ISSN Print: 2394-7489
ISSN Online: 2394-7497
IJADS 2022; 8(1): 382-387

www.oraljournal.com
Received: 05-12-2021
Accepted: 12-01-2022

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Evaluation of skeletal maturation based on permanent mandibular second molar calcification

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DOI: https://doi.org/10.22271/oral.2022.v8.i1f.1453

Abstract
Objective: To investigate the relationship between permanent mandibular second molar calcification stages and cervical vertebrae skeletal maturity and to evaluate whether second molar calcification stages can be used as a reliable diagnostic tool to determine skeletal maturity.
Materials and Methods: The Panoramic and intraoral periapical radiographs and lateral cephalogram of 240 subjects (120 males and 120 females) with age group between 8 to 17 years were examined. The Calcification stages of mandibular second molar were rated according to Demirjian index (DI) and skeletal maturity were determined using Cervical vertebrae maturation index (CVMI) given by Hassel and Farman.
Results: A highly significant association was found between CVMI and DI. DI stage E corresponded to stage 2 of CVMI (pre-pubertal growth spurt). DI stages F and G corresponded to stage 3 and 4 of CVMI (pubertal growth spurt). DI Stage H corresponded to CVMI stage 5 and 6 (end of pubertal growth spurt).
Conclusion: A highly significant association exists between DI and CVMI. Mandibular second molar DI stages can be used as a reliable indicator for growth evaluation in clinical practice.

Keywords: Skeletal maturation, mandibular second molar, Demirjian index, cervical vertebrae maturation indicators

1. Introduction
The growth is a critical variable in diagnosis and treatment planning especially in the field of orthodontics [1]. The human growth is very complex in nature being influenced by multiple factors like hereditary, functional, environmental, sexual, nutritional and metabolic factors [1, 2]. The strong relationship between pubertal growth and acceleration in the growth of the craniofacial skeleton have been suggested by orthodontists [3].
The assessment of maturational status can have a considerable influence on diagnosis, treatment planning and the eventual outcome of orthodontic treatment [4, 5]. The considerable variation in development among children of the same chronological age have led to the concept of biological or physiological age. Physiological age is the rate of progress towards maturity that can be estimated by somatic, sexual, skeletal and dental maturity [4, 6].
The somatic maturity is recognized by the annual growth increments in height or weight. The changes of secondary sexual characteristics, voice changes in boys and menarche in girls are characterized as sexual maturity [4, 7]. The usefulness of two maturity indicators has limited value because these can be applied only after the serial recording of height or inception of puberty [4].
The technique for assessing skeletal maturity consists of a visual inspection of the developing bones, including their initial appearance and their subsequent ossification related changes in shape and size. In the orthodontic field, the hand-wrist and the cervical vertebrae radiographs are commonly used for skeletal assessment [4, 6]. Hassel and Farman [8] suggested six stages of skeletal maturity based on the shapes of cervical vertebrae (C2, C3, C4) on lateral cephalogram.
The dental maturity can be determined by the stage of tooth eruption or the stage of tooth formation. The tooth formation is proposed as a more reliable criterion for determining dental maturation [4, 9].

For the evaluation of tooth mineralization, classification stages proposed by Demirjian [9] appears to be best suited since stages are defined by changes in form and independent of possibly speculative length estimates.

The correlation between the calcification stages of individual teeth and skeletal maturity has been reported previously [10,11] It was reported that the stages of mandibular second molar calcification showed the highest correlation with the stages of skeletal maturity as compared to other teeth [4, 6, 10]. Previous studies have also suggested the racial variations in their relationship. Mappes *et al*. [12] indicated that the predominant ethnic origin of the population, climate, nutrition, socioeconomic levels and urbanization are causative factors of these racial variations.

The ease of recognizing dental developmental stages, together with the availability of intraoral periapical or panoramic radiographs in most orthodontic dental practices, are practical reasons for attempting to assess physiologic maturity without resorting to hand wrist radiographs [4, 5]. In addition, radiation exposure is high when specialized radiographs like lateral cephalogram and hand wrist radiograph are used making their use questionable according to ALARA principle.

2. Materials and methods

Lateral cephalogram, orthopantomogram (OPG) and intraoral periapical (IOPA) radiographs of 240 subjects (120 males & 120 females) of age group between 8 to 17 years were obtained. There are 10 groups of 24 subjects in each group and each age group contains 12 males and 12 females. The inclusion criteria includes normal overall growth and development of the individual, absence of abnormal dental conditions such as impactions, transposition and congenitally missing tooth, absence of previous history of trauma or diseases related to face, no previous history of orthodontic treatment and permanent teeth extraction.

