

EXPERIMENTAL PRODUCTION OF CLASS III IN RHESUS MONKEYS

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Introduction

For some time the orthodontist has desired more definitive information about the effects of various kinds of orthodontic treatment on the growth of the craniofacial skeleton. A number of hypotheses seem possible if satisfactorily rigid testing could be designed.

- a. Nothing alters the development of the craniofacial skeleton; only the teeth are moved during treatment.
- b. Bony growth is stimulated by orthodontic therapy.
- c. Therapy optimizes the growth potential within the limits of the individual's genetic pattern.
- d. Orthodontic treatment alters the vectors of growth without altering the amount of growth.
- e. An alteration in the timing of growth events occurs but no differences are produced in final size by orthodontic treatment.
- f. Growth is inhibited in some regions by treatment, resulting in a relative change of parts.
- g. The response to orthodontic treatment is primarily a pathologic reaction.

Which, if any, of these or other hypotheses is correct would obviously depend on the type of orthodontic force used, the age of the patient, the direction and site(s) of force application, etc. It is also possible that varying answers are correct for varying circumstances.

There are many reasons why, after all these years, our knowledge in this area is quite incomplete. The principal reason, however, is our inability to measure and control the large number of operating variables in the patients we treat and study. Accordingly, it has been necessary to turn to research on animals, chiefly higher primates. Although this has been rewarding, there has been a general lack of elegance of experimental design for a number of reasons.

- a. Insufficient number of animals - primate research is costly.

- b. Lack of control animals or control standards of primate growth.
- c. Failure to identify accurately the age of the animals.
- d. Use of adult animals to study growth phenomena.
- e. Failure to quantify the methods of analysis.
- f. Lack of standardization of experimental conditions.
- g. Improper or inadequate measuring methods.

For the past four years a multi-disciplinary group of scientists at The Center for Human Growth and Development, University of Michigan, under the direction of Professor Donald Enlow, has been studying some of the basic aspects of craniofacial growth. One sub-group is studying the role of the neuromusculature in craniofacial growth. A matter of interest is how alterations in the sensory input to normal reflex functions affect growth of the face and jaws.

The Problem

This paper, one of several studies on the role of the muscles in facial growth, reports findings on Class III malocclusions produced experimentally in the rhesus monkey (*Macaca mulatta*) by contrived alterations in the occlusal pattern which prompt changes in the sensory input to the midbrain.

Materials and Method

- (1) Cephalometric methods - Cephalograms are taken at monthly intervals with a specially designed cephalostat (ELGOYHEN *et al.*) All animals have a series of tantalum implants placed in the manner described by BJÖRK (1955). Duplicate cephalograms are taken at least once on each animal to test the error of the cephalometric method. Each cephalogram is enlarged three times on Kodak Translite film before tracing to reduce measuring errors (ELGOYHEN *et al.*) A series of specially designed measures are used for assessing the response to each experiment (*Fig. 1*).
- (2) Vital Dye method - All animals receive either tetracycline HCl and/or Procion as a vital bone marker (BEAR and GAVAN 1968).
- (3) Electromyography - EMG records are taken routinely on the experimental animals by a method developed for this study.
- (4) Photographs, both intra- and extra-oral, are taken at the start and end of each experiment and on special occasions (ELGOYHEN *et al.*).
- (5) Hand-wrist radiographs are taken at monthly intervals (ELGOYHEN *et al.*).
- (6) Histologic sections are made of selected areas from one half of each head. Ground sections are made of selected areas from the other half head for study of bone growth and remodeling by ENLOW's methods (1968).

