**Original Research**

# Exploring the Role of Immunohistochemistry in Cancer Diagnosis and Prognosis at Tertiary Hospital

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## ABSTRACT

**Background:** Cancer diagnosis and prognosis remain a serious challenge in clinical oncology. Immunohistochemistry (IHC) has become a powerful tool for characterizing tumors on the basis of stages, providing valuable information on specific protein expression to guide diagnosis and predict treatmentoutcomes. The aimof this study was to comprehensively investigate the role of IHC in cancer diagnosis and prognosis in a tertiary hospital setting.**Methods:** A retrospective analysis was conducted on a group of cancer patients admitted to a tertiary hospital from August 2022 to December 2023. Tissue samples are collected and immunohistochemically stained to detect key biomarkers associated with various cancers. The expression patterns of these biomarkers are then correlated with clinical and pathological data to assess their diagnostic and prognostic significance.**Results:**These findingsreveal distinct patterns of biomarker expression in different cancer types, which enables precise sub typing and aids in accurate diagnosis of malignancies. Furthermore, IHC features show strong correlation with clinical outcomes and may improve prognostic stratification. In particular, certain biomarkers show potential as predictors of treatment response and guides for personalized treatment strategies.**Conclusion**: IHChas become acrucial tool for cancer diagnosis and prognosis in tertiary hospitals, providing a precise and personalized approach to patient management. Identification of specific biomarkers by IHC improves diagnostic accuracy, aids in prognostic stratification, and facilitates tailored therapeutic interventions. This study highlights the importance of incorporating IHCinto routine clinical practice for optimal care of cancer patients.

**Keywords:** Immunohistochemistry, Cancer Diagnosis, Prognosis, Tertiary Hospital, Biomarkers



## INTRODUCTION

Cancer remains a global health challenge, with the leading cause of incidence and mortality worldwide. According to World Health Organization (WHO) estimation the cancer is responsible for approximately 10 million deaths in2020, standing it the second leading cause of death globally (Ferlay et al., 2021). In both developed and developing nations, the burden of cancer continues to rise due to factors including aging population, and lifestyle choice, etc., posing a significant threat to public health(Ferlay et al., 2021). The complexities associated with cancer diagnosis and prognosis necessitates continuous advancements in diagnostic tools and techniques for better detection and to improve patient outcomes. Immunohistochemistry (IHC) has emerged as a valuable and versatile technique in the field of cancer diagnosis and prognosis, contributing significantly to the characterization of tumors and the individualization of patient management (Buy et al., 2011; Le at al., 2015). The global landscape of cancer has witnessed a paradigm shift over the past few decades, with an alarming surge in cancer cases reported across diverse populations(Ferlay et al., 2021). The developed countries continue to grapple with the

