

# Evaluation of Hematological and Coagulation Parameters in Oral Squamous Cell Carcinomas: A Study at a Tertiary Care Centre

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## ABSTRACT

**Introduction:** Oral cancer is one of the most common cancers worldwide and a major health problem in India accounting for 30% of all types of cancer. Platelet plays an essential role in physiological and pathological processes such as coagulation, inflammation and thrombosis. Platelet count (PC) and platelet indices (PI) such as, mean platelet volume (MPV), platelet crit (PCT), platelet distribution width (PDW) and platelet large cell ratio (PLCR) are known to be affected in various clinical conditions and recently their role as an inflammatory marker in cancer cases is being studied. However, data regarding oral cancer is limited. Authors say activated platelets with higher volume are associated with increased thrombotic potential through the activation of the coagulation cascade.

**Objectives:** To evaluate PC, platelet indices and coagulation profile in patients of oral cancers and compare them with healthy controls. Also, to study the relationship of platelet indices and coagulation parameters with clinicopathological framework in oral cancers.

**Results:** Statistically strongly significant differences on comparing health individuals with oral cancer patients were observed in MCH, RBC, WBC, PDW, MPV and APTT. Furthermore, RBC count emerged as a parameter of significance between different histological grades of oral cancer.

**Conclusion:** To summarise, simple and cost effective yet integral methods like platelet, haematological and coagulation parameters should be considered as an essential part of the work up of oral cancer patients in order to arrest delay in diagnosis henceforth propelling timely diagnosis, commencement of treatment and improving the overall prognosis of oral cancer patients.

**Keywords:** Oral cancer, Platelet indices, Coagulation cascade, Clinicopathological

## INTRODUCTION

Oral cancer is one of the most common cancers worldwide and a major health problem in India accounting for 30% of all types of cancer.<sup>1</sup> Cancer is generally related to platelet activation and thromboembolic events.<sup>1</sup>

Platelet, an important element of blood, plays an essential role in physiological and pathological processes such as coagulation, inflammation and thrombosis. Platelet count (PC) and platelet indices (PI) such as, mean platelet volume (MPV), platelet crit (PCT), platelet distribution width (PDW) and platelet large cell ratio are a group of parameters obtained from automatic haematology analysers and act as an important tool to measure the total number of platelets, morphology and proliferation kinetics.<sup>2</sup> Among these, MPV is most extensively researched and an early marker of platelet activation.<sup>3</sup> MPV reflects the average size of platelets while PDW depicts the heterogeneity in platelet volume.<sup>4</sup> PCT acts as an indicator of platelet mass/unit volume and is calculated as  $PC \times MPV/10,000$ .

Platelet indices are known to be affected in various clinical conditions like diabetes mellitus, pre-eclampsia and myocardial infarction.<sup>4</sup> Recently, their role as an inflammatory marker in cancer cases is being studied. These are clinically more important for both tumour prognosis and diagnosis. Platelet indices are studied in various cancers as they are important markers of inflammation but the data regarding oral cancer is very limited. On extensive literature search, we came across a single study from India that is based on study of platelet indices in oral cancer.

Platelet indices which reflect platelet morphology, namely PDW, and PCT also play a significant role in atherosclerosis and thrombosis.<sup>5</sup> Authors say activated platelets with higher volume are associated with increased thrombotic potential through the activation of the coagulation cascade.<sup>4,6</sup> Coagulation state has been studied in various cancers however on review of

literature, we could not come across any study on coagulation in oral cancer patients. The aim of this study was to evaluate the haematological parameters including platelet count (PC), platelet indices (MPV, PDW, PCT) and coagulation profile (APTT, PT and INR) in patients of oral cancers vs healthy controls and to analyse the relationship of these parameters with clinicopathological framework in oral cancers.