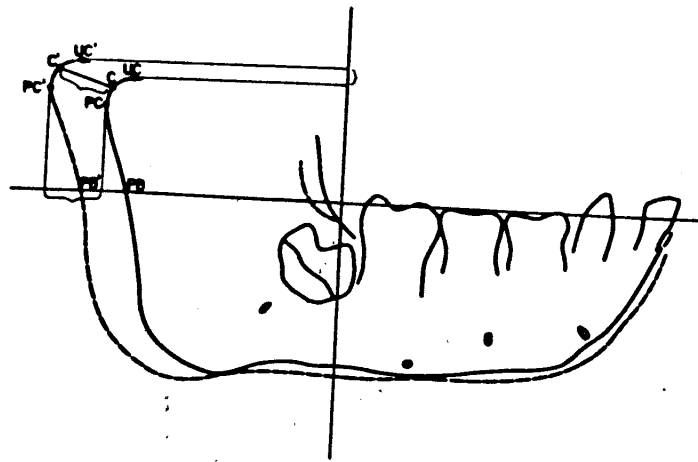


Fig. 1. Measuring method. The horizontal plane is the functional occlusal plane. The vertical plane is a perpendicular to the functional occlusal plane. This grid is used as a template by relating to the implants in successive radiographs.
 PB - intersection of functional occlusal plane and posterior border of the ramus
 PC - the most posterior point on the condyle
 UC - the most superior point on the condyle
 C - the most posterior-superior point on the condyle.



Fig. 2. Occlusal appliance with the mandible occluded protrusively.

Each animal is studied by these methods for at least three months prior to the start of a particular experiment to provide control growth data. Additional control data are available from a control sample (N = 13 and 18; see Table 1) of animals on whom all these routine data (except EMG) are gathered (ELGOYHEN *et al.*).

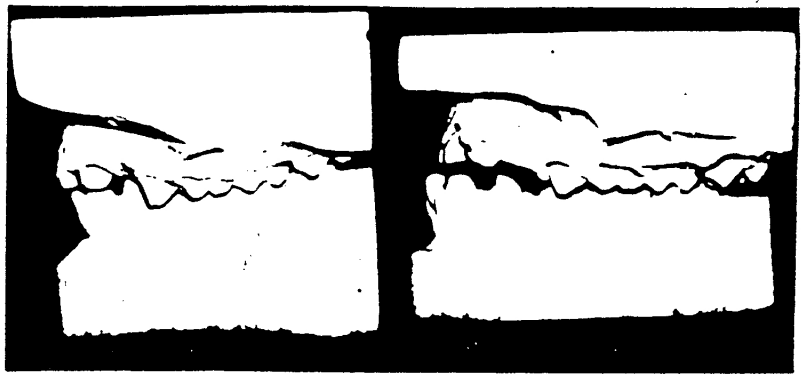


Fig. 3. Casts at the start of the experiment and after five months.

For this experiment six animals were used with identical developmental age (dentitionally) (SWINDLER 1961). New maxillary occlusal configurations were contrived in gold castings carefully made to prompt, reflexly, all occlusal function a specified distance forward with complete occlusal balance and just enough vertical opening to permit the desired mandibular displacement and functional use of the appliance (*Fig. 2*). Two types of splints have been used. Each appliance was designed in wax on an articulator and the occlusion was carefully equilibrated after insertion of the device to insure balanced occlusal function in the experimental position only. In different experiments the mandible has been displaced forward 2, 3, and 4 mm.

Findings

This paper will report some cephalometric and electromyographic findings only: details of other findings are being reported separately.

Dentitional Findings

After three months all animals displayed a typical Class III molar relationship, which was no longer a functional protrusion since the mandible could not be manipulated posteriorly when the animal was anaesthetized (*Figs. 3 and 4*).

At this time the mandibular incisal relations are atypical of Class III since their axial inclination has not altered and they are not crowded. The mandibular teeth have not migrated mesially (*Fig. 3*). The maxillary dentition showed little change in its relationships to supporting bone at the end of three months (*Fig. 4*).

After the appliance has been in place five months the findings are essentially the same except for vertical dento-alveolar growth in a continuing direction and some mesial migration of the mandibular buccal teeth (*Fig. 4 and 5*).

