ASIAN JOURNAL OF PHARMACEUTICAL AND CLINICAL RESEARCH

INNOVARE ACADEMIC SCIENCES
Knowledge to Innovation

Vol 16, Issue 6, 2023

Online - 2455-3891
Print - 0974-2441

Research Article

STUDY OF THYROID PROFILE, URIC ACID, CREATININE AND LIPID PROFILE IN PRE-ECLAMPSIA PATIENT

CHANCHLESH DEHARIYA[1], CHANDRASHEKHAR JAISWAL[2], RITESH UPADHYAY[3], DILEEP DANDOTIYA[3]*

[1]Department of Pathology, CIMS, Chhindwara, Madhya Pradesh, India. [2]Department of Dermatology, CIMS, Chhindwara, Madhya Pradesh, India. [3]Department of Community Medicine, CIMS, Chhindwara, Madhya Pradesh, India. Email: dr.dileep85@gmail.com

Received: 4 March 2023, Revised and Accepted: 16 April 2023

ABSTRACT

Objectives: (1) To evaluate and compare the thyroid profile and lipid profile in preeclampsia in a normotensive pregnant female in the third trimester of pregnancy. (2) To estimate and compare serum uric acid and serum creatinine in preeclampsia in normotensive pregnant females.

Methods: In the present study, 154 patients, of which 79 preeclampsia (cases) and 75 (controls), were selected randomly and matched with their gestational age in patients who attended an ANC clinic at the Department of Obstetrics and Gynecology, NSCB Medical College, Jabalpur, from March 2016 to August 2017. All patients were evaluated for their lipid profile, thyroid profile, serum uric acid, and serum creatinine. The study includes patient's specific detailed clinical history, physical examination findings, and a report of relevant investigations recorded from the patient who presented with pre-eclampsia and normotensive pregnant female attending the ANC Clinic of N.S.C.B. Medical College, Jabalpur.

Results: The most common age group for preeclampsia was 20–24 years. Preeclampsia was most common in the nullipara as compared to the multipara. Pre-eclampsia cases commonly occur in low-socioeconomic and rural areas. In this study, pre-eclamptic women showed higher serum concentrations of total cholesterol (221.60±22.55), low density lipoprotein (LDL) (134.44±16.96), Very LDL (48.48±6.88) and Triglyceride (243.50±32.33) when compared to normotensive women, which was statistically significant. In this study, a statistically significant decrease in HDLC (34.55±5.37) was observed in pre-eclamptic women. An increase in serum creatinine was observed in women with pre-eclampsia. The mean serum creatinine level was (1.21±0.47).

Conclusion: Our study concludes that nulliparous and low-socio-economic women had a higher risk of pre-eclampsia. Maternal age between 20 and 24 years are more prone to develop pre-eclampsia. Serum uric acid levels are significantly higher in pre-eclampsia patients and could be a useful indicator of maternal and fetal complications in hypertensive patients.

Keywords: Thyroid profile, Uric acid, Creatinine, Lipid profile, Pre-eclampsia.

 DOI: http://dx.doi.org/10.22159/ajpcr.2023v16i6.48268. Journal homepage: https://innovareacademics.in/journals/index.php/ajpcr

INTRODUCTION

Pre-eclampsia continues to be a major obstetric challenge in present-day of medical practice. It is one of the leading causes of maternal, fetal/neonatal morbidity and mortality worldwide. Pre-eclampsia is a multisystem disorder of pregnancy that is characterized by hypertension (blood pressure >140/90 mm Hg) and proteinuria (urinary protein excretion of >300 mg in 24 h urine) after 20 weeks of gestation in previously normotensive nonproteinuric pregnant women [1,2]. The major cause of fetal compromise in pre-eclampsia is a reduction in utero-placental perfusion [3,4]. Pre-eclampsia occurs in 7–10% of pregnancies worldwide. In India, the incidence is reported to be high, approximately 8–10% of the pregnancies [5].