## MATERIALS & METHODS

The study was conducted in the department of Pathology, ESIC Medical college and Hospital, Faridabad, Haryana. It was a hospital based, cross sectional and analytical study conducted for a period of 3 months. 30 patients of oral cancers were included with similar number of age and gender matched controls. Clinico-pathological details (age, sex, tumour grade, stage, type, lymph node status etc) of all patients were recorded wherever available. Venous blood samples were collected under aseptic conditions in potassium EDTA and citrate vacutainers for estimation of haematological indices and coagulation parameters respectively. Samples were tested within 1 hour of collection to minimize variations. Complete blood count with platelet indices estimation was done using 5 part haematology analyzer (Sysmex XN 1000). APTT, PT and INR measurement was done with the coagulation method on a fully automated coagulometer (Stago STA Compact Max). All biopsy proven cases of oral cancer were included in the study. Nutritional anaemia can be a cause of reactive thrombocytosis thereby, increasing MPV, so, male patients with Hb less than 13 gm%, female patients with Hb less than 12 gm% were excluded from the study. Other patients excluded from the study were those suffering from bleeding disorders like Haemophilia, patients on anti-platelet drugs such as aspirin and clopidogrel etc and patients with any other diagnosed malignancy or thrombocytopenia.

The study was conducted after approval from Institutional Ethics Committee. Objective of the study was explained and confidentiality & anonymity was assured to the participants and a written informed consent was taken from the participants prior to sample collection. All data was compiled and analysis was done using appropriate statistical methods. Level of significance was set as p-value <0.05. Data was analysed using SPSS software.

**RESULTS**

The study comprised of 30 patients of oral cancer patients. In the present study, oral cancer was seen more in males (76.6%) than females (23.3%) and the most common age of presentation was >60 years (53.3%). Majority of them were smokers (66.6%) with 60% of them having a smoking duration of >20 years. In addition to smoking the most frequently observed substance abuse was tobacco chewing (6.6%). Base of tongue (26.6%) was the most commonly involved site followed by buccal mucosa (23.3%) and oropharynx (13.3%). (Table I)

CLINICAL PROFILE	NO. OF CASES & %
<b>GENDER</b>	
Male	23 (76.6)
Female	7 (23.3)
<b>AGE (years)</b>	
21-30	2 (6.6)
31-40	3 (10)
41-50	6 (20)
51-60	3 (10)
>60	16 (53.3)
<b>SUBSTANCE ABUSE</b>	
Smoking (exclusively)	20 (66.6)
Smoking + tobacco/alcohol	10(33.33)
<b>SMOKING</b>	
<10 yrs	1 (5)
11-20 yrs	7 (35)
>20 yrs	12 (60)
<b>SITE</b>	
Base of tongue	8 (26.6)
Buccal mucosa	7 (23.3)
Epiglottis	2 (6.6)
Gingivobuccal sulcus	1 (3.3)
Oropharynx	4 (13.3)
Larynx	1 (3.3)
Tonsillar fossa	1 (3.3)
Palate	3 (10)
Posterior pharyngeal wall	1 (3.3)
Vocal cord	1 (3.3)
Vallecula	1 (3.3)