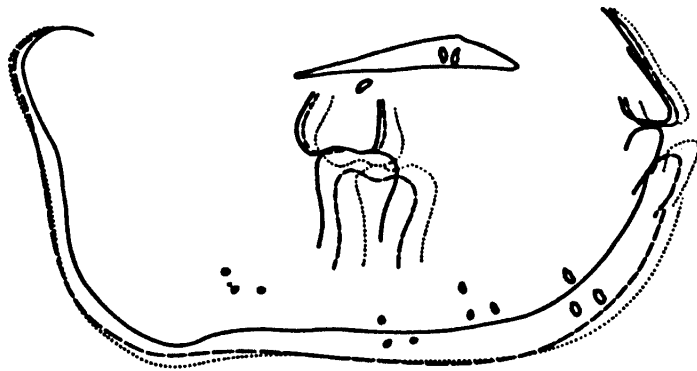


Fig. 4. Mandibular tracing superposed on maxillary implants. Solid line at start, dashed line after three months, dotted line after five months. Note, the appliance was in place at three months but removed at five months.

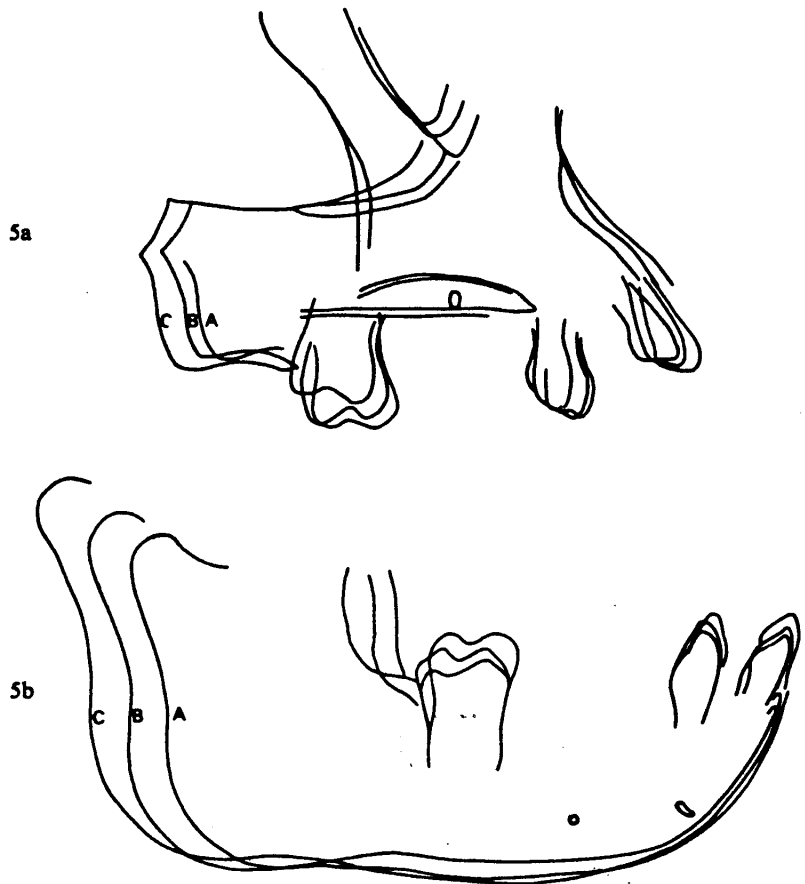


Fig. 5. Upper maxillary tracings superposed on maxillary implants. Lower mandibular tracings superposed on mandibular implants.
 (A) five months before appliance
 (B) at time of insertion of appliance
 (C) five months after insertion of appliance.

Table 1.

Comparisons in mandibular growth after three months.

Variables	Control (N=13)			Experimental (N=5)			\bar{D}	t	P
	\bar{X}_1	S ₁	R	\bar{X}_2	S ₂	R			
Mandible									
P.C.	5.6	2.40	1.6-10.1	9.1	1.31	7.3-10.5	3.4	3.178	.01
U.C.	3.8	1.88	.7- 7.5	6.2	2.82	2.6- 9.2	2.5	2.140	.05
C.	6.4	1.92	3.4- 8.4	10.6	1.43	9.2-12.7	4.2	4.472	.001
P.B.	5.8	1.83	3.3- 9.6	7.7	1.33	6.0- 9.5	1.9	2.094	.1

Skeletal Findings

The Class III skeletal relationship is established by three months and is maintained until the appliance is removed (Fig. 4).