2.1 Evaluation of Dental Maturity on Panoramic and Introral Periapical Radiograph

In this study, mandibular left permanent second molar was used as a sample. The dental maturity was evaluated based on mandibular second molar calcification stages according to Demirjian index in which one of the eight stages of calcification (A to H) were assigned to the tooth.

2.1.1 Dental calcification stages using Demirjian Index (DI)

Stage A: Calcification of single occlusal points without fusion of different calcifications

Stage B: Fusion of the mineralization points; the contour of the occlusal surface is recognizable.

Stage C: Enamel formation has been completed at the occlusal surface, and dentin formation has commenced. The pulp chamber is curved and no pulp horns are visible.

Stage D: Crown formation has been completed to the level of the cementoenamel junction. Root formation has commenced. The pulp horns are beginning to differentiate, but the walls of the pulp chamber remain curved.

Stage E: The root length remains shorter than the crown height. The walls of the pulp chamber are straight, and the pulp horns have become more differentiated than in the previous stage. In molars, the radicular bifurcation has commenced to calcify.

Stage F: The walls of the pulp chamber now form an isosceles triangle and the root length is equal to or greater than the crown height. In molars, the bifurcation has developed sufficiently to give the roots a distinct form.

Stage G: The walls of the root canal are now parallel, but the apical end is partially open. In molars, only the distal root is rated.

Stage H: The root apex is completely closed (distal root in molars).The periodontal membrane surrounding the root and apex is uniform in width throughout the root length.

2.2 Evaluation of Cervical Vertebrae Maturity on Lateral Cephalogram

The cervical vertebrae maturity was evaluated by classifying C2, C3, C4 into six groups depending on their maturation patterns using the classification of Hassel and Farman.

2.2.1 Cervical Vertebrae Maturation Indicators (CVMI) by Hassel and Farman

Stage 1 (Initiation)

Very significant amount of adolescent growth expected (80% to 100%). C2, C3 and C4 inferior vertebral body borders are flat, vertebrae are wedge-shaped and superior vertebral borders are tapered posterior to anterior.

Stage 2 (Acceleration)

Significant amount of adolescent growth expected (65% to 85%). Concavities are developing in the inferior borders of C2 and C3. The inferior border of C4 is flat. The bodies of C3 and C4 are nearly rectangular in shape.

Stage 3 (Transition)

Moderate amount of adolescent growth expected (25% to 65%). Distinct concavities are seen in the inferior borders of C2 and C3. A concavity is beginning to develop in the inferior border of C4. The bodies of C3 and C4 are rectangular in shape.

Stage 4 (Deceleration)

Deceleration of adolescent growth spurt. Small amount of adolescent growth expected (10% to 25%). Distinct concavities in the inferior borders of C2, C3 and C4. C3 and C4 are nearly square in shape.

Stage 5 (Maturation)

Final maturation of the vertebrae takes place during this stage. Insignificant amount of adolescent growth expected (5% to 10%). Accentuated concavities of inferior vertebral body borders of C2, C3 and C4. C3 and C4 are square in shape.

Stage 6 (Completion)

Adolescent growth is completed (little or no growth expected). Deep concavities are seen in inferior border of C2, C3 and C4. C3 and C4 heights are greater than widths.

3. Results

The data was analyzed with SPSS software version 16. Descriptive statistics were obtained by determining the mean and standard deviation of the chronological ages for the six stages of CVMI. Chi Square test and Pearson contingency coefficient were estimated to determine the relationships between DI and CVMI.

Table 1 shows the distribution of average chronological ages for both male and female subjects grouped by CVMI staging. Each CVMI stage appeared earlier in female subjects than in the male subjects except in CVMI stage 6 where similar age is seen in both genders.

Table 2 and graph 1 shows correlation between CVMI and DI for overall subjects. The Chi square test value of 464.735 with Pearson contingency coefficient of 0.812 (P value < 0.01) indicated a highly significant association between CVMI and DI for overall subjects. Stage D and E of DI showed high distribution of 53.1% and 42.9% in CVMI stage 1. Stage E showed high distribution of 60% in CVMI stage 2. Stage F and G showed high distribution of 40% and 58.3% in CVMI stage 3. Stage G showed high distribution of 81.8% in CVMI stage 4. Stage H showed high distribution in both stage 5 (97.6%) and stage 6 (100%) of CVMI.

