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Hospitals and Public Health: Conflicting Or Complementing Each Other

Inayat H. Thaver, Nadia Khalid

Hospitals - the original facilities for the sick date back to the temples dedicated to "healing gods" in Egyptians and Greeks. The oldest architectural evidence can be dated back to 9th Century AD in Sri Lanka. The word "hospital" comes from the Latin word "hoses" for host or "hospitium" meaning a place to entertain[1]. While medical schools were established in Greece in the 6th Century BC, there is consensus that the first teaching hospital was founded at Gondisapur in present day Iran in 300 AD Ref). Similarly, the evolution of Public Health can be traced to the origins of various religions wherein purity, cleanliness and hygiene had been part of the religion. The Greek and Indus Valley civilization (3500-1500 BC) also emphasized the sanitation and drainage system and building of wells[2]. However, the concepts of "treating the sick" and "preventing the healthy from getting sick" could not be combined and had been treated as separate entities.

The transition from agrarian society to industrialized society with increase in population including the change in life styles –all have resulted in emergence of both communicable and non-communicable diseases and thus increased morbidity and mortality (the "double burden") [3]. This then followed the "pill for every ill" and "sick care behavior" by the health practitioners, focusing more and more on the treating the diseases either through stationary clinics or through hospitalization. Thus, early days hospital cultures can also be traced back to religious institutions and philanthropy. Some of the countries took the responsibility of maintaining the health of their citizens as one of the social welfare responsibilities; but few in real terms and many only as political slogans. However, due to lack & improper services, the private for profit and not for profit institutions emerged to take care of the sick and fill that gap[4]. Medical students as well as doctors have been taught to battle disease and treat chronic illness to the end; some efforts for promoting community based education and integrated teaching had been made, but mostly the focus has been on treating the sick. In addition, we have amazing technologies and treatments that have been proven to save lives, but our costs and health outcomes do not seem to match what we pour into the system.

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Traditionally public health professionals are focusing on preventing diseases and maintaining good health. Therefore, they have scorned hospitals as the antithesis of community health. The hospital care remains notably distanced from public health practice and policy[5]. Hospitals consume more than 50% of health budget and just over a quarter of population have a contact with hospitals[6]. This can be illustrated with the pyramid that is followed in Pakistan including many other developing countries; the community health forms the basis and tip of pyramid as the tertiary care hospitals. However, the allocation of budget is lop-sided, i.e. more money going towards the curative care as compared to basic primary health care[7]. The problem gets compounded and grave when many of the secondary and tertiary care facilities provide the basic primary care mainly due to lack of an effective and operational referral system. Though we do have an effective primary health care program in all the provinces with the Lady Health Workers and Community midwifes and Basic Health Units (BHUs), but the government health facilities with grass root workers are not fully covering many rural, and most of the urban areas, especially the urban squatter areas and peri-urban areas[7].The urban areas mostly have hospitals which are overcrowded with patients which can easily be managed at primary care settings. The worst scenario is the fact that there is a booming and unregulated private sector including the quacks; the preference for seeking the primary and basic curative care is higher for these practitioners.

Apparently, it appears that doctors working in tertiary care hospitals and public health practitioners focusing on preventive health have conflicting roles. Both the principles and practices of curative and preventive health can play a mutually supportive role if widen our focus of approach to the problem. Unfortunately, in Pakistan the Public Health field has not been fully nurtured (my letter Rise and fall of public health in Pakistan. The role of epidemiologist in infection control at a hospital level is increasing[8]; similarly, identification of an epidemic in the catchment community followed by its management can much better be addressed if we work together. In addition, roles of hospital in PHC has been recognized more than three decades back [8] and now for achieving Universal Health Care (UHC) in a country, hospital can play a major role. Health education and preventive care sessions can play a crucial role for improving the disease pattern. Above all clinician (who are apparently busy and firefighting) can join hands in conducting research, which can improve not only practices but also overall quality of care of the patients.

The first step must be taken by the clinician to open their doors to public health practitioners, by appreciating their

roles and including them as part of their team. Reciprocally, the Public Health practitioners should also be seeking the guidance of their fellow clinicians while targeting diseases interventions. It is thus mandatory that this synergy be considered at both policy as well planning and implementation levels. It has been long recognized that prevention of cancer by having screening programmes, early identification of diseases and raising awareness of the people will not only improve the health status but also save the government lot of money[9] which it would have spent if the preventive services are ignored or side-lined. In addition, all the health cadres, varying from paramedics, nurses, OT technicians, surgeon, specialists etc., can easily apply the public health approach. Let us accept that we get away from a "pill for every ill" approach.

Role of public health practitioners in mass communication, campaigns, raising awareness of the emerging infectious and epidemic diseases cannot be undermined. The pre-service training, i.e under-graduate level training of various cadres of health practitioners is mandatory by having more integrated teaching sessions and bringing the relevance and importance of combined approaches.

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Original Article

Estimation Of Salivary Superoxide Dismutase Level In Oral Submucous Fibrosis: A Clinical And Biochemical Study

Humera Akhlaq, Muhammad OwaisIsmail, Muhammad Abdul Samad

ABSTRACT:

Objective: The objective of the study was to compare the salivary Superoxide dismutase (mU/L) level among stage one patients of Oral sub mucous fibrosis and healthy controls.

Study Design and Setting: It was a comparative cross sectional study design conducted at Outpatient clinic of Dental Department at Ziauddin University Hospital from January 2012 till December 2013.

Methodology: Eighty histo-pathologically confirmed patients of clinical stage one Oral Sub mucosal fibrosis (OSMF) and eighty healthy controls were matched for age and gender to recruit in the study. Socio demographic information (i.e. age, gender, ethnicity, education and occupation), oral health status, measurement of mouth opening (mm) and Superoxide dismutase (mU/L) were recorded. The study was conducted after the approval granted from the ethical review committee of Ziauddin University Hospital. Data was entered and analyzed using SPSS version 21 (IBM).

Result: The standard mean (+/-SD) of Superoxide dismutase (mU/L) in both groups was 4.99 (+/-2.18). The mean of Superoxide dismutase (mU/L) in confirmed cases of oral sub mucosal fibrosis was 3.02 (+/-0.44) which was significantly lower as compared to the control group which was 6.96(+/-1.24) and the calculated p value was 0.001.The strong positive correlation was identified in mouth opening (mm) and superoxide dismutase (0.842).

Conclusion: It was concluded that the levels of salivary superoxide dismutase was significantly lower in the sample of OSMF and lower levels of superoxide dismutase (mU/L) was the indication of decrease in mouth opening among the cases of OSMF. Therefore, salivary superoxide dismutase can be used as a biomarker for the early detection as well as successful treatment of OSMF.

Keywords: Free radicals, Lipid peroxidation, mouth opening, oral submucosal fibrosis (OSMF), salivary, Superoxide dismutase (SOD).

INTRODUCTION:

Oral sub mucosal fibrosis (OSMF) being a chronic, progressive and irreversible disease.[1]The common features associated with OSMF are blanching (marble like appearance) due to impairment in local blood vessels along with the stiffening and fibrosis of any part of the oral cavity.[2] Oral sub mucosal fibrosis is characterized by progressive fibrosis of oral mucous membrane involving soft palate, buccal and lips mucosa, and anterior pillar of fauces.[3] Decrease mouth opening is an adverse clinical outcome associated with OSMF.[3]

Oral cancer is a significant public health concern and account for 2-4% of all malignant tumors worldwide.[4]It is a significant wide reaching health concern in Asia as well. In Southeast Asia, oral cancer accounts for 40% of all cancers.[5]The epidemiological findings indicated as this disease is a concern in South Asia. The inhabitants of the urban population of India reported the prevalence of 0.2% to 1.2% at the dental practices.[6-7]Multiple cases have also been reported among Pakistani population. A retrospective analysis of Karachi population evidenced that oral cancer is the second most common cancer in both genders.[8]

Tobacco intake induces generation of free radicals and reactive oxygen species (ROS).[9]Lipid peroxidation is produced by free radicals and responsible for oxidative degradation of lipids, the end product of lipid peroxidation can be carcinogenic or mutagenic.[10]Superoxide dismutase (SOD) is an antioxidant acts as a defense mechanism by restricting the initiation of free radical chain reactions.[11] At a very initial stage the SOD inactivates the superoxide ion by transforming it into the hydrogen peroxide (H_2O_2) which is being catabolized into dioxygen (O_2) and water (H_2O) by the action of catalase and peroxidase.[11]The production of hydrogen peroxide by the action of SOD triggers the antioxidant defence mechanisms; thereby SOD acting as a key enzyme of natural defense against free radicals.[12-15]

Despite the high prevalence of OSMF in Pakistan, with a rising trend and potential to undergo malignant transformation, OSMF has not been widely investigated

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with respect to lipid per oxidation and antioxidants. Moreover, previous studies have estimated the level of superoxide dismutase (mU/L) from the blood samples. Considering, saliva sample as a cost effective, non-invasive and associated with low discomfort and fear; the present study was conducted to compare the Superoxide dismutase (mU/L) levels in the cases of OSMF stage 1 and the healthy controls.

METHODOLOGY:

A comparative cross sectional study was conducted and the participants were recruited from the outpatient clinic of Dental department, Ziauddin University Hospital Karachi from January 2012 till December, 2013. Eighty cases and eighty controls were matched by following similar criteria of age and gender. OSMF cases were histo-pathologically confirmed as a clinical stage 1 Oral Sub mucosal fibrosis (OSMF). The study was conducted after the approval from the ethical review committee of Ziauddin University Hospital. Written informed consent was obtained from all participants prior to execute the study. The participants were completely briefed about the purpose of the research and procedures involved. The study was conducted according to the ethical guidelines of Helenski declaration and Pakistan Medical research Council (PMRC). Anonymity and confidentiality of the study participants were maintained throughout the research.

The subjects with age greater than18 years, either gender and had addiction of chewing habits (i.e. pan, ghutka and betel nuts) were recruited in this study. Pregnant women and patients with inflammatory conditions i.e. arthritis and periodontal inflammation or with any systemic illness and the subjects which received any prior therapy for OSMF were also excluded.The data was recorded on a pre-designed proforma. Socio demographic information included (i.e. age, gender, ethnicity, education and occupation) were recorded. In addition, oral health status (i.e. burning sensation, addiction or chewing habits, duration and frequency of addiction and habits, brushing frequency) and mouth opening (mm) and Superoxide dismutase (mU/L) were recorded for cases and controls. Early morning saliva samples were obtained from the study participants before that mouth was rinsed with water thoroughly, and this same water was collected in the sterilized container in which PBS solution was dropped for the maintenance of PH. All these samples were kept in storage at temperature of minimal 80^0C. Finally using the Superoxide dismutase level Kit assay, salivary superoxide dismutase (mU/L) level was estimated.

Data was entered and analyzed using SPSS version 21 (IBM). Once the data was entered in the analytical software it was weighted twice for incorrect entries. Qualitative or categorical data was presented as frequency and percentage while quantitative data was presented as mean ± standard deviation. Qualitative variables were compared between cases and controls using chi square statistics. If the assumptions of chi square statistics were not satisfied Fisher exact test was used. Independent t test was used to compare the quantitative variables between cases and controls. Correlation of addiction duration, habit frequency and mouth opening with Superoxide dismutase (mU/L) were performed and correlation co-efficient were reported. For inferential statistics p-value < 0.05 was considered significant.

RESULTS:

The table 1 gives details of the comparison of socio demographic characteristics of cases and controls enrolled in this study. There was no significant difference in mean age in years, gender, ethnicity, and occupation between cases and controls. However, significant difference was found in mean education years, with controls having higher mean years of education (10.65 years) as compared to cases (9.25). Similarly, significant difference was also found in education year categories with greater proportion of controls (45%) attained thirteen or more years of education as compared to cases (15%). The table 2 gives details of the comparison of burning sensation, addiction (chewing habits), duration of addiction in years, habits frequency, brushing frequency and mouth opening (mm) between cases and controls. Significant difference was found in burning sensation, addiction, duration of addiction in years, habits frequency, brushing frequency and mouth opening between cases and controls. Greater proportion cases (67.5%) had burning sensation compared to controls (2.5%) with p-value = 0.001. Moreover, among eighty cases, around sixty three percent were addicted to Ghutka compared to only around thirty three percent among controls. The mean addiction duration in cases and controls were (6.55 Vs. 3.80; p-value = 0.001) with mean frequency habit significantly higher among cases (5.43) compared to controls (3.38). The brushing frequency was significantly lower among cases compared to controls (1.75 Vs. 1.98; p-value = 0.001). Importantly, the overall mean mouth opening among participants enrolled was 36.66 mm, however cases had significantly lower mouth opening (31.10) compared to controls (42.23) with p-value = 0.001. No cases had mouth opening greater than 45 mm compared to around eighteen percent controls in the similar category. The Figure 1 showed comparison of mean Superoxide dismutase (mU/L) between cases and controls. The overall mean (SD) of Superoxide dismutase (mU/L) for participants enrolled were 4.99 (2.18). Cases with confirmed oral sub mucosal fibrosis had mean/SD of Superoxide dismutase (mU/L) as 3.02(0.44) which was significantly lower compared to controls 6.96 (1.24); the difference was significant with p-value = 0.001. The table 3 give details of correlation of addiction duration (years) and habit frequency with Superoxide dismutase (mU/L). For the participants enrolled in this study moderate negative correlation existed between addiction duration in years with Superoxide dismutase (- 0.303); moderate negative correlation existed between habits frequency with Superoxide dismutase (-0.460); and strong positive correlation between mouth

opening (mm) with Superoxide dismutase (0.842). Significant weak positive correlation also existed between addiction duration (years) with Superoxide dismutaseboth among cases (0.274) and controls (0.333).

DISCUSSION:

The present study findings highlighted that cases with confirmed oral submucosal fibrosis (OSMF) had lower mean Superoxide dismutase(mU/L) levels as compared to controls.

Socio demographic Characteristics	Cases (n = 80)	Controls (n = 80)	Total (n* = 160)	P-value
Age in years	22.53 ± 2.76	23.03 ± 3.67	22.78 ± 3.25	0.332
Age Categories				
< 20 years	8 (10)	8 (10)	16 (10)	
20-25 years	64 (80)	50 (62.5)	114 (71.2)	0.016
>25 years	8 (10)	22 (27.5)	30 (18.8)	
Gender				
Male	46 (57.5)	54 (67.5)	100 (62.5)	0.253
Females	34 (42.5)	26 (32.5)	60 (37.5)	
Ethnicity				
Urdu speaking	20 (25)	26 (32.5)	46 (28.8)	
Sindhi	22 (27.5)	14 (17.5)	36 (22.5)	
Punjabi	18 (22.5)	20 (25)	38 (23.8)	0.157
Balochi	16 (20)	10 (12.5)	26 (16.2)	
Pathan	4 (5)	10 (12.5)	14 (8.8)	
Education years	9.25 ± 3.50	10.65 ± 3.78	9.95 ± 3.70	0.016
Education Categories				
= 5 years	24 (30)	12 (15)	36 (22.5)	
6-12 years	44 (55)	32 (40)	76 (47.5)	0.001
= 13 years	12 (15)	36 (45)	48 (30)	
Occupation				
Skilled	48 (60)	50 (62.5)	98 (61.2)	0.871
Unskilled	32 (40)	30 (37.5)	62 (38.8)	

Table 1: Comparison of socio demographic characteristics of Cases and Controls

Oral health status	Cases (n = 80)	Controls (n = 80)	Total (n = 160)	P-value
Burning Sensation				
Yes	54 (67.5)	2 (2.5)	56 (35)	0.001
No	26 (32.5)	78 (97.5)	104 (65)	
Addiction (Chewing habits)				
Pan	10 (12.5)	8 (10)	18 (11.2)	0.001
Ghutka	50 (62.5)	26 (32.5)	76 (47.5)	
Betel nuts	20 (25)	46 (57.5)	66 (41.2)	
Addiction duration (years)	6.55 ± 4.20	3.80 ± 1.17	5.18 ± 3.37	0.001
Habits frequency (packets/ day)	5.43 ± 2.26	3.38 ± 1.17	4.40 ± 2.16	0.001
Brushing frequency	1.75 ± 0.44	1.98 ± 0.27	1.86 ± 0.38	0.001
Brushing frequency Categories				
Once a Day	20 (25)	4 (5.1)	24 (15.2)	0.001
Twice a Day	60 (75)	74 (94.9)	134 (84.8)	
Mouth opening (mm)	31.10 ± 2.48	42.23 ± 3.48	36.66 ± 6.34	0.001
Mouth opening (mm) Categories				
26-35 mm	76 (95)	2 (2.5)	78 (48.8)	0.001
36-45 mm	4 (5)	64 (80)	68 (42.5)	
> 45 mm	0 (0)	14 (17.5)	14 (8.8)	

Table 2: Comparison of Oral health status of Cases and Controls

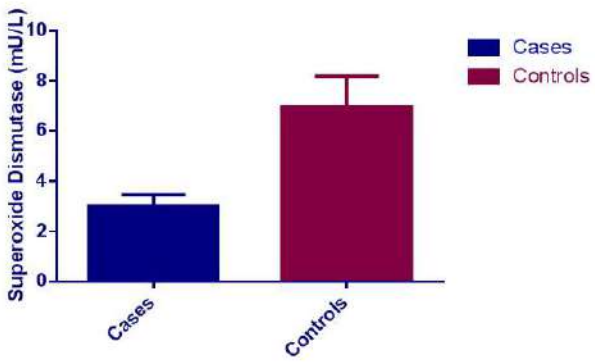

Figure 1: Comparison of Superoxide dismutase (mU/L) between Cases and Controls

Oral health status	Cases (n = 80)	Controls (n = 80)	Total (n = 160)
Addiction duration (years)	0.274*	0.333**	- 0.303**
Habits frequency (packets/day)	-0.034	0.015	- 0.460**
Mouth opening (mm)	0.096	0.024	0.842**

Table 3: Correlation of Addiction duration (years) and Mouth Opening (mm) with Superoxide dismutase (mU/L) among Cases and Controls

Moreover, strong negative correlation between addiction duration in years and habits frequency with SOD was found, thereby indicating increase in addiction duration as well as habit frequency leads to decrease in superoxide dismutase (mU/L) levels. Moreover, strong positive correlation was found between mouth opening and SOD, thereby indicating that increase in Superoxide dismutase (mU/L) levels leads to increase in mouth opening.

In the present study; it was identified that cases with OSMF had mean (SD) Superoxide dismutase (mU/L) as 3.02 which was significantly lower compared to controls 6.96 with the difference being highly significant. The results are consistent with the evidence in the literature. A case control study conducted in a tertiary care setting of India that recruited forty cases (patients with OSF) and forty controls reported that SOD (mU/L) levels were significantly lower among cases compared to controls.[16] The study reported that patients with OSF had mean SOD of 2.46 mU/L while controls had significantly higher mean SOD of 3.46 mU/L.[16]Another clinical study reported a significant decrease in SOD (mU/L) levels in OSF, oral leukoplakia and oral cancer group as compared to the control group.[17]Another clinical and biochemical study reported that SOD (mU/L) levels were significantly lower in cases as compared to controls.[18] A recent study in which venous blood sample was collected to estimate the SOD levels using an ultraviolet spectrophotometer and revealed 204.2 nmol/dl mean serum SOD levels in healthy volunteers whereas in OSMF group mean serum SOD was 82.7 nmol/dl; and the estimated difference was highly significant.[19]The similar study also reported that increase in staging of OSF the mean serum SOD levels significantly decreases thus exhibiting disease progression being associated with decrease in serum SOD levels. The mean serum SOD level for OSF stage 1 (123.4 n/ mol), for OSF stage 2 (88.9 n/ mol) and least for OSF stage 3 (67.7 n/ mol).[19]

The enzymatic as well as non enzymatic antioxidants scanvage lipid peroxidation byproducts formed both under physiological as well as pathological conditions.[20-21] The decrease in SOD levels in OSF can be accounted due to the utilization of these antioxidants by tissues being affected or combating excessive oxidative stress in circulation.[21-24] Thereby, SOD can be a potential biochemical index for evaluating the disease progress.[25-27]

The study had certain limitations. Firstly, cases i.e. of stage one oral sub mucosal fibrosis were recruited only. Secondly, the sample size was limited and was a single centered study. participants were recruited from only Therefore, it is recommended that comparative cross sectional study with greater number of cases and controls should be conducted in future being from multiple clinical sites.

CONCLUSION:

From the present study it was evident that Superoxide dismutase (mU/L) levels were significantly lower among patients with oral submucosal fibrosis. Moreover, progressively decrease in Superoxide dismutase (mU/L) levels leads to decrease in mouth opening among patients with OSMF. Thus, salivarySuperoxide dismutase can be used as a biomarker for the early detection as well as successful treatment and management of OSMF, thereby arresting it at an early stage and reducing the possible consequences of malignant transformation.

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Original Article

Introducing Evidence Based Orthodontics Journal Club Using A Structured Pre And Post Test

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ABSTRACT

Objective: To evaluate the impact of introducing evidence based orthodontics journal club on the performance of postgraduate residents using a structured pre-test and post-test.

Study Design and Setting: The comparitive cross sectional study was conducted among the orthodontic postgraduate residents (n=30) of third year and final year at the orthodontic department at de'Montmorency College of Dentistry, Lahore.

Methodology: Present study was conducted among the orthodontic postgraduate residents (n=30) of third year and final year at the orthodontic department at de'Montmorency College of Dentistry, Lahore. Questions were extracted from the journal club articles. These questions were structured and used in journal club as pre-test and post-test during the academic year 2015-16 and comparison of the performance in the pre-test and post-test over the course of the year was done.

Results: The results of pre-test showed a statistically significant increase during the academic year (p=0.031). Performance in the post-test also showed a statistically significant increase during the academic year (p=0.001).

Conclusion: The redesigning of structured pre and post test in orthodontic journal club resulted in significant improvement in the performance of postgraduate orthodontic residents.

Keywords: *Evidence Based; Journal Club; Orthodontic; Test.*

INTRODUCTION:

The postgraduate training program in orthodontics requires teaching the knowledge and skills necessary for the understanding and critical reviewing of the orthodontic literature and regular journal club is one of the method to accomplish this goal.[1]There is a need to reorganize the regularconduction of these monthly journal clubs because of the ongoing issues like attendance, presentation, and active participation of the postgraduate orthodontic residents.

The journal club is an established teaching modality, usually consists of discussion on scientific articles on regular basis by group of residents in a closed environment.[2]Traditionally the structure of journal club is to present article by one of the assigned residents followed by discussion on it with other residents.[3] Its structure can be improved by redesigning it in such a way that it improves performance of residents at the end of academic year. From which one such way is to re-structure the monthlyjournal club by introduction of some sort of evaluation at the start and end of sessions.

A journal club has often a format used to teach evidence orthodontics to residents.[1]Journal club is a powerful tool to motivate reading behavior of postgraduate residents, and helped in teaching epidemiology and biostatistics to postgraduate residents.[4, 5]Success of journal clubs depends of many factors, such as, regularity, high participation, longevity, availability of food, pre-journal club knowledge of residents, presence of supervisor during journal club.[6-9]

A journal club is an integral part of most postgraduate training programs.[10,11]The advantages of journal clubs are: learner-centric, problem-oriented, relevant to the resident's recent knowledge, relate theoretical component to clinical, active participation of residents, and enable long-term learning.[12-16] Postgraduate orthodontic trainees were surveyed at our institute, as a part of needs assessment, to find out issues with the conduct of monthly journal club. The suggestion was reorganizing of monthly journal club by evaluating the performance of postgraduate residents using a structured pre-test and post-test.

The rationale of the present study is that at our orthodontic center, teachers involved with conduct of these monthly journal clubs noted ongoing issues with the presence, presentation, and active participation of the postgraduate orthodontic residents. The postgraduate trainees were

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surveyed at our orthodontic centre, as a part of needs assessment, to find out issues encountered during monthly journal club. Following this rationale, the objective of present study was to evaluate the impact of introducing evidence based orthodontics journal club on the performance of postgraduate residents using a structured pre-test and post-test.It may also enhance their knowledge and skills.

METHODOLOGY:

Present study was conducted on the orthodontic postgraduate residents (n=30) of third year and final year at the orthodontic department of de'Montmorency College of Dentistry, Lahore. The information of first and second year orthodontic postgraduate residents was collected but not included as they were not informed and trained about such structural changes in the journal club.

TheMultiple choice questions (MCQs)were chosen as a method of evaluating the impact of introducing evidence based orthodontics journal club on the performance of postgraduate residents during the academic year 2015-16. The Multiple choice questions were extracted from the journal club articles assigned to the presenters. The chosen articles were from three orthodontic journals i.e. American journal of orthodontics and dentofacial orthopedics, The Angle orthodontists, and The Seminars in orthodontics. These three journals were chosen because of their well known high impact scores in the field of orthodontics.[17-21]

Well structured MCQs were chosen to demonstrate validity and reliability. Content validity was accomplished through selection of MCQs including the key concepts of evidence based orthodontics. The items were prepared by the head of journal club and prepared both for the pre and post tests.

The data analysis was done using S.P.S.S. version 19.0. Normality of the data was checked. The mean was calculated for each month both for pre and post tests over the course of year.Data analysis was performed by using paired t –test to compare the performance by pre-test and post-test. Results were presented using the mean for each month over the course of the year.

RESULTS:

From the total (n=30) of third year and final year residents of post graduate orthodontic residents; the analysis of the data showed that data were normally distributed so pre-test scores was presented using the mean for each month. Pre-test performance improved from 49% to 77 % over the course of year with the significant linear trend.Results of the pre-test showed a statistically significant increase during the academic year (p=0.031).

Post-test performance improved from 59% to 82 % over the course of year with the significant linear trend. Performance in the post-test also showed a statistically significant increase during the academic year (p=0.001). (Table 1)

Month	Mean Pre %	Mean Post %	% Difference
1	49.34	59.76	18.45
2	45.32	61.45	35.67
3	59.09	73.52	24.55
4	54.78	74.56	32.55
5	61.03	74.56	26.45
6	53.4	72.45	39.43
7	73.45	75.67	49.34
8	71.34	65.42	10.45
9	74.63	75.96	1.56
10	75.21	78.43	4.57
11	76.45	79.54	2.53
12	77.56	82.66	4.79
P value	0.031	0.001	0.039
Mean	58.23	73.24	-
S.D	9.32	18.32	-

Table I: Comparison of scores in the Orthodontic journal club (n=30)

DISCUSSION:

Results of the present study found that pre-test showed a statistically significant increase during the academic year i.e. pre-test performance improved from 49% to 77 % over the course of year with the significant linear trend. This is in accordance with the results of study by Cramer et al., showed that the incorporation of a pre-test/post-test structure resulted in significant increase of the performance of the residents in pre-test during the academic year of Evidence Based Medicine.[22] The pre-test allowed trainees to begin to track in a structural manner and it pointed out the trainees with problems or lack of active participation.

In our study the performance in the post-test also showed a statistically significant increase during the academic year i.e. post-test performance improved from 59% to 82 % over the course of year with the significant linear trend. These results were in accordance with the results of study by Cramer et al., who showed that the incorporation of a pre-test/post-test structure resulted in significant increase of the performance of the residents in post-test during the academic year of Evidence Based Medicine.[22] The post-test allowed trainees to reinforce and facilitate positive change in structuring of journal club.

Limitations of the study were its small sample size, and was conducted at one orthodontic center. Further large scale multi-centric studies with better methodology are suggested.

Our recommendation is that structural changes should be applied in the orthodontic journal club using pre-test and post-test, as it may improve the performance of residents at the end of academic year.

CONCLUSION:

The redesigning of structured pre and post test in orthodontic journal club resulted in significant improvement in the performance of postgraduate orthodontic residents.

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Original Article

Adenoidectomy Before Or After Myringotomy In A Single Stage Procedure: Does The Sequence Matter?

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ABSTRACT:

Objective: To evaluate the sequence of performing adenoidectomy or myringotomy first in a single stage procedure has any bearing on the per- operative outcome on middle ear effusion and subsequent grommet insertion in a patient of otitis media with effusion (OME). The hypothesis was that initially performed adenoidectomy allowed the middle ear fluid to drain passively and precluded grommet insertion.

Study Design and Setting: Comparative study conducted at Department of ENT, Combined Military Hospital Sialkot and PNS Shifa Hospital Karachi, from Jun 2016 to Jun 2017.

Methodology: One hundred and twenty patients (218 ears) with OME and adenoid hypertrophy, meeting the inclusion criteria, were inducted in this study and divided into 2 groups. Group A (60 patients with 110 ears) had adenoidectomy first followed by myringotomy and or grommet insertion, while Group B (60 patients with 108 ears) had myringotomy with or without grommet insertion first and followed by adenoidectomy.

Result: There were 76 males and 44 female patients with a ratio of 1.7:1 and the age range was 3 to 14 years with a mean age of 4.81 ± 0.77 years. There was a significant difference between the two groups. Out of a total of 110 ears in group A, 74 ears (67.2%) had no mucoid fluid or dry tap on myringotomy in contrast with group B where out of total 108 ears, only 26 ears (24.1%) had dry tap or no mucoid fluid (p = 0.001).

Conclusion: Adenoidectomy performed before myringotomy significantly reduced the need for grommet insertion. Larger studies however are needed to corroborate these findings.

Key Words: Adenoidectomy, Grommet, Myringotomy, Otitis Media with effusion.

INTRODUCTION:

Otitis media with effusion (OME) or glue ear is presence of sterile effusion in the tympanic cavity without signs or symptoms of an acute ear infection. It is considered as one of the foremost source of acquired yet preventable hearing loss in the paediatric population, primarily between the ages of 1 and 3 years, averaging 30 dB[1]. It usually occurs as a result of poor eustachian tube function, chiefly secondary to adenoid hypertrophy or following an attack of acute otitis media. Most of the cases resolve naturally in 2 to 3 months, but up to 30% to 40% of the patient may develop recurrent OME and among them 5% to 10% may last longer than 1 year. It is this latter group of patients that require treatment.[2] Patients developing OME earlier in life or those having bilateral or recurrent disease will show effects of impaired reading comprehension along with phonetic and speech articulation difficulties[3].

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The management of OME varies from watchful waiting to conservative treatment to surgery[4]. To date there is no convincing evidence to indicate the benefits of grommet insertion over antibiotic use[5]. Surgical management of OME includes, myringotomy with or without grommet insertion and adenoidectomy if the adenoids are enlarged. There is as yet no consensus on the way surgery for OME/Adenoid hypertrophy/ Myringotomy/ Grommet insertion is performed[6]. Some surgeons perform myringotomy first considering it to be a clean sterile procedure, followed by adenoidectomy; while others do it the other way round.

