ORIGINAL ARTICLE

Estimation of dental and skeletal age in iron-deficient anemic Patients of Iraqi sample.

HEIDER HEMEED ABBAS¹, SELMA MERZA HASAN², NOOR MOHAMMED HASAN³

¹Department of Basic Medical Sciences, College of Dentistry, Babylon University¹

²Department of Orthodontics, College of Dentistry, University of Kufa²

³Department of Orthodontics, College of Dentistry, University of Baghdad³

Correspondence to Dr. Heider Hemeed Abbas, Assistant Professor, E.mail: dent.heider.hemeed@uobabylon.edu.iq hhemeed@yahoo.com

ABSTRACT

Background: The orthodontist concern with growing and non-growing individuals. Greatest of the information regarding growth are obtained from hand wrist radiograph lateral cephalogram. The relationship of different chronological, dental, and skeletal ages is significant indicator in diagnosis and treatment and the differences of dental and skeletal ages from known chronological age refer to alterations in the normal growth pattern.

Aim: To evaluate the dental and skeletal age in iron deficient anemic patient who seeking orthodontic treatment in comparison to their chronological age.

Methods: One hundred and nineteen 67 females and 52 males Participant selected randomly were aged between (8-15) year who attend these center seeking different orthodontics treatment. They were clinical examined by specialist to symptoms of anemia. The Dental age were determined according to The Demerijian method which is most widely used technique were obtained from panoramic radiographs used for pubertal growth evaluation. Bone age were determined on lateral cephalometric radiograph, which taken as a patient record, the third (C3) and fourth (C4) cervical vertebrae were traced and the subsequent parameters were measured rendering to Mito et al. (2002). **Result**: the statistical analysis of data show that there was significant difference among different age groups that's mean that the iron deficiency anemia can effect significantly on bone and dental age of patients. **Conclusion**: Dental and bone age delay was an important criteria in iron-deficient anemic patients. Dental and bone age are

considered as respected parameters in evaluating the total growth of the child.

Keywords: Chronological age, Bone age, dental age, orthodontic patients, iron-deficiency anemia.

INTRODUCTION

Growth indicatesincrease in the size of tissues, while development indicate increase inmaturation of functions¹. The orthodontist concern with growing and non-growing individuals. Greatest of the information regarding growth are obtained from hand wrist radiograph lateral cephalogram². The relationship of different chronological, dental, and skeletal ages is significant indicatorin diagnosis and treatment and the differences of dental and skeletal ages from known chronological age refer to alterations in the normal growth pattern³.

The chronological age of an individual may be enhanced of slowed due to the personal disparity of the exact time of pubertal growth spurt². For that reason, chronological age considered as undependable indicator for the determination of the child maturity⁴. Thus, physiological age was considered⁵.

The lateral cephalometric radiographs usedto determine the skeletal maturity by examining the cervical vertebrae, it has been found to be dependable and valid as the hand-wristtechnique⁶. Skeletal age is a pointer of physiological development and is different from the chronological age². Bone age indicate the overall osseous maturation of individual. The appearance and unification of various centers of calcification according tospecific order and schedule from birth to complete maturation⁷. Bone maturation in a person can be defined by using the radiographic pictures to study these centers of calcification according to the amount of maturation⁸. The bone age is, therefore important in approving a diagnosis. Bone and dental maturation is critical for dentists in framing treatment

plan, and is suitable and supportive data for several medical specialties⁹.

Dental age detection is beneficial in various clinical and scientific specialty such as orthodontics, pediatric dentistry, forensic dentistry and others¹⁰, this can be done by examine a series of identifiable events that happened in the same order from primary occasion to a finish point. Age of an unknown person can be determined by correlating theskeletal, dental, and physical maturity of a person, so measuring dental age directly is important tool to determine the chorological age of a child who has unknown date of birth¹¹. Mineralization rate of teeth is a constant event: development of individual teeth is a more desirable than teeth eruption. Numerous systems are present to detect the dental age according to the amount of calcification. Demirjian system has been widely used by many experts to detect the dental age¹². In this method tooth formation is classified in to eight stages, the left mandibular seven teeth was scored in each stage and the sum of the scores assess the subject's dental maturity and the dental age was measured according to the gender specific tables. Toward the end of dental maturation less stage contribute more;therefore, a solitary stage change can lead to a big jump in dental age¹³.