Table 3 and graph 2 shows correlation between CVMI and DI for male subjects. The Chi square test value of 228.328 with Pearson contingency coefficient of 0.810 (P value < 0.01) indicated a highly significant association between CVMI and DI for male subjects. Stage D and E of DI showed high distribution of 50% and 46.2% in CVMI stage 1. Stage E showed high distribution of 56.5% in CVMI stage 2. Stage G showed high distribution of 65.7% in stage 3 and 78.6% in stage 4 of CVMI. Stage H showed high distribution of 100% in both stage 5 and 6 of CVMI.

Table 4 and graph 3 shows correlation between CVMI and DI for female subjects. The Chi square test value of 244.647 with Pearson contingency coefficient of 0.819 (P value < 0.01) indicated a highly significant association between CVMI and DI for female subjects. Stage D of DI showed high distribution of 56.5% in CVMI stage 1. Stage E showed high distribution of 63.6% in CVMI stage 2. Stage F and stage G showed high distribution of 52% and 48% in CVMI stage 3. Stage G showed high distribution of 84.2% in CVMI stage 4. Stage H showed high distribution of 96% in stage 5 and 100% in stage 6 of CVMI.

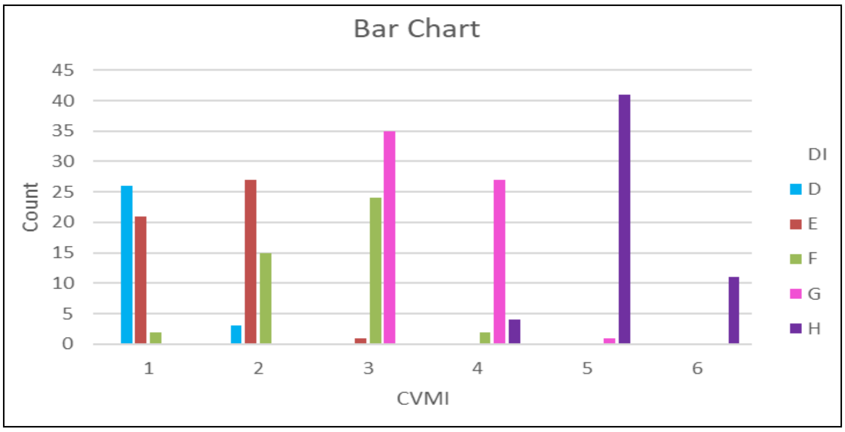
Table 1: Distribution of mean chronological age for all subjects grouped by CVMI stages

Gender and CVMI Staging		No. of Subjects	Mean Age in years	Standard deviation	Standard error
Males	Stage 1	26	8.769	0.815	0.160
	Stage 2	23	10.739	1.096	0.229
	Stage 3	35	12.886	1.231	0.208
	Stage 4	14	15.357	0.842	0.225
	Stage 5	17	16.118	0.781	0.189
	Stage 6	5	17.000	0.000	0.000
	Total	120	12.500	2.884	0.263
Females	Stage 1	23	8.652	0.647	0.135
	Stage 2	22	10.364	1.049	0.224
	Stage 3	25	12.200	1.041	0.208
	Stage 4	19	14.211	0.918	0.211
	Stage 5	25	15.840	0.850	0.170
	Stage 6	6	17.000	0.000	0.000
	Total	120	12.500	2.884	0.263

Table 2: Correlation between CVMI and DI for overall subjects

CVMI staging		DI staging					Total
		D	E	F	G	H	
1	n	26	21	2	0	0	49
	%	53.1	42.9	4.1	0	0	100
2	n	3	27	15	0	0	45
	%	6.7	60	33.3	0	0	100
3	n	0	1	24	35	0	60
	%	0	1.7	40	58.3	0	100
4	n	0	0	2	27	4	33
	%	0	0	6.1	81.8	12.1	100
5	n	0	0	0	1	41	42
	%	0	0	0	2.4	97.6	100
6	n	0	0	0	0	11	11
	%	0	0	0	0	100	100
Total	n	29	49	43	63	56	240
	%	12.1	20.4	17.9	26.3	23.3	100

Chi square value = 446.735, Pearson contingency coefficient = 0.812; *p*< 0.01 (highly significant)

