Journal of Clinical & Experimental Pathology

Bere et al. J Clin Exp Pathol 2021, 11:4

Research Article Open Access

Relationship between Pre-Intervention Plasma Fibrinogen Levels and Clinico-Histologic Features in Patients with Oral Squamous Cell Carcinoma

Silas K Bere[1*], **Onyango JF**[2], **Njiru AW**[2] **and Dimba E**[2]

[1]*Department of Dentistry, University of Zimbabwe, College of Health Sciences Parirenyatwa Group of Hospitals, Mazowe Street, Belgravia, P.O Box A178 Avondale Harare, Zimbabwe*

[2]*Department of Oral and Maxillofacial Surgery/Oral Pathology/Oral Medicine, School of Dental Sciences, University of Nairobi, P.O Box 19676-00202, Nairobi, Kenya*

[*]**Corresponding author:** Silas K Bere, Department of Dentistry, University of Zimbabwe, College of Health Sciences Parirenyatwa Group of Hospitals, Mazowe Street, Belgravia, P.O Box A178 Avondale Harare, Zimbabwe, E-mail: silasbere@students.uonbi.ac.ke

Received date: July 08, 2021; **Accepted date:** July 22, 2021; **Published date:** July 29, 2021

Abstract

Objectives: The purpose of this study was to evaluate the relationship between pre-treatment plasma fibrinogen levels and common prognostic clinico-pathological parameters in patients presenting with OSCC.

Materials and Methods: Thirty-one participants were recruited at presentation to Kenyatta National and University of Nairobi Dental Hospitals. All patients had detailed medical history, thorough clinical examination, radiographs, and histopathological examination of their biopsies. Socio-demographic and Clinico-pathologic parameters were recorded. Plasma fibrinogen concentration was determined on a blood sample from the participants by the prothrombin-time- derived fibrinogen (FIB_{PT}) assay.

Results: Most of the patients were above the age of 40 years and an almost 1:1 male to female ratio. The mean pre-intervention plasma fibrinogen level was 543.7 mg/dl, with a range of 299 mg/dl to 911 mg/dl. Thirty patients (96.8%) had elevated pre-intervention plasma fibrinogen levels above the upper reference level of 362 mg/dl. There was a statistically significant higher mean plasma fibrinogen level among female patients than the male (CI, p=0.017). The difference in plasma fibrinogen levels among patients above and those below 50 years was not statistically significant. There was a general up-regulation of plasma fibrinogen levels in the more severe disease status. The difference in fibrinogen levels in the more severe disease and less severe disease status was not statistically significant in this study.

Conclusion: Consistent with other studies, it was shown that plasma fibrinogen levels were up- regulated in the OSCC. Further studies are necessary to determine the significance of elevated pre- intervention plasma fibrinogen levels in the prognostication of the disease.

Keywords: Computed tomography; Chi-squared test; Mann-Whitney U test; Pathology

Introduction

Hemostatic alterations are frequently seen in patients with cancer, the degree of which has been found to correlate with the clinical progression of the disease [1]. Tumour cells produce tissue factor, cancer procoagulant, plasminogen activators and other factors that interact with the coagulation system, the fibrinolytic system and vascular or blood cells such that they disrupt the normal homeostasis and balance between activation and inhibition of the coagulation and fibrinolytic systems [2-4]. Rather than serving as a mere trigger for increased thromboembolic events, cancer-induced hemostatic activity has also been shown to promote tumour growth and cancer cell dissemination [5,6]. Indeed, both coagulation assays and high levels of circulating biomarkers indicative of coagulation and fibrinolysis activation have been associated with decreased survival in several tumour types [7-12]. One of the biomarkers for this up-regulation is plasma fibrinogen. Several studies have associated elevated plasma fibrinogen levels with tumour progression, invasiveness, metastasis, and poor prognosis, suggesting that plasma fibrinogen concentration could be used as a risk indicator for some tumors [13-19]. Moreover, other studies have also associated plasma fibrinogen levels with tumour response to therapy indicating that plasma fibrinogen levels could be used as a predictor for tumour response to therapy for some malignancies [20-22]. However, the relationship between plasma fibrinogen levels and prognosis in oral cancer remains uncertain [11]. Furthermore, there are hardly any similar studies in the Kenyan population. The purpose of this study was to evaluate the relationship between pre-treatment plasma fibrinogen levels and common prognostic clinico- pathologic parameters in patients presenting with OSCC at two tertiary teaching hospitals in Nairobi.

