Association of gamma glutamyl transferase with prostate specific antigen levels in patients with prostatic disorders

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Abstract

Introduction: Prostate related diseases like prostatitis, Benign Prostatic Hyperplasia (BPH), prostatic carcinoma are observed most commonly in elderly males. Prostatic specific antigen (PSA) is a common biomarker of prostate related disorders but due to various limitations, new biomarkers are in need for prostate diseases diagnosis. Gamma Glutamyl Transferase (GGT) is also known to be synthesised by the prostate gland. In this study we evaluated the role of GGT as a biomarker of prostatic disorders by observing the association of GGT with PSA.

Materials and Methods: This case control study was conducted at Sri Ramachandra Medical college & Research Institute, Sri Ramachandra Institute of Higher Education and Research, Chennai In this study the case group consisted of 38 males aged between 45-90 years with prostate disorders (prostatitis, BPH, Prostatic cancer). The control group consisted of 187 healthy males of age 45-90 years. Patients with high ALP were excluded to rule out hepatobiliary disease. Relevant patient details and laboratory investigations were collected and statistical analysis done.

Results: The correlation coefficient, r value between PSA and GGT in the control group was −0.076 and in the cases group was -0.049. There was no statistically significant difference between the PSA and GGT levels in both the study groups (p value in control group=0.300; p value in cases group=0.769).

Conclusion: From this study we conclude that GGT cannot be used as a prostate diseases biomarker as there was not much statistical significant difference between the PSA and GGT values among both the controls and cases group.

Keywords: Prostate disorders, Benign prostatic hyperplasia (BPH), Prostatic cancer, Gamma glutamyl transferase, Prostate specific antigen.

Introduction

Prostatic disorders are a common problem in elderly males. BPH (Benign Prostatic Hyperplasia) is a common benign disease of the prostate in elderly males. BPH presents with symptoms of difficulty in urination like urgency, dribbling of urine. About 50% of males above 60 years and 80% males above 80 years of age have BPH.[3] Among the various cancers observed in men, prostatic cancer is very common. It has been found out as the second most common cause of death in males with cancers in the United States.[1] Various diagnostic modalities exist for diagnosing prostatic disorders which includes radiological imaging like Computed Tomography (CT), Magnetic Resonance Imaging (MRI), bone scan and common biomarkers like Prostatic Specific Antigen (PSA).[2]

Prostate specific antigen (PSA) is a serine protease, synthesised by the prostatic epithelium and hence elevated in prostatic disorders. It was considered as the best tumour marker to diagnose prostatic cancers though it has some limitations. It can be synthesised by other normal and tumour cells also.[3]

Gamma glutamyl transferase (GGT) is synthesised by liver, kidneys and pancreas. It is elevated in hepatobiliary diseases.[4] It was generally used as a marker of alcoholism and indicator of liver damage.[3] It is now suggested as an oxidative stress marker as it plays a role in glutathione resynthesis by enhancing cysteine availability. It is proinflammatory, catalyses LDL oxidation and has been observed in atheromas.[5]

GGT is also synthesised by the prostate and its levels have been suggested to correlate with prostatic disorders like prostate cancer.[3]

Aim of the study was to observe the correlation of PSA with GGT levels and to assess the statistical significance of the correlation. This study aims to evaluate if GGT can be used as a biomarker for prostatic diseases.

Materials and Methods

This is a single centre, case control study

Study Population

The study population consisted of males aged between 45-90 years attending Sri Ramachandra Institute of Higher Education & Research, Chennai. The study population included two groups, a case group with high PSA levels and a control group with normal PSA levels.

Case Group

Inclusion Criteria

38 males aged 45-90 years attending Urology OPD at Sri Ramachandra Institute of Higher Education & Research with prostatic disorders such as prostatitis, benign prostatic hyperplasia (BPH) and prostatic cancer were selected.

Exclusion Criteria

Patients who underwent surgical procedures like TURP

(Trans Urethral Resection of Prostate), radical prostatectomy, rectal biopsy, alcoholics and known hepatobiliary diseased patients were excluded from the study.

Control Group

Inclusion Criteria
187 apparently healthy males aged 45-90 years attending master health check up at Sri Ramachandra Institute of Higher Education & Research, Chennai were selected.

Exclusion Criteria
Alcoholics and known hepatobiliary diseased patients were excluded from the study.

Period of the Study
The study was conducted over a period of 6 months from april 2018 till september 2018.

Materials and Methods
After Institutional Ethical approval and consent, history and laboratory data of serum PSA, GGT were taken from all patients under the study. The PSA normal cut-off used was <4 ng/ml. The normal cut-off for GGT was < 55 U/L.

Serum ALP levels were also taken to rule out hepatobiliary disease. So patients with high ALP (>120 U/L) were excluded from the study so that increase in GGT levels are not due to hepatobiliary disease. Alcoholics were excluded from the study, by asking the history of alcoholism from the patient. History of any recent surgical procedures like TURP, radical prostatectomy, rectal biopsy was also asked. The levels of PSA and GGT obtained were correlated for both the groups.

Serum PSA levels were done by a two-site immune enzymatic (sandwich) assay in Beckman Coulter DXI 800 analyser. Serum GGT levels were estimated by an enzymatic method-gamma glutamyl-3-carboxy-4-nitroanilide as a substrate and the gamma glutamyl group from the substrate is transferred by GGT forming the products L-γ-glutamylglycylglycine and 5-amino 2-nitrobenzoate.

Serum ALP levels were estimated by the pNPP (p-nitrophenyl phosphate) method. Both GGT and ALP were done with Beckman coulter AU 5800/680 analysers. All the analyses were done at the Clinical Biochemistry Lab, Sri Ramachandra Laboratory Services at Sri Ramachandra Institute of Higher Education and Research.

Statistical Analysis
The statistical analysis was done using SPSS software 20.0. Student t test and Pearsons correlation were done. A p-value < 0.05 was considered statistically significant.
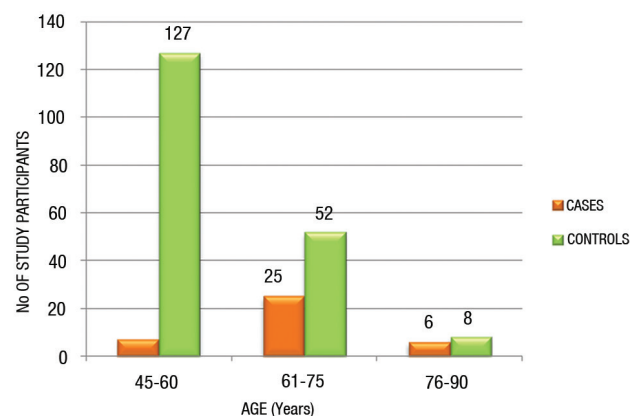
Results
Baseline Characteristics

Age Distribution
Among the 38 cases, 7 were in the 45-60 years age group, 25 were in the 61-75 years age group and 6 were in the 76-90 years age group. Among the 187 controls, 127 were in the 45-60 years age group, 52 were in the 61-75 years, 8 were in
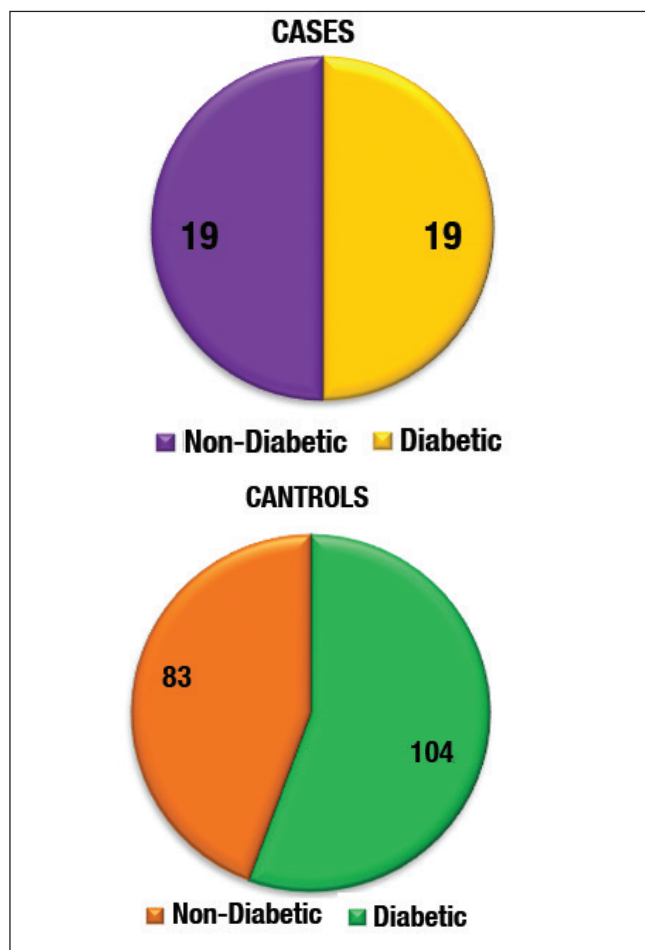
the 76-90 years age group.

Diabetics

Fig. 1: Cases and controls distribution across various age groups

In this study, 19 individuals had diabetes in the case group and 104 individuals had diabetes in the control group.

The mean age was 67.5 ± 8.4 years in the case group and 60.0 ± 38.8 years in the control group. The p value was 0.237

Fig. 2: Distribution of individuals with diabetes among cases and controls

Table 1: Mean and Standard deviation of Age, PSA, GGT, ALP in case and control group

Mean ±SD	Age (Years)	PSA (ng/ml)	GGT (U/L)	ALP (U/L)
Cases (n=38)	67.5± 8.4	11.0 ±12.5	35.4± 21.9	73.6 ± 21.2
Controls (n=187)	60.0 ± 38.8	0.94 ± 0.72	27.0 ± 12.14	82.0 ± 18.6
p value	0.237	0.0001	0.0011	0.014

Table 2: Association of age with PSA and GGT

| Age	PSA (ng/ml)		GGT (U/L)	
	Controls (n=187)	Cases (n=38)	Controls (n=187)	Cases (n=38)
r value	0.164	0.010	− 0.042	− 0.046
p value	0.025	0.952	0.564	0.784

which was statistically not significant. The mean PSA levels were 11.0 ± 12.5 ng/ml in the case group and 0.94 ± 0.72 ng/ml in the control group. The p value was 0.0001 which was statistically significant. The mean GGT levels were 35.4 ± 21.9 U/L in the case group and 27.0 ± 12.14 U/L in the control group. GGT levels were significantly different between the cases and controls (p value-0.0011).

The mean ALP levels were 73.6 ± 21.2 U/L in the case group and 82.0 ± 18.6 U/L in the control group. The p value was 0.014 which was statistically significant. The ALP values in the case group was lesser than in the control group, but within the biological reference interval. Even though p value is significant between the groups, it can be ignored since

Table 3: Association of PSA with GGT levels

PSA (ng/ml)	GGT (U/L)	
	Controls (n=187)	Cases (n=38)
r value	−0.076	−0.049
p value	0.300	0.769

participants who had ALP limits within biological reference interval alone were included in the study. Mean value in the case group is lesser probably due to smaller sample size compared to control group.
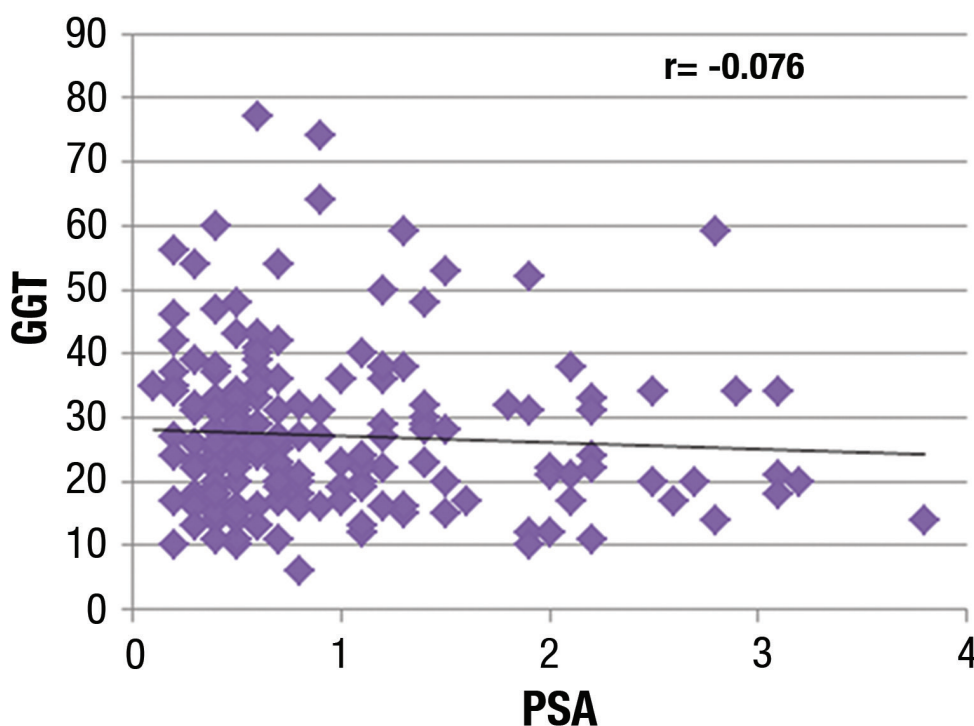
Fig. 3: Correlation between serum PSA and GGT in control group
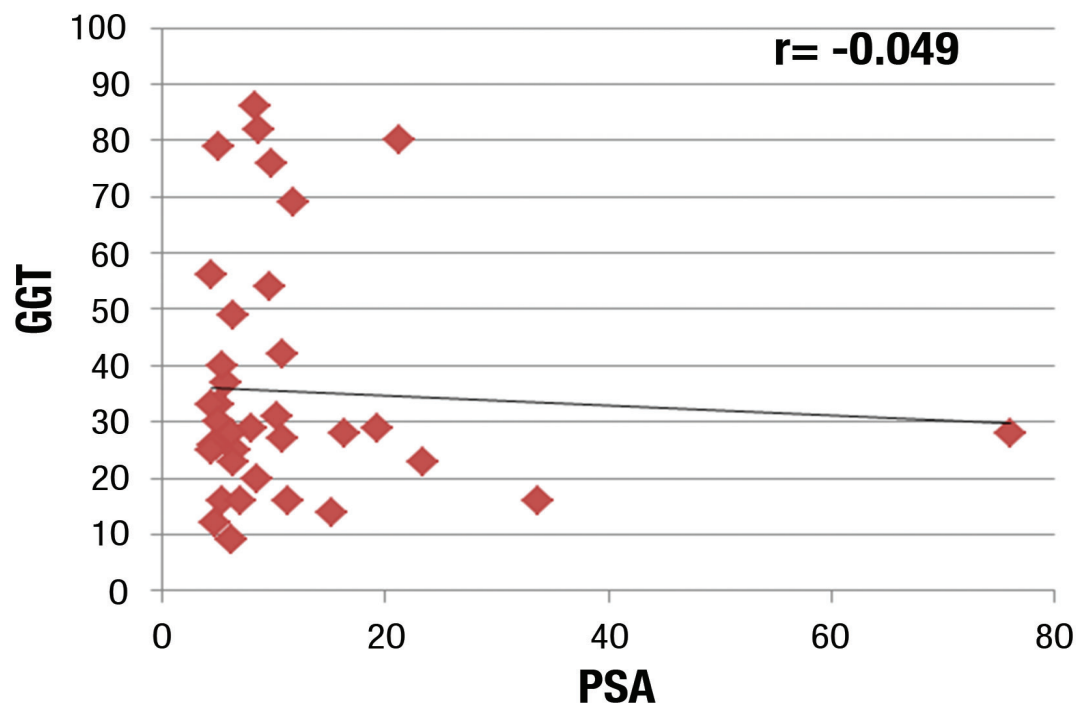
Fig. 4: Correlation between serum PSA and GGT in case group

The correlation coefficient, r value between age and PSA in the control group was 0.164 and 0.010 in the case group. The p-value for significance between age and PSA was 0.025 in the controls and 0.952 in the cases. There was no significant correlation and statistical significance between age and PSA values in both the study groups.

The r value between age and GGT was -0.042 in the controls and -0.046 in the cases. The p-value for significance between age and GGT was 0.564 in the controls and 0.784 in the cases. There was negative correlation and no statistical significance between age and GGT values in both the study groups.

The correlation coefficient, r value between PSA and GGT in the control group was -0.076 and the p value was 0.300. The correlation coefficient, r value between PSA and GGT in the control group was -0.049 and the p value was 0.769. There was negative correlation and no statistical significance between PSA and GGT values in both the study groups.

Discussion

Prostate diseases are a common problem in males especially BPH (Benign Prostatic Hyperplasia) and prostatic cancer. Other conditions like prostatitis, prostatic infections are observed less commonly in males. Prostate Specific Antigen (PSA) belongs to the human kallikrein family. It is a 33 kDa glycoprotein, a serine protease. It is synthesised by epithelial cells of the prostate and the periurethral glands. PSA exists in free and ligand bound forms. Main physiological role is to help in spermatozoa release. It is by seminal coagulum liquefaction. After 1980's PSA widely replaced Prostatic acid phosphatase as a tumour marker for prostate diseases.

PSA has limitations in diagnosis as a tumour marker due to its poor specificity. Though various modifications of PSA like age-adjusted PSA, PSA velocity, PSA density have been used, new biomarkers are needed for screening and diagnosis of prostatic diseases especially for differentiating benign conditions from malignancy.[6,7]

Mishra et al suggested new biomarkers like Human kallikrein-related peptidase 2 (hK2), α-methylacyl-coA racemase (AMACR), TGF-β1, early prostate cancer antigen (EPCA), insulin-like growth factors and binding proteins (IGFBP-2 and IGFBP-3), prostate-specific membrane antigen (PSMA),cytokine interleukin-6 (IL-6) with its receptors, urokinase plasminogen activator (uPA) with receptor (uPAR), enhancer of zeste homolog 2 (EZH2) for prostate cancer evaluation.[8] But a common cost effective biomarker is in urgent need for prostate diseases evaluation.

GGT (Gamma Glutamyl Transferase) is a membrane bound, cytoplasmic enzyme. It plays role in the normal and cancer cells growth. It is needed for the metabolism of glutathione and also other gamma glutamyl group containing molecules. It is synthesised in liver, kidney, pancreas, prostate etc.[9] Prostasomes are seen in storage vacuoles of prostate epithelium. They are membranous vesicles and found to contain sphingomyelin, cholesterol, calcium, and several enzymes. A major source of this enzyme is from the prostate where GGT is bound to the prostasomal membranes.[10,11] There is increased activity of GGT in prostasomes. The seminal plasma has 1000 fold increased GGT activity when compared with serum.[9]

So in our study we aimed to evaluate if GGT can be used as a biomarker for prostatic diseases and whether its values

correlate with the well studied prostatic biomarker PSA. In our study, the maximum individuals were in the 45-60 years age group. There were 19 diabetics in the prostatic cases and 104 diabetics in the control group. In our study there was negative correlation between PSA and GGT values and the values were not statistically significant.

Strasak et al observed the incidence of cancer in a large population based study, with the study population consisting of healthy men. They observed the relation of increased GGT in men with cancer incidence. Further they put forth that GGT levels are affected by diet. Certain fruits can reduce GGT levels so these factors have to be considered in GGT estimation. They concluded that elevated GGT may be associated with various site specific incidence of cancer especially in males <65 years old. Increased GGT is associated with increased possibility of overall incidence of cancers. Thus elevated GGT can be used for monitoring and decisions on intervention of cancer patients according to the study group.[13] Preyer et al studied the role of GGT as a pro oxidant and the role in carcinogenesis of breast. They evaluated the role of GGT irrespective of alcoholism as a risk factor. And they concluded that GGT is associated with breast cancer.[14]

Rubae et al in 2006, observed a positive correlation between PSA and GGT in prostate disease patients with BPH and prostatic cancer. GGT is needed for the growth and maintenance of prostate cells. It is also utilised by prostate neoplastic cells for its growth. The membrane bound enzyme will be released from the damaged cancer cells, into the circulation. The enzyme is increased in circulation in proportion to the damaged cells in neoplasm. GGT was observed to increase in epithelial tumours arising from colon, breast, prostate etc. The GGT released from neoplastic cells have a change in its normal activity. So this distinction can help to differentiate between normal and cancer cells. They suggested that GGT can be used as a general epithelial tumour marker. It was finally suggested in their study to include GGT as a prostatic cancer biomarker.[9]

Adekola et al from Nigeria observed no statistically significant difference between the prostatic cases and healthy control groups in the study. Serum levels of PSA, GGT and Malondialdehyde (MDA) showed no correlation in the study groups. MDA is a PUFA (Poly Unsaturated Fatty Acids) peroxidation end product which is stable since it is an aldehyde. It can be used as an oxidative stress marker as oxidative stress plays important role in case of many diseases including cancers. They concluded the study with the suggestion that GGT is not a good biomarker for prostate diseases though it can be comparatively seen to be increased in males.[3]

Kyojiro kawagami et al evaluated extracellular vesicles (exosomes) activity of GGT in serum whether it could be used as a prostatic cancer biomarker. Exosomes are 40-150 nm microvesicles that is known to contain various DNA, RNA, proteins and is synthesised by various cells. In their study they also correlated serum PSA and serum GGT values in prostate disease patients. Their study showed good

correlation between serum exosomal GGT and GGT1, which was isolated using differential centrifugation. GGT1 and GGT were shown to be elevated in prostate cancer patients when compared with BPH. They concluded that serum GGT may not be a good biomarker, but serum exosomal GGT may be a good biomarker to differentiate prostatic cancer from the benign condition, BPH.[2]

Liu et al. stated the importance of exosomes rich in other membrane proteins like PSMA (Prostate specific membrane antigen), which can serve as prostate tumour markers.[12]

Conclusion

In our study there is no statistically significant association between PSA and GGT levels in prostatic disorders Hence I conclude from my study that GGT cannot be used as a biomarker for prostatic diseases, as it needs further research. There are not much studies to confirm its use as a prostatic disease biomarker. More research is needed to evaluate its role in prostate diseases.

Limitations of the Study

Sample size may be increased to observe statistical significance for correlation between GGT and PSA levels.

Conflict of Interest: None.

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