PREVALENCE OF CARIES SECONDARY TO THE ORTHODONTIC TREATMENT IN PATIENTS VISITING A PRIVATE DENTAL HOSPITAL-A RETROSPECTIVE STUDY

Type of study: Short Research Article
Running Title: A retrospective study of prevalence of caries secondary to the orthodontic treatment in patients visiting a private dental hospital.

ABSTRACT:

INTRODUCTION:
Orthodontic treatment involving fixed appliance is a long term process. This treatment has a maximum risk of development of caries as proper oral hygiene maintenance would not be noticed. This increases the number of streptococcus mutans and lactobacillus which would form a biofilm. The armamentarium used in this treatment like molar band, brackets etc. are plaque stagnant areas, which over a period of time leads to caries, gingivitis etc. Thus, proper oral hygiene should be maintained by patients.

AIM:
The main objective of the study is to assess the prevalence of caries secondary to the orthodontic treatment in patients visiting a private dental hospital.

MATERIALS AND METHOD:
A descriptive study which was performed under a university setting where all the patients reported to Private Dental college and hospital, Chennai. The data was collected by reviewing patients records and analysed the data of 140 patients who underwent orthodontic appliance treatment between May 2019- December 2020**.** The population size of the study was found to be n=80. Data was collected, tabulated, statistical analysis was done using SPSS-IBM.

RESULTS AND DISCUSSION:
From the statistical analysis, it is observed that the total number of caries that could be seen after the orthodontic appliance treatment was more in males (58.23%) when compared to females and the most commonly affected age is between 18-25 years with 49.37%. Moreover, the common tooth that affected were molars with 49.37%.

CONCLUSION:
Within the limitations of the current study, it is found that molars are the most commonly affected teeth and the age group which is affected the most is between 18-25 years. Thus, it is the

imperative for dentists to provide preventive resin restorations, fluoride varnish and also proper instructions for maintenance of oral hygiene during the orthodontic treatment.

KEYWORDS:
Orthodontic appliance, biofilm, gingivitis, periodontitis , innovative technology, novel method

INTRODUCTION:

Orthodontic treatment, especially the fixed appliance is most likely to cause plaque accumulation, increasing the risk of caries and periodontitis. It makes the conventional method of brushing difficult and also decreases the removal of plaque by the saliva (1). It could be seen that the majority of the population undergoing fixed appliance is under the age group of 25 years. This type of the orthodontic treatment has made the maintenance of the oral hygiene difficult, altered the environment of the oral cavity and has also increased the caries activity due to increase in the salivary concentrations of lactobacilli which is the source of acid for enamel demineralisation (2).The ecological changes that place in the oral microbiota affect the composition, metabolic activity and pathogenicity of the biofilm formed (3). The biofilm which forms on the tooth surface induces caries, gingivitis and periodontitis. This problem arises mainly due to the presence of the archwires, multiple loops and different types of elastics which makes it complicated to maintain and hence those areas become more susceptible for caries formation (4). The insertion of the wire during the ortho treatment, will make new surfaces available for the biofilm to form (5). Patients who are under orthodontic treatment are more prone to develop caries when compared to others. However, the distribution of lesions and the severity varies from person to person (6). The most commonly affected site would be the buccal and lingual surfaces especially in the anterior and lower premolar areas, the pits and fissures of both premolars and molars (7). The use of topical fluorides and mechanical plaque control methods have been incorporated (8). It has been noticed that streptococcus mutans and Lactobacillus are the major constituents of the pathogenic bacteria flora in causing dental caries (9). The capacity for S.mutans to produce caries is the ability to synthesize water-insoluble glucans, the main component essential for its cariogenicity (10), (11). Many studies have revealed that changes in the development of S.Mutans and Lactobacillus are complex and unpredictable during the first 2 months of the orthodontic treatment (12). In clinical studies, increase in the incidence of incipient carious lesions and generalised gingival inflammation have been noticed in patients undergoing fixed orthodontic treatment (13). Researches have revealed

that material and properties of brackets have an effect on the bacterial attachment and plaque retaining capacity and microbial diversity (14),(15). Initial bacterial accumulation is a pre-condition for the further development of the biofilm which on severe progression would result in dental caries and periodontitis. The main reason for the deterioration of the periodontal tissue is that these orthodontic bands are engaged near the supra and subgingival areas (16). Patients undergoing orthodontic treatment will most commonly experience caries at the end stage of their treatment (17). Severe biofilm formation would thus lead to periodontitis , caused by the periodontopathogenic bacteria and another condition is gingivitis, which is usually a milder one when compared to periodontitis, and is largely reversible (18),(19). This gingival enlargement can lead to increase in the subgingival surface (20),(21). Hence, biofilm formation is one the major problems for inducing caries and is said to be high in the patients undergoing orthodontic treatment. The aim of the study is to assess the prevalence of caries secondary to the orthodontic treatment in patients visiting a private dental hospital.

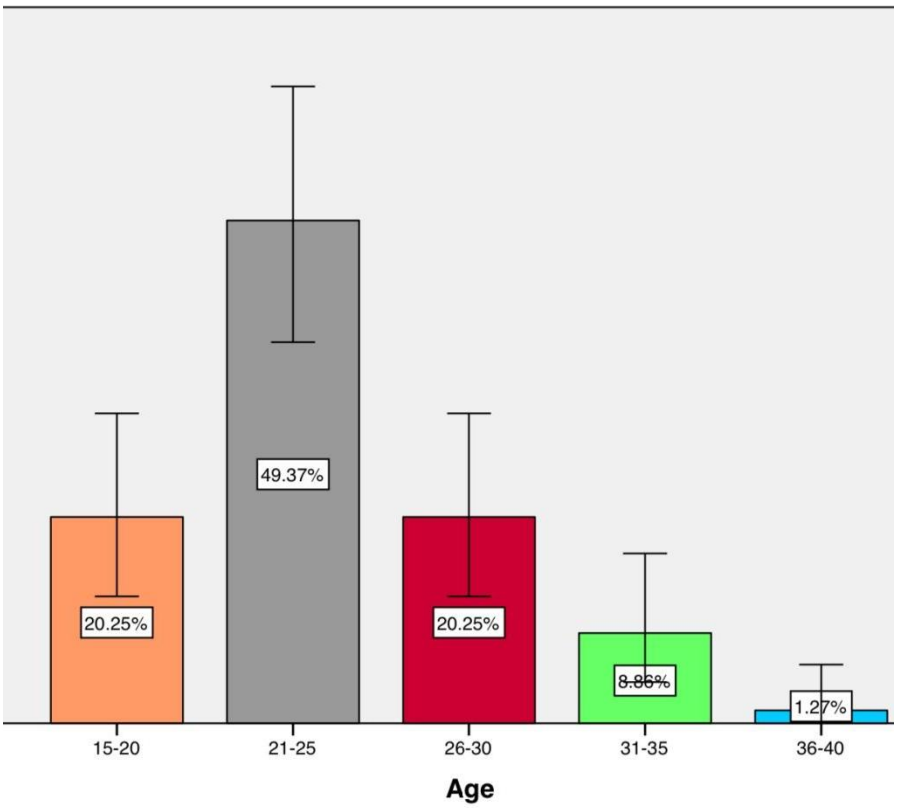
MATERIALS AND METHOD:

This was a descriptive study which was performed under a university setting where all the patients between 15-40 years of years reported to Private Dental College and hospitals, Chennai, India. The data was collected by reviewing the patient's post treatment records and analysed the data of 140 patients who underwent orthodontic appliance treatment between May 2019-December 2020 in which sample size of the study who had caries post orthodontic treatment was found to be n=80 whereas rest 60 patients were found to be caries free. The ethical approval was obtained from the Institutional Ethical Committee. The data also gives the different duration of the orthodontic treatment which could be taken as 2,3,4,5,6,7,8 months and 1 year.The data was cross verified with photographs and was compiled for statistical analysis on SPSS software. The minimising sampling bias was done using the random sampling method. There was a high internal validity and low external validity in our study. The inclusion criteria for the study is patients between 15-40 years of age who underwent only orthodontic fixed appliance treatment. Exclusion criteria is patients with existing caries before the start of orthodontic treatment and patients who had other orthodontic treatment like removable appliance etc. Chi square test was used to compare the groups with $p<0.05$ was considered significant and the results were interpreted.

