Research Article

Foxp3 and p53 Expression in Prostate Carcinoma and Their Correlation with Histological Grades Among Archived Tissue Blocks at Uganda Cancer Institute

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Abstract: Prostate carcinoma remains a leading cause of cancer-related morbidity and mortality among men in Uganda. Immunohistochemical markers such as Foxp3 and p53 have emerged as potential indicators of tumor aggressiveness, but their expression patterns in Ugandan prostate carcinoma blocks remain underexplored.

To determine the immunohistochemical expression of Foxp3 and p53 and assess their correlation with histological grades in prostate carcinoma tissue blocks at the Uganda Cancer Institute.

We conducted a cross-sectional study on formalin-fixed paraffin-embedded prostate carcinoma tissue blocks that were diagnosed between December 2024 to January 2021. Hematoxylin and eosin staining was used for histological grading, and immunohistochemistry was performed to detect Foxp3 and p53 expression. The correlation between markers' expression and histological grade was analyzed using Spearman's correlation test, and a p-value of ≤ 0.05 was deemed significant.

Foxp3 expression was absent in 81.9% of blocks and detected in only 18.1%, predominantly in Grade Group 1 tumors. However, the association between Foxp3 expression and histological grade showed a negative correlation. In contrast, p53 was expressed in 100% of the tissue blocks analyzed and showed a statistically significant positive correlation with higher histological grade. The aggressiveness of prostate carcinoma was shown by overexpression of p53 and the absence of Foxp3 in most of the prostate tissue blocks.

This study gives information on the relevance of Foxp3 and p53 expressions in prostate cancer diagnosis and prognosis in Uganda.

Keywords: Foxp3, p53, histological grades, prostate Carcinoma , Uganda.

1. Introduction

Prostate carcinoma (PCa) is a malignancy of the prostate gland, a common tumor that affects the male reproductive system, and is distinguished by the uncontrolled proliferation of cells in the prostate gland (Ljokjel et al., 2022). It is the second most frequently identified malignancy and the fifth leading cause of fatalities due to cancer in males globally (Sekhoacha et al., 2022).

In 2020, prostate carcinoma was the fourth type of cancer, with the highest incidence of 1.41 million blocks globally (Ferlay et al., 2021). Males over 50 years old have the highest risk of developing prostate carcinoma, and African-American men have the highest incidence rates and more aggressive kinds of prostate carcinoma than white men (Jemal et al., 2021, Rawla, 2019).

It is the sixth leading cause of cancer death among African men and the most frequent cancer in East Africa where Uganda lies with a relative death rate of 29 per 100,000 men and an age-standardised incidence rate (ASR) of 40.6 per 100000 (Mbugua et al., 2021) (Cassell et al., 2019) (Jemal et al., 2021, Rawla, 2019) . As of 2023, PCa was the second most common male cancer in Uganda (Nakaganda et al., 2023).

However, diagnosis of PCa is by biopsy that renders it to histology processes, staining with routine staining technique (Heamatoxyline and Eosin) that is amorphological stain and finally microscopic examination and grading where by its having shortcomings such as poor specificity since it doesnot directly asses protein expression of markers such as Foxp3 and p53. This limits molecular insights that can be offered by immune-histochemistry.Nevertheless,Foxp3 and p53 markers have shown to be good markers in the prediction of the disease aggressiveness relatively to H&E alone (Murray et al., 2019),(Rana et al., 2020),(Cimadamore et al., 2019). (Surintrspanont and Zhou, 2022).

Foxp3 transcriptionally inhibits Cellular myelocytomatosis oncogene (C-Myc) expression and Tuberous Sclerosis Complex 1(TSC1) which regulates the phosphorylation of C-Myc in prostate carcinoma cells, and dysfunction of TSCI and Foxp3 synergistically increases tumour progression of the PCa through regulating transcription and stabilization of C-My(Wu et al., 2019),(Hao et al., 2024).

On the other hand, the p53 protein, known as the "guardian of the genome," plays a pivotal role in preventing cancer development by regulating cell cycle progression and apoptosis (Capuozzo et al., 2022). This is done by inhibiting and promoting the tricarboxylic acid cycle and oxidative phosphorylation in the prostate epithelium (Qu et al., 2025, Chen et al., 2021). Dysregulation of p53 protein may contribute to prostate carcinoma development and progression (Bishop et al., 2014).

Despite the increasing burden of prostate cancer in Uganda more so at Uganda Cancer Institute (UCI) which is the biggest and most comprehensive cancer treatment centre in East Africa, PCa diagnosis is confirmed histologically by examining Hematoxylin and Eosin-stained sections. Biomarker stratification for Foxp3 and p53 remain scarce due to a lack of awareness about their implications in prostate carcinoma management, and their correlation with histological grades of PCa is not known. This underscores the clinicopathological relevance of these markers. Foxp3 and p53 have shown to be good prognostic markers of PCa for predicting advanced/metastatic disease (Iovanna, 2021). Therefore, in this current study, our main objective was intended to determine Foxp3 and p53 expression in prostate carcinoma and their correlation with histological grades among archived tissue blocks at UCI.

2. Methodology

Study design and setting

We conducted a laboratory-based Cross sectional study at UCI pathology laboratory. This laboratory is internationally accredited under the facility accreditation number: M0921. UCI acts as an East African Cancer treatment centre and pathology laboratory as a Ugandan national referral centre for pathology samples.

It is located along Upper Mulago Hill, central division, Kampala, Uganda, about 5km northeast of the central business district (Kampala) of the city at coordinates of $00^0 20'29.0''$ N,$32^0 34'40.0''$ E (latitude: 0.341389; longitude: 32.577778). The

Pathology Department has five pathologists, 4 technologists, and some residents.