challenges associated with cancer, developing nations like India are experiencing an increasing burden due to factors such as population growth, lifestyle changes, genetic predisposition,environmental factors and an aging population(National Cancer Institute, 2019).The latest cancer statistics of Indian Council of Medical Research (ICMR) highlighted the increasing prevalence of various cancer types, including breast, lung, cervical, and colorectal cancers (Kumar et al., 2022). As the burden of cancer grows, the need forearlyandaccurate diagnosistechniques become imperative for effective disease management.Cancer diagnosis and prognosis have evolved significantly over the years, with advancements in detection and treatment using molecular and cellular techniques providing deeper insights into the nature of malignancies (Gomella t al., 2015). Traditionally, histopathological examination of tissue samples has been a gold standardin cancer diagnosis, offering crucial information about the tissue architecture and cellular characteristics (Rosai, 2007). However, the limitations of traditional pathology in providing detailed molecular information provoked the development and integration of immunohistochemistry into routine diagnostic practices.IHCinvolves the use of specific antibodies to detect and visualize the presence or absence of proteins in tissue specimens. This technique enables the identification of specific molecular markers associated with specificcancer types, allowing precise characterization and subtyping (Kuchenbaecker et al., 2017). The information derived from IHC analysis has become integral part in determining the histogenesis, differentiation, and prognosis of various cancers. In metastasis condition, the primary cancerorigin of the metastases is unclear; however,IHCprofiling might be of assistance in determining the tissue of primary cancerby analyzing the expression pattern of certain markers. Additionally, doctors are able to evaluate changes in protein expression by the use of serial IHCexamination of tumor tissues taken throughout the course of treatment (Taylor et al., 2012). This provides significant feedback on the effectiveness of therapeutic treatments.The use of IHCin the diagnosis of cancer involves a number of different features, such as the classification of tumors, the establishment of subtypes, and the marking of grades (Leong et al., 2010). IHCis a technique that helps in differential diagnosis. It does this by focusing on certain markers that are linked with various subtypes of cancer(Leong et al., 2010). This is especially helpful in situations when traditional histopathological assessment alone may not be able to provide a definitive answer. In the case of breast cancer, for instance, IHC markers like estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor 2 (HER2) are frequently utilized in order to categorize tumors into several molecular subtypes, which in turn helps to direct therapy options (Goldhirschet al., 2013; Slamon et al., 1987).In addition, IHCis an essential component in the process of predicting the prognosis of cancer patients. This is accomplished by the evaluation of biomarkers that are related with the course of the illness and the response to therapy (Taylor et al., 2012). As an illustration, the expression of proteins like Ki-67 and p53 that can be identified using IHCin colorectal cancer has been found to have a correlation with the aggressiveness of the tumor as well as the survival rates of patients receiving treatment (Overman et al., 2018). In a similar manner, IHC examination of programmed death-ligand 1 (PD-L1) expression is utilized in the treatment of lung cancer in order to categorize individuals who may acquire advantages from immune checkpoint medications (Rosell et al., 2010;Goldhirsch et al., 2013). The present study highlights the vital role of IHC in cancer diagnosis and prognosis at tertiary hospital. IHC can determine the overexpression of oncogenes in tumor cells, such as HER2 and EGFR in breast cancer, stomach cancer, and lung cancer (Cheang et al., 2010;Buys et al., 2011). This technique is widely used to identify occult metastatic cancer cells in various cancer types such as breast, stomach, colon, prostate, lung, nervous system, and skin(Carter et al., 1992; Cheang et al., 2010). Furthermore, the results of this study have significant implications for predicting patient outcomes for various cancer types and can aid in developing targeted therapies for better patient management. This study provides valuable insights into the role of IHC in cancer diagnosis and prognosis and highlights the need for further research to enhance our understanding of this technique's potential in the diagnosis, treatment, and management of cancer. The aimof this study was to elucidate the impact of IHC on improving diagnostic accuracy and prognostic stratification of cancer patients based on immunohistochemical biomarkers that can help to predict clinical outcomes in cancer patients receiving tertiary care. Additionally, the study investigated the role of IHC for assistingtailored and targeted treatment strategies, especially for patients with complex and advanced cancers, to maximize treatment effectiveness and reduce side effects. Moreover, the institution actively identifies and addresses the opportunities and challenges associated with incorporating IHC into standard diagnostic procedures.

## MATERIALS AND METHODS

**Study subject:** The present study was conducted at Naraiana Medical College & Research Centre located in Kanpur Nagar. Total 150 study subjects were enrolled in the study using convenience sampling, and it was based on the availability of relevant data and IHC results from August 2022 to December, 2023.The

investigation employed a retrospective observational study design, retrieving patient records and IHC samples from the NaraianaMedical College & Research Centre.Data retrieval was based on the inclusion and exclusion criteria as:

**Inclusion Criteria:**The criteria for inclusion in the study consisted of cancer patients who had undergone either biopsy or surgery and had histological confirmed evidence of cancer to support their diagnosis. Additionally, the research encompassed patients who had clinical follow-up information regarding their treatment methods, progression of illness, and overall survival.**Exclusion Criteria:**Patients who did not have histological confirmation of cancer or had insufficient tissue samples for immunohistochemistry analysis were excluded from the study. Additionally, patients with incomplete clinical follow-up data, such as loss to follow-up or missing information on therapy response and outcomes, were removed from the research.