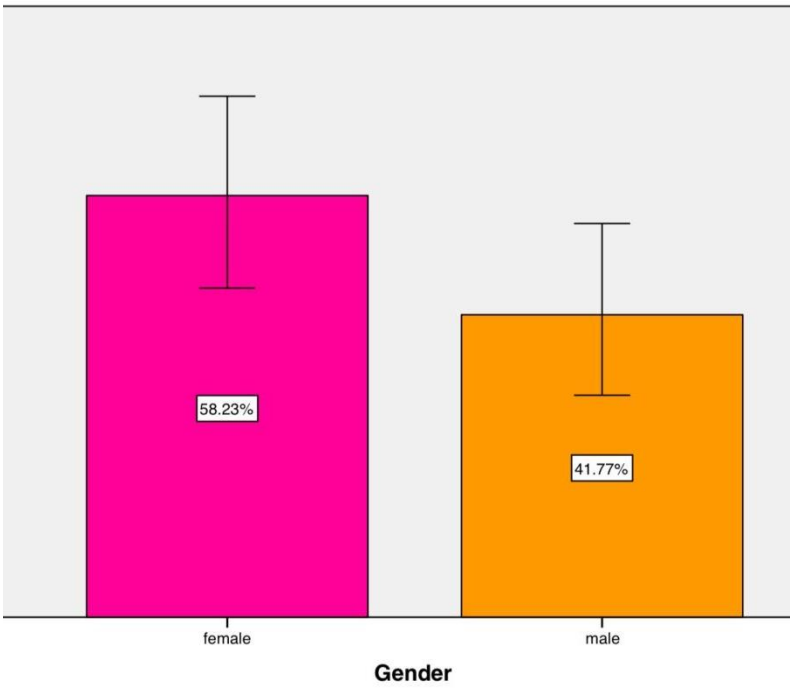
RESULTS:

The current study shows the data for the caries which have been developed after the orthodontic treatment. Graph 1 the distribution of patients having caries after the orthodontic treatment in relation to age. Patients in 21-25 years of age had been maximum affected (49.37%) whereas those in 15-20 years were less affected (20.25%) when compared to 21-25 years of age group. The data reveals that 58.23% of the females were most commonly affected with caries (Graph 2).

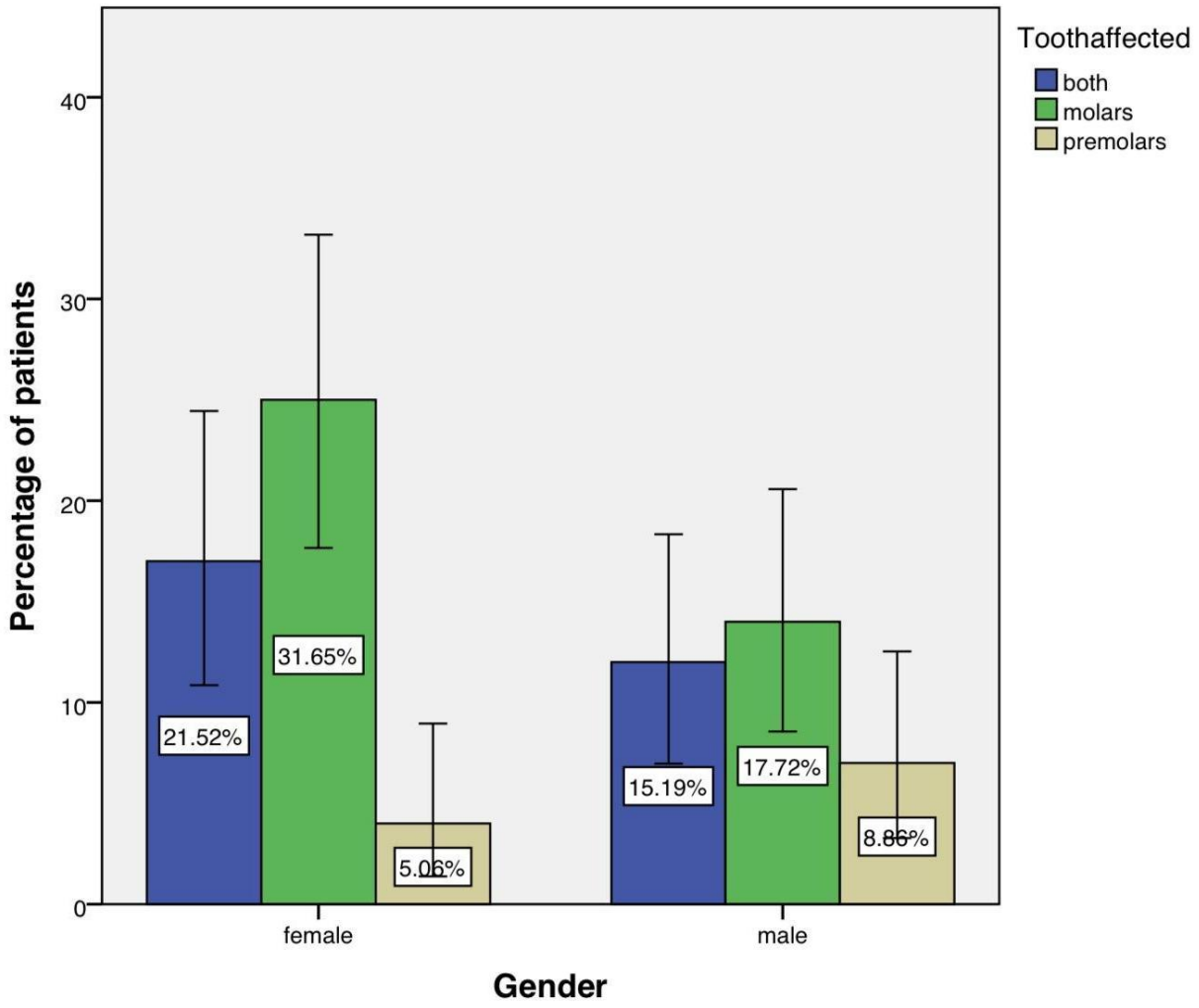
Molars were most commonly affected among both females and males in which females have a higher percentage of 31.65% whereas males have 17.72%. (Graph 3).Most affected teeth were patients having both the molars and premolars which were affected simultaneously, in which females are 21.52% whereas males have 15.19% affected due to caries with p value of 0.2, being not significant.Correlation between the gender and the arch affected showed Both maxillary and mandibular arches had been simultaneously affected for males and females in which females show a high percentage of 29.11% whereas males show 18.99%.(Graph 4) Mandibular arch was affected next with 21.52% in females and 13.92% in males and also has a p = 0.6, which was not significant. Correlation between the gender and the treatment duration found that females were more affected than males. 22.78% of females got caries in a period of 3 months treatment duration whereas 15.19% of males developed caries in a duration of 6 months of orthodontic treatment (Graph 5). These two percentages were the highest in the treatment duration and with the p value of 0.08, which was not significant. Correlation between the age and the tooth affected showed that molars were most affected teeth in the age groups between 15-30 years. 22.78% of them affected belong to 21-25 years, 10.13% of them in 15-20 years and 12.66% of them in 26-30 years for which the p value = 0.3, not significant (Graph 6). Also the correlation between the age and arch was seen. Both maxilla and mandibular arch had been simultaneously affected for the majority of patients. Around 18.99% of those affected were in 21-25 years, 12.66% of them belonged to 26-30 years and 8.86% were under 15-20 years. Moreover, the mandible arch had been affected the most in 21-25 years patients (22.78%) when compared to the other groups (Graph 7). correlation between the age and the treatment duration. Caries development in 6 months duration seemed to be highest in certain groups like 21-25 years (17.72%) and 15-20 years (5.06%), whereas on the other hand 3 months duration was the highest in age group 26-30 years (7.59%), 31-35 years (3.80%) and 36-40 years (1.27%) (Graph 8).



