RESEARCH ARTICLE

FGFR2 as A Prognostic and Predictive Marker in Colorectal Adenocarcinoma Based on TILs Grade

Andi Marcha Rianti[1], Muhammad Husni Cangara[1,2], Amalia Yamin[1,3], Haslindah Dahlan[1,2], Muhammad Riza Ilyasa[4], Upik Anderiani Miskad[1,2,*]

[1]Department of Anatomical Pathology, Faculty of Medicine, Universitas Hasanuddin, Jl. Perintis Kemerdekaan KM. 10, Makassar 90245, Indonesia
[2]Anatomical Pathology Laboratory, Universitas Hasanuddin Hospital, Jl. Perintis Kemerdekaan KM. 10, Makassar 90245, Indonesia
[3]Anatomical Pathology Laboratory, Dr. Wahidin Sudirohusodo Hospital, Jl. Perintis Kemerdekaan KM. 10, Makassar 90245, Indonesia
[4]Undergraduate Program of Medical Study, Faculty of Medicine, Universitas Hasanuddin, Jl. Perintis Kemerdekaan KM. 10, Makassar 90245, Indonesia

*Corresponding author. Email: upik.miskad@med.unhas.ac.id

Received date: Jan 6, 2025; Revised date: Mar 9, 2025; Accepted date: Mar 11, 2025

Abstract

BACKGROUND: Colorectal cancer remains a serious health problem due to its high incidence and mortality rate each year. Histopathological grades and tumor-infiltrating lymphocytes (TILs) are associated with patient's outcome. Fibroblast growth factor receptor 2 (FGFR2) overexpression is correlated with a worse prognosis in colorectal adenocarcinoma patients. Unfortunately, there are not many studies investigating the relationship between FGFR2 with histopathological grade and TILs in Indonesia. This study was conducted to analyze the correlation between FGFR2 expression with histopathological grade and TILs grade in colorectal adenocarcinoma.

METHODS: Immunohistochemistry examination using FGFR2 rabbit polyclonal antibody was performed on 94 paraffin-embedded colorectal adenocarcinoma blocks and its expression was examined using a light microscope. The relationship between FGFR2 expression with histopathological grade and TILs grade in colorectal adenocarcinoma was statistically analyzed.

RESULTS: Of the 94 samples examined, low grade adenocarcinoma was more common (n=76), of which 60.5% showed high FGFR2 expression. While in high grade adenocarcinoma, 83.3% of the samples showed high FGFR2 expression. In low grade TILs (n=30), 80% showed strong FGFR2 expression. While in high grade TILs (n=17), 64.7% showed weak FGFR2 expression. Based on statistical analysis, there was a significant correlation between FGFR2 expression and TILs grade (p=0.008). However, there was no significant association with histopathological grade (p=0.127).

CONCLUSION: The significant correlation between FGFR2 expression and TILs grade suggests that FGFR2 may be used as a prognostic and predictive marker in colorectal adenocarcinoma.

KEYWORDS: FGFR2, Colorectal adenocarcinoma, TILs, Histopathological grade

Indones Biomed J. 2025; 17(2): 188-96

Introduction

Colorectal cancer is one of the most prevalent malignancies, whose incidence is increasing each year. GLOBOCAN data from 2022 revealed an increase in the incidence of colorectal cancer compared to previous data from 2020. (1,2) It is due to several factors, including lifestyle, dietary factors, family and personal history of previous diseases, environmental factors, and an imbalance of gut flora.(3–5) There are several pathogenesis of colorectal carcinoma. These include chromosomal instability (CIN), microsatellite instability (MSI), and CpG island methylator phenotype (CIMP).(6–8) Chromosomal instability represents the most prevalent pathogenesis of colorectal cancer, with recurrent mutations in the adenomatous polyposis coli (APC), Kirsten

rat sarcoma viral oncogene homolog (KRAS), and tumor protein p53 (TP53) genes being a notable feature.(9)

Despite advances in diagnosis and treatment, the prognosis for patients with colorectal cancer remains variable.(10,11) It depends on several factors, including the stage of the disease when the patient is first diagnosed, histopathological grade, and the anti-tumor immune response.(12,13)

