Prostate-specific antigen (PSA) is a widely used marker for staging and monitoring of prostate cancer. Slightly PSA is a protease secreted by both normal and malignant prostatic epithelial cells with much secreted into the seminal fluid during ejaculation. Minor amounts normally leak into circulation although the release of PSA into circulation increases with prostate disease. Cancer of the prostate (CaP) is the second most commonly diagnosed cancer among men in the world. Its identification has been characterised by false positives associated with benign prostatic hyperplasia (BPH).1,2

The importance of PSA as a diagnostic tool for CaP came into light following a study in USA that determined elevation of total PSA in men with newly diagnosed, untreated prostate cancer. The sensitivity of cancer detection when tPSA is in the “grey zone” of 4-10 ng/mL range as compared to the use of tPSA alone with f/tPSA ratio being more predictive. The use of PSA remains uncertain. Notwithstanding resource limitations, there is a need for larger national cohort studies which may run concurrently with CaP awareness campaigns.

METHOD
In this cross-sectional study, 44 men presenting at Harare Central Hospital Urology Clinic were enrolled in the study. The mean age of the participants was 70 years (Range: 46 to 92). These men would have initially presented themselves to the Casualty officer at the hospital’s out patients department and had been thereafter referred to the Urology Clinic for further diagnosis and management. At the clinic, a PSA screen test would be ordered if digital rectal examinations (DRE) were abnormal. Participants were included in the study if they met the following criteria:

- Adult men aged 18 years and above
- Men with a tPSA level between 3 – 10 ng/mL drawn at time of initial evaluation
- Participants with a prostate biopsy request
- Men who would have consented

Participants excluded in the study were: men under treatment for prostate cancer, men less than 18 years, men with a tPSA level between 3 – 10 ng/mL from blood drawn at time of initial evaluation and men who did not consent to participate.

Written informed consent was sought from each participant and when granted questionnaires soliciting demographic and clinical parameters were administered. Consent ing men with a tPSA level of between 3 and 10ng/mL and a prostate biopsy request were enrolled in the study. Left over blood samples from the tPSA level determination was accessed for the determination of fPSA and cPSA. Patients’ records were also accessed to ascertain
the prostate biopsy findings. One urologist did the transurethral resection of the prostate (TURP) biopsies and two pathologists read the histology. To determine the outcome, PSA, tPSA, cPSA and their calculated ratios were used to determine the diagnostic ability of these isoforms in correlation to the biopsy results.

Total PSA assay was done using a Maglumi Total PSA (CLIA) (Shenzhen New Industries Biomedical Engineering Co. Ltd, Shenzhen, China) kit from the instruments supplier. The principle of the test is based on a sandwich immunoluminometric method. Free PSA assay was done using a Maglumi free PSA (CLIA) (Shenzhen New Industries Biomedical Engineering Co. Ltd, Shenzhen, China) kit. The principle of the test is also based on a sandwich immunoluminometric method. The cPSA assay was done using the Advia Centaur cPSA reagent pack (Siemens Healthcare Diagnostics, Deerfield, IL), which is a two-step sandwich assay using direct chemiluminescent technology. Bio-Rad Lymphocheck Tumor Marker Plus Control level 1 and 3 (BIO-RAD Laboratories, California, United States) were used as controls in the Total PSA assay. Sample results were only taken if the control values obtained for levels 1 and 3 were within ±3SD. A kit internal control was used as control material for the fPSA assay. Sample results were only taken if the control values were within the manufacturer’s reference range. Bio-RAD Lymphocheck Tumor Plus Control level 1 and 3 (BIO-RAD Laboratories, California, United States) were used as controls for the complexed PSA assay. Results were acceptable as levels 1 and 3 were within ±3SD.

RESULTS

Data used in the analysis was drawn from a cross-sectional study on 44 male participants attending Urology Clinic at Harare Hospital. The R version 3.0.1 (2013-05-16) statistical software was used in developing most of the statistical analysis with the exception of the ROC analysis. SPSS was used in ROC curve analysis. The study used non-parametric statistical methods such as Mann-Whitney test estimate the possibility of differences between two population samples (prostate cancer and BPH cases), exact logistic regression was used to obtain odds ratios. Only 5 out of 44 (11.4%) of the participants had histologically confirmed prostate cancer (adenocarcinoma). Explanatory variables measured were: PSA mean 14.9 (SD 1.8) as shown in Table 1. Table 2: Summary Statistics

| Variable (mean; SD) | Biopsy results | Prostate cancer (n=5) | BPH (n=39) | p-valuea |
|---------------------|----------------|-----------------------|------------|----------|
| Total (n=44) | 5.9; 2.1 | 6.2; 2.6 | 5.7; 2.1 | 0.7532 |
| PSA 0.9; 0.6 | 0.3; 0.1 | 1.0; 0.7 | 0.0033** |
| cPSA 4.1; 2.0 | 5.6; 3.2 | 3.9; 1.9 | 0.2365 |
| Ratio PSA (f/tPSA) 14.9; 1.8 | 4.8; 0.6 | 16.2; 12.8 | 0.0024** |

The p-value was determined using the Mann-Whitney two-sample mean comparison. A single asterisk(*) represents a significant difference, while double asterisks (**) represent very significant differences and triple asterisks (***') depict highly significant differences.

Variables found significant using the Mann-Whitney test (alternative to the t-test, used when assumptions of the t-test were not met) were PSA (p=0.003) and consequently as a ratio to total PSA (p=0.002). Total PSA and cPSA were not significant (p=0.753 and 0.237 respectively).

The exact logistic regression analysis was used to measure association between the occurrences of prostate cancer with the isoforms and ratio (f/tPSA). Results show that an increase in the fPSA significantly lowered the risk of developing prostate cancer (OR 0.01, p=0.003). However, the ratio of f/tPSA is more predictive of the occurrence of prostate cancer. An increase in the ratio is likely to reduce the chances of prostate cancer as depicted in Table 2.

| Variable | Odds ratio | p-valuea |
|----------|------------|----------|
| fPSA 1.07 | 0.7553 |
| cPSA 0.01 | 0.0031** |
| f/tPSA 1.37 | 0.1228 |
| f/tPSA 0.44 | 0.0005*** |

The p-value was determined using the Exact logistic regression. A single asterisk(*) represents a significant difference, while double asterisks (**) represent moderately significant differences and triple asterisks (***') depict highly significant differences.

A ROC was fit in SPSS 17 to compare diagnostic ability of PSA and ratio f/tPSA to the biopsy outcome as depicted in Figure 1. The area under curve (AUC) was used as a measure of comparison between the tests.

DISCUSSION

The major limitation in this study was the small sample size as the prostate cancer cases in the study population are generally low. This is in accordance to the WHO (2002) data table for cancer impact by country that reported an incidence of 30/100 000 in Zimbabwe whereas in the United States 166/100 000 were reported. Another reason for low sample size than anticipated was that some participants could not afford the cost of referring biopsies to private pathologists. This resulted in our tests having reduced power. There are limited biopsies done at government hospitals due to shortage of both human and financial resources. This has been compounded by the socio-economic and socio-political dynamics whose cascade effect impacted negatively on health service delivery. The post year 2000 era heralded a phase of brain drain and reduced budgetary allocations to government institutions as a result of the international sanctions imposed on the Zimbabwean government. In this paper, cut off points could not be established for the different parameters. However, some meaningful conclusions were drawn from the data, whose information can be used to generate new
hypothesis for further studies in the same area of study.

Here we found a prevalence of 11.4% for prostate cancer, which was slightly lower as compared to other studies within the same tPSA range\textsuperscript{15,17} this could be as a result of the methods used in the inclusion criteria. In this study a tPSA range of 3.0 -10 ng/ml was used while most studies included participants with any tPSA value. However, a study by Thakur et al.\textsuperscript{18} reported a prevalence of 13.0% although they did not use a specific tPSA range in their study. The information obtained in this study support finding from previous studies in terms of tPSA, fPSA, cPSA and f/tPSA ratio relative to differences between cases of prostate cancer and other benign neoplasms. This research found BPH cases to have lower tPSA (mean 5.7 versus 6.2 ng/ml) and cPSA (mean 3.9 versus 5.6 ng/ml) than in prostate cancer cases but the differences are statistically not significant (p>0.05). In a retrospective study of 180 patients, Chen ZD et al.\textsuperscript{19} also came to the same conclusion since the mean value of tPSA was not statistically different between patients with prostate cancer and BPH (p>0.05).

The mean fPSA was significantly lower in prostate cancer than in BPH cases (0.3 versus 1.0 ng/ml) and consequently the mean f/tPSA values (4.8 versus 16.2 %) Sakai et al.\textsuperscript{20} also reported that f/tPSA ratio was significantly lower in prostate cancer patients than in BPH patients. The mean cPSA was lower in prostate cancer patients than in BPH patients (5.6 versus 3.9 ng/ml) although the values were not statistically significant (p>0.05). In their study of 54 patients, Tamimi et al.\textsuperscript{21} reported no significant association between high cPSA values and prostate cancers and they concluded that cPSA presented little advantage over tPSA for discriminating between BPH and prostate cancer in the population that they studied. However, such conclusions might arise due to small sample size as other researchers actually reported cPSA being statistically significant between patients with prostate cancer and BPH.\textsuperscript{21}

The sensitivity and specificity values were in the range of what has been found in other studies. Both fPSA and f/tPSA had high and equal sensitivity value of 80%. However, the f/tPSA ratio had a significantly higher specificity (90% versus 79%). Results for specificity were on the contrary as many of the studies have noted that these biomarkers have high sensitivity and low specificity in the tPSA range of less than 10 ng/ml. A systematic review and meta-analysis reported a specificity of 18% in the 4-10 ng/ml range and 6% in the 2-4ng/ml range at a sensitivity of 95% \textsuperscript{21}. The optimal cut off points for fPSA, cPSA and f/tPSA ratio could not be established to classify individuals as having prostate cancer or not again as a result of low power in our study.

There is need to carry out cross-sectional studies \textit{major} stake holders and reproductive health organizations may support such a CaP\textsuperscript{23,24} initiative. An interesting dimension during this cohort study would be to relate CaP to HIV as other researchers have found a positive correlation.\textsuperscript{25,26,27} Such an integral approach will maximize resource utilization whilst increasing project feasibility.\textsuperscript{28}

**Conclusion**

This paper is in conformity with other reports that f/tPSA ratio can aid in the diagnosing of prostate cancer in the 3-10 ng/ml range as compared to the use of tPSA alone. The use of f/tPSA ratio was found to be more predictive because its association with the occurrence of prostate cancer was highly significant (p<0.05). The use of cPSA remains controversial, hence warranting further study. The diagnostic ability of the use of the isoforms so as to reduce unnecessary biopsies was not ascertained as cut offs of these isoforms and their ratios could not be established. This could also have been a result in bias of the statistical methods, as most of the assumptions to carry them were not protected by the available data. To that end, results in this study cannot be used to make direct recommendations for clinical practices, but for the formulation of new hypothesis to be conducted in using larger cohorts. This also entails effective resource mobilization for an inclusive program that may be run along a national CaP awareness campaign in both urban and rural areas.

**Competing interests**

All authors declare that they have no competing interests.

**Authors’ contributions**

HTM was involved in designing the study, supervision of fieldwork and data analysis. SU contributed in proposal writing, laboratory based assays and data analysis. PJ contributed in the discussion and data analysis. CM was involved in drafting the manuscript and discussion of the Public Health implications of the study. The final manuscript was read and approved by all authors.

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