The hypothesis that adenoidectomy performed first would allow the middle ear fluid to drain passively and preclude grommet insertion prompted us to carry out this study and to evaluate whether the sequence of performing adenoidectomy or myringotomy first has any bearing on the per-operative findings of middle ear effusion and subsequent need for grommet insertion.

METHODOLOGY:

This study was conducted at the Department of ENT, Combined Military Hospital Sialkot and PNS Shifa Hospital Karachi, from Jun 2016 to Jun 2017. The inclusion criteria for the study were patients of both gender with diagnosis of otitis media with effusion (with type B graph on tympanogram) and enlarged adenoids (confirmed on x-ray soft tissue nasopharynx, lateral view) and failure to respond to at least 3 months of medical treatment. The exclusion criteria were OME without adenoid hypertrophy, OME

secondary to acute otitis media, Type A and C on tympanogram, patients with nasal allergy, cleft palate or craniofacial abnormalities, severe retraction pocket and history of previous myringotomy and tube extrusion. The approval of ethical review committee of the institute was taken before the start. One hundred and twenty patients attending the ENT OPD of CMH Sialkot and PNS Shifa Karachi; and meeting the inclusion criteria, were registered for the study.

The objective, process, hazards and benefits of the study were explained; confidentiality was ensured, and informed consent was obtained from the attendants or parents of the patients. A complete history was elicited followed by a thorough and immaculate ENT examination. The presenting complaints, status of tympanic membranes and any signs of effusion in the middle ear were documented. Pure tone audiogram (PTA) was performed only in elder children where possible and tympanogram was performed in all patients. The patients with conductive deafness and type B tympanogram were included in the study. X-ray soft tissue nasopharynx (lateral view) was done in all the patients and adenoid hypertrophy was graded according to the Clemens classification.

The study population was divided into two groups, A and B. In Group A (n=60/ 110 ears), adenoidectomy was performed first, followed by myringotomy with or without grommet insertion. In Group B (n=60 / 108 ears) myringotomy with or without grommet insertion was performed first and subsequently adenoidectomy was performed. Per-operatively the soft palate was retracted using an oro-nasal Foley's catheter and adenoidectomy was carried out by curettage in all the cases. Myringotomy was performed in the antero-inferior quadrant in all ears and grommet was inserted if the mucoid discharge was physically observed coming out from the middle ear cavity. The data was noted on a proforma and all the statistical analysis was done by using SPSS version 23 and a p- value of < 0.05 was considered significant.

RESULTS:

In this study, a total number of 120 patients with a total of 218 ears were included (as in 22 patients only one ear was involved). There were 76 males and 44 female patients with a ratio of 1.7:1. Group A had 36 males and 24 females while group B had 40 males and 20 female patients. The age range was 3 to 14 years with a mean age of 4.81 ± 0.77 years. Fig 1 shows the gender and age distribution in each group A and B. Majority of the patients were between the age of 3 to 6 years (n = 85, 70.8%), 24 (20%) were between the age of 7 to 10 years and 11 (9.2%) were between the age of 11 to 14 years.

Fig 2 depicts the common symptoms by which patient came for consultation. Hearing impairment or inattentiveness in school and home was the most common symptom in 89 patients (74.1%) while mouth breathing, or nasal obstruction was the second common symptom in 81 patients (67.5%). The other symptoms were rhinolalia clausa (n= 78, 65%), snoring (n = 63, 52.5%) and learning problems (n = 38, 31.6%).

Out of 120 patients, 98 (81.6%) had bilateral ear disease (50, 83.3% in group A and 48, 80.0% in group B) while 22 had only one ear disease (10, 16.7% in group A and 12,

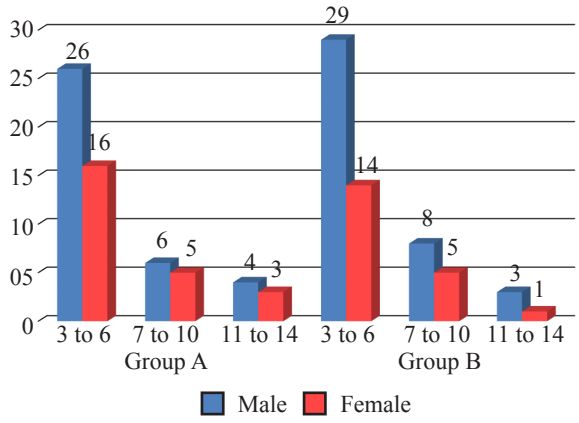

Fig. 1. Age and gender distribution in both groups A and B (n = 120)

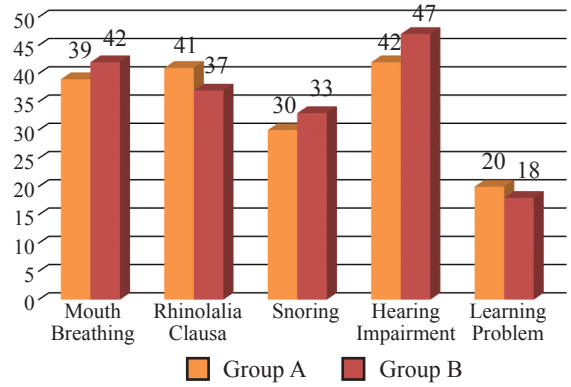

Fig. 2. Presenting symptoms of the patients in each group

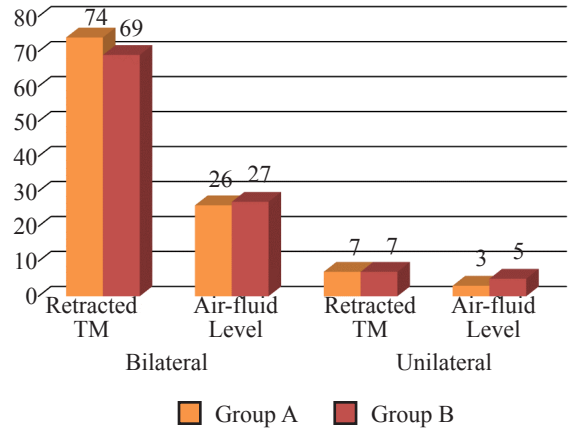

Fig. 3. Otoscopic findings of tympanic membrane in each group (n = 218 ears)

	Mucoid fluid present			Mucoid fluid absent or Dry tap			Grand Total
	Bilateral	Unilateral	Total	Bilateral	Unilateral	Total	
Group A	32	4	36	68	6	74	110
Group B	74	8	82	22	4	26	108
Total	106	12	118	90	10	100	218

Table 1. Per-operative findings on myringotomy in each group A and B (n = 218 ears)

20.0% in group B). On otoscopic examination, dull or retracted tympanic membrane was seen in 157 ears (72%) and air-fluid level was seen in 61 ears (28%). Fig 3 shows the findings of otoscopic examination of the tympanic membrane in each group A and B. On plain x-ray soft tissue nasopharynx (lateral view) adenoid hypertrophy was measured according to the classification of Clemens et al. where grade III hypertrophy was seen in 98 (81.66%) patients and grade II hypertrophy in 22 (18.33%) patients.

Table 1 depicts the overall per-operative findings on myringotomy in each group A and B. It is clearly showing out of total 110 ears in group A, 74 ears (67.2%) had no mucoid fluid or dry tap on myringotomy in contrast with group B where out of total 108 ears, only 26 ears (24.1%) had dry tap or no mucoid fluid ($p = 0.001$).

DISCUSSION:

Otitis media with effusion is a prevalent clinical entity in kids because of eustachian tube dysfunction and enlarged adenoids is the most important cause for dysfunction of the eustachian tube. The ventilation and drainage of natural mucus production in the middle ear is effected during the upper respiratory tract infection until the age of 5 years. The age range in our study was 3 to 14 years and majority of our patients were between the ages of 3 to 6 years which is commensurate with the findings of Timna[7] (3-5 years) and partly with Humaid[8] (age less than 8 years). There is no consensus about the predominant gender involved. Kocyigit[9] claims a female, while Khayat[10] documents a male predominance, consistent with our study. The main complaints in our study are also consistent with other similar studies[11-14]. The Otoscopic findings in our study i.e. predominantly dull or retracted tympanic membrane followed by air fluid level also correspond with most other studies[15,17].

Mechanical obstruction of the eustachian tube is among the chief recognizable causes of eustachian tube dysfunction, particularly due to enlarged adenoids in children. The existence of enlarged adenoids can obstruct the opening of the eustachian tube and leads to negative pressure in the middle ear cavity, and ultimately mucosal transudation. Currently adenoidectomy is one of the most commonly performed pediatric surgical procedure for the treatment of enlarged adenoids. The ratio of size of the adenoid in relation with the size of the nasopharynx is important to assess adenoids enlargement and subsequent surgical removal[14]. Certain studies have stressed the need for screening of the general population for otitis media with effusion as Kocyigit[9] recommends regular screening because almost 16% were positive. Balbani et al[16] noted that hearing loss secondary to OME results not only in impaired reading comprehension but also phonetic and speech articulation difficulties while Butler et al[17] claims that there is no positive implication of screening general population in the first 4 years of life regarding behaviour and language development.

Adenoidectomy is strongly linked with relieving the middle ear effusion in most studies[18,19], while Gates[12] claims that the size of adenoid is not directly proportional to the relief obtained following adenoidectomy. Few studies claim that grommet insertion provides no permanent or significant relief [20] while others believe that not only it drains the effusion but is also cost effective and additionally precludes revision surgery[21-25]. Regarding the effect of the combination of adenoidectomy and grommet insertion, there are both opponents[26,27] and proponents[28-30]**.** However, both schools of thought agree that more extensive and long-term studies are required to be conducted to arrive at a conclusive decision.

Regarding the sequence of surgery in a single stage procedure whether adenoidectomy should be done first or myringotomy first, there are no clear guidelines in the literature and this study is the first of its kind. Removal of enlarged adenoids alone is also recommended in children with OME who had associated postnasal obstruction and chronic rhinosinusitis and have previously undergone surgery for OME. It is reported to be of utmost value in children less than 3 years of age. Adenoid hypertrophy and its closeness to the opening of the eustachian tube is considered as significant factor in the development of OME. The bacterial biofilm around the adenoids and opening of the eustachian tube is important for is dysfunction. The main objective of adenoidectomy in patients of otitis media with effusion is not only to restore patency of the eustachian tube but also to eliminate this biofilm. We have found in our study that proper adenoidectomy causes release of mucoid secretion through the eustachian tube very effectively.

CONCLUSION:

Adenoidectomy performed prior to myringotomy significantly reduces the need for grommet insertion.

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Original Article

Burden Of Cysts And Tumors Around Impacted Third Molars

Muhammad Asif Shahzad, Adnan Aslam, Imtiaz Ahmad, Daud Mirza

ABSTRACT:

Objective: To assess the frequency of cysts and tumors around impacted third molars.

Study Design and Setting: Cross-sectional study was conducted at the OMFS department, Lahore Medical & Dental College, from August 2015 to January 2018.

Methodology: A total of 2057 patients were operated for the removal of 2354 impacted third molars and analyzed for their associated pathological lesions(cysts/tumors). Patients were evaluated with thorough history, clinical and radiological examination. Panoramic and periapical radiographs were primarily used to assess the site of third molar impactions (maxilla/mandible) and their associated pathologies confirmed with histopathological examination. Data was analyzed using SPSS version 20.

Results: The patients' ages were between 17 to 62 years (Mean±SD, 28.12±8.585). There were n=709 (34.47%) females and n=1348 (65.53%) male with female to male ratio of 1.9:1. The mandibular to maxillary impacted third molars ratio was 1.66:1. An overall frequency of 1.36% for cysts and 0.72% for tumor associated with impacted third molars was demonstrated in the current study. The most frequently diagnosed cyst was dentigerous whereas ameloblastoma was the most commonly identified tumor.

Conclusion: A relatively lower frequency (2.08%) of pathological lesions was found around third molars. The study reported a smaller number of pathological lesions affecting a significant minority of patients. It is recommended that a dentist/oral maxillofacial surgeon should be consulted at the earliest if any symptoms in the third molar region arise along with imaging of the area.

Key words: Ameloblastoma, dentigerous cyst, pathological lesions, third molars,.

INTRODUCTION:

The third molar is the most common tooth to become impacted[1]. There have been different hypotheses behind why the third molars are the most commonly impacted teeth[2]. Impacted third molars display a range of anatomical variation in terms of their pattern and position and can lead to a hoist of often diverse pathological occurrences[3,4]. Despite being of little benefit in terms of function, the question whether they should be removed is contentious and the debate is still continues[5,6].

A number of studies conducted worldwide have emphasized on the removal of impacted third molars irrespective of presence of any symptoms or associated pathologies[7,8,9]. While, on the contrary, recent literature have advocated that removal of asymptomatic impacted third molar is not essential, keeping in view the relatively low risk of development of pathologies around these teeth[10,11]. However, another group of researchers concluded that removal of asymptomatic third molars is still controversial and dubious because of paucity of reliable data regarding the frequency of pathologies associated with these third molars[12,13,14].

The objective of this current study was to observe the frequency of cysts and tumors around the impacted third molars

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METHODOLOGY:

This cross-sectional study was conducted from August 2015 to January 2018 at the department of oral maxillofacial surgery (OMFS), Lahore Medical & Dental College, Lahore, a total of 2057 patients were operated for the removal of 2354 impacted third molars. The patients of both gender and age 17 years or older having clinical or radiological evidence of complaint/pathology associated with third molars were included in the present study. While selecting the patients, the normal third molar eruption time/age was kept in view. Patients having historyof maxillofacial trauma, any associated systemic orcraniofacial anomaly or syndrome, were excluded.

The patients, who came to our unit as a primary care setup or referred by dental practitioners, were first evaluated in the Oral Diagnosis department according to the institute protocol. Patients having their presenting complaint associated with impacted third molars and those who showed clinical

& radiographic findings of associated pathology were referred to Oral and Maxillofacial Surgery Department for further evaluation and management.

For each patient, the assessment of impacted third molars and their associated pathologies/radiolucency was done by a thorough history, clinical examination and radiographs, including periapical and panoramic views, according to departmental protocol. CT/CBCT scans were prescribed if needed. On pericoronal radiographs; the radiolucency measuring <4mm, representing follicular space, was taken as normal. Patients' demographic details (age and sex), site of impaction and associated pathologies/radiolucency (maxilla/mandible) were recorded (Figure 1,2).

The patients having clinical and radiographic evidence of impacted third molar associated pathologies/radiolucency were informed about their lesion and an informed consent was taken for the biopsy procedure (incisional/excisional) to obtain the specimen under local anesthesia. For histopathological examination, the specimen was sent to Department of Oral Pathology and only after histopathological report; the diagnosis for cyst and tumor was confirmed.

The data collected was analyzed using SPSS version 20. The qualitative variables like gender, side of impaction, and associated pathologies were presented as frequency and percentages. Quantitative variables were presented as mean and standard deviation. With a confidence interval of 95%, the value of $p<0.05$ was considered significant. We did not apply any inferential test as the study was descriptive in nature.

RESULTS:

Over a 29 months period from August 2015 to January 2018 at the Department of OMFS, LMDC, Lahore, 2354 impacted third molars in 2057 patients were removed.

The patients' ages were between 17 to 62 years (Mean±SD, 28.12±8.585). There were 709 females and 1348 male with female to male ratio of 1.9:1. The mandibular to maxillary impacted third molars ratio was 1.66:1.

There were n=32 cysts (1.36%) and n=17 tumors (0.72%), that were found associated with 2354 impacted third molars.

Among the 32 patients who were identified as having cysts, there were n=14 (43.75%) females and 18 (56.25%) male. The ages of these patients were between 18 to 62 years(mean, 31.19). There were n=9(28.12%) localized in maxilla and 23 (71.88%) in mandible. Out of these, 21 (65.63%) were found to be dentigerous cysts, 09 (28.12%) odontogenic keratocysts and 2 (06.25%) were identified as calcifying cysts (odontogenic) (Table 1&2).

A total of 17 patients were diagnosed as having tumors associated with third molars.There were 9 (52.94%) females and 8 (47.06%) males. Their ages ranged from 17 to 53(mean, 29.47 years). Among the 17 tumors, the maxilla was involved in 3 (17.65%) while the mandible was affected in 14 (82.35%) patients. There were 11(64.71%) ameloblastomas, 02 (11.77%) odontogenicmyxomas, 2 (11.76%) calcifying epithelial odontogenic tumor, 1(5.88%) ameloblastic fibromas and 01(05.88%) odontoma. No malignant tumor was found in our study. (Table 3&4)

An overall frequency of 1.36% for cysts and 0.72% for tumor associated with impacted third molars was demonstrated in the current study. The most frequently diagnosed cyst was dentigerous where as ameloblastoma was the most commonly identified tumor.

Types of Cysts	**(n=32)**	**%**
Dentigerous cyst	21	65.63
Odontogenickeratocyst	09	28.12
Calcifying odontogenic Cyst	02	06.25

Table 1: Frequency Of Cysts Around Impacted Third Molars

	Site			
Gender	**Maxilla** n	**Mandible** n	**Total**	**%**
Female	03	11	14	43.75
Male	06	12	18	56.25

Table 2: Gender and site distribution of cysts

Tumors	(n=17)	%
Ameloblastoma	11	64.71
Calcifying epithelial odontogenic tumor	02	11.77
Odontogenicmyxoma	02	11.76
Ameloblastic fibroma	01	05.88
Odontoma	01	0.588

Table 3: Frequency Of Tumors Around Impacted Third Molar

Gender	**Mandible** (Patients No.)	**Maxilla** (Patients No.)	**Total**	**Percentage**
Female	08	01	09	52.94
Male	06	02	08	47.06

Table 4: Gender And Site Distribution Of Tumors

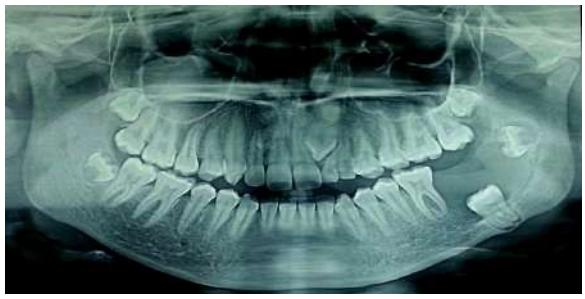

Figure 1: Radiograph showing a radiolucent lesion in left mandible diagnosed as dentigerous cyst after histopathological examination.

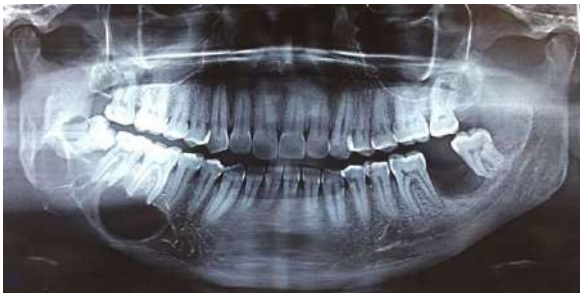

Figure 2: Radiograph showing a multilocular radiolucent Lesion in right mandible diagnosed ameloblastoma after histopathological examination.

DISCUSSION:

Impacted third molars display a range of anatomical variation in terms of their pattern and position, and can lead to a hoist of often diverse pathological occurrences[3,4]. The question whether they should be removed is contentious/dubious and a debate over this still continues[12,14].

Most of the literature regarding the incidence and frequency of cysts and tumors around third molars has shown and documented a significant variation according to the patient's age and the population being studied. The studies performed by Mourshed[15] and Shear[16] have reported and revealed a lower incidence of 1.44% and 0.001% respectively regarding the cysts & tumors around impacted third molars. These recorded findings are in accordance and supported by the studies executed by Goldberg[17] and Chiapasco[18] who demonstrated a frequency of 2% and 1.5% respectively. While Guven[19] (2.31%) and Lysell[20](3%) have almost highlighted the same results. On the contrary, Nordenram[21] (4.5%) and Dachi[22](11%) have reported a relatively higher incidence in their conducted studies. The current study has revealed a frequency of 1.36% of cyst development around impacted third molars. These findings are comparable and corroborated by the studies done by Chiapasco[18] (1.5%) and Mourshed[15] (1.44%) among others who also documented the similar results.

On the other side, the findings of a study done Bruce[23] has exposed that frequency of cyst development around third molars is an age dependent phenomenon which varies significantly among different age groups. He demonstrated and documented an overall incidence of 6.2% with a lower incidence (1.5%) in the younger age group with a mean age of 20 year whereas the highest incidence was recorded in the older age group with a mean age of 46.5 years. Girod[24]in his executed study identified that the progression and eventual development of cysts around third molars is a time related process. He advocated a possible assumption that the impacted third molars persisting for a longer period (2-13 years) might have an increased risk of developing a pathologic lesion (cyst/tumor).

To differentiate a hyperplastic follicle (dental) from an earlier dentigerous cyst, at times, can be difficult as no uniform and definite criteria for their differential diagnosis has been set and popularized and still controversy does exist. In this regard, Kotrashetti[25] conducted a study in 115 asymptomatic healthy patients having 120 impacted third molars. He reported that dentigerous cyst was identified in 1.1% of patients, while odontogenickeratocyst and calcifying cyst (odontogenic) were found in 2.5% and 6.6% of patients respectively. Follicular epithelium was diagnosed to be normal in the remaining patients.

Stoelinga[26], on the other hand, regarding a study on keratocysts hypothesized and revealed that these pathological lesions emanate either from the epithelial proliferation of overlying oral mucosa or offshoots/remnants of dental lamina.

Regarding the incidence and frequency of tumor development around third molars, Lysell et al [20] reported a figure of less than 1%. Their findings were reinforced by Regezi[27] who also found an incidence of 0.14%. The results of Shear[16] (2%) and Weir[28] (2%) were also in harmony with that of Lysell and Regezi. In the current study, we found a frequency of 0.79% for tumor developing around third molars which is in agreement with the results shown by Regezi[27] and Lysell[20].

The development of malignant tumor associated with impacted third molars is a rare phenomenon[29,30]. Only 2 cases of malignant tumors, with an incidence of 0.02%, were recorded in a study done by Guven[19]. However, we were not able to diagnose and find any case of malignancy in our study.

The decision regarding the removal of third molar or not is dependent on the patient compliance as well as local circumstances; which may influence the policy regarding impacted third molars. This in turn will help to prioritize treatment in patients with such pathologies and rationalize decision making in relation to removal of impacted third molars. A regular follow up along with imaging of the third molar region is of utmost importance while adopting a wait and see policy. This fact has been corroborated and advocated by recent literature in worldwide conducted studies. It is recommended that a dental/oral surgeon should be consulted at the earliest, if any symptoms in the third molars region arise, along with regular follow up.

CONCLUSION:

This study concluded a relatively lower frequency (2.08%) of pathological lesions around third molars. An overall frequency of 1.36% for cysts and 0.72% for tumor associated with impacted third molars was demonstrated in the current study. The most frequently diagnosed cyst was dentigerous whereas ameloblastoma was the most commonly identified tumor.The study also highlighted that although a smaller number of pathological lesions were reported but still affecting a significant minority of patients.

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Original Article

Gender Distribution Of Depression Among Undergraduate Medical Students By Using PHQ-9 Scale

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ABSTRACT

Objective: To evaluate the depression on different stages (mild, moderate and severe) among undergraduate students of pre-clinical & clinical settings on the basis of gender.

Study Design and Setting: The cross sectional study was carried out among undergraduate medical students of Bahria University medical and dental College (BUMDC) Karachi from September 2017 – December 2017.

Methodology: The participants were 125 medical students of first year MBBS and 125 students of final year MBBS which were labeled as pre-clinical and clinical groups respectively. The survey instrument used was PHQ-9 scale. This scale divides depression into mild, moderate and severe categories with the help of scores. Percentage of students in different levels of depression was calculated in both the groups.

Results: Mild depression was experienced more (39%) in pre-clinical medical students and (32%) in clinical students. On the other hand clinical medical students showed an upward trend for "severe depression" (19%) as compare to preclinical students (6%) as showed in Table 1. Gender based comparison was done between male and female students in pre-clinical and clinical groups separately. Strikingly, the results showed that females were more depressed than males in both cohorts.

Conclusion: It was concluded that severe depression was equal in males and females in the preclinical group as compared to clinical group. Gender based comparison showed that severe depression was higher in females than males of clinical years whereas mild and moderate depression was also more frequents in females of clinical years.

Key words: Depression**,** Pre-clinical, PHQ-9scale.

INTRODUCTION:

Studying medicine is no doubt a privilege but on the other hand it is considered to be one of the toughest courses. Medical students face sheer mental stress[1, 2] because of various factors including academic burden, clinical responsibilities, assessments and examinations in addition to intellectual demands. All the above stress factors make them prone to poor mental health which leads to major depressive disorders.[3] Depression is a common mental disorder which is expressed as depressed mood, loss of interest or pleasure, disturbed appetite, disturbed sleep, decreased energy, feeling of guilt or low self-worth and poor concentration.[4] Physically the person might show symptoms of vertigo, tachycardia, sweating and tremors. Depressive symptoms may have serious outcomes like dropping out of medical school, drug addiction, and suicidal thoughts.[1] According to Patient health questionnaire 9 (PHQ-9) scores of 5, 10. 15 and 20 represent mild, moderate and moderately severe and severe depression.[5] Ample researchers have observed that young adults studying in medical schools have higher percentage of depressive symptoms than their age fellows studying in non-medical courses.[6] Moreover, an ascent in the severity is seen from first year to final year of medicine.[6,7,8] High physical, intellectual and emotional demands of medical courses augmenting the risk of developing some types of psychological illnesses as major depressive disorders.[9]

The percentage distribution of depression among medical students in public universities has been estimated to 10.4% in Greece[10], 15.2% in USA[11], 24% in UK[12]; 21.7% in Malaysia[9], 29.1% in India[13] and as high as 60% in Pakistan according to Inam et al.[14]

Anxiety and depression were found to be present in 70% and 60% according to a Pakistani study.[15] Previous studies conducted in Pakistan also revealed that higher number of female medical students were screened positive for depression and suicidal ideation as compared to males.[16] It is imperative for medical educators to know the magnitude and causative factors of depression among basics and clinical medical students which not only affect their health and academic performance but may also, have serious consequences like suicide.[17] It is of utmost important that medical curriculum should be formulated on a student friendly which can provide healthy learning environment without imposing extraordinary

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stress on students. This study will create a baseline for educators to identify the gravity of the problem, hence design the curriculum accordingly.[18]

The aim of the study was to compare depression among first year and final year students in clinical and pre-clinical environment based on gender distribution.

METHODOLOGY:

This cross sectional questionnaire based survey was carried out among undergraduate medical students of (BUMDC) Karachi from September 2017 – December 2017.The participants were 125 medical students of first year MBBS and 125 students of final year MBBS. The first 2 years of basic sciences classes have little exposure to clinical teaching and therefore were termed as "preclinical " , while 3rd,4th and final year were termed as "clinical" as they spend ample time in the clinical setting of a hospital. The questionnaire was distributed in their classrooms after the lecture. Out of 250 students, 200 students were in attendance and received the questionnaire. Non-attendees and incomplete responses were excluded from the study. Informed consent was included containing participants right to decline altogether or leave the questions unanswered. The questionnaire did not incorporate name, address or signature of the participants. The study was approved by Institutional Review Committee of BUMDC.

The PHQ-9 instrument derived from the primary care evaluation of mental disorders (PRIME-MD). It is a multipurpose brief self-report tool for screening, diagnosing, monitoring and measuring the severity of depression among patients in a primary care setting.[5] The PHQ-9 questions are about the level of interest in doing things, feeling down or depressed, difficulty in sleeping, energy levels, eating habits, self-perception, ability to concentrate, speed of functioning and thoughts of suicide.[1]

Responses were ranges from "0" (Not at all) to "3" (nearly every day).The total sum of responses suggests the varying levels of depression .Scores were ranges from 0-27. In general, a total 10 or above is suggested as the presence of depression.

RESULTS:

According to this study there were n=54 (27%) males and n=146(73%) females. Mild depression was experienced more (39%) in pre-clinical medical students and (32%) in clinical students. On the other hand clinical medical students showed an upward trend for "severe depression" (19%) as compare to preclinical students (6%) as showed in Table 1. Gender based comparison was done between male and female students in pre-clinical and clinical groups separately. Strikingly, the results showed that females were more depressed than males in both the groups. The pre-clinical female students outnumbered the male students in the category of "no depression", "mild depression" and "moderate depression" except for "severe depression" where the number was equal (3%). Similarly, clinical females were more depressed than the male participants for all the degrees of depression except for "moderate depression" where the difference was not very significant (table 1).

Scale	Depression Level	Pre-clinical			Clinical		
		Male	Female	Total	Male	Female	Total
0-4	No depression	5	23	28	11	20	31
5-9	Mild	12	27	39	8	24	32
10-14	Moderate	5	22	27	7	11	18
15-27	Severe	3	3	6	3	16	19

Table 1: Distribution of pre-clinical and clinical medical students according to PHQ-9 scale on the basis of gender

Comparison between male students of one group with males of the other revealed that a smaller number of pre-clinical students were not depressed (n=5%) than the clinical students (n=11%).

DISCUSSION:

This study surveyed the percentage distribution of depression in medical students in preclinical and clinical years, as very limited data is available in our country on this topic. The demands of undergraduate student training programs in medical schools create an ever increasing burden on students nowadays due to the competition between the medical colleges, over loaded curriculum and clinical rotations in early years of training which results in mental and physical deterioration of health.[18] Our findings showed the evaluation of depression on different stages (mild, moderate and severe) among undergraduate students of pre-clinical & clinical settings on the basis of gender distribution (table 1). It was found that females of both the groups having depressive symptoms outnumbered males of their respective groups (table 1).Systematic studies done in Canada and USA revealed that psychosocial distress was found higher among female medical students.[6] Local studies also show higher female preponderance in severely depressed category[19] Some recent study done in India found that prevalence of depression is higher in males but the difference was not very significant.[20]

Another finding of this study was related to the year of study in the medical school and it was revealed by the results that students of clinical group were more severely depressed than pre-clinical students (table 1).These findings were coherent with the results of previous studies that stress level and extent of emotional disturbances amongst clinical medical students is high[12, 21] and it increases with increasing year of study. These outcomes could be due to academic burden because students have to compete in clinics along with non-clinical schedules. Whereas, another study suggests that anxiety and depression was higher among newly entered students than in senior students. Higher levels of anxiety can be related to the start of new challenging journey of education[14]. The smaller sample size and unicentered study was the limitations of the study. However we need to conduct this study on a larger scale in order to identify and prevent

the causative agents of this stress which might lead to psychiatric disorders in young generation. The utilization of PHQ-9 which was the validated tool was the strength of the study. As this study was done in a single medical university of Pakistan, the results cannot be generalized. Further studies should be done to explore the factors associated with depression.