Many childhood disorders and diseases, mainly those associated with growth problems, display distinctive relation between bone and chronological age. Iron-deficiency anemia associated with poor growth level and delay in the development of humans and animals¹⁴.

The objective of this study was to evaluate the dental and skeletal age in iron deficient anemic patient who seeking orthodontic treatment in comparison to their chronological age.

PATIENTS AND METHODS

This study done on sample of Iraqi patients who attendedspecific centersseeking different orthodontic treatment. One hundred and nineteen 67 females and 52 males Participant were aged between⁸⁻¹⁵ years; they were clinical examined by specialist to symptoms of anemia. Those patients with signs and symptoms of anemia weresend to a pediatrician to confirm the diagnosis of iron-deficiency anemia through laboratory tests and clinical examination. The laboratory tests serum ferritin, transferrin, iron and hemoglobin level, as well as morphology of red blood cells (RBCs) and hemoglobin. Informed consent that contain all the information about the study introduced to the parents.

Inclusion criteria include(1) with no medical history of systemic diseases or nutritional maladies² with no absent left mandibular teeth. While exclusion subjects were (a) sever medical illness (psychiatric problems, endocrine diseases), (b) permanent teeth extraction (c) Face trauma (d) ankylosedor impacted teeth (e) congenital abnormalities (f) physically or mentally handicapped patients, and (g) sever malocclusion.

Estimation of dental age: The most common method used for children is Demirjian's method. This method centered onorthopantomograms to assessment the amount of mineralization of the tooth and the chamber shape of seven left permanent mandibular teeth. It is also appropriate for estimation the maturity of individual dental tissues with a known age whether is delayed or advanced rather than for estimation of an unknown age^{15,16,17}.

The Demerijian method(18)is most widely used technique were obtained from panoramic radiographs used for pubertal growth evaluation, Teeth on OPG are rated according to8-stage system (A to H). This depend on tooth follicle shape, pulp chamber, dentin deposit and root formation. The maturity score may be altered directly into dental age (DA), by reading on the horizontal gauge the age at which the 50% percentileachieves a given maturity score^{19,20}.

After obtaining OPG for the orthodontic patient with iron deficiency anemia, they were wisely examined under the X-ray viewer to evaluate the level of tooth calcification of seven teeth in accordance with the Demirjian system. The calcification stages of the teeth were estimated, and each tooth was labelled with a certain score as per the Demirjian table (Tables 1.1 and figure 1.1]. The summation of marks of the seven teethcalled the maturity score of person and the summation of scores which delivers an assessment of an person's dental maturity on a scale from 0 to 100were transformed into dental age according to the conversion chart. The summation of. The scores were evaluated for boys and girls separately²¹. **Bone age determination:** On lateral cephalometric radiograph, which taken as a patient record, the 3rd (C3) and 4th (C4) cervical vertebrae were traced and the subsequentparameters were measured rendering toMito et al.(22):anterior height of the vertebral body (AH), vertebralbodyheight (H), posteriorheightof the vertebral body (PH), and the length of vertebral bodyantero posteriorly (AP).In order to evaluate the measurement, following formulas are achieved to obtain cervical vertebral bone age by using C3 and C4 measurements (23):

Cervical vertebral bone age for male = $1.775 + 13.557 \times$ AH3/AP3 + $6.808 \times$ H4/AP4

Cervical vertebral bone age for female = $1.582+7.920 \times AH3/AP3 + 10.110 \times AH4/AP4$.