Graph 1: The bar chart represents the distribution of patients having caries after their orthodontic treatment in relation to age. Most of the people who were likely affected were between 21-25 years (49.37%), 15-20 years (20.25%), 26-30 years (20.25%), 31-35 years (8.86%).



Graph 2: This bar chart represents the distribution of patients affected with caries after their orthodontic treatment in relation to gender. Nearly more than half of the females (58.23%) were affected when compared to males (41.77%).

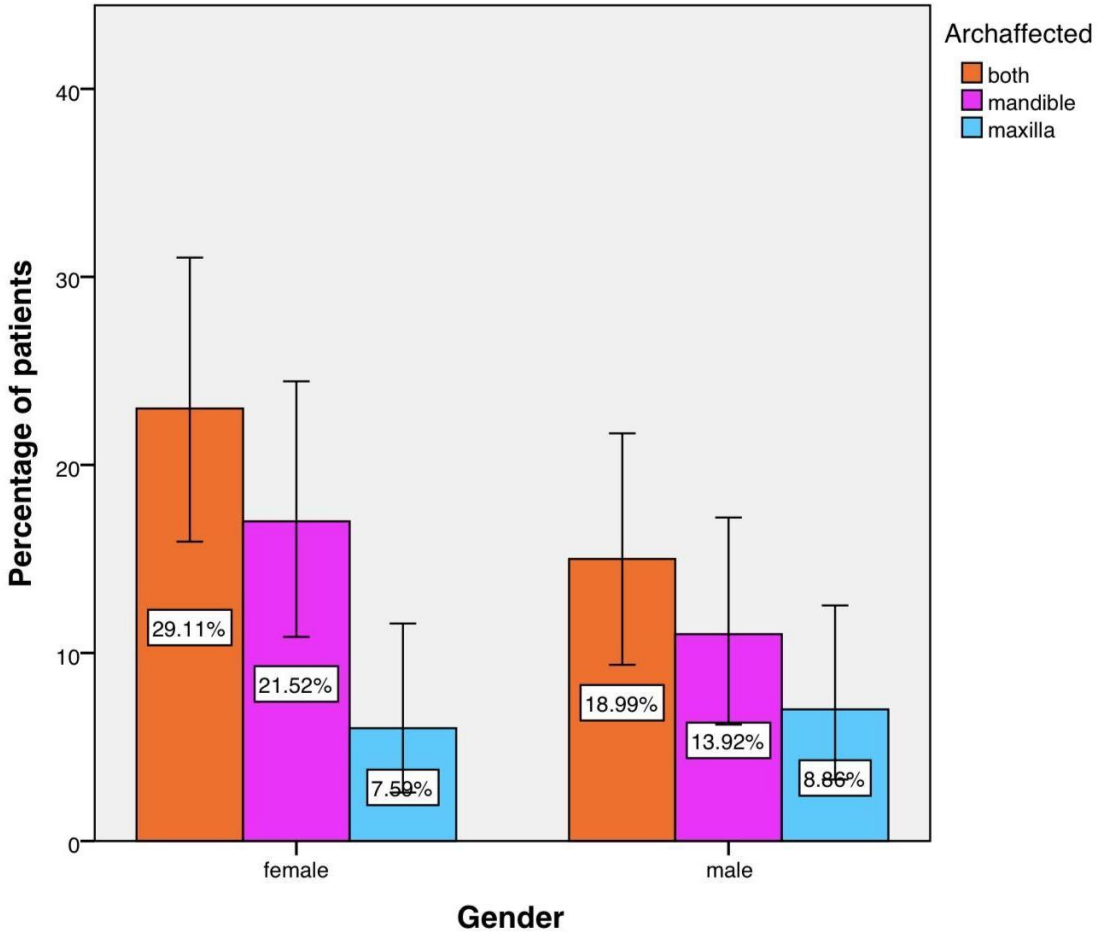


Chi-Square Tests

	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	2.717[a]	2	.257
Likelihood Ratio	2.691	2	.260
N of Valid Cases	79		

Graph 3: The bar chart represents the correlation between the gender and the tooth affected. The x axis represents gender and the y axis represents the count of the tooth affected. The count of molar teeth affected was found to be the highest in which females had a largest count of 31.65% and males had 17.72%. Next affected tooth was premolars with 5.06% in females and 8.86% in

males. However, a large percentage is present for both the teeth affected which was 21.52% for females and 15.19% in males. (Chi square test ; p value=0.2; p>0.05; hence not significant). From this we can infer than females are more prone to the caries especially in molars after the orthodontic treatment (31.65%).