In cases of nonintervention, pre-eclampsia may progress to the dreaded complications like eclampsia, HELLP syndrome, pulmonary edema, abruptio placentae, postpartum circulatory collapse, acute renal failure, hepatic rupture, cerebral hemorrhage, and visual disturbances, including death. These conditions are also major causes of neonatal morbidity and mortality [6]. Despite considerable research, the etiology of pre-eclampsia remains obscure. Many theories have been proposed to explain the pathophysiology of pre-eclampsia [7]. Pregnancy is a physiological process. In pregnancy, maternal physiological adjustments of the multi-organ system are seen. The adjustments are metabolic, circulatory, and hormonal [8]. The pathogenesis of pre-eclampsia appears to occur in two phases: abnormal implantation of the placenta leading to impaired placental blood flow, which in turn includes the release of pathogenic factors in the maternal circulation resulting in endothelial injury to many maternal organs, especially the kidney, and liver [9]. In pre-eclampsia the liver, kidneys, and brain are mostly affected. Due to autointoxication, functional disorders in these organs are evident [10]. This injury leads to various biochemical changes. The alteration of serum lipid profiles in pre-eclampsia is well documented. An abnormal lipid profile is known to be strongly associated with atherosclerotic cardiovascular diseases. It has a direct effect on vascular endothelial dysfunction [11]. The liver, kidneys, and muscles are the main organs of peripheral deiodination from T4 to T3. The serum concentrations of T4 and T3 may differ in pre-eclampsia from normal pregnancy [12]. There are several potential origins for uric acid in pre-eclampsia. Abnormal renal function, increased tissue breakdown, acidosis, and increased activity of the enzyme xanthine oxidase/dehydrogenase causes a rise in uric acid [13,14]. Creatinine is an anhydride of creatine. Serum creatinine concentrations depend on the balance between their production and excretion. The rise in creatinine in preeclampsia is due to increased production and a reduction in excretion [15]. The above-mention biochemical and hematological parameters affect maternal and fetal morbidity and mortality directly or indirectly. A successful pregnancy outcome requires frequent monitoring of these parameters to avoid complications throughout the pregnancy. Thus, in the present study, we evaluated the serum lipid profile, creatinine, uric acid, and thyroid hormonal concentration in pre-eclampsia women and compared them with those of normal pregnant women.

METHODS

154 patients, 79 pre-eclampsia (cases) and 75 (controls), were selected for the present study by a simple random sampling method during the

study period of March 1, 2016, to August 31, 2017. All patients were evaluated for the lipid profile, thyroid profile, serum uric acid, and serum creatinine.

Type of study
Prospective cross-sectional study.

Period of study
March 1, 2016, to August 31, 2017 (1 year and 6 months).

Place of study
Department of Obstetrics and Gynaecology, Netaji Shubhash Chandra Bose Medical College, Jabalpur, Madhya Pradesh.

Study subjects
Total of 154 patients, including79 pre-eclampsia (cases) and 75 normal pregnant women in The third trimester (control).

Sampling method and source of data-patients
Attending ANC clinics at the Department of Obstetrics and Gynaecology who fulfil the inclusion criteria were selected for the study by simple random sampling after obtaining written informed consent. Study variable, included demographic data, family and personal histories of pre-eclampsia, hypertension, diabetes mellitus, and investigations details were recorded in the study proforma. After consent, 5 mL of blood was drawn under aseptic precaution from the antecubital vein and collected in an EDTA and a plane tube.

OBSERVATION AND RESULTS

In this present study, a total of 154 cases were included. 79 were presented with pre-eclampsia and 75 were included as a control group with a normal pregnancy. They were analyzed and compared using different parameters. The observations are tabulated and analyzed as follows:

Table 1 shows the age distribution among the PE and normal control groups. They were included in four major groups (<20, 20–24, 25–29, and 30–34). In the Mahakoushal region, a high incidence of pre-eclampsia was observed between 20–24 and 25–29 years of age. The mean age in pre-eclamptic patients was 24.51±3.707 years.

This Table 2 shows distribution of tri-iodothyronine (T3) in pre-eclampsia cases and the normal group. It is observed that (31.6%) of pre-eclampsia cases show a higher T3 value and 68.4% show a T3 value normal. The values are compared with normal cases, 52% of cases showing a higher T3 value and 48.0% of cases showing a normal T3 value. The observation suggests a normal T3 value of 68.4% and a 31.6% higher T3 in pre-eclampsiacases cases. The value in comparison to higher T3 was 52% and normal T3 was 48% in the normal control group.

The observation is significant at —p<0.01.

