara et al.,7 if it is true that the morphologic characteristics of the face are genetically determined, the final expression would then depend on the balance between the active divergent forces of growth and the resistance provided by the facial structures. In the skull, this relationship exists between the thrust caused by the frontal lobe growth versus the convergent forces of local bones and muscles.

The temporal muscles influence the transverse development of the upper and middle thirds of the face. The muscles of facial expression antagonize the sagittal growth forces of the craniofacial skeleton. Furthermore, the tongue affects the development of the mandible.

These concepts are reinforced by observations made on patients with unilateral facial paralysis of congenital or obstetrical origin, in whom the involved side shows greater growth than the healthy side. This phenomenon is the result of the inactive muscles of facial expression and the lack of their antagonistic effect on growth forces.

In this way, the effect of the genetic message may be modified by alterations in the balance between the different acting forces.

During the 20 years that we have followed patients with Treacher Collins syndrome, we have observed, even in those patients in whom bone grafts that were placed for malar reconstruction are well preserved, that the deformity becomes more evident with time. By means of anthropometric and cephalometric studies, we have confirmed that these patients experience

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an anterior growth of the maxilla that is greater than average (centrofacial hyperprojection).

In view of this and with the hypothesis that the lack of continuity of the zygomatic arch might have some causal relationship with the facial alterations observed in patients with the Treacher Collins syndrome and similar malformations, we studied the effects of surgical resection of the zygomatic arch in a group of experimental animals.

**MATERIALS AND METHODS**

Male Wistar rats (82 days old) were randomly placed into three groups: Group I consisted of 14 control rats on which no resections were made. Group II, composed of 33 rats, underwent surgical resection of the left zygomatic arch. Group III, with 34 rats, underwent bilateral resection of the zygomatic arches.

The operations were performed on the first day of life using inhalation general anesthesia with ether. Under microscopic magnification, a vertical incision was made between the eye and the ear (Fig. 2), and the zygomatic arch with its periosteum was resected. Our aim also was to preserve the branches of the facial nerve and the attachments of the masseter muscle. The skin was closed with 7-0 silk sutures.

In groups II and III, 21 animals were excluded from the study and an additional 22 animals died. Of the 21 rats excluded, 10 sustained injury to the facial nerve, and 11 were excluded because of damage to the masseter muscle. Of the 22 rats who died, 15 were killed by their mothers and 7 died in the postoperative period. Fifteen rats remained in group II and 8 rats in group III. All experimental animals were subjected to the same nutritional and environmental influences.

At 124 days of age, the rats were sacrificed with ether. The heads were placed in boiling water for 4 hours and then immersed in a 35% sodium hypochlorite solution for an average of 9 hours.
Once the skulls were dry, they were weighed on an analytic scale. A homogeneous sample was observed, with weight ranges between 2.6 and 3.14 gm (mean = 2.86 gm) (Fig. 3).

To perform cephalometric measurements, constant points were determined on all skulls. Conventional points were used, and these were augmented with additional landmarks. Using a Vernier millimetric caliper, we obtained the distances shown in Table I.

Because of the technical difficulties involved when measuring angles on a dry skull, radiograms were taken with a long-cone dental machine using a constant target distance of 8.5 cm. The following points on each radiogram were precisely traced on acetate sheets: incisive foramen, mesial root of the first molar, posterior nasal spine, and midpoint of the rim of the occipital foramen (Fig. 4).

Starting from the incisive foramen, straight lines were drawn to the mesial root of the first right molar, to the mesial root of the first left molar, and between these two molar points to create an anterior palatine triangle. A midline

![Fig. 8. Dry skulls of group I, control; group II, unilateral resection of zygomatic arch; and group III, bilateral resection.](image-url)
Fig. 4. Radiograph of rat skull with following reference points: (1) incisive foramen, (2) mesial root of first molar, (3) posterior nasal spine, and (4) midpoint of anterior border of occipital foramen.

was drawn from the incisive foramen to the midpoint of the rim of the occipital foramen, dividing the anterior palatine triangle in two halves. The anterior angle of each half triangle was recorded in degrees.