Maxillary growth occurred during the experimental period in a pattern similar to that observed prior to the start of the experiment except that there was an increased rate of growth in the maxillary tuberosity during the experiment and some inhibition of vertical growth of the maxillary molar region (Fig. 5a).

The primary finding is a significantly greater growth upwards and backwards in the condylar region and to a lesser extent backwards at the posterior border of the ramus (Table 1 and Fig. 5b).

All experimental animals displayed changes in the angle of clivus which seemed to be different from the changes observed in the control animals.

Incremental analysis

Incremental differences were significantly greater in three condylar measures for the first three months but no differences between experimental and control animals were seen during the fourth and fifth months (Table 1 and Fig. 6). No correlation could be shown between the amount of forward displacement and the rate or amount of condylar growth, nor were differences between types of appliances shown. Some animals have had the mandible protruded in function for a second experimental period. Although the results will be reported separately (MOYERS *et al.*), it is interesting to note at this time that they have demonstrated a second period of increased rate of condylar growth.

Neuromuscular findings

The electromyographic data are much more difficult to obtain and analyze quantitatively; therefore, they are being reported in detail separately (KURODA *et al.*, MOYERS *et al.*). However, it has been found that such primitive reflex functions as mandibular posture and the unconditioned swallow may be altered within an hour by the conditions of this experiment (Fig. 7).

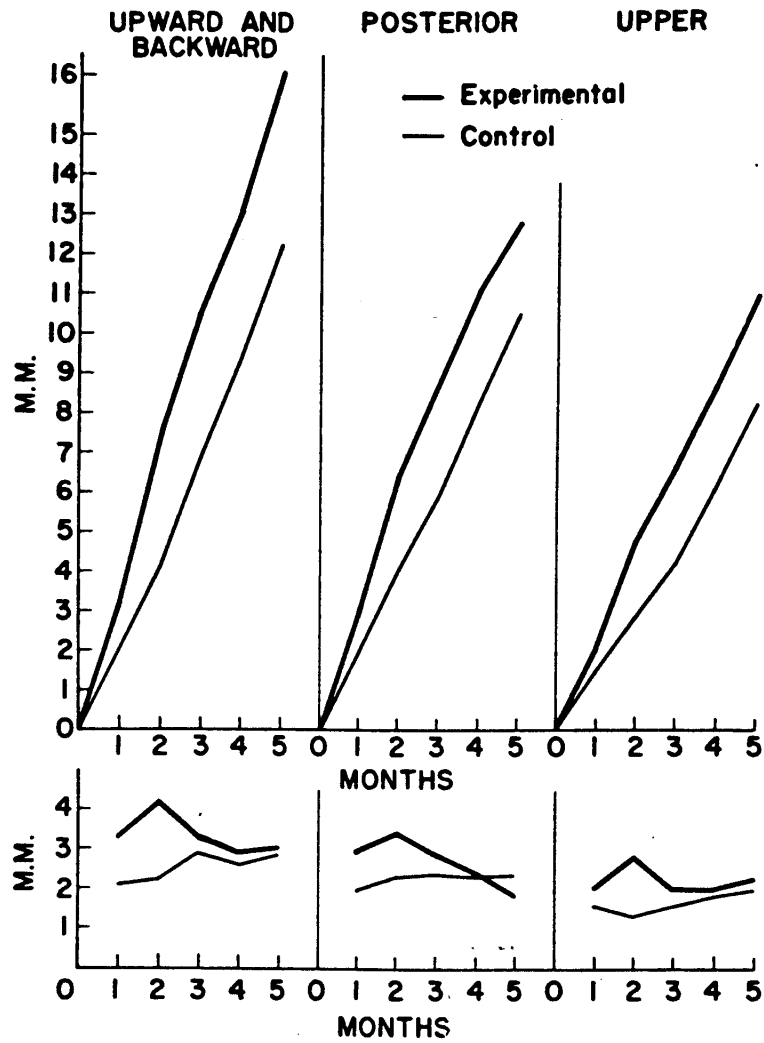


Fig. 6. Comparisons of growth in control and experimental animals. Upper - summated growth at the condyles measured three different ways (see fig. 1). Lower - incremental growth at the condyles measured three different ways (see fig. 1). This table reports results of five experimental animals only. A sixth animal was also done but not included.