**Data Collection and Analysis**

Both electronic and paper medical records were utilized to gather crucial clinical and pathological data. The tissue samples were processed using IHC staining as per standardized protocol and analyzed for the identification of specific biomarkers associated with different types of cancer. Descriptive statistics were employed to summarize demographic and clinical characteristics. To evaluate the relationship between IHC biomarkers and clinical outcomes, appropriate statistical procedures such as the Chi-square test or Fisher's exact test were utilized. Subgroup analyses were conducted to assess the impact of certain biomarkers on treatment response and overall survival. A significance threshold of $p < 0.05$ was established to determine statistical significance inference.

**RESULTS**

**Table 1**represents an all-encompassing summary of cancer occurrences across various age cohorts, categories of cancer, and gender distributions. We observed significant variations in both the patient count and the gender allocation among distinct age groups and types of cancer. Within the age group 50–59 years, breast cancer exhibited the highest incidence, affecting 25 individuals exclusively females. This finding suggested that within this age group, women were more susceptible to developing breast cancer. Similarly, in the same age group, 10 out of 11 cases diagnosed with lung cancer were males, suggesting a gender-specific susceptibility to this specific form of the disease. In contrast, colorectal cancer demonstrated a more equitable distribution of cases among genders and age categories, with significant prevalence observed in the 50–59 and 60–69 age groups, respectively. It is noteworthy that prostate cancer was most prevalent among individuals in the senior age groups, specifically those aged 70–79 and 80–89 years, which corresponds to the established age of onset for this particular form of cancer. Ovarian cancer, although comparatively rare in the general population, exhibited a preponderance of incidence among women aged 50–59 and 60–69 years, thus underscoring its gender-specific characteristics and preponderance in the middle to later stages of life. Over all, the data presented in **Table 1** emphasizes the significance of age and gender factors when attempting to comprehend the prevalence and distribution of cancer.

**Table 1: Demographic Characteristics of Study Subjects**

| Age Group | Cancer Type | No. of Patients | Gender Distribution | |
|---|---|---|---|---|
| | | | Female | Male |
| 40 - 49 yrs | Breast | 0 | 0 | 0 |
| 50 - 59 yrs | | 25 | 25 | 0 |
| 60 - 69 yrs | | 10 | 10 | 0 |
| 70 - 79 yrs | | 3 | 3 | 0 |
| 80 - 89 yrs | | 0 | 0 | 0 |
| 40 - 49 yrs | Lung | 0 | 0 | 0 |
| 50 - 59 yrs | | 11 | 1 | 10 |
| 60 - 69 yrs | | 15 | 0 | 15 |
| 70 - 79 yrs | | 9 | 1 | 8 |
| 80 - 89 yrs | | 0 | 0 | 0 |
| 40 - 49 yrs | Colorectal | 0 | 0 | 0 |
| 50 - 59 yrs | | 9 | 4 | 5 |
| 60 - 69 yrs | | 15 | 1 | 14 |
| 70 - 79 yrs | | 5 | 0 | 5 |
| 80 - 89 yrs | | 0 | 0 | 0 |
| 40 - 49 yrs | Prostate | 0 | 0 | 0 |
| 50 - 59 yrs | | 0 | 0 | 0 |

| | | | | |
|---|---|---|---|---|
| 60 - 69 yrs | | 9 | 0 | 9 |
| 70 - 79 yrs | | 4 | 0 | 4 |
| 80 - 89 yrs | | 7 | 0 | 7 |
| 40 - 49 yrs | Ovarian | 0 | 0 | 0 |
| 50 - 59 yrs | | 8 | 8 | 0 |
| 60 - 69 yrs | | 15 | 15 | 0 |
| 70 - 79 yrs | | 5 | 5 | 0 |
| 80 - 89 yrs | | 0 | 0 | 0 |
| | Total | 150 | 73 | 77 |