Table 1:	Determining	the	dental	developing	stages	according	to	the
biological	criteria from A	to ⊢	l.(18)					

Stage	Description
A	Establishment of calcification can be seen at the superior level of the crypt, in the form of an inverted cone or cones. In both uniradicular and multiradicular teeth and there is no union of these calcified plugs.
В	At this level, the union of the calcified points forms one or numerous cusps, which fuse to form occlusal surface with regularly outline.
С	 a- Formation Enamel is finished at the occlusal surface. Its addition and convergence toward the cervical region. b- The establishment of a dentinal deposit can be seen. c- At the occlusal border, The pulp chamber outlines has a curved shape.
D	a- Complete formation of the crown to the cementoenamel junction. b- In the pulp chamber, the superior border in single root teeth has a clear curved form, start concave to the cervical area. The outline of the projection of pulp horns, seem like an umbrella top if present. The pulp chamber has a trapezoidal form in molars. c- Starting of root formation canseen in the shape of a spicule.
E	In single root teeth: a. The outlines of the pulp chamber give straight lines, its continuity is interrupted by the existence of the pulp horn, that's larger than in the earlier stage. b. The length of the root is a smaller amount than the crown height. Molars: a. Primary formation of the radicular bifurcation is apparent in the form of either a semilunar shape or a calcified point. b. The length of the root is quiet less than the crown height.
F	Uniradicular teeth: a. The pulp chamber wallsnow give a more or less isosceles triangle and funnel shape at the apex ends. b. Thelength of root is equivalent to or larger than the crown height. Molars: a. The calcified area of the bifurcation has progressed further down from its semilunar stage to forma more distinct outlines ofthe roots that have funnelshaped endings. b. The length of the root is identical to or larger than the height of the crown.
G	a- Parallel walls of the root canals (distal root in molars). b- The ends of the root canals apically are still partly open (distal root in molars).

Table 2: Represent the value of each stage of tooth development according to Demirjian method:

Tooth		Stage							
Boys	0	А	В	С	D	Е	F	G	Н
M2	0.0	1.7	3.1	5.4	8.6	11.4	12.4	12.8	13.6
M1				0.0	5.3	7.5	10.3	13.9	16.8
PM2	0.0	1.5	2.7	5.2	8.0	10.8	12.0	12.5	13.2
PM1		0.0	4.0	6.3	9.4	13.2	14.9	15.5	16.1
С				0.0	4.0	7.8	10.1	11.4	12.0
LI				0.0	2.8	5.4	7.7	10.5	13.2
CI				0.0	4.3	6.3	8.2	11.2	5.1
Girls									
M2	0.0	1.8 3	3.1	5.4	9.0	11.7	12.8	13.2	13.8
M1				0.0	3.5	5.6	8.4	12.5	15.4
PM2	0.0	1.7	2.9	5.4	8.6	11.1	12.3	12.8	13.3
PM1		0.0	3.1	5.2	8.8	12.6	14.3	14.9	15.5
С				0.0	3.7	7.3	10.0	11.8	12.5
LI				0.0	2.8	5.3	8.1	11.2	13.8
CI				0.0	4.4	6.3	8.5	12.0	15.8



Figure 1: The stages of tooth development.

RESULT

SPSS version 21 was used to evaluate differences between studied age groups via ONE WAY ANOVA test; differences within groups were inspected by using multiple comparison method (L.S.D.), which represented in the comparison table as small letters.

Table 1 explain the distribution of our sample according to the gender, female represent 67 patients which form about 56.3% from the total sample, while 52 patients were male who form about 43.6 from the total sample.

In table 2 and figure 1 show that there was significant variance among different age groups of male, there was significant difference between dental age and chronological and significant difference between chronological and bone age and the dental age differ significantly from bone age.

In table 3 and figure 2 show that there was significant difference among different age groups of female, there was significant difference between dental age and chronological and significant difference between chronological and bone age and the dental age differ significantly from bone age.

Table 3: Gender distribution of the patients.

Gender	n	%age		
Female	67	56.302		
Male	52	43.697		
Total	119	100		

Table	2:	Represent	the	mean	and	standard	deviation	of	male
param	ete	rs.							

Parameters	Mean±SD	P value	
chronologic age	10.31±1.907		
bone age	9.053±1.618a	<0.01**	
Dental age	7.951±1.325ab		

a=differ significantly from chronologic age, b= differ significantly frombone age.





Table 3: Represent the mean and standard deviation of female parameters.

Parameters	Mean±SD	P value
chronologic age	10.63±2.272	
bone age	9.142±1.869a	<0.01**
Dental age	8.015±1.553ab	

a=differ significantly from chronologic age, b= differ significantly frombone age.

Figure 2: Distribution of the different ages of female in years.