Currently, medical curricula of our country do not cater personal and behavioral growth of the students. It is recommended for effective learning environment that workshops on stress management and awareness sessions about psychological well-being be conducted in medical schools throughout the country.

CONCLUSION:

It was concluded that severe depression was equal in males and females in the preclinical group as compared to clinical group. Gender based comparison showed that severe depression was higher in females than males of clinical years whereas mild and moderate depression was also more frequents in females of clinical years.

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Original Article

Indications of Primary Cesarean Section In Multipara

Shazia Aftab, Nazish Ali, Fehmida Saleh, Saira Ghafoor, Aasha Mahesh, Sheena Memon

ABSTRACT

Objective: To evaluate the indications of primary cesarean section in multipara and to assess the obstetric outcome including maternal, fetal morbidity and mortality, perinatal outcome.

Study Design and Setting: It was a hospital based study of primary caesarean sections (CS) done on multiparous patients in duration of two years between January 1, 2016, and December 2017 at Jinnah medical college hospital Karachi.

Methodology: Multiparous patients were those who had delivered through vaginal route one or more times (i.e. 28 weeks of gestation or above) or had 1–4children and grand-multiparous are those who had 5 or more children. All the cases included in the study were hospital based and cesarean section was decided by specialist. The procedure was performed by registrars and specialists. The selected patients were followed up till they were discharge from the ward with minimum hospital stay of three days. Data was compiled and results were carried out by SPSS version 23.

Results: During the two years of study period, the number of total deliveries were 2064. The primary CS rate in multipara was 37.17%. These women have more likely to have an emergency cessarean sections compared to elective i.e. 85% and15%. The mean age of women was 29.5 years, booked cases were 72.5% and unbooked were 27.5%. Regarding indications for cesarean sections, non-progress of labour ranked first 25.5% followed by fetal distress 20%, pre eclampsia 12 % and ante partum hemorrhage 10.5% etc. Increase incidence of morbidity and mortality was seen in patients undergoing cesarean section due to different reasons.

Conclusion: Primary caesarean sections in multipara comprise only a small percentage (37.17%) of total deliveries but were related to high maternal and fetal morbidity.

Keywords: *cessarean section rate, indications of cessarean section, multiparous.*

INTRODUCTION:

Cesarean section is the most commonly performed surgical procedures; in many cases it can be life-saving for the mother, fetus or both[1]. Cesarean section is generally performed these days, when a vaginal delivery would lay the baby's or mother's life or health at risk.[2]

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In the past years there has been a significant rise in the rate of caesarean section (CS) in both developed and developing countries rising from about 5% in developed countries to more than 50% in some regions of the world.[3]

According to World Health Organization (WHO) study during the period of 2007–8 the rates of caesarean section in China and other Asian countries were 46% and 27%, respectively[4] in spite of 10–15% suggested by WHO.[10]

With the passage of time there has been a change in the indications for caesarean section and the rate of both primary and repeat caesarean delivery have been on the rise, A study by Emma L Barber et al concluded that primary caesarean births accounted for 50% of the increase in caesarean section rate.[5]

It is essential to evaluate the several indications, maternal and fetal outcome related with a cesarean delivery as several studies have established that cesarean section causes a greater risk of maternal morbidity, and mortality in comparison to vaginal deliveries.[1]

Primary caesarean section in multipara means first caesarean section done in the women who had delivered through vaginal route previously a viable fetus. Since these women (multiparous) have had previous uneventful labours, a sense of false security prevails in them and as a result such multiparous mothers often overlook there regular antenatal checkup and labour. There are still many doctors with an attitude of satisfaction that once a woman had passed through

her first pregnancy and labour, she had practically nothing to worry about her subsequent childbirths.[6] The rapid rise in the rate of cesarean section in the current years warrant serious concern. Pakistan being a developing country have shown an alarming increase in the rate of cesarean section deliveries Haidar G et al from Hyderabad and Shamshad from Abbotabad Pakistan reported caesarean section rate as high as 67.7%[7] and 45.1% in 2007.[8] The rationale of the study was to assess the CS in Karachi.

METHODOLOGY:

It was a hospital based study of primary caesarean sections done on multiparous patients in duration of two years between January 1, 2016, and December 2017 at Jinnah medical college hospital which is a tertiary care hospital. Multiparous patients are those who had delivered through vaginal route one or more (i.e. 28 weeks of gestation or above) or had 1–4 children and grand-multiparous were those who had 5 or more children. All the cases included in the study were hospital based and cesarean section was decided by specialist. The procedure was performed by registrars and specialists. The selected patients were followed up till they were discharged from the ward with minimum hospital stay of three days. The patient's information was collected with the help of doctor present on the duty. The demographic data; included were age, parity, gravidity, maternal medical history; specific information on maternal or fetal pregnancy-related complications; booked and unbooked status, mode of delivery, gestational age (measured according to the last menstrual period), (and it was confirmed by an ultrasound examination within 20 weeks of gestation or by the first trimester ultrasound measurement of the crown-rump length of the fetus), all primary indications for cesarean sections, the newborn's sex, birth weight and apgar score; and the maternal and perinatal outcomes and the need for ICU admission. All adverse maternal and fetal outcomes were recorded. All those females who were primigravida and previous cesarean section were excluded from the study. Informed consent was obtained from all participants. The study was approved by the hospital's research and ethics committee.

Statistical analysis was conducted using SPSS version 23. For continuous variables minimum, maximum, mean, and standard deviation were calculated. Chi-square test was used for categorical variables.

RESULTS:

During the two years total deliveries were 2064 out of which vaginal deliveries were 1278 i.e. 61.91 % and total cessarean section were 786 with rate of 38.08. The multiparous women were 200 in number in which primary cesarean section was done and cesarean section rate came out to be 37.17%.

These women have more likely to have an emergency cessarean sections compared to elective i.e. 85% and15%. The overall incidence of primary emergency and elective caesarean section rate was shown in table no 1. The mean age of women was 29.5 years with range from 15 to 45 years, the 66% of women presents between 26-35 years of age and 72 % presents between 31-40 years. Among 200 multiparous patients which undergoing cessarean section, 87.5% presents with parity 1, 2, 3 and 4 while grand multiparity (5+ births) prevalent in 12.5% of all women. Prevalence of cesarean section according to parity was present in table no 2. Booked cases were 402 (72.5%) and unbooked were 136 (27.5%). The overall indications for cesarean sections were shown in table no. 3, in which non progress of labour ranked first (25.5%) followed by fetal distress (20%) etc.

Increase incidence of morbidity and mortality was seen in patients undergoing cesarean section due to different reasons. The number of patients who had, blood transfusion were fifteen, patients with prolong hospital stay were six due to (wound infection, obstructed labour, blood pressure and sugar monitoring), two patients had obstetrical hysterectomy and forty three babies were admitted in NICU due to fetal distress, neonatal jaundice, hypoglycemia, growth restriction and neonatal sepsis.

DISCUSSION:

A woman who had normal vaginal delivery still may require a caesarean section for safe delivery. The average labor curve continues to change from low parity to multiparity but not toward an ever improved progress[9].

	Frequency	Percent
Total no. of Primary Cesarean sections	538	26.06
Primary Cesarean section in multiparous	200	37.17
Total emergency Cesarean section	456	84.75
Total elective Cesarean section	82	15.24
Primary emergency Cesarean section in multiparous	170	85
Primary elective Cesarean section in multiparous	30	15

Table 1: Frequency of primary caesarean section

Parity	Age / no. of pt	Age / no. of pt	Total no. of pt %
Para1	15-20 (12)	21-25 (17)	29 (14.5%)
Para 2	21-25 (16)	26-30 (24)	40(20%)
Para 3	26-30 (15)	31-35 (41)	56 (28%)
Para 4	31-35 (22)	36-40 (28)	50(25%)
Para 5	36-40 (15)	-	15(7.5%)
Para 6	36-40 (8)	41-45 (2)	10(5%)

Table 2: Parity And Frequency Of Caesarean Section

Indication	Parity						Total	Total multi	%
	Primig-ravida	2	3	4	5	6 and more			
Npol	105	19	20	10	2	0	156	51	25%
PE, Eclampsia	10	5	4	9	4	2	34	24	12%
Iugr	19	2	3	4	0	0	28	9	4.5%
Twin	8	2	2	0	0	0	12	4	2%
Chorioamnionitis	4	0	1	0	0	2	7	3	1.5%
Cpd	15	1	1	2	3	1	15	8	4%
Maternal Request	25	4	1	0	0	0	25	5	2.5%
Boh	0	0	1	1	0	0	2	2	1%
Post Term	15	2	0	0	0	0	17	2	1%
Fetal Distress	70	18	15	5	1	1	116	40	20%
GDM	6	2	5	4	4	2	24	17	8.5%
Failed Induction	25	0	2	0	0	0	27	2	1%
Breech	23	4	3	1	0	0	31	8	4%
Placentprevia	5	2	4	1	2	1	15	10	5%
Placenta Abruptio	7	4	2	2	3	0	18	11	5.5%
Obstructed Labor	1	1	1	0	2	0	5	4	2%
Total	338	66	65	39	21	9	538	200	

Table 3: Indication and Parity Crosstabulation

The primary caesarean sections in multipara comprise small proportion of total deliveries i.e 9.6 % in our study which was relatively less than primary caesarean in primipara, but were actually associated with high maternal and fetal morbidity it is of concern.

In the study primary lscs in multipara constitutes 37.17%, however it is still higher than the World Health Organization recommendation of 15%[10] but is in the range of cesarean sections performed in United States i.e 34 %[2] but low than a highest level of 46% in China[4], and other parts of Pakistan 67.7%[7] and 45.1% in 2007[8]. During labour it is now easier to determine the risks relating to the mother and the baby earlier due to increased use of technology, which can be somewhat related to increase in the amount of cesarean sections.

The rate of emergency caesarean section is much higher 85 % than the elective caesarean section i.e.15 % this is similar to earlier studies in Pakistan[12,13,] Saxena N et al study[11] and Nigeria[14] etc and might be because of the prevalence of such factors as cephalo-pelvic disproportion and prolonged obstructed labour which are diagnosed in labour another probable explanation could be the great aversion to operative delivery in this environment which makes women 'surrender' to surgery as a last result .

The rate of emergency caesarean section is much higher 85 % than the elective caesarean section i.e.15 % this is similar to earlier studies in Pakistan[12,13,] Saxena N et al study[11] and Nigeria[14] etc which might be because of the prevalence of factors such as prolonged labour or cephalo-pelvic disproportion which are diagnosed in the labour are could be the possible explanation for emergency cesarean section instead of instrumental vaginal delivery.

In the present study maximum number of women undergoing primary caesarean section were in the age group of 31-40 years i.e. 72 % and 66% presents bw 25-35 years which is also found in other researches i.e. Partha Saradhi et al study where as In Adnan A. Abu Omar series maximum number of patients were in the age group of < 25 years[15].

Prolonged labor and fetal compromise remained the major indications for emergency cesareans.The commonest indications observed in this study were failure to progress 25.5%, fetal distress 20% , pre eclampsia 12 %, APH 10.5%etc which are similar to findings from other studies. A study conducted in the US[16], in urban Bangladesh[17] and by Boyle A, Reddy UM and Landy HJ, etc[18] concluded the same indications for primary cesarean deliveries. In the majority of our patients i.e 72.5% were booked cases while 27.5% were unbooked which is comparable to 78.4% of Partha Saradhi et al study.

Increase incidence of morbidity and mortality is seen in patients undergoing cesarean section due to various reasons.

In the study postoperative complication rate was 11.5% which is nearer to 12% cases of Sethi Pruthwiraj et al study[1], patients who had blood transfusion were 7.5%, patients with prolong hospital stay were 3% due to (wound infection, obstructed labour, blood pressure and blood sugar monitoring) and two patients had obstetrical hysterectomy due to postpartum hemorrhage. Regarding to neonatal morbidity 21.5% were admitted in NICU due to fetal distress, neonatal jaundice, hypoglycemia, intra uterine growth restriction and neonatal sepsis similar to results observed in other studies[5,1]. There was no maternal death observed in the study. This may be because of the skilled obstetrician attendant at birth,

effective care during labour, management of pregnancy complications, availability of antibiotics, blood transfusion facilities and effective neonatal intensive care and early referral. The limitation of the study were the one public healthcare hospital in one territory. Further analysis in respective areas about indications and prevelance of primary caesarean section in the multiparous may be performed.

CONCLUSION:

Primary caesarean sections in multipara comprise only a small percentage of total deliveries but are related with high maternal and fetal morbidity. Due to previous normal deliveries these woman passes in a subnormal state of health throughout their pregnancy and labour, so they should be emphasized for good antenatal and intrapartum care and expert supervision periodically for any unforeseen emergencies.

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Original Article

Correlation Between Automated And The Westergren Method For Determination Of ESR

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ABSTRACT:

Objective: To determine the correlation between automated and the Westergren method for determination of ESR in symptomatic patients.

Study Design and Setting: This cross sectional study conducted at Hematology Department of PNS Shifa Hospital Karachi.

Methodology: Sample size was determined by using WHO sample size calculator. Westergren method was performed according to ICSH's specifications. ESR vacuum tubes containing sodium citrate were used. The sedimentation of the RBC's was recorded visually after 1 hour using a timer. Automated ESR was performed by TEST1 automated ESR analyzer (ALIFAX SpA, Italy). It uses K3-EDTA or K2-EDTA as an anticoagulant. Bias was controlled by inclusion and exclusion criteria and the confounding factors like temperature of lab and effect of wind were controlled by taking appropriate measures. Pre-designed proforma was used to collect the data that was analyzed through SPSS version 16. Correlation between the two methods was measure through Pearson correlation coefficient analysis.

Results:. The ESR measured by Westergren method ranged from 2 mm/1st hour. to 131 mm/1st hour, with a mean of 39.94±29.60 mm/1st hour. while ESR measured by automated method ranged from 2 mm/1st hour. to 120 mm/1st hr. with a mean of 41.67±32.43 mm/1st hour. There was significantly strong correlation between automated and the Westergren method for determination of ESR (r =0.945; p = <0.001). Similar results were observed across all ages, gender, duration and type of symptoms groups.

Conclusion: **A**utomated and the Westergren method are strongly correlated. Automated method can be used in place of Westergren method for determination of ESR.

Keywords: Automated Method, Erythrocyte Sedimentation Rate, Westergren Method.

INTRODUCTION:

Erythrocyte sedimentation rate (ESR) is being used to evaluate the prognosis and diagnosis of certain diseases. It indicates the chronic inflammation that may be the pathological process in many ailments[1].

Although it is non-specific, but this inexpensive test is commonly used in the diagnosis and monitoring of certain disabling conditions including Multiple myeloma, Tuberculosis, Giant cell arteritis, and Rheumatoid arthritis. Moreover, in Hodgkin's disease it is helpful in prediction of relapse. It also has clinical significance in stroke and prognostic value in coronary artery disease[2,3].

The Wetergren method for ESR measurement was first described by Westergren and Fahraeus, henceit is named so. This method is still considered as a reference standardized method[4]. According to the recommendation of the International Council for Standardization in Hematology (ICSH) Westergren method is the method of choice for ESR measurement[5]. And subsequently introduced techniques for ESR measurement are being evaluated against this method. But, despite being a standard technique, it has certain disadvantages which limit its application. Therefore newer techniques of automated systems for measuring ESR have been introduced to overcome the disadvantages of the original technique.

In Westergren method the use of citrate, a liquid-based anticoagulant may affect the accuracy of ESR readings. The automated method uses EDTA as an anticoagulant, which is more reliable than the primarily used sodium citrate[6,7]. Other modifications introduced in an automated analyzer include the use of closed blood collection tubes, vacuum-controlled aspiration of the sample that more reliably provide a correct dilution of sample with the anticoagulant and automated mixing. The automated ESR method helps in avoiding unnecessary exposure of blood samples and therefore reduces the risk to the sample handlers against the blood borne infections such as infectious hepatitis and HIV, This method also minimize the possibility of external influence like temperature, dust particles, positioning of tube and diluents' ratio on the final reading. It also gives the ESR readings in much shorter time.

Studies have been conducted to compare the results of two

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methods but they show difference outcomes. Some concluded that there is agreement between two methods. While other show significant difference between the readings[8]. The available data is still inconclusive therefor this study was aimed to detemine the coorelation between automated and the westergen method for dertermination of ESR in symptomatic patients.

METHODOLOGY:

This cross-sectional study was conducted at Hematology Department of PNS Shifa Hospital Karachi for ESR evaluation, between Jan 2017 to Nov 2017.

Total 300 blood samples from the patients referred at Hematology Department for evaluation of ESR and fulfilled the inclusion criteria, were selected by Non-Probability, Consecutive Sampling, included in the study. Sample size was calculated according to WHO sample size calculator by applying formula**:** $n = (u + v)^2 \times (SD\ of\ difference)^2 / (\mu 1-\mu 2)^2$

For calculation of sample size, Correlation coefficient was taken as 0.72 with 95 % confidence interval. Further calculation was performed by considering significance level of 5% and the Power of study 90% (Beta error=10%). Correlation coefficient, confidence interval, values of Mean and SD were determined after consulting from published study of similar nature.

Both male and female patients of all age groups suffering from more than three days duration of symptoms including: Fever, malaise, cough, muscular, joint pain or generalized weakness associated with above mentioned symptoms were included in the study.

Blood samples taking more than 30 seconds while collection or samples with excessive venous stasis samples without proper proportion to the anticoagulant were excluded from the study. Furthermore, on visual inspection samples with hyperbilirubinemia, Lipemia or hemolysis were also excluded from the study.

Approval was taken from the ethical committee of the hospital to conduct the study. Written consent was taken from each included patient after explaining them the purpose and procedure of the study. 3ml blood sample was obtained by venipuncture into Tri potassium Ethylene diamine tetra -Acetic acid (K_3-EDTA) vacuum tubes. The samples were thoroughly mixed at the time of venipuncture and also just before analysis. Samples were kept at room temperature and run within the 4-hours period after venipuncture as per ICHS recommendation.

Westergren method performed according to ICSH's specification was considered as standard/reference. Blood was transferred manually from an EDTA tube into ESR vacuum tubes containing sodium citrate. The sedimentation of the RBC's was recorded through visual determination, after 1 hour using a timer.

Automated ESR was performed by TEST1 automated ESR analyzer (ALIFAX SpA, Italy). This analyzer functions on the principle of photometrical capillary stopped flow kinetic analysis. It uses K3-EDTA or K2-EDTA as an anticoagulant.

Bias was controlled by inclusion and exclusion criteria and the confounding factors like temperature of lab, effect of wind were controlled by taking appropriate measures.

Both readings were entered in the proforma along with the demographics and the duration of symptoms. If the patients was already receiving treatment, than it was also documented.

All the collected data was recorded in a proforma specially designed for this study.

All the collected data was analyzed through SPSS version 16. Numerical variables; age, duration of symptoms and ESR value by both the methods have been presented by mean ±SD. Categorical variables i.e. gender and type of symptoms been presented by frequency and percentage. Pearson correlation coefficient analysis has been used to determine correlation between automated analyzer and Westergren method. Data has been stratified for age, gender, symptoms and duration of symptoms to address effect modifiers. Post stratification Correlation coefficient has been recalculated.

RESULTS:

The mean age of the patients was 40.58±20.13 years. There were 188 (62.7%) male and 112 (37.3%) female patients in the study group. Among the causes for measurement of ESR: Malaise was the most frequent symptom, observed in 249 (83.0%) patients followed by fever (77.0%), generalized weakness (62.3%), myalgia (30.0%), cough (25.3%) and arthralgia (15.3%). The duration of symptoms ranged from 7 days to 21 days with a mean of 13.80±4.03 days as shown in Table 1.

The ESR measured by Westergren method ranged from 2 mm/1st hour. to 131 mm/1st hour. With a mean of 39.94±29.60 mm/1st hour, while ESR measured by automated method ranged from 2 mm/1st hour. to 120 mm/1st hour. With the mean of 41.67±32.43 mm/1st hour. There was significantly strong correlation between automated and the Westergren method for determination of ESR (r=0.945; p= <0.001) as shown in Table 2. Similar results were observed across all age, gender and type of symptoms groups as shown in Tables 2 and 3.

DISCUSSION:

ESR is considered as a poorly understood test and despite the advent of newer techniques, no method certainly ruled out the effects of confounding factors like variations in relative erythrocyte volume or shape[9]. But, despite limitations, it remains a widely used test for the screening and monitoring of various conditions that affect plasma proteins and the sedimentation rate.

Characteristics	Study Population n=300
Age (years)	**40.58±20.13**
Age Groups	
• =25 years	65 (21.7%)
• 25-50 years	147 (49.0%)
• >50 years	88 (29.3%)
Gender	
• Male	188 (62.7%)
• Female	112 (37.3%)
Symptoms	
• Malaise	249 (83.0%)
• Fever	231 (77.0%)
• Generalized Weakness	187 (62.3%)
• Myalgia	90 (30.0%)
• Cough	76 (25.3%)
• Arthralgia	46 (15.3%)
Duration of Symptoms (days)	**13.80±4.03**
• =7 days	19 (6.3%)
• 7-14 days	143 (47.7%)
• 14-21 days	138 (46.0%)

Table 1: Baseline Characteristics of Study Subjects

		n	Automated	Westergren	Correlation
ESR(MEAN±SD)		300	41.67±32.43	39.94±29.60	0.945**
Age	<25 yrs	65	30.70±30.94	29.11±27.12	0.960**
	25-50yrs	147	38.05±29.77	36.63±28.01	0.950**
	>50 yrs	88	55.81±33.29	53.47±29.37	0.913**
Gender	Male	188	36.48±32.06	35.42±30.31	0.952**
	Female	112	50.39±31.28	47.52±26.84	0.928**

Table 2: Correlation between Automated and Westergren ESR P=<0.001 ** Correlation is significant at the 0.01 level (2-tailed).

Symptom		n	Automated	Westergren	Correlation
Malaise	Yes	249	43.55±34.11	41.15±30.94	0.946**
	N o	51	32.53±20.37	34.00±21.21	0.944**
Fever	Yes	231	41.95±34.12	39.41±30.85	0.949**
	N o	69	40.75±26.18	41.70±25.09	0.933**
Gen. Weakness	Yes	187	47.39±32.65	45.71±29.37	0.932**
	N o	113	32.22±29.87	30.38±27.56	0.962**
Myalgia	Yes	90	35.62±29.51	34.53±27.68	0.938**
	N o	210	44.27±33.33	42.25±30.16	0.947**
Cough	Yes	76	37.66±32.72	34.80±28.00	0.961**
	N o	224	43.04±32.29	41.68±29.99	0.941**
Arthralgia	Yes	46	50.65±33.53	48.57±32.39	0.937**
	N o	254	40.05±32.02	38.37±28.86	0.946**

Table 3: Correlation between Automated and Westergren ESR across Symptoms

** Correlation is significant at the 0.01 level (2-tailed)

In developed part of the world, the traditional Westergren method is not commonly used in routine laboratories. But, it is considered as the gold standard technique for measuring ESR. Due to various disadvantages, its applications is now limited. To resolve this issue, several techniques to introduce automated systems for measuring ESR have been developed over the last few decades. Some of these involve automation of the Westergren method with diluted or undiluted samples while others use very new technologies. The modification includes the use of undiluted EDTA samples, easier use, closed sample manipulation, conservation of time, vacuum-controlled aspiration of the sample (which intends to provide a correct dilution with the anticoagulant) and automated mixing.

In the last two decades prevalence of infectious diseases such as Hepatitis B and HIV is increasing, their transmission by blood is a cardinal hazard for medical staff. It is therefore imperative to ensure the safety of laboratory staff against these infectious diseases. Automated techniques offer more benefits in reduction of biohazard.

Another advantage of automated techniques is protection against external influence (temperature, dust particles, position of tube, and diluents' ratio) on the final reading. Moreover, in a specified time the number of evaluated samples is higher with this method than the manual method; a significant advantage in any tertiary care hospital with a heavy workload. But in order to substitute the standard ESR method, these automated instruments must be validated against the standard Westergren method.

In this study significantly strong correlation between automated and the Westergren method for determination of ESR (r=0.945; p= <0.001) has been noted. The ESR measured by Westergren method ranged from 2 mm/1st hr. to 131 mm/1st hour with a mean of 39.94±29.60 mm/1st hour. While ESR measured by automated method ranged from 2 mm/1st hr. to 120 mm/1st hour, with a mean of 41.67±32.43 mm/1st hour.

In the previously conducted studies, some authors found good correlation and agreement between two methods while few were noted significant discrepancies in the readings of the two methods. Asif et al. (2012) reported significant strong correlation between Westergren and automated methods (r=0.97, p= 0.00) similar to the present study [9]. Drashti in 2016[10], Kamal in 2018[11] and Hashemi et al. in 2014 (r=0.987; p<0.001)[12], Sönmez et al. in 2014 (r=0.978; p<0.05)[13], Cerutti et al. in 2011 (r=0.816, p<0.05)[14] and Wiwanitkit in 2001 (r=0.98; p<0.05)[15], also observed similar correlation between these two methods. Horsti et al. in 2010 reported relatively lesser correlation (r=0.72; p<0.01) between these two methods[16].

Venapusa[17], in his study determined that automated and Westergren method are correlated at 95%. But, simultaneously he noted higher ESR values with automated method.

Although, he associated his findings with the presence of systemic bias, but failed to determine its nature. Al fadhli[18], observed marked discrepancy in readings between the reference and the automated methods, but mostly with higher ESR readings; While, readings for normal and slightly raised ESR values were about similar by both methods. Similar variations were also noted by Plebani[19]. This study has few limitations: as, it was performed on TEST1 automated ESR analyzer (ALIFAX SpA, Italy), that functions on the principle of photometrical capillary stopped flow kinetic analysis. Therefore, we are unable to comment about validity of other auto analyzers that function on different principles. Furthermore, few samples with very high ESR readings were included so it was difficult to comment on the discrepancies in two methods with higher ESR readings.

It can be thus advocated that automated method is also reliable and can be used for routine estimation of ESR. To determine more reliability on the automated analyzer, further validation experiments and studies, would be required.

CONCLUSION:

The results of the present study determined the significant correlation between the Westergren method and automated method of ESR estimation.

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Original Article

Dental Mangement Of Pregnant Patients: An Obstetrician's Perspective

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ABSTRACT:

Objective: The aim of the study was to investigate obstetricians' perspective regarding dental management of pregnant patients.

Study Design and Setting: It was questionnaire based study and was directed towards practicing obstetricians of Karachi.

Methodology: A seven item questionnaire was formulated by focusing on different aspects of the oral healthcare of pregnant patients including need for regular dental visits, dental radiograph safety, awareness about the adverse pregnancy outcomes due to untreated dental infections, safe use of local anesthesia, antimicrobials and analgesics and the safe trimester for dental extractions. A total of 74 responses received were entered and descriptive analysis was done by SPSS version 16.

Results: Out of 74 respondents, n = 47 (63.5%) were aware that untreated dental infection may lead to adverse pregnancy outcome, n = 20 (27%) actually advise regular dental check-ups to their patients; n = 29 (39.2%) respondents were in favor of leaving the decision for dental x-rays to the dentist and n = 14 (18.9%) respondents allowed dental x-rays anytime during pregnancy to their patients. Out of n = 74, n = 21 (28.4%) respondents permitted dental extractions under local anesthesia with adrenaline by taking some precautions. Co-Amoxiclav (Amoxicillin and Clavulanic acid) was considered the safest antimicrobial by n = 45 (60.8%) respondents whereas Acetaminophen as the safest analgesic by n = 66(89.2%) respondents.

Conclusion: This study concluded that there was a lack of knowledge among obstetricians regarding oral health care of the pregnant patients and this study emphasized the need for oral health care awareness among obstetricians.

Keywords: Adverse pregnancy outcome, antimicrobials, analgesics in pregnancy, periodontal disease, X-rays in pregnancy.

INTRODUCTION:

According to Pakistan Economic Survey 2016-17, the crude birth rate of Pakistan in 2017 was estimated to be 25.2 (per 1000 persons),[1] whereas the World Factbook of Central Intelligence Agency (CIA) estimates it to be 21.9 births/1000 population in 2017.[2] Naseem et al and Gary Armitage listed important oral health problems among pregnant patients in their reviews including caries, gingivitis, periodontitis, tooth mobility and pregnancy oral epulis/tumor; along with the importance of modification in dental chair positioning, prescribed drugs and radiographs during different stages of pregnancy.[3, 4]

Pregnancy results in increased secretion of estrogen by 10 fold and that of progesterone by 30 folds, which in turn leads to a number of changes in different systems of the body including oral cavity.[5] The alterations in cardiovascular system, include increased cardiac output, plasma volume and heart rate. Pregnant patients are also at risk of developing hypertension and diabetes.[6] The microorganisms causing periodontal diseases like gingivitis and periodontitis have the tendency to colonize, produce tissue destruction and escape host defenses in areas distant from the oral cavity.[7,8] Adverse pregnancy outcomes include preterm birth, stillbirth, spontaneous abortion, induced abortion, low birth weight, pre-eclampsia etc.[9]

A systematic review by Ide and Papapanou has shown a significant association between maternal periodontal health and adverse pregnancy outcomes like pre-eclampsia, low birth weight and preterm birth.[10]

Oral health has been found to be neglected among pregnant patients even in developed parts of the world.[11] Thomas and Middleton revealed that only 30% of their study participants had a dental visit once during their last pregnancy.[12] Somewhat similar pattern has also been observed in America.[13]

While working in Oral & Maxillofacial Surgery Department, we have experienced that there are various misconceptions among the patients as well as their attending obstetricians regarding the oral health care leading to avoidance of receiving dental treatment by the patients. Suri and Rao[14] have commented in their study that the obstetricians despite being knowledgeable about the prenatal oral health; did not demonstrate it in their practice behaviors. In a questionnaire

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based study by Hashim and Akbar, 73% participants considered radiographs and 60% considered local anesthesia unsafe during pregnancy.[15]

Delay in dental treatment may lead to adverse pregnancy outcomes but these can be easily avoided by an adequate advice to the pregnant patient from the attending obstetrician with consensus from dental professionals. Curtis et al[16] have also emphasized on the importance of a consensus statements regarding prenatal oral health through inter-professional collaboration in order to provide better dental care to pregnant patients. Therefore the purpose of our study was to investigate the obstetricians' knowledge and point of view regarding different aspects of oral healthcare including regular dental visits, dental radiographs, use of local anesthesia, use of antimicrobials/analgesics and dental extractions during pregnancy.