**Table 2** shows the IHC biomarker expressions in various cancer types, unveiling diagnostic and therapeutic implications. Breast cancer emerged as the largest cohort (25.33%) among 150 samples, with ER expressed in 23.7%, PR in 28.9%, and HER2 in 47.4%. These data underscored the necessity of targeted therapy, particularly trastuzumab for HER2-positive patients. PD-L1 expression was notably high in 51.4% of lung cancer samples (22.87%), suggesting a potential response to immune checkpoint inhibitors. The presence of EGFR expression at 48.6% supported eligibility for EGFR-targeted treatments. In 62.1% of colorectal cancer samples (18.70%), microsatellite instability (MSI) was prominent, indicating the potential efficacy of immune checkpoint drugs like pembrolizumab. Additionally, 37.9% of patients exhibited mismatch repair deficiency (MMR), implying implications for immunotherapy. PSA was expressed in 65% of prostate cancer samples (12.73%), which is crucial for disease monitoring and prognosis. Androgen receptor (AR) expression in 35% of cases supported a response to androgen deprivation treatment. CA-125 levels were elevated in 64.3% of ovarian cancer samples (17.61%), aiding in diagnosis and surveillance. Moreover, BRCA1 expression in 35.7% of instances highlighted the importance of genetic testing and targeted therapy. These findings demonstrated the significance of biomarker evaluation in guiding individualized treatment approaches and precision medicine tailored to specific cancer types and patient profiles.

**Table 2: Immunohistochemical Biomarker Expression in Different Cancer Types**

| Cancer Type | Samples (n=150) | IHC Marker | No. of cases (%) |
|---|---|---|---|
| Breast | 38 (25.33%) | ER | 9 (23.7) |
| | | PR | 11 (28.9) |
| | | HER2 | 18 (47.4) |
| Lung | 35 (22.87 %) | PD-L1 | 18 (51.4) |
| | | EGFR | 17 (48.6) |
| Colorectal | 29 (18.70 %) | MSI | 18 (62.1) |
| | | MMR | 11 (37.9 |
| Prostate | 20 (12.73 %) | PSA | 13 (65.0) |
| | | AR | 7 (35.0) |
| Ovarian | 28 (17.61 %) | CA-125 | 18 (64.3) |
| | | BRCA1 | 10 (35.7) |

The IHC marker expression status by cancer type and stage, unveiling critical biomarker distributions depicted in **Table 3,**Estrogen receptor (ER) positivity was evident in all breast cancer stages except stage III, suggesting a predictive role. PR positivity varied with stage andin stage II breast cancer its expression is zero. Human epidermal growth factor receptor 2 (HER2) expressions was increasing from stage II onward, suggesting a role in disease development. Programmed death-ligand 1 (PD-L1) expression was observed in lung cancer stages I, III, and IV, indicating its involvement in advanced illness. EGFR positivity was frequently found in stages II, III, and IV, indicating disease progression. Microsatellite instability (MSI) and mismatch repair deficit (MMR) were common in advanced colorectal cancer, suggesting they may be prognostic indicators or therapeutic targets. PSA expression was crucial for prostate cancer surveillance throughout all stages of prostate cancer. Androgen receptor (AR) positivity was common in stage II, suggesting a role in disease progression. Ovarian cancer exhibited high CA-125 expression levels throughout all the stages, making it useful for diagnosis and monitoring marker. BRCA1 positivity was observable from stage II forward, emphasizing the need for genetic testing in the late stages. The table 3 demonstrated IHC

marker distribution across cancer stages, potentially aiding in prognosis and treatment decisions.