Table 4: Represent the difference between male and female parameters.

Deremetere	Ger	Byoluo	
Farameters	Male	Female	Fvalue
chronologic age	10.31±1.907	10.63±2.272	0.420ns
bone age	9.053±1.618	9.142±1.869	0.786ns
dental age	7 951+1 325	8 015+1 553	0.814ns

Data were represented as mean \pm SD, ns=No significant differences at P \leq 0.05

Figure 3, 4, 5: Comparisons of age in respect to gender were done by using unpaired (independent) t-test. No significant differences were observed between male and female all studied parameters.













DISCUSSION

Bone and dental age estimation permanently take up larger importance in clinical Dentistry and Medicine and in many social events such as marriage, start a job, a birth documentation, finding criminal and legal accountability, and others. It represent an important measurement to determine the whole growth and development of persons. Dental and skeletal age are very valuable measurements to detect the general maturation of an individual¹³.

Many systemic disorders influence the growth rate in children. Iron-deficiency anemia is consider one of the systemic disorders, which result in growth retardation in children. Bandhu *et a*^{P^4} suggest that iron-deficiency anemia in children caused momentous mental and physical growth failure. Aukett *et a*^{P^5} discoverd that growth failure can be inverted in children with iron-deficiency anemia, while Moffatt *et a*^{P^6} approved that treatment for children with iron-deficiency will convey the psychomotor development. Knowing the growth of the orofacial area is significant when design orthodontic treatment because it assist the orthodontic to determine the type of orthodontic appliance which used to correct different types of maloccusion.

Dental age can determined by Demirjian's method using dental radiograph on it we can determine the stage of dental mineralization. We used Demirjian's method in our examination because many studies by Hagg and Matsson²⁷, Nanda and Chawla²⁸, and Hegde and Sood²⁹ recommended that the Demirjian method provide more accurate and larger reliability for dental age estimation. Dental mineralization is more dependable to calculate dental age when compared to dental eruption method. Tooth mineralization is a constant route, which can be detected by radiographs whereas tooth eruption is a changeable event where its precise time is challenging to analyze¹². Emerging teeth in orthopantamograph are suitable method because a single radiograph gives the developmental condition of teeth in children, so it always used to evaluate dental maturity and estimation age. Age evaluation has become progressively important to detect the age of individuals. Digital OPGs were used as the images can be magnified to make analysis easier¹³.

Skeletal maturation is a basic portion of individual forms of growth and development. Difference in the maturation station is highly associated with variation in the scheduling and amount of growth. As a result to individual dissimilarity in duration, timing, and speed of growth, skeletal age calculation is important in framing practicable orthodontic treatment procedures³⁰.

The patient population age groups presence the orthodontic clinic aged between 8 to 15 years. Below age eight, lateral cephalometric radiographs were infrequently taken as dental records, also children before this age is difficult to implement in practical terms (31). Clinical choices concerning the use of functional forces and appliances, extraction against nonextraction treatment or orthognathic surgery are as minimum as depend on growth concerns³². Every growing child develops in specific manner and numerous means are offer as indicators to assess the maturity. These are the chronological age⁸ dental age^{33,34, 35,36} Hand and wrist^{37,38} also the body mass index^{39,12} sexual maturation; Cervical vertebrae² and the

Frontal sinus^{2,40} and newly biomarkers⁴¹. Bacettee et al⁴² and other researchers^{2,43,44} introduced the cervical vertebral maturation method. In our study we estimate the bone age depending on cervical vertebral from lateral cephalometric radiograph to avoid exposing the patient to extra radiation that associated with hand-wrist radiograph. The cervical vertebrae can be used to measure personal skeletal maturation is achievement that mentioned in many literature^{45,46,47}. The predictable cervical vertebral maturation process for measuring of skeletal maturation is depend on personalassessment of the form and proportions of cervical vertebrae⁴⁰. The determination of cervical vertebral bone age suggest better benefit of accurately estimating the osseous maturation from lateral cephalometric radiographs by calculating the dimensional considerations of C3 and C422, also this method is more dependable and precise than the use of chronological age in evaluating an individual's advancement to maturity⁴⁸.