Fibroblast growth factor receptor 2 (FGFR2) is a tyrosine kinase receptor consisting of 3 domains. They are the extracellular, transmembrane, and intracellular domain.(14,15) The extracellular domain comprises three immunoglobulin-like domains, designated D1, D2, and D3. This is the point at which FGFR2 binds to its ligand, fibroblast growth factor (FGF).(14) The binding between FGFR2 and FGF will cause FGFR2 activation. Activation of this receptor will result in the initiation of a cascade of signaling pathways, including the Ras/mitogen-activated protein kinase (RAS/MAPK), the extracellular signal-regulated kinase/protein kinase B (ERK/AKT), the phospholipase C gamma (PLC γ) and the Janus kinase/signal transducers and activators of transcription (JAK-STAT) pathway. These signaling pathways ultimately lead to the processes of proliferation, differentiation, survival, migration, and angiogenesis.(16–18) Point mutations, amplifications, gene fusions, and gene rearrangements can lead to FGFR2 overexpression. FGFR2 overexpression plays a role in the development of various malignancies including gastric cancer, lung cancer, urothelial cancer, cholangiocarcinoma, colorectal cancer, and others.(17,19) FGFR2 overexpression is associated with a worse prognosis in colorectal adenocarcinoma patients.(20) In colorectal cancer, FGFR2 and some other growth factors, such as epidermal growth factor receptor (EGFR), are known for their contribution to tumor growth, but the key difference is that FGFR2 is less frequently mutated or amplified in colorectal cancer compared to EGFR, indicating that it might be a more specific marker for colorectal cancer.

In accordance with the World Health Organization (WHO) Classification System for Colorectal Adenocarcinoma 2019, the histopathological grade is based on the degree of tumor cells differentiation and is divided into two grades: low grade and high grade. Low grade if the glandular formation of tumor cells is 50% or more and high grade if the glandular formation of tumor cells is less than 50%.(6) The histopathological grade of a tumor is closely related to the prognosis of the patient, with a higher grade tending to indicate a worse prognosis.(6)

Tumor-infiltrating lymphocytes (TILs), are defined as lymphocytes that can be found in and around cancer cells and play a role in tumor-fighting defense mechanisms.(21,22) TIL is one of the prognostic markers in colorectal adenocarcinoma.(23) High TILs counts are associated with better prognosis in colorectal adenocarcinoma, whereas low TILs counts are associated with a less favourable prognosis.(23,24) Many recent studies have concentrated on examining TILs specifically in colorectal adenocarcinoma. Not only to determine the prognosis of patients but also closely related to the development of immunotherapy which is promising in colorectal cancer treatment in addition to other conventional therapies.(25)

There are many studies evaluating TILs in different types of cancer using various methods, however studies using the method suggested by the International TILs Working Group in breast cancer 2014 are rarely used before. This method allows to assess the prognostic utility of TILs on Hematoxylin-Eosin (HE) stained sections in colorectal cancer, similarly to the approach employed in breast cancer. Moreover, it standardizes the methodology for evaluating TILs.(26) The aim of this study was to ascertain the correlation between FGFR2 expression and histopathological grade, as well as TILs grade, in colorectal adenocarcinoma.

Methods

Study Design and Data Collection

This study was an analytical observation study with cross-sectional design to determine FGFR2 expression in primary tumor preparations of colon and/or rectal adenocarcinoma. The number of samples examined was 94 samples of paraffin block preparations from resection surgery of colon and/or rectal tumor tissue that met the inclusion criteria, and were taken from January 2021 to May 2024 from the Anatomical Pathology Laboratory of Dr. Wahidin Sudirohusodo Hospital, Universitas Hasanuddin Hospital, and Makassar Pathology Diagnostic Centre. The Ethics Committee of the Faculty of Medicine has approved this study (Protocol #UH24060469 – Registry No. 607/UN4.6.4.5.31/PP36/2024).

Subjects' Inclusion and Exclusion Criteria

Paraffin blocks preparations of colon or rectal tumors with lymph nodes, from colon or rectal tissue resection surgery with examination results showing the diagnosis of colon or rectal adenocarcinoma with or without lymphovascular invasion and with or without lymph node metastasis, through

HE staining that has been examined by two anatomical pathologists. Meanwhile, the paraffin block preparations of colon or rectal tumors that are depleted or damaged during processing were excluded from the study.

Evaluation of Histopathological Grade and TILs Grade

The histopathological grade was the degree of tumor cell differentiation, based on glandular formation, divided into a low grade (if glandular formation is ≥50%) and a high grade (if glandular formation is <50%).(6) TILs are the presence of lymphocytes among the tumor stroma which can be assessed by histopathological examination. The percentage score of TILs was categorized into three groups based on the International TILs Working Group (ITWG): Low (0-10%), Intermediate (15-50%), and High (55-100%).(26)

Based on Tumor, Node, and Metastasis (TNM) staging of digestive system tumors in the 5th edition of the WHO Classification, the depth of invasion is measured based on how deeply the tumor cells invade the lining of the colon or rectum: pTis if the tumor cells invade the lamina propria; pT1 if the tumor cells invade up to the submucosa; pT2 if the tumor cells invade up to the muscularis propria; pT3 if the tumor cells invade into the subserous layer or into non-peritonealized pericolic or perirectal tissues; and pT4 if the tumor cells directly invade other organs or structures and/or perforates the visceral peritoneum.(6)

FGFR2 Immunohistochemistry (IHC) Examination

Slides for IHC examination were prepared from paraffin blocks, which were cut to a thickness of 3 µm, then deparaffinised. The blocks were stained with FGFR2 rabbit polyclonal antibody (Cat. No. E-AB-60590; Elabscience, Wuhan, China), using a dilution of 1:200. FGFR2 has membranous and cytoplasm reactivity, and its expression was assessed using a light microscope at 200x magnification. The expression was analyzed by two pathologists who were not informed of the subjects's clinical data.