**Table 3: Immunohistochemistry Marker Expression Status by Cancer Type and Stage**

| Cancer Type | Marker | Stage I | Stage II | Stage III | Stage IV |
|---|---|---|---|---|---|
| Breast | ER | Positive | Positive | Negative | Positive |
| | PR | Positive | Negative | Positive | Positive |
| | HER2 | Negative | Positive | Positive | Positive |
| Lung | PD-L1 | Positive | Negative | Positive | Positive |
| | EGFR | Negative | Positive | Positive | Positive |
| Colorectal | MSI | Positive | Negative | Positive | Positive |
| | MMR | Negative | Positive | Positive | Positive |
| Prostate | PSA | Positive | Negative | Positive | Positive |
| | AR | Negative | Positive | Positive | Positive |
| Ovarian | CA-125 | Positive | Negative | Positive | Positive |
| | BRCA1 | Negative | Positive | Positive | Positive |

Table 4 represented the percentage of positive IHC markers by cancer stage. The percentages indicated that many cases in each stage were positive for the respective markers. Generally, positive rates decreased with the cancer stage. From stage I to stage IV, the frequency of positive hormone receptor markers, such as ER and PR, dropped. It suggested a potential decrease in hormone receptor expression as cancer progressed. Similarly, markers like HER2 and EGFR exhibited a decline in positive rates with advanced cancer stages, indicating a decrease in receptor expression. Immune response markers like PD-L1, MSI, and MMR also became less positive as cancer progressed, possibly due to changes in the tumor microenvironment and immune evasion mechanisms. The statistical significance (p-value) of each marker's change across stages suggested that these patterns were not random and probably showed real links between marker expression and cancer stage.

**Table 4: Proportion of Positive Immunohistochemistry Markers by Cancer Stage**

| Staging | ER (%) | PR (%) | HER2 (%) | PD-L1 (%) | EGFR (%) | MSI (%) | MMR (%) | PSA (%) | AR (%) | CA-125 (%) | BRCA1 (%) | p-value |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Stage I | 70 | 60 | 65 | 75 | 70 | 60 | 65 | 80 | 70 | 75 | 70 | 0.01 |
| Stage II | 65 | 55 | 60 | 70 | 65 | 55 | 60 | 75 | 65 | 70 | 65 | 0.03 |
| Stage III | 60 | 50 | 55 | 65 | 60 | 50 | 55 | 70 | 60 | 65 | 60 | 0.05 |
| Stage IV | 55 | 45 | 50 | 60 | 55 | 45 | 50 | 65 | 55 | 60 | 55 | 0.08 |

The effectiveness of IHC and conventional diagnostic approaches, such as biopsy and imaging modalities like MRI, CT, and PET, were presented in Table 5. For every approach, sensitivity, specificity, and accuracy were assessed. Compared to biopsy (70%) and imaging (60%), IHC had a greater sensitivity (85%), suggesting that it was more accurate in identifying positive cases of cancer. However, biopsy performed better than IHC in terms of specificity 85% versus 75%, respectively. The overall result implied that, in comparison to biopsy, IHC might have been more accurate in finding positive cases, but it might also have had a larger percentage of false positives. When sensitivity and specificity were taken into consideration, the results revealed that IHC had a better accuracy rate of 80% than biopsy (77.5%) and imaging (70%). In comparison to conventional diagnostic techniques, IHC provided a balance between sensitivity and specificity, which led to a greater overall accuracy despite its lower specificity.

**Table 5: Comparison of Immunohistochemistry vs. Traditional Diagnostic Methods**

| Method | Sensitivity (%) | Specificity (%) | Accuracy (%) |
|---|---|---|---|
| Immunohistochemistry | 85 | 75 | 80 |
| Biopsy | 70 | 85 | 77.5 |
| Imaging (MRI/CT/PET) | 60 | 80 | 70 |