Materials and Methods

In this study 31 participants (15 men and 16 women) were recruited consecutively from among oral cancer patients presenting at the Kenyatta National and University of Nairobi Dental Hospitals over a one-year period. A detailed medical history, thorough clinical examination, chest X-ray, Computed Tomography (CT) of the head and neck and histopathological examination of OSCC biopsies were conducted for all patients. The following details of clinico-pathological parameters were also recorded: age, sex, history of smoking, tumour site and size, nodal status, distant metastasis, tumour stage and differentiation. Patients suffering from acute infectious diseases were

excluded from the study due to the acute phase response characteristics of fibrinogen.

Plasma fibrinogen measurement

Five milliliters of peripheral venous blood for pre-operative fibrinogen analysis were collected in Vacutainer tubes (Becton Dickson, Rutherford, NJ, USA) containing 3.8% sodium citrate. The plasma fibrinogen concentration was determined by the prothrombin-time-derived fibrinogen (FIBPT) assay using an automated photometric coagulation analyzer; ACL Elite-Pro® and thromboplastin reagent; HemosILRecombiPlasTin (Instrumentation Laboratories), according to the manufacturer's instruction [23,24]. HemosILRecombiPlasTin contains thromboplastin that is a liposomal preparation of recombinant human tissue factor in a synthetic phospholipid blend, calcium chloride buffer and a preservative. The addition of this reagent to the test plasma initiates the activation of the extrinsic coagulation pathway which ultimately results in the conversion of fibrinogen to fibrin.

This method is not a direct determination of plasma fibrinogen but an estimation of the fibrinogen concentration from the absorbance changes at 450 nm (delta OD) during the clotting process of the PT on an automated photo optical coagulometers [23-26]. To perform the FIBPT assay, the analyzer measures the total change in sample absorbance at 671 nm from the endpoint of the prothrombin reaction. The total change in optical density is directly proportional to the fibrinogen content of the sample [26]. The reference range was set between 242 mg/dl and 362 mg/dl. Plasma fibrinogen levels higher than 362 mg/dl indicated hyperfibrinogenemia.

Statistical analysis

The continuous data were expressed as the mean ± standard deviation and the categorical data were expressed as percentages (%). Continuous variables were compared using unpaired t-test and one way analysis of variance. Categorical variables were compared using chi-squared test, Mann-Whitney U test, Fishers exact test or Kruskal-Wallis test where appropriate. A p-value<0.005 was considered as having been statistically significant in all statistical analyses. SPSS for Windows version 9.0 computer program (SPSS Inc., Chicago, Illinois, USA) was used for statistical analysis.

Results

Baseline patient characteristics

The socio-demographic and clinico-pathologic parameters evaluated, including sex, age, tumour site, tumour size, histological grade, tumour stage and the pretreatment plasma fibrinogen level are shown in Table 1. There were 31 patients among whom 16 were female and 15 were male with a median age of 56.7 years (range: 28-77 years, mean: 56.5 years). The most frequently involved anatomical sites were the tongue (excluding the base) 10 (32%) and gingivae 8 (26%). The other sites accounted for the remaining 13 cases (48%) which were evenly distributed. Out of the 31 patients 24 (77.4%) had stage 4 disease, 1 (3.2%) had stage 3 disease and 6 (19.4%) had stage 2 disease. Twenty-two (75.9%) patients had well differentiated (grade 1) tumors, seven (24.1%) had moderately differentiated (grade 2) tumors and two did not have histological grading available.