FGFR2 expression was obtained by multiplying the intensity score and proportion score which will result in a total immunostaining score (TIS) (score 1-12). The intensity score is divided into 0: not stained, +1; weakly and faintly stained, +2: moderately stained, +3: strongly stained. The proportion score 0: Stained 0-5%; 1: Stained 6-25%; 2: Stained 26-50%; 3: stained 51-75%; 4: stained 76-100%. (27) In the double-blind method, five high visual fields were randomly selected to read the results sequentially, and the average of the five fields scores was used as the final score. FGFR2 expression was classified as weak if TIS<6, and strong if TIS≥6.

Statistical Analysis

Statistical analysis was performed using SPSS 27 software (IBM Corporation, Armonk, NY, USA). The data were presented univariately, as frequencies and distribution tables of clinicopathological characteristics, and bivariately to analyze the association of FGFR2 expression with histopathological grade, TILs grade, and other clinicopathological data. Chi-square, Mann-Whitney, and Kruskal-Wallis tests were used to analyze the data, with a $p<0.05$ indicating a statistically significant result.

Results

Characteristics of Colorectal Adenocarcinoma Subjects

The distribution of colorectal adenocarcinoma subjects according to age, sex, tumor location, histopathological grade, TILs grade, depth of invasion and FGFR2 expression were presented in Table 1. From 94 subjetcs' samples examined, based on age, subjects with age categories under 50 years were 28.7%, while subjects above the age of 50 years were 71.3%. Based on gender, colorectal adenocarcinoma was more common in males (55.3%) than females (44.7%).

The most common tumor location was the distal colon (37.2%), followed by the rectum (29.8%) and proximal colon (25.5%). For histopathological grade, low-grade colorectal adenocarcinoma was found in 76 subjects (80.9%), while high-grade colorectal adenocarcinoma was found in 18 subjects (19.1%). For the depth of invasion, most subjects were belong in pT2 (55.3%), followed by pT3 (41.5%) and pT1 (3.2%) (Figure 1). Based on the grading of TILs, intermediate grade TILs were found in 50% of subjects, followed by low grade (31.9%) and high grade (18.1%) TILs. FGFR2 with weak expression was found in 33 subjects (35.1%) and strong expression was found in 61 subjects (64.9%).

FGFR2 Expression was Correlated with TILs Grade in Colorectal Adenocarcinoma

Table 2 showed that age, sex, tumor location, and depth of invasion did not have any significant association with FGFR2 expression ($p>0.05$). In 76 low grade colorectal adenocarcinoma subjects, weak FGFR2 expression was found in 30 subjects (39.5%) and strong FGFR2 expression was found in 46 subjects (60.5%). In 18 high grade colorectal adenocarcinoma subjects, weak FGFR2 expression was found in 3 subjects (16.7%) and strong FGFR2 expression was found in 15 subjects (83.3%).

Table 1. Characteristics of subjects.

Characteristics	n (%)
Age	
≤50 years	27 (28.7)
> 50 years	67 (71.3)
Gender	
Male	52 (55.3)
Female	42 (44.7)
Tumor Location	
Proximal	24 (25.5)
Distal	35 (37.2)
Rectum	28 (29.8)
Rectosigmoid	7 (7.4)
Histopathological Grade	
Low	76 (80.9)
High	18 (19.1)
TILs Grade	
Low	30 (31.9)
Intermediate	47 (50.0)
High	17 (18.1)
Depth of Invasion (pT)	
pTis	0 (0)
pT1	3 (3.2)
pT2	52 (55.3)
pT3	39 (41.5)
pT4	0 (0)
FGFR2 Expression	
Weak	33 (35.1)
Strong	61 (64.9)
Total	**94 (100)**

In low grade TILs, weak FGFR2 expression was found in 6 subjects (20.0%) and strong FGFR2 expression was found in 24 subjects (80.0), in intermediate grade TILs, weak FGFR2 expression was found in 16 subjects (34.0%) and strong FGFR2 expression was found in 31 subjects (66.0%). And in high grade TILs, weak FGFR2 expression was found in 11 subjects (64.7%) and strong FGFR2 expression was found in 6 subjects (35.3%).

The relationship between FGFR2 expression with histopathological grade and TILs grade was evaluated. The results demonstrated that there was no significant relationship between FGFR2 expression and histopathological grade (p=0.127 and p=0.115). A significant association was observed between FGFR2 expression and TILs grade, with p=0.008 and p=0.039, respectively.