The UCI pathology laboratory provides services to people who come from all regions of Uganda, plus its neighboring countries of Rwanda, Tanzania, Kenya, South Sudan, and the Democratic Republic of Congo. It receives about 300 prostate carcinoma biopsies every year.

Study Period: We retrieved PCa tissue blocks from December 2024 to January 2021

Study Population: We used prostate Carcinoma blocks that were in the archival; each PCa tissue block represented a single carcinoma case.

Sampling Criteria:

A purposive sampling for all achieved prostate carcinoma blocks in the Pathology laboratory of UCI until the sample size attained was used. Archived prostate tissue blocks histologically diagnosed with prostate carcinoma and documented in Laboratory information management system (LIMS) were traced and retrieved using their corresponding laboratory numbers.

Inclusion Criteria:

All formalin-fixed paraffin-embedded prostate carcinoma tissue blocks archived in the Pathology Laboratory of UCI were included in the study.

Exclusion Criteria:

I.   Poorly fixed and processed blocks and not feasible for IHC.
II. Prostate carcinoma tissue blocks that were depleted after sectioning.
III. Blocks that didn't show the prostate carcinoma tumour following repeat with the H&E staining technique.

Sample size Estimation:

We used Cochran, 1977 formular, sample size was calculated based on the number of prostate carcinoma specimens received at UCI annually and later corrected to finite population,

and final minimum sample size for the study was 116 prostate carcinoma tissue blocks. However, during retrieval of the blocks, we managed to get 127 PCa cases.

Work flow of laboratory procedures:

These procedures involved during our study include; retrieving PCa results from LIMS along with corresponding tissue block, and later assigned new study identifier numbers.This was followed by microtomy, floating out , H&E staining, IHC , examination of stained slides and reporting of the study findings.

Staining with H&E

We used regressive staining, and tissue sections were microscopically examined by the patholologist to confirm the prostate carcinoma and grade as per the standard. PCa sections were dewaxed, rehydrated then stained with Hematoxylin for 10 minutes, differentiated in with 1%acid alcohol until the nucleus was fully stained and then washed with water to stop differentiation, bluing with running tap water for 5minutes, and then stained with Eosin for 60 seconds . Sections were then rinsed in 95% alcohol to remove excess eosin stain, rapidly dehydrated using absolute alcohol in 2 changes, dealcoholized using Xylene in two changes and used Dibutylphthalate Polystyrene Xylene (DPX) to mount.

Foxp3 and p53 Immunohistochemistry staining:

Selected sectioned PCa tissues were made at a thickness of 3 micrometre, floated out with a positively charged slide (Bio-optica company), then left to air dry. This was followed by baking them at 60^0 for 2hours. Sections were dewaxed with 2 changes of xylene for 3 minutes in each , hydrated through decreasing changes of alcohols and then gently rinsed in distilled water.

Pca sections were then taken for antigen retrieval in a Biocare Medical decloaking chamber containing Epitope retrieval solution containing 0.01M sodium citrate buffer of pH

9.0 at temperature of 95^0c for 45 minutes.Upon cooling, slides were removed and cleaned with Tris buffer solution (TBS) for 60 sections fellowed by blocking (blocking endogenous peroxidases and other protein binding sites) . Immunohistochemistry was performed by applying primary antibodies (Foxp3 which was a monoclonal from abcam (236A/E7) and p53 (AB1101)) on one of the two slides prepared for IHC as per case for 15minutes . Later we washed off the antibodies with TBS then followed by secondary antibody (Horsh reddish peroxidase) for 10minutes then washed , applied achromogen (Diaminobenzidine solution) for 40 seconds and washed off. Sections were then counterstained with 4 dips of Mayer's hematoxylin for a minute then blued blued in in tap running water , dehydrated using increasing concentrations of alcohol, cleared with xylene in two changes and then mounting was done using DPX.

We used known controls for staining; H&E, we used a colon tissue. Foxp3 antibody, a normal spleen tissue section was used for positive control, and a skeletal muscle tissue block as a negative control. For the p53 antibody, we used a colorectal carcinoma known tissue block and a known benign breast tissue block.

H&E grading, Foxp3 and p53 scoring:

In this study, we used the Current ISUP grading of prostate biopsies was utilised: Grade Group 1 = Gleason 6 (or less), Group 2 = Gleason 3+4=7**,** Group 3 = Gleason 4+3=7, Group 4 = Gleason 8, and Group 5 = Gleason 9-10 (Samaratunga et al., 2016). Some of the photomicrographs are shown below **(Figure 1 A-E)**
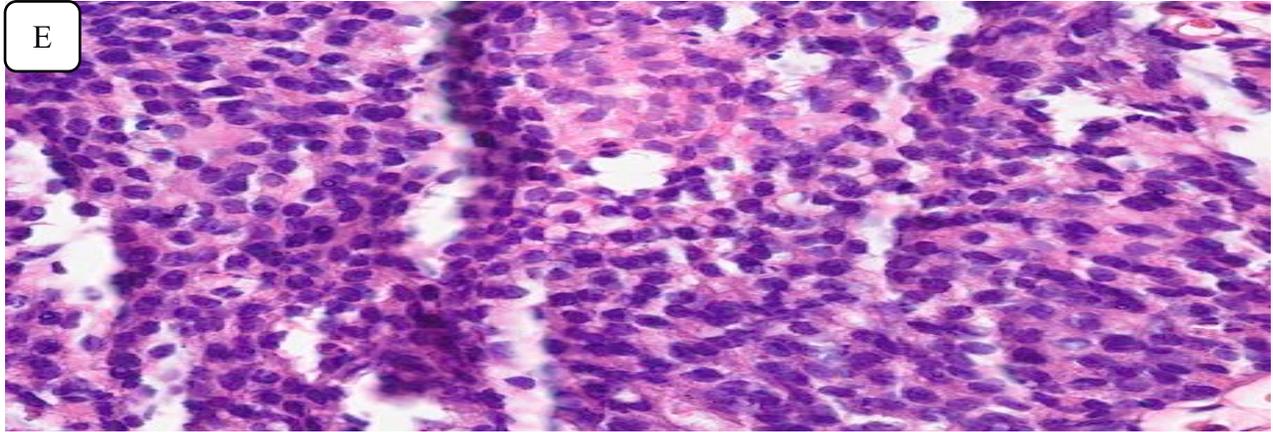
Figure 1:
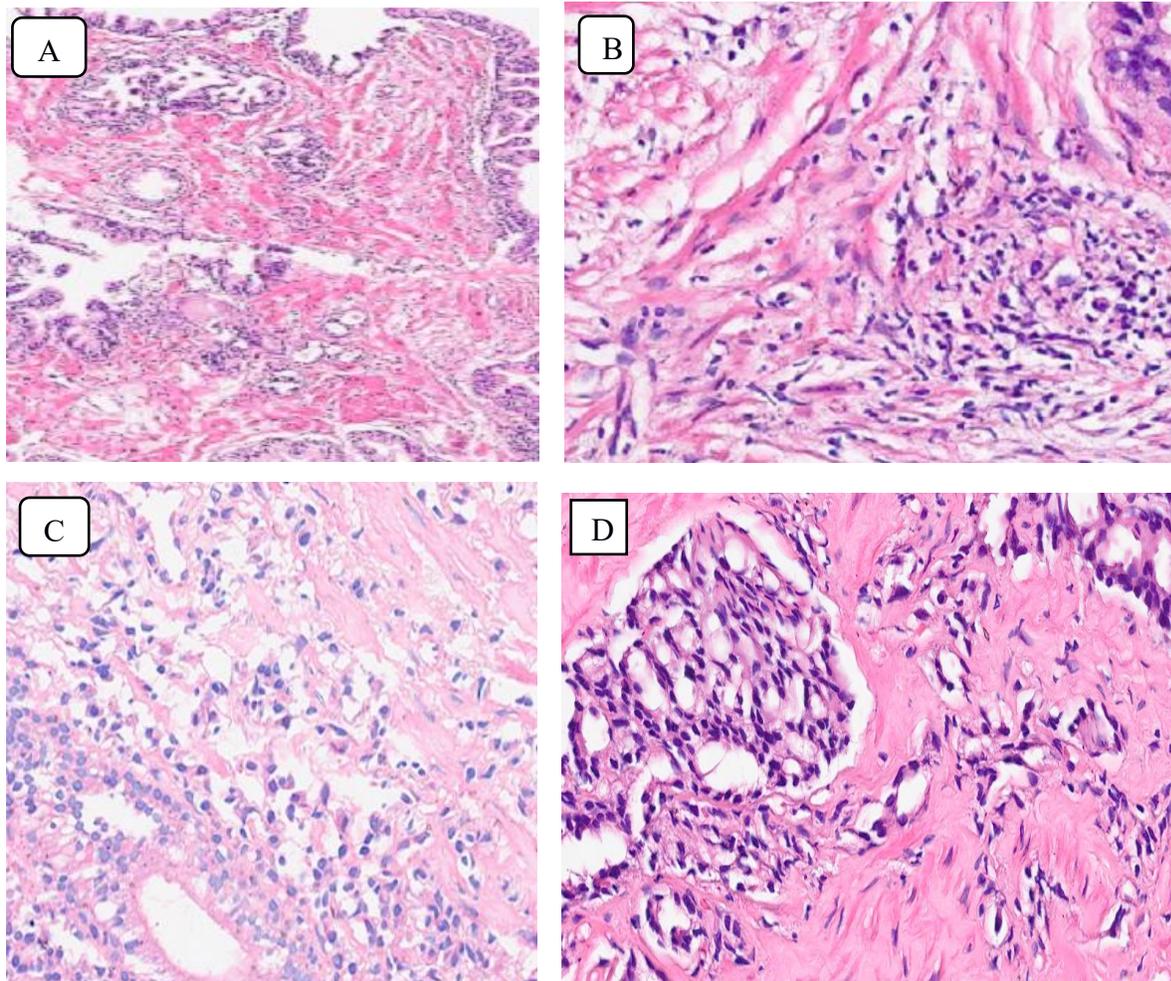
FIGURE 1 : Magnification: X200, **A-** Histological grade 1**:** Glands well formed with no fusion**. B-** Histological grade 2**:** Some glandular fusion and poorly formed glands**. C-** Histological grade 3: Almost 50% with poorly formed glands**. D-** Histological grade 4: Nearly all glands are fused and poorly formed**. E-** Histological grade 5**:** No glandular formation observed.