METHODOLOGY:

The questionnaire was directed towards practicing obstetricians. The seven closed ended items addressing various aspects of dental management of pregnant patients included need for regular dental visits, prescribed dental radiographs, prescribed drugs, awareness about the adverse pregnancy outcomes due to untreated dental infections, awareness about the safe use of local anesthesia, antimicrobials and analgesics and the safe trimester for dental extractions. The questionnaire with a consent form was approved by the Ethical Review Board of Hamdard University. The questions were pilot tested and validated from five practicing dentists and obstetricians for clarity and reproducibility. The corrections from obstetrician were incorporated in the final questionnaire. Total 80 hard copies of the questionnaire were distributed by hand among the obstetricians from different hospitals in Karachi. It was also uploaded on Google Forms to get maximum number of responses[17]. In our study; trainees in Obs/gynecology department and consultants who were providing prenatal care regardless of their duration of experience and involvement in teaching were included in this study. Extensive literature search showed similar studies on the prenatal healthcare workers including midwives who were practicing with gynecologists/obstetricians at all levels.[5, 11, 14, 15, 16, 21, 22, 33] Total number of responses were n = 74 out of 104 forms distributed manually and electronically. Data was entered and descriptive analysis was done by SPSS version16.

RESULTS:

The response rate of the study was 71%. In our study, out of 74 respondents, n = 47 (63.5%) were aware of the possibility of adverse pregnancy outcome as a result of dental infections and n = 20 (27%) practically advise regular dental check-ups to their patients (Table 1). Regarding the safest time during pregnancy for dental x-rays, n = 29(39.2%) respondents were in favor of leaving that decision to the dentist followed by n = 14 (18.9%) respondents who would allow dental x-rays anytime during pregnancy (Table 2).When asked for the permission of dental extractions under local anesthesia with adrenaline during pregnancy, n = 30 (40%) of the participants were not sure, followed by n = 21 (28.4%) of the respondents who would allow this procedure with some precautions (Table 3).

Amoxicillin/clavulanic acid was considered as the safest antimicrobial by n = 45 (60.8%) respondents (Figure 1) and Acetaminophen as the safest analgesic by n = 66 (89.2%) respondents (Figure 2).

DISCUSSION:

Our study was focused on the knowledge and behavior of obstetricians towards their patients' oral health. Relationship of poor oral health and adverse pregnancy outcomes including pre-eclampsia, low birth weight and preterm birth have been vastly researched worldwide but limited researches were found in our local literature.

Han[18] conducted a study based review on this subject. The epidemiological studies in his review showed dual predictors namely periodontal disease and presence of microorganisms including Lactobacilli and Actinomyces species in the saliva for the adverse pregnancy outcomes. His survey of mechanistic studies showed that adverse pregnancy outcomes can be the result of intrauterine infection but the source of this infection could be oral bacteria namely Fusobacterium Nucleatum instead of vaginal microflora.

Harjunmaa et al conducted the study on 1391 pregnant patients and found that patients having periapical infection had significant short mean pregnancy duration, infants with low mean birth weight, neonatal length and neonatal head circumference as compared to the infants of mothers free of periapical infection.[19]

The risk of adverse pregnancy outcomes due to untreated dental infection puts heavy responsibility over the obstetricians/prenatal health care providers. They have to ensure that the pregnant female should have disease free oral cavity during pregnancy.

In our study, out of n = 74 respondents, n = 47 (63.5%) were aware of the possibility of adverse pregnancy outcome due to compromised oral health and only n = 20 (27%) practically advised regular dental check-ups to their patients. Morgan et al[20] revealed in an obstetrician directed questionnaire based study that although 84% of their study participants acknowledged the relationship between the periodontal disease and adverse pregnancy outcome,73% of them had never inquired their patients about their routine dental checkup during last 12 months and 38% participants did not advise their patients for routine dental checkup during pregnancy. Similarly Patil and Thakur in their questionnaire based survey commented that gynecologists' inability to appreciate the impact of poor oral health on pregnancy outcome is because of lack of knowledge (38%) and time (28%) devoted for this purpose.[21]

Decision regarding dental x-rays during pregnancy is a complicated issue and the obstetrician as well as patient should be educated in this context. There are mainly two factors for avoidance of dental treatment during pregnancy namely patient's fear of the dental treatment and obstetrician's concerns regarding congenital defects in newborns developing from x-ray exposure. In our study, when inquired about the safest time for dental x-rays during pregnancy; n = 29 (39.2%) respondents were in favor of leaving that decision to the dentist and n = 14 (18.9%) respondents permitted their patients for dental x-rays anytime during pregnancy. In a questionnaire-based survey by Strafford et al[22]regarding the awareness of dental health during pregnancy among patients, dentists and obstetricians, 92% of the obstetrician group agreed upon the provision of dental x-rays throughout pregnancy with proper shielding and collimation.

According to National Commission for Radiation Protection (NCRP); the recommended cumulative radiation exposure to fetus is not to exceed the limit of 0.2Gy to prevent congenital defects during pregnancy.[23] Abbott[24] commented that 32nd to 37th day (4-5 weeks) of gestation is related to the organogenesis and is the most sensitive period for radiation damage to fetus. A significant radiation exposure for fetal damage is considered in two ways. First the developing fetus should be in the path of radiation and second a radiation dose would exceed 10 μ Sv. The fact that an intraoral periapical radiograph has an effective dose of 4 μ Sv and a panoramic film has 7 μ Sv; therefore dental x-rays should be allowed if indicated and with proper precautions like shielding and collimation to avoid legal issues.[13]

Dental radiographs with proper shielding and collimation have been considered safe during pregnancy by the Committee of American College of Obstetricians and Gynecologists 2013[13] and the evidence based practice guidelines by California Dental Association Foundation and supported by CDA.[25]

In our study while inquiry about the permission to carry out dental extractions under local anesthesia with adrenaline during pregnancy, n = 30 (40%) of our participants were not sure and n = 21 (28.4%) of the respondents permitted it with some precautions. Various factors should be considered when a pregnant patient is advised for dental extraction including the type/dose of local anesthetic and vasoconstrictor agents. In addition, the stage of pregnancy should also be considered. Lidocaine lies under category B of pregnancy risk which means it poses no danger on humans.[26] It is recommended that the dose of lidocaine with adrenaline should be kept below 500mg.[27]

Ample literature recommended dental extractions as a safe procedure with proper positioning in 2nd and 3rd trimester of pregnancy, as untreated infected teeth may lead to more serious complications.[3, 13]

In our study, when participants were asked about the safely prescribed medications during pregnancy for dental treatment; Amoxicillin/clavulanic acid was considered as the safest antimicrobial drug by n = 45 (60.8%) respondents and acetaminophen as the safest analgesic by n = 66 (89.2%) respondents.

Cyclooxygenase is a dilator for ductus arteriosus and pulmonary resistant vessels. It is inhibited by NSAIDs which result in premature closure along with reduced perfusion of fetal kidneys.[28] This is the reasons why Ibuprofen and Naproxen are avoided during 2nd and 3rd trimesters of pregnancy.

Although acetaminophen is considered safe during pregnancy by FDA.[29] Toda K[30] in his review article discussed the rare possible complications of using acetaminophen during pregnancy like autism spectrum disorders, neuro-developmental problems and lower performance intelligence quotient. It was also emphasized in his study that acetaminophen should be used with caution and minimum possible dose and duration as it is the safest analgesic available at the moment during pregnancy.

It has been recommended by multiple resources that amoxicillin plus clavulanic acid, cephalosporin and clindamycin may be used safely during pregnancy along with the metronidazole if required.[25,31, 32]

George et al mentioned that the obstetricians should be aware with of the importance of oral health screening but were unable to perform this on their own due to lack of training and also because they prioritized other health issues over their patients' oral health.[11] Jackson et al[33] while conducting a prenatal oral health program (pOHP) involving faculty members, residents and third year medical students, highlighted the importance of collaborative approach for providing prenatal healthcare for better management of pregnant patients in terms of their oral health.

Despite of using electronic means for distribution of questionnaires, the number of responses we received was less than our expectation; which was the limitation of our study. It was recommended that there is a dire need for oral health awareness of the pregnant patients among entire workforce of prenatal care providers.

CONCLUSION:

This study concluded that there was a lack of knowledge among obstetricians regarding oral health of the pregnant patients.

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Original Article

Localization Of Cyst Of Myelomeningocele Among Pediatric Patients

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ABSTRACT:

Objective: To determine the frequency distribution of myelomeningocele cyst location among pediatric patients at a tertiary care hospital of Karachi.

Study Design and Setting: This cross sectional study was conducted in outpatient department of neurosurgery ward of JPMC.

Methodology: All fifty male and female babies having myelomeningocele, newly born to more than 12 months of age, who presented in outpatient department of neurosurgery ward of JPMC during the period of six months were included in the study. Verbal informed consent of the parents was taken and babies were examined for the location, size and associated complains of myelomeningocele.

Results: The patients presented mainly in the age group of newly born to 3months. Majority of them were males. Atonic bladder and bowel along with paralysis of lower limbs were uniformly found associated features. Among male patients distribution of myelomeningocele was 3% each in cervical and thoracic while 96% in lumbar region whereas female patients had 6% cervical and 84% lumbar cysts with no cyst in the thoracic region. The average size of MMC cyst in lumbar region was 4.0×4.2 cm.

Conclusion: Cyst of myelomeningocele was found to be more in male children up to 3months of age with location in the lumbar region. The average size of cyst was 4.0×4.2 cm and accounted for atonic bladder and bowel along with paralysis of lower limbs.

Keywords: Atonic bladder, Cyst, Gender, limb paralysis, location, Myelomeningocele

INTRODUCTION:

The leading cause of infant mortality is due to congenital anomalies.[1] A group of birth defects characterized by failure of fusion of midline structure is referred to as Spinal Dysraphism.[2] These anomalies are usually referred to as Neural Tube Defects (NTDs). It is the most frequent abnormality and a vital issue of well being in children.[3] It occurs due to inappropriate neural tube closure. Neurulation is an embryonic process in which a notochord forms a broad, uniform neural plate. The neural plate in turns, wrap inside to form a neural tube. It is this neural tube which is the precedent to the central nervous system, later on forming brain and spinal cord.[4] The growth and closure of neural tube occurs after 28 days of fertilization. Neural tube defects (NTD) occur due to inappropriate closure of neural tube.[5] NTDs are usually referred to as "Spina Bifida" a defect in the vertebral arches, may be covered by skin, meninges or underlying neural tissue. Myelomeningocele (MMC) is the most commonly occurring form of spina bifida in which meninges and cerebrospinal fluid along with neural elements buldge out through the sac.[6] Many factors are involved in the etiology of disease, of them, folic acid deficiency has been found to be the common cause.[7] Various authors have published different frequency figures regarding abnormalities of central nervous system varying from 1.3% in Pakistan[8,9] to 8.8% in Tanzania.[10] Worldwide prevalence of NTDs varies from 0.5-10 per 1000 live births.[11] The prevalence of the disease has greatly been reduced worldwide by periconceptional use of folic acid.[12] The cyst of myelomeningocele could be present on any part of vertebral column.

Since much literature is not available on the current topic at local level therefore present study was designed to determine the frequency distribution of myelomeningocele cyst location among pediatric patients.

METHODOLOGY:

This cross-sectional study was a part of "Molecular variations of Myelomeningocele in relation to VANGL1 gene". After approval of IRB letter# *IRB-556/DUHS/Approval/2015/130* of Dow University of Health Sciences, male and female children with ages from newborn to more than 12 months having MMC were enrolled from Neurosurgery O.P.D. of Jinnah Postgraduate Medical Center (JPMC) Karachi. This

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study was conducted for a period of six months. All fifty cases who presented with rare problem of myelomeningocele were included in the study after verbal informed consent of the parents during the specified period.

Children were subjected to physical (local) examination of the cyst regarding location, size and associated complains of myelomeningocele.

RESULTS:

The number of patients between ages 0-3, >3-6, >6-9, >9-12 and >12 months were 20, 15, 7, 5 and 3 respectively. The frequency of male patients was 31 and that of female patients was 19 (Table 1). The cyst of MMC was mostly located at lumbar region (92%) with mean size 4.0×4.2 cm followed by cervical cysts (6%) having mean size 2.6×3.6cm and thoracic region (2%) having mean size 4×4cm (Table 2). The uniformly associated features found in all patients with MMC were atonic bladder, bowel and lower limb paralysis. The age wise distribution of patients having myelomeningocele at lumbar region is, 20 patients newly born -3months, 14 patients >3-6 months, 7 patients >6-9 months, 6 patients > 9-12 months and 3 patients were >12months of age. The gender wise distribution of patients of myelomeningocele was 3% male patients each had cervical and thoracic cysts while 96% had lumbar myelomeningocele whereas regarding female patients 6% had cervical, 84% lumbar and no cyst in thoracic region (Table 3).

S.No.	Parameter	Frequency	%
1	**Age in months**		
	Newly born-3	20	40
	>3-6	15	28
	>6-9	7	14
	>9-12	5	12
	>12	3	6
2	**Gender**		
	Male	31	62
	Female	19	38

Table 1: Age and Gender wise Distribution of Study Patients N=50

Location	Frequency	%	Mean Size(cm)	Associated features
Cervical	3	6	2.6×3.6	Atonic bladder, bowel & lower limb paralysis
Thoracic	1	2	4×4	Atonic bladder, bowel & lower limb paralysis
Lumbar	46	92	4.0×4.2	Atonic bladder, bowel & lower limb paralysis

Table 2: Location, Size & Associated features of Myelomeningocele N=50

Gender	Cervical	Thoracic	Lumbar
Male	1(3.2%)	1(3.2%)	30(96%)
Female	2(6.4%)	0	16(84%)

Table 3: Gender wise distribution of myelomeningocele

DISCUSSION:

Neural tube defects (NTDs) comprises of fatal anomalies leading to lifelong disabilities and death of babies. Myelomeningocele has been reported as the most diagnosed cases and is related with the greatest degree of impairment among NTDs.[13] Myelomeningocele (MMC) is an inborn error of central nervous system (CNS) which occurs due to inappropriate closure of spinal column and the neural elements buldge out in the form of pouch through the bone and skin. Worldwide prevalence of neural tube defects is 1-10 per 1000 live births.[14]

The most common age group at which the babies presented in our study was newly born to 3months. Majority (46) of the patients had cyst on the lumbar region followed by cervical and thoracic regions. Results are in consistent with study conducted in 2016, *Ullah W* suggested lumbar region as the most common site followed by sacral, thoracic and cervical regions.[15] Other authors have also documented the same.[16-19] In contrary to this, *Asindi* found thoraco lumbar being the commonest site.[20]

Patients suffering from MMC usually have sensory and motor neurological defects underneath the lesion. It may lead to weakness of lower limbs or paralysis that hinders or restrain from walking and the chances of pressure sores increases due to lack of sensation.[21] The issues of bowel and bladder incontinence are frequent because the desire for defecation is although vanished but the recto anal inhibitory reflex is sustained.[22] Due to paralysis of external anal sphincter, fecal soiling is inescapable when internal anal sphincter relaxes.[23] This is coinciding with our study results as all fifty patients had complains of atonic bladder and bowel along with paralysis of lower limbs.

The associated features were atonic bladder, bowel and lower limb paralysis present in all patients irrespective of cyst at different sites in our study. Schletker has supported the view of such patients suffering from neurogenic bladder and bowel.[24] This dysfunction occurs virtually in all children having MMC irrespective of the site of location of the cyst. In case of bladder dysfunction there is failure of urine storage or failure to empty the urine. This in turn may be related to bladder itself or with the external sphincter of bladder or with both. Consequently there is increase risk of urinary tract infection following failure to empty the bladder properly and adequately. This sequelae culminates overtime into urinary reflux, hydronephrosis , renal damage and ultimately renal failure. Similar findings are reported by other researchers.[25,26] The findings of neurogenic bladder and bowel are consistent with our study results. We have found that the most common age period of patient presentation in the tertiary care hospital with lumbar myelomeningocele was newly born to 3 months. The same is the finding of *Chand MB*.[16] This could be justified by the statement that MMC is a congenital anomaly so patients usually present in the hospital settings with such little age.

Majority were males (31%) in this study. Hidrosefalis[27] and Ghani[28] supported the view of males being more commonly affected by myelomeningocele than females. Whereas Nnadi DC[29] and Sachdeva S[30] have documented that females were more commonly affected than males with neural tube defects in their studies. Pre-pregnancy counseling and administration of folic acid throughout pregnancy, awareness regarding neural tube defects and myelomeningocele in the community through print and electronic media are small steps that can play a major role in combating this problem. Early and timely referrals of such babies to specialized units can also improve the quality of life of these patients. Large multicenteric studies on this subject are open avenues for future research in our country.

CONCLUSION:

Frequency distribution of cyst of myelomeningocele is found to be more in male children in the age group of newly born to 3months with location in the lumbar region. The average size of cyst was 4.0×4.2 cm and accounted for atonic bladder and bowel along with lower limb paralysis.

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Original Article

Effect Of Withania Coagulans And Liraglutide On Serum Glp-1, Postprandial And Fasting Blood Glucose In Streptozotocin Induced Diabetic Rats

Abdul Samad, Noor Nasir Rajpoot, Hira Ayaz, Noman Sadiq

ABSTRACT

Objective: To evaluate effect of Withania coagulans and liraglutide on serum Glucagon like peptide-1, Postprandial and Fasting Blood Glucose levels in streptozotocin induced diabetic rat.

Study Design and Setting: This randomize control trile was conducted at Islamic International Medical College in collaboration with National Institute of Health Islamabad.

Methodology: This randomized controlled study was performed on a total of forty male Sprague dawly rats, which were initially divided into two groups; Group A (n=10) and Experimental Group (n=30). Diabetes in the Experimental group B was induced by intraperitoneal administration of streptozotocin for 5 days (30mg/kg/day). Diabetes was checked in experimental group by measuring fasting blood glucose (mg/dl) on day 6. Experimental group was further divided into Group B (Diabetic control), Group C (Withania coagulans-treated) and Group D (Liraglutide-treated). Blood sampling was done at day 30 and serum GLP-1, postprandial and fasting blood glucose levels were measured and compared in all groups.

Results: Fasting and postprandial blood glucose levels of group C and D were significantly reduced as compared to group B. Serum GLP-1 levels were significantly increased in group C and D as compared to group B.

Conclusion: Withania coagulans reduces hyperglycemia in diabetic rats through increasing GLP-1 hormone.

Keywords: Diabetes, GLP-1, Liraglutide, Withania coagulan

INTRODUCTION:

Diabetes mellitus is a metabolic disorder affecting protein, fat and carbohydrate metabolism with a global prevalence of 8.8%.[1] It is characterized by relative or absolute lack of insulin leading to hyperglycemia.[2] Type 2 diabetes is a chronic disease in which an individual exhibits a decrease in pancreatic beta cells function, insulin resistance and failure to inhibit postprandial glucagon secretion.[2] Diabetes mellitus is a global health issue which if left untreated leads to serious morbidity and mortality.[3] Studies have shown that insulinotropic function of incretin hormones especially Glucagon like Peptide-1 (GLP-1) are severely diminished in type 2 diabetes causing postprandial and fasting hyperglycemia.[4] Various groups of drugs are currently in use for treating type 2 diabetes like sulphonylurea, biguanides and thiazolidinedione.[5] These group of drugs have several clinical limitations, the most serious one is the eventual need for insulin replacement therapy. Since 2007 a new class of drug, known as GLP-1 mimetics (liraglutide and exenatide) has been in practice for treating diabetes. GLP-1 mimetics targets and enhances incretin hormone (GLP-1) and have shown remarkable result in treating type 2 diabetes.[6] Liraglutide, in addition to controlling hyperglycemia also upsurges glucose dependent insulin secretion (GSIS) from pancreatic beta cells.[7] However high cost and parenteral administration are the two main factors which limits the use of these remarkable GLP-1 analogues.[8]

Due to the risk profile of antidiabetic agents, herbal medicines in are popular in practice for treating diabetes as they are well known to be free from toxic effects.[9] Withania Coagulans (paneer doda) is a famous herb of family Solanaceae and is cultivated in Pakistan, Afghanistan, India and Iran.[10-11] Various components of Withania Coagulans including flowers, seeds, and fruit berry extract have been studied for its several biochemical properties.[12] Antidiabetic property of Withania Coagulans (aqWC) have been studied previously by using Aqueous extract of its dried fruits.

Previous Studies on animals as well as on humans have revealed that the presence of Withanolide, an active ingredient in the aqueous extract of Withania Coagulans resulted in significant improvement in blood HbA1C level. Effect of Withania Coagulans extract on reducing glucose levels were similar to the reesults produced by using biguanides and sulphonylureas.[13] Moreover different studies in animal model have also shown that aqueous extract of Withania Coagulans (aqWC) significantly improves pancreatic islets cells architecture.[14] However Anti-diabetic effect of aqueous extract of Withania Coagulans on incretin hormone has not

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been studied yet. Moreover studies comparing the effect of Withania Coagulans and GLP-1 mimetics on fasting and postprandial blood glucose levels are also lacking. So the current study was conducted with an aim to investigate the effect of aqueous extract of Withania Coagulans (aqWC) and Liraglutide on serum GLP-1, postprandial and fasting blood glucose levels.

METHODOLOGY:

This randomized control trial was conducted at multidisciplinary lab of Islamic International Medical College, Rawalpindi in collaboration with Animal house at National Institute of Health (NIH), Islamabad from 1st April 2016 to 31st March 2017 after getting approval from Ethical Review Committee of Riphah International University (RIU), Islamabad.

Male Sprague dawly rats having weight 200-300 grams were included in the study while overweight or underweight rats were excluded. A total of 40 selected adult male Sprague dawly rats were divided randomly into two groups; Group A (n=10) and Experimental Group (n=30). Group A received standard diet for five days while experimental group received normal diet along with streptozotocin at a dose of 30mg/kg/day intraperitoneally for Five days. On sixth day diabetes in experimental group was confirmed by assessing Fasting blood glucose levels (mg/dl) and its levels was compared with that of group A. The diabetic Experimental group was then randomly divided into group B, group C and group D (n=10). Group B received standard diet for 25 days while group C rats received normal diet besides aqueous extract of Withania coagulans (1000mg/kg/day) orally mixed in drinking water. In addition to normal diet Group D rats received liraglutide drug (0.3mg/kg/day) subcutaneously via Victoza pen for 25 days. Levels of postprandial glucose (mg/dl), fasting blood glucose (mg/dl) and serum GLP-1 level (pg/ml) was determined after 30 days of treatment by taking blood sample from group A, B, C and D.

Withania coagulans dried fruits were purchased from local market. The herb was then authenticated and identified by National Agriculture Research Center (NARC), herbarium section Islamabad. Whole fruits of W. coagulans (1 kg) were kept and soaked in distilled water for overnight after removal of pedicle and calyx. Water extract after the process of filtration was vaporized at temperature of 55 to 60c over magnetic stirrer for seven hours. As a result a semisolid material with a yield 16% wt/wt was obtained. This yield was further diluted with distilled water at a dose of 250 mg/ml. The diluted extract of Withania coagulans was then used for further work.

Blood sampling was done through Intracardiac sampling and 2 mL of blood was drawn for measuring levels of postprandial glucose, fasting blood glucose and serum GLP-1 level at day 30. Rat tail vein was used for drawing blood sample at day 6 for measuring serum fasting blood glucose level.

Statistical analysis of the data was done by using SPSS version 21 and Results were documented as mean + SEM. Independent sample t-test was used for the comparison of mean among groups. A p-value of <0.05 was considered as statistically significant.

RESULTS:

Glucagon like Peptide-1 levels (Mean ± SEM) in all groups (A, B, C, D) is shown in Figure -1. GLP-1 levels of group B rats (931 ± 64.12 pg/ml) were significantly reduced (P<0.05) as compared to GLP-1 levels of group A rats (1646 ± 42.36 pg/ml) depicting that diabetes in group B resulted in lowering of GLP-1 levels. On the other hand GLP-1 levels of group C (1158 ± 27.28 pg/ml) and group D (1290± 0.17 pg/ml) rats were significantly raised (P<0.05) as compared to the GLP-1 levels of group B (931 ± 67.94 pg/ml) reflecting the effectiveness of GLP-1 mimetics and of withania coagulans in improving GLP-1 levels in diabetes.

Comparison of Mean ± SEM of Fasting and Post prandial blood glucose (mg/dl) levels of the study groups is displayed in Table 1. Fasting blood glucose levels in group C rats (98 ± 1.80mg/dl) and group D rats (102 ± 2.04) were significantly lower (P<0.05) than group B rats (131 ± 3.05mg/dl). Post prandial blood glucose levels of group C rats (183± 6.30mg/dl) and Group D rats (163 ±3.95 mg/dl) were significantly reduced (P<0.05) as compare to group B (330± 15.95mg/dl) rats.

DISCUSSION

Type 2 diabetes is an area of concern worldwide as it results in significant health-care cost. Medical management of type 2 diabetes comprises of oral hypoglycemic drugs (metformin,

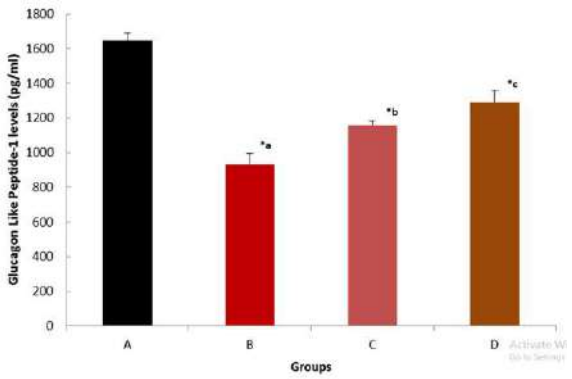

Figure 1: Comparison of Glucagon like Peptide-1(GLP-1) levels (Mean ± SEM) in various groups:
Group A: Control
Group B: Diabetic control
Group C: Withania Coagulans Treated
Group D: Liraglutide -Treated
*a= Group A vs B
*b= Group B vs C
*c =Group B vs D
*=P < 0.05 is considered statistically significant.

Parameters	Group A (Control)	Group B (Diabetic)	Group C (Withania Coagulans treated)	Group D (Liraglutide treated)
Fasting blood glucose (mg/dl)	80 ± 3.19	131 ± 3.05*[a]	98 ± 1.80*[b]	102 ± 2.04*[c]
Post Prandial blood glucose (mg/dl)	143 ±5.34	330± 15.95*[a]	183± 6.30*[b]	163 ±3.95*[c]

Table 1: Comparison of Mean ± SEM of Fasting Blood Glucose (mg/dl) and Postprandial glucose (mg/dl) levels in all four Groups (A, B, C, D)

thiazolidiendiones and sulphonylurea) and parenteral drugs (Insulin and GLP-1 mimetics). In the present study antidiabetic effect of withania coagulans and GLP-1 mimetic (liraglutide) on serum GLP-1, fasting and postprandial blood glucose levels was evaluated.

Current study shows that the use of aqeous extract of Withania coagulans resulted in significant reduction in fasting and postprandial blood glucose levels which is in accordance with the work done by Jaiswal et al,.(2009) who explored the effect of aqeous extract of withania coagulans (1000 mg/kg body wt) in streptozotocin induced diabetic rats.[15] Our study also supports the finding of study done by Alam et al who orally gave 10 gram powder of withania coagulans to diabetic patients which resulted in significant reduction in their fasting and postprandial blood glucose levels.[16]

Hypoglycemic property of withania coagulans fruit can be explained by Boltzmann distribution law because of the substantial amount of calcium and magnesium present in the aqueous extract. Expression of insulin gene is enhanced due to the presence of significant amount of calcium ion via Calcium Responsive Element Binding protein (CREB) which is principally accountable for exocytosis of stored insulin from pancreatic beta cells.[17]

Findings of currents study is also in line with the study conducted by Hamaltha et al who administered aqueuous extract of withania coagulans and proved its antidiabetic effect by measuring postprandial and fasting blood glucose levels. However they administered the aqueous extract for only seven days instead of 30 days as in our case. Moreover in their study levels of serum GLP-1 was not explored. However they explored the anti-hyperlipidemic effect of withana coagulans and stated that reduction in lipid peroxidation activity and presence of antioxidants is accountable for the protection of pancreatic beta cells from oxidative damage which is the underlying cause of hyperglycemia.[18]

Our Study also seconds the findings obtained by Shukla et al who described the effect of aqueous extract of withania coagulans at a dose of 500mg/kg/wt on glucokinase and phosphfructokinase (carbohydrate metabolic enzymes) and concluded that withania coagulans improves blood glucose levels.[19]

Shimoda et al conducted a study on diabetic rats and explored the effect of GLP-1 analogue on pancreatic cell histology and GLP-1 level. The level of GLP-1 were significantly elevated after treatment with GLP-1 analogue. Findings of current study is parallel to this study results with a distinction that instead of only GLP-1 analogue we used aqueous extract of withania coagulans along with liraglutide (GLP-1 analogue).[20]

Datta et al., (2015) mentioned in their study that oral administration of hydroalcoholic extract of withania coagulans dried fruits to streptozotocin induced diabetic male albino rats for 4 weeks cause significant reduction in postprandial and fasting glucose levels. Withania coagulans treated group also showed the recovery of destructed pancreatic beta cells in comparison with untreated diabetic rats. They used hydroalcoholic extract of withania coagulans because in addition to hypeglycemic they also explored the antihyperlipidemic effect of withania coagulans.[14]

Our study results also supports the findings of study done by Lee et al (2002) who concluded that administration of maglitol to the type 2 diabetic subjects increases the post meal GLP-1 levels.[21]

CONCLUSION

Withania coagulans increases level of GLP-1 hormone while reducing both fasting as well as postprandial blood glucose levels. In comparison with GLP-1 mimetics the lack of adverse effects, cost effectiveness and oral mode of ingestion makes withania coagulans as a better treatment option for type 2 diabetes.

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Original Article

Colour Blindness Among Male Professional Drivers: A Cross-sectional Study In Karachi

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ABSTRACT:

Objective: To determine the frequency of color blindness among male professional drivers of Karachi- Pakistan and the difficulties they experience while driving.

Study Design and Setting: A cross-sectional study was conducted aomng 300 male drivers with valid driving licenses operating on local and long routes in Karachi.

Methodology: The study was done at various locations in the city of Karachi from September to November 2017. The subjects were tested for vision and color blindness with the help of Snellen's chart and Ishihara chart respectively. A précised questionnaire was developed keeping in view the study objectives. SPSS version 21.0 was used for data entry and its analysis. Each questionnaire was thoroughly studied and seen for missing data or information before entry. A written informed consent about the study was provided to the subjects in the form of a plain language statement attached to the questionnaire. This study was approved by the ethical review board of Karachi Institute of Medical Sciences (KIMS).

Results: Amongst the 300 participants 7 (2.33 %) of the drivers were found to have complete color blindness while 2 (0.66%) were partially colour blind. They preferred day time driving and experience difficulties in recognizing traffic signals and vehicle lights.