FGFR2 Intensity Score and Grading of TILs

In Figure 2, FGFR2 intensity score could be assessed in the membrane and cytoplasm of tumor cells, where tumor cells with strong FGFR2 intensity score (+3) are stained dark brown, moderate intensity score (+2) is stained light brown/yellowish, and weak intensity score (+1) is stained weakly and faintly. A score of negative (0) indicates no staining at all. In this study, 47 subjects showed strong intensity, 42 subjects showed moderate intensity, and 4 subjects showed weak intensity and 1 subject showed no color (negative).

The TILs grade in colorectal adenocarcinoma was assessed in the stroma surrounding the tumor, where 30 subjects showed low TILs, 47 subjects showed intermediate TILs, and 17 subjects showed high TILs (Figure 3).

FGFR2 Expression was Significantly Correlated with TILs Grade

Spearman rank coefficient test was performed to examine the correlation between FGFR2 expression and TILs using

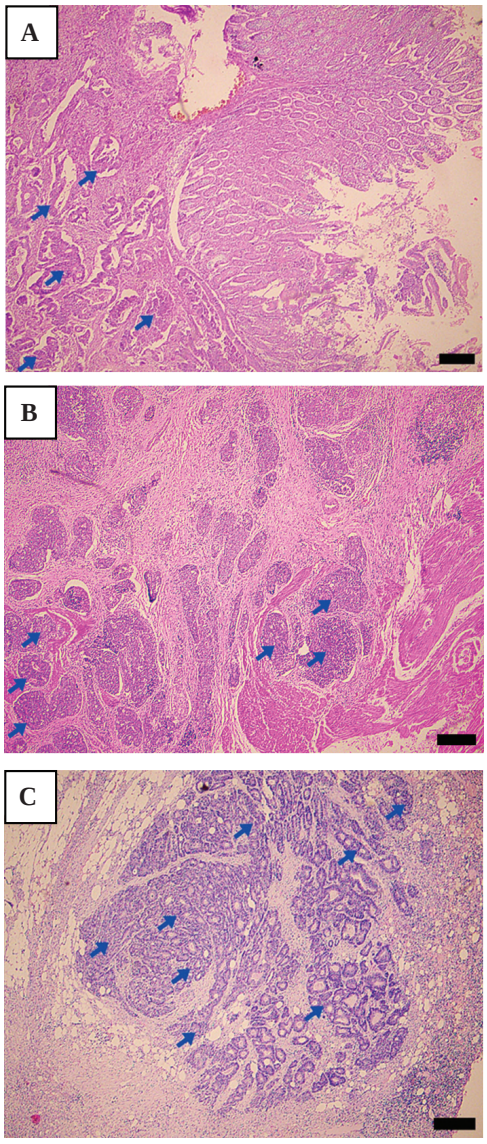

Figure 1. Depth of invasion in colorectal adenocarcinoma. A: pT1 ; B: pT2; C:pT3. Blue arrows: the depth of invasion. Black bar: 20 µm.

Table 2. Relationship of FGFR2 expression with histopathological grade and TILs grade.

Characteristics	n	FGFR2 Expression [n (%)]		*p*-value[a]	TIS FGFR2 [Mean±SD]	*p*-value
		Weak	Strong			
Age (yrs)						
<=50	27	10 (37.0)	17 (63.0)	0.992	5.74±2.23	0.961[b]
> 50	67	23 (34.3)	44 (65.7)		5.75±2.13	
Sex						
Male	52	20 (38.5)	32 (61.5)	0.589	5.56±2.06	0.395[b]
Female	42	13 (31.0)	29 (69.0)		5.98±2.25	
Tumor Location						
Proximal	24	8 (33.3)	16(66.7)	0.942	6.00±2.13	0.900[c]
Distal	35	12 (34.3)	23 (65.7)		5.80±2.31	
Rectum	28	11 (39.3)	17 (60.7)		5.50±2.03	
Rectosigmoid	7	2 (28.6)	5 (71.4)		5.57±2.15	
Histopathological Grade						
Low	76	30 (39.5)	46 (60.5)	0.127	5.59±2.20	0.115[b]
High	18	3 (16.7)	15 (83.3)		6.39±1.79	
TILs Grade						
Low	30	6 (20.0)	24 (80.0)	0.008*	6.17±1.74	0.039*[c]
Intermediate	47	16(34.0)	31 (66.0)		5.85±2.30	
High	17	11(64.7)	6 (35.3)		4.71±2.11	
Depth of Invasion (pT)						
pT1	3	2 (66.7)	1 (33.3)	0.304	4.67±1.15	0.200[c]
pT2	52	20 (38.5)	32(61.5)		5.48±1.95	
pT3	39	11 (28.2)	28 (71.8)		6.18±2.39	

[a]Analyzed with Chi-square; [b]Analyzed with Mann-Whitney; [c]Analyzed with Kruskal-Wallis; *Significant if $p<0.05$.