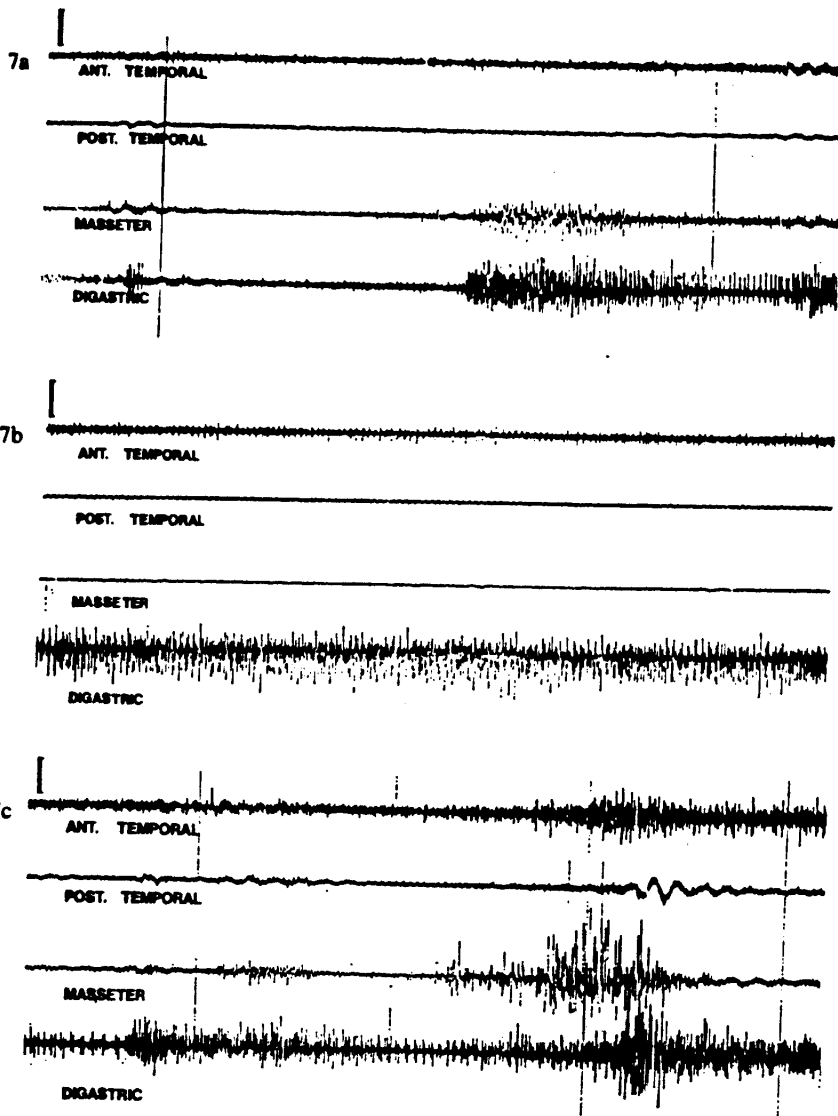


Fig. 7a. Mandibular posture and unconscious swallow on the same day as the experiment, but prior to insertion of the appliance. Vertical lines are one second apart. (Animal \neq 259)

Fig. 7b. Mandibular posture 8 hours after the start of the experiment (animal \neq 259).

Fig. 7c. Unconscious swallow 8 hours after the start of the experiment (animal \neq 259). Note particularly the differences in timing and amplitude of the anterior temporal and masseter records when compared to *fig. 7a*. The calibration is 500 mv. in all figures.