Clinico-pathological Variable		N %	Plasma fibrinogen levels (mg/dl)				P-value
			<362	>362	Mean	SD(+/-)	
Age							0.34
	<50	8 (25.8%)	1	7	496.7	145	
	>50	23(74.2%)		23	557.5	145.8	
Gender							0.017
	M	15(48%)	1	14	480.7	116.4	
	F	16(52%)		16	602.9	148.2	
Site							0.587
	Tongue	10(32.2%)		10	530.6	148.7	
	Gum	8(25.8%)		8	581.4	180.3	
	Floor of mouth	3(9.7%)		3	634	87.2	
	Palate	3(9.7%)	1	2	424	77.2	
	Multiple site	4(12.9%)		4	548	175.4	
	Other sites	3(9.7%)		3	511	50.1	
Size							0.411
	X ≤ 20 mm	4(13%)	1	3	452.3	84.6	

	20 mm<X ≤ 40 mm	9(29%)		9	550.7	189.5	
	X>40 mm18(58%)			18	560.6	130	
Grade							0.239
	Well differentiated	22(75.9%)	1	21	535.9	139	
	Mod. differentiated	7(24.1%)		7	611	158.3	
Stage							0.25
	Early Disease (1 and 2)	6(19.4%)	1	5	481.2	211.7	
	Late disease (3 and 4)	25(80.6%)		25	560	128.7	

Table 1: Relationship between Pre-intervention plasma fibrinogen levels and socio-demographic/Clinico-pathologic parameters.

The mean plasma fibrinogen was 543.7 mg/dl (+/- 145.5), with a range from 299 mg/dl to 911 mg/dl. Out of 31 patients 30 (96.8%) had elevated plasma fibrinogen levels above the reference range (>362 mg/dl) and only one (3.2%) patient had a plasma fibrinogen level of 299 mg/dl. The relation between plasma fibrinogen concentration and clinic-pathologic variables is summarized in the table below. Except for gender, there was no significant association between plasma fibrinogen concentration and clinico-pathologic characteristics including age, sites, sizes, stage, or grade.

Discussion

Fibrinogen, a soluble 340-kDa glycoprotein synthesized in the liver is transformed through the activity of activated thrombin into insoluble fibrin which is a key coagulation factor in platelet aggregation, clot formation, wound healing and coagulation [27-29]. Several studies have shown that plasma fibrinogen levels are up-regulated in various cancer types including head and neck cancer [30,31] and may indicate cancer progression, metastasis, and recurrence [32-36].

The pathogenesis of this coagulation impairment is complex and involves general risk factors which are common to all categories of patients such as advanced age, prolonged immobility, a prior history of thrombosis, high leukocyte and platelet counts, obesity, immobility, and co- morbid conditions; and other cancer-specific factors such as tumour type, grade, and disease stage. It is the relationship between pretreatment plasma fibrinogen levels and common prognostic clinic-pathological parameters in patients presenting with OSCC cell carcinoma that was the subject of this investigation.

The baseline socio-demographic and clinico-pathologic characteristics of our patients were typical of this population [37] with most of the patients aged over 40 years and an almost 1:1 male to female ratio. The plasma fibrinogen levels were up regulated in 97% of the patients. This differs markedly from a similar study [33] in which 61% of patients presented with low serum fibrinogen concentration and 31% presented with high serum fibrinogen concentration. Our study also confirmed the effect of age and gender on serum fibrinogen levels [38]. Younger patients tended to have lower serum fibrinogen levels compared to older patients and mean plasma fibrinogen levels between males and females clearly demonstrated a gender bias. A few studies have shown that the female gender, age, body mass index (BMI) and alcohol abstinence are directly associated with fibrinogen levels [38]. Most importantly, our results support the findings in other studies that have shown a positive correlation between serum fibrinogen concentration and cancer prognostic factors [11,39-41]. Although statistical significance could not be demonstrated the mean serum fibrinogen levels were higher in the larger tumors, the higher grades, and the more advanced disease.

Conclusion

Consistent with other studies, this study has shown that plasma fibrinogen levels are up regulated in OSCC. Furthermore, the levels of plasma fibrinogen were higher in the more severe diseases indicating that plasma fibrinogen levels could serve as an independent biomarker for oral cancer prognosis. However, further studies with larger samples are required to confirm this.

Acknowledgement and Disclosure Statement

Our sincere thanks go to the administrative organs of the Kenyatta National Hospital and School of Dental Sciences, University of Nairobi, for the support accorded in the conduct of this work. This study was approved by The Kenyatta National Hospital and The University of Nairobi, Ethics and Research Committee (KNH/UoN-ERC: P70/02/2015).

Declaration of Interest

Authors have no financial or any other interest in the publication of this work.

Authors Contribution

Dr. SK Bere: Designed and carried out clinical and laboratory research.