This Table 3 shows the distribution of tetra-iodothyronine (T4) in pre-eclampsia cases and the normal group. It is observed that (38%) of cases of pre-eclampsia show a higher T4 value, and 62% show a normal T4 value normal. The values are compared with normal cases, with 30.7% of cases showing a higher T4 value and 69.3% of cases showing a normal T4 value. The T4 value evaluation is not significant at p>0.05.

Table 4 indicates the distribution of thyroid profile values amongst the pre-eclampsia and control groups for evaluation with mean values. An abnormal Thyroid profile is observed in pre-eclampsia cases in comparison to the normal group. The observation is significant as having —p<0.05.

Table 5 shows the distribution of serum uric acid among the pre-eclampsia and normal groups. High s. uric acid >6 mg/dL was observed in (92.4%) of cases, while only 7.6% of PE showed normal s. uric acid values. In normal group, 51.9% of cases showed a normal s. uric acid value, while 48.1% of cases showed a high s. uric acid value.

The observation is significant as —p<0.0001.

Table 6 shows the distribution of serum creatinine among pre-eclampsia and the normal group. High serum creatinine >1.2 mg/dL was observed in (19.0%) of cases, while 81.0% of pre-eclampsia showed a normal S.

Table 1: Preeclampsia age wise distribution

Age group	Group (%)		Total (%)
	Case	Control	
<20	4 (5.10)	4 (5.30)	8 (5.20)
20–24	37 (46.80)	41 (54.70)	78 (50.64)
25–29	26 (32.90)	24 (32.00)	50 (32.46)
30–34	12 (15.20)	6 (8.0)	18 (11.70)
Total	79	75	154

χ^2=3.007; p<0.05

Table 2: Tri-iodothyronine values in case and control group

T_3	Cases (%)	Control (%)	Total (%)
60–200 ng/dL	54 (68.40)	36 (48.00)	90 (58.40)
>200 ng/dL	25 (31.60)	39 (52.00)	64 (46.10)
Total	79	75	154

χ^2=6.563; p<0.01. T_3: Tri-iodothyronine

Table 3: Tetra-iodothyronine values in case and control group

T_4	Cases (%)	Control (%)	Total (%)
4–12 μg/dL	49 (62)	52 (69.30)	101 (65.60)
>129 μg/dL	30 (38.00)	23 (30.70)	53 (34.40)
Total	79	75	154

χ^2=0.910; p>0.05. T_4: Tetra-iodothyronine

Table 4: Thyroid profile amongs preeclampsia cases and control group

Parameters	Study group (n=79), mean±SD	Control group (n=75), mean±SD	p
T_3 ng/dL	183.20±30.02	195.40±33.72	<0.01
T_4 μg/dL	10.73±2.20	10.56±1.90	>0.05
TSH μIU/mL	5.16±1.50	4.08±0.96	<0.0001

SD: Standard deviation, T_4: Tetra-iodothyronine, T_3: Tri-iodothyronine, TSH: Thyroid stimulating hormone

Table 5: Serum uric acid level in case and control group

Serum uric acid	Cases (%)	Control (%)	Total (%)
1.5–6 mg/dL	6 (7.60)	74 (98.70)	80 (51.90)
>6 mg/dL	73 (92.40)	1 (1.30)	74 (48.10)
Total	79	75	154

χ^2=127.836; p<0.0001

Table 6: Serum creatinine level in case and control group

Serum creatinine	Cases	Control	Total
<0.8–1.2 mg/dL	64 (81.00)	75 (100.00)	139 (90.30)
>1.2 mg/dL	15 (19.00)	0	15 (9.70)
Total	79	75	154

χ^2=15.777; p<0.0001

creatinine value. In the normal group, almost 100% of cases showed a normal S. creatinine value. The observation is significant as —p<0.0001.

Table 7 indicates the distribution of lipid profile values amongst the pre-eclampsia and control groups for evaluation with mean values. An abnormal lipid profile is observed in pre-eclampsia cases in comparison to the normal group. The observation is significant at —p‖ <0.05.