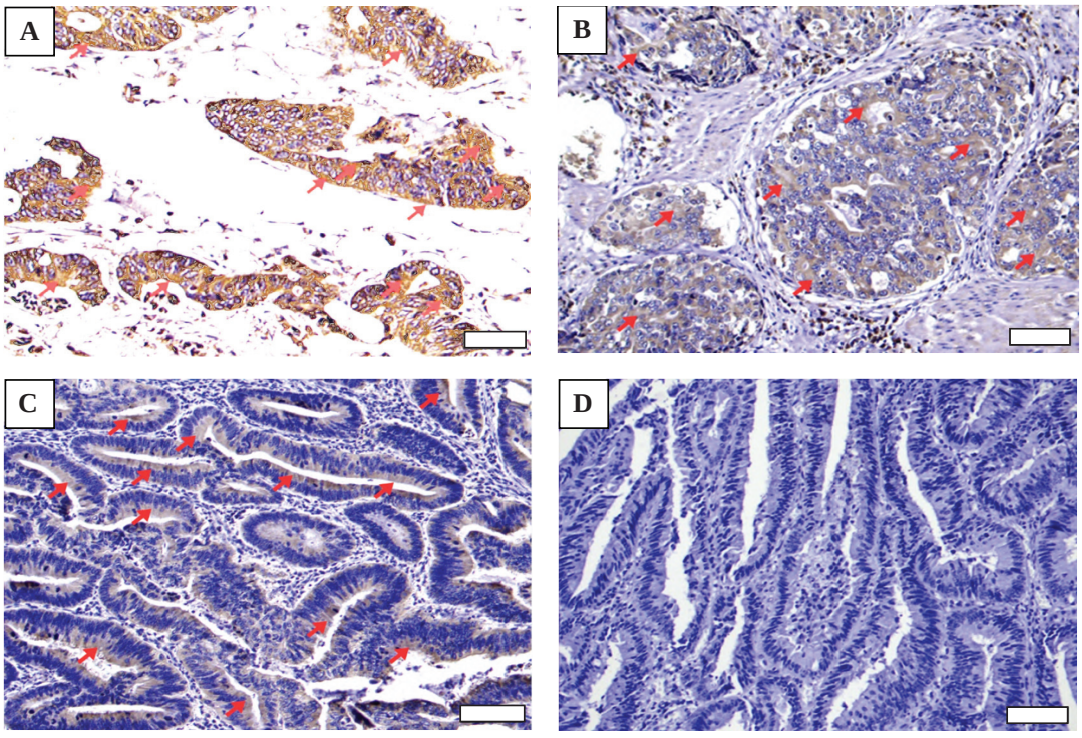

Figure 2. FGFR2 intensity in colorectal adenocarcinoma. A: Strong (+3); B: Moderate (+2); C: Weak (+1); D: Negative (0). Red arrows: FGFR2 Expression in membrane and cytoplasm. White bar: 100 µm.

quantitative data from the FGFR2 TIS and TILs score. The results demonstrated a negative correlation between FGFR2 TIS and TILs score (r=-0.270). It means that the higher the FGFR2 expression, the lower the TILs grade, with p=0.008.

Discussion

The study about FGFR2 has been growing rapidly lately. This is because FGFR2 is proven to play an important role not only in various normal body mechanisms but also in the process of oncogenesis in various malignancies including colorectal cancer.(14) Furthermore, currently, several FGFR inhibitors have been tested in a clinical trial phase, some have even been approved by the Food and Drug Administration (FDA) as targeted therapy in cancer, making research on FGFR even more interesting.(17)

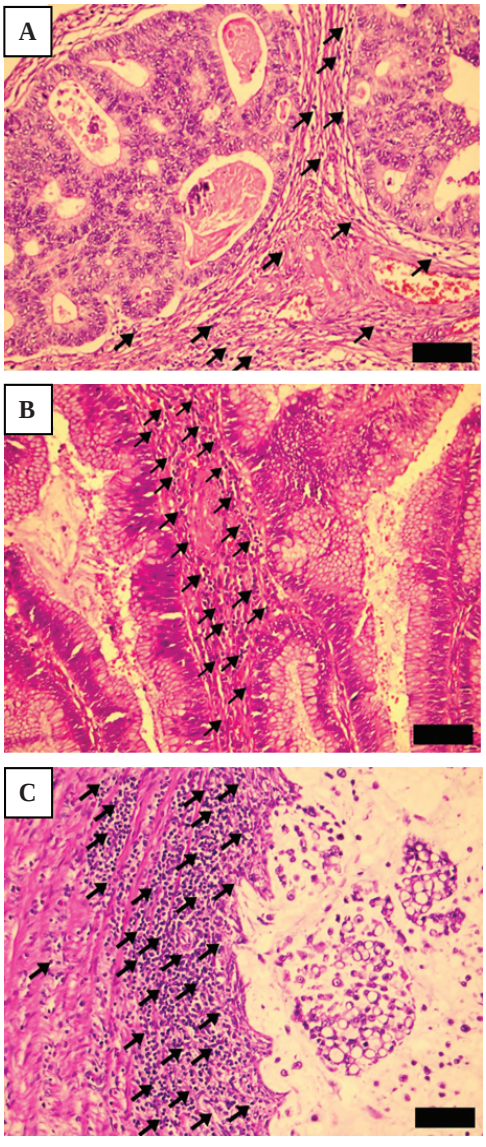

Figure 3. TILs grade in colorectal adenocarcinoma. A: Low (score 1) ; B: Intermediate (score 2); C: High (score 3). (Black arrows: TILs. Black bar: 100 µm.