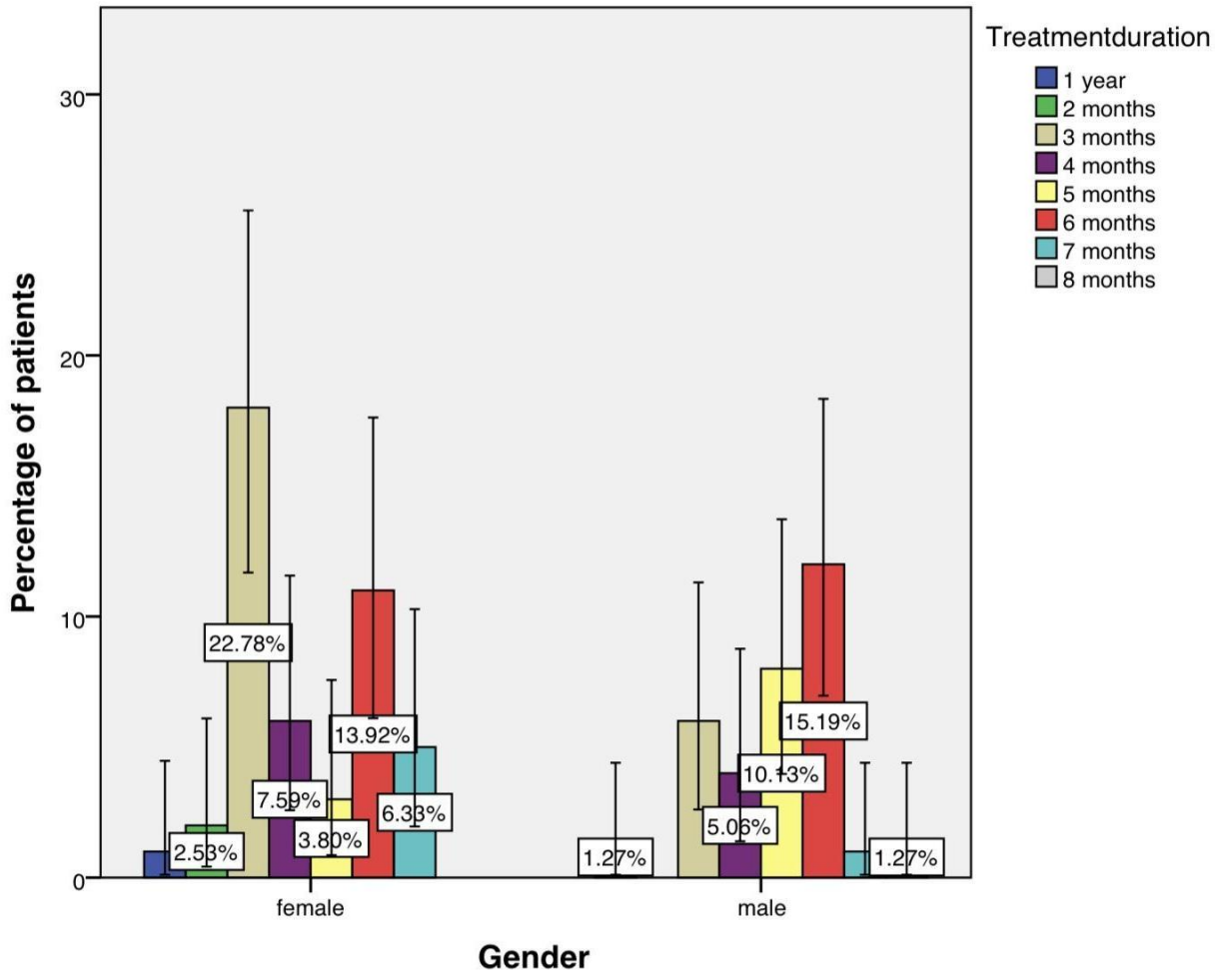


Chi-Square Tests

	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	.933[a]	2	.627
Likelihood Ratio	.921	2	.631
N of Valid Cases	79		

Graph 4: The bar chart represents the correlation between the gender and the arch affected. The x axis represents gender and y axis represents the count of the arch affected. The count of

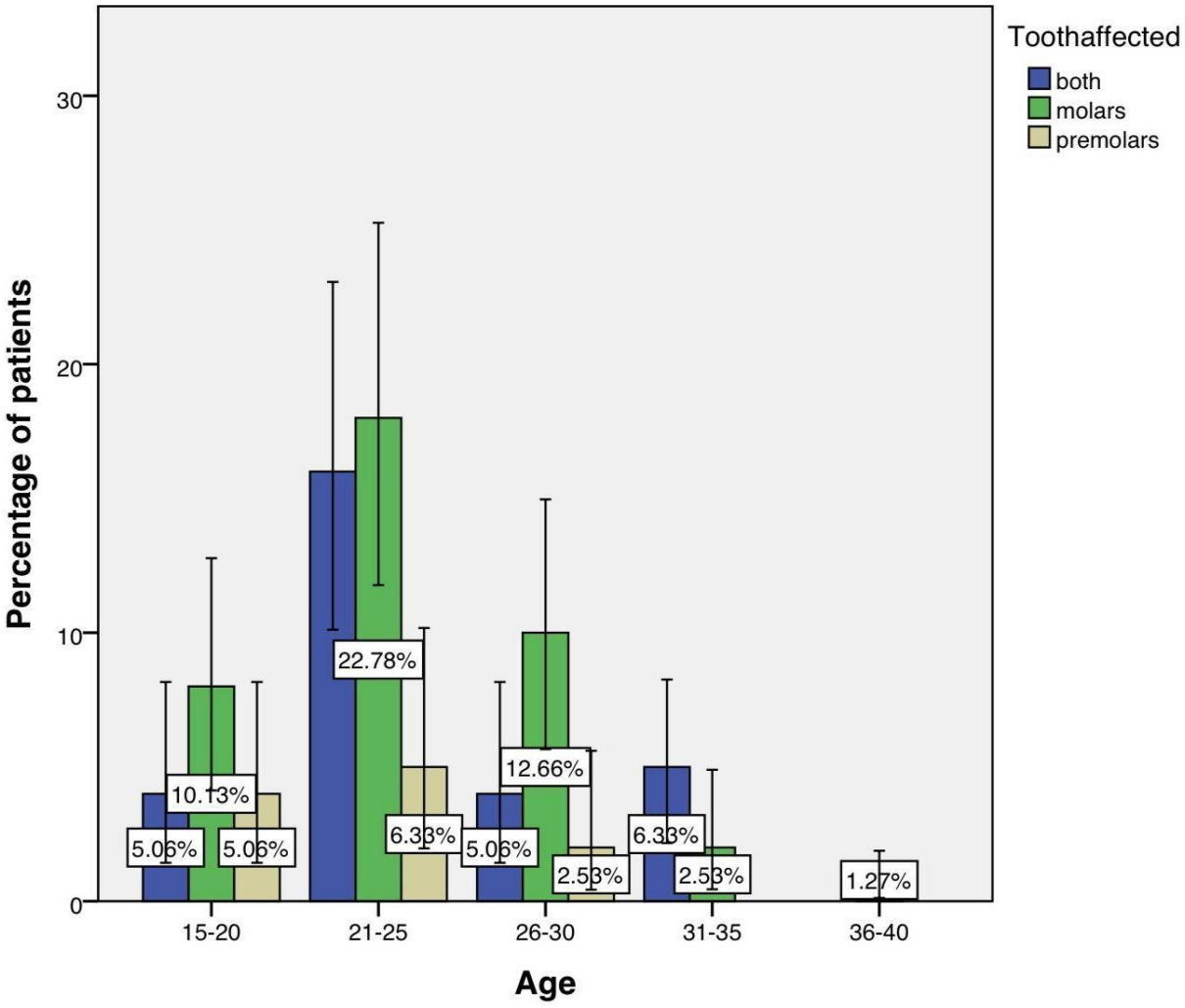
maxillary arch only was found to be 7.59% in females and 8.86% in males. Followed by the count of mandibular arch only was found to be 21.52% in females and 13.92% in males. The count was seen highest in both the arches affected which was 29.11% in females and 18.99% in males.(Chi square test ; p value=0.6; p>0.05; hence not significant). From this graph, we can infer that females are affected the most by caries in both the maxillary and mandibular arches (29.11%)



Chi-Square Tests

	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	12.584[a]	7	.083
Likelihood Ratio	14.004	7	.051
N of Valid Cases	79		