DISCUSSION

Hypertensive disease in pregnancy is a major cause of maternal and fetal morbidity and mortality1. Pre-eclampsia adversely affects the maternal and fetal outcomes due to its wide-spread multiorgan involvement. Pre-eclampsia is a complication in approximately 7–8% of all pregnancies. In our study, we observed a correlation between lipid profile, thyroid profile, serum uric acid, and serum creatinine levels and the development of pre-eclampsia. Hence, estimation of different parameters in the early second trimester can be a cost-effective way of diagnosing pre-eclampsia well in advance, because early identification of pre-eclampsia still remains the cornerstone to preventing maternal and fetal morbidity and mortality due to hypertensive disorders. In this present study, we found Table 1 that 59.1% of patients below 24 year of age and between 25 and 29 years of age developed pre-eclampsia, respectively. This suggests younger patients have a high the incidence of pre-eclampsia. It is of great significance as p<0.0001. Sheraz *et al*. [16] also observed and reported the same finding and stated that PE is more frequent in patients younger than 21 years of age and those older than 35 years. A similar observation was also reported by Duckitt *et al*. [17]. Who found teenage pregnancy to be one of the risk factors for PIH and eclampsia. In this study, pre-eclamptic women showed higher serum concentrations of total cholesterol, low density lipoprotein (LDL), Very LDL and Triglyceride (TG) when compared to normotensive women, which was statistically significant. In this study, a statistically significant decrease in HDLC was observed in pre-eclamptic women. Estrogen is responsible for the induction of TG and HDL. Estrogen levels fall in pre-eclampsia. The low level of HDL is, however, not only because of hypoestrogenemia but also due to insulin resistance [18]. The present study also showed a significant rise in TG in pre-eclamptic patients in comparison to normotensive pregnant women. The other studies that support our study are Vani *et al*. [19], Somani *et al*. [20], and Gohil *et al*. [21].

In the present study, we also found an increased serum uric acid level in pre-eclamptic women, which was statistically significant. Our result is in agreement with other studies (Sayyed and Sontakke, Sunita *et al*., Sirajwala *et al*., Bainbridge and Roberts). Elevated serum uric acid often precedes clinical manifestations of the disease. Bainbridge and Roberts observed hyperuricemia in 75% of women with clinically diagnosed pre-eclampsia [23].

Uric acid, an end product of purine catabolism catalyzed by xanthine oxidase, is filtered, reabsorbed, and secreted by the kidney. In pre-eclampsia, glomerular endothelial lesions lead to diminished renal blood flow and glomerular filtration rate, as well as impaired tubular reabsorption. Hyperuricemia in pre-eclampsia is thus primarily due to decreased renal clearance and increased tubular reabsorption because of the reduction in glomerular filtration rate [24]. On the other hand, an elevated level of uric acid reflects increased uric acid production. In pre-eclampsia due to placental hypoxia, the degree of placental cell destruction increases, which is one of the rich sources of purine for the production of uric acid by xanthine oxidase [25]. This could also explain the increased uric acid concentration. Increases serum creatinine is observed in women with pre-eclampsia. These differences were significant compared with noromotensive pregnant women because pre-eclampsia is associated with a reduction in plasma renin activity and plasma renin concentration if compared with normal pregnancy [26], while the circulating level of angiotensin II may be normal during pre-eclampsia [27]. Other studies found that reducing uteroplacental perfusion pressure could increase renal sensitivity to angiotensin II through a reduction in NO or prostacyclin synthesis or by enhanced formation of TXA2 and endothelin [28].

Table 7: Complete lipid profile evaluation in preeclampsia and control group

Parameters (mg/dL)	Study group (n=79), mean±SD	Control group (n=75), mean±SD	P
TG	243.50±32.33	129.63±18.76	<0.0001
HDL	34.55±5.37	42.20±4.48	<0.0001
LDL	134.44±16.96	83.96±8.64	<0.0001
VLDL	48.48±6.88	29.14±5.08	<0.0001
Cholesterol	221.60±22.55	167.25±8.48	<0.0001

SD: Standard deviation, TG: Triglyceride, HDL: High density lipoprotein, LDL: Low density lipoprotein, VLDL: Very LDL

Thyroid profile			
Study	**T_3**	**T_4**	**TSH**
Kaveti *et al*. [29]	266±101 (ng/dL)	16.53±3.87 (μgm/dL)	3.15±1.57 (min/dL)
Pasupathi *et al*. [30]	185±49 (ng/dL)	12.75±5.08 (μgm/dL)	5.24±2.58 (min/mL)
Das *et al*. [31]	183±39 (ng/dL)	12.71±5.02 (μgm/dL)	5.22±2.48 (min/mL)
Present study	183.20±30.02 (ng/dL)	10.73±2.20 (μgm/dL)	5.16±1.50 min/mL)

TSH: Thyroid-stimulating hormone

Compared to the level of women with normotensive pregnancy, the TT4 level was marginally raised when TT3 was significantly lower (p<0.05). The mean thyroid-stimulating hormone (TSH) level for pre-eclamptic women was significantly higher than that in normal pregnancy (p<0.0001). Dr. Das *et al*., Kaveti [T3, T4, TSH] increased, Khanam *et al*. [32] TSH significant increased, T3, T4 not significant.