Linear measurements were made to the nearest tenth of a millimeter with the Vernier caliper. Statistical analysis of these data was performed in the following manner: The analysis of variance (ANOVA) test was used to compare measurements, and then Fisher's LSD multiple-comparison test was used to determine statistical significance.

RESULTS

Of the 16 analyzed measurements, we will only refer to the 7 most significant ones: 2 on the vault and 5 on the base of the skull (Fig. 5).

Nasion-lambda distance determines the anteroposterior growth of the vault of the skull. In the control group, we found a mean of 22.02 mm with a standard error of ±0.16 mm compared with a mean of 22.32 mm in group II and a standard error of ±0.22 mm. In group III, the mean was 22.68 mm with a standard error of ±0.16 mm. The \( p \) value (for the overall analysis of variance) was 0.03 (Fig. 6 and Table II).

The transverse parietal diameter had a mean of 10.06 mm in the control group with a standard error of ±0.08 mm. In group II, the mean was 10.05 mm with a standard error of ±0.07 mm. In group III, the mean was 9.68 mm with a standard error of ±0.10 mm. The \( p \) value was 0.016 (Fig. 7).

To determine the longitudinal growth of the base of the skull, the distance from the incisive foramen to the rim of the occipital foramen was measured. The mean distance in group I was 39.5 mm; in group II, 40.49 mm; and in group III, 40.16 mm. Differences between groups were statistically significant (\( p = 0.012 \)). With this parameter, we observed that the anteroposterior growth was greater in the operated groups than in the control group (Fig. 8).

To determine the longitudinal growth of the lateral aspect of the skull, measurements were made from the infraorbital foramen to the base of the styloid process. The right and left sides were measured separately with the following results: The mean was 31.77 and 31.74 mm, respectively, for each side of the control group. The standard error was ±0.19 mm for each. In group II, the mean was 32.52 mm for the right side and 32.90 mm for the left side with a standard error of ±0.18 mm. In group III, the mean was 32.47 mm for the right side and 32.45 mm for the left side with a standard error of ±0.25 mm. Thus rats of groups II and III grew more anteroposteriorly than those in the control group. In group II, the operated side grew more than the contralateral side. The difference between means resulted in a \( p \) value of 0.001 (Fig. 9).

The anterior transverse palatine diameter, measured from the anteromedial alveolar border of the first molar on one side to the same point on the opposite side, showed a mean of 5.84 mm in the control group, a mean of 5.77 mm in group II, and a mean of 5.56 mm in group III (Fig. 10). Dividing the measurements into right and left halves, significant differences were observed between both sides. In the control group, the mean was 2.93 mm on the right side and 2.91 mm on the left side with a standard error of ±0.04 mm on both sides. In group II, the mean was 2.83 mm on the right side and
Fig. 5. Seven most significant measurements: (left) cranial vault: (a) nasion-lambda distance, and (b) transverse parietal diameter; and (right) cranial base: (c) distance between infraorbital foramen and base of styloid process, (d) distance between incisive foramen and border of occipital foramen, (e) anterior transverse palatine diameter, (f) distance between midline and anteroinferior alveolar border of first molar, and (g) anterior palatine triangle.

TABLE II
Nasion-Lambda Distance for Individual Rats

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2.94 mm on the left side with a standard error of ±0.03 mm. In group III, the mean was 2.78 mm on each side, and the standard error was ±0.05 mm. The p value was 0.05 in each of the three studied groups.

The discrepancy in the anterior transverse palatine diameters was more easily demonstrated by measuring the anterior angle of each half in degrees. In the control group, the mean was 13.9 degrees for the right side and 14.9 degrees for the left side, respectively, with a standard error of ±0.3 degrees in each. The mean obtained in group II was 13.4 degrees on the right side and 15.8 degrees on the left with a standard error of ±0.29 degrees. In group III, the mean was 14 and 14.9 degrees, respectively, with a standard error of ±0.4 degrees (Figs. 11 and 12).

DISCUSSION

Previous studies have emphasized the relationship between the base of the calvarium and the facial skeleton. However, we were unable to find a reported study on the effects of early resection of the zygomatic arch on craniofacial development.