Conclusion: The frequency of colour blindness was found to be low but it was observed that colour blind drivers face a lot of difficulties while driving. Therefore, easy and practical changes should be made, like shape-coding of signals, and continuous research-based modifications are required to aid these drivers.

Keywords: *Colour Vision Deficiency (CVD)/Colour blindness, professional drivers*

INTRODUCTION

Commercial transport businesses have a colossal lucrative effects on the country's transportation industry and on the overall economy.[1] Most governments invest a lot in every sector to make it advance further through continuous monitoring and evaluation that emanates better policies for improvements.[2] The most common problem being observed in this profession is the poor physical and mental health of professional drivers.[3-5] Long working hours, difficult and stressful driving conditions, monotonous and repetitive work, lack of education and low socio-economic status all contribute to deteriorate their mental and physical health.[6]

While talking about physical health of drivers, colour blindness is an important health related issue that is under consideration to be solved appropriately.

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Colour blindness or colour vision deficiency (CVD) is the deficiency to detect certain colors from what is seen with normal vision. Colour vision deficiency is a condition that affects the perception of colours that are specifically associated with electromagnetic radiations of specific wavelengths which a human eye can visualize.[7] The retina is lined by red, blue and green cone cells which perceive the colours and the signals are generated and send to the brain to create a wide spectrum of colours that we see. Genetic CVD is due to defected photopigments in these three different kinds of cones that respond to red, blue and green light.[8] Mostly colour blind people are not aware of their deficiency specially those who do not have severe type of colour vision impairment unless tested in clinic or lab. Colour blindness is mostly hereditary but, it can also be caused by chemical or physical damage to the eye, to the optic nerve or to the part of brain that process colour information. Men are more affected by genetic CVD than women due to the mutation of X chromosome. Sometimes aging or cataract may lead to decline in colour vision.[9]

The most common type of colour blindness is red-green followed by blue-yellow while complete colour blindness is rare.[10,11] There are three main types of color blindness, that is entailed by the photopigment defects in the three different kinds of cones. The commonest being the red-green color blindness that is followed by blue-yellow type of color blindness. Total color blindness is rare which is the complete absence of colour vision.

Worldwide colour blindness approximately affects 1 in 12 (8%) of men while 1 in every 200 women is affected by CVD.[12] Similarly, among European Caucasians of Chinese and Japanese ethnicity, it is around 8% in men while 0.4% in women.[11,12] Although, different studies are being carried out throughout the world to observe the health status of drivers[13,14] but, It is commonly assumed that drivers with colour identification problems drive as safely as other drivers with normal eyesight.[15] This is owing to the reasons that colour blind drivers hardly complaint, no accident records are kept and above all no policies have been developed regarding CVD for the vehicle licensing system therefore, it is assumed that colour impaired drivers easily compensate. An old study showed that in Canada only, around 500,000 drivers were having difficulty in recognizing some colors specially red and green.[16] Similarly, a study of 1992 shows that at least seven million drivers in North America had difficulty in identifying red and green lights.[15] Some three million drivers were suffering from colour blindness in British Colombia in addition to those with hereditary colour blindness.[17]

Individuals having colour blindness experience a lot of difficulties in everyday life and at work. They take a longer time to judge certain colours than normal individuals.[18] A study revealed that the chances of rear-end collision is doubled in colour blind people.[19] The traffic lights seem satisfactory and are easy to follow for a normal person but for the color impaired person they are difficult to judge. For instance, red blind drivers have difficulty in critical stops due to poor recognition of red lights on signals and car brake lights.[17]

Keeping this in view a cross-sectional study was conducted in 2017 in Karachi, Pakistan, among 300 male drivers. This study observed the frequency of colour blindness among male drivers. These drivers were also tested for their eyesight and specifically tested for colour blindness using Ishihara's chart.

METHODOLOGY:

To study the frequency of colour blindness amongst male drivers, a cross-sectional survey was conducted from September to November 2017. Data was collected from 300 participants. The age limit was from 18-60 years as visual acuity lowers with increasing age while CVD may also occur. Data was collected from various sites within Karachi city through convenient sampling. The subjects included male drivers with permanent and valid driving license, either driving on local routes or long routes. In our society, professional driving is solely dominated by males therefore, the study does not include any female drivers. Participants who had history of severe head injury or neurological or ocular surgeries were excluded from the study. Drivers using antituberculosis drugs at the time of survey were also excluded.[20] They were briefed about the purpose and procedure of the study. Those who volunteered signed a written informed consent and filled a small proforma regarding personal information including age, education, years of driving, type of vehicle and route of driving. The participants were also inquired about their eyesight (with or without glasses; if applicable) and if they know that they had colour vision deficiency. Those who were found to have or were aware of their CVD were also inquired if any of their family members have the same problem.

The drivers' visual acuity was measured through Snellen's Chart while colour blindness was investigated through Ishihara's Chart. Those who could not read the numbers (never attended school) on Ishihara's Chart were asked to draw on paper using a pencil, what they saw. The participants who found to be having any of the above deficiencies were advised to consult eye clinics for further evaluation.

This study was approved by the ethical review board of Karachi Institute of Medical Sciences (KIMS). Data was compiled for analysis using IBM SPSS statistics version 21.0 (IBM Corp., Armonk, N.Y., USA). Frequency was calculated as simple percentages.

RESULTS:

Table 1 summarizes the characteristics of the study population and CVD frequency among them. The cross-sectional survey included 300 male professional drivers amongst whom 37% (n=111) were between 31-40 years of age followed by 31% (n=93) who were between 41-50 years. CVD frequency was found in the same age groups (n=5, n=4 respectively). 21% (n=63) were unable to read or write however, majority of the CVD positive individual (n=6) had received 6 to 10 years of education (n=108, 36%). Complete CVD was found amongst drivers who had 6 to 10 years of driving experience.

The study included both long and local route vehicles (n=120 and n=180 respectively) among which mostly were minibus and coach drivers but, most of the CVD was seen in truck drivers of both, long and local routes (Table 2).

Table 3 enumerates the frequency of visual impairment and CVD amongst professional drivers. Out of 300 individuals, 26 were found to be having impaired vision and only 3 used to wear glasses among them and needed reevaluation. All of the 26 drivers were somewhat aware of their visual in-acuity. Partial CVD was observed in 2 (0.66%) participants who showed difficulty in recognizing red and green colours while, 7 out of 300 (2.33%) were suffering from complete CVD. None of them were aware of their deficiency nor did they know if anyone in their family had the same problem.

DISCUSSION:

This study was done to determine the frequency of partial and complete colour blindness in professional drivers. For the detection of CVD, Ishihara's Chart was chosen because of its validity for accurate assessment in mass screenings.[7] Moreover, it is easy and quick to perform as the individuals had to identify numbers rather than the colours.

Age in years	Frequency	Percent %	CVD frequency
20-30	69	23.0	0
31-40	111	37.0	5*
41-50	93	31.0	4*
51-60 years	27	9.0	0
Education			
Not educated at all	63	21.0	0
Primary, up to 5 yrs	84	28.0	1
Secondary, up to 10 yrs	108	36.0	6*
Higher education, above 10 yrs	45	15.0	2*
Years of driving			
1-5 Years	39	13.0	3*
6-10 years	57	19.0	5
11-20	120	40.0	0
> 20	84	28.0	1*

Table 1: CVD frequency related to characteristics of drivers N=300
*Partial CVD included

Types of vehicles	Frequency	Percent %	CVD Frequency
(a) Long route vehicles	**120**	**40.00**	**5**
Bus (coach)	61	50.83	3*
Truck	35	29.16	2
Trailer	24	20.00	0
(b) Local route vehicles	**180**	**60.00**	**4**
Bus (mini/coach)	93	51.66	0
Taxi	56	31.11	1
Truck	31	17.22	3

Table 2: CV frequency related to types of vehicles and driving routes (N=300)
*Partial CVD included

	Frequency	Percent
Visual impairment	26	8.66
Partial CVD (red-green CVD)	2	0.66
Complete CVD	7	2.33
Visual impairment with CVD	0	0.00

Table 3: Visual impairment and Colour vision deficiency (N=300)

The prevalence of complete CVD was found to be 2.33% while partial CVD came out to be 0.66%. Our results are comparable with the results of a study by Siddiqui at el.[21] That showed 3.1% CVD amongst male students of Pakistan.[21] In 2017, another similar frequency was given during a pre-employment screening in a tertiary care health centre of Pakistan. It reported a 0.9% overall prevalence of CVD (32/3437) with 1.4% males and 0.4% females.[11] These frequencies are much less than the results of earlier studies done in various countries. A study done among Indian Muslims in 2013, showed 8.73% of CVD in males but, their results differed in various Muslim ethnicities of India.[22] Another 10 year study was done in 2016 among science students of ASC Rahuri College India, showed 0.303% of colour Blindness in male students.[10] A Turkish research also observed 3.2% red-green and 6.7% CVD in males . In 2006, Balasundaram et al. revealed a prevalence rate of 6.7 colour blindness in male health professionals of Seremban, Malaysia.[7] The differences in frequencies in all the above studies might be due to the race, ethnicity and/or geographical locations.

It is commonly believed that colour blindness hardly create any trouble while driving. It is mostly due to the fact that drivers often identify lights according to their arrangements on the signal. However, several studies have shown that drivers do face difficulties while driving.[9,24] Therefore, those who had partial or complete CVD were further interviewed about their driving issues.

In this study, individuals with impaired vision acuity were fully aware however, seemed careless about it. Vision impairment due to refractive errors is easier to self-detect and can be overcome by wearing glasses but, colour blindness is difficult to detect unless tested in clinic or labs especially, when congenital.[25] All of the CVD positive individuals in the study were also not aware of their deficiency although, most of them were middle aged and had more than 5 years of education. Previous researches have revealed that these people develop some strategies and clues about certain colours since their childhood.[11,18] Despite the colour identification strategies, our colour blind participants were facing particular problems while driving that they never realized before then. Sudden inversion of signal lights from red to green specially when there is no yellow light in between, were difficult to detect for most of them. This is a common difficulty found in most of the colour blind drivers.[24] Similarly, almost all of them accepted that they could not recognize brake lights which delays brake time while critical stops.

Drivers declared that they had faced RTAs in their early driving years. Among them were mostly bus and truck drivers who had difficulty to identify car brake lights that were at lower level compared to their own vehicles. The small sample size of our study restricted us to infer CVD as a risk factor of RTAs. These results are in agreement with some previous studies.[26,27] As CVD positive drivers were not aware of their deficiencies, they did not know if any of their family members had the same problem.

It was recommended that research based modifications, like shape-coding of traffic signs, should take place involving all the stakeholders including traffic engineers and medical and public health practitioners to counter the driving issues of CVD positive drivers. A complete eye checkup will be helpful, before the issuance of driving license and at renewals, in order to get recent statistics and information.

CONCLUSION:

It was concluded that the frequency of colour blindness amongst male drivers was found to low but, most of them

remain unaware throughout their life. They, themselves, not only face a lot of difficulty while driving but are a huge risk for others as well.

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Original Article

Students' Perspective On Using Internet Based Dental Educational Videos As An Adjunct In Dental Education

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ABSTRACT

Objective: The aim of this study was to observe students' perspective regarding the role of internet-based education as a supplementing tool in dental education.

Study Design and Setting: A cross-sectional survey was designed and carried out among the dental students of Margalla Institute of Health Sciences, Rawalpindi in March 2018.

Methodology: Approximately two hundred and five regular undergraduate dental students of 2nd, 3rd and final year BDS were included with an overall response rate of 100%. A self-administered close-ended questionnaire was used regarding the student's perspective of using internet based educational videos during their course of dental education. Data was analyzed using SPSS version .24. Descriptive statistics was calculated.Chisquare test was used. $P<0.05$ was taken as significant.

Result: Majority of the students (97.6%) considered internet based dental educational videos (IBDEVs) was a significant supplementing tool in dental education and agreed that the knowledge gained by internet based dental educational videos was retainable and reproducible. A greater frequency of students (88.8%) searched for videos based on clinical procedures than those for theory-based topics. No significant difference was observed between males and females as well as students of different years regarding opinion about significance of IBDEVs and their frequency of use ($P>.05$).

Conclusion: Internet based dental educational videos (IBDEVs) were used as a beneficial supplementing tool by dental undergraduate students to enhance their understanding of a topic and improve overall learning.

Keywords: Dental Education, Surveys and Questionnaires, Visual Aid, Educational Technology

INTRODUCTION:

The need and benefits of technology in today's era cannot be overemphasized. Use of internet has seeped into our daily lives and made us dependent on it. Students today are more digitally oriented and driven than those of pre-information technology age. They tend to be attracted to and engaged in activities that involve use of technology especially internet, and expect a greater use of multimedia for teaching and learning. Literature reveals that use of internet is more frequent among contemporary dental undergraduate students than medical students.

Owing to the widespread use of portable gadgets such as smartphones and laptops, students have access to internet all the time 'on the go', making it easier to access information within no time. This generation of students are technologically savvy and immersed in the web, heavily engaged in gaming and internet surfing and social networking applications. Two widely used social media platforms are Facebook and Youtube. The students tend to prefer pictures and videos over lengthy demonstrations, expect immediate feedback, and demand instructions that are fun, interactive and non-linear.

Due to the pressure exerted by advances in technology, faculty shortages and student demand for flexible learning, the momentum of educational technological growth is likely to continue to increase in the future. The same applies to dental education with respect to digital or video-based learning. The term 'video-based learning' refers to the technology that is electronically taking and distributing sequences of educational image displaying scenes in motion. The videos are a rich and powerful tool for computer-assisted learning. Infect, an official you-tube channel of American Dental Association has been launched and offering free access to innumerable videos covering a wide range of topics such as oral hygiene, oral care for pediatric and geriatric patients, dental care during pregnancy etc. These videos that are now instrumental for student learning which were previously been used only for educating patients.

Advancements in technology have brought a paradigm shift in medical education with greater emphasis on a student – centered approach. Clinical education and training is primarily obtained through senior faculty professors giving verbal or written instruction along with live demonstrations, lectures,

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laboratory and clinical sessions. Clinical education of specific skills continues to be the most time-consuming aspect of education in dentistry. Today, not only instructors but students need to be better equipped to cater the changing educational requirements. Use of videos as a supplementing tool in education is not a new concept. However, content that was previously restricted or available on payment is now available free of cost, easy to access and is of standard and evidenced content. For subjects that involve psychomotor skills in clinical subjects of dentistry; the educational videos can serve as a supplemented guide to improve learning and clarify concepts.Incorporating the use of instructional videos made for specific clinical skills have been advocated to assist students to achieve better academic out comes and better clinical skill performances even as also cost effective by decreasing the need of faculty.

Currently self – directed learning is being incorporated in curricula, utilizing supplementary educational videos and internet content becomes mandatory for the student. Although the impact of video and multimedia technologies in educational outcomes is afield of ongoing research, a summary of the impact of videos can be defined by three key concepts; interactivity with content, engagement and knowledge transfer and memory. Today, video plays a significant role in education in terms of its integration into traditional classes, and become the principal delivery system of information in classes particularly in online courses as well as serving as a foundation for many blended classes.

Research has formerly been focused to evaluate the effectiveness of such videos in teaching clinical skills and achieving learning objectives. However, students' appraisal of these videos as a learning aide needs to be considered. The aim of this paper was to obtain students' perspective regarding the role of education videos as a supplementing tool in dental education. The outcome of this study would assist teachers while tailoring their teaching strategies which guide their pupils towards effective self – directed learners along with the dental school administrators to understand the perspectives of dental students while formulating decisions about dental school educational strategies.

METHODOLOGY:

Ethical approval of this study was taken from the college's ethical committee. A cross-sectional survey was designed and carried out among the students of Margalla Institute of Health Sciences, Rawalpindi in March 2018. Based on previously published data, keeping confidence interval (1-á) at 95%, absolute precision (d) at 0.06, and anticipated population prevalence (P) at 0.257, a sample size of 205 was calculated using WHO sample size calculator. The regular undergraduate students of 2nd, 3rd and final year BDS students were included in the survey whereas students of physiotherapy, pharmacy, faculty members, house officers and postgraduate trainees were excluded from the study. A self-administered close-ended questionnaire was used for data collection tool. Face validation of the questionnaire was done by faculty members of different dental departments and ambiguous questions were removed. The questionnaire was also pilot – tested to ensure its validity, reliability and relevance. A total of eleven questions were asked that encompassed different perspectives of students based on how much beneficial internet based educational videos are during their course of dental education to supplement it. The purpose of study was explained to the students and individual inquiries were answered. The information was answered anonymously, no personal data except age and gender were asked. Data was analyzed using SPSS version 24. Descriptive statistics were calculated. Categorical variables were presented as frequency and percentages. Post-stratification Chisquare test was used to control effect modifiers such as gender and year of education. $P<0.05$ was taken as significant.

RESULTS:

The questionnaire was filled and returned by two hundred and five dental undergraduate students with an overall response rate of 100%. There were seventy-nine students (38.5%) from 2nd year, sixty students (29.3%) from 3rd year and sixty-six students (32.2%) from final year BDS.Out of two hundred and five respondents, one hundred and sixty nine (82.4%) were females and thirty six (17.6%) were males. Majority of the students (n=200) considered internet based dental educational videos (IBDEVs) a significant supplementing tool in dental education and that the knowledge gained by IBDEVs was retainable and reproducible. Table 1 highlights the general opinion of students regarding IBDEVs. A greater frequency of students (n=182) searched for videos based on clinical procedures than those for theory-based topics (11.2%). Most of students (n=115) found the content of IBDEV to be moderately relevant while 60.2% (n=123) found the videos to be moderately clarifying when they specifically search for a topic (see figure 1, 2). Regarding the frequency of access of IBDEVs, 39.3% (n=81) replied that they required assistance of such videos on a weekly basis (see figure 3) while 80% (n=164) preferred searching for IBDEVs at their home. No significant difference ($P>0.05$) was observed between males and females as well as students of different years regarding opinion about significance of IBDEVs (table 2) and their frequency of use (table 3).

DISCUSSION:

One of the ways to improve the effectiveness of dental education is by introducing videos in the lectures. It has been suggested that video-based teaching is appreciated and widely acknowledged by dental undergraduate students as a good way of attaining knowledge. The present study, however, aimed to know the student's perspective with regards to the use of internet – based dental educational videos as a supplementing tool in their dental education.

Question	N	Response (%)	
		Yes	No
Are internet-based dental educational videos a significant tool in dental educational?	205	97.6%	2.4%
Do you think internet-based dental educational videos help retain the knowledge for longer period?		96.1%	3.9%
Is the knowledge learned by internet based educational videos reproducible?		95.1%	4.9%
Have you personally looked for internet based educational videos on any other topic NOT COVERED in class?		77.2%	22.8%
Are you satisfied with the quality of the video content available?		83.5%	16.5%

Table 1: Opinion of dental undergraduate students regarding internet based dental educational videos (IBDEV)

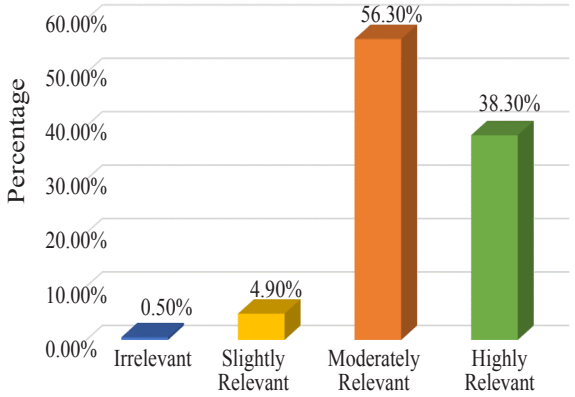

Figure 1: Relevance to clinical practice of content learned from Internet-based dental educational videos

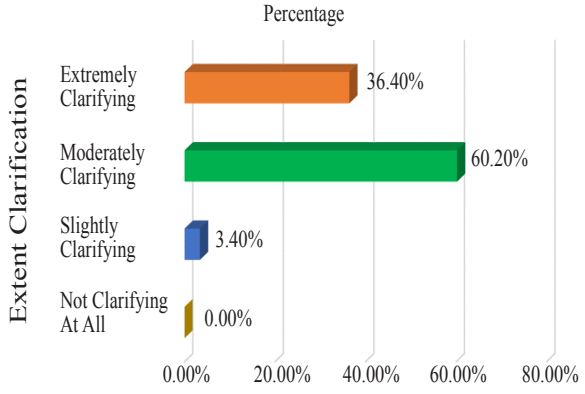

Figure 2: Extent to which IBDEVs clarify students' concepts

In the present study, majority (n=200, 97.6%) students found IBDEVs a useful supplementing tool in dental education. These findings corroborated with Al-Jandan et alin which 91.4% of students found videos a beneficial instrument in their education. Similar results were reported by Kalwitzki et alin which students favored video-based learning over lectures. A systematic review on effectiveness of various

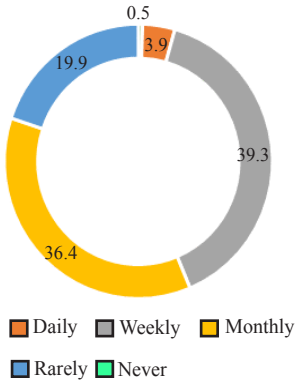

Figure 3: Frequency of access of IBDEVs as a supplementing tool

Variable	Response	Gender		P value
		Male	Female	
IBDEVs are a significant tool in dental educational	Yes	34	167	0.179
	No	2	3	
Frequency of use of IBDEV	Never	2	2	0.825
	Rarely/Once in a while	4	35	
	Daily	1	7	
	Weekly	15	66	
	Monthly	14	61	

Table 2: Cross-tabulation of variables with respect to gender

Variable	Response	Year of Education			P value
		Year 2	Year 3	Year 4	
IBDEVs are a significant tool in dental educational	Yes	75	61	65	0.586
	No	3	2	2	
Frequency of use of IBDEV	Never	2	2	2	0.358
	Rarely/Once in a while	10	15	13	
	Daily	5	2	2	
	Weekly	36	19	23	
	Monthly	25	23	24	

Table 3: Cross-tabulation with respect to year of education

teaching methods by Gopinath and Nallaswamy[11] also observed video-based teaching as a useful adjunct to other teaching and learning strategies.

In response to questions about retention of gained knowledge, 96.1% subjects in the present study agreed that IBDEVs helped retain the knowledge for a longer period of time. Contradictory results were reported by Al-Jandan et al that majority of (68.5%) students agreed that knowledge gained from videos is retained for a longer period but both males and females appeared rather unclear about the question. This may be attributed to varying retention capabilities of males and femalesas well as a difference in learning approaches that work for different students such as VARK. It has been suggested that males prefer multiple modes of instructions

while females prefer a single mode, with a greater predilection for kinesthetic learning.

Approximately 60% subjects in the present study reported that IBDEVs 'moderately clarified' their concepts while 36.4% students found them 'extremely clarifying'. These findings are endorsed by those of Chi et alwho compared student learning outcomes associated with video-based learning in comparison to paper – cases. They reported that video – based learning significantly enhanced learning in cognitive and affective domains, improving the overall learning outcomes. Comparable results were also found by Naseri et althat video-assisted clinical instruction in dentistry (VACID) is an effective method to improve student learning and it minimizes the number of students who approach the teacher with questions. A similar study was carried out in Sweden by Rystedt et alby evaluating students' perception of video-based clinical demonstration where students expressed that video – based teaching helped in integrating theoretical knowledge with clinical understanding.

In dental education, a noteworthy problem is how to bridge the gap between theory and practice. Students can find it difficult and intimidating to appreciate the clinical relevance of preclinical courses and to visualize clinical scenarios. Although clinical demonstrations can be a useful tool in this regard, but limited access to oral cavity makes it difficult for all students to see adequately what is being demonstrated. This difficulty can be overcome by the use of internet-based dental educational videos as demonstrated by the present study. Such videos can serve as a supplementing tool to augment understanding. Further queries can be addressed individually during hands-on practice. The present study suffers from the weaknesses inherent in a questionnaire – based survey. The responses were subject to reporting bias of study participants. Moreover, the sample size can be increased to include a more diverse sample and to generalize the results to the entire population.

With the changing paradigms of health professional education, incorporating latest teaching and learning strategies has become even more important. Internet based educational videos can play a major role in student learning. However, since the content of IBDEVs cannot be regulated or supervised, such videos should only be used as a supplementing tool and not as the primary source of learning. Any ambiguities must be discussed with the instructor and concepts verified. Moreover, quality and content of IBDEVs can be improved and regulated if prominent dental associations, organizations or teaching universities in the country upload videos tailored to the needs of preclinical and clinical students.

CONCLUSION:

Based on the findings of this study, it was concluded that dental undergraduate students considered internet based dental educational videos (IBDEVs) a significant supplementing tool in dental education and that the knowledge gained by IBDEVs was retainable and reproducible. Students preferred to search for videos based on clinical procedures and found the videos to be moderately clarifying when they specifically searched for a topic. Students accessed IBDEVs on a weekly basis, found them moderately relevant and were satisfied with the content of available videos.No significant difference was observed between males and females as well as students of different years regarding opinion about significance of IBDEVs and their frequency of use (P>.05).

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Original Article

Frequency of Dyslipidemia In Type 2 Diabetic Patients In Karachi

Sehrish Shafique, Daud Mirza, Summayya Shawana, Shahneela Tabassum, Naveed Faraz

ABSTRACT:

Objective: To find out the frequency of dyslipidemias in type 2 diabetic patients.

Study Design and Setting: This was cross-sectional study and conducted in a different clinics in Karachi during 3 months period.

Methodology: Those patients who fulfilled the inclusion criteria and attended the OPD were selected. Patient were advised to bring their lipid profile report in next visit if not already done. The total dyslipidemias were presented by their frequencies and percentages with 95% confidence interval.

Results: Out of 383 patients with diabetes mellitus 210 (55 %) had dyslipidemia. Among which 76% had elevated low-density lipoprotein (LDL), 66.6 % had elevated serum cholesterol, 57% patient had elevated triglyceride. p- value of 0.05 was taken as statistically significant.

Conclusion: It was concluded that increased frequency of dyslipidemias, with elevated low-density lipoprotein, cholesterol and triglycerides levels seen in diabetic patients.

Keyword: coronary heart disease, Diabetes mellitus, dyslipidemia, HbA1C.

INTRODUCTION:

Diabetes is one of those disease that is of high concerns because of its increasing number of cases. It is expected that number of diabetics will increase from 382 million in 2013 to 592 million by 2035[1]. According to International Diabetes Federation estimates in 2013,35 countries out of 219 have about 12% prevalence of diabetes. Among the Asian population 20% is currently affected due to the disease. In our country 7 million people suffer from hyperglycemia. The prevalence of the disease in our country is 18-46%. The occurrence of diabetes with metabolic syndrome is 46-75% in Pakistan.[2] It is one of the chronic metabolic diseases which is characterized by hyperglycemia, occurring due to defects in either insulin secretion, action, or both.[3,4] Poorly controlled diabetes can result in many long-term complications which can lead to damage and dysfunction or failure of many vital organs, especially the eyes, kidneys, nerves, heart, brain, blood vessels which are more affected along with increased risk of local and systemic infections in these patients.[4] Because of developing these complications, the patients are also at a high risk of disability and premature death.[4] Among these complications, Cardiovascular disease is of high concern in type 2 diabetics, as more than 60-70% die from coronary heart disease.[5,6] According to American Heart Association, at 70%of the people with diabetes aged 65 or older die of diseases due to atherosclerotic changes in arteries supply heart and CNS.[7] Diabetic patients have four times more chances to have coronary heart disease than non- diabetes. Thus diabetes is considered as equivalent to coronary artery disease.[8]

Dyslipidemia is common in diabetics.[6,7,8] In diabetic dyslipidemia there are lipid and lipoprotein abnormalities. It is characterized by elevated fasting and non-fasting triglycerides (TG) and TG-rich lipoproteins, e.g., chylomicrons and very-low-density lipoprotein (VLDL), low high-density lipoprotein cholesterol (HDL-C) and often also elevated low-density lipoprotein cholesterol (LDL-C) concentrations, increased small dense LDL particles.[8,9] The pathophysiology behind these abnormalities is that they occur due to impaired VLDL secretion and there is decreased hepatic uptake of chylomicrons and their remnants also.[8,9,10] Furthermore, lipolysis in these patients is also suppressed due to the presence of insulin resistance in adipose tissue which further contributes to diabetic dyslipidemia. So, there are number of factors which can contribute to deranged lipid markers from normal value in human body in diabetics, apart from insulin resistance, there is also deficiency of adipocytokines, which are also considered as the contributing factors in causing the alteration in lipid metabolism.[8] The exact pathophysiology is still poorly understood, but it is seen that because of insulin resistance there is activation of

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intracellular lipases which in turn increase the release of no esterified fatty acids (NEFA) from the stored triglycerides in the adipose tissue and increases the hepatic triglyceride production which also causes increase in apolipoprotein B production.[10,11] So, the normal inhibitory effect of insulin on hepatic apolipoprotein B production and triglyceride secretion in VLDL is lost, and the VLDL that is secreted is larger and more triglyceride-rich. There is also reduced VLDL catabolism which also increases the triglyceride levels.[9,10] Lipoprotein lipase which is an enzyme located on vascular endothelium is responsible for the rate of removal of triglycerides from the circulation, but it is downregulated in diabetics due to insulin resistance or deficiency.[10] This reduction in lipoprotein lipase activity further also contributes to deranged lipid markers from normal value in human body.

Recent studies suggest that low HDL cholesterol is also an independent factor not only for causing cardiovascular disease but also for the development of diabetes itself.[12,13,14,15] In diabetic patients, improvement in the glycemic control not only cause reduction in cholesterol and triglyceride levels but also increases the catabolism of LDL through the upregulation of its receptors and reduced glycation.[14,16] The rationale of our study was to find out the frequency of deranged lipid markers from normal value in patients with diabetes as earlier diagnosis of such abnormalities in diabetics can minimize and prevent the mortality due to coronary artery disease. Also, both of these diseases are commonly prevalent in our society and their mortality increase many times when they occur simultaneously.

METHODOLOGY:

The study was conducted in a private clinic in Karachi for a period of three months. All diagnosed cases of diabetes mellitus for more than 3 years with Hb A1C of 7 % or more, irrespective of age, gender were included and the patients suffering from cancer, chronic liver disease, hypothyroidism, patients on lipid lowering drugs and chronic renal disease were excluded from the study. The ethical approval was taken from the ethical review committee. Consent was taken before collection of data and also explaining the significance of this study and its procedure. Those patients who were selected after inclusion criteria and attending the OPD were selected. Patients were advised to bring their lipid profile report in next visit if not already done. The total deranged lipid markers from normal value in human body were presented in the form of frequencies and percentages with 95% confidence interval and compared with standard NCEP chart given below. For data analysis SPSS version 20 were used. Different variables were calculated through frequency and percentage.