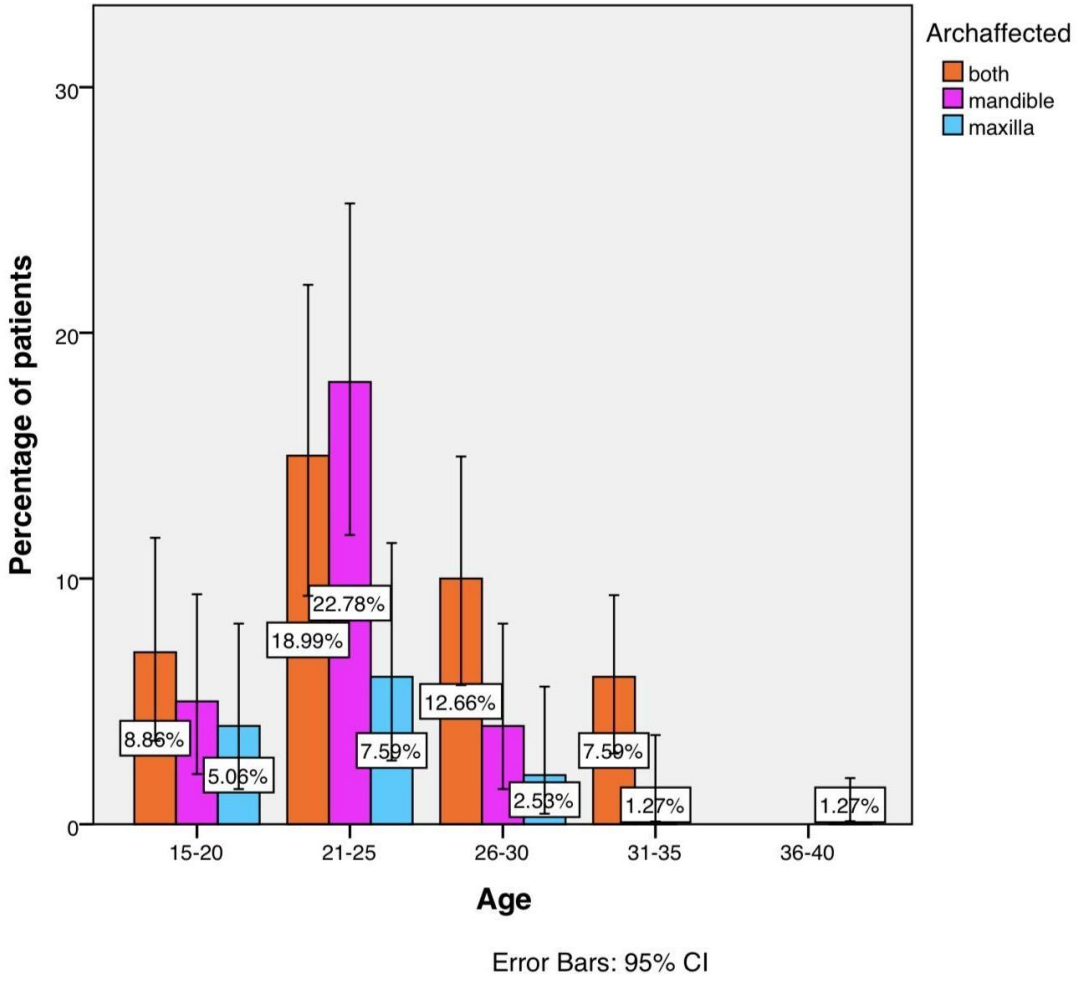
Graph 5: The bar chart represents the correlation between the gender and the treatment duration. The x axis represents the treatment duration and the y axis represents the count of the affected teeth in various treatment duration. The count for 3 months was found to be highest in which it is 22.78% in females and 7.59% in males. The count of affected teeth was found to be highest in 6 months for males with 15.69%. (Chi square test ; p value=0.08; p>0.05; hence not significant). From this graph, we can infer that females have developed more caries within 3 months of their treatment (22.78%) and males have developed caries in 6 months of their treatment (15.19%).



Chi-Square Tests

	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	8.414[a]	8	.394
Likelihood Ratio	9.216	8	.324
N of Valid Cases	79		

Graph 6: The bar chart represents the correlation between the age and the tooth affected. The x axis represents the age groups and y axis represents the count of the tooth affected. It was found that molars only are the highest being affected and are 10.13% in 15-20 years, 22.78% in 21-25 years and 12.66% in 26-30 years. The least affected was premolars and the second most affected were both molars and premolars only.(Chi square test ; p value=0.3; p>0.05; hence not significant). From this graph, we can infer that patients in the age group 21-25 years are most affected by caries especially in molars (22.78%).