In this study we depend on Mito method²², which are of large significance because it permits skeletal age to be considered in an objective way. It is more popularity in latest year; it is depend on the morphological features of the cervical vertebrae at different stages of development. These stages are characterized by different growth degrees in facial configurations. Furthermore, this CVM procedure cover the whole circum pubertal period for male and female by covering all important stages in craniofacial maturation through adolescence and young adulthood. In this method, C3 and C4 were chose for estimation due to the difficulty in detecting and assessing morphological body variations in the first two vertebrae and the usual absence of the lower cervical vertebrae in predictable lateral cephalometric radiographs⁴⁹. Mito et al. observed only Japanese female due to sex-dependent variation with respect to the timing of morphological deviations in cervical vertebral dimevntions⁵⁰. This was in agreement with Alhadlaq and Al-Maflehi⁵¹, who consider Saudi male only to prevent any sex-related disparities in timing of maturational changes and growth pattern of the cervical vertebrae. In this study, we use this method for both boys and girls to assess the vertebral body dimensions but the measurement were compared separately for both genders.

The results of our study represent that there were a significant difference between skeletal age (by cervical vertebral measurement using Mito method) and the chronological age (according to the birthday), and also significant difference between dental age (determined by Demirjian's method by using orthopantamograph) and chronological age and also significant difference between dental and bone age. This result mean that the bone and dental age are significantly affected by iron deficiency anemia, this may attributed to that Iron is one of the vital elements of life and Iron metabolism is associated to bone metabolism⁵². These result was in agreement with Vinod et al²⁰, who stated that Dental and bone age was significantly inferior in comparison to chronological age and the relation among the three ages were positive in both genders. While study by Mei-Lien Panetal⁵³ shown that patients with a history of iron deficiency anemia had a higher occurrence of osteoporosis in comparison to individuals without iron deficiency anemia, by about two-times risk for osteoporosis. The causes of iron deficiency anemia include infections, malnutrition, vegetarian diets, GI tract diseases, and acute or chronic blood loss, or other etiologies⁵⁴. All these causes may associated with retardation in bone formation.

The result showed that the chronological, dental, and bone ages were definitely correlated. These findings are supported by related studies such as those of Vallejo-Bolanos *et al*⁹) Green⁸, Hegde and Sood²⁹ and Prabhakar *et al*¹². Also many earlier studies haveconfirmed that different growth maturation systems display positive association.

REFERENCES

- 1. Ghai OP, Gupta P, Paul VK. Essential Pediatrics. 6th ed. New Delhi: CBS Publisher; 2004.
- Hayder A. Hashim, Hussain Mansoor, Mohamed H. Hashim Mohamed. Assessment of Skeletal Age Using Hand- Wrist Radiographs following Bjork System, 2019
- Bala M, Pathak A, Jain RL. Assessment of skeletal age using MP3 and hand- wrist radiographs and its correlation with dental and chronological ages in children. J Indian SocPedodPrev Dent 2010;28:95- 9.
- Macha M, Lamba B, Avula JS, Muthineni S, Margana PG, Chitoori P, et al. Estimation of correlation between chronological age, skeletal age and dental age in children – A cross- sectional study. J ClinDiagn Res 2017;11:ZC01- 4.
- Baccetti T, Franchi L, De Toffol L, Ghiozzi B, Cozza P. The diagnostic performance of chronologic age in the assessment of skeletal maturity. ProgOrthod 2006;7:176-88.
- Hassel B, Farman AG. Skeletal maturation evaluation using cervical vertebrae. Am J OrthodDentofacialOrthop 1995;107:58- 66.
- Boeyer ME, Sherwood RJ, Deroche CB, Duren DL. Early maturity as the new normal: A century- long study of bone age. ClinOrthopRelat Res 2018.
- 8. Green LJ. Interrelationship among height, weight and chronological, dental and skeletal age. Angle Orthod. 1961;31:189–93.
- Vallejo-Bolanos E, Espana-Lopez AJ. The relationship between dental age, bone age and chronological age in 54 children with short familial stature. Int J Paediatr Dent. 1997;7:15–7.
- Bagic IC, Sever N, Brkic H, Kern J. Dental age estimation in children using orthopantomograms. Acta Stomatol Croat. 2008;42:11–8.
- 11. Lewis AB, Garn SM. The relationship between tooth formation and other maturational factors. Angle Orthod. 1960;30:70–7.
- Prabhakar AR, Panda AK, Raju OS. Applicability of Demirjian's method of age assessment in children of Davangere. J Indian SocPedodPrev Dent. 2002;20:54–62.
- Rezwana Begum Mohammed, P. V. Krishnamraju, 1 P. S. Prasanth,2 Praveen Sanghvi,3 M. Asha Lata Reddy,4 and S. Jyotsna5 Dental age estimation using Willems method: A digital orthopantomographic study. Contemp Clin Dent. 2014 Jul-Sep; 5(3): 371–376.
- 14. Beard JL, Zhan CS, Brigham DE. Growth in iron-deficient rats. ProcSocExpBiol Med. 1995;209:65–72.
- Koshy S, Tandon S. Dental age assessment: The applicability of Demirjian's method in south Indian children. Forensic Sci Int. 1998;94:73–85.
- Liversidge HM, Speechly T, Hector MP. Dental maturation in British children: Are Demirijian's standards applicable? Int J Paediatr Dent. 1999;9:263–9.
- 17. Eid RM, Simi R, Friggi MN, Fisberg M. Assessment of dental maturity of Brazilian children aged 6 to 14 years using Demirjian's method. Int J Paediatr Dent. 2002;12:423–8.
- Demirjian A, Goldstein H, Tanner JM. A new system of dental age assessment: Human biology. Hum Biol. 1973;45:211–27.