RESULTS:

In our study a total of 383 cases were selected among which 210 were found to have deranged lipid markers from normal value in human body(table1). The minimal age is 30 years and maximum age is 70 years. Most of the selected patients were in the age group of 46-55yrs (65%) followed by 56-70yrs age group (53%) as shown in graph 2. There were 200 females and 183 male patients included in our study, among which 110 females (55%) and 100 males (55%) were found to have deranged lipid markers from normal value in human body graph 2.

DISCUSSION:

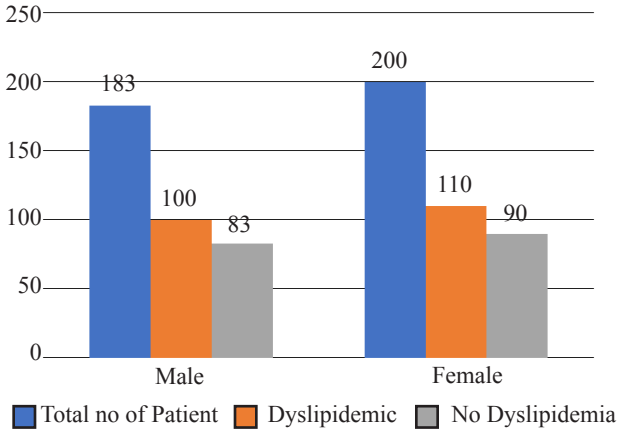
This study has been conducted to see the association of deranged lipid profile in patients with diabetes mellitus. In our study it is observed that out of total 383 patients with diabetes mellitus, 210 (55%) were found to have deranged lipid markers from normal value in human body. The results showed in our study that in diabetic patients the levels of different lipid markers were above the normal required range seen in human body and the outcome was also consistent with previous studies.[17] In our study there were slightly more females 200, among which 55% had deranged lipid markers from normal value in human body, as far as in males out of total 183 patients, 55 % had deranged lipid markers from normal value in human body (graph1). The frequency of deranged lipid markers from normal value in human body was 55% in diabetic patients in our study is consistent with two different previous studies which also shows the presence of deranged lipid markers from normal value in human body in diabetic patients.[18,19] Sedentary life style, increasing shift

Deranged lipid markers from normal value in human body	Frequency	Percentage
YES	210	55%
NO	173	45%

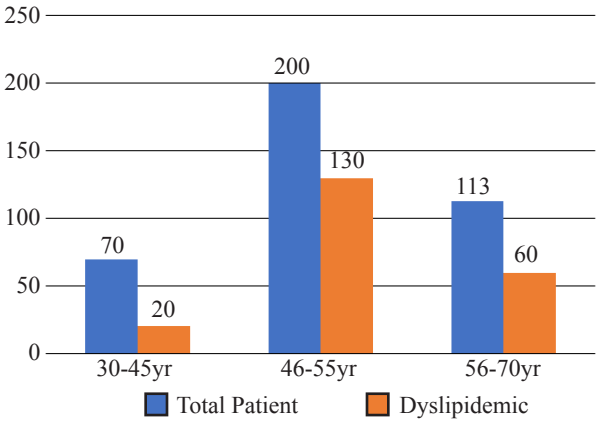
Table:1 frequency & percentage of have deranged lipid markers from normal value

Classification of lipids	Total cholesterol mg %	LDL-C mg%	HDL -C mg %	TG mg%
Desirable	< 200	< 100	>60	< 150
Near optimal	-	100- 129	Higher value is better	-
Borderline High	200 - 239	130 – 159	-	150- 199
High	240 and above	160- 189	59- 40	200- 499
Very high	-	190 and above	< 40	500 and above

Classification of lipids NCEP ATP 111



Graph 1: Distribution of deranged lipid markers from normal value in human body according to Gender



Graph 2: Distribution of deranged lipid markers from normal value in human body according to age group

Total No of Patients with deranged lipid markers from normal value in human body (210)	No of patients with elevated Serum cholesterol 140	Percentage % 66.6%
	Total no of patients with elevated Serum triglycerides (TG) 120	57%
	Total no of patients with elevated LDL-C 160	76%

Table 2: Percentage of Derangement in Lipid profile in Diabetics

of population from simple village life to mechanize life of cities, minimal physical activity due to the use of these hi-tech appliances and change of pure diet plan using junk meal causes increase weight that leads to development of tendency to have increase blood sugar level.[20] The risk of cardiovascular diseases in diabetic patients is more likely as compared to normal human beings. Lipid abnormalities (increased level of low-density lipoprotein, triglycerides and decrease levels of High-density lipoprotein) are a predisposing factor for deposition of these lipid in arteries. Deranged lipid markers from normal value may be due to unbalanced metabolic state in diabetes and better control of diabetes does result in progressive decline in diabetes-associated deranged lipid markers from normal value in human body. In our study elevated low density lipoprotein (76%) followed by the next common abnormalities being increase in serum cholesterol (66.6%) and TG level. (57%) (table 2) is also consistent with studies done in the past.[21,22] In the other study done in past showed elevated triglycerides more than serum cholesterol.[23] In our study maximum (68.5%) of the sample population were 40 years. Comparing in different age groups, among diabetics, patients who are specially in 46 to 55 years of age, were more found to have deranged lipid markers from normal value in human body (65%) (graph 2) then other age groups. The next age group which was more affected was 56 -70 years of age in which 53% of patients were having deranged lipid markers from normal value in human body (graph2). Previous studies also showed that increasing life span plays a major role in the risk of developing hyperglycemia.[17,24] Mortality in diabetics is associated with deranged levels of different of lipid marker.[19,25,26,27,28] So early detection and treatment of deranged lipid markers from normal value in human body especially with hyperglycemia can prevent the progression of disease and limit the morbidity and mortality due to cardiovascular events, and cerebrovascular accident.[29] The limitation of the study was the small sample size in one lab and the financial constrains. It was recommended that patients with diabetes should be considered to be given lipid lowering therapy like statins along with life style modifications to prevent them from coronary artery disease especially in an age group of 55-70 years.

CONCLUSION:

It was concluded that diabetes was associated with higher frequency of deranged lipid markers from normal value in human body and was associated with increased atherosclerotic changes in coronary arteries.

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Original Article

Emergence And Management Of Muti-Drug Resistant Pseudomonas Aeruginosa Isolated From Intensive-Care Unit

Saman Nadeem, Faisal Hanif, Yasmeen Taj, Sana Yousuf, Nadia Midhat Zehra, Pervaiz Asghar

ABSTRACT

Objective: To evaluate the antibiotic resistance pattern of Pseudomonas aeruginosa and its prevalence from samples received from Intensive Care Units (adult and neonatal) at PNS Shifa Hospital Karachi.

Study Design and Setting: This cross sectional study was carried out at PNS SHIFA hospital.

Methodology: Samples including blood, pus, wound swab, sputum and endobronchial washing were received for culture and sensitivity. Isolates were cultured on blood and MacConkey agar. Antibiotic susceptibility testing was performed by Kirby-Bauer and broth microdilution and then analyzed on SPSS version 23.Results were confirmed by VITEK 2.

Results: Out of 674 positive clinical specimens 97(14.39%) were positive for Pseudomonas aeruginosa growth. The most susceptible antibiotic against Pseudomonas aeruginosa was Polymixin b (94.854%).The least effective antimicrobial was aztreonam (40.21%sensitive).

Conclusion: The prevalence of Pseudomonas aeruginosa from the samples of Intensive Care Units was found to be 14.39%. The most susceptible antibiotic against pseudomonas was Polymixin B. The least effective antimicrobial was aztreonam.

Keywords: Antibiotic resistance pattern, Culture and sensitivity, Pseudomonas aeruginosa.

INTRODUCTION:

Pseudomonas aeruginosa is an opportunistic pathogen causing a wide range of infections in the human host.[1,2] It is saprophytic in nature and is found in both hospital and community settings.It can survive in adverse conditions because of its extremely flexible physiological and physical nature. [2] *Pseudomonas aeruginosa* is one of the most important pathogen causing soft tissue infections in burns, diabetic patients and the immunocompromised host. Although not a human flora it usually causes super-infection in diabetic, burns and cystic fibrosis patients.[3]

About 9% of all healthcare associated infections are caused by *Pseudomonas aeruginosa.*[4] Pathogenicity of organism is directly linked to the toxins and enzymes produced by it, and its ability to resist phagocytosic immune capacity of the host.[3,4] Emerging antibiotic resistance against different antimicrobials contributes to its increased morbidity and mortality. Formation of biofilms, increased drug permeability, multi-drug efflux mechanism and acquisition of certain enzymes are the factors mainly responsible for acquisition of antibiotic resistance.[1-4]

According to records from the Centre of Disease Control and National Nosocomial Infection Surveillance System (NNIS) *Pseudomonas aeruginosa* is the second principal cause of pneumonia (17%), third cause of UTI (7%)fourth cause of surgical wound infection(8%) and the seventh leading cause of sepsis (2%) in the hospital settings. Pseudomonas aeruginosa is one the most common cause of health care associated infections specifically from intensive care units.Multidrug resitant strains and carabapenem resistant isolates have been isolated from intensive care units with limited treatment options available.[5]

Emerging resistant strains of pseudomonas have strongly restricted the treatment possibilities available. This study was aimed to evaluate the antibiotic resistance pattern of Pseudomonas aeruginosa and its prevalence from samples received from Intensive Care Units (adult and neonatal) at PNS Shifa Hospital Karachi.

METHODOLOGY:

The cross sectional study was performed from January 2016 to June2017 on patients admitted to the Intensive Care Units (both adult and neonatal) in PNS Shifa hospital. After approval of ethical committee of the hospital we included samples received from patients admitted to the neonatal and adult intensive care units.They include body fluids, blood,

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and pus,endobronchial washing, sputum, collected and submitted to the microbiology department for culture and sensitivity.

Samples received from different body sites were inoculated on Blood, MacConkey and Chocolate agar and incubated at 37 C for 24 hours both aerobically and anaerobically. Standard biological methods were carried out in order to identify *Pseudomonas aeruginosa* colonies as outlined by CLSI. Biochemical test like oxidase, urease, citrate utilization test, nitrate reduction and indole production were also performed. Confirmation of *Pseudomonas aeruginosa* was done by using API 20NE (Biomeriux).

Antimicrobial susceptibility testing was carried out by Kirby-baeur disc diffusion method and broth microdilution as outlined by clinical laboratory standard international guidelines.[6] Isolated colonies of *Pseudomonas aeruginosa* were picked up by a sterile loop and 0.5McFarland suspension was prepared. It was then inoculated on the surface of Mueller-Hinton agar (MHA) plate using sterile cotton swab and incubated at 37 C for 24 hours after placing different antibiotic discs.

Eleven antibiotic discs used in this study were Polymixin b (Oxoid) (300 units), imipenem (Oxoid) (10 µg) tazobactam-piperacillin (Oxoid) (110 µg) , aztreonam (Oxoid) (30 µg) ceftazidime (Oxoid) (30µg) ceftazidime (Oxoid) (30µg), cefepime(Oxoid)(30µg), gentamicin(Oxoid)(10µg), amikacin(Oxoid)(30µg),ciprofloxacin(Oxoid) (5µg), tazobactam/piperacillin(Oxoid)(110µg), cefoperazone/sulbactam75/10mcg),meropenem(Oxoid)(10µg), imipenem (Oxoid) (10µg), aztreonam (Oxoid) (30µg). The results were interpreted as per CLSI guidelines.[8] Control strains used were *Escherichia coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853.

All isolates identification and antimicrobial susceptibility patters has been confirmed by VITEK 2(bioMérieux).

Data was analyzed using statistical software SPSS Version 23. P value of less than 0.05 was considered significant.

RESULTS:

In this study 97 (14.39%) out of 674 samples were positive for the growth of *Pseudomonas aeruginosa*. Clinical specimens showing positive growth for *Pseudomonas aeruginosa* include pus (49%),blood (25%), sputum (13%), endobronchial washing (13%) (Figure: 1). Among them 33% of samples were received by females and 67% by males.The results of antimicrobial susceptibility testing is shown in (Figure:2). The most susceptible antibiotic against pseudomonas is Polymixin B (94.85%) sensitive followed by gentamicin(89.69%) and amikacin(84.54%) imipinem (77.32%) cefoperazone/salbactum(75.26), meropenem (71.13%), cefepime(68.04%) ciprofloxacin(56.7%). The least effective antimicrobial is aztreonam (40.21%sensitive) followed by ceftazidime(56.7%) sensitive.

DISCUSSION:

Pseudomonas aeruginosa is one of the most important opportunistic pathogen. It has imposed a huge burden on healthcare with increase in mortalities both in hospitals and community settings.[1-4.] In our study, 67% isolates were from males and 33% from females which is comparable to a study carried out in Baroda[1], however these figures determined are in disagreement with the values observed in Nepal where female prevelance is superceding in *Pseudomonas* Spp.[7] This change may be due to the difference in cultural practices.

Most of samples showing the growth of *Pseudomonas aeruginosa* (49%) were from pus followed by blood(29%) in intensive care unit.These results are in slight variation

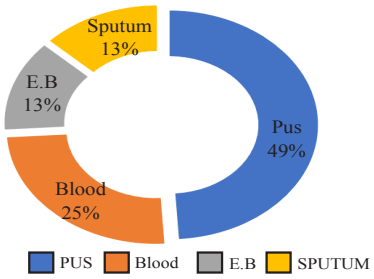

Figure 1 Sample wise prevalence of pseudomonas isolates

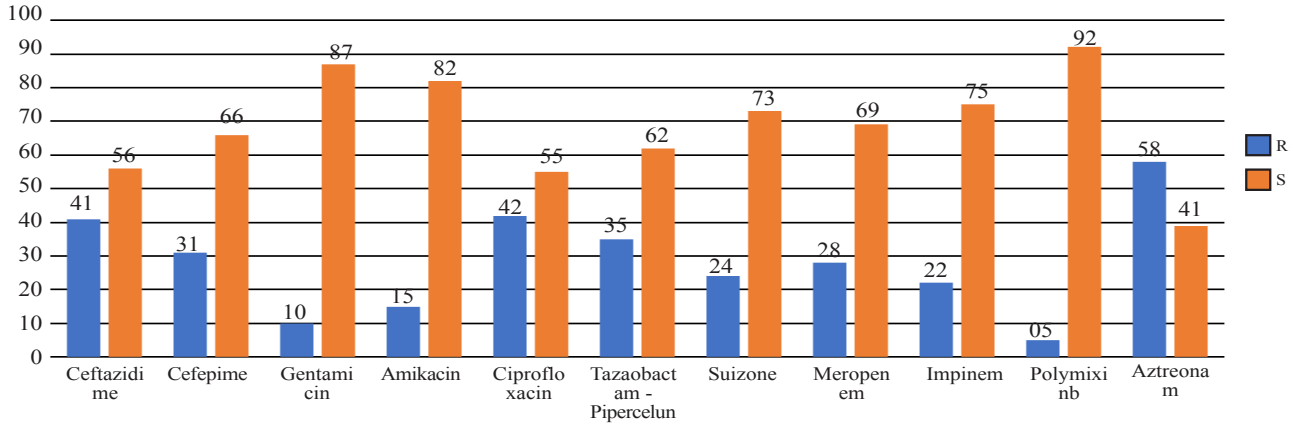

Figure 2:Antimicrobial susceptibility pattern of pseudomonas aeruginosa isolates

with a study carried out in Baroda, revealed *Pseudomonas* growth (70%) from wound swab.[1] Although in our setup, pus samples revealed the growth of *Pseudomonas* as wound swabs are not an appropriate sample for culture and sensitivity.Pseudomonal infections are difficult to treat as the organism exhibit intrinsic resistance towards many antimicrobials. Microorganisms have developed numerous mechanisms to counter the effect of antimicrobials making them resitant. The antibiotic susceptibility differs from hospital to hospital,of different regions and may also alter with time. Patients came in contact with various antibiotics in rural or urban areas in sub-optimal doses and exploitation of some over the counter drugs are the contributing factors towards emerging resistance. *Pseudomonas aeruginosa* resistance to antimicrobials is either through acquiring carbapenemases or aminoglycoside-modifying enzymes.[2,4,5] Resistance is conferred by the shift of extra-chromosomal DNA between different isolated generating a population of bacteria that becomes more difficult to treat. In our study, the most effective antibiotic found was Polymixin b.This finding may be due to the judicial use of this antibiotic to treat more resistant isolates, as it is the last resort for the clinicians to treat such resistant infections. Moreover Polymixin b is not available in oral form in Pakistan and it has major side effects like nephrotoxicity which is also one of the reasons of its limited use and poor patient compliance. These can be the causes contributing towards its highest sensitivity observed against pseudomonal isolates in our study. Quinolones are one of the best treatment options for UTI mainly because of the easy accessibility in oral formula,good patient compliance due to its pharmacokinetics and cheaper cost. However, emerging quinolone resistant strains of *Pseudomonas aeruginosa* have been reported due to the changes in target sites of the bacteria and active efflux pumps operated to prevent the entry of the drugs.[8,9] These are the common factors contributing towards increasing resistance against ciprofloxacin. In our study resistance against ciprofloxacin is 43.3% which is quite similar to the figures reported in one of their studies carried in India which revealed the resistance rate of 31.5%.Resistance shown by these isolates are either acquired or due to the overexpression of MexAB-OprM efflux pump.[10] One of the recent studies carried out in Malaysia reported a high resistance pattern of ciprofloxacin against pseudomonas (92%).[11] Slight discrepancy observed between the results might be due to the patient awareness and clinician's approach to treat the patients admitted in intensive care units .

One striking observation in our study is the rising trend of resistance of pseudomonal isolates against carbapenems (meropenem 28.87% resistantt and imipinem 22.86% resistant.The figures observed are quite identical with another research conducted in Pakistan in 2018 showing a huge reistance rate due to the production of blaVIM Metallo-â-lactamase.[12]Carbapenem-resistant strains of *Pseudomonas aeruginosa* have been reported in various countries like Korea and China.[13,14] These strains resist carbapenems (both Imipenem and Meropenem). Regular use of Imipenem alters the outer membrane absorbency or when given along with Piperacillin, changes the target site of *Pseudomonas aeruginosa* which then eventually turned them resistant to carbapenems.

Results obtained for resistance towards the antibiotic amikacin(15.46%). are also in accordance with a study conducted in Pakistan in 2017 in Peshawar showing amikacin the most sensitive antimicrobial against pseudomonas isolates.[15] Higher resistance against aminoglycosides are also reported in studies carried out in Bangladesh and India.[16] Good sensitivity pattern is observed with the antimicrobials cefoperazone-salbactum (75.26%) which is in close similarity with a study conducted in China showing cefoperazone-sulbactam and amikacin are the choice of drugs available for carbapenem resistant strains,although their combination have no added advantage.[17] Cefoperazone acts on the cell wall, and salbactum acts as a beta-lactamase inhibitor, to amplify the bactericidal activity of cefoperazone against beta-lactamase-producing isolates.

The most resistant antibiotic found in our study is aztreonam 59.79%. Same resistance pattern was also reported in Saudi Arabia (63.4%).[18] Aztreonam has excellent efficacy against Gram-negative aerobic bacteria. Aztreonam has found to be more active than cefamandole in certain infections and quite same in efficacy to tobramycin or gentamicin.[19] Aztreonam has been proven to be an effective antibiotic in treating pseudomonal infections in most patients including burns and soft tissue infections (except in cystic fibrosis).[20] However aztreonam reistance has been emerging due to variable genetic mutations in pseudomonal isolates.[21] Combination therapies like aztreonam–monobactam can be a good option available for such resitant strains,but still under clinical trials for the treatment of serious infections caused by metallo-beta lactamase producing pseudomonas.[22] Therefore the inadequate number of pseudomonal infections available for our study from intensive care units thwarts any conclusions as to the relative efficacy of aztreonam compared with other antibiotic options available.

The antibiotic susceptibility pattern observed thin down the most sensitive drugs from the most resistant ones and our study can be used to provide a more detailed and comprehensive attitude to opt the most appropriate antibiotic therapy against *Pseudomonas aeruginosa*. Aztreonam, is an antibiotic used primarily to treat infections caused by *Pseudomonas aeruginosa*. The high resistance pattern observed in our study is quite alarming for both clinicains and microbiologist.We suggest a judicial and restricted use of this antibiotic in hospital settings. Regular monitoring of antibiotics with proper de-escalation should be adopted in order to decrease the emergence of such resistant strains.

CONCLUSION:

The prevelance of Pseudomonas aeuginosa isolated from the samples received at intensive care units(adult and neonatal) was 14.39%. Polymixin B was the only antimicrobial showing highest susceptibility trends against Pseudomonal isolates. Piperacillin-tazobactam is the treatment of choice for non-resistant isolates. Aminoglycosides like amikacin and gentamicin can also be one of treatment options available but cannot be used alone due to their poor intracellular penetration.

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Review Article

Carbapenem Resistance Of *Pseudomonas Aeruginosa*: A Review

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ABSTRACT

Carbapenem resistance towards the gram negative microorganisms especially *Pseudomonas aeruginosa* is alarming and on-going public health problem all over the world. It may be intrinsic or acquired through transcription of genes among microorganisms. These genes are spreading rapidly and are responsible for serious out-breaks. So selection of antibiotics is limited to treat these resistant cases. Resistant genes are commonly extended in Europe, Asia (Turkey, India, China, Pakistan and so on) and South America. In Pakistan carbapenem resistance in Pseudomonas aeruginosa isolates is increasing among hospitalized patients. It shows a progressive trend in multidrug resistance (even towards last resort drug carbapenem). In this article, we offer an in-depth review of carbapenem resistant *Pseudomonas aeruginosa*. This will facilitate the readers to take effective measures in order to control infection and appropriate use of antibiotics.

Keywords: appropriate antibiotics, Carbapenem resistant, transcription of genes.

INTRODUCTION:

Pseudomonas aeruginosa is opportunistic and non fastidious pathogen. This bacterium is a threat in particular to those hospitalized patients who are dependent on devices as ventilators, blood catheters, urinary catheters, i/v (intra vascular) catheters etc.[1]It leads to pneumonia, meningitis, skin infections, urinary tract infections, endophthalmitis and malignant otitis externa.[2] Different anti-pseudomonal drugs are available but it exhibits multidrug resistance. It has natural aptitude to adopt new ways to resist treatment. So carbapenem is a good choice but many surveillance studies conducted in USA and Europe have shown the increasing prevalence of CRPA (Carbapenem Resistant Pseudomonas Aeruginosa).[3, 4] Beta- lactamase and different genes are responsible for resistance.[5, 6, 7] Annually, worldwide antimicrobial resistance was projected to cost over $ 105 billion dollars.[8] In Pakistan,isolates of Pseudomonas aeruginosa are also detected to have a progressive tendency towards carbapenem resistance.[9] WHO publishes an antibiotic resistant list in which *P. aeruginosa* exhibits carbapenem resisttance is considered as critical pathogens.

METHODOLOGY:

Google and Google scholar search engine were employed with numerous key words and idioms to search articles related to antibiotic resistance epidemiology in the Gram negative rod shaped bacilli *Pseudomonas aeruginosa.* Articles were selected from 2013 to 2018 for write up of this review. Key words such as carbapenem resistance, *Pseudomonas aeruginosa* modified Hodge test and genes detection were used. A total of 200 articles included 5 review articles and 195 original articles. Among 200 articles 40 were short listed on the basis of correlation with my work. A major content of this article is based on genotype detection of antibiotic resistance by Molecular detection of genes that code for enzymes that cause resistance towards traditionally used antibiotics.

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LITERATURE REVIEW:

All over the World carbapenem starts to show its resistance especially hospitalized patients.[10]

Pathogenesis consists of different virulence factors like endotoxin, exotoxin, enzymes, adhesions, biofilms and pigments etc. Endotoxin A is responsible for tissue necrosis, it restrains protein synthesis through ADP-ribosylation of elongation factor-2. Different enzymes eg elastase and proteinases (zinc-metalloprotease and metalloendopeptidases) that make easy incursion into blood-stream. Adhesion through pili can kick off the biofilm formation and phagocytosis. Pigments especially pyocynin can disrupt the movement of cilia and facilitate the secretion accumulation.

LPS (lipopoly-saccharides) are recognasized by TLR4-MD2-CD14 which are located on macrophages and dendritic cells, so these take part in inflammation. Flagellum also plays in adhesion,invasion, biofilm formation and mediation of inflammation. Type IV pili binds to glycosphingolipid located on host epithelial cell-membranes.this facilitates the internalization of *P. aeruginosa.* Type IV pili contains pilin protein and indulges in inflammation.

Different drugs are in practice to control *pseudomonal* drugs eg β lactum antibiotics (penicillin, cephalosporine and carbapenem), fluoroquinolone and aminoglycosides. P. aeruginosa is defiant to multiple antibiotics because of unnecessary and inadequate use of these antibiotics.

CARBAPENEM ANTIMICROBIALS:

Carbapenems are β-lactam antibiotics. It binds to pencillin-binding proteins (PBP) and hinder the production of cell wall of micro organism.It is effective against both gram positive and gram negative bacteria[11,12]

Carbapenems were produced from thienamycin that is derivative of streptomyces cattleya. Carbapenems are analogous of pencillin but the sulfur atom in position 1 of the structure has been substituted with a carbon atom. Imipenem is the other antibiotic in this group which was introduced clinically in the United States of America in the year 1985.[13]

Carbapenems cannot move through bacterial cell wall. It pierces through porins. While crossing through the periplasmic space, carbapenems undyingly acylate the PBP (pencilline binding proteins). Carbapenem acts as inhibitors of the enzymes of PBP. At the end cell death of microorganism occurs due to osmotic pressure.[14]

Carbapenems are effective as Empiric monotherapy for ventilator associated pneumonia, intraabdominal infections and infections in cancer patients.[15] It is contraindicated in those who are hypersensitive because of increased risk of seizure.[16] Carbapenem can interact with live typhoid vaccine and probenecid. Different mechanisms are involved in carbapenem resistance like:

a. Loss of outer-membrane porins the upregulation of an efflux pump:

Pseudomonas aeruginosa exhibits resistance to carbapenem due to impermeability through cell-membrane. This impermeability is mediated by MexA-MexB-OprM. Gram negative organism's membrane is naturally designed with pores of Opr-M. MexB protein facilitates the exit portal. This pump comes across impermeability of drugs by upregulation of Mex A-MexB-OprM.This upregulation crop up as a result of nalB mutation.On the other hand Pseudomonas aeruginosa is also set up to lack Opr D proteins. These OprD pores allow the entrance of carbapenems. Whenever these pores are lost, carbapenems have to face the challenges of resistance.

b. Enzymes β-lactamases

β-lactamases are the major reason of microbial resistance to β-lactam medicine. There are four molecular classes of β-lactamases A, B, C and D according to Ambler classification. Three classes out of four posses serine amino acid at active site. These classes are A, C and D.

Class A β-lactamases are the most assorted and widely allocated class of the β-lactamases.[18] This class belongs to different enzymes which are chromosomally encoded, for instance NmcA (not metalloenzyme carbapenemases A), SME (Serratia marcescens enzyme), IMI-1 (Imipenem-hydrolyzing β-lactamases), SFC-1(Serratia fonticola carbapenemases-1). But plasmid encoded enzymes are KPC (kleibsiella pneumoniae carbapenemases) and GES (Guiana extended spectrum). Carbapenemases A is monomeric enzyme consisting of 265-269 amino acids. These enzymes inactivate β-lactams by hydrolysis before it reaches the PBP targets. In serine β-lactamases hydroxyl group breaks β-lactam ring. But in case of class B requires Zn^{+2} to facilitate the process of hydrolysis. KPC had spread all over the World especially Asia, NorthAmerican, European countries and Africa.[19]

Class B carbapenemases consists of enzymes New Delhi metallo- β-lactamase 1(NDM-1), Imipenem-resistant pseudomonas (IMP)-type carbapenemases, VIM (Verona integron-encoded metallo-β-lactamase), GIM (German imipenemase) and SIM (Seoul imipenemase). The NDM-7 carbapenemase has been recognized in 2008 in Escherichia coli in France.NDM-1 producing P. aeruginosa isolates are major intimidation to human beings.[20] . These enzymes were detected in pseudomonas, Acinetobacter and Enterobactericeae.[21]

Class D carbapenemases carbapenemases are OXA (Oxacillinase) enzymes. These enzymes are stumbled on P. aeruginosa and Acinetobacter bauminnii. OXA was first discovered in 1985 in Edinburgh, Scotland, by Pton et al. OXA type carbapenemases are majorly discrete in *P aeruginosa* and *A. baumannii.* Oxacillinase hydrolyses the isoxazolylpencillin oxacillin. Presently 121 variants of class D β-lactamases have been well-known on the basis of protein and 45 of them reveal carbapenem-hydrolysing activities.[22]

c. Intrinsic resistance

Pseudomonas aeruginosa acquires high intrinsic resistance. This resistance towards antibiotic inflicts financial burden and patient health.[23] intrinsic resistance occurs as a result of genes from naturally existing bacteria to clinical pathogen.

THREATS FOR ACQUIREMENT OF CARBAPENEM RESISTANCE

Resistant isolates of *Pseudomonas aeruginosa* can cause infections in immune-supressed conditions (neutropenia), elderly patients, hospital acquired infections (mechanical ventilation and organ transplantation), previous exposure to antibiotics, inadequate use of antibiotics and unnecessary use of antibiotics. In order to over-come the problems of antibiotic resistance, nonjudicious use of antibiotics in animals and plants should be avoided. Carbapenem resistance is common in developing countries in sub-saharan Africa.[24]

First report of carbapenem resistance from an aeromonas hydrophila isolate was observed in 1980 in Japan. Consecutively followed in London (1982) from Serratia marcescens (SME-1), IMI-1 from Enterobacter cloacae in Calfornia (1984) and NMC-A from Enterobacter cloacae in France (1990). Carbapenem resistant KPC was firstly recognized in 1996 in United States. Out breaks of KPC

were existing in Israel, Greece, Colombia, Canada, Australia and New-zealand.[25]From 2000-2010, infection rates has boosted from 1% to 12% in United States.[26] KPC gene positive *Pseudomonas aeruginosa* had found majorly from 2009-2012, 231 KPC- positive strains in 2010, 368 in 2011 and 293 in 2012 were reported[27] Class D carbapenemases are recently classified under four sub-groups, sub-group-1 is made up of OXA-23, OXA-27 and OXA-49, sub-group-2 is composed of OXA-24, OXA-25, OXA-26, OXA-40, sub group 3 is made up of OXA-51 and sub group 4 is made up of OXA-58. In Turkey, carbapenemase resistance is at high rate and resistance is screened for IMP, VIM, OXA-23, OXA-27, OXA-49, OXA-25, OXA-26, OXA-40 andOXA-48. The bla OXA-48 gene in *pseudomonas aeruginosa* isolates were discovered from December 2015 to January 2017 in Khartoum state (Sudan).[29] OXA-48 producing *Klebsiella pneumoniae* is recently monitored in France, Belgium, Israel, Russia and the Netherland and exhibit resistance to carbapenem. The bla OXA-48 gene has been recognized as insertion sequenceIS1999 in *Klebsiella pneumonia*. The bla_{OXA-48} was part of transposon (Tn1999) was practical. Now Tn1999 has been discovered in an E. coli in Italy.[30] .But now OXA-48 producing isolates have been documented in Lebanon, Sultanate of Oman, Saudi Arabia and Kwait. Resistance to colistin in *Pseudomonas aeruginosa* is uncommon but has been occurred.[31] Resistance to all antibiotics except the polymyxin is now common in numerous hospitals. Daplano et al explained outburst of panresistant *Pseudomonas aeruginosa* in an intensive care unit in Belgium. In Japan, surveillance for multi-drug-resistant *Psedomonas aeruginosa* described the metallo-β-lactamase gene bla_{SMP-1} involved in resistance from 2004 to 2006.[32]

PHENOTYPIC AND GENOTYPIC METHODOLOGY

It can be detected by disc diffusion, MICs, selective agar, modified Hodge test, synergy test, spectrometry, genome sequencing and molecular methods.

