



Original Article

The Association of ACTN3 Rs1815739 Polymorphism with Various Malocclusion Phenotype

Hussain Nihad Izaddin Alalim¹ , Başak Funda Eken² , Korkut Ulucan² , Fulya Özdemir¹ 

¹Department of Orthodontics, Marmara University, Faculty of Dentistry, İstanbul, Turkey

²Department of Medical Biology and Genetics, Marmara University, Faculty of Dentistry, İstanbul, Turkey

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Main Points

- A relationship is present with some traits of the face and alpha-actinin-3 (ACTN3) rs1815739 polymorphism.
- This relationship is present in the sagittal position of the maxilla and inclination of the maxillary incisors.
- The prognathic maxilla was related to RR genotype.
- The retrognathic maxilla was found to be related to RX and XX genotypes.
- Subjects having normal incisor angulation in the maxilla had no RX genotype but had RR and XX.

ABSTRACT

Objective: A functional polymorphism on the 16th exon of the alpha-actinin-3 gene has an effect on the protein structure and cellular signaling and therefore on muscle contraction. In this study, we aimed to analyze the alpha-actinin-3 rs1815739 polymorphism in 3-dimensional malocclusions and different craniofacial skeletal patterns.

Methods: Forty-nine volunteering subjects enrolled for the study. Genotyping of alpha-actinin-3 rs1815739 polymorphism was performed using real-time polymerase chain reaction. Pre-orthodontic cephalometric radiographs were traced using NemoTech cephalometric tracing software. IBM SPSS Statistics for Windows was utilized to carry out statistical analysis. $P < .05$ was considered to be statistically significant.

Results: The respective numbers and the percentages of alpha-actinin-3 rs1815739 polymorphisms for RR, RX, and XX genotypes were 39 (79.6%), 4 (8.2%), and 6 (12.2%), respectively. Twenty-one patients had low angle vertical patterns and 17 patients had Class I and the same number of the patients had Class III facial patterns. But none of these had statistically significant difference in terms of alpha-actinin-3 rs1815739 polymorphism and in vertical or sagittal facial patterns, and mandibular incisor inclination. When we examined the maxillary anteroposterior position, we found a significant difference between rs1815739 polymorphisms ($P < .05$). Also, we detected a significant difference between rs1815739 polymorphism and maxillary incisor inclination ($P < .05$).

Conclusion: Maxillary incisor inclination and maxillary anteroposterior position are associated with alpha-actinin-3 rs1815739 polymorphism in a Turkish cohort.

Keywords: ACTN3, facial pattern, malocclusion, polymorphism

INTRODUCTION

Malocclusion is a complex trait in both its phenotypic expression and its genetic etiology, and several genes are associated with the phenotype. Dental and skeletal malocclusions, especially bone, teeth, skeletal muscles, and other soft tissues, are affected by transcription and growth factors combined and additional environmental factors. Our knowledge of the etiology of many of the resulting malocclusion is very limited. The effect of muscle tissues on the bone structure of the face and therefore malocclusions has been a subject that has been included in textbooks for years.¹ In studies conducted in the last decade, it has been shown that facial muscles are quite effective on facial morphology.^{2,3} Therefore, gene variants related to muscle composition and activity have also

been studied and have been associated with different craniofacial skeletal phenotypes.⁴ Alpha-actinin-3 (*ACTN3*) is one of these genes. Proteins expressed by these genes are closely related to muscle function. The presence of *ACTN3* was found to be associated with strong and rapid contraction in type II muscle fibers.⁵ The rs1815739 polymorphism (R577X) of the *ACTN3* gene has been associated with Class II deep-bite malocclusion⁶ and sagittal and vertical craniofacial skeletal pattern.⁷

Actinins are actin-binding proteins and have important functions in muscle contraction. In humans, there are 4 different types of actin, and each type of actin is encoded by specific genes.⁸ The product of the *ACTN3* gene, ACTN3 protein, is localized in the Z lines of the sarcomers and is responsible for binding the actins more closely to the Z line.⁹ The result of the variation in codon coding for amino acid 577 of exon 16 of *ACTN3* (R577X; dbSNP rs1815739) leads to a change of arginine amino acid (R) to a stop codon (X). This transformation results in shorter protein formation than normal form.¹⁰