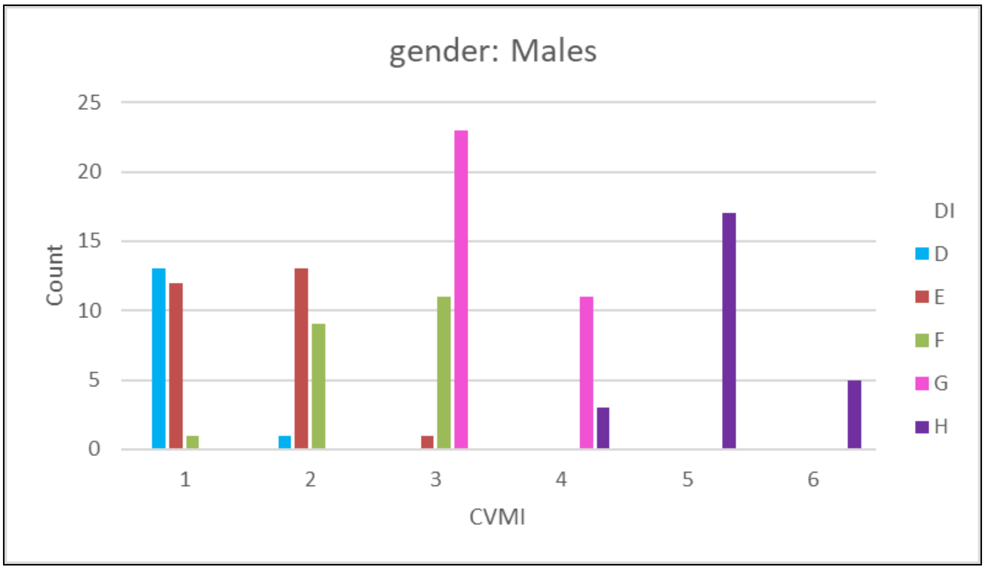


Graph 1: Correlation between CVMI and DI for overall subjects

Table 3: Correlation between CVMI and DI for male subjects

CVMI staging		DI staging					Total
		D	E	F	G	H	
1	n	13	12	1	0	0	26
	%	50	46.2	3.8	0	0	100
2	n	1	13	9	0	0	23
	%	4.3	56.5	39.1	0	0	100
3	n	0	1	11	23	0	35
	%	0	2.9	31.4	65.7	0	100
4	n	0	0	0	11	3	14
	%	0	0	0	78.6	21.4	100
5	n	0	0	0	0	17	17
	%	0	0	0	0	100	100
6	n	0	0	0	0	5	5
	%	0	0	0	0	100	100
Total	n	14	26	21	34	25	120
	%	11.7	21.7	17.5	28.3	20.8	100

Chi square value = 228.328, Pearson contingency coefficient = 0.810; $p < 0.01$ (highly significant)

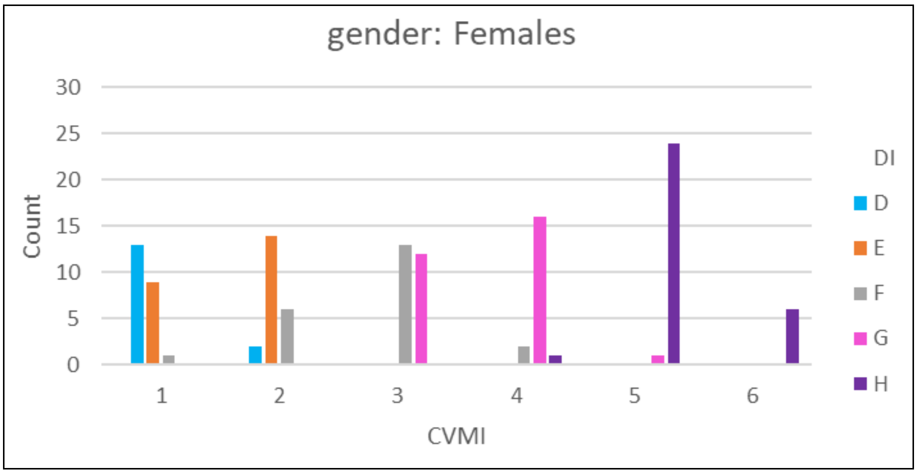


Graph 2: Correlation between CVMI and DI for male subjects

Table 4: Correlation between CVMI and DI for female subjects

CVMI staging		DI staging					Total
		D	E	F	G	H	
1	n	13	9	1	0	0	23
	%	56.5	39.1	4.3	0	0	100
2	n	2	14	6	0	0	22
	%	9.1	63.6	27.3	0	0	100
3	n	0	0	13	12	0	25
	%	0	0	52	48	0	100
4	n	0	0	2	16	1	19
	%	0	0	10.5	84.2	5.3	100
5	n	0	0	0	1	24	25
	%	0	0	0	4	96	100
6	n	0	0	0	0	6	5
	%	0	0	0	0	100	100
Total	n	15	23	22	29	31	120
	%	112.5	19.2	18.3	24.2	25.8	100

Chi square value = 244.647, Pearson contingency coefficient = 0.819; $p < 0.01$ (highly significant)



Graph 3: Correlation between CVMI and DI for female subjects

4. Discussion

Many studies have reported high correlation between tooth calcification stages and skeletal maturity indicators, which would allow the clinicians to more easily identify pubertal growth stages from panoramic or intraoral periapical radiographs [4, 10, 13-15].