- 19. Samuel P. Tam. Craniofacial differences between obese and non-obese orthodontic patients by D.M.D. 2019.
- Vinod Kumar, Harish Haridas,1 Prahlad Hunsigi,2 Umar Farooq,3 Sridhar R. Erugula,4 and Kranti K. R. Ealla4. Evaluation of dental and bone age in iron-deficient anemic children of South India J IntSocPrev Community Dent. 2016 6(5): 430–435.
- Ali I. Al-Bustani B.D.S., M.Sc. (1)Application of periapical radiographic view of 4mandibular permanent teeth in orthodontic diagnosis. J Bagh College Dentistry, 2011,Vol. 23(1).
- 22. Mito T, Sato K, Mitani H. Cervical vertebral bone age in girls. Am J OrthodDentofacialOrthop 2002;122:380–5.
- 23. Dr. AfafAbd El- Raouf Shaaban1and Prof. Ossama Sayed Ahmed El-Shall2. Age Estimation Based on Some Cervical Vertebral Measurementsin a Sample of Egyptian Children, Ain Shams Journal of Forensic Medicine and Clinical Toxicology Jan 2017, 28: 72 -87.
- Bandhu R, Shanker N, Tandon OP. Effect of iron on growth in iron deficient anemic school going children. Indian J PhysiolPharmacol. 2003;47:59–66.
- Aukett MA, Parkes YA, Scott PH, Wharton BA. Treatment with iron increases weight gain in psychomotor development. Arch Dis Child. 1986;61:849–57.
- Moffatt ME, Longstaffe S, Besant J, Dureski C. Prevention of iron deficiency and psychomotor decline in high risk infants through use of iron-fortified infant formula: A randomized clinical trial. J Pediatr. 1994;125:527–34.
- Hagg U, Matsson L. Dental maturity as an indicator of chronological age: The accuracy and precision of three methods. Eur J Orthod. 1985;7:25–34.
- Nanda RS, Chawla TN. Growth and development of dentitions in Indian children. I. Development of permanent teeth. Am J Orthod. 1966;52:837–53.
- 29. Hegde RJ, Sood PB. Dental maturity as an indicator of chronological age: Radiographic evaluation of dental age in 6 to 13 years children of Belgaum using Demirjian method. J Indian SocPedodPrev Dent. 2002 Dec;20:132–8.
- Flieger R, Matys J, Dominiak M. The best time for orthodontic treatment for Polish children based on skeletal age analysis in accordance to refund policy of the Polish National Health Fund (NFZ). AdvClinExp Med 2018;27:1-6.
- 31. Al-EmranS : Dental Age Assessment of 8.5 to 17 Year-old Saudi Children Using Demirjian's Method. J. Contemp. Dent.Pract.; 2008. 3 (9): 64-71.
- 32. De Stefani A, Bruno G, Siviero L, Crivellin G, Mazzoleni S, Gracco A. Middle phalanx maturation of the third finger (MPM) method in the evaluation of skeletal age in a growing orthodontic patient. Int Orthod 2018;16:499-513.
- Nolla CM. The Development of Permanent Teeth. In: University of Michigan; 1952.
- Demisch A, Wartmann P. Calcification of the mandibular third molar and its relation to skeletal and chronological age in children. Child Dev 1956;27:459-73.
- 35. Demirjian A, Goldstein H, Tanner JM. A new system of dental age assessment. Hum Biol 1973;45:211-27.
- 36. Demirjian A, Goldstein H. New systems for dental maturity based onseven and four teeth. Ann Hum Biol 1976;3:411-21.