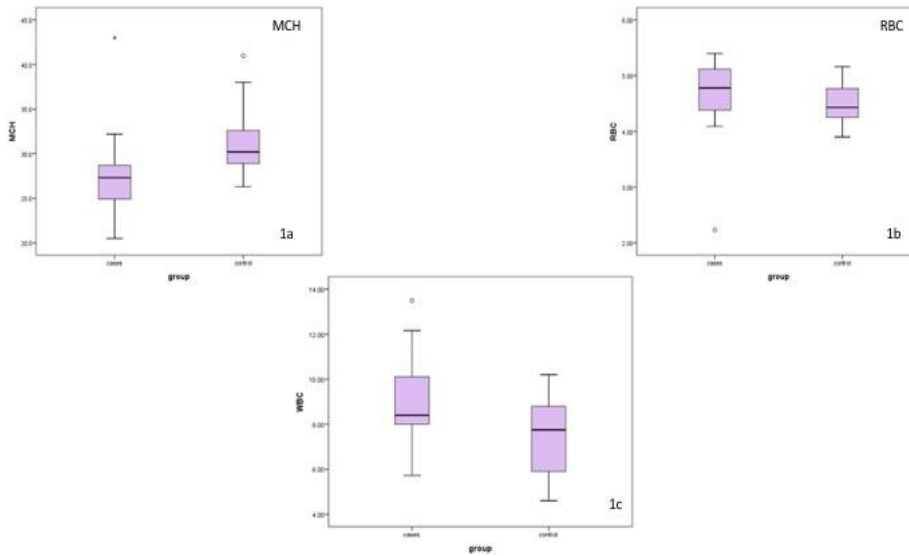
**Table I: Distribution of cases among various clinical parameters**

On comparing the various hematological parameters among oral cancer patients and healthy individuals, significant p values were observed in MCH (0.001), RBC (0.019) and WBC (0.027) (hematological) (Figure 1). Significant values among platelet

parameters were seen in PDW (0.0001) and MPV (0.004) (platelet) and APTT (Figure 2) was the only coagulation parameter with a significant p value of 0.0001. On plotting ROC curves for these parameters (Figure3) APTT (AUC: 0.868, p<0.0001)

demonstrated better predictive power as per other parameters.  
area under curve of ROC as compared to

**Figure 1: Box plot showing significant p values: a) MCH, b) RBC and c) WBC.**



**Figure 2: Box plot showing significant p values: a) PDW b) MPV and c) APTT.**

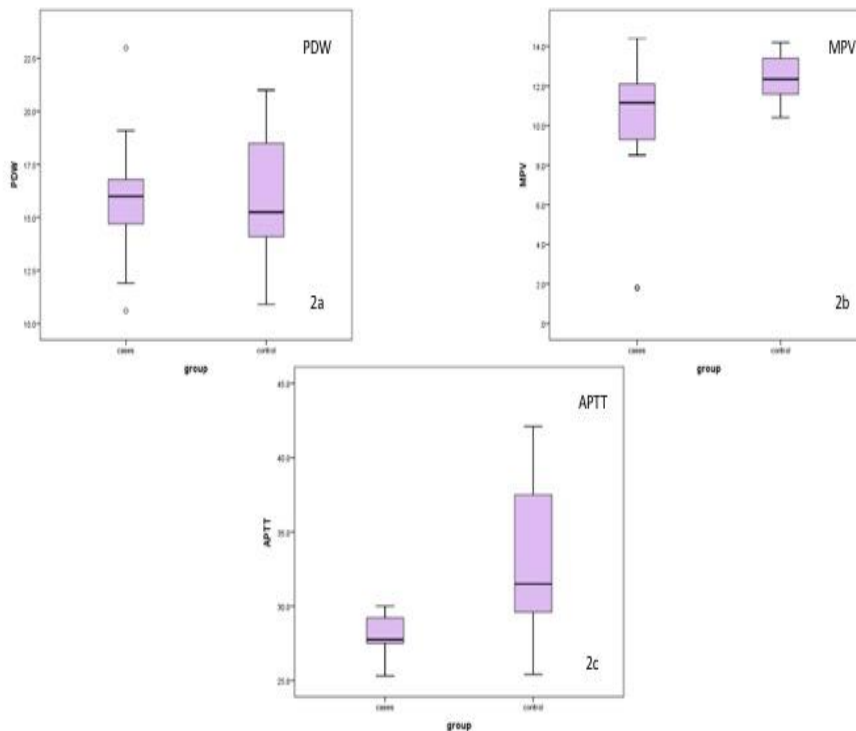
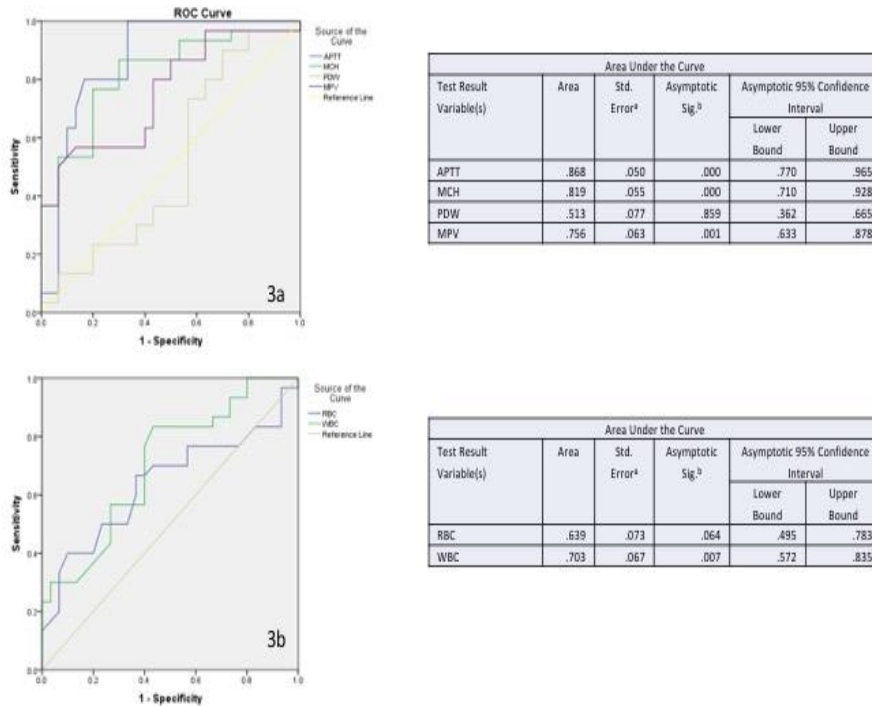


