THE EFFECT OF UNILATERAL NASAL BLOCKAGE ON THE GROWTH OF THE RAT CRANIOFACIAL COMPLEX - A QUANTIFIED GROSS AND HISTOLOGICAL MICROSCOPIC ASSESSMENT

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William Nigel Chalk

Department of Preventive Dental Science

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The effect of unilateral nasal blockage on the growth of the rat craniofacial complex - a quantified gross and histological microscopic assessment

by

William Nigel Chalk

ABSTRACT

The functional stimulus of nasal airflow on the growth and development of the rat calvarium was investigated. Unilateral surgical closure of the external nares altered the local functional environment. It was hoped to demonstrate the effects, if any, of the functional stimulus of the airflow or, alternatively, to illustrate the effects of surgery, or a combination of the two, on the growth of the craniofacial complex.

A sample of 88 male Long Evans rats of starting weight 90 ± 3 grams was studied, 71 animals having unilateral nasal blockage, 5 having a sham nasal operation unilaterally and 12 non-operated animals as a control. In this mixed longitudinal investigation 18 animals were sacrificed serially as a cross-sectional group. All the rats, except the nine animals in the soft tissue sample, were given intraperitoneal injections of NN'-di-(Carbomethyl) aminomethyl fluorescein (Dcaf), alizarin red S, terramycin, and terramycin and alizarin red S on days 0, 4, 11 and 25, respectively, after attaining the starting weight of 90 ± 3 grams, and were sacrificed on day 46 of the experimental period.

The general and local changes or reactions were investigated using weight gains, bacteriological, pH, temperature and soft tissue data. The cross-sectional data were used to evaluate the morphological changes over the experimental period, using photographic, radiographic and microscopic analyses. It was, therefore, possible to describe statistically the changes due to nasal blockage.

Finally, by a quantified microscopic analysis of the serial bone marking agents, a histological measurement of the effect of nasal blockage was made. A total of 32 paired anatomical sites in four coronal regions in the snout of the rat was investigated. The statistical evaluation of the growth differences bilaterally was achieved using an analysis of variance and, from a Duncan multiple range test, the students t test.

The following conclusions were made from this investigation -

1). The surgical technique was found to be satisfactory in producing unilateral nasal blockage with rapid post-operation recovery.

2). The histological examination showed no abnormal tissue changes except local post-surgical reparative processes.

3). The general health, as measured by the gain in weight, was found to be significantly inhibited by unilateral nasal blockage.

4). The pattern of bone growth of the nasomaxillary region was found to be complex and occurred by external apposition with concomitant internal resorption. Area relocation, differential growth and resorption were also demonstrated.

5). Quantitative coordinate analyses of the photographic and microscopic material, demonstrated as polygons, were effective in showing the growth changes cross-sectionally.

6). No gross skeletal changes were found following the surgical procedures and the minor changes reduced throughout the experimental period.

7). The bone thicknesses and bone increments were not significantly different when statistically compared bilaterally or between the groups.

8). This experimental technique has demonstrated that the effect of nasal blockage on the bone growth of the snout was so small as not to be significant statistically. While this does not rule out the possibility of a dynamic functional stimulus of the airflow, it does suggest that nasal blockage per se has a minimal effect on nasal growth.

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INTRODUCTION

There has been much controversy over the effect of chronic nasal blockage on the overall growth and development of the craniofacial complex. As the majority of children do pass through stages of partial nasal obstruction, this makes the issue important practically and clinically. The obstruction is usually brought about by engorged blood vessels of the mucous membrane linings of the nasal cavity and turbinates or by chronically inflamed or enlarged lymphatic masses such as the adenoids and tubular tonsils. It has been pointed out that, during the development of the permanent dentition, the child's lymphatic tissue is twice that of its mature amount and the airway is still growing.

Many investigators have concluded that nasal obstruction in young children will produce abnormal growth and development of the bony facial form including the shape of the dental arches. Other authors vigorously dispute this hypothesis.

Recent expansions of the functional matrix theory have postulated that such functioning units as the airway and sinuses have an intimate dynamic relationship with the surrounding tissues and play an important role in their growth and development.

Because of the myriad of other environmental factors which may be involved, it is understandable that there have been few human clinical studies in this field. Animal experiments have the advantage that controlled alterations of functional activity can be studied.

The aim of this investigation was to evaluate the effect of artificial, unilateral closure of the external nares in young male Long Evans rats on the normal growth and development of the craniofacial complex. This was a mixed longitudinal study of an altered functional environment on the rat skeletal form. With this experimental technique it was hoped to demonstrate the effects, if any, of the functional stimulus of the airflow or, alternatively, to illustrate the effects of surgery, or a combination of the two, on the growth of the craniofacial complex.