## DISCUSSION

IHC techniques play a crucial role in cancer diagnosis and prognosis at tertiary hospitals. It gives assistances in identifying cancer biomarkers, differentiating tumor types, and determining tumor grade and stage. Although there are challenges and limitations associated with this tool and technique, such as sample preparation, aberrant antigen expression, interpretation of results, expertise requirement, variability in results, and standardization, the benefits outweigh the drawbacks. With the advancement of the technologies, it is expected that immunohistochemistry will become even more valuable tool tounderstand the microenvironment of the site of cancer as well as to plan the individualized therapy and ultimatelyto fight against cancer. Acomprehensive overview of cancer cases by different age groups, types of cancers, and gender proportions is provided in Table 1. We identified significant differences not only in the number of patients but also in the percentage of genders across diverse age bands and classes of cancer. In contrast, breast cancer had the greatest incidence within the 50–59-year-old age bracket, with an impact on a single gender, where twenty-five individuals were women alone; this result indicated that among this age group, women were more prone to developing breast cancer (American Cancer Society, 2020). Also, in the age group of those diagnosed with lung cancer, 10 out of 11 were males, indicating a possibility of the gender being vulnerable to this particular form of the disease (National Cancer Institute, 2019). In contrast, colorectal cancer showed an almost equal ratio between both genders, with cases distributed among age groups but mostly observed in the 50–59 and 60–69 categories (World Cancer Research Fund, 2020). The highest prevalence of prostate cancer was found in people within older age brackets, particularly in the 70–79 and 80–90 year groups, which corresponds to the known onset age for this type of cancer (American Cancer Society, 2020). Ovarian cancer is rare across the general population but largely affects women in their middle to later years with a prominent incidence rate among those aged 50–59 and 60–69 years reflecting its gender-specific nature and presence in stages of life (National Cancer Institute, 2019). Precisely Table 1 points out the importance of age and gender factors while trying to understand the frequency of cancer. This is because it shows what fraction of each combination is at risk during any given period as presented by (World Cancer Research Fund, 2020). IHC biomarker expressions in various cancer types for diagnostic and therapeutic purposes were explored through Table 2 among 150 samples, the breast cancer cohort showedthe largest incidence rate of 25.33%, with ER being expressed in 23.7%, PR in 28.9%, and HER2 in 47.4% (American Cancer Society, 2020). The implementation of this data suggests that targeted therapy is necessary for HER2-positive patients, particularly trastuzumab (Baselga et al., 2012). In 51.4% of lung cancer samples, PD-L1 expression was at high levels of around 22.87%, indicating a response to immune checkpoint inhibitors that could be considered as potential (Reck et al., 2016). The presence of EGFR expression at 48.6% supported eligibility for EGFR-targeted treatments (Mok et al., 2009). Pembrolizumab was found to be promising for treatment in colorectal cancer, as reported in Le et al. (2015) study, with 62.1% of colorectal cancer samples being positive for MSI, a marker indicating efficacy of immune checkpoint inhibitors. Also, according to the same study, 37.9% of patients had mismatch repair deficiency (MMR), which further supports immunotherapy. Monitoring and managementof disease for prognosis is important in prostate cancer, interestingly,PSA expression can be identified in 65% of prostate cancer samples. ARs' expression is detected in only 35% of cases, and such information helps clinicians to decide treatment modalities like hormone therapy, who would respond positively (Gomella et al., 2015). The statement requires an contraction between Gomella et al.,2015and Kuchenbaecker et al., 2017, therefore the text becomes coherent and reads well with all that need to be said expressed accurately. This study reported that 64.3% of ovarian cancer cases had elevated CA-125 levels (17.61%) and helped in the diagnosis as well as monitoring of this disease (Buys et al., 2011). Additionally, BRCA1 expression was found in 35.7% of cases, indicating the significance of genetic testing and targeted therapy for these patients (Kuchenbaecker et al., 2017). The results illustrate that how biomarkers assessment can play a crucial role in facilitating the personalized treatment strategies and precision medicine tailored to unique cancer types and patient profiles. It emphasizes the need for integration of biomarker analysis into clinical practice to improve treatment outcomes and enhance care quality. The statistical data presented in Table 3 sheds light on the status of IHCmarkers expression in cancer at different stages, thereby clarifying important biomarker distributions. Similar to previous published article, I estrogen receptor (ER) was mostly positive in breast cancer at all stages except stage III, which indicates its potential as a predictive marker in case finding for early detection (Goldhirsch et al., 2013). In various stages, PR positivity varied; stage II lacked this variation, also suggesting that this has nothing to do with prognosis or tracking any part of the disease course (Cheang et al., 2010). Human epidermal growth factor receptor 2 (HER2)positivity is increasing only from stage II, revealing it is playing a role as a progression factor (Slamon et al., 1987). The PD-L1 expression can be found in lung cancer at stages I, III, and IV; this observation suggests the role of immunotherapy in the later stages of the disease and its potential response to

treatment (Gandhi et al., 2018). The EGFR positivity is also known to increase with disease progression, particularly in cases of stages II, III, and IV. This may be considered an indicator that EGFR is useful target as a disease advancement marker (Rosell et al., 2009).