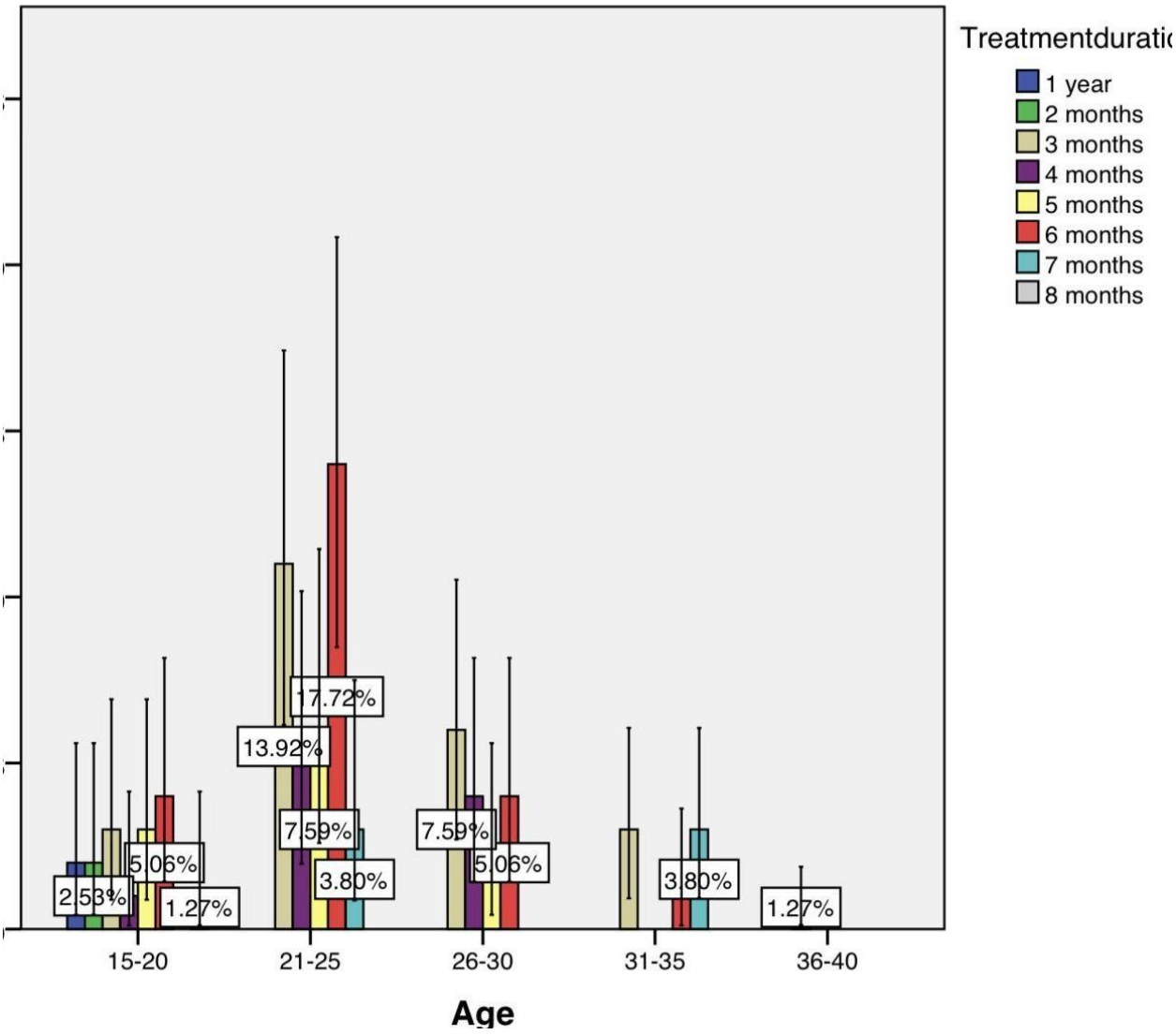


Chi-Square Tests

	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	13.401[a]	8	.099
Likelihood Ratio	12.817	8	.118
N of Valid Cases	79		

Graph 7: The bar chart represents the correlation between the age and arch affected. The X axis presents the age groups and the y axis represents the count of the arch affected. The highest percentage was seen in the both maxillary and mandibular arch affected and that was about 18.99% in 21-25 years, 8.86% in 15-20 years, 12.66% in 26-30 years and 7.59% in 31-35 years.(Chi square test ; p value=0.09; p>0.05; hence not significant). Thus, we can infer that

patients in the age group 21-25 years are the most affected by caries especially in their mandibular arch (22.78%).



Chi-Square Tests

	Value	df	Asymptotic Significance (2-sided)
Pearson Chi-Square	43.193[a]	28	.033
Likelihood Ratio	37.587	28	.106
N of Valid Cases	79		

Graph 8: The bar chart represents the correlation between the age and the treatment duration. The x axis represents the age group and y axis represents the treatment duration. It was found to be highest in 6 months which was 17.72% in 21-25 years, 5.06% in 15-20 years, 5.06% in 26-30 years.(Chi square test ; p value=0.03; $p<0.05$; hence significant). From this graph, we can infer that patients in the age group 21-25 years are having more caries in 6 month duration of their treatment (17.72%).

DISCUSSION:

Orthodontic treatment with a fixed appliance increases the risk of developing plaque retention and thus increases the risk for caries and periodontitis. Bacterial adhesion to these materials has the ability to form the biofilm on foreign bodies which leads to infections, caries and other conditions (22). It includes braces, wires and attachments which would retain more food particles and provide retentive sites for dental plaque. It makes it more difficult to maintain oral hygiene and thereby there is a marked increase in the S.mutans and lactobacillus seen. It could be noticed that self ligating braces restrict wires into slots using self ligating components that reduces friction and provide less space for plaque accumulation when compared to other dental ligating components.Biofilm process starts initially with the bacterial attachment. The glucan binding protein contributes to the initial attachment.

In this study, we could find that caries after the ortho treatment was seen that 21-25 years was the most affected age group (Graph 1). In a study by Ameberg et al, states that 19-25 years are affected as they face greater challenges in maintaining good oral hygiene. Contrast study of weiting chen and Yu Zhou states that the orthodontic treatment with the fixed appliance decreased the risk of caries. This could be due to increase in the caries level mainly depending on the individual susceptibility, dietary, oral hygiene and could also be due to lack of basic manual skills and intellectual abilities that preclude adequate practices such as brushing. Oral health education is a must before placing an orthodontic appliance inorder to obtain an optimum oral hygiene achievement (23). We observed that females are more affected by caries after their ortho treatment when compared to males (Graph 2). Previous study of Kristian Tadic et al, reveals that caries due to ortho treatment were seen in 60% of females and 40% of males. Another study contrast to this study revealed that males (58%) had more caries when compared to females (42%). Dental anxiety and negligence is seen more commonly in females. Although, growing up females tend to understand the importance of oral hygiene when compared to males.There are various factors contributing to the dental anxiety such as infrequent visits, time taken for the procedure, encountering any painful experience, anxiety in falling of brackets, intricacy of the procedure (24).

This study reveals that 31.65% of females and 17.72% of males have got molars affected (p value=0.2; $p>0.05$; not significant) (Graph 3). In the study of Mustafa et al, molar teeth had more caries which affected both the males and females equally. Another study by Macek et al, stated

that caries in molars are more susceptible when compared to the incisors, canines and premolars. In a contrast study of Eur J et al, reveal that more caries could be seen in the distal surfaces of the central incisor, lateral incisors and canines. The reason for caries buccal surfaces of the maxillary molars and the lingual surfaces of the mandibular molars is due to the intricate morphology and difficult access for good oral hygiene (25). It could also be seen that greater food impactions are there between archwire and soft tissue (26).

Both the arches were equally affected in males (18.99%) and females (29.11%) with p value=0.6; p>0.05, not significant (Graph 4). In a study of Safa Tuncer et al, females are more affected (59.1%) by caries in both the maxillary and mandibular arch. A contrast study proposed by Manji et al, stated that lower molars were severely affected in the entire dentition. The reason for developing caries could mostly be due to the many reasons. It could be due to the female sex hormones. In a study of Lukacs, stated that hormones play a significant role in the cavity formation. Cumulative effects of estrogens, fluctuations at puberty and high levels during pregnancy can induce caries. Yet another reason could also be due to the less saliva production by females when compared to males, thus reducing the food residue in the teeth and also there is a rate of flow of saliva during pregnancy (27). It was found that the most affected patients with caries is during their 3 months for females (22.78%) and 6 months duration for males (15.19%) of their orthodontic treatment with p value of 0.08 (Graph 5). Another study revealed high caries in females within 5 months of the orthodontic treatment (28). There are no such contrasting studies correlating the gender and the duration of the orthodontic treatment. Reasons could be mainly due to the anxiety females face and related to the dental anxiety issues which females face apart from the biofilm formation.

In relation to the tooth affected with age, molars seem to show the highest percentage for all the age groups with the p value=0.3; p>0.05, not significant (Graph 6). In the study of lange et al, states that caries was highest in the age group 16-20 years (29). In a similar study of Blerim kamberi et al, it was found that prevalence of dental caries was highest in the 18-34 year group (77.90%). A contrast study suggested the high rate of caries was found to be in the age of 45-70 years (80.1%). The reasons could be reduced brushing frequency, not following instructions given by the orthodontist, reduced frequency of dental check ups and most importantly lack of knowledge in the brushing pattern and maintenance of the appliance (30). In this study, the correlation between the age and arch affected reveals that both the arches were equally affected which was seen highest in the age group 21-25 years with 18.99% and p value=0.09; p>0.05; not significant. (Graph 7). A similar study of Yu Zhou revealed the caries affecting both the maxillary and mandibular arches equally, especially the molars of both in teenagers. In a contrast study of Falci et al, stated that mandibular arch was affected in the age group 35-45 years (31) It could be morphology of the molars with deep pits and fissures that causes increased risk of caries especially when enamel decalcification takes place during the treatment which creates stagnation of bacteria and biofilm formation (32).