Discussion

The change in occlusion during the experimental period occurred because of an increased rate of condylar growth and subtle changes in dento-alveolar growth. The changes in cranial base flexure and tuberosity growth are being analyzed further, since they are particularly interesting findings which we find difficult to explain.

Comparison of our findings with other studies is a bit difficult due to differences in methodology. HINNIKER and RAMFJORD (1966) reported traumatic periodontal changes and distal movement of maxillary cuspids and only a minimal pathologic temporomandibular response, but they used adult monkeys. JOHO's fine study (1968) is the most comparable to ours, yet differs in two important ways: (1) his measures did not include that portion of the condyle we found most actively responding (2) his experimental appliance did not provide continuous occlusal function. Our animals all chewed well on the same diet within a few days and grew at the same rate as the control animals. We feel our measuring methods reveal findings likely present in several other studies. The enlargement of the cephalograms before tracing greatly reduced the error of the present method.

Extrapolations to orthodontic practice should be made with care. Our animals were all approximately six years human age. The skeletal and neurologic response was so rapid and over so soon, it would seem we should study the methods we use in functional jaw orthopaedics; perhaps we should replace activators more frequently.

It is important to remember that the midface growth in the monkey is much greater than in man; therefore, it probably is a relatively more difficult task to get these results in rhesus than it would be in homo (ELGOYHEN *et al.*).

We use a similar clinical approach to treat Class II to Class I, but in this experiment the occlusion went from Class I to Class III, which is not exactly the same.

It is difficult to believe, however, that the results are not of interest to those who attempt clinically to alter skeletal patterns and growth.

The neurophysiologic findings are most provocative and draw attention to the theories propounded by AHLGREN (1960) and HERREN (1965). Our electromyographic findings are being reported separately (KURODA).

SUMMARY AND CONCLUSIONS

Experimentally produced Class III malocclusions appeared in six monkeys who, after three months, functioned in a forward occlusal position. The change was largely due to a significant increase in the rate of condylar growth for the first three months only. Thereafter the increments were similar to those of the con-

trois mois. Alterations in reflex control of posture and the unconditioned swallow have been seen within the first hour.

We conclude that (1) we have much yet to learn about the relationship between variation in function and craniofacial growth; (2) the possibilities for exploiting clinically the neuromuscular effects on bone growth seem even greater than we may have realized; and (3) while it is easy to measure grossly in bone the results of altered neuromuscular function, it is much more difficult to explain *how* such changes are effected neurologically and to understand any local control mechanisms at the sites of bony deposition and resorption.

RESUME

Les occlusions dentaires défectueuses (Class III) produites artificiellement, sont apparues après trois mois d'expérience sur six singes (*Macaca mulatta*) qui ont été mis en occlusion protrusive.

Le changement est intervenu en grande partie sur la croissance condylienne d'une façon significative pendant les trois premiers mois seulement.

Après cela, les augmentations ont ressemblé à celles des animaux témoins.

On a vu dès la première heure, des modifications du réflexe contrôlé de posture mandibulaire et de la déglutition.

Les Conclusions sont :

(1) Qu'on a encore beaucoup à apprendre au sujet des rapports entre la fonction et la croissance cranio-faciale.

(2) Qu'il y a plus de possibilités d'investigations cliniques concernant les effets neuro-musculaires sur la croissance osseuse qu'on ne pouvait se l'imaginer au commencement.

(3) Et s'il est facile de mesurer macroscopiquement sur l'os les résultats de l'action de la fonction neuro-musculaire remaniée, il est beaucoup plus difficile d'expliquer ces remaniements et de comprendre les mécanismes de contrôle local dans les régions d'apposition et de résorption osseuses.

DISCUSSION

PROF. HERREN said that he was very much impressed by the paper and would like to make a few remarks. First, it was interesting to see that the results that Professor Moyers had obtained in producing Class III anomalies so very much resembled the results in treating Class II cases and trying to produce a normal jaw relationship. In his view papers from BREITNER up to the present day demon-

strated that the basic problem is a biological one and that the differentiation between Class I, II and III jaw relationships is an arbitrary one and not purely biological.