For IHC's scoring, we adopted a semi-quantitative grading system for Foxp3 and p53 expression as was used by (Cunha et al., 2012) and (Wang et al., 2009), respectively. The percentage of cells showing p53 expression was counted using a 10x objective, and the expression was graded as negative if stained sections showed no positive reaction. When less than 25% of cells stain positive, they were graded as weakly expressed (+). When 26%-50% of the cells were expressed, the section was graded as moderately expressed (++), and with greater than 50% expression, they were graded as strongly expressed (+++) (Wang et al., 2009b). **(Figure 2 A- C)**

**Figure 2: Scanned slides showing different p53 staining scores.**
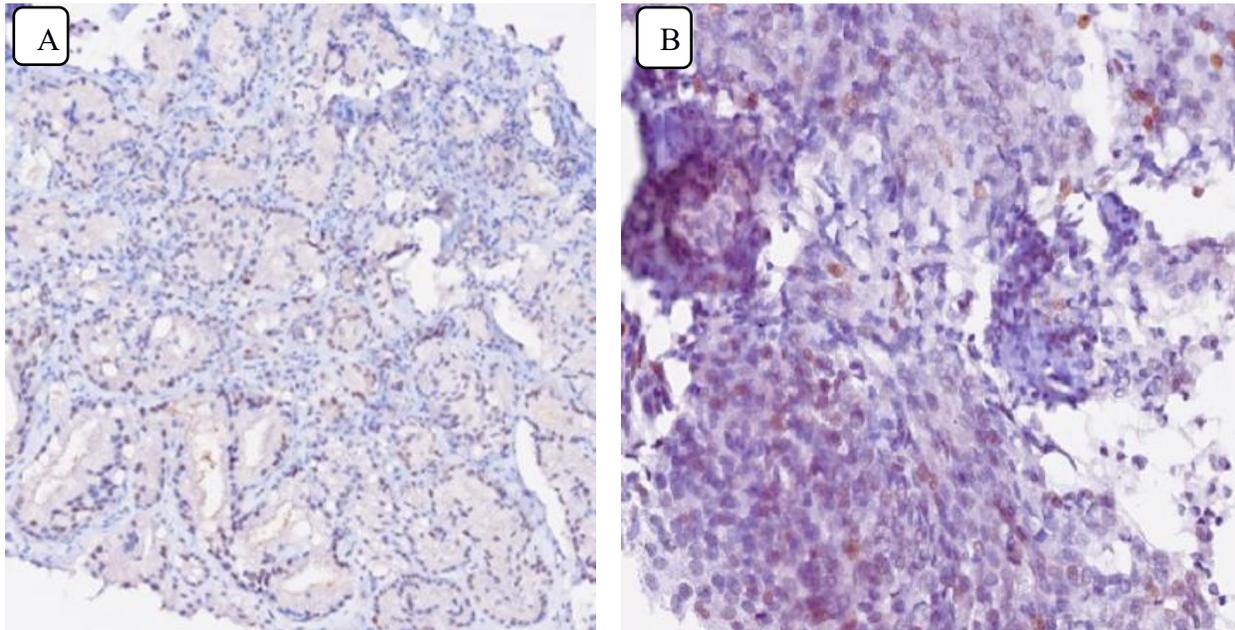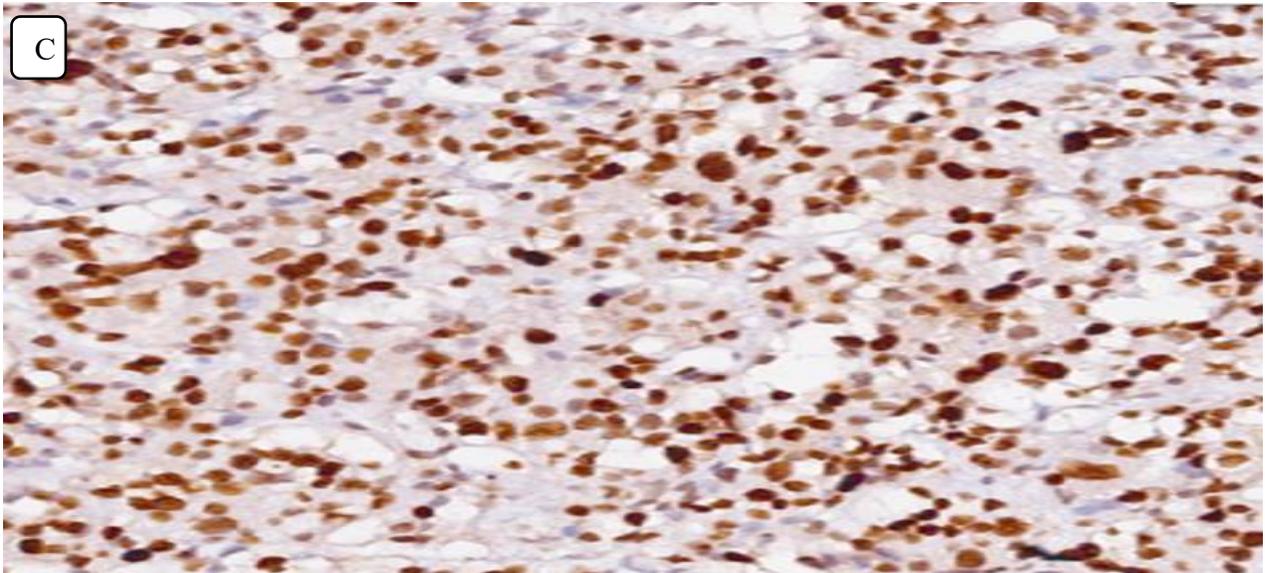
p53 expression scored: Magnification: X200, **Slide A**-<25 cells stained weakly with p53, **Slide B**-26-50% stained moderately, and **Slide C**- >50% had a strong staining intensity with p53.

Foxp3 scoring was borrowed from Cunha et al. (2012), who looked at differentiated thyroid carcinomas, and scoring was as follows: 0 = no positively stained cells; 1 = up to 10% positively stained cells; 2 = 10 to 30% positive cells; and 3 = more than 30% of cells stained with Foxp3 antibody as shown in **(Figure 3 A-C)** below.