In our study, there is a correlation between the treatment duration and age which shows that 6 months duration is the highest in 21-25 years with 17.72% and p value=0.03; $p<0.05$, significant (Graph 8). A similar study stated that orthodontic treatment was associated with active caries with the prevalence rate of 72.3%. There were no contrast studies present for the correlation between the age and the duration of the orthodontic treatment. The longer the duration of the orthodontic treatment, the higher occurrence of active carious lesions. Various treatment options are available in order to prevent the caries formation. Preventive resin restoration before undergoing the orthodontic treatment, usage of orthodontic brushes will definitely prevent these caries development (33). The level of caries might even go as deep as we would have never imagined, so, prevention is quite important. Proper brushing technique and rinsing method should be taught to the patients to decrease the risk of caries (34). Preventive resin restoration can be given based on the affordability of the patient. High fluoride varnish can also be given to prevent white spot lesion (35). Weekly review is a must for these orthodontic patients. The limitations of our study is unicentred with a limited demographic area of smaller sample size. By investigating the prevalence of caries and its association with preventive measures would also might help broaden existing knowledge about the various preventive measures for such caries development and hence would also improve our clinical management to minimize false interpretations.

CONCLUSION:

Within the limitations of study, it can be concluded that patients undergoing orthodontic treatment, especially fixed appliance, molars are the common affected teeth and also between the age group 21-25 years of age. This is one of the most recent and perhaps the first study to evaluate the prevalence of caries secondary to the orthodontic treatment among 15-40 years of age visiting the outpatient department of Private Dental College and Hospitals. Hence, patients should have proper oral hygiene maintenance especially during their orthodontic treatment as they are prone to develop in such patients and at the same time, duty of the dentists to provide preventive measures like preventive resin restoration, mouth washes, orthodontic brushes and fluoride varnish.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

REFERENCES:

1. Scheie AA, Arneberg P, Krogstad O. Effect of orthodontic treatment on prevalence of Streptococcus mutans in plaque and saliva [Internet]. Vol. 92, European Journal of Oral Sciences. 1984. p. 211–7. Available from: http://dx.doi.org/10.1111/j.1600-0722.1984.tb00881.x

2. Schwaninger B, Vickers-Schwaninger N. Developing an effective oral hygiene program for the orthodontic patient: Review, rationale, and recommendations [Internet]. Vol. 75, American Journal of Orthodontics. 1979. p. 447–52. Available from: http://dx.doi.org/10.1016/0002-9416(79)90167-2

3. Yamunadevi A, Pratibha R, Rajmohan M, Ganapathy N, Porkodisudha J, Pavithrah D, et al. Molecular Insight into Odontogenesis in Hyperglycemic Environment: A Systematic Review. J Pharm Bioallied Sci [Internet]. 2020 Aug;12(Suppl 1):S49–56. Available from: http://dx.doi.org/10.4103/jpbs.JPBS_159_20

4. Westergren G, Krasse B. Evaluation of a micromethod for determination of Streptococcus mutans and Lactobacillus infection. J Clin Microbiol [Internet]. 1978 Jan;7(1):82–3. Available from: https://www.ncbi.nlm.nih.gov/pubmed/342541

5. Antony JVM, Vini Mary Antony J, Ramani P, Ramasubramanian A, Sukumaran G. Particle size, penetration rate and effects of smoke and smokeless tobacco products – an invitro analysis [Internet]. Vol. 7, Heliyon. 2021. p. e06455. Available from: http://dx.doi.org/10.1016/j.heliyon.2021.e06455

6. Wisth PJ, Nord A. Caries experience in orthodontically treated individuals. Angle Orthod [Internet]. 1977 Jan;47(1):59–64. Available from: http://dx.doi.org/10.1043/0003-3219(1977)047<0059:CEIOTI>2.0.CO;2

7. R H, Hannah R, Ramani P, Ramanathan A, Jancy MR, Gheena S, et al. CYP2 C9 polymorphism among patients with oral squamous cell carcinoma and its role in altering the metabolism of benzo[a]pyrene [Internet]. Vol. 130, Oral Surgery, Oral Medicine, Oral Pathology and Oral Radiology. 2020. p. 306–12. Available from: http://dx.doi.org/10.1016/j.oooo.2020.06.021

8. Zachrisson BU. Fluoride application procedures in orthodontic practice, current concepts. Angle Orthod [Internet]. 1975 Jan;45(1):72–81. Available from: http://dx.doi.org/10.1043/0003-3219(1975)045<0072:FAPIOP>2.0.CO;2

9. Tanner ACR, Sonis AL, Lif Holgerson P, Starr JR, Nunez Y, Kressirer CA, et al. White-spot lesions and gingivitis microbiotas in orthodontic patients. J Dent Res [Internet]. 2012 Sep;91(9):853–8. Available from: http://journals.sagepub.com/doi/10.1177/0022034512455031

10. Umashankar K, Abilasha, Hannah, Ramani P, Gheena. Knowledge and attitude about COVID-19 pathogenesis among oral pathologists in Chennai. Int J Curr Res Rev [Internet]. 2020;12(19):143–51. Available from: https://ijcrr.com/uploads/2964_pdf.pdf

11. Suvarna K, Abilasha R, Gheena S. Analysis of Prevalence of Oral Squamous Cell Carcinoma in Patients with History of Chronic Irritation of Oral Tissues-A Retrospective Study. Indian Journal of [Internet]. 2020; Available from: http://search.ebscohost.com/login.aspx?direct=true&profile=ehost&scope=site&authtype=crawler&jrnl=09739122&AN=148410023&h=XYlwRUZ0p54Y6mZHESI7LVi228pkq%2FRhoPO4Jt4231oGPh6KuqA4bM6e9ZslMs9HCBZY6ZF8tl2TlR3cgyExXQ%3D%3D&crl=c

12. Topaloglu-Ak A, Ertugrul F, Eden E, Ates M, Bulut H. Effect of Orthodontic Appliances on Oral Microbiota—6 Month Follow-up. J Clin Pediatr Dent [Internet]. 2011 Jul 1 [cited 2021 May 11];35(4):433–6. Available from: https://meridian.allenpress.com/jcpd/article/35/4/433/78329

13. Alexander SA. Effects of orthodontic attachments on the gingival health of permanent second molars. Am J Orthod Dentofacial Orthop [Internet]. 1991 Oct;100(4):337–40. Available from: http://dx.doi.org/10.1016/0889-5406(91)70071-4

14. Atack NE, Sandy JR, Addy M. Periodontal and microbiological changes associated with the placement of orthodontic appliances. A review. J Periodontol [Internet]. 1996 Feb;67(2):78–85. Available from: http://dx.doi.org/10.1902/jop.1996.67.2.78

15. Monica K, Gheena S, Ramani P. IN SILICO GENE EXPRESSION ANALYSIS OF CRUCIAL CELL CYCLE CONTROL GENE CDKN2A AND CDKN2B IN HEAD AND NECK SQUAMOUS CELL …. Annals of [Internet]. 2020; Available from: https://www.journal.atmph-specialissues.org/abstract.php?article_id=8921