CONCLUSION

Nulliparous and low-socio-economic women had a higher risk of pre-eclampsia. Maternal age between 20 and 24 years makes them more likely to develop pre-eclampsia. Serum uric acid levels are significantly higher in pre-eclampsia patients and could be a useful indicator of maternal and fetal complications in hypertensive patients. There is a strong correlation between dyslipidemia and the development of pre-eclampsia. Hence, estimation of the lipid profile in the early second trimester can be a cost-effective way of diagnosing pre-eclampsia well in advance because early identification of pre-eclampsia still remains the cornerstone to preventing maternal and fetal morbidity and mortality due to hypertensive disorders. Hence, we conclude that maternal dyslipidemia in pregnancy is a very good predictor of pre-eclampsia. Pre-eclamptic patients had significantly higher levels of TSH and low levels of T3 and T4 in comparison to normotensive pregnant women. However, the changes in thyroid hormones did not correlate with the severity of pre-eclampsia. Urinary micro albuminuria should be considered an important indicator for pre-eclampsia, so it can be used as a screening test.

ACKNOWLEDGMENT

We are thankful to the whole Pathology and OBGY department, NSCB, Jabalpur; without their support, this study would not have been possible.

AUTHORS CONTRIBUTIONS

CD: Concept and design of the study; aims and objectives; reviewed the literature; prepared the first draft of the manuscript; arranged all the references; and this is his own dissertation work; RU Contributed regarding the conception or design of the study, developing the consent form, data collection, interpreting the results, and manuscript preparation; DD: Concept, coordination, statistical analysis and

interpretation, preparation of the manuscript, and revision of the manuscript.

CONFLICT OF INTEREST

None declared.

FUNDING OF SOURCES

Nil.