Prof. JF Onyango: Supervised the research and edited the final report.

Dr. AW Njiru: Assisted in the supervision of the clinical research.

Dr. EO Dimba: Assisted in the supervision of the laboratory research.

References

1. Zacharski LR, Wojtukiewicz Mz, Costantini V (1992) Pathways of coagulation fibrinolysis activation in malignancy. Semin Thromb Hemost 18: 104-116.
2. Falanga A, Marchetti M, Vignoli A (2003) Clotting mechanisms, and cancer: Implications in thrombus formation and tumor progression. Clin Adv Hematol Oncol 1: 673-678.
3. Rickles FR, Falanga A (2009) Activation of clotting factors in cancer. Cancer Treat Res 148: 31-41.
4. Falanga A, Russo L (2012) Epidemiology, risk, and outcomes of venous thromboembolism in cancer. Hamostaseologie 32: 115-25.
5. Gale AJ, Gordon SG (2001) Update on tumor cell procoagulant factors. Acta Haematol 106: 25-32.
6. Amirkhosravi A, Meyer T, Amaya M (2007) The role of tissue factor pathway inhibitor in tumor growth and metastasis. Semin Thromb Hemost 33: 643-652.
7. Taguchi O, Gabazza EC, Yasui H (1997) Prognostic significance of plasma D-dimer levels in patients with lung cancer. Thorax 52: 563-565.
8. Blackwell K, Haroon Z, Broadwater G (2000) Plasma D-dimer levels in operable breast cancer patients correlate with clinical stage and axillary lymph node status. J Clin Oncol 18: 600-608.
9. Oya M, Akiyama Y, Okuyama T (2001) High preoperative plasma D dimer level is associated with advanced tumor stage and short survival after curative resection in patients with colorectal cancer. Jpn J Clin Oncol 31: 388-394.
10. Sampson MT, Kakkar AK (2002) Coagulation proteases and human cancer. Bio Chem Soc Trans 30: 201-207.
11. Peng P, Shen J, Dong JB (2014) Preoperative plasma fibrinogen predicts cervical metastasis in patients with stage I/II carcinoma of the tongue. Int J Oral Maxillofac Surg 43: 393-398.
12. Gharat L, Rathod GP (2013) Quantitative estimation of serum fibrinogen degradation product levels in oral premalignant and malignant lesions. J Int Oral Health 5: 65-72.
13. Takeuchi H, Ikeuchi S, Kitagawa Y (2007) Pretreatment plasma fibrinogen level correlates with tumor progression and metastasis in patients with squamous cell carcinoma of the esophagus. J Gastroenterol Hepatol 22: 2222-2227.
14. Polterauer S, Seebacher V, Hefler-Frischmuth K (2009) Fibrinogen plasma levels are an independent prognostic parameter in patients with cervical cancer. Am J Obstet Gynecol 200: 647.e1-647.e7.
15. Polterauer S, Grimm C, Seebacher V (2009) Plasma fibrinogen levels and prognosis in patients with ovarian cancer: A multicenter study. Oncologist 14: 979-985.
16. Ghezzi F, Cromi A, Siesto G (2010) Prognostic significance of preoperative plasma fibrinogen in endometrial cancer. Gynecol Oncol 119: 309-313.
17. Zhao J, Zhao M, Jin B (2012) Tumor response and survival in patients with advanced non-small-cell lung cancer: the predictive value of chemotherapy-induced changes in fibrinogen. BMC Cancer 12: 330.
18. Son HJ, Park JW, Chang HJ (2013) Preoperative plasma hyperfibrinogenemia is predictive of poor prognosis in patients with non-metastatic colon cancer. Ann Surg Oncol 20: 2908-2913.
19. Zhu JF, Cai L, Zhang XW (2014) High plasma fibrinogen concentration and platelet count unfavorably impact survival in non-small cell lung cancer patients with brain metastases. Chin J Cancer 33: 96-104.
20. Qiu J, Yu Y, Fu Y (2012) Preoperative plasma fibrinogen, platelet count and prognosis in epithelial ovarian cancer. J Obstet Gynaecol Res 38: 651-657.