In our study, greater longitudinal growth was found in the groups where the zygoma was resected. In groups II and III, the distance between the nasion-lambda and the incisive foramen-occipital foramen showed a major increase in the anteroposterior dimension of the cranial vault and base, while the transverse parietal and the anterior transverse palatine diameters showed decreases. Thus major growth
Fig. 6. Nasio-lambda distance shows that longitudinal growth of cranial vault increased from 22.02 mm in the control group to 22.68 mm in group III (bilateral resection of zygomatic arches). The p value is for the overall analysis of variance.

\[ \text{Mean} \]
\[ P = 0.03 \]

Fig. 7. Transverse parietal diameter shows that transverse growth of the cranial vault decreased from 10.06 mm in the control group to 9.68 mm in group III.

\[ \text{Mean} \]
\[ P = 0.016 \]

occurred in the anteroposterior direction that was accompanied by minor transverse development.

The longitudinal growth of the lateral aspects of the base of the skull was measured by recording the distance between the infraorbital foramen and the styloid process. This distance increased bilaterally in group III, but the greatest growth was found on the left side in group II (resected side).
Fig. 8. Anterior incisive foramen to occipital foramen edge distance shows that longitudinal growth of the skull base increased from 39.5 mm in the control group to 40.49 mm in group II and 40.16 mm in group III.

\[ P = 0.012 \]

The bigger dimension of the anterior angle of the palatine triangle also was on the left side of this group.

Comparing the width of the halves of the palate, an increase in breadth on the resected side was evident in group II. But this dimension was reduced bilaterally in group III. Thus the evidence shows that the zygomatic arch resection in the newborn rat can bring about a greater anteroposterior facial growth with a de-
crease in its transverse dimension (Fig. 13). The greatest anteroposterior growth of the cranial base was observed in group II.

In order to explain this, the "counterpart principle" of craniofacial surgical growth is applied. It states that the growth of any facial or cranial part relates specifically to the other structural and geometric "counterpart." Thus imbalances are produced by differences in respective amounts or directions of growth between parts and counterparts. Therefore, a lack of enlargement of a particular part produces compensatory growth of its counterpart.¹¹ Not only the mechanical principles of balance exist.
Fig. 12. Skull base view. Variations in the anterior palatine angle show evidence of increase in longitudinal growth in the resected side with rotation to none on the operated side in group II.

Fig. 13. Growth differences between dry skulls from each group.
because the growth process itself is under genetic control. A series of morphogenetic structural genes under specific regulation exists, and this could be induced or repressed by means of nuclear, cytoplasmic, or membrane receptors produced by regulatory genes.12

Thus interaction of cells with their environment exists. The cells are surrounded by growth factors (ionic gradient, fibronectin, glycoproteins, etc.), forming the extracellular matrix, stimulating cellular reproduction and protein transcription, but there are also factors that inhibit growth, such as cell adhesive factor.13,14

Another growth control factor is the "functional matrix." The order of action of these three factors on growth are genetic regulation, mechanic regulation, and functional matrix.

In our study, the resection of one zygomatic arch produces more compensatory growth on the cranial base than bilateral resection. According to the "counterpart principle," this happened because the number of cell receptors is decreased, so counterpart overgrowth is less important than when the contralateral part exists, to be submitted to genetic and epigenetic control.

The measurements and alterations obtained in our study are very significant if we transfer them to the scale of the human skull. We can conclude that resection of the zygomatic arch disturbs the normal craniofacial growth. It does not stop or delay its growth. It is associated with a more anteroposterior growth. This growth potential originates in the center of the craniofacial skeleton. The zygomatic arch is a moderator of morphologic development and growth of the face. This thesis is supported by the fact that the most affected measurements were those of the anterior portion of the skull.

Extrapolating these results to humans, we can infer that the congenital absence of the zygomatic arch disturbs craniofacial growth and definitely affects the final adult Treacher Collins expression, independently of its etiology. It could be the cause of the reduction in the transverse diameter of the face and of the progressive facial hyperprojection observed in these patients.15,16

Early reconstruction of the zygomatic arch might prevent this additional deformity and result in a more harmonious development that surely would require simpler treatment and probably would require fewer surgical procedures.

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REFERENCES