Longitudinal growth changes have been studied in a number of ways but only serial bone marking agents will reveal the sites and the amount of apposition at a histological level. Quantitative normative data have been evaluated for the direction and amounts of bone growth for male Long Evans rats using sequential staining in the midsagittal and coronal planes.

The objectives of this investigation were -

1). To consider concomitant local and general changes or reactions following unilateral nasal blockage, using the weight gains, soft tissue and bacteriological data.

2). To evaluate when the morphological changes occurred, using cross-sectional data.

3). To describe the gross morphological alterations following unilateral nasal blockage, using a quantitative analysis of the standardized photographs and radiographs of the rat crania.

4). To describe internal and external morphological changes in different anatomical regions from microscopic measurements of the hard tissue sections.

5). To give an explanation of the gross changes at the tissue level by a detailed microscopic measurement of the bone increments.

REVIEW OF THE LITERATURE

Human Implications of Nasal Obstruction

Tomes in 1872 suggested that the palatal and dental deformities found in malocclusions may be a direct consequence of nasal obstruction by enlarged tonsils. The anomaly, he felt, was caused by muscle pressures. Kingsley (1879) and Talbot (1894) contested this view and claimed that the facial musculature would be in the opposite direction to the deformity described.

Collier (1898) and Lane (1900) rejected the perioral muscle hypothesis and postulated a pressure hypothesis. They claimed that blocked nasal passages would have a marked negative air pressure during inspiration and that this force would be so great that the turbinates would almost be compressed to the nasal septum. Franke (1893), as cited by Brash et al. (1956), showed experimentally that the normal variations in air pressure within the nasal cavity are comparatively slight and rejected the mechanical pressure hypothesis as being exaggerated.

Spicer (1889) claimed that the lack of function was the cause of deformities following nasal obstruction. The normal functioning airway allowed the full development of the nasal passages and chronic disuse stunted this realization. This view was supported in the works of van der Klaauw (1948) and Moss (1954).

Most early authors felt that the facial pattern of the child would be affected by chronic nasal obstruction, leaving a flat skeletal profile, a narrow maxillary dental arch and a high vault. Angle (1907) claimed that mouth breathing was the most potent, constant and varied factor in the causation of malocclusions. Many of the authors noted how wider systemic conditions were often associated with nasal blockage. Some of these earlier authors were Brady (1902), Tilley (1908), Hoggan (1914) and Johnson (1936) and they described decrease in resistance to infection, failure to gain weight and many other symptoms of poor physical development.

Duke, in 1930, claimed that nasal allergy produced deformities of the face which were different from the types of deformity due to enlarged tonsils and adenoids, and were due to a lack of aeration of the paranasal sinuses. Todd (1936, 1938) and Cohen (1937) agreed that, in fact, growth was retarded in all three dimensions. Straub (1944) felt from his studies that allergies must be considered as an aetiological factor in the growth of the nasal passages and paranasal sinuses and hence the face and jaws. Rosenberger (1934) divided the nasal obstruction into anterior and posterior types, both affecting the growth of the nasomaxillary complex. He did not feel that heredity was the cause of the anomalies found, as was frequently claimed.

Many clinicians have felt that the maxillary malformations gave rise to respiratory inhibition and thus vital embarrassment, and recommended intra oral expansion appliances. Macary (1954), treating 15 patients in this way, felt a marked improvement was achieved. Isaacson et al. (1964) and Haas (1970) also suggested improvements in the upper respiratory airflow, while Wertz (1968) felt the changes were minimal.

Among the recent investigators, the role of enlarged lymphoid masses obstructing the airway and affecting the growth and development of the dentition and facial skeleton has continued to be controversial.

Subtelny and Sakuda (1966), Ricketts (1958), Yip (1969) and Linder-Aronson (1974) have all noted neuromuscular compensations in the tongue and jaw posture before and after surgical removal of these masses. Yip and Linder-Aronson felt that these compensations before and after surgery were reversible, the latter suggesting a direct influence on the maxillary incisors and the mandibular position, the maxillary width and the depth of the nasopharynx. Undoubtedly, the changes would be expected to be relatively small but possible changes in patterns of deglutition as found by Yip may be sufficient to direct morphological change.

Respiration is the primary function of life which, after birth, is the only natural means of survival. Bosma (1960, 1963) showed how 30 newborn children aerated their lungs by gulping in response to their need. He stated that infants with Pierre Robin Syndrome should not have tracheostomies as the normal neuromuscular patterns would not develop. Removal of the tracheostomy tubes necessitated the opening of the oronasal airway as in the newborn. Turner (1913) reported on children who had worn tracheostomy tubes for long periods and concluded that the lack of nasal breathing had not interfered with nasal development of the dental arches and maxilla.