REFERENCES

1. Cunnigham FG, Leveno KJ, Bloom SL, Hauth JC, Gilstrap LC, Wenstrom KD, *et al*. Hypertensive disorders in pregnancy. In: Cunnigham FG, Leveno KL, Bloom SL, editors. Williams Obstetrics. 22nd ed., ch. 34. New York: McGraw-Hill; 2005. p. 1237.
2. Sibai B, Dekker G, Kupferminc M. Pre-eclampsia. Lancet 2005;365:785-99. doi: 10.1016/S0140-6736(05)17987-2, PMID 15733721
3. Bellany L, Casas JP, Hingorani AD, Williams DJ. Pre-eclampsia and risk of cardiovascular disease and cancer in later life: Systematic review and meta-analysis. BMJ 2007;335:974.
4. Solomon CG, Seely EW. Brief review: Hypertension in pregnancy: A manifestation of the insulin resistance syndrome? Hypertension 2001;37:232-9.
5. Gohil JT, Patel PK, Gupta P. Estimation of lipid profile in subjects of preeclampsia. J Obstet Gynaecol India 2011;61:399-403.
6. Arias F, Daftary SN, Bhide AG. Hypertensive Disorders of Pregnancy. Practical Guide to High Risk Pregnancy and Delivery. 3rd ed. p. 398.
7. Lampinen KH, Ronnback M, Groop PH, Kaaja RJ. A relationship between insulin sensitivity and vasodilation in women with a history of preeclamptic pregnancy. Hypertension 2008;52:394-401.
8. Kumar A, Ghosh BK, Murthy NS. Maternal thyroid hormonal status in preeclampsia. Indian J Med Sci 2005;59:57-63. doi: 10.4103/0019-5359.13904, PMID 15738611
9. Malarewicz A, Gruszka O, Szymkiewicz J, Rogala J. The usefulness of routine laboratory tests in the evaluation of sudden threat of pregnant woman and fetus in pre-eclampsia. Ginekol Pol 2006;77:276-84. PMID 16875033
10. Rahman MH, Chowdhury MA, Alam MT. Serum thyroxine and triiodothyronine levels in normal pregnancy and pre-eclampsia. TAJ J Teach Assoc 2007;20:6-10.
11. Jayanta D, Mukhopadhyay A, Saha PK. Study of serum lipid profile in pregnancy induced hypertension. Indian J Clin Biochem 2006;21:165-8.
12. Kumar A, Ghosh BK, Murthy NS. Maternal thyroid hormonal status in preeclampsia. Indian J Med Sci 2005;59:57-63. doi: 10.4103/0019-5359.13904, PMID 15738611
13. Stamilio DM, Sehdev HM, Morgan MA, Propert K, Macones GA. Can antenatal clinical and biochemical markers predict the development of severe preeclampsia? Am J Obstet Gynecol 2000;182:589-94.
14. Dekker G, Sibai B. Primary, secondary, and tertiary prevention of pre-eclampsia. Lancet 2001;357:209-15. doi: 10.1016/S0140-6736(00)03599-6
15. Zilva JF, Pannal PR, Mayne PD. Clinical Chemistry in Dignosis and Treatment. 5th ed., vol. 18. London: Edward Arnold;. p. 147.
16. Sheraz S, Boota M, Shahzad S. Eclampsia. Prof Med J 2006;13:27-31. doi: 10.29309/TPMJ/2006.13.01.5052
17. Duckitt K, Harrington D. Risk factors for pre-eclampsia at antenatal booking: Systematic review of controlled studies. BMJ 2005;330:565. doi: 10.1136/bmj.38380.674340.E0, PMID 15743856
18. Opitasari C, Andayasari L. Parity, education level and risk for (pre-) eclampsia in selected hospitals in Jakarta. Health Sci J Indones 2014;5:35-9.
19. Vani I, Gayathri A, Nagamani T, Sunieeta H. Lipid profile parameters in normal and pre-eclampsia complicating pregnancies-a prospective observational study. Ame J Sci Med Res 2015;1:61-6.
20. Somani SG, Somani VC, Babu PS, Vijayalaxmi A. To determine association of lipid profile, serum uric acid and body mass index as a marker for pre-eclampsia. Int J Curr Respir Res 2015;7:
21. Gohil JT, Patel PK, Gupta P. Estimation of lipid profile in subjects of preeclampsia. J Obstet Gynaecol India 2011;61:399-403.
22. Bainbridge SA, Roberts JM. Uric acid as a pathogenic factor in preeclampsia. Placenta 2008;29;Suppl A: S67-72. doi: 10.1016/j.placenta.2007.11.001, PMID 18093648
23. Sayyed A, Sontakke A. Study of lipid peroxidation and antioxidant status in preeclampsia. J Krishna Inst Med Sci Univ 2013;2:69-76.
24. Bargale A, Ganu JV, Trivedi DJ, Nagane NS, Mudaraddi R, Sagare A. Serum hs-CRP and uric acid as indicator of severity in preeclampsia. Int J Pharm Bio Sci 2011;2:340-5.
25. Shah DM. Role of the renin-angiotensin system in the pathogenesis of preeclampsia. Am J Physiol Renal Physiol 2005;288:F614-25.
26. Irani RA, Xia Y. Renin angiotensin signaling in normal pregnancy and preeclampsia. Semin Nephrol 2011;31:47-58. doi: 10.1016/j.semnephrol.2010.10.005, PMID 21266264
27. Redman CW, Beilin LJ, Bonnar J. Renal function in preeclampsia. J Clin Pathol 2000;29:91-4.
28. Kaveti SM, Kumar VS, Lakshmi AR, Audi A, Rajashree TK. Correlation of thyroid and lipid profile in preeclampsia patient. MRIMS J Health Sci 2014;2:84.
29. Pasupathi P, Mathiyalagan D, Rani P, Sankar KB, Kumar SP. Evaluation of serum lipids and thyroid hormone changes in non-pregnant, pregnant and PE women. Thyroid Sci 2009;4:CLS1-6.
30. Das S, Char D, Sarkar S, Saha TK, Biswas S. Evaluation of thyroid hormone changes in non-pregnant, normotensive pregnant and pregnancy with preeclampsia. IOSR J Dent Med Sci 2013;11:16-8.