In addition, *ACTN3* interacts with the signal protein calcineurin to influence fiber-type ratios during growth, causing changes in muscle function.¹¹ Alpha-actinin-3 binds to the calsarcin family of signal proteins on the Z disc and binds to calcineurin to activate specific gene expression pathways of the muscle fiber type that determine muscle fiber types and size.^{12,13}

The aim of our study is to investigate the potential relationship between 3-dimensional malocclusion phenotypes and *ACTN3* gene that may play a role in craniofacial development. For this purpose, the *ACTN3* rs1815739 polymorphism, which is expressed in muscle cells and is related to muscle structure by having important functions in providing appropriate muscle movements, is examined in adults with malocclusions to determine its clinical effect in a Turkish population.

METHODS

The Participants

Forty-nine (18 male and 31 female) orthodontic patients, who have applied to the Department of Orthodontics at Marmara University between January and October 2019 and have agreed to be a part of this project, participated in this study. The study protocol was in agreement with the Helsinki Declaration 2 (2015) guidelines and approved by Uskudar University Non-Interventional Ethics Committee (61351342/2019-575). The volunteers participating in the study were given detailed information about the analyses and outputs before the study, and their consent forms were obtained.

The following inclusion and exclusion criteria were followed to include or discard patients who were not in line with the purpose of this study. Inclusion criteria were being older than 18 years, being in permanent dentition, having no previous orthodontic treatment, and having available pre-orthodontic records. Exclusion criteria were having a history of orthodontic or orthognathic treatment, an aesthetic/plastic operation or trauma in the facial area, having any hereditary genetic disease in its self and

in their first-degree relatives, having uncontrolled medical systemic disease or diseases, cleft lip and palate and having lost more than 1 permanent tooth.

Pre-orthodontic cephalometric radiographs were used in this study to evaluate the sagittal and vertical malocclusion of the patients. All cephalometric radiographs were traced using NemoTech Cephalometric tracing software (Version 10.4.2, Madrid, Spain) by a single examiner (HA).

CEPHALOMETRIC MEASUREMENTS USED IN THIS STUDY

Vertical

1. Sum of inner angles: the collective sum of the saddle angle, articulare angle, and the gonial angle
2. Gonion–Menton–SN: the angle between the anterior cranial base and the plane passing through the gonion and menton points
3. ANS-Me/N-Me: the ratio of anterior facial height to lower anterior facial height
4. Jarabak ratio: the ratio of posterior facial height to anterior facial height
5. FMA: the angle between the Frankfort horizontal plane and the mandibular plane
6. Maxillary height: The angle between the nasion, center of face, and A point

Sagittal

1. SNA: the angle between the S-N and the N-A plane
2. SNB: the angle between the S-N and the N-B plane
3. ANB: the angle between the N-A and the N-B plane that determines the sagittal relationship between maxilla and mandible
4. Wits: a measure of jaw relationships in an anteroposterior plane, by drawing perpendiculars from A and B points on the occlusal plane
5. N per A: distance between A point from a line drawn perpendicular from nasion with respect to the Frankfort horizontal plane

Dental

1. UI-SN: the angle between the axis of maxillary central incisor and the SN plane
2. IMPA: the angle between the axis of the lower central incisors and the mandibular plane
3. LI-UI angle: the inter-incisal angle, the angle between the axis of maxillary and mandibular incisors
4. UI-OP angle: the angle between the axis of upper incisor and occlusal plane
5. LI-OP angle: the angle between the axis of lower incisors and occlusal plane

For the assessment of vertical discrepancy, cephalometric radiographs were used to categorize the patient into normal, high and low angle vertical pattern. The analyses that were used are total inner angle, GoMe-SN, Jarabak, ANSMe/NMe, Frankfort mandibular plane angle, and Maxillary height. Since there are many variations in these analyses, all of the measurements were used

to categorize the patient into 1 of the 3 vertical patterns in order to come up with a more accurate diagnosis for the patients.