Congenital anomalies which help the understanding of the growth and development of the nasal cavity are rare. Among these is choanal atresia, first reported in 1829 by Otto. Since then over 500 cases have been reported. While this anomaly may be bilateral or unilateral, it would serve as a non-surgical natural form and function model. Morgenstern (1940) described a case of left sided blockage in which the left side of the face was less well developed than the right.

Radiographs revealed an underdevelopment of the inferior turbinates, left frontal and maxillary sinuses. In other studies McGovern (1950), Cracovander and Goodman (1956), Craig and Simpson (1959), Flake and Ferguson (1964) and Ransome (1964) with over 100 cases did not note or did not find any obvious asymmetry, but stated that these were rare. Most authors, however, did note how these patients had a high infant mortality.

McLaughlin (1949) in reporting a case of absence of a nasal septum assumed it to be acquired as no cases had previously been presented of true congenital absence. He mentioned the high possibility of birth damage and noted poor or retarded nasal development. Moss (1968) reported two cases of congenitally absent nasal septa and suggested that the nasal septum played only a secondary compensatory role in nasal growth.

While much research has been done in the field of human form and function, there is no clear picture of a causal relationship between nasal blockage and orofacial skeletal deformity. Brash et al. (1956) recognized the limitations of the clinical view and believed that nasal blockage might lead to pathological conditions which might interfere with the normal development of the nose or of the body as a whole.

Assessment and Measurement Techniques

A knowledge of the degree of maturity is essential in studies of growing animals. There are no definite criteria for determining the stages of developmental processes leading up to the attainment of maturity. Maturity is taken as meaning the full structural integrity of form with functional development. As body growth and development is an integrated continuum, different aspects have been used to relate this degree of development. Chronological age is not a maturity indicator, but a time frame for comparing indicators. There is always a positive correlation of these indicators with growth but it can often be very low, to such an extent as to be of little value.

While most investigators using a maturity indicator have used body weight because it is the simplest to measure, it is probably the most susceptible to change due to environmental stress factors. Morphological changes were utilized in many of the earlier studies, e.g. Jackson and Lowry (1912), Sugita (1917), Donaldson (1912, 1924) and Cameron (1925), but their relations were merely guidelines. Jackson and Lowry pointed out that the head of the newborn rat comprised 23% of the body size, reaching a maximum at seven days of 26%. By adulthood, the overall head proportion of body size had dropped to 10% and then to 8% in animals over 300 grams.

As with most animal species, the rat shows a sexual dimorphism, with the male growing to a larger size than the female. The age of sexual maturity and sexual characteristics have been utilized in studies by Engle et al. (1937) and Hughes and Tanner (1970). The fact that there is a basic sexual dimorphism is the reason why mixed sex sample data should perhaps not be pooled as done in 1956 by Baer and Moss, using their data of 1954 and the data of Massler and Schour (1951).

Various techniques have been used to assess the degree of skeletal development. Among these, Bhaskar et al. (1950) used the tibia and Hughes and Tanner did a more comprehensive study of Long Evans rats studying the skull, limbs, pelvis and noserump lengths to evaluate the skeletal norms of development. They calibrated the equivalent hand wrist radiographical tabulation of ossification centres as had been used for children.

De Groot (1963) and Mosier (1969) used the tail length as the criterion for relative growth of the body under normal and in operational studies of the catch-up phenomena. Hammett et al. (1923) and Dienstein (1956) stated that while the eruption pattern of the teeth in man is a relatively good guide to dental development, its assessment in the rat is practically difficult and there is only one dentition erupting over a short period of growth.

Burr and Mortimer (1938) and Jarabak (1942) described a cephalometric technique on a living rat but did not use it to study growth. Spence (1940) in a longitudinal growth study of 62 Wistar rats pointed out two drawbacks of vital cephalometrics, i.e. the difficulty in accurately positioning the rat skull and the lack of normative data.

Baer (1954), Ford et al. (1959), Asling et al. (1963), Cleall et al. (1968) and Hanada (1969) all used serial longitudinal cephalometric studies of rats, often together with other branches of research. Their overall findings were similar, showing to varying degrees of accuracy the relative two-dimensional growth, but with no good estimate of growth rates because of the difficulty of superimposition and deciding which structures should be used for superimposition.

Although vertex basillar and lateral radiographs have been used in rat craniofacial cephalometrics, the limitations of the small skull, using the loose and fragile tympanic bullae for the positioning of the earrods, will always leave a technical error too large for refined growth evaluation. In any case such radiographical techniques will never reveal the specific regions of growth. Implant markers have been used in