21. Xiao B, Ma LL, Zhang SD (2011) Correlation between coagulation function, tumor stage and metastasis in patients with renal cell carcinoma: A retrospective study. Chin Med J 124: 1205-1208.
22. Lu KY, Zhu L, Sheng L (2011) Serum fibrinogen level predicts the therapeutic response and prognosis in patients with locally advanced rectal cancer. Hepato gastroenterological 58: 1507-1510.
23. Sobas F, Hanss M, Ffrench P (2002) Human plasma fibrinogen measurement derived from activated partial thromboplastin time clot formation. Blood Coagul Fibrinolysis 13: 61-68.
24. Rossi E, Mondonico P, Lombardi A (1988) Method of determination of functional (clottable) fibrinogen by the new family of ACL coagulometers. Thrombosis Research 52: 453-468.
25. Chitolie A, Mackie IJ, Grant D (1994) Inaccuracy of the "Derived" fibrinogen measurement. Blood Coagul Fibrinolysis 5: 955-957.
26. Lawrie AS, Mc Donald SJ, Purdy G (1998) Prothrombin time derived fibrinogen determination on Sysmex CA6000. J Clin Pathol 51: 462-466.
27. Lawrence SO, Simpson-Haidaris PJ (2004) Regulated de novo biosynthesis of fibrinogen in extrahepatic epithelial cells in response to inflammation. Thromb Haemostas 92: 234-243.
28. Collen D, Tytgat GN, Claeys H (1972) Metabolism and distribution of fibrinogen. I Fibrinogen turnover in physiological conditions in humans. Br J Haematol 22: 681-700.
29. Koenig W (2003) Fibrin (ogen) in cardiovascular disease: an update. Thromb Haemost 89: 601-609.
30. Selzer E, Grah A, Heiduschka G (2016) Pretherapeutic fibrinogen levels are of prognostic significance in locally advanced head and neck cancer. Wien Klin Wochenschr 128: 320-328.
31. Holzinger D, Danilovic I, Seemann R (2016) Prognostic Impact of Pretreatment Plasma Fibrinogen in Patients with Locally Advanced Oral and Oropharyngeal Cancer. PLoS One 11: e0158697.
32. Yamamoto M, Kurokawa Y, Miyazaki Y (2016) Usefulness of Preoperative Plasma Fibrinogen Versus Other Prognostic Markers for Predicting Gastric Cancer Recurrence. World J Surg 40: 1904-1909.
33. Mei Y, Zhao S, Lu X (2016) Clinical and Prognostic Significance of Preoperative Plasma Fibrinogen Levels in Patients with Operable Breast Cancer. PLoS One 11: e0146233.
34. Troppan KT, Melchardt T, Wenzl K (2016) The clinical significance of fibrinogen plasma levels in patients with diffuse large B cell lymphoma. J Clin Pathol 69: 326-330.
35. Zhang D, Zhou X, Bao W (2015) Plasma fibrinogen levels are correlated with postoperative distant metastasis and prognosis in esophageal squamous cell carcinoma. Onco Target 6: 38410-38420.
36. Zhu LR, Li J, Chen P (2016) Clinical significance of plasma fibrinogen and D-dimer in predicting the chemotherapy efficacy and prognosis for small cell lung cancer patients. Clin Transl Oncol 18: 178-188.
37. Muange P, Chindia M, Njiru (2014) Oral Squamous Cell Carcinoma: A 6-Month Clinico- Histopathologic Audit in a Kenyan Population. Czas Stomatol 4: 475- 483.
38. Nascetti S, Elosua R, Pena A (2001) Variables associated with fibrinogen in a population- based study: Interaction between smoking and age on fibrinogen concentration. Eur J Epidemiol 17: 953-958.
39. Perisanidis C, Psyrri A, Cohen EE (2015) Prognostic role of pretreatment plasma fibrinogen in patients with solid tumors: A systematic review and meta-analysis. Cancer Treat Rev 41: 960-970.
40. Tang LQ, Chen QY, Guo SS (2014) The impact of plasma Epstein-Barr virus DNA and fibrinogen on nasopharyngeal carcinoma prognosis: an observational study. Br J Cancer 111: 1102-1111.
41. Jagielska B, Symonides M, Stachurska E (2011) Coagulation disorders in patients with locally advanced head and neck cancer-should they really be disregarded? Neoplasma 58: 129-134.