Figure 3: ROC curve showing better predictive power as per significant area under curve of ROC, a) APTT (AUC: 0.868) and b) WBC (AUC: 0.639)



On the other hand, there wasn't any statistically significant difference among normal and oral cancer patients when it came to the rest of the hematological,

platelet and coagulation parameters such as Hg, HCT, MCV, MCHC, and platelet count, PCT, PTT and INR (Table II).

PARAMETER	TEST (n=30)	CONTROL (n=30)	p VALUE
MCH (pg)	27.18±4.57	30.86±3.97	0.001
RBC (10 <sup>6</sup> /microliter)	4.79±0.63	4.43±0.52	0.019
WBC (10 <sup>3</sup> /microliter)	8.56±2.39	7.36±1.65	0.027
Hg (g/dl)	13.17±1.81	12.79±0.80	0.297
HCT (g/dl)	40.85±5.54	38.38±2.40	0.028
MCV (fl)	86.39±12.55	91.40±5.74	0.051
MCHC (g/dl)	32.87±5.15	31.94±2.07	0.362
Platelet count (lacs/mm <sup>3</sup> )	2.39±7.98	2.18±8.97	0.923
PDW (fl)	18.72±3.82	14.82±3.51	0.0001
MPV (fl)	10.5±3.38	12.42±1.20	0.004
PCT (%)	0.28±0.08	0.34±0.29	0.279
APTT	28.48±1.06	32.83±4.62	0.0001
PTT	13.85±0.79	13.96±1.48	0.720
INR	1.12±0.17	1.07±0.09	0.159

Table II: Comparison of hematological, platelet and coagulation parameters between oral cancer patients and healthy individuals (test vs control)

On classification of oral cancers (using Broder's classification for squamous cell carcinoma), 56.67% cases were moderately differentiated, followed by well differentiated (36.67%) and 6.67% cases were poorly differentiated. Additionally, morphological distribution of oral cancers

was assessed, RBC was the only parameter to have a significant p value (0.009) on studying all the haematological, platelet and coagulation parameters among well, moderately and poorly differentiated oral cancers. (Table III)