Figure 3: Scanned slides showing different Foxp3 staining scores.
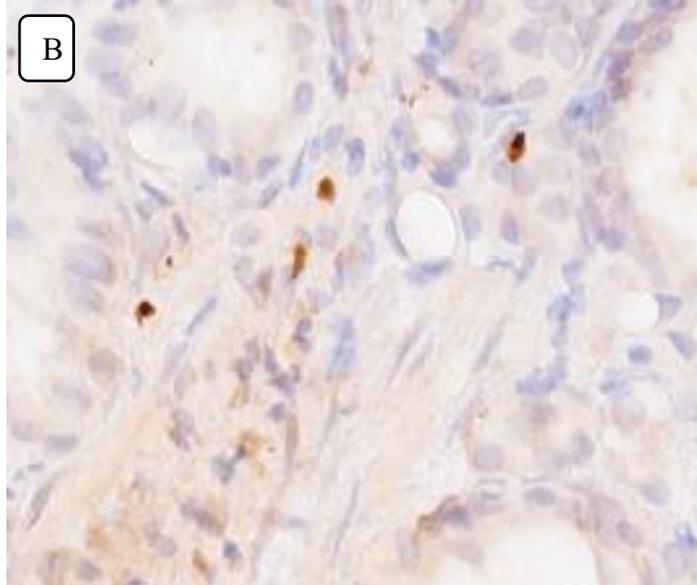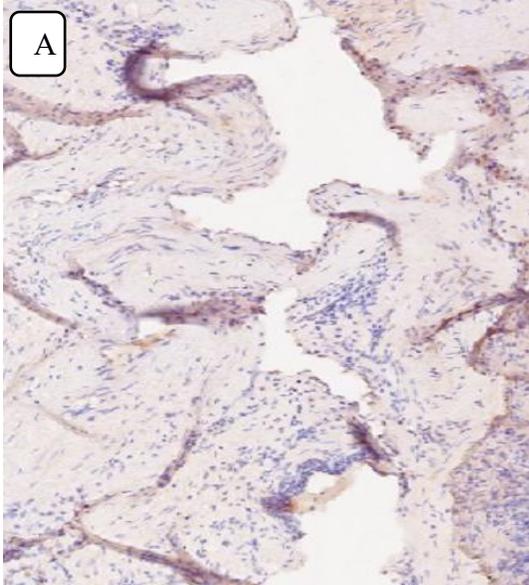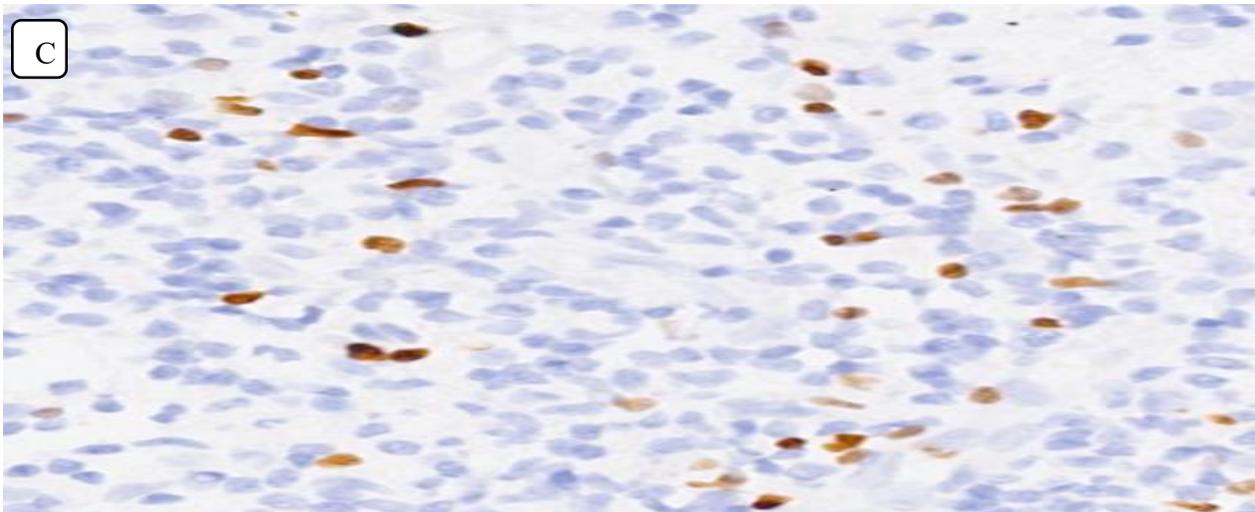
Magnification: X200, **A**-Slide with Negative stained Foxp3 cells attained counterstain colour, **B-** slide with 1-10% cells stained around 6-7 cells only stained. **C-** Slide with 11-30% cells stained with 28 cells stained with Foxp3.

Data analysis and management

We collected data using a data collection tool that contained all the parameters of our interest. After data was entered to an excel spreadsheet that was exported into STATA version 17 for analysis and for the safety of our data, computer used had a password that was known by the main principal investigator. For objective one, the expression of Foxp3 and p53 levels was presented in the form of frequencies and percentages. To achieve objective two, we expression levels of Foxp3 and p53 were correlated with histology grades. A categorical variable of Foxp3 and p53 was correlated with a categorical variable of histological grades using Spearman's correlation test and a p-value of ≤ 0.05 was deemed statistically significant.

Ethical consideration

We sought letters of approval from the Department of Medical Laboratory Science (MLS), the Faculty of Research Committee (FRC), MUST-Research Ethics Committee (REC) reference number MUST-2024-1714, and an administrative clearance/ Permission letter to use archived blocks from UCI administration/ UCI – REC.