TILs are lymphocytes found among tumor cells, including Cytotoxic T cells ($CD8^+$ T cells), Helper T cells ($CD4^+$ T cells), B cells, natural killer cells (NK cells), regulatory T cells (Tregs).(21) $CD8^+$ cytotoxic T cells are a subset of T cells that play a crucial role in immune responses. They are the primary agents in fighting intracellular pathogens, including tumor cells. These cells directly eliminate cancer cells by recognizing specific antigens on the surface of cancer cells.(21) Some of the substances produced by $CD8^+$ T cells include perforin, granzyme, granulysin, Fas ligand, and tumor necrosis factor α (TNF-α), which are responsible for the process of cytotoxicity.(21) $CD4^+$ helper T lymphocytes assist in organizing the immune response by activating cytotoxic T cells and B cells.(21) These cells then develop into Th1, Th2, Th17 subsets, follicular helper T cells (Tfh), and regulatory T cells (Treg).(21)

NK cells represent a component of innate immunity. They are capable of playing a role in the early stages of cancer development, whereby they can kill cancer cells without the need to recognize specific antigens.(21) B cells represent a component of the body's adaptive humoral immunity, whereby they produce antibodies that can target pathogens. However, their role in killing cancer cells remains a topic of contention and is currently undergoing further investigation.(21) The presence of TILs is closely associated with the host's immune response against the tumor.(23) A number of studies have demonstrated that TILs are an important prognostic factor in patients with colorectal adenocarcinoma.(28,29)

In this study, we analyzed the relationship between FGFR2 expression with histopathological grade, depth of invasion and TILs grade. Based on Chi-square test and Mann-Whitney test, there was no significant association between FGFR2 expression and histopathological grade and depth of invasion. However, there is a trend that the higher the histopathological grading and the deeper the invasion of tumor cells, the higher the FGFR expression.

This result is different from previous studies that showed a significant relationship between FGFR2 expression with histopathologic grade and depth of invasion. The difference in results of this study with the results of previous studies can be caused by the presence of intratumor heterogeneity and dynamic tumor microenvironment factors. Intratumor heterogeneity refers to the genetic, phenotypic, and functional diversity among cancer cells

within a single tumor. This diversity makes tumors highly complex and have behaviors that are difficult to predict. Tumor microenvironments are the highly complex and dynamic surroundings of cancer cells. This environment consists of various interacting components including blood vessels, immune cells, stromal cells, extracellular matrix, and the presence of other proteins that interact and influence each other.(30)

Based on TILs grade, Chi-square test and Kruskal-Wallis test showed a statistically significant correlation between FGFR2 expression and TILs grade. This can be explained by the fact that FGFR2 overexpression will activate a number of different signaling pathways, including the RAS/MAPK, the phosphatidylinositol 3-kinase/protein kinase B (PI3K/AKT), the JAK/STAT3 and the PLC γ pathway.(13,17,18)

The increased of programmed cell death ligand 1 (PD-L1) expression on the tumor cell membrane will then bind to its receptor, programmed cell death protein 1 (PD-1), which is located on the surface of T cells.(31–33) Under normal circumstances, the binding between PD-L1 and PD-1 represents a physiological mechanism that serves to maintain the homeostasis of the immune system. Their bonding reduces autoimmune attacks into the body itself.(34) However, tumor cells exploit this mechanism for their own benefit, allowing them to evade the host immune response and continue to grow, survive, invade and metastasis.(31,34)

The binding of PD-L1 to PD-1 will result in the inhibition of T cell activation, a reduction in proliferation and survival of T cell, and a decrease in the secretion of cytotoxic substances.(35) PD-L1 binding to PD-1 on T cells triggers a series of intracellular events that ultimately inhibit T cell activation. When PD-L1 binds to PD-1, phosphorylation of tyrosine residues within the immunoreceptor tyrosine-based switch motif (ITSM) in the intracellular domain of PD-1 occurs.(36) This phosphorylation creates a binding site for the protein tyrosine phosphatases Src homology region 2 domain-containing phosphatase (SHP)-1 and SHP-2. Subsequently, SHP-1 and SHP-2 bind to the phosphorylated PD-1.(34,36) SHP-1 and SHP-2 dephosphorylate key molecules in the T cell receptor (TCR) and CD28 co-stimulator signaling pathways including Zeta chain of TCR, CD3, zeta-chain-associated protein kinase 70 (ZAP-70), PI3K.(34,36)