PARAMETER	WELL DIFFERENTIATED (n=11)	MODERATELY DIFFERENTIATED (n=17)	POORLY DIFFERENTIATED (n=2)	p VALUE
MCH (pg)	26.09±3.96	27.57±5.02	29.5±3.25	0.543
RBC (10 <sup>6</sup> /microliter)	4.92±0.41	4.77±0.70	3.37±0.71	0.009
WBC (10 <sup>3</sup> /microliter)	7.80±2.51	9.08±2.37	7.82±0.32	0.364
Hg (g/dl)	13.53±1.50	13.07±2.05	12.1±0.28	0.564
HCT (g/dl)	42.50±3.77	40.45±6.30	35.5±2.47	0.230
MCV (fl)	86.50±6.69	86.59±14.76	83.85±22.41	0.959
MCHC (g/dl)	31.67±1.13	33.75±6.56	31.1±0.28	0.516
Platelet count (lacs/mm <sup>3</sup> )	2.48±6.54	2.26±5.30	2.53±1.13	0.994
PDW (fl)	15.37±5.22	15.78±2.99	17.1±1.55	0.845
MPV (fl)	11.44±1.98	9.68±4.01	12.4±0.56	0.291
PCT (%)	0.27±0.05	0.27±0.10	0.31±0.12	0.818
APTT	28.75±0.82	28.37±1.22	28±0.14	0.530
PTT	13.79±0.83	13.91±0.82	13.7±0.14	0.113
INR	1.13±0.20	1.12±0.16	1.07±0.01	0.903

**Table III: Hematological parameters**

## DISCUSSION

The progression of carcinoma and circulating platelets in venous blood are closely associated with each other which establish the role of platelets besides the usual hemostatic function.<sup>7,8</sup> Platelets perform a variety of functions per se act as a mediator of angiogenesis, wound healing & immune modulation. They secrete cytokines & growth factors such as TGF β, VEGF, MMP-2, PF-4 & PDGF. (9-12) Epithelial, mesenchymal transition, angiogenesis, cell migration & proliferation are considered to be the most significant factors determining carcinoma progression.<sup>13</sup> Thrombopoiesis is stimulated by the actions of soluble mediators IL-6 and GM-CSF which are a product of platelet derived TGF β, a promoter of carcinoma metastasis. (14-17)

The present study investigated an association between platelet parameters as well as coagulation and haematological parameters between test and control groups along with their relationship with the clinicopathological framework (in respect to site of the tumour and history of smoking).

We found out men to be affected more than women (76.6%). This finding was in concordance with Khatib et al<sup>18</sup> (63.55%) and also Park et al<sup>19</sup> (70%). Maximum cases belonged to the age group >60 years.

The results were similar to Park et al<sup>19</sup> (median age 66 years). One of the most common etiological factors for oral cancer is substance abuse such as smoking (bidi/cigarette), vaping, tobacco/areca nut/betel nut chewing and alcohol consumption. The present study included oral cancer patients with history of smoking, tobacco chewing and alcohol consumption. The most common substance abuse in the said study was exclusive smoking (sans any other associated substance abuse) which was found in 66.6% patients of oral cancer, these results were comparable with Madani et al (72%).<sup>20</sup> Duration of smoking/number of pack years and oral cancer are directly proportional to each other. Duration of smoking >20 years (60%) was observed in the study which was in sync with Nirmala CJ et al.<sup>21</sup> Base of tongue (26.6%) was the most frequently affected site in patients of oral cancer which was similar to Dhanuthai K et al (25.4%).<sup>22</sup>