3. Results

Although the determined sample size was 116 prostate carcinoma blocks, we managed to retrieve 127 blocks that also passed the inclusion criteria; hence, 127 blocks were used as the total sample size. Most of the study participants were above 60 years, and a few (15%) were less than 60 years of age. The biggest age group with prostate carcinoma was between 71-80 years, which accounted for 35.4%. On histological grading, most of the blocks examined were in histological grade five (50.4%), and a few with histological grade one, 12 blocks (9.4%), as shown in the table1 below:

Table 1: Demographic Characteristics of Prostate carcinoma blocks at Uganda Cancer Institute**.**

Variable	Frequency (Percentage,%) n=127
Age (years): Mean ±SD	70.8±10.3
Age groups (Years)	
≤60	19 (15.0%)

61-70	43 (33.9%)
71-80	45 (35.4%)
>80	20 (15.7%)
Histological grading	
Grade one	12 (9.4%)
Grade two	16 (12.6%)
Grade three	13 (10.2%)
Grade four	22 (17.3%)
Grade five	64 (50.4%)

Abbreviation: SD, Standard Deviation

Age was presented as a continuous variable with the corresponding Mean ± SD, since our age was normally distributed across the study participants.

Levels of Foxp3 expression in prostate carcinoma blocks at UCI.

The majority of the IHC prostate carcinoma blocks subjected to Foxp3 IHC showed a negative expression 104/127 (81.9%), while 23/127 (18.1%) showed a positive expression, and on the staining score, Cells stained were less than 30%, as illustrated in Table 2

Table 2: Foxp3 expressions in prostate carcinoma blocks at UCI.

Variable	Frequency n=127	(percentages, %)
Foxp3 un stained cells	104	81.9%
1-10% stained	16	12.6%
11-30% stained	7	5.5%

Expression Levels of p53 in Prostate Carcinoma Blocks at UCI

All tissue blocks that passed the exclusion criteria on IHC 100% (127 blocks) showed expression with p53, with most of them moderately stained, 26-50% of cells stained counted for 54.3% (69 blocks) as indicated in Table 3

Table 3: Levels of p53 expression in prostate carcinoma blocks at UCI

Variable	Frequency n=127	(Percentages, %)
p53 cells stained		
<25%	37	29.1
26-50%	69	54.3
>50%	21	16.5)

Correlation between Foxp3 expression and Prostate carcinoma histological grades

A significant correlation of Foxp3 expression with different prostate histological grades was observed in No stained cells versus grade one and grade five (Spearman's rho (Prob>t) (-0.1976(0.0260)), (0.1877 (0.0346)) respectively. A positive correlation in 1-10% of the stained cells with histological grade one 0.2830 (0.0013), and a negative correlation with histological grade five (-0.2403 (0.0065)) was also identified, as summarized in Table 4 below

Table 4: Correlation between Foxp3 expression and Prostate carcinoma histological grades

Fox p3	Histological grading
Expression	

	Grade1 Spearman's rho(Prob>t)	Grade 2 Spearman's rho(Prob>t)	Grade 3 Spearman's rho(Prob>t)	Grade 4 Spearman's rho(Prob>t)	Grade 5 Spearman's rho(Prob>t)
No stained cells	-0.1976 (0.0260)*	-0.1295 (0.1466)	-0.0436 (0.6268)	0.0532 (0.5527)	0.1877 (0.0346)
1-10%	-0.2830 **(0.0013)***	0.0704 (0.4317)	0.1066 (0.2328)	-0.0484 (0.5891)	-0.2403 (0.0065)
11-30%	-0.0780 (0.3833)	0.1163 (0.1930)	-0.0816 (0.3620)	-0.0194 (0.8288)	-0.0326 (0.7160)

*Statistically significant (P<0.05)

Correlation between p53 expression and Prostate carcinoma histological grades

A statistical significance correlation of p53 expression with different prostate histological grades was observed in > 50% stained cells versus grade one as summarized in Table 5 below.

Table 5: Correlation between p53 expression and Prostate carcinoma histological grades

P53 Expression	**Histological grading**				
	Grade1 Spearman's rho(Prob>t)	Grade 2 Spearman's rho(Prob>t)	Grade 3 Spearman's rho(Prob>t)	Grade 4 Spearman's rho(Prob>t)	Grade 5 Spearman's rho(Prob>t)
1-25%	-0.0294(0.7429)	0.1221(0.1714)	0.0122 (0.8921)	0.0728 (0.4158)	-0.1264 (0.1569)
26-50%	-0.1362 (0.1269)	0.0146 (0.8703)	-0.0033(0.9708)	0.0437 (0.6253)	0.0388 (0.6647)
>50%	0.2185 (0.0016)	0.1690(0.0575)	-0.0105(0.9071)	0.1477(0.0974)	0.1025 (0.2516)

*Statistically significant (P<0.05)

4. Discussion

We discussed key findings obtained in our study on Foxp3 and p53 immunohistochemical expression in prostate carcinoma tissue blocks and their correlation with histological grades among prostate cancer cases at the Uganda Cancer Institute (UCI). We interpreted these results in comparison with other researchers' findings.

Table 6 Summary of our findings in comparison with other studies' findings attached

Our findings	**Similar findings**	**Dissimilar findings**
Foxp3 expression in prostate carcinoma was 18.1% with CI= 12.3-25.9	Foxp3 expression was 17% in prostate carcinoma blocks from Sweden (Erlandsson et al., 2023)	In USA, 31.5% of Foxp3 expression was observed in prostate carcinoma blocks by (Wang et al., 2009a) . 88.8% Foxp3 expression was observed in Germany (Flammiger et al., 2013).
p53 expression showed 100%	100% Indonesia (Singh et al., 2017)	49.8% United States (Gesztes et al., 2023)

A significant negative correlation of Foxp3 with histological grade 5	(Qiu et al., 2022), China (Bulten et al., 2022), China	Study not found
Positive correlation of p53 and histological grades with prostate carcinoma biopsy was found	(Gesztes et al., 2025), USA (Bhat et al., 2023) (Asia)	None found

Foxp3 expression in prostate carcinoma blocks at the Uganda Cancer Institute.