- 37. Björk A, Helm S. Prediction of the age of maximum puberal growthin body height. Angle Orthod 1967;37:134-43.
- Hägg U, Taranger J. Skeletal stages of the hand and wrist asindicators of the pubertal growth spurt. ActaOdontolScand1980;38:187- 200.
- 39. Hunter CJ. The correlation of facial growth with body height and skeletal maturation at adolescence. Angle Orthod 1966;36:44-54.
- Baccetti T, Franchi L, McNamara Jr JA. The cervical vertebral maturation (CVM) method for the assessment of optimal treatment timing in dentofacial orthopedics. SeminOrthod 2005;11:119–29.
- Perinetti G, Baccetti T, Di Leonardo B, Di Lenarda R, Contardo L. Dentition phase and chronological age in relation to gingival crevicular fluid alkaline phosphatase activity in growing subjects. ProgOrthod 2011;12:100-6.
- Baccetti T, Franchi L, McNamara JA Jr. An improved version of the cervical vertebral maturation (CVM) method for the assessment of mandibular growth. Angle Orthod 2002;72:316-23.
- 43. Soegiharto BM, Moles DR, Cunningham SJ. Discriminatory ability of the skeletal maturation index and the cervical vertebrae maturation index in detecting peak pubertal growth in indonesian and white subjects with receiver operating characteristics analysis. Am J Orthod DentofacialOrthop 2008;134:227-37.
- 44. Caldas Mde P, Ambrosano GM, HaiterNeto F. Computer assisted analysis of cervical vertebral bone age using cephalometric radiographs in Brazilian subjects. Braz Oral Res 2010;24:120-6.
- 45. Gandini P, Mancini M, Andreani F. A comparison of hand-wrist bone and cervical vertebral analyses in measuring skeletal maturation. Angle Orthod 2006;76:984–9.
- 46. Chen L, Liu J, Xu T, Long X, Lin J. Quantitative skeletal evaluation based on cervical vertebral maturation: a longitudinal study of adolescents with normal occlusion. Int J Oral Maxillofac Surg 2010;39:653–9.
- Fudalej P, Bollen AM. Effectiveness of the cervical vertebral maturation method to predict postpeak circumpubertal growth of craniofacial structures. Am J Orthod Dentofacial Orthop 2010;137:59–65.
- Schmeling A, Reisinger W, Loreck D et al., Effects of ethnicity on skeletal maturation: consequences for forensic age estimations. Int. J. Legal Med.; 2000, 113(5): 253-8.
- San-Roman RP, Palma JC, Oteo MD and Nevado E: Skeletal maturation determined by cervical vertebrae development. Eur. J.Orthod. 2002;24:303–11.
- 50. Beker L: Principles of growth assessment. Pediatr. Rev. 2006;27:196–197.
- Alhadlaq AM and Al-Maflehi NS (2013):New model for cervical vertebral bone age estimation in boys. King Saud Univ. J. Dent. Science; 4: 1–5.
- LinBoaZhichunLiuaYingbinZhongbJianHuangbBinChencHanW angbYoujiaXuc. Iron deficiency anemia's effect on bone formation in zebrafish mutant, 2016.
- Camaschella C. Iron-deficiency anemia. N. Engl. J. Med. 2015;372:1832–1843.