Anaemia in an untreated cancer patient occurs due to a variety of reasons, related or unrelated to the malignancy, such as defective marrow function due to impaired production of erythropoietin, tumor encroachment, myelofibrosis or marrow necrosis, low iron availability due to nutritional deficiency or enhanced retention

of iron in the reticuloendothelial system, increased RBC loss due to haemorrhage or haemolysis, or as a paraneoplastic syndrome.<sup>23</sup>

On comparison of haematological parameters between case and control groups, MCH and RBC turned out to be of significant p values (0.001 and 0.019 respectively) which was in conjunction with Bhattathiri et al.<sup>24</sup> Additionally, WBC count also showed a difference between mean values among test and control groups ( $8.56\pm 2.39$  and  $7.36\pm 1.65$  respectively), this was in concordance with Shankar et al.<sup>25</sup>, who could associate oral cancer related mortality with high WBC counts. Recent studies have provided evidence that platelet activation is clinically significant in some malignant tumors. In particular, in a series of processes in which cancer cells disseminate through blood circulation, platelets play an important role for cancer cells to aggregate and be discharged from blood vessels and spread to other organs.<sup>26</sup> In the current study out of the platelet parameters, PDW ( $18.72\pm 3.82$  vs  $14.82\pm 3.51$ , 0.0001) and MPV ( $10.5\pm 3.38$  vs  $12.42\pm 1.20$ , 0.004) showed significant p values and differences in mean between test and control groups respectively. Strong association of raised PDW and MPV was seen even in association with oral cancer prognosis and elderly patients along with a usual relation with frequency of oral cancers by Hirahara et al.<sup>27</sup>, Park et al.<sup>28</sup> and Tham T et al.<sup>29</sup> However this was different from a study conducted by Kannar V et al.<sup>30</sup> who witnessed equal results between test and control groups for PDW. APTT, in the present study also turned out to reveal strongly significant p value (0.0001) and differences in mean between test ( $28.48\pm 1.06$ ) and control ( $32.83\pm 4.62$ ) groups. Vylliotis A et al.<sup>31</sup> studied effect of thrombosis-related gene polymorphisms upon oral cancer however to the best of our knowledge ours is the first study establishing a correlation between APTT and oral cancer. On calculating mean and p value for differences in PDW (0.66-p value)

and MPV (0.36-p value) values between oral cancers occurring at base of tongue vs elsewhere was obtained, which was not significant. Additionally, the current study also evaluated the difference between these parameters between the morphological subtypes (well, moderately and poorly differentiated carcinoma, Broder's classification system)<sup>32</sup> which showed a significant p value (0.009) only for RBC count.

## CONCLUSION

To summarise, the present study emphasizes on the evaluation of routine platelet, haematological and coagulation parameters in determining frequency, progression and furthermore prognosis of oral cancer using these rapid and cost-effective tests. Statistically significant differences were observed in MCH, RBC, WBC, PDW, MPV and APTT between cases and controls. Data generated from the present study would provide a baseline data and would serve as a reference for any future study of similar nature. Multi-institutional studies on a larger sample size are required to establish the role of these novel parameters and determine the cut-off values for diagnosis.

## Contributor's Statement:

- Idea & design:** Dr Varsha Chauhan, Dr Charu Agarwal, Dr Mukta Pujani, Dr RK Chandoke
- Data acquisition:** Dr Varsha Chauhan, Kunal Jain, Dr Kanika Singh, Dr Mitasha Singh
- Analysis:** Dr Charu Agarwal, Dr Mukta Pujani, Dr Kanika Singh
- Interpretation of findings:** Dr Varsha Chauhan, Dr Charu Agarwal, Dr Kanika Singh, Dr Mitasha Singh
- Preparation of manuscript:** Dr Charu Agarwal, Dr Mukta Pujani, Dr Varsha Chauhan, Kunal Jain
- Critical revision:** Dr Mukta Pujani, Dr Kanika Singh, Dr RK Chandoke

## Declaration by Authors

**Ethical Approval:** Approved

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**Conflict of Interest:** There is no conflict of interest

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