Phenotypic detection

First of all *pseudomonas aeruginosa* is detected by gram staining from different samples as gram-negative organisms. The organisms can grow on blood agar and MacConkey agar. On blood agar, colonies are grey and irregular as fig no 1A.

MacConkey is used to differentiate fermenters from non fermenters, as *Pseudomonas aeruginosa* is non fermenters as per fig 1B. *Pseudomonas aeruginosa* can also be detected by biochemical test eg oxidase test as per fig 1C. It turns purple within few seconds. For disc diffusion, organism is inoculated on Muller-Hinton Agar.[33] Then organism is incubated overnight after placing antibiotic discs. Next day inhibitory zone around antibiotic disc is measured according to CLSI.[34]

The modified Hodge Test (MHT) is inexpensive and practicable method. It is used for carbapenemase detection phenotypically and recommened by CLSI.[35]

Break points for carbapenem is revised every year according to CLSI and EUCAST.[36,37]Most Eurpean, Asian and African countries use EUCAST and CLSI as guidelines to detect carbapenemase producing isolates. Whenever these

Fig 1A.

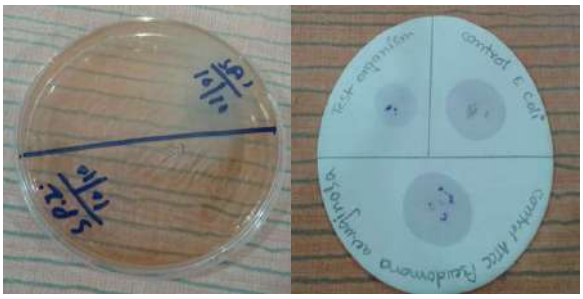

Fig 1B. Fig 1C

Antimicrobial Agents	Disk content	Zone diameter Break points			MIC Break points			Comments
		(Sens)	(Inter)	(Resist)	(Sens)	(Inter)	(Resist)	
Doripenem	10µg	>19mm	16-18mm	<15mm	<2mm	4mm	>8mm	Based on dosage regime of 500mg every 8hrs
Imipenem	10µg	>19mm	16-18mm	<15mm	<2mm	4mm	>8mm	1gm for 1hr and 500mg every 6hrs
Meropenem	10µg	>19mm	16-18mm	<15mm	<2mm	4mm	>8mm	1gm every 8hrs

Zone Break Points And Mic Break Points As Per CLSI

Table 1: As per CLSI

recommendations are delayed, clinicians and researchers come across problems. Modified Hodge Test is time consuming and unable to detect MBL, but 100% sensitive for class A and D.[38,39]

Genotype based technique

Molecular techniques are the most reliable methods for confirmation of carbapenemase production and resistance. Colonies of *pseudomonas aeruginosa* are detected by polymerase chain reaction within 4-6 hours with specific sensitivity and specificity. Wang et al informed a real time PCR with 100% sensitivity and specificity. Plasmid located genes like NDM, VIM, IMP and class D serine carbapenemase OXA-48 need to be identified on molecular basis. This procedure will improve the detection of unidentified genes and variants of these genes. A molecular technique consists of different methods like multilocus sequence typing, multilocus enzyme electrophoresis and DNA finger printing methods. OXA-48 is carbapenem-hydrolyzing oxacillinase and mostly ubiquitous carbapenemase in Europe and Middle East. Several variants of OXA-48, but most frequent OXA-48 like carbapenemases, have been documented which change from OXA-48 on basis of amino acids e.g, OXA-162, OXA-163, OXA-181 and OXA-204.[40]

It is confirmatory test for detection of carbapenem resistance. It is fast and reliable methods helping diagnosis and treatment.[41] Molecular methods can detect and differentiate carbapenemase consisting of NDM, KPC and OXA-48 mediated resistance, which is significant for epidemiological investigations.[42]

TREATMENT ALTERNATIVES IN CARBAPENEM RESISTANT PSEUDOMONAS AERUGINOSA

Pseudomonas aeruginosa is majorly involved in serious hospital-acquired infections. It builds up Sresistance to multiple antibiotics. Colistin is used as salvage therapy of *Pseudomonas aeruginosa* infections when resistance is common towards carbapenems. This drug reveals 98.8% susceptibility according to the U.S survey. According to retrospective cohort study, colistin was used in 23 seriously ill patients with multiple –drug resistance *Pseudomonas aeruginosa* infections; response was 61% with 3 patients showed resistance. Ceftolozane-tazobactam indicates excellent afficacy against many multi-drug resistant isolates. C/T was active against 95.2% CRPA (Carbapenem resistant *Pseudomonas aeruginosa*) clinical isolates. C/A (ceftazidime-avibactam) and C/T (ceftolozane-Tazobactam) are effective in case of over expression of oprD, efflux pumps and chromosomal ampC among non carbapenemase-producing CRPA clinical isolates.[43] Thirty seven patients were treated for bacteremia with carbapenem resistant (CR) pseudomonas *aeruginosa*. Among these patients, 65% of isolates exhibited multi-drug resistance.[44]

MEASURES REQUIRED TO OVERCOME CARBAPENEM RESISTANCE

Resistance towards carbapenem is identified as one of the greatest threats to human health over the World. Almost 2 million Americans per year suffer from nosocomial infections, leading to 99,000 deaths as a result of antibiotic resistance. Coordinated intrusion should be designed to get better use of antibiotics. Unecessary and inappropriate antibiotics should be avoided. We should discontinue the use of antibiotics for rapid growth and prevention of diseases in animals as well as in crops. The intimidation must be controlled by strict infection control policies and guidelines planned for every country. Skillful identification of carbapenemase productions by microbiology laboratories plays a vital role in infection control.[45] Therapy of resistant pathogen should be selected with regarding pharmacokinetics and pharmacodynamics.[46] *Pseudomonas aeruginosa* resistant genes have been detected globally, especially in Europe, South Asia and America. Hence there is an urgent need for infection control stewardship policy to control dissemination of resistance.

CONCLUSION:

Pseudomonas aeruginosa has the capacity of transferring antibiotic resistant genes to susceptible population of microorganisms.

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Review Article

Comparision Of BRAF V600E, COX–2 and p53 As Biomarkers For The Early Detection Of Colorectal Cancer

Hina Wasti, Summaya Shawana

ABSTRACT:

Colorectal cancer (CRC) is one of the most common types of gastrointestinal cancer. Almost two million new cases of CRC are diagnosed every year, making CRC the third most common cancer and the fourth most common cancer-associated cause of mortality in the world. The onset and development of CRC is induced by a combination of genetic and environmental factors including social, cultural and lifestyle factors. Age is considered as main risk factor for the colorectal cancer, there is remarkable increase past the fifth decade of life. Because of its high incidence and mortality rate worldwide, colorectal cancer (CRC) has become a global public health problem. Patients with CRC are typically asymptomatic and therefore it is difficult to diagnose disease until advanced stages, where the disease becomes incurable. Early diagnosis and therapy is able to decrease the risk of CRC in this asymptomatic population; however, early diagnosis of CRC remains a challenge in clinical practice. This review article was a comparative study and aims to explore the ability of the selected markers for early diagnosis of colorectal cancers for long term survival. Hence, identification of novel non-invasive diagnostic methods for early tumor detection in CRC is required. Screening of average-risk individuals can reduce CRC mortality by detecting cancer at an early curable stage. There is need for the implementation of new speci?c and more sensitive biomarkers in upcoming future which will improve diagnostic strategies and allowing clinicians to detect CRC cases in the earliest stages of the disease, to improve the prognosis of thousands of patients.

Keywords: BRAF V600E, COX-2, p53, Colorectal carcinoma, Early detection.

INTRODUCTION:

Colorectal cancer is the most prevalent cancer of gastrointestinal tract globally and is the leading cause of cancer-related death. Every year around two million new cases are diagnosed all over the world. The risk of colorectal cancer increases gradually and it is related with some demographic features like age, gender, disease history and lifestyle. There is diversity in genetic predisposition for colorectal carcinoma cases. There is transition of normal mucosa into a premalignant polyp and ultimately develops in to a cancer due to certain genetic and epigenetic changes.[1,2]

Recent evidence suggests that one third of sporadic colorectal cancers are thought to arise from the progression of premalignant serrated lesion.[3,4] There are various mutations involving the tumour suppressor genes,proto oncogenes and the genes responsible for DNA repair mechanisms.The mechanisms accountable for the pathogenesis of colorectal cancer are chromosomal instability (CIN), CpG islandmethylator phenotype (CIMP) and microsatellite instability (MSI). Mutations in APC, MYC, KRAS, BRAF, TP53, COX-2and TGF signaling genes are detected frequently in CRCs. [5] The reported 5-year survival rate of colorectal cancer is about 90% and it can decreases up to 14% for the early stage of this lesion. [6] CRC may develops from a benign adenoma, and the estimated time for the development of adenoma into adenocarcinoma is approximated 5–10 years.[7,8]At present colonoscopy is the gold- standard early diagnostic test to determine the colonic pathology.[9,10]Screening colonoscopy is estimated to reduce colorectal cancer incidence by 69% and mortality by 68%.[11]As colorectal cancer is a heterogeneous disease for which chemotherapy is considered to be the backbone of treatment, specific biomarkers now have been established for detection of tumors at an early stage of disease and also to predict the treatment adequacy to improve the survival time.[12,13] Some of such biomarkers include BRAF V600E, COX-2 and, P53.

BRAF (v-Raf murine sarcoma viral oncogene homolog B1) is a member of the RAS/RAF family, an which has coding an enzyme, and suggested as integral component of the cascade. BRAF oncogene has presented as direct effector of RAS and promotes the phosphorylation of MEK, which further causes tumour growth and survival. For colorectal cancer,BRAF mutations have accounts for about 8 % of cases. [14] At present >95% of BRAF point mutations have been observed to at BRAF V600 in which GTG>GAG substitution results in V600E amino acid change i-e the substitution of glutamate by the valine at residue 600. [15,16]

COX-2 has been recognized as important member of Cyclooxygenases (COXs) family which appears as an important regulator of cell proliferation. COX-2 is an inducible enzyme which accounts for the development of epithelial cell dysplasia, carcinoma, invasion and metastasis thus contributing to the development or progression of malignancy.[17]

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In various studies p53 has been recognized as a cellular SV40 large T antigen-binding protein. [18] The p53 signaling has been frequently impaired in CRC. p53 is a well-known tumor suppressor which promotes the transcription of various targeted genes namely p21 and p27. It also has a significant central role in cell cycle control, senescence, and apoptosis and in the prevention of cellular stress by mediating up-regulation of p21 and PTEN, inhibition of AKT, and decrease of cyclin E/CDK2.[19] Early detection of CRC improves the 5 years survival rate from 12-13% in stage IV metastatic disease to 90% in stage I-II early stage disease.[20] This review article was a comparative study and aimed to explore the ability of the selected markers for early diagnosis of colorectal cancers for long term survival.

METHODOLOGY:

A Literature search was done by using Pubmed and Google scholar from 2009 – 2018.Key words and phrases used were ,colorectal cancer, early diagnosis, biomarkers, BRAF-V600E, P53, COX-2. Multiple studies were scrutinized for the use of immunohistochemistry in early detection of colorectal cancer. 47 relevant articles are included for write up of this review article. The articles were analyzed and then composed the review article to assess the comparative analysis of these biomarkers for the early detection of colorectal cancer.

LITERATURE REVIEW:

BRAFV600E:

The BRAF oncogene is an integral component of the MAP kinase signaling pathway (RAS-RAF-MEK-ERK).Oncogenic activation of BRAF leads to constitutive kinase activity and phosphorylation of downstream targets of the RAS/RAF/MAPK signaling pathway.BRAF mutation constitues an alternative molecular pathway in the early carcinogenesis and accounts for 15% cases of sporadic colorectal cancer.V600E mutation is assumed an early event in serrated pathway of tumourigenesis and is greatly associated with proximal location, female gender, CpG island methylator phenotype and microsatellite instability (MSI). [21,22]

KRAS and BRAF are prime oncogenic drivers for colorectal cancer. Mutational analyses of these two important proto-oncogens have been a centre of research interest in recent years. [23] The *BRAF*V600E are generally mutually exclusive with another proto-oncogene such as KRAS, both of them have been implicated in the equivalent downstream effects in tumorigenesis . Mutations of these genes might play distinct roles in tumor initiation and/or maintenance. The activating *BRAFV600E* mutations have been revealed to play a role in tumor invasion and evasion of apoptosis.

BRAF mutations are found in 7% of cancers, with *BRAF* V600E accounting for >90% of mutations in *BRAF*-mutated cancers. Between 8% and 12% of metastatic CRC (mCRC) cases harbor a *BRAF* mutation. [24]*BRAFV600E* significantly increases the DNA methylation of CIMP-associated markers in primary colorectal tumors. Moreover these *BRAF* mutations and have been observed in early precursor lesions of colorectal cancer. [25] This mutation has been observed as an important predictive factor for adjuvant therapy for colorectal cancer thereby the mutation status of the tumor should be screened right before starting the treatment. The mutation status of BRAF has shown great diversity among different populations and regions.[26] BRAF mutations could be considered as a stratification factor for the adjuvant therapy.For patients having MMR-deficient (dMMR) CRC, BRAFV600E mutation revealed a sporadic origin.[27] Therfore both BRAF mutation and mismatch repair (MMR) statuses should be determined in all CRC to differentiate sporadic tumors from Lynch syndrome-related tumors.[28]

The early screening of *BRAF V600E* might improve the evaluation of the risks for colorectal cancer and give the effective management of the patients and also important in predicting the prognosis of early CRCs.[29] A study revealed BRAF mutations in 4.0% of colorectal cancers.[30] Previous studies have suggested *BRAF V600E* mutation as an independent prognostic factor which is significantly linked with prolonged DFS (disease free survival).The association of *BRAFV600E* and MSI phenotype showed a better survival for earlier tumor stage. [31]Various studies have proved that for (HNPCC) diagnostics, (a hereditary condition) *BRAF V600E* mutation within BRAF has been proposed as a convenient, reliable, fast, and low cost strategy which simplifies genetic testing for HNPCC and therefore should be recommended for early diagnosis as it can improve the efficiency of genetictesting for HNPCC. [32]

At present BRAF VE1 immunohistochemistry has been identified as a useful screening tool for the detection of *BRAF V600E* mutation in CRCs. The BRAF VE1 IHC is more cost-effective and less time-consuming than *BRAF* sequencing studies. [33] It reveals good diagnostic performance and excellent sensitivity on IHC (sensitivity, specificity, and positive predictive values are 96.1%, 94%, and 89.15 respectively).Moreover the BRAF *V600E* mutation has been appraised as an early event in colorectal cancer with multifaceted roles for progression, diagnosis and the prognosis of colorectal cancer. [34,35]

P53:

A well- known tumor suppressor gene p53 has been recognized as important components of our body's defense system which has been working for cancer progression control. Multiple studies have been proposed that the CRC carcinogenesis has significantly involves the mutations in various recognized proto-oncogenes namely the *K-Ras*, *APC* and *p53*. Among them the p53 mutation is playing an important role in colorectal carcinogenesis. It also helps in determining the biologic basis of the disease which is implicated in the early stages of ulcerative colitis and tumorogenesis of the colorectum.[39]

This marker has been considered as a good competitor for early detection marker panel of colorectal cancer. Along with it the antibodies to p53 tumor suppressor protein have been identified as early biomarker for colorectal cancer.[40]

The protein expression of p53 in dysplastic crypts may serve as an important biomarker for colorectal cancer.0–85% of colitis-associated cancers have defective *p53* gene which can be recognized via immunohistochemistry. Now a days screening method like Immunohistochemistry for the p53 biomarker in tissue samples has been considered as a useful tool for estimating the risk of morphological changes, distinction of intraepithelial neoplasms, and the progression in to malignant neoplasm involving the colonic epithelium. Also Immunohistochemistry (IHC) has been identified as a fast, most convenient and reliable method in detecting the level of p53 mutations in early precursor lesions for colorectal carcinogensis.[41]

The Overexpression of p53 in colonic epithelia has been identified as a most valuable tissue biomarker in surveillance of colorectal carcinogenesis. Also p53 has been recommended for a better quantification of the risk for colon cancer. The reactivation and remodeling of p53 function has an unconventional role in colorectal carcinoma. Proper understanding of screening of this marker may allow a better stratification of early dysplastic changes and invasive carcinoma, in order to personalize treatment and surveillance.[42]

COX -2:

The levels of cyclo oxegenase-2 in early stages of colorectal cancer have been proposed for the early detection of CRC. [43]It has been recognized as a useful diagnostic marker for the CRC patients with Stage I or II disease. Cox-2 has been found as potential blood markers and may be useful in identifying early stage CRC.[44] Targeting the inhibition of COX-2 expression may help to control the progression of carcinoma, including colonic carcinoma.[45]

Review of numerous studies reveales that expression of COX-2 is related with some important clinicopathological parameters namely the lymphovascular invasion, serosal involvement, metastasis of multiple lymph nodes, Duke's stage, and poorly differentiated cancer. It has synergistic effects in colorectal cancer carcinogenesis. COX-2 influences different steps in cancer progression. It increases the production of prostaglandin, inhibits tumor cell apoptosis and promotes cell proliferation and tumor angiogenesis, and activates the prototype of carcinogenic substances. Significantly higher levels of COX2 in precancerous lesions and carcinoma in situ point towards an early event in tumorigenesis.[45]COX-2 has also been recognized as a useful prognostic marker for colorectal cancer and its highest levels of expression may correspond with tumor recurrences. Routine screening of COX-2 may provide an effective index for prognosis of those at higher risk of disease metastases.

Further *BRAF V600E* is currently under focus as a potential prognostic and predictive biomarker which may improve assessment of colorectal cancer risk and guiding tool for patient management.

CONCLUSION:

BRAF V600E, p53 and COX-2 have been recognized as markers for early detection for colorectal cancer. Amongst these markers mutated *BRAFV600E* is now considered as the most promising tool especially those associated with MSI.

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Review Article

Diagnostic Utility Of Various Biomarkers For Prostate Cancer: A Review

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ABSTRACT:
Prostate carcinoma is one of the most widespread occurring cancers in males and is the second most common cause of cancer related mortality in men around the world. The therapeutic success rate for prostate cancer can be greatly improved if the disease is detected at an early stage. Therefore, a successful therapy depends immensely on the clinical indicators (biomarkers) for early diagnosis and progression of the disease, as well as the prognosis after the clinical intervention. Despite of its limitations, prostate biopsy is the "gold standard" for diagnosis of prostatic carcinoma. Beyond the shadow of doubt, the advent of PSA level has marked a new era for the efficient screening of prostatic lesions, but PSA alone cannot be considered as an authentic tool for diagnosis. Therefore, the use of new imaging techniques and molecular markers are of great importance for an accurate diagnosis of prostate cancer. Recent advances in molecular techniques have provided new tools facilitating the discovery of new biomarkers for prostate cancer. The aim of this review is to examine the current status of prostate cancer biomarkers, with special focus the on their diagnostic utility and therefore determine a panel of two or three markers for the prompt diagnosis of prostatic carcinoma.
Keywords: biomarker, risk factor, prostate cancer, screening of prostate cancer

INTRODUCTION:

Prostate, lung and colorectal carcinoma comprise 42% of all cases reported in men, with 1 in every 5 case being that of prostate cancer. Approximately, 164,690 new cases of prostatic carcinoma were reported in the year 2018 in United States of America causing an estimated number of 29,430 deaths in men. In the last ten years, the overall incidence of cancer in men has decreased by 2% annually, with a remarkable decline in prostatic carcinoma incidence of approximately 10% every year from 2010 to 2014.This decline in the incidence of prostatic carcinoma is accredited to effective screening measures by monitoring PSA levels in elderly men.[1] According to the annual cancer registry report ,published by ShaukatKhanum Memorial Cancer Hospital and Research Centre, prostate cancer is the second most commonly occurring malignancy in Pakistani men.[2,3]

Risk Factors:

Several risk factors might be involved in the carcinogenesis; these comprise of men above the age of 50 years, androgens, genetic factors, environmental factors, family history, high fat diet, alcohol consumption, cigarette smoking and acquired somatic mutations[4,5]. Androgens are of paramount importance. Carcinoma of the prostate does not develop in males castrated before puberty, which strongly suggests that androgens somehow provide the "soil," the cellular context, within which prostate cancer develops. Environment moreover plays a necessary role as validated by the fact that in Japanese emigrants travelling to United States of America, the incidence of this lesion increases. It is not prevalent within Asian population and is most frequent amid blacks and in the Scandinavian countries .Genetic factors also play a pivotal role, as there is a greater threat among first-degree family members of patients suffering from prostate tumor.

Acquired somatic mutations, comparable to other carcinomas, are the real drivers of cellular transformation.TMPRSS2-ETS fusion genes are found in about 40% to 50% of prostate cancers; it is assumed that the unregulated over expression of ETS transcription factors intervenes in the differentiation of prostatic epithelial cell.[4] Dietary factors include an increased intake of whole milk and dairy products is associated with aggressive prostate cancer lesions.[6] Likewise; an increased consumption of a high fat diet may also lead to the development of prostatic carcinoma.[7]

The human prostate gland is about the size of a walnut, positioned below the urinary bladder surrounding the urethra; it is approximately 30 centimeters in length and about 20 grams in weight, composed of glands with double layer, a flat basal cell layer and an overlying columnar secretory cell layer. The prostatic stroma consists of a mixture of smooth muscles and fibrous tissue. The prostate gland can be divided into various biologically distinct regions, of which the peripheral and transition zones are of prime importance. The peripheralzone makes up about 70% of the glandular prostate. Almost 70% to 80% of the carcinomas occur in this peripheral zone, whilehyperplasticlesions are most likely to be found in the inner transition zone.[8,9]

Prostatic carcinomas can be classified into two major groups: (1) carcinoma of large ('primary') ducts (2) adenocarcinoma of peripheral ('secondary') ducts and acini. This morphologic distinction has conservatively been based on the belief of a different site of origin for the two tumors. However, two patterns are sometimes seen together in the same tumor, and it is believed the two types may coexist in the same prostate

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as anatomically separate lesions. Therefore, it is assumed that it is the site of the growth rather than the originthat governs the tumor architecture.[9]

Androgens have a vital role in the stimulation of the prostatic cancer cells, which includes testosterone and dihydrotestosterone (DHT). Majority of the androgens are secreted by the testicles, whereas the adrenal glands also produce a minute amount of male sex hormones mainly () and . Prostate is responsible for converting testosterone into dihydrotestosterone (DHT). Alterations in the androgen receptors are responsible for the development of prostate cancer.[10]

Prostate cancer can be classified into various histological types; these comprise of acinaradenocarcinoma, ductal adenocarcinoma, transitional cell, squamous cell and small cell carcinoma.The Gleason Score is the grading system is utilized to determine the grade of the cancer. This system helps the clinician to select the effective treatment modality for this lesion. The most frequent sites of metastatic prostate carcinoma are lymph nodes and bone. Metastatic prostate carcinoma to the lung usually takes the form of multiple small nodules or diffuse lymphatic spread rather than large metastatic deposits.[11, 12, 13]

The clinical diagnosis of carcinoma of the prostate can be extremely difficult on prostate needle biopsy alone, and therefore the advent of immunohistochemistry has revolutionized the diagnosis of prostatic cancer. Prostatic carcinoma is developed due to various quantitative and structural genetic alterations. A number of genes have been involved in prostate cancer pathogenesis these include, ELAC2 (HPC2), MSR1, and RNASEL (HPC1) genes that have germ line mutations in prostate cancer; AR, ATBF1, mitochondria DNA, p36, PTEN, RAS BRCA1, BRCA2,, CYP17, CYP1B1,NKX3.1,CDKN1B(p27), and MYC. In this context, there is gain of function mutation as seen in AMACR and loss of function mutation as depicted by NKX3.1 AND p63. More genes relevant to prostate cancer remain to be appropriately analyzed these groups. Recognition and classification of these genes will be crucial for ameliorating the prompt diagnosis and management of this disease.[14, 15, 16]

Certain protective dietary factors have been recognized these include consumption of the isoflavonoidgenistein found in many cruciferous vegetables, family Brassicaceae such as cauliflower, cabbage, broccoli, brussels sprouts, legumes, lycopene present in tomatoes, and inhibitor of cholesterol biosynthesis (statin drugs).[17]

PSA levels and digital rectal examination (DRE) have been consistently used for the screening of prostate cancer. The DRE emphasizes on volume and consistency of the prostate gland. A major number of tumors originate in the peripheral zone and are easily palpable on DRE. Carcinomas are generally hard, nodular, and irregular.[18]About 25% of males having positive findings on DRE have more chances of being diagnosed as prostatic cancer. PSA testing is a comparatively economical technique and has more individual compliance. The prime benefit of this test is its remarkable sensitivity and a major drawback is its low specificity since some pathological conditions such as benign prostatic hyperplasia, acute and chronic prostatitis can also exhibit abnormally raised PSA levels. Such false-positive results in over diagnosis leading to further evaluation for confirmation of the diagnosis, causing an increased expenditure and further utilization of invasive procedures [19, 20]

A biomarker is described as, "a characteristic that can be measured objectively and evaluated as an indicator of normal physiological processes, pathological processes or pharmacologic responses to a therapeutic intervention" This definition explains the role of the biomarker, which includes population screening, diagnosis and prognosis.[21] Disease biomarkers may vary in their types and include visual inspection (e.g, blood in urine); biochemical, enzymatic, spectrometric, or immunological measurements; and molecular changes. The hypothesis behind the use of biomarkers is that a measurement that can be used as an indicator of a physiological process and as a proxy that whether a particular pathology is existent or not.[22]

METHODOLOGY:

A systematic search was performed using the PUBMED NCBI database. Forty two relevant publications were selected from this literature search. Recent studies were analyzed for the use of immunohistochemistry and other techniques in the early detection and diagnosis of prostate cancer. The articles were analyzed and methodologically assessed and a review article was composed to assess the basis of diagnostic utility of various markers used for the detection of prostate cancer.

LITERATURE REVIEW:

Important Biomarkers For The Diagnosis Of Prostate Cancer:

AMACR (alpha-methylacyl-CoAracemase):

AMACR (alpha-methylacyl-CoAracemase), is an enzyme which is localized to both mitochondria and peroxisomes. It is responsible for the beta oxidation of branched fatty acids, is especially over expressed in prostatic carcinoma. Increased expression of AMACR in the premalignant lesions i.e prostatic intraepithelial neoplasia tends to potentiate the risk of adenocarcinoma of prostate.[23] Alpha-methyl acyl-CoA racemase AMACR has been routinely over expressed in prostate cancer epithelium of premalignant and malignant lesions, hence it becomes an important definitive biomarker prostate cancer.[24]

NKX3.1:

NKX3.1 gene is located on chromosome 8p21, plays an important role in normal prostate development, regulating

proliferation of glandular epithelium and in the formation of ducts in prostate. It binds to androgen receptor at the ERG gene breakpoint blocking the contact of the TMPRSS2 and ERG gene loci; preventing their recombination.[25] NKX3.1 promotes the recruitment of cellular proteins that initiate homology-directed DNA repair. Absence of NKX3.1 results in the accumulation of the ERG gene breakpoint which leads to error-prone non homologous end-joining. Genetic studies carried on malignant lesions of prostate gland reveal that loss of NKX3.1 is positively correlated with TMPRSS2–ERG rearrangement.[26] NKX3.1 is seen to be under expressed in majority of high-grade prostate carcinomas and loss of expression is found in approximately 65% to 78% of metastatic prostate carcinomas.[25, 26]

p63:

The prostate gland is made up of two main epithelial cell types; the luminal and basal cells. The luminal cells are columnar; secrete prostate-specific antigen and specific markers, namely NKX3.1. Prostatic basal cells play an essential part in maintaining integrity of the ducts and survival of luminal cells; they express markers; p63 and cytokeratin.[28]p63 is an analogue of the tumor suppressor gene p53, which is necessary for formation of squamous epithelium. It is over expressed in normal prostate epithelial cells and is down-regulated in the presence of malignant lesion. Loss of the expression of p63 is strongly associated with the progression from benign tumors to carcinomas.[29]

Transforming growth factor-β1 (TGF-β1):

TGF-β1 is a family of polypeptides which is composed of a group of cytokines that take part in the various steps of tumorogenesis and increased levels of TGF-β1 have been seen in patients of several malignancies.[30]. Specific immunohistochemical studies have demonstrated overexpression of TGF-β1 in neoplastic lesions as to the normal prostate tissue.[31] H

igher levels TGF-β1 have been recorded in aggressive prostate carcinoma. Various studies reported that these levels were markedly raised in lesions of prostate cancer shown to have extraprostatic expansion involving the seminal vesicle and distant metastasis to lymph nodes.[32]

Glutathione S-transferase P1 (GSTP1):

This class of ubiquitous multifunctional enzymes is responsible for the conjugation of reactive substances with reduced glutathione (GSH) and detoxifying them. They play an important role in protecting the cells from free radical injury, and thereby up regulated in such conditions signify them as an excellent biomarker.[33]*GSTP1* methylation is frequently associated with tumor development or poor prognosis in a variety of tumors including neuroblastoma, hepatocellular carcinoma, breast, endometrial and prostate cancers. It is involved in the initial process of tumorogenesis in prostate cancer and its methylation has been inordinately researched. Different studies have revealed that about 70–80% of prostate cancer cases are methylated, whereas, cases hyperplasia of prostate are usually seen to be hypomethylated.[34] PCR analysis done in some studies revealed the presence of methylated GSTP1 in the urine samples of men that underwent prostatic biopsy. [33, 34]

Chromogranin A:

Chromogranin A is an acidic protein is a member of the granin family of proteins. It has been identified in all neuroendocrine cell types studied and is secreted in greater quantities than all other proteins synthesized by such cells. It is also called secretory protein I, which is encoded by the CHGA gene in human beings. The growth of prostate cells has been found to be regulated by peptides derived from chromogranin A. It has been analyzed as a marker for the diagnosis of prostate cancer.[3]

INTERLEUKIN-6:

Interleukin 6 is a cytokine produced by various types of cells and mediates distinct physiologic phenomena which includes; immune responses, cellular proliferation and their differentiation. Under normal circumstances, IL-6 levels in cells are nominal, although a number ofsignals mediate the marked expression of IL-6; for instance, the acute inflammatory responses to certain infections, for the activation of acute phase proteins etc. Elevated systemic levels of IL-6 have been correlated with diseases such as autoimmune disorders e.g hepatitis, arthritis, Crohn's disease, ulcerative colitis, pancreatitis, and various tumors.[36] Studies utilizing immunohistochemistry have elucidated that levels of IL-6 were18 folds increased in localized prostate cancer tissue.[37]

EPCA:

Early prostate cancer antigen (EPCA) is a nuclear matrix protein which is associated with prostate cancer. EPCA expressed only in the patients of prostate cancer, absent in the normal prostatic tissue. Furthermore, previous studies have evaluated that EPCA could aid in the diagnosis of carcinoma of prostate even at the initial phase of carcinogenesis.[38] Two EPCA subtypes have been recognized, the first one is termed as EPCA, and the second one is known as EPCA-2. Various studies have also revealed the possible benefits of EPCA-2 levels in blood samples of prostate cancer patients for clinical management of this lesion.[39]

PTEN:

The phosphatase and tensin homolog gene *(*PTEN) is situated on chromosome 10q23.3 and is localized to the mitochondria. It plays an essential role in cell cycle regulation and cell death by means of apoptosis, heavily regulated by post-translational modifications, which includes oxidation, ubiquitination and aceylation.[40]It is a negative regulator of the PIK3/AKT survival pathway and is the most commonly tumor suppressor gene in prostate cancer. PTEN expression and function, including transcriptional and post-transcriptional

regulation, post-translational modifications, and protein–protein interactions, have been shown to be altered in human prostate cancer.[41] PTEN is either lost or mutated in 50–80% of primary prostate cancer in majority of cases.[42] Despite some advancement, several limitations still exist with the current technology that hinders the discovery and development of new biomarkers for all forms of cancer including prostate cancer. A critical point that has been reiterated is the fact that an ideal biomarker has to show a high level of specificity and sensitivity to prevent false-positive screening tests, which will create anxiety in patients and lead to more expensive and invasive testing. Therefore, combining markers is thought to be the next best thing to improve the accuracy of diagnosing, treating, and surveillance of recurrence of prostate cancer.