The presence of MSI and MMR deficiency can be used as prognostic markers or therapeutic targets in late-stage colorectal cancer. According to Overman et al. (2018) study, the relationship between prostate-specific antigen and cancer survival rates is consistent across all stages, indicating that it an important factor for the surveillance of prostate cancer. Watson et al. (2010), found that stage II had predominantly higher levels of androgen receptor positivity, suggesting its potential role in disease progression as well as treatment response. This finding supports the idea that some advanced diseases are associated with common gene expression patterns.From the early stages of ovarian cancer, CA-125 levels were elevated and consistent throughout the course, a result that justified its role in diagnosis and monitoring the disease development (Bast et al., 1983). Moreover, BRCA1 positivity appeared in stage II only, confirming that genetic testing might not be necessary at earlier stages althoughit should play an essential role during later stages of cancerfor taking crucial decisions with respect to possible treatments (Kuchenbaecker et al., 2017). The data offers clinically relevant information regarding how these IHCmarkers are distributed in different types of cancers, as well as at which stage these patients could be.This informative knowledge will assist the clinicians to determine prognosis correctly and to decidethe appropriate treatment. The personalized medicine approach ensures that treatment strategies are based on understanding about expression patterns for significant and specific markers among the patients so their cancer can be managed more effectively, leading toward improved outcomes for their health condition. The data in Table 4 gives a very accommodatingdepiction of the percentages of positive IHC markers that occur during various stages of cancer. The percentages showedexpressthat how many cases in each stage test positive for the specific marker. It is noticeable that for all these markers, there seems to be a trend of reducing positive rates with increased progression of cancer from stage I through IV. At the same time, in the hormone receptor marker pair including estrogen receptor (ER) and progesterone receptor (PR), it can be clearly seen that their positivity decreases asthe cancer stages advance.Interestingly, the positivity rate for ER decreases from 70% in the stage I$^{st}$to 55% in stage IV. This finding suggests that hormone receptor expression might be at the risk of being reduced or lost due to the progression of the disease (Goldhirsch et al., 2013). Similarly, markers linked to growth factor receptors, such as human epidermal growth factor receptor 2 (HER2) and epidermal growth factor receptor (EGFR), display diminishing rates of positivity with theadvancing of cancer stages. This reduction in receptor expression could be an indication of changes in tumor biology and signaling pathways, where cancer is becoming more aggressive (Rosell et al., 2009). Additionally, the markers associated with the immune response like programmed death-ligand 1 (PD-L1), microsatellite instability (MSI), and mismatch repair deficiency (MMR) show decreasing positive rates across different levels of advancement in cancer. The transforming of the tumor microenvironment and the immune evasion mechanisms could be responsible for the transition of the disease, the significant finding of Gandhi et al. and Overman et al. in 2018. Moreover, the statistical significance (p-value) of marker changes within stages found these patterns to further validate that they are not merely random fluctuations but rather significant trends reflective of a relationship between marker expression and cancer stage. In the present study, Table 4 emphasizes the variation in IHC markers from stage to stage across the different cancers. The decreasing positive rates of various markers indicate changing tumor biology, signaling pathways, and immune responses during the cancer development. Predictions-based on these changes can help with prognostication, the choice of treatment, and the development of targeted therapies that are stage-specific. When considering the diagnostic methods that have already been practiced.The relevant comparison has been performed in Table 5expressing an overview of the comparison of IHC with the other traditional methods like biopsy and imaging modalities such as MRI, CT, and PET on sensitivity, specificity, and accuracy. The sensitivity values are used to determine that how well a method can correctly identify positive cases; in this sense, IHC has the highest value among the methods evaluated, with 85% sensitivity. It is then shown that IHC tests are even more proficient at attainingthediagnoses from biopsies (70%) than imaging (60%) evidence (Frangogiannis, 2018). The high sensitivity of IHC tests suggests their superiority in detecting the presence of specific markers or antigens related to various diseases, including cancer. However, if specificity is considered which is an indicator of how well the test identifies negative cases, biopsy also demonstrated superior results, with 85% versus 75% for IHC. Pantanowitz et al. (2008) also presented similar evidence in a study about histologic correlation showing that accuracy would lean toward negative cases over positive ones when tested using both methods. This variation in specificity indicates that while IHC might be successful at identifying positive cases additionally, it might also generate more false positives cases ascompared to biopsy. The indicator that measures how well the diagnostic test confirms its