Foxp3,amember of the the FOX protein family, acts as amaster regulator in the growth of and operation of regulatory T cells (Treggs) These cells play acrucial role in immune system reactions , Particularly in suppressing anti- tumour immunity with in the tumor microenvironment.. In our current study, Foxp3 expression was detected in 18.1% of the prostate carcinoma cases. Our findings are in consistent with a study research that was conducted in Sweden that found out that expression of Foxp3 was at 17% (Erlandsson et al., 2023) . The close similarity of these results might be stemmed from the use of the same

anti-Foxp3 (Tregs) monoclonal antibodies utilized in both studies came from the same supplier, Abcam, Cambridge, UK.

However, other studies conducted in USA and Germany had dissimilar results with our findings of 31.5% and 88.8% respectively (Wang et al., 2009a) and (Flammiger et al., 2013). These discrepancies highlight potential influencesbeyond antibody clone. Differences in patient cohorts, such as the smaller sample size in some comperative studies(fore example, 92blocks Vs our 127 blocks), or prior to treatment like androgent deprivation therapy (ADT) before prostatectomy(Flammiger et al., 2013), could have significantly altered the immune landscape and Foxp3 expression.

Additionally, varriations in IHC techniques, including antigen retrieval protoccols and scoring methodologies , could have contributed to these divergent findings . The biological context of Foxp3 expression in Pca is complex; its presence often indicates an

immunosuppression microenvironment that can promote tumor progression by dampening effective anti- tumour resposes. The rerelatively low expression in our cohort compared to some studies might suggest adifferent immune profile in Ugandan Pca patients or specific characteristics of the tumor microenvironment in these archived samples.

Expression of p53 in Prostate Carcinoma Blocks at UCI

In our present study, p53 showed 100% expression in prostate carcinoma blocks.These findings aligns with a study conducted in Indonesia by Singh et al., 2017 that found 100% p53 expression in all prostate carcinoma blocks tested using IHC. This high concordance might be attributed to the posiblity of using similar monoclonial antibodies and detection systems. However, its crucial to differentiate between p53 accumulation (Detected by IHC) and actual p53 gene mutation . While p53 overexpression usually correlates with amutated p53 protein that has along half life , it can occur with wild- type of p53 under cellular stress, leading to protein stablisation . The consistent 100% expression in our study suggests awidespread alteration in p53 pathway regulation within these PCa cases, potentially indicating early pervasive genomic instability or a high prevalence of p53 mutations in this cohort. Conversely, Gesztes et al., 2023 reported 49.8% expression of p53 in their prostate tissue blocks, despite using an African target population . This significantly differences from our findings could be due to the diverse types of biopsies they included such as radical prostatectomies, prostate chips , metastatic deposits , and core biopsies. Our study exclusively utilized core biopsies ,

which might represent amore localised orr specific tumor area compared to to the broader sampling radical prostatectomy specimen. Different Tissue processing and fixing protocols could have also influenced antigenicity and detection. The biological implication of asuch high p53 expression in our cohort warrants further investigation in future , potentially through genetic sequencing , to confirm mutation status and understant its functional concequences in PCa Progression.

Correlation between Foxp3 expression and histological grades of Prostate carcinoma

The correlation analysis between Foxp3 expression and prostate carcinoma histological grades revealed a grade-specific pattern of association. Using Spearman's rank correlation, our results highlighted both positive and negative relationships between varying levels of Foxp3 expression and the severity of histological grading.

In Grade 1 tumors, a significant negative correlation was observed between Foxp3 "no stain" and histological grade (rho = -0.1976, p = 0.0260), suggesting that the absence of Foxp3 expression may be more common in lower-grade tumors. Conversely, a significant positive correlation was noted between the 1–10% expression level and Grade 1 (rho = 0.2830, p = 0.0013), indicating that mild Foxp3 expression is more prevalent in well-differentiated tumors.This mild infiltration of Tregs might of treggs might represent an early immune response to the nascent tumor, or it could contribute to the initial establishment of immune tolerance , allowing the tumor to invade early immune surveillance.

As histological grade increased, the strength and significance of these correlations diminished. In Grade 2 tumors, none of the Foxp3 expression levels showed statistically significant correlations. For Grade 3 and Grade 4, all expression categories had weak and non-significant associations with histological grade, implying a more heterogeneous or less consistent expression pattern of Foxp3 in moderately differentiated

tumors.This variability might reflect atransitoinal phase phase where the tumor micro environment is undergoing complex changes , potentially involving shifts in immune cell recruitment , activation and suppression, making aclear correlation with Foxp3 expression challenging . Other immune evasion mechanisms might also become more dominant , reducing the singular impact of Treg infiltration.

Interestingly, in Grade 5 tumors, a significant positive correlation was found with "no stain" (rho = 0.1877, p = 0.0346), suggesting an increase in Foxp3-negative stained blocks in high-grade tumors. Additionally, there was a significant negative correlation with 1–10% expression (rho = -0.2403, p = 0.0065), indicating a reduction in mild Foxp3 expression at higher tumor grades. These findings may reflect a potential immune escape mechanism where tumors either suppress Treg infiltration or alter their immunogenic profile to facilitate progression (Quinn et al., 2015). Alternatively , these highly aggressive tumors might have evolved to rely on other, more potent immunosuppressive mechanisms, making the presence of Tregs less critical for their survival and growth. The lack of significant correlation with moderate (11–30%) Foxp3 expression across all grades suggests that intermediate levels of Foxp3 do not distinctly characterize any particular histological group. This could reflect adynamic , transitional immune state within the tumour microenvironment , whereTreg populations are fluctuating or their functional roles are being modulated by other factors (Sakaguchi et al., 2009).

Overall, our findings suggest that Foxp3 expression shows a non-linear and grade-dependent relationship with prostate carcinoma differentiation. Mild expression (1–10%) appears to be more common in early, low grade tumors, whereas the absence of expression becomes more frequent in high-grade tumors. These patterns support the hypothesis that immune modulation via regulatory T cells may be more pronounced in

early-stage tumors, with potential suppression or evasion occurring in advanced stages (Qiu et al., 2022). Therefore further studies intergrating functional assays of Tregs and analysis of other immune cell populations would be of great importance to provide amore comphrensive understanding of these complex dynamics.

5.5 Correlation between p53 expression and Prostate carcinoma histological grades

The expression of the p53 protein, as assessed through immunohistochemical staining, demonstrated grade-specific associations with prostate carcinoma histological differentiation. Spearman's correlation analysis revealed that statistical significance was achieved only in Grade 1 tumors, particularly in the group with >50% of cells positive for p53 staining (rho = 0.2185, p = 0.0136). This suggests a unique pattern of p53 dysregulation in low-grade prostate carcinoma tumors compared to higher-grade lesions.

In Grade 1 (well-differentiated) tumors, the positive correlation with high-level p53 expression (>50%) may reflect a critical finding. This high expression typically reflects the accumulation of mutant p53 protein, which often manifests a longer half-life and resistance to degradation compared to wild – type p53. This leads to its nuclear stabilization and IHC staining. This implication is that p53 pathway alteration, likely through mutation , can be an early event in in prostate tumor genesis (Garreffa and Lee, 2024). This observation challenges the traditional view that p53 mutations are predominantly late-stage occurrence in PCa progression. Instead, it supports growing evidence that molecular changes such as p53 alterations, can precede significantly histological differentiation (Ofner et al., 2025). The presence of significant p53 overexpression in these low- grade tumors suggests that these alterations could be initiating the events, driving early cellular changes that compromise genomic integrity

and contribute to the neoplastic process from its nascent stages.

In contrast, Grades 2 through Grade 5 showed no statistically significant correlations between p53 expression and tumor grade, regardless of the expression category. The absence of clear patterns across these intermediate and high grades suggests a more complex or heterogeneous role of p53 as tumors progress.

Interestingly, for Grade 2, there was a negative trend for the >50% expression group (rho = –0.1690, p = 0.0575), though this did not reach statistical significance. Similarly, Grade 4 showed a borderline negative correlation (rho = –0.1477, p = 0.0974), suggesting that in some higher-grade tumors, elevated p53 expression may inversely associate with differentiation, albeit modestly.

Our general findings suggested that p53 was expressed in all prostate carcinoma histological grades, and this was in agreement with a study that used 189 radical prostatectomy specimens. Gesztes and colleagues reported that 49.8% of prostate tumors showed focal p53 expression across all grade groups 1 to 5. Higher expression (>5%) correlated with lymphovascular invasion and poorer metastasis-free survival, indicating p53 expression is present in all grades but with prognostic significance increasing in aggressive tumors, possibly reflecting early genetic instability (Gesztes et al., 2022). Our findings and Gesztes et al's findings were also in keeping with another study in Asia which found that p53 overexpression was detected in all prostate adenocarcinoma specimens, 30 blocks (100%), emphasizing its role as a biomarker distinguishing malignant from benign prostate tissues and correlating with tumor aggressiveness (Bhat et al., 2023).

In 2025, Smith and the team found that p53 expression was present across different Gleason scores, with intensity correlating with worse clinical outcomes irrespective of the percentage of positive cells, demonstrating that p53 expression is not limited to high-grade tumors (Gesztes et al., 2025).

5. Study limitations

Cold ischemia or fixation time, however, we had no control over it since the samples were already processed.

Missing blocks that generated a selection bias however a sensitivity analysis was employed to mitigate it.

6. Conclusion

We found out that Foxp3 expression was 18.1% in all prostate carcinoma blocks analyzed, showing anon-linear, grade dependent relationship. Mild Foxp3 expression (1-10%) was positively correlated with Grade1 tumors, and grade5 tumors. This was suggesting a dynamic role for regulatory T cells in the PCa immune microenvironment, potentially contributing to early immune torelence in low grade tumors indicating immune evasion in high- grade tumors.

Conversely, p53 was expressed in 100% of the PCa blocks analyzed, most of them showing moderate staining. A significant positive correlation was observed between high p53 expression (>50%) and Grade 1 tumors. These findings suggest that p53 pathway alterations may are early events in PCa tumor genesis, even in well-differentiated lesions, challenging the notion that p53 dysregulation is solely a late stage phenomenon. The pervasive expression of p53 across all grades underscores its fundamental role in PCa development in this Pearl of African cohort (Uganda).

Abbreviations

ADT, Androgen Deprivation Therapy; c-MYC, Cellular myelocytomatosis oncogene; Foxp3, Forkhead Box P3; H&E, Hematoxylin and Eosin; LVI, Lymph Vascular Invasion; p53, Tumor protein 53; PCa ,Prostate carcinoma; TILs, Tumor Infiltrating Lymphocytes; TSC1, Tuberous Sclerosis Complex 1; UCI, Uganda Cancer Institute.

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Author contributions

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Human ethics and Consent to participate declaration

Not applicable

Data availability

The data sets supporting the findings of this study are safely stored and can be given out by the corresponding author upon reasonable request.

Competing interests

As authors, we declare no conflicting interests

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