Most of the studies of dentition have used either mandibular canines [11, 15] or third molars [16, 17] for dental age assessment, but these two parameters exhibit some drawbacks. Apex closure of mandibular canine occurs by the age of 13 years, whereas growth in most of the children extends upto the age of 16–17 years. On the other hand, third molars are the most commonly missing teeth in human dentition, which makes it an unreliable parameter for maturity assessment. Therefore, the mandibular second molar was used for the assessment of dental maturity because this tooth offers an advantage over other teeth which tends to continue its development over a longer period and until a later age. Apex closure generally extends up to the age of 16 years in normal children.

Sushil kumar *et al.* [6] and Chamania *et al.* [5] reported a highly significant correlation between mandibular second molar calcification stages and skeletal maturity. Each CVMI stage appeared earlier in females compared to males. Stage E of DI corresponds to stage 2 of CVMI which signifies pre-peak pubertal growth spurt. Stage F and G of DI corresponds to stage 3 and 4 of CVMI which represents the peak pubertal growth spurt. Stage H of DI corresponds to stage 5 and 6 of CVMI which is similar to the present study.

Bhardwaj *et al.* [18] found statistically significant correlation between DI stages and CVMI staging. Males tend to have a more advanced dental maturity compared to females considering the same CVMI stage. Males particularly in CVMI stage 3 and 4 (pubertal growth phase) were in advanced stage of DI which is similar to the present study.

Giri *et al.* [19] found a statistically significant association between mandibular second molar calcification and CVMI method in both males and females. Average chronological age of males was greater compared to females in each group of CVMI except stage 4. It was found that stage 3 and 4 of CVMI which represents the peak skeletal growth spurt corresponded with stage F and G of DI for females and stage G of DI for males which is similar to the present study.

Aunhomi *et al.* [20] investigated the relation between dental development of left mandibular canines, first and second premolars and second molars and cervical vertebral maturation stages. Mandibular second molars had the strongest correlation with CVMI in both genders. Stages F and G of DI in females and Stage G of DI in males for the mandibular second molar represents the peak of pubertal growth spurt which is similar to the present study.

The findings of Uysal *et al.* [10] and B. Rai *et al.* [21] indicated that the maturation patterns of tooth development in males tend to be more advanced than females in relation to skeletal maturity stages which is similar to the present study. Mithun *et al* [22] found significant correlation between mandibular second molar calcification stages by DI and CVMI method by Hassel and Farman. The chronological age distribution into six stages of CVMI showed that each stage appeared earlier in females than in males. CVMI stages in male subjects had a more advanced trend in DI and opposite pattern seen in female subjects.

Mittal *et al.* [13] investigated the relation between the calcification stages of various teeth (canines, first and second premolars, second and third molars) and skeletal maturity stages. The second molar showed the highest correlation in both males and females. The appearance of each CVMI stage is consistently earlier in the females than in the males.

The unique and significant findings from the present study imply that the stages of mandibular second molar calcification as observed on panoramic and intraoral periapical radiographs gives very accurate results and can be considered as a reliable indicators of skeletal maturity. This method can be easily incorporated into clinical practice by using Introral periapical radiograph (IOPA) for initial growth assessment of an individual.

In orthodontic perspective, the orthopaedic treatment is possible during stage 2 of CVMI which represents the pre-peak of pubertal growth. Myofunctional therapy can be done during the peak of pubertal growth (stage 3 and 4 of CVMI) where more of skeletal changes will be seen. In stage 5 and 6 which represents the end of pubertal growth spurt, only dental effects will be seen.

5. Conclusion

- A highly significant association was found between CVMI and DI of permanent mandibular second molar for both males and females.
- The appearance of each CVMI stage was consistently earlier in females than in male subjects.
- The dental calcification stages were advanced in male subjects as compared with female subjects at the same CVMI stage.
- DI stage E corresponds to stage 2 of CVMI which represents the pre-pubertal growth spurt. DI stages F and G corresponds to stage 3 and 4 of CVMI which represents the pubertal growth spurt. DI Stage H

corresponds to CVMI stage 5 and 6 which represents the end of pubertal growth spurt.

- The calcification stages of permanent mandibular second molar can also be used as a reliable indicator for evaluation of growth stages in clinical practice.

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