He was very pleased to hear that the views that he had expressed contrary to those of HAUPL in the discussion in 1956 on functional jaw orthopaedics fitted in quite well with the findings of Moyers' experiments.

Professor Moyers hesitated to equate the results of his experimental findings in apes with the clinical results that one could expect in the human but Professor Herren wondered whether he need be so hesitant. If one observes Class II cases treated with activators in a construction bite where there has been overcompensation one can see that it is possible to create Class III cases. This can occur if the patient fails to attend for checks on treatment progress and is, in consequence, over-treated. This results in an experimental Class III relationship produced from an initial Class II case. Fortunately if these cases are seen in time and the activator is removed the condition relapses in two or three weeks but he could recall one case in which the patient did not return early enough and after starting Class II treatment it was subsequently necessary to initiate Class III treatment in order to restore a normal occlusion. Prof. Moyers had observed that varying the relationship between the upper and lower jaw from 2 to 6 mm. resulted in no variation in the amount of growth subsequently observed. Professor Herren and his colleagues had observed the results of treatment in patients wearing activators with a construction bite of 4, 6 and 10 mm. and it was clear that there was a more rapid reaction with the larger overcompensation and that the treatment progressed further.

It was interesting to see the important rôle played by the digastric muscle which confirms his own hypothesis about the mode of action of the activator. It is a pity that it is not possible to study the action of the lateral pterygoid by this method. Professor Herren believed that these two muscle groups are the antagonists most involved in this process and that they should be observed simultaneously. If a method could be devised to do this it would represent a big step forward.

DR. WITT said he would like to ask Professor Moyers whether he had observed similar reactions in the masseter and the digastric. He and his colleagues had carried out observations on cases of Class II malocclusion being treated with appliances and they had made electromyographic observations of the reaction of the masseter muscles at regular intervals and after treatment had been completed. They believed that the adaptation of the muscle was of outstanding significance in determining stability after treatment.

They had also observed that at the beginning of treatment with functional appliances there was a marked increase in the muscle activity when the appliance was being worn. At some stages a so-called normalisation began but he questioned whether it was as simple as that and whether one could really call the new position of muscle balance a normalisation. He believed that this was a

very difficult problem and he wondered if Professor Moyers could throw any further light on the question of stability.

DR. STOCKLI said that he was most intrigued by the papers. Three years ago he and his colleagues had carried out a very similar experiment and believed that the results had been very much the same as those described by Professor Moyers. Professor Moyers had made a very positive statement about the increment of condylar growth and Dr. Stockli wondered whether he would make any comment at this point about the mechanism particularly the histological mechanism of this adaptive process.

DR. AHLGREN said that he had been fascinated by the paper and was very glad to have heard it. Professor Moyers had referred to similar experiments that RAMFJORD had made. As Professor Moyers knew, these experiments did not show any increased growth in the mandible, it had been found that the mandible went back concomitant with alveolar removal or remodelling. Did the clasps that Professor Moyers had used hinder the mandible and affect its posture in the saggital plane. He would also like to know what the monkey did with the electrodes. How did Professor Moyers manage to keep them in place. Dr. Ahlgren had carried out experiments on a human subject by fitting clasps to the molars which forced the mandible to function in a protruded position but he found that after about three weeks the clasp was bitten down and the mandible had taken up its original position similar to the results that RAMFJORD had found in monkeys. Finally he believed that fitting an activator in the mouth resulted in an increased activity in the digastric and the anterior temporal muscles.

DR. PATRIKIOS said that Professor Moyers had produced a Class III malocclusion from an original Class I malocclusion: We can assume that Class I has a normal amount and direction of growth. If you apply the same technique of bringing the jaw forward with activators in a Class II malocclusion, where in some cases there is probably deficient mandibular growth, will you get the same response as the mandible moves from a Class II to a Class I relationship. He would also like to know what effect an activator would have in cases with a high mandibular plane angle.

PROF. MOYERS said that first he would like to thank all his questioners. Some of the questions were very perceptive, he did not have answers to all of them but would like to comment on some of them.