16. Boyd RL, Leggott PJ, Quinn RS, Eakle WS, Chambers D. Periodontal implications of orthodontic treatment in adults with reduced or normal periodontal tissues versus those of adolescents. Am J Orthod Dentofacial Orthop [Internet]. 1989 Sep;96(3):191–8. Available from: http://dx.doi.org/10.1016/0889-5406(89)90455-1

17. Sinduja P, Ramani P, Gheena S, Ramasubramanian A. Expression of metallothionein in oral squamous cell carcinoma: A systematic review. J Oral Maxillofac Pathol [Internet]. 2020 Jan;24(1):143–7. Available from: http://dx.doi.org/10.4103/jomfp.JOMFP_137_19

18. Kouraki E, Bissada NF, Palomo JM, Ficara AJ. Gingival enlargement and resolution during and after orthodontic treatment. N Y State Dent J [Internet]. 2005 Jun;71(4):34–7. Available from: https://www.ncbi.nlm.nih.gov/pubmed/16146305

19. Ramani P, Krishnan RP, Karunagaran M, Muthusekhar MR. Odontogenic sarcoma: First report after new who nomenclature with systematic review. J Oral Maxillofac Pathol [Internet]. 2020 Jan;24(1):157–63. Available from: http://www.jomfp.in/text.asp?2020/24/1/157/283952

20. Ramani P, Gheena S, Karunagaran M, Hannah R. Clear-cell variant of oral squamous cell carcinoma: A rare entity. J Oral Maxillofac Pathol [Internet]. 2021 Mar 1 [cited 2021 May 15];25(4):22. Available from: https://www.jomfp.in/article.asp?issn=0973-029X;year=2021;volume=25;issue=4;spage=22;epage=26;aulast=Ramani

21. Ramasubramanian A, Ramani P, Sherlin HJ, Premkumar P, Natesan A, Thiruvengadam C. Immunohistochemical evaluation of oral epithelial dysplasia using cyclin-D1, p27 and p63 expression as predictors of malignant transformation. J Nat Sci Biol Med [Internet]. 2013 Jul;4(2):349–58. Available from: http://dx.doi.org/10.4103/0976-9668.117011

22. E A, Aswani E, Gheena S, Pratibha R, Abilasha R, Hannah R, et al. Overexpression of HNRNPA2B1 is Associated with Poor Prognosis in Head and Neck Squamous Cell Carcinoma [Internet]. International Journal of Current Research and Review. 2020. p. 15–8. Available from: http://dx.doi.org/10.31782/ijcrr.2020.122502

23. Costa MR, Silva VC, Miqui MN, Sakima T, Spolidorio DMP, Cirelli JA. Efficacy of ultrasonic, electric and manual toothbrushes in patients with fixed orthodontic appliances. Angle Orthod [Internet]. 2007 Mar;77(2):361–6. Available from: http://dx.doi.org/10.2319/0003-3219(2007)077[0361:EOUEAM]2.0.CO;2

24. Arici S, Alkan A, Arici N. Comparison of different toothbrushing protocols in poor-toothbrushing orthodontic patients. Eur J Orthod [Internet]. 2007 Oct;29(5):488–92. Available from: http://dx.doi.org/10.1093/ejo/cjm038

25. Baumgartner S, Menghini G, Imfeld T. The prevalence of approximal caries in patients after fixed orthodontic treatment and in untreated subjects [Internet]. Vol. 74, Journal of Orofacial Orthopedics / Fortschritte der Kieferorthopädie. 2013. p. 64–72. Available from: http://dx.doi.org/10.1007/s00056-012-0111-2

26. Geiger AM, Gorelick L, Gwinnett AJ, Benson BJ. Reducing white spot lesions in orthodontic populations with fluoride rinsing. Am J Orthod Dentofacial Orthop [Internet]. 1992 May;101(5):403–7. Available from: http://dx.doi.org/10.1016/0889-5406(92)70112-N

27. Behera A, Hannah R. Association of the Depth of Invasion wth Lymph Node Metastasis in Oral Squamous Cell Carcinoma Patients-A Retrospective Study. Indian Journal of [Internet]. 2020; Available from: http://search.ebscohost.com/login.aspx?direct=true&profile=ehost&scope=site&authtype=crawler&jrnl=09739122&AN=148410053&h=nwnHkKvd5DGQXazZu6t5p6l8D0PbUqebPTLzhmdSl6FyXTWsSzGTNdBhQxTZUG9F%2BD8YcgHrW%2FSaXuV%2BCg9vCA%3D%3D&crl=c

28. You C-A, Zheng P, Hu N, Su Q. [Clinical analysis of caries status of the mandibular second molar]. Shanghai Kou Qiang Yi Xue [Internet]. 2014 Apr;23(2):233–6. Available from: https://www.ncbi.nlm.nih.gov/pubmed/24935851

29. Lange J de, de Lange J. Third molars and second molar distal caries [Internet]. Vol. 39, International Journal of Oral and Maxillofacial Surgery. 2010. p. 630. Available from: http://dx.doi.org/10.1016/j.ijom.2010.02.005

30. Krishnan RP, Ramani P, Sukumaran G, Ramasubramanian A, Karunagaran M, Hannah R. Workplace violence among dental surgeons - A survey. 2021 May 19 [cited 2021 May 19]; Available from: https://www.ijdr.in/preprintarticle.asp?id=315914;type=0

31. Falci SGM, de Castro CR, Santos RC, de Souza Lima LD, Ramos-Jorge ML, Botelho AM, et al. Association between the presence of a partially erupted mandibular third molar and the existence of caries in the distal of the second molars [Internet]. Vol. 41, International Journal of Oral and Maxillofacial Surgery. 2012. p. 1270–4. Available from: http://dx.doi.org/10.1016/j.ijom.2012.03.003

32. Thamilselvan S, Abilasha R, Ramani P, Gheena S, Hannah R. Evaluation of Accuracy between Habit History and Incidence of Oral Squamous Cell Carcinoma [Internet]. International Journal of Current Research and Review. 2020. p. 30–5. Available from: http://dx.doi.org/10.31782/ijcrr.2020.122503

33. A study on the variability of drug responsiveness to anti inflammatory drugs - A pilot survey. Int J Pharm Res [Internet]. 2020 Oct 2;12(02). Available from: http://www.ijpronline.com/ViewArticleDetail.aspx?ID=17202

34. Bergenholtz A, Gustafsson LB, Segerlund N, Hagberg C, Ostby N. Role of brushing technique and toothbrush design in plaque removal. Scand J Dent Res [Internet]. 1984 Aug;92(4):344–51. Available from: http://dx.doi.org/10.1111/j.1600-0722.1984.tb00901.x

35. Weintraub JA, Ramos-Gomez F, Jue B, Shain S, Hoover CI, Featherstone JDB, et al. Fluoride varnish efficacy in preventing early childhood caries. J Dent Res [Internet]. 2006 Feb;85(2):172–6. Available from: http://dx.doi.org/10.1177/154405910608500211

Abirami, S., Kumar Jain, R. and Girija, A. S. S. (2020) “Effect of Two Different Mouth Rinses on S. mutans Counts in Subjects Undergoing Orthodontic Treatment – A Pilot Study”, Journal of Pharmaceutical Research International, 32(15), pp. 148-155. doi: 10.9734/jpri/2020/v32i1530638.