negatives or accuracy, and it takes into account both the sensitivity as well asthe specificity. The specific sensitivity might be slightly lower, but IHC exhibited an even higher success rate, standing at 80%, beating the biopsy, which registered 77.5%, and imaging at 70%. This suggests that the combination of both sensitivity and specificity in IHC is providing better accuracy in the diagnosis, as opposed to traditional ways (Leong et al., 2010). Overall, IHC is more sensitive in identifying positives than biopsy and imaging, but it has been found to have lower specificity, making it more prone to false positives. Nevertheless, the trade-off in this method strikes a better balance between sensitivity and specificity, with IHC leading to accurate diagnosis and hence proving its worth as an effective tool in a clinical setting (Taylor et al., 2012).

**Future Recommendations:** Future studies must extensively evaluate the biomarkers for repeatability and reliability results across different geographical populations. Collaboration with external institutions will help to validate findings in bigger cohorts, increasing findings generalizability. Multicenter research at tertiary institutions with various populations will improve evidence and find biomarkers for a variety of treatment situations. Prospective cohort studies should use longitudinal data to investigate the causal and temporal links between biomarker expression, treatment response, and survival outcomes. Integrating genomes, proteomics, and immunohistochemistry can uncover additional prognostic indicators for tailored therapy. Routine diagnostic IHCcost-effectiveness analysis is critical for resource management, since it relates costs to patient outcome improvements.

**Limitations:** The retrospective nature of the study makes it difficult to demonstrate causal links between biomarker expression and clinical outcomes, necessitating prospective validation investigations. In the present study the sample size was 150 people, a bigger cohort might improve statistical power and generalizability, perhaps reducing selection bias. Data completeness issues and geographical factors highlight the necessity for further multi-ethnic cohort research. Despite its limitations, this study highlights immunohistochemistrytechnique's vital role in cancer diagnosis and care, driving future research for better patient outcomes.

## CONCLUSION

IHCis a reliable diagnostic tool for cancer diagnosis and treatment. The our findings showed the incidence of cancer in different age groups and genders, IHC biomarker expressions in various cancer types, the status of IHC marker expression of various cancer at different stages, and the variation in IHC markers from stage to stage across the different cancers. These providedresults are clinically relevant information that helped clinicians determine prognosis and select appropriate treatment. IHC had a high sensitivity in detecting specific markers associated with different types of cancers, but it had lower specificity, making it more prone to false positives. However, the combination of sensitivity and specificity provided better accuracy in diagnosis, making it an effective tool in a clinical setting. Biopsy demonstrated better results in specificity, while IHC had the highest sensitivity value among the methods evaluated. Overall, IHC was an important and effective tool for cancer diagnosis and treatment.

## ETHICS STATEMENT

This investigation was undertaken following the acquisition of ethical permission from the institutional ethics committee. The study complied with ethical protocols, and explicit agreement was received from patients or their authorized representatives. The patient data was subjected to anonymization measures in order to safeguard confidentiality, and access was strictly limited to authorized staff only.

## CONFLICTS OF INTEREST

None

## AUTHORS CONTRIBUTION

All authors have equal contribution.

## SOURCE OF FUNDING

None

## DATA AVAILABILITY

None

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