In reply to PROF. HERREN, he would not comment on the first and second

points which he thought were rather more declarative than inquisitive and he happened to agree with Professor Herren. The third point related to the differing responses according to the amount of anterior positioning. Professor Moyers would like to point out that what Professor Herren had found in humans may not be different from what they had found in monkeys. In the first place, the monkeys' response is quite fast and they took cephalograms only at monthly intervals; therefore it is quite possible that what Professor Herren found was all over by the second month in the animal experiments and the investigators did not even see it occur. Furthermore, one can inhibit the gamma efferents in monkeys very easily with less forward positioning than in humans.

The fourth point, about the pterygoid; he was just as interested as Professor Herren but he would not like to show his monkey pterygoid electromyographs at this time with some of the eminent workers in the field present in his audience since only recently were they able to get routinely good E.M.G. records of monkey lateral pterygoids. If the first good records are indicative of those to come, then Professor Herren would be interested and in agreement with the findings.

Replying to DR. WITT Professor Moyers thought that there may be a slight language problem since he was replying to the question as he had heard it on the simultaneous translation. He would like to point out that while he used activators he did not feel that one could draw direct comparisons between the result of their experiments and the use of activators because the appliance in the experiments is cemented in place, unlike the activator which is free to drop when the mouth is opened.

There was also a semantic problem in the use of the word normalisation. He had taken a quick look at his text and he thought that the phrase he used was 'more nearly normal'. He would not wish to convey the impression that the muscles at the end of the experiment were completely as they had been at the outset.

In reply to the question by DR. STOCKLI, Professor Moyers thought that the question of how the increments are produced was the most difficult question of all to answer, because, although he could describe what happened, to account for how it happened was another question altogether. There are in the team better histologists, anatomists, and bone biologists than he and if Dr. Stockli was interested he suggested that he look out for the papers that will be produced by Professor Enlow, Dr. McNamara and others on these points.

In reply to DR. AHLGREN. First the comparison of his experiments with those of SIGURD RAMFJORD'S studies. RAMFJORD had used adult animals and these were pretty old adults so he did not feel that it was reasonable to make any direct comparison with their own experiments except to say that none of their young animals thus far had responded like RAMFJORD'S adult animals.

RAMFJORD had achieved primarily a pathological response and an actual retraction of the maxillary dentition. It was his intention however to repeat these experiments on older animals soon for more accurate comparisons. Since Professor RAMFJORD is a colleague at Michigan, there is interest in the experiment. Professor Moyers said that they had been particularly careful to ensure that the gold casting interfered with the antero-posterior posture of the mandible as little as possible. Clearly, however, he had not emphasised this point in the paper and he was very grateful to Dr. Ahlgren for raising the question. Fitting the electrodes to the monkeys is quite a problem. The monkey is seated in a plastic chair with a collar that prevents him from reaching the electrodes. Professor Moyers said that two of the orthodontists, DR. KURODA and MCNAMARA, are extremely clever at handling the animals and he cannot give too much credit to their ingenuity and skill in this part of the experiment. He had no comment to make on the patient in Malmo who was treated for Class II malocclusion and whose castings wore down except to say that in the monkey experiments the castings did not show any marked wear. Professor Moyers agreed that the electromyographic response was similar to that obtained when using an activator as demonstrated in Ahlgren's own splendid studies. Professor Herren had already commented on the comparison between the monkey and the human.

In reply to DR. PATRIKIOS, Professor Moyers said if he had to guess – and that is all that he could do – about the bony response in the monkey experiments he would guess that the localised bony regulatory mechanisms must be the same, though the initial skeletal pattern differed. He was sure that several people must have seen what Professor Herren had described as a Class II case looking 'Class III-ish' at the end of treatment. Finally the high mandibular plane problem, Professor Moyers did not know the answer. He personally did not use an activator in treating these cases, he would use extra-oral traction and high-pull head gear. Monkeys do not have high mandibular plane angles so that he had not been able to study this in the animal experiments. He could see no advantage at the present time in using an activator in the clinical treatment of these particular cases.

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