CONCLUSION:

It was conculded that the increasing importance of biomarkers in screening and diagnosis of prostate cancer to reduce invasive follow-up procedures, help clinicians and pathologists for a prompt diagnosis and treatment. The application of biomarkers to prostate cancer is at the forefront of the research field because of the distinctive relationship between the genomic changes in the cancer cells and the disease progression.

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Case Report

Amyotrophic Lateral Sclerosis: The Most Common And Lethal Form Of Motor Neuron Disease-a Case Report From Middle East

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ABSTRACT:

A neurodegenerative disorder which is fatal, rapidly progressive and has no effective treatment till date is amyotrophic lateral sclerosis. Almost 90% of all cases occur in the sporadic form, with the rest occurring in the familial form. It is a devastating disease leading to death within 3-5 years in most cases. The diagnosis of AML is difficult to made in spite of acknowledgment for 140 years. It is diagnosed by clinical presentation which is a combination of upper and lower motor neuron signs and electro diagnostic studies which gives information about diffuse motor axonal injury. This neurodegenerative disorder results in degeneration of corticospinal tracts and anterior horn cells and involving motor neurons of the cerebral cortex, brainstem, and spinal cord. There are a variable signs and symptoms of this disease, so the diagnosis is very important for the management and better outcome of the patients. Cause of death in these patients is usually respiratory failure.

KEYWORDS: Amyotrophic Lateral Sclerosis, Motor Neuron disease, Neurodegenerative disorder, MRI

INTRODUCTION:

The most common form of is **Amyotrophic lateral sclerosis (ALS)**, which is also called **Lou Gehrig disease**,[1-3] which is causing progressive weakness and death of the patient due to respiratory failure. Others are also prone to the pseudobulbar affect.[1] It is a neurodegenerative disorder which is rapidly progressive and fatal resulting in degeneration of corticospinal tracts and anterior horn cells and involving motor neurons of the cerebral cortex, brainstem, and spinal cord. The diagnosis of AML is difficult to make in spite of acknowledgment for 140 years. The Escorial criteria are commonly used for obtaining a definitive diagnosis of ALS.[2] Within three years, more than 60% of patients died with AML. The treatment of choice for AML are Riluzole but it is effective for only two to three months.[3] The physician of patients with AML should manage the illnesses which can be treatable along with neuropathy. The underlying etiology is not known. Several genetic mutations have been found to be associated with familial ALS (FALS) which accounts for 10% of ALS.[4] With the combination of pathophysiology and newer treatment options, other diagnostic tests should be considered for the early detection of AML.

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CASE REPORT:

We have encountered a patient with age of 48 years who presented two months after the onset of bilateral lower extremity weakness which was progressive as it was initially started in his right leg and then progressed to his left leg. After that, patient felt muscle twitching. There was no known past medical or family history and no any history of trauma or insect bite. On Physical examination, the patient was thin, cachexic and well oriented with time, place, and person. He was fully conscious and no any loss of memory noted. However, decrease in strength is noted which was mild at the hips and knees but more pronounced at ankles and toes bilaterally, more marked on right side. There was fasciculation in both arms and legs. On sensory examination, sense of vibration was decreased in legs and sense of position was decreased in toes. Achilles tendon reflexes were absent but other reflexes were present and normal. No significant abnormality seen on cerebellar examination. He was unable to walk without support and unable to stand from sitting position. CT scans of the abdomen and chest were done which showed no significant abnormality. So the diagnosis of acquired polyneuropathy was made for which treatment was started with 60 mg steroids and 120 mg azathioprine daily. But no positive change in the disease process was seen and the patient was deteriorated with progressive decrease in strength in arms, legs as well as intrinsic muscles of hands. Further work up was done with MRI brain in axial, coronal and sagittal planes which showed increased signal intensity in corticospinal tracts bilaterally on T2WI and DWI. Thus, on the basis of clinical history, examination and MRI brain findings, diagnosis of Amyotrophic lateral sclerosis was made.

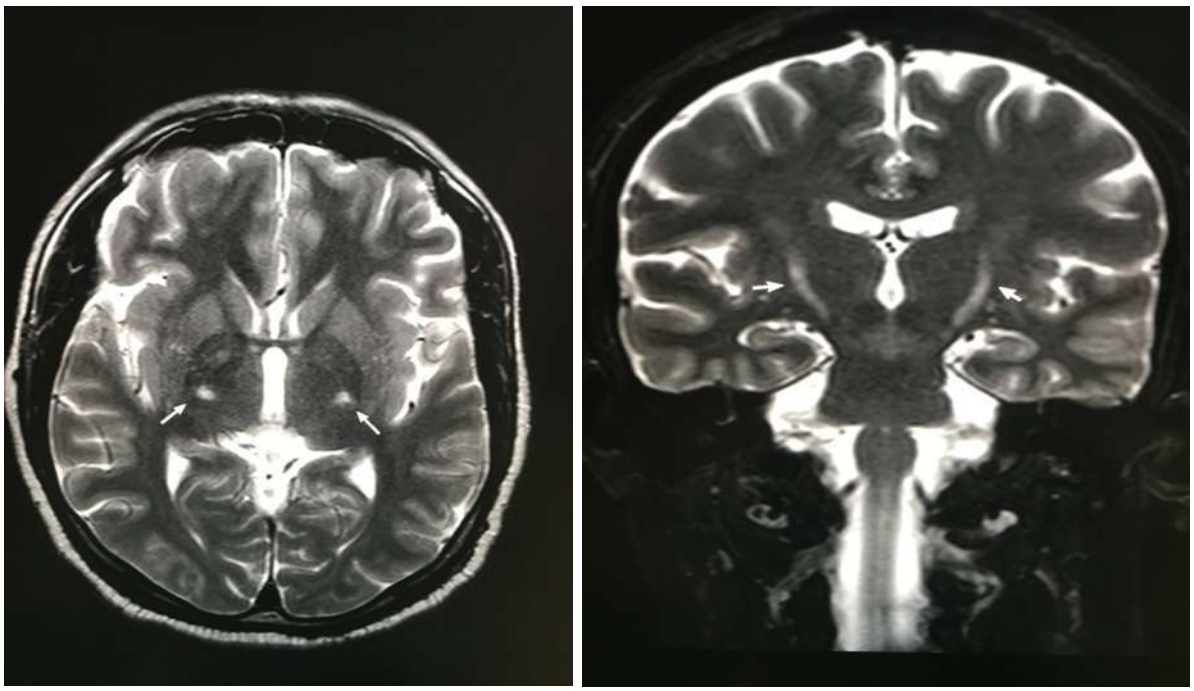

Figure 1: T2WI in Axial and Coronal sequences showing high intensity signals in corticospinal tract (internal capsule).

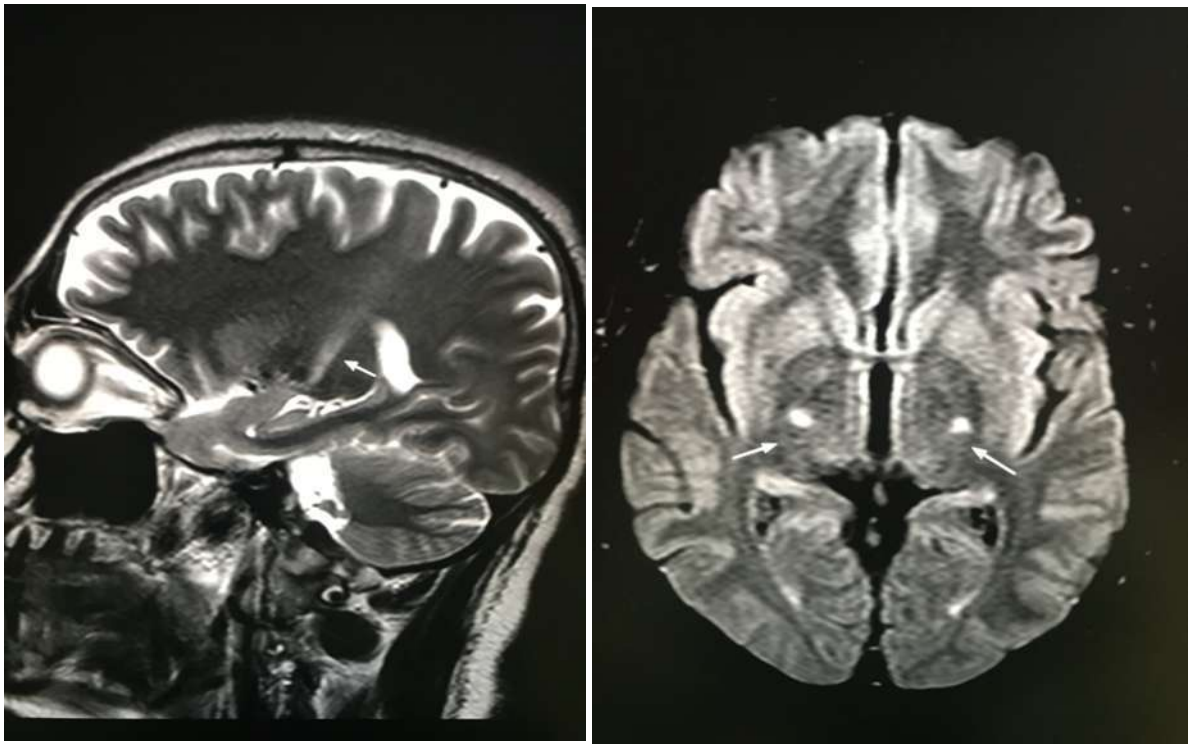

Figure 2: T2WI and DWI in Sagittal and Axial planes showing high intensity signals in corticospinal tract (internal capsule).

DISCUSSION:

ALS is diagnosed on the basis of signs and symptoms of upper and lower motor neurons dysfunction along with laboratory evidence. No any significant test is present for the diagnosis of AML which is widely accepted. It is diagnosed with electro diagnostic studies. Neuro imaging and lab tests are used for the exclusion of other diseases.

Signs and symptoms of ALS include asymmetric weakness of extremities (60%– 80%), bulbar symptoms (20%), respiratory muscle weakness (1%–3%), generalized weakness

in limbs and bulbar muscles (1%–9%), axial onset with head drop or truncal-extension weakness, muscle atrophy, fasciculations, and cramps.[5-6] Although diagnostic criteria for ALS were revised in 2000, the unavailability of the specific diagnostic test is a big hindrance to detect ALS in the early stages.[7]

Weight loss is also seen in AML which is a late finding and is due to difficulty in swallowing and difficulty in respiration during eating along with weakness of muscles of mastication and increase metabolic state.[8-9] Due to the insufficiency of dietary intake and weight loss, the state of catabolism and weakness of respiratory muscles become worsen progressively. So the vicious cycle is continued and the disease get worsen. The poor prognostic factor is weight loss and BMI =18.5 kg/m2. The life of ALS patient is associated with the function of respiratory muscles. The episodes of apnea and dyspnea are poor factor for the quality of life of patients. With the combination of clinical history, physical examination and MRI brain, the diagnosis of AML was made. AML was treated with corticosteroid with symptomatic treatment, but due to the involvement of respiratory muscles the patient is died due to respiratory insufficiency.[10-11]

Various questions in a survey were asked from university students regarding ALS which showed some heard about this disease first time, only some of them have basic information about this disease, some had knowledge about signs and symptoms and only some of them have knowledge regarding its diagnosis and treatment strategies. These types of awareness studies helpful for medical student about disease, symptoms, treatment and precaution etc.[12-17]

CONCLUSION:

In Patients with rapidly progressive muscle weakness and neuropathic symptoms, one should suspect AML. The diagnosis of AML should be made early because the disease should be arrested before progression. The occurrence of the disease is increasing every year, so the attempt should be made for the research and awareness of the disease process.

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Importance of Quality in Medical and Dental Institutes

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ABSTRACT:

There is a dire need to incorporate quality improvement measures in medical education of our country as now worldwide the paradigm is shifted from the students' outcome to the continuous quality improvement (CQI) measures. The ultimate goal of CQI is to provide optimal patient care. This is the responsibility of the accreditation council (PMDC) to ensure the true application of quality improvement indicators in medical institutes. The quality document introduced by the PMDC is the true reflection of the world accreditation bodies WFME, WHO. By ensuring the true application of this quality document, Pakistan Medical and Dental Council will be able to compete with the International standards of medical education through competent future physicians.

Key words: Accreditation, quality, quality document.

INTRODUCTION:

"A physician who is excellent academically with the science of medicine but incompetent in the art of medicine due to lack of experience and updated knowledge is like a one winged bird who is inept to fly high in the sky". (Shushruta Samhita 300-400 BC3)

The profession of physician is the most respectful in every era of mankind. For the sake of credibility of this relation, it is imperative to be skilled and updated. This is difficult to establish direct link between the education quality and accreditation. [2-7]

The political, social and economic challenges have poorly impacted the quality of medical education in our country, so this is the time to revise and reassure the true application of the quality standards [1,8] to face these challenges by ensuring optimal patient care.[2] The accreditation body of the country is sole responsible to bring the quality standards, framework and professional guidelines in medical education to shape the physicians of today and tomorrow. [1]

Quality is a subjective and vibrant term so a bit difficult to express and has multiple meanings for various stakeholders. In medical education, quality is defined as "to adapt certain standards in medical education approved by an accreditation council or an external agency"[1]

Moreover, quality in education is defined as value for money to reach certain bench marks. Quality can be estimated by number of quality researches in an educational Institute.[8]

Furthermore, quality in education is the achievement towards mission and vision of an Institute.[9]

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Quality in medical education.

The purpose of considering quality in medical education in Pakistan is to develop the today's and future doctors as a competent healthcare personnel. It is already emphasized in "Tomorrow's Doctor'; that there is an emerging need to develop a framework to ensure the quality and competency among future physicians.[9] This framework should reflect the bench marks and standards approved by the WFME (World Federation of Medical Education) and the WHO (World Health Organization).[2,10] These are the international accreditation agencies of medical education. Quality in Medical Education is a growing demand of all the stakeholders due to following reasons. [2,10]

- To ensure that the students admitted in the Institute will become competent after qualifying.
- The future doctors would be able to compete with the international standards of medical education.
- The institutes have to accomplish and adopt the criteria of an accreditation council.

The PMDC council is the accreditation body in Pakistan. To compete with the world standards of Medical Education, it has formulated the quality document approved by the WFME by the help of the competent team of medical educationalist which then followed and adopted by the entire medical Institutes of Pakistan. The core outcome to bring quality in medical education is patient centered care.[2, 10-12]

Quality document of PMDC:

Following factors are considered by PMDC[13,14] while formulating the quality document for medical colleges of Pakistan. This quality document is addressing the core competencies of the physician as prescribed by WFME.

- To ensure the homogenous training of future physicians.
- To establish the transparent mode of entrance examination.
- Development of centrally regulated curriculum.

- Predefined framework of self evaluation and academic audit conducted within Institute and for external audit.
- To enhance teaching strategies which make the future doctors a critical thinker, problem solver, decision maker and overhaul a competent healthcare provider.
- To ensure the transparent induction of qualified teachers in medical and dental Institutes.
- There should be well established department for monitoring and evaluation which ensure quality assurance in course evaluation, peer evaluation and assessment. The following task should be performed by this department.[1,5]
  - This department is responsible for the supervision of valid and reliable assessment system.
  - Self evaluation and quality assurance.
- Infrastructure of the Institute is reflecting the facilities for the students and the catered community.
- There should be sufficient manpower in the Institute. (Trained faculties along with the subject specialist).
- Activities related to teaching and learning should be standardized.
- To ensure structured assessment methods to appraise the attitudes, skills and knowledge which are properly aligned with learning strategies and learning outcomes.
- An objective and structured peer evaluation is incorporated in a regular feedback of students and alumni of the course.
- The university has to make arrangements for external examiners to conduct final university examinations.

How to determine the quality of undergraduate and post graduate institutes across Pakistan?

To determine the quality of undergraduate or post graduate medical education across Pakistan is judged through the student's outcomes by passing rate of Pakistani graduate physician in USMLE, national examination and other foreign licensure exams which is frequently measures in literature.[15] Other way of determining the quality of undergraduate and post graduate education in Pakistan is the framework of accreditation standards in PMDC which is highlighted by following parameters.

- Mission statement should be clearly displayed on the Institute's Website and well written in prospectus.
- Outcomes of the undergraduate program should be well defined which explain the differentiation of the similar courses with other Institutes.
- Curriculum document of the Institute will reflects the Institutional autonomy and academic freedom by policy document, implementation of curriculum, educational outcomes of the program, appropriate uses of curriculum resources and evidence based educational pedagogy.
- Curriculum design should address the national and international needs of the community and student should get the exposure with the patients in early years of education.
- Curriculum should be designed according to the framework proposed and prescribed by the International accreditation agencies.
- Half of the course will be based on the clinical sciences.
- Learning in clinical sciences should be structured and planned.
- Quality improvement course content will be integrated from very initial years of undergraduate medical education.
- Mission and vision of the program should be aligned with the curriculum design; which should be outcome based, community oriented patient centered and will promote health and prevents disease.
- To ensure the implementation of learning strategies which enhance active learning like PBL, student centered learning, self directed learning, e-learning, simulation, inter professional learning, peer learning.[5]
- Ensure organized and systemic learning in clinical settings by incorporating electives in curriculum.
- Educational content of the program should be developed with the consensus of the cohort of subject experts.
- Educational content should be aligned with core competencies of the accreditation agency, should be outcome based and should be relevant to the need.
- Educational content will highlighted the patient safety, professionalism and ethics, research, life saving skills, evidence based medicine, behavioral sciences and communication skills to produce safe healthcare providers.
- Integrated curriculum should be formulated by the subject experts from the clinical and basic sciences of that year of undergraduate program.
- Study skills, leadership skills should be the part of the educational content.
- Curriculum management should be maintained and managed by the curriculum committee[6].
- Every undergraduate year should have separate study guides which express the details of learning objectives, mode of teaching, modes of assessment.
- Assessment ensures the balance between formative and summative assessment.
- Wide range of assessment methods should be inculcated in educational content like portfolios, mini.
- Assessment method should be compatible with the educational strategy and learning outcome.
- Workplace based training should be structured and encouraged.
- Standard setting processes should be inculcated in examination items.
- There should be a student support program in every Institute like Mentoring.
- Students should comply the admission policy in concordance with the international standards.
- To ensure the availability of student counselor in every Institute.
- To ensure the presence of student representative in every committee related to student
- To maintain the quality standards, timely inspection is recommended.
- The training of the future doctor is reflected by the

deliberate practice instead of unstructured training for that every institute should ensure to make training electives more outcomes based and individual patient care.

- Availability of simulated patient is essential to practice as many times as needed to become skilled as cognitive psychological research on expertise evidenced that to become expert consistent practice is essential which is not possible on live patients.
- Continuous supervision and feedback should be provided to every student.
- Effective learning practice should be ensured by performing tasks repeatedly.
- Wide range of assessment methods should be inculcated in educational content to assess competencies like portfolios, 360^0 feedback, and mini-clinical evaluation exercise with a positive attitude.
- To ensure the quality in research standards with relevant interdisciplinary collaboration.
- The national journals have to raise the standards of journals and so the original research.

CONCLUSION:

It is concluded that the quality document introduced by the PMDC is the true reflection of the world accreditation bodies WFME, WHO. By ensuring the true application of this quality document, Pakistan Medical and Dental Council will be able to compete with the International standards of medical education through competent future physicians.

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Emergence Of XDR Typhoid: An Alarming State To The Health Professionals

Ayesha Shakeel Ahmed

Dear Sir,

Keeping in mind the outbreak of typhoid fever in Pakistan, we would like to bring an important issue to your attention. Typhoid is a serious illness and is a major cause of morbidity and mortality in infants and adults. It is endemic in areas where the establishment of pure water supplies and sewage control are insufficient, namely, the Indian sub-continent, South-East Asia, Africa, the Middle-East, South and Central America.[1] Disease is spread via contaminated food and water and poor hygiene, resulting in a series of manifestations. Disease remains endemic in our country with a higher health and financial burden to the community. Despite improvement in the access to the health facility and literacy, prevalence of typhoid remains unchanged. Pakistan has the highest incidence (451.7 per 100,000 persons/year) of typhoid fever.[2] Asia also has the highest regional frequency rate of 274 cases per 100,000 populations, which is five times greater than the second highest, Latin America.[3]

Typhoid carries a high incidence of morbidity and mortality; it can affect any age group, but the greatest incidence is found in the pediatric age group.[4] Symptoms include high fever, headache and, abdominal pain, diarrhea/constipation, cough and loss of appetite. It is also known to have very serious complications such as internal bleeding, perforation and encephalopathy. Initially, typhoid was easily treated by the first line antibiotics, like amoxicillin, chloramphenicol and co-trimoxazole.[5] These drugs remained the drug of choice for patients with typhoid for many years, but due to excessive use, Multidrug Resistance (MDR) strands of Salmonella Typhi emerged. This resulted in a shift to 3rd generation cephalosporins like ceftriaxone, cefixime and fluoroquinolones. This group of antibiotics remained to show sensitivity for nearly the last two decades. However, irrational use of these drugs too, led to the evolution of Extensively Drug Resistant (XDR) strands which were resistant to these medications as well. Reduced susceptibility to Fluroquinolones and 3rd generation Cephalosporins and first, the emergence of Multidrug Resistance (MDR) and now Extensively Drug Resistant (XDR) strands has complicated treatment. In November 2016, an outbreak of XDR typhoid fever began in Hyderabad and spread to the city of Karachi and to several districts, leading to several deaths. . Despite the rapid spread of highly resistant typhoid across Sindh, the rural areas are deprived of any public sector facility capable of carrying out the gold standard laboratory test for typhoid, a blood culture. During 2016-18, 8188 cases of typhoid have been reported out of which 64% were XDR typhoid. Now, there is evidence to believe that strands resistant to the drugs that worked against XDR are emerging. Apart from parenteral Carbapenems, Azithromycin is the only oral/outpatient treatment option available for XDR Typhoid.

These facts were stressed on the need for a through history to be taken and detailed investigations to be performed to rule out XDR typhoid, before starting treatment, as azithromycin is the only oral drug available for XDR typhoid and the growing resistance to it is worrisome. Emphasis should also be placed on the administration of the typhoid vaccine in patients with increased risk of exposure. It is a parenteral polysaccharide Vi vaccine, administered as a single 0.5mL intramuscular injection, at anytime after the age of 6 months. We would like to strongly suggest judicial and restricted use of azithromycin for 'non-typhoid' infections, such as upper respiratory tract infections. These measures can play a great role in decreasing the incidence of XDR typhoid. We also suggest regular monitoring of antibiotic usage, with the aim that suitable reduction in the use of these antibiotics be adopted in order to reduce the emergence of resistant salmonella strains in our community.

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Instructions to Author

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1. Abstract

It should have no more than 150 words for unstructured abstracts or 250 words for structured abstracts. The comprised heading of abstract would be (objective, Study design and setting, methodology, results and conlcusion. Below the abstract provide, 3-10 key words that will assist indexers in cross-indexing the article and may be published with the abstract in alphabetical order.

2. Introduction:

State the global and national dataon the title which summarize the rationale for the study. Give only strictly pertinent references (at least 15) and do not include data or conclusions from the work being reported.

3. Methodology:

Describe your selection of the observational or experimental subjects (patients or laboratory animals, including controls) clearly. Identify the age, sex, and other important characteristics of the subjects. Identify the methods, apparatus (give the manufacturer's name and address in parentheses), and procedures in sufficient detail to allow other workers to reproduce the results. Identify precisely all drugs and chemicals used, including generic name(s), dose(s), and route(s) of administration. For randomized clinical trials provide information on all major study elements, including the protocol (study population, interventions or exposures, outcomes, and the rationale for statistical analysis), assignment of interventions (methods of randomization, concealment of allocation to treatment groups), and the method of masking (blinding). Authors submitting review manuscripts should include a section describing the methods used for locating, selecting, extracting, and synthesizing data. These methods should also be summarized in the abstract. All studies must be approved by the relevant Ethics Committee/Institution Review Board of the respective institutions. Specify any general-use computer programs employed for analysis.

4. Results:

Present your results in logical sequence in the text, tables, and illustrations based on the objective of the study. Do not repeat in the text all the data in the tables or illustrations; emphasize or summarize only important observations. Describe appropriate indicators of measurement error or uncertainty such as confidence intervals, P values. Report complications of treatment and dropouts from a clinical trial.

5. Discussion and Conclusion:

Emphasize the new and important aspects of the study and the result. Do not repeat in detail data or other material given in the Introduction or the Results section. Include in the Discussion section the implications of the findings and their limitations, including implications for future research (recommendation). Relate the observations to other relevant studies.

Conclusion should linked with the goals of the study.

6. Acknowledgment:

List all contributors who do not meet the criteria for authorship, such as a person who provided purely technical help, writing assistance, or a department chair who provided only general support. Financial and material support should also be acknowledged.

7. Authorship

Authorship credit is based only on the criteria laid down by International committee of Medical Journal Editors (http://www.icmje.org/recommendations/browse/roles-and-responsibilibies/defining-the-role-of-authore-and-contributors. html).1) substantial contributions to conception and design, or acquisition of data, or analysis and interpretation of data; 2) drafting the article or revising it critically for important intellectual content; and 3) final approval of the version to be published. 4) Agreement to be Accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. All Conditions must be met. Authors should provide a description of what each contributed.

8. Conflict of interest

All authors have to disclose and submit any financial /personnel relationship that might bias and inappropriately influence their work.

9. References

Majority of the references must be from last five years. Local references must also be included. Vancouver style should be followed. Examples are:

a) Standard journal article

List the first six authors followed by et al.
I)Less than 6 authors:

Vega KJ, Pina I, Krevsky B. Heart transplantation is associated with an increased risk for pancreato-biliary disease. Ann Intern Med 1996; 1;124 (11):980-3

II) More than six authors:

Parkin DM, Clayton D, Black RJ, Masuyer E, Friedl HP, Ivanov E, et al. Childhood leukaemia in Europe after Chernobyl: 5 year follow-up. Br J Cancer 1996;73:1006-12

b) Organization as author

The Cardiac Society of Australia and New Zealand. Clinical exercise stress testing. Safety and performance guidelines. Med J Aust 1996; 164: 282-4

c) No author given

Cancer in South Africa [editorial]. S Afr Med J 1994;84:15

d) Chapter in a book

Phillips SJ, Whisnant JP. Hypertension and stroke. In: Laragh JH, Brenner BM, editors. Hypertension: pathophysiology, diagnosis, and management. 2nd ed. New York: Raven Press; 1995. p. 465-78

e) Newspaper

Hasan Mansoor. Excessive use of drugs creating resistance to antibiotics. The Dawn 2013, 24 June; sect. Metropolitan (col.1-4)

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Type or print out each table with double spacing on a separate sheet of paper. Number tables consecutively in the order of their first citation in the text and supply a brief title for each. Give each column a short or abbreviated heading. Place explanatory matter in footnotes. Explain in footnotes all nonstandard abbreviations that are used in each table. Identify statistical measures of variations, such as standard deviation and standard error of the mean. Do not use internal horizontal and vertical rules.

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Units of Measurement

Measurements of length, height, weight, and volume should be reported in metric units. Temperatures in degrees Celsius, Blood pressure in millimeters of mercury and all hematologic and clinical chemistry measurements in the metric system in terms of the International System of Units (SI).

Abbreviations and Symbols

Use only standard abbreviations. Avoid abbreviations in the title and abstract. The full term for which an abbreviation stands should precede its first use in the text unless it is a standard unit of measurement.

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3	Original Article	Structured (250)	3-10	2500-3000	20-30	4	3
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		3. Reproducible work (guide lines, questionnaire)	Mention Source, Accessed on, Retrieval date				
5	Short Communication /Commentary/ Opinions/ Perspective	-	-	1200-1500	15-20	2	1
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