ANB and Wits analyses were utilized to classify the patients into Class I, Class II, and Class III relationship. SNA, maxillary depth, and nasion perpendicular A were utilized to categorize the patients into normal, prognathic, and retrognathic maxillary position groups, and the overall maxillary position of the patients were decided by considering all 3 analysis.

Incisor SN angle and maxillary incisor-occlusal plane angle were utilized to determine maxillary incisor inclination and categorized them into normal, proclined, and retroclined incisor inclination groups. Similarly, incisor-mandibular plane angle and incisor-occlusal plane angles were utilized to determine mandibular incisor inclination and categorizing patients into normal, proclined, and retroclined incisor inclination groups.

ACTN3 RS1815739 GENOTYPING

DNA Isolation

Oral epithelium cells were collected by DNA collection swabs from the volunteers who participated in the study, and DNA isolation was completed by using PureLink DNA isolation kit (Invitrogen, Van Allen Way Carlsbad, Calif, USA). Briefly, 20 µL proteinase K was vortexed by adding 10 µL of RNAase to 200 µL of DNA isolation. After 2 minutes at room temperature, 200 µL of binding buffer was added and homogenized with stirring. After incubation for 10 minutes in a 55°C water bath, 200 µL of ethanol was added and vortexed for 5 seconds. It was taken to the filtered tube and centrifuged at 10 000 g for 1 minute. The supernatant was discarded and 500 µL of washing buffer was added to the pellet and centrifuged at 10 000g for 1.15 seconds. Later, 80 µL of elution buffer was added and incubated and centrifuged at maximum speed for 1 minute. The DNA samples obtained were stored at -20°C until the analysis of the respective gene regions was completed. An average of 20 ng of DNA was isolated from

each sample, and the isolated DNAs were evaluated according to the OD260/280 spectrophotometric ratio. The DNA samples obtained were stored at -20°C until the analysis of the relevant gene regions was completed.

Genotyping of ACTN3 rs1815739

Genotyping of ACTN3 rs1815739 polymorphism was performed from the isolated DNA by using 7500 Fast Real-Time PCR System (Applied Biosystem, Foster City, Calif, USA). Taqman Genotyping Assays (Applied Biosystems) genotyping kit was used for allelic determination.

Statistical Analysis

IBM SPSS-Statistical Product and Service Solutions (Statistics for Windows, Version 19.0., Armonk, NY, IBM Corp.) was used to conduct statistical analysis. Descriptive analyses were presented using means, standard deviations, median, and minimum and maximum values for continuous data, and frequencies and percentages for categorical data. The variables investigated using Kolmogorov-Smirnov test to determine whether or not they were normally distributed. The Fisher’s exact test was used to compare the proportions of the groups. Since the variables were not normally distributed, Kruskal-Wallis test was used to compare medians of the groups. A 5% type I error level was used to infer a statistical significance.

RESULTS

Vertical Facial Pattern

There was no statistically significant relationship between ACTN3 rs1815739 polymorphism and vertical pattern $P > .05$ (Table 1).

When the overall vertical pattern was analyzed, the low angle group had 21 patients, normal group had 9 patients, and the high angle group had 19 patients. In the low angle group, 18

Table 1. Genotype and Percentage Distribution of ACTN3 rs1815739 Polymorphism in Vertical Pattern Groups

			Genotype			Total	P
			XX	RX	RR		
Overall vertical pattern	Low	Patients	1 _a	2 _a	18 _a	21	.560
		Percentage	4.8	9.5	85.7	100.0	
		Percentage within all patients	16.7	50.0	46.2	42.9%	
	Normal	Patients	2 _a	0 _a	7 _a	9	
		Percentage	22.2	0.0	77.8	100.0	
		Percentage within all patients	33.3	0.0	17.9	18.4	
	High	Patients	3 _a	2 _a	14 _a	19	
		Percentage	15.8	10.5	73.7	100.0	
		Percentage within all patients	50.0	50.0	35.9	38.8	
Total	Patients	6	4	39	49		
	Percentage	12.2	8.2	79.6	100.0		
	Percentage within all patients	100.0	100.0	100.0	100.0		

$P < .05$ indicates statistically significant difference. Same subscript numbers indicate statistically insignificant relationships among genotype groups.

Table 2. Genotype and Percentage Distribution of ACTN3 rs1815739 Polymorphism in Sagittal Pattern Groups

			Genotypes			Total	P
			XX	RX	RR		
Maxillary Position	Normal	Patients	1 _a	0 _a	16 _a	17	.044*
		Percentage	5.9	0.0	94.1	100.0	
		Percentage within all patients	16.7	0.0	41.0	34.7	
	Prognathic	Patients	0 _a	0 _a	9 _a	9	
		Percentage	0.0	0.0	100.0	100.0	
		Percentage within all patients	0.0	0.0	23.1	18.4	
	Retrognathic	Patients	5 _{a,b}	4 _b	14 _a	23	
		Percentage	21.7	17.4	60.9	100.0	
		Percentage within all patients	83.3	100.0	35.9	46.9	
Total	Patients	6	4	39	49		
	Percentage	12.2	8.2	79.6	100.0		
	Percentage within all patients	100.0	100.0	100.0	100.0		

*P < .05 indicates statistically significant difference. Same subscript numbers indicate statistically insignificant relationships among genotype groups.

patients had RR, 2 had RX, and 1 had XX genotype. In high vertical group, 14 had RR, 2 had RX, and 3 had XX genotype. In normal vertical group of patients, 7 had RR and 2 had XX. No RX genotype was detected in the normal group.

In XX genotype, 50% consisted of high-angle patients, 33.3% normal vertical, and 16.7% low-angle patients. Low- and high-angle groups shared RR genotype each 50%. RR genotype was seen 46.2% in low angles, 17.9% in normal vertical, and 35.9% in high angles.

Sagittal Facial Pattern

In the sagittal plane, 17, 15, and 17 patients had Class I, Class II, and Class III malocclusion, respectively. There was no statistically significant relationship between ACTN3 rs1815739 polymorphism and sagittal pattern using ANB or Wits appraisal $P > .05$.

Maxillary Anteroposterior Position

There was a statistically significant relationship between ACTN3 rs1815739 polymorphism and overall maxillary sagittal position $P < .05$ (Table 2). The people who had retrognathic maxilla had significant proportional difference between XX and RX, and between XX and RR. In the prognathic maxilla group, all the patients had RR genotype and 23% of all RR genotype was expressed in these 9 prognathic patients. In the normal maxillary sagittal position group, RR genotype percentage was 94.1% and 100% of RX and 83.3% of XX were represented by 4 and 5 patients out of total 23 retrognathic patients, respectively.

Maxillary Incisor Inclination

There was a statistically significant frequency distribution difference between ACTN3 rs1815739 polymorphism and incisor inclination using I-SN ($P < .05$) (Table 3). The subjects who had normal incisor angle had statistically significant proportional difference

Table 3. Genotype and Percentage Distribution of ACTN3 rs1815739 Polymorphism in Maxillary Incisor Inclination Groups

			Genotype			Total	P
			XX	RX	RR		
I-SN	Normal incisor angle	Patients	4 _a	0 _{a,b}	5 _b	9	.024*
		Percentage	44.4	0.0	55.6	100.0	
		Percentage within all patients	66.7	0.0	12.8	18.4	
	Proclined Upper Incisor	Patients	1 _a	3 _a	24 _a	28	
		Percentage	3.6	10.7	85.7	100.0	
		Percentage within all patients	16.7	75.0	61.5	57.1	
	Retroclined upper incisor	Patients	1 _a	1 _a	10 _a	12	
		Percentage	8.3	8.3	83.3	100.0	
		Percentage within all patients	16.7	25.0	25.6	24.5	
Total	Patients	6	4	39	49		
	Percentage	12.2	8.2	79.6	100.0		
	Percentage within all patients	100.0	100.0	100.0	100.0		

*P < .05 indicates the statistically significant difference.

between RX and XX ($P < .05$) and between RX and RR ($P < .05$). In the normal incisor angle group, we detected no RX genotype. Also, 66.7% had XX and 12.8% had RR genotype in this group, presenting in 4 and 5 subjects, respectively. In the proclined group, 85.7% and in the retroclined group, 83.3% of subjects had genotype RR, presenting in 24 and 10 subjects, respectively.

On the other hand, there was no statistically significant frequency distribution difference between *ACTN3* rs1815739 polymorphism and incisor inclination when UI-OP measurement was used ($P > .05$).

Mandibular Incisor Inclination

There was no statistically significant frequency distribution difference between *ACTN3* rs1815739 polymorphism and incisor inclination using IMPA or LI-OP ($P > .05$). In volunteers with normal incisor angle, all of them had RR genotype RR.

DISCUSSION

Identifying the cause and early detection of malocclusion is valuable in the effective treatment, management of malocclusions, as well as public health planning. Many researchers have pointed out to date the relationship of facial bones and facial muscles. Therefore, the aim of the present research was to investigate the impact of *ACTN3* rs1815739 polymorphism, which has an effect on the muscle performance and on the configuration of the facial bones.

In order to describe the configuration of facial pattern, cephalometric radiographs were utilized because they are inexpensive and powerful tools, readily available in the patient files, and give information about both the vertical and sagittal relationship of the facial bones. Well-established diagnostic angular measurements in orthodontics were selected to determine the skeletal malocclusion and the facial type in the present study.

Wolff's¹⁴ law points out that the internal structure and the shape of the bone is closely related to function and defines a relationship between bone shape and muscle function. Following this rule, one of the most established concepts in orthodontics is that there is an effect of facial muscles on the facial skeleton and malocclusion.^{2,3,15,16}

In previous research, it was shown that facial morphology was related to the fiber type of masticatory muscles.^{17,18} The relationship of fiber type and genetic variations were revealed by other researchers.^{4,19,20}

In the present study, when the overall vertical pattern was analyzed, there was no statistically significant relationship of *ACTN3* rs1815739 polymorphism and vertical facial pattern. Also, 50% of XX genotype consisted of high-angle patients, 33.3% normal vertical, and 16.7% were low-angle patients, which showed tendency of XX genotype to have a higher frequency of high-angle vertical pattern supporting the findings of Cunha et al.²¹ who have stated that the XX genotype is associated with dolichofacial phenotype. It was stated in the recent literature that *ACTN3* rs1815739 polymorphism resulted in a lack of *ACTN3* protein expression.

The loss of this protein has been shown to lead to smaller type II fiber diameters in masseter muscles and an increased expression of ENPP1, a negative regulator of mineralization, which was related to a small-sized mandible and reduced bone mass in the mandible.^{22,23,24} This condition was found to be similar to Class II openbite morphology in humans. Cunha et al.²¹ have also studied *ACTN3* rs678397 and rs1815739 and have concluded that polymorphisms in *ACTN3* were found to be associated with sagittal and vertical craniofacial skeletal patterns, and this could vary according to the patient's ethnicity. In our study, as 2-dimensional measurements were carried out, mandibular bone volume analysis could not be performed.

For the sagittal facial pattern, there was no statistically significant relationship between *ACTN3* rs1815739 polymorphism results and sagittal relation using ANB or Wits. Also, 35.9% and 48.7% of RR genotype consisted of Class III patients. Recent studies reported the association of XX genotype which was found with higher frequency of Class II patients.^{6,21} Zebrick et al.⁶ has studied *ACTN3* rs678397 which is a cytosine to thymine transition in the intronic region of *ACTN3*.

Rs678397 and rs1815739

Even though there was no relationship between *ACTN3* rs1815739 polymorphism and sagittal classification, there was a statistically significant relationship between *ACTN3* rs1815739 polymorphism results and overall maxillary sagittal position in our study. It was found that the prognathic maxilla group was only associated with RR genotype. For incisor inclination, there was a statistically significant relationship between *ACTN3* rs1815739 polymorphism and incisor inclination in the I-SN parameter ($P < .05$). The subjects who had normal incisor inclination had significant proportion difference between RX and XX and between RX and RR. It showed an association of XX genotype with normal incisor inclination phenotype and RR genotype with proclined upper incisor inclination phenotype. A master's thesis in 2018 has stated that the genotype for polymorphisms rs678397 and rs1815739 was associated with both weak lips and a skeletal Class II phenotype, with a protrusive maxilla, and that genotype and soft tissue were significantly associated with skeletal phenotype. There was a significant association between the position of the maxilla and the strength of the labial musculature, and associations were found between markers in gene *ACTN3* and skeletal measures.²⁵ The findings of this study, especially about lip weakness, may explain why maxilla was prognathic and upper incisors were proclined in patients with rs1815739 polymorphism in the present study.

ACTN3 rs1815739 Polymorphism

There are some limitations in our study. Although the study investigated a relatively large number of patients, the study protocol separated participants into subgroups for comparisons. This resulted in a limitation of the study for the statistical comparisons. Future studies should include larger subject numbers to further understand the relationship of genetic variants with facial phenotype.

This study used conventional lateral cephalograms to determine morphologic differences in the pattern of craniofacial skeletal

patterns. These imaging modalities are routinely utilized in radiographic evaluation of dental patients. Although cone beam computed tomography is more precise, for this study, routine and already available records were evaluated. Future studies may use 3D evaluations of the dentofacial complex to unfold relations of genetics and morphology of the face.

CONCLUSION

There was no statistically significant frequency distribution difference between *ACTN3* rs1815739 polymorphism and facial pattern except:

- Overall maxillary position measured by SNA, maxillary depth, and nasion perpendicular A:
 - The prognathic maxilla was related to RR genotype.
 - The retrognathic maxilla was found to be related to RX and XX genotypes
- Maxillary incisor inclination measured by I-SN:
 - In the normal incisor angle group, there was no RX genotype.
 - In the proclined group, 85.7% and in the retroclined group, 83.3% of subjects had genotype RR.
 - Both proclined (85.7%) and retroclined (83.3%) groups were dominated by RR genotype.

Ethics Committee Approval: Ethics committee approval was received from the Non-Interventional Ethics Committee of Üsküdar University (61351342/2019-575).

Informed Consent: Informed consent was obtained from the participants.

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REFERENCES

- Graber TM. The "three M's": muscles, malformation, and malocclusion. *Am J Orthod.* 1963;49(6):418-450. [\[CrossRef\]](#)
- Brotto M, Johnson ML. Endocrine crosstalk between muscle and bone. *Curr Osteoporos Rep.* 2014;12(2):135-141. [\[CrossRef\]](#)
- Sacomanno S, Antonini G, D'Alatri L, D'Angelantonio M, Fiorita A, Deli R. Causal relationship between malocclusion and oral muscles dysfunction: a model of approach. *Eur J Paediatr Dent.* 2012;13(4):321-323.
- Klingenberg CP, Leamy LJ, Cheverud JM. Integration and modularity of quantitative trait locus effects on geometric shape in the mouse mandible. *Genetics.* 2004;166(4):1909-1921. [\[CrossRef\]](#)
- Ulucan K, Bayyurt GM, Konuk M, Güney AI. Effect of alpha-actinin-3 gene on Turkish trained and untrained middle school children's sprinting performance: a pilot study. *Biol Rhythm Res.* 2014;45(4):509-514. [\[CrossRef\]](#)
- Zebrick B, Teeramongkolgul T, Nicot R, et al. ACTN3 R577X genotypes associate with class II and deepbite malocclusions. *Am J Orthod Dentofacial Orthop.* 2014;146(5):603-611. [\[CrossRef\]](#)
- Cunha A, Nelson-Filho P, Marañón-Vásquez G, et al. Genetic variants in ACTN3 and MYO1H are associated with sagittal and vertical craniofacial skeletal patterns. *Archives of Oral Biology.* 2018;97:85-90.
- Ulucan K. Literature review of Turkish sportsmen in terms of ACTN3 R577X polymorphism. *Clin Exp Health Sci.* 2016;6(1):44-47. [\[CrossRef\]](#)
- Eroğlu O, Zileli R, Nalbant MA, Ulucan K. Prevalence of alpha actinin-3 gene (*ACTN3*) R577X and angiotensin converting enzyme (*ACE*) insertion/deletion gene polymorphisms in national and amateur Turkish athletes. *Cell Mol Biol (Noisy-le-grand).* 2018;64(5):24-28. [\[CrossRef\]](#)
- North KN, Beggs AH. Deficiency of a skeletal muscle isoform of α -actinin (α -Actinin-3) in merosin-positive congenital muscular dystrophy. *Neuromuscul Disord.* 1996;6(4):229-235. [\[CrossRef\]](#)
- Vincent B, De Bock K, Ramaekers M, et al. ACTN3 (R577X) genotype is associated with fiber type distribution. *Physiol Genomics.* 2007;32(1):58-63. [\[CrossRef\]](#)
- Frey N, Olson EN. Calsarcin-3, a novel skeletal muscle-specific member of the calsarcin family, interacts with multiple z-disc proteins. *J Biol Chem.* 2002;277(16):13998-14004. [\[CrossRef\]](#)
- Swoap SJ, Hunter RB, Stevenson EJ, et al. The calcineurin- NFAT pathway and muscle fiber-type gene expression. *Am J Physiol Cell Physiol.* 2000;279(4):C915-C924. [\[CrossRef\]](#)
- Wolff J. The classic: on the inner architecture of bones and its importance for bone growth. *Clin Orthop Relat Res.* 2010;468(4):1056-1065. [\[CrossRef\]](#)
- Hall BK, Herring SW. Paralysis and growth of the musculoskeletal system in the embryonic chick. *J Morphol.* 1990;206(1):45-56. [\[CrossRef\]](#)
- Rot-Nikcevic I, Reddy T, Downing KJ, et al. Myf5-/-:MyoD-/- amyogenic fetuses reveal the importance of early contraction and static loading by striated muscle in mouse skeletogenesis. *Dev Genes Evol.* 2006;216(1):1-9. [\[CrossRef\]](#)
- Rowlerson A, Raoul G, Daniel Y, et al. Fiber-type differences in masseter muscle associated with different facial morphologies. *Am J Orthod Dentofacial Orthop.* 2005;127(1):37-46. [\[CrossRef\]](#)
- Sciote JJ, Horton MJ, Rowlerson AM, Ferri J, Close JM, Raoul G. Human masseter muscle fiber type properties, skeletal malocclusions, and muscle growth factor expression. *J Oral Maxillofac Surg.* 2012;70(2):440-448. [\[CrossRef\]](#)
- Konno M, Sato K, Mito T, Mitani H. Relationship between the direction of mandibular growth and masseter muscle conduction velocity. *Am J Orthod Dentofacial Orthop.* 2005;128(1):35-43. [\[CrossRef\]](#)
- Shastri BS. SNPs: impact on gene function and phenotype. *Methods Mol Biol.* 2009;578:3-22. [\[CrossRef\]](#)
- Cunha A, Nelson-Filho P, Marañón-Vásquez GA, et al. Genetic variants in ACTN3 and MYO1H are associated with sagittal and vertical craniofacial skeletal patterns. *Arch Oral Biol.* 2019;97:85-90. [\[CrossRef\]](#)
- Arino F, Sciote J, Houweling P, North K. Effects of Actn3 KO on mandibular condyle microarchitecture and bone morphology. *J Dent Res.* 2017;96A:1769.
- Yang H, Alnaqeeb M, Simpson H, Goldspink G. Changes in muscle fibre type, muscle mass and Igf-1 gene expression in rabbit skeletal muscle subjected to stretch. *J Anat.* 1997;190(4):613-622. [\[CrossRef\]](#)
- Nicot R, Chung K, Vieira AR, Raoul G, Ferri J, et al. Condyle modeling stability, craniofacial asymmetry and ACTN3 genotypes: contribution to TMD prevalence in a cohort of dentofacial deformities. *PLoS ONE. Public Library of Science.* 2020;7(7):e0236425. [\[CrossRef\]](#)
- Hansen SE. *The Influence of Genotype and Perioral Musculature on Maxillary and Mandibular Development*; 2018:25-31.