Dephosphorylation of these molecules inhibits the activation of downstream signaling pathways that are crucial for T cell activation, including the PI3K/AKT pathway, MAPK pathway, and PLC γ pathway. The inhibition of these signaling pathways ultimately decreases the activation of transcription factors such as nuclear factor of activated T cells (NFAT), activator protein-1 (AP-1), and nuclear factor kappa-B (NF-κB). This results in a reduction in the production of essential cytokines such as interleukin (IL)-2 and interferon (IFN)-γ, as well as effector molecules such as perforin and granzyme B that are required for T cell cytotoxic function. In addition, PD-1 signaling can also increase the expression of pro-apoptotic proteins such as Bim, which contributes to T cell death.(36) The binding of PD-L1 to PD1 has the ability to inhibit T cell activation and cause T cell apoptosis.(25) As a consequence, the number of T cells will be reduced. Histopathologically, this can be observed as a low number of TILs.(28)

Therefore, this mechanism can explain how high FGFR2 expression can lead to low TILs grading, which is statistically shown as a significant relationship between FGFR2 expression and TILs grading where there is a negative relationship between the two. High FGFR2 expression correlate with low TILs grade, and low FGFR2 expression correlate with high TILs grade. To prove this hypothesis, the Spearman correlation test was conducted, where it was found that there was a significant negative correlation ($r=-0.270$) between total FGFR2 immunostaining score (TIS) and TILs score with $p=0.008$. Based on the results, we conclude that FGFR2 expression can be used as a prognostic and predictive biomarker in terms of its ability to influence TILs grade in colorectal adenocarcinoma.

In this study, TILs as a mechanism of immune response to tumors was only evaluated by HE examination. Future studies should add other markers involved in the mechanism of immune response to tumors using IHC examination. These proteins include PD-L1, PD-1, $CD8^+$, $CD4^+$, and others, in order to gain a more comprehensive understanding of the host immune response in influencing tumor development, invasion and metastasis in colorectal adenocarcinoma. Analysing the correlation between tyrosine kinase inhibitors and immune checkpoints inhibitor (ICI) might also necessary to obtain more comprehensive mechanism related to FGFR2 and its role in cancer treatment.

Conclusion

There is a significant relationship between FGFR2 expression and TILs grade where high FGFR2 expression shows low TILs grade and conversely weak FGFR2 expression correlated with high TILs grade. However, there

was no significant relationship between FGFR2 expression and histopathological grade. The significant association between FGFR2 expression and TILs grade suggests that FGFR2 may be used as a prognostic and predictive marker in colorectal adenocarcinoma.

Acknowledgments

We would like to thank the Department of Anatomical Pathology, Faculty of Medicine, Hasanuddin University, Indonesia and all of our teachers for their guidance.

Authors Contribution

AMR and UAM conceptualized and planning the study. AMR, UAM, MHC, AY, HD, and MRI were contributed in the methodology, data collection and analysis, and drafting of the manuscript. All authors participated to manuscript revisions and approved the final version of the manuscript.

References

1. Bray F, Laversanne M, Sung H, Ferlay J, Siegel RL, Soerjomataram I, *et al.* Global cancer statistics 2022: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2024; 74(3): 229–63.
2. Harjanti DA, Murtono C, Suryawinata K, Halim A, Wiryokusuma MF, Djitro TB. John Cunningham virus T-antigen expression on mild and severe dysplasia adenomatous polyp, low and high grade adenocarcinoma of the colon. Mol Cell Biomed Sci. 2019; 3(1): 7–12.
3. Roshandel G, Ghasemi-Kebria F, Malekzadeh R. Colorectal cancer: Epidemiology, risk factors, and prevention. Cancers. 2024; 16(8): 1530. doi: 10.3390/cancers16081530.
4. Miskad UA, Akil F, Cangara MH, Dahlan H, Hutasoit GA, Ilyasa MR. Snail expression is positively correlated with depth of invasion in colorectal carcinoma. Indones Biomed J. 2024; 16(6): 553–9.
5. Tedjasaputra TR, Hatta M, Massi MN, Natzir R, Patellongi I, Simadibrata M, *et al.* Risk assessment in hereditary colorectal cancer family by using APC and MSH2 mRNA gene expression and Bayesian analysis. Indones Biomed J. 2020; 12(4): 368–75.
6. Nagtegaal ID, Arends MJ, Salto Tellez M. Tumours of the colon and rectum. In: WHO Classification of Tumours Digestive System Tumours. 5th ed. Geneva: World Health Organization; 2019.
7. Odze RDr, Goldblum JR. Epithelial neoplasms of the large intestine. In: Odze and Goldblum Surgical Pathology of the GI Tract, Liver, Biliary Tract, and Pancreas. 3rd ed. Philadelphia: Elsevier Saunders; 2014.
8. Rhee YY, Kim KJ, Kang GH. CpG island methylator phenotype-high colorectal cancers and their prognostic implications and relationships with the serrated neoplasia pathway. Gut Liver. 2017; 11(1): 38–46.
9. Kang S, Na Y, Joung SY, Lee SI, Oh SC, Min BW. The significance of microsatellite instability in colorectal cancer after controlling for clinicopathological factors. Medicine. 2018; 97(9): e0019. doi: 10.1097/MD.0000000000010019.
10. Theodora I, Sudiana IK, Budipramana VS, Erwin F, Dewi S, Novita BD. Tumor differentiation is correlated with estrogen receptor beta (ERβ) expression but not with interleukin-6 (IL-6) expression in colorectal carcinoma. Indones Biomed J. 2024; 16(5): 481–6.
11. Wulandari F, Hanifa M. Integrative bioinformatics reveals the lactate dehydrogenase B (LDHB) significance in colon adenocarcinoma. Mol Cell Biomed Sci. 2023; 7(2): 98–108.
12. Langrand-Escure J, Diao P, Garcia MA, Wang G, Guy JB, Espenel S, *et al.* Outcome and prognostic factors in 593 non-metastatic rectal cancer patients: a mono-institutional survey. Sci Rep. 2018; 8(1): 10708. doi: 10.1038/s41598-018-29040-2.
13. Li CJ, Zhang X, Fan GW. Updates in colorectal cancer stem cell research. J Can Res Ther. 2014; 10(8): 233–9.
14. Tiong KH, Mah LY, Leong CO. Functional roles of fibroblast growth factor receptors (FGFRs) signaling in human cancers. Apoptosis. 2013; 18(12): 1447–68.
15. Dai S, Zhou Z, Chen Z, Xu G, Chen Y. Fibroblast growth factor receptors (FGFRs): Structures and small molecule inhibitors. Cells. 2019; 8(6): 614. doi: 10.3390/cells8060614.
16. Zhou WY, Zheng H, Du XL, Yang JL, Zhou WY, Zheng H, *et al.* Characterization of FGFR signaling pathway as therapeutic targets for sarcoma patients. Cancer Biol Med. 2016; 13(2): 260–8.
17. Ratti M, Orlandi E, Hahne JC, Vecchia S, Citterio C, Anselmi E, *et al.* Targeting FGFR pathways in gastrointestinal cancers: New Frontiers of Treatment. Biomedicines. 2023; 11(10): 2650. doi: 10.3390/biomedicines11102650.
18. Chae YK, Ranganath K, Hammerman PS, Vaklavas C, Mohindra N, Kalyan A, *et al.* Inhibition of the fibroblast growth factor receptor (FGFR) pathway: the current landscape and barriers to clinical application. Oncotarget. 2017; 8(9): 16052–74.
19. Pacini L, Jenks AD, Lima NC, Huang PH. Targeting the fibroblast growth factor receptor (FGFR) family in lung cancer. Cells. 2021; 10(5): 1154. doi: 10.3390/cells10051154.
20. Porta R, Borea R, Coelho A, Khan S, Araújo A, Reclusa P, *et al.* FGFR a promising druggable target in cancer: Molecular biology and new drugs. Crit Rev Oncol Hematol. 2017; 113: 256–67.
21. Bai Z, Zhou Y, Ye Z, Xiong J, Lan H, Wang F. Tumor-infiltrating lymphocytes in colorectal cancer: The fundamental indication and application on immunotherapy. Front Immunol. 2022; 12: 808964. doi: 10.3389/fimmu.2021.808964.
22. Lin B, Du L, Li H, Zhu X, Cui L, Li X. Tumor-infiltrating lymphocytes: Warriors fight against tumors powerfully. Biomed Pharmacother. 2020: 132: 110873. doi: 10.1016/j.biopha.2020.110873.
23. Clearesta KE, Mambu T, Tjandra F, Langi FG. Hubungan antara tumor-infiltrating lymphocytes dengan stadium dan derajat diferensiasi adenokarsinoma kolorektal. Medical Scope Journal. 2023; 6(1): 99–105.
24. Miskad UA, Hamzah N, Cangara MH, Nelwan BJ, Masadah R, Wahid S. Programmed death-ligand 1 expression and tumor-infiltrating lymphocytes in colorectal adenocarcinoma. Minerva Med. 2020; 111(4): 337–43.
25. Alsaab HO, Sau S, Alzhrani R, Tatiparti K, Bhise K, Kashaw SK, *et al.* PD-1 and PD-L1 Checkpoint signaling inhibition for cancer immunotherapy: Mechanism, combinations, and clinical outcome. Front Pharmacol. 2017; 8: 561. doi: 10.3389/fphar.2017.00561.
26. Iseki Y, Shibutani M, Maeda K, Nagahara H, Fukuoka T, Matsