Elevation of free prostate - specific antigen (FPSA) in cancer patients adenosine deaminase (ADA) Binds and stimulates plasminogen (PG) activation on 1-LN Human prostate cancer cells

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Abstract

The Prostate Specific Antigen (PSA) is the best and most commonly used tumor maker for prostate in clinical practice these days.[1-2] It is a glycoprotein has molecular weight of 33KD. PSA is very routinely used in the diagnosis and follow up of prostate disease, specially prostate cancer.[3-4] Prostate Specific Antigen (PSA) is formed in two different molecular types in serum; they are free PSA (fPSA) and complex PSA (cPSA), the serum of which form the total PSA (tPSA).fPSA is mostly used in the detection of Prostate cancer.[5] Free PSA 28 kda is eliminated by glomerular filtration. The date revealed that the patients with end stage of renal dysfunction prolonged dialysis with increase percent of fPSA,[6-9]

Introduction: Adenosine Deaminase (ADA) is expressed in intracellular by all the cells. It is also associated with the cell surface multifunctional. Glycoprotein CD_{26}/ dipeptidyl peptidase IV. $CD_{26\ is}$ expressed on the surface of human prostate cancer. 1-LN cells acting as a receptor for plasminogen (pg). Since ADA and pg bind CD_{26} of distinct nearby sites. A possible interaction is found between these two proteins on the surface of 1-LN cells. Human ADA binds to CD_{26} on the surface 1-LN cells. Thus PSA, ADA may be factors regulating events in prostate cancer cells that occur when pg binds to the cell surface and in activated in prostate cancer,[10-13]

Materials and Methods: A total number of 201 patients with age group of (Median age 58 years, interquartile age range 47-68) with Chronic Kidney Disease (CKD). Their Glomerular Filtration Rate (GFR) was 24 ml/min/1.7cm^2. (Interquartile range is 18-34) by determined by iohexol clearance method. A separate biochemical and quantitative estimation was done for serum Adenosine Deamine (ADA) and Plasminogen (pg) was done by fluroimmunoessay method and are found elevated as serum Adenosine Deamine (ADA) > 10Iu/L and plasminogen (pg) > 0.2 Iu/L in prostate cancer patients.[14-17] Control included 350 subjects, attended a positive cancer screening test with no diagnostic prostate cancer.

Results: Mediant PSA levels and percent of f PSA were significantly higher ($p<0.001$) in patients with renal dysfunction, 0.45µg/L and 48.2%. Compared to control, 0.28µg/L and 28.8% respectively. Effect of the interaction between Plasminogen (pg) and Adenosine Deamine (ADA) on plasminogen activation or Adenosine Deamine (ADA) enzymatic activity that plasminogen (pg) binds to CD_{26} on1-LN cells[18-20] The present study shows the effect of increasing concentration of ADA on pg activation by 1-LN cells monolayer are single pg concentration 0.2 µM and also on pg activation by t-PSA.

Conclusion: The present study of percent of free Prostate Specific Antigen (fPSA) Adenosine Deamine (ADA) on plasminogen (pg) are significantly influenced by moderately impaired renal function in patients with Chronic Kidney Disease (CKD) and in prostate cancer.

Keywords: Prostate specific antigen (PSA), Adenosine deaminase (ADA), Plasminogen (pg), CD26, 1-LN Cells, Chronic kidney disease (CKD), Glomerular filtration rate (GFR) and prostatic cancer.

Introduction

The estimation of serum prostate specific antigen (PSA), Adenosine Deaminase (ADA) and Plasminogen (pg) are the important clinical methods for the detection and monitoring prostate cancer. PSA is found in two different molecular forms in the blood, free prostate specific antigen (fPSA), and complexed prostate specific antigen (cPSA). Free PSA (fPSA) with a molecular weight of ~28 kDa and Complexed PSA (cPSA) ~90 kDa. Complexed PSA (cPSA) alpha-1-antichymetrypsin, alpha-1-antitrypsin and alpha-2-macroglobulin.The serum of fPSA and cPSA correspond to the conventional immunodetected total PSA (TPSA). Adenosine Deaminase (ADA) is expressed intracellular by all cells. It is also associated with the cell surface multifunctional glycoprotein CD_{26} /dipeptidyl peptidase IV. CD_{26} is expressed on the surface of human prostate cancer 1-LN cells acting as a receptor for plasminogen (pg). Since ADA and pg bind CD_{26} as distinct nearby sites. A possible interaction is found between these two proteins on the surface of 1-LN cells. Thus prostate specific antigen (PSA) Adenosine Deaminase (ADA) and Plasminogen (pg) are the factors regulating events in prostate cancer cells that occurs when Plasminogen (pg) binds to the cell surface and are activated in prostate cancer.

Materials and Methods

Men with chronic kidney disease (CKD) undergone medical examination at the department of urology Princess Esra Hospital a teaching hospital to Deccan College of Medical Sciences, Hyderabad, Telangana State, India. Blood samples for PSA, ADA and plasminogen analysis were collected at the same time as the routine determination of glomerular

filtration rates (GFRs). The blood samples were allowed to clot for 15 mints at room temperature subsequently centrifuged at 3500 rpm for 10 mints and then immediately stored at -70˚C for pending analysis. Measurement of serum creatinine, t –PSA, f-PSA, ADA and plasminogen were carried in all blood samples. Fluroimmunoessay technique was used to measure t-PSA and f-PSA, from which percent of f-PSA was calculated. A quantitative estimation of creatinine was determined by enzymatic method using the enzymes creatinine amidohydrolase and creatinine amidohydrolase. The reference range was 0.5-1.5md/dl. The enzymatic activity of ADA was determined by phosphate buffer pH 7.4 containing 100 mm adenosine. ADA activity was expressed as a 265 nm/min. Binding of ADA to pg attached to CD26 and inhibition of its binding to pg by pg structures modules. Increasing concentration of pg was added to CD26 with a single concentration of ADA. It is suggested that ADA facilitates pg activation. It is only the stimulation of pg activation by ADA. When pg is bound to 1-LN cells increase the concentration of ADA which was added to 1-LN cells.

Results

The characterisation study of patients and controls are present in Table 1. In the current study evaluated PSA levels and percent of f-PSA is 201 men with chronic kidney disease (CKD). The majority of patients had moderated to severe renal insufficiency 96 % has GFR < 59 ml/min/1.73m.^{2} The median GFR was 23ml/min/1.73m.^{2}

There was no significant difference in t-PSA in the study patients compared with controls (median 1.08 ug/L versus 0.95 ug/L ; P=0.15). Both median f-PSA and PSA, how were significantly higher (pc=0.01) in the study patients (0.45 ug/L and 47.8 % respectively) than in the controls (0.29 ug/L and 29.9 % respectively). Among the study patients, 69(68 %) has percent f-PSA > 40 % and 39(39 %) has percent f-PSA>50 %.

Hence there is association between GFR and percent f PSA. Regression analysis showed significant association of lower GFR with higher percent Fpsa (p=0.036). Both pg2 and ADA bind to CD26 and ADA binds directly to pg2, the capacity of pg2 to influence the binding of ADA to CD26, increasing concentration of pg enhance the binding of ADA to the complex CD26-pg2. Stimulation of pg activation by ADA when pg is bound to 1-LN cell or immobilized CD26 .An increase concentration of ADA were added to 1-LN cells pg2 (0.2u) at 37˚C.

Discussion

Total Prostate specific antigen (tPSA), Percent free prostate-specific antigen (fPSA) are frequently measured to discriminate of prostate cancer from begin prostate disorder. It has been confirmed a higher percent fPSA indicating a less risk of cancer. By using a cut off of < 25 %. Prostate cancer detection in men with t-PSA in the range 4-10 ug/L. Diminished renal elimination of fPSA may, therefore effect percent fPSA and an accuracy as a diagnostic marker for prostate cancer. This study shows that men with chronic kidney disease and impaired renal function have significantly higher serum levels of fPSA (median 0.45 ug/L versus 0.29ug/L) and significantly higher percent fPSA (median 45 % versus 30 %) compared to control. As many as 69% of the study patients present with a percent of fPSA > 40 %. In addition to the above study ADA binding influences the function of CD_{26} - 1-LN cells.

The binding region for pg and ADA is CD_{26} are in close proximally. The study suggests a novel role for the complex CD_{26}-ADA. In this centre the levels of CD26 and ADA have been evaluation in prostate cancer. Prostate cancer may be higher levels of the protease plasmin, resulting from higher levels of CD_{26} and ADA which facilitates pg localization and activation on the surface of the cells.

Table 1: Characteristics of the study participants

Characteristic	Study Patients (n=201)	Controls (n =350)	p-value
Age	58 (47,68)	58(55-64)	-
GFR[a](ML/min/ 1.73m^2)	24 (16,33)	-	-
Creatinine (ug/dl)	1.8 (0.5-1.5)	-	-
Total PSA (ug/L)	1.08(0.28,1.87)	0.95(0.01,1.55)	0.15
Free PSA (ug/L)	0.45(0.28,0.76)	0.29(0.20,0.43)	<0.001
Percent free PSA	47.2(37.5,55.1)	29.9(22.4,38.8)	<0.001
Renal Diseases, No.of patients (%)			
Diabetic Nephropathy	25(25%)	-	-
Renal Transplants	20(20%)	-	-
Glomerulonephritis	22(22%)	-	-
RenalDisease (non-specific)	15(15%)	-	-
Nephrosclerosis	16(16%)	-	-
Miscellaneous renal disease	2(2%)	-	-

GFR measured by iohexol clearance. P-values were obtained by linear regression, with adjustment for age.

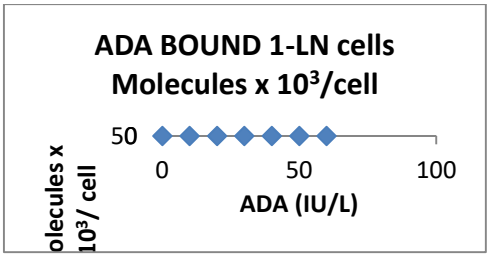

Fig: 1 ADA bound 1-LN cells molecules x 10^3/cell

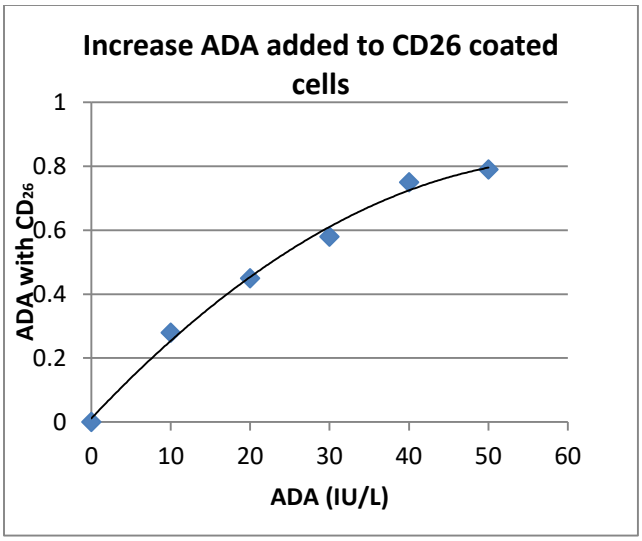

Fig: 2 Increase ADA added to CD26 coated cells

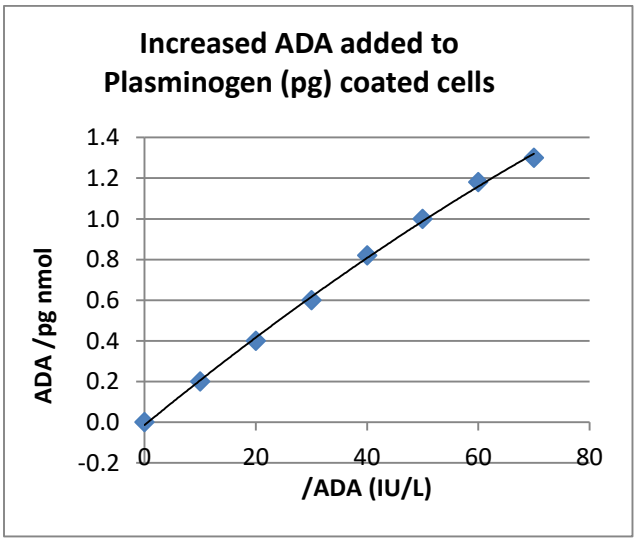

Fig: 3 Increased ADA added to plasminogen (pg) coated cells

Table 2: Adenosine Deaminase, (ADA) CD_{26} (1-LNCells) and Plaminogen (Pg) In patients and controls of prostate cancer

	Patients	Control	P-values
Adenosine Deaminase (ADA),	40±10	8±3	1.7
CD_{26} (1-LN Cells)	0.8±0.2	0.2+0.1	<0.001
Plasminogen (pg),	1.4±0.3	0.2+0.1	<0.001

Source of Funding
None.

Conflict of Interest
None.

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How to cite: Kumar P, Trailokya A Gupta C, Amrit B Karmarkar[2] et al. Elevation of free prostate - specific antigen (FPSA) in cancerpatientsadenosine deaminase (ADA) Binds and stimulates plasminogen (PG) activation on 1-LN Human prostate cancer cells *IP Journal of Urology, Nephrology & Hepatology Science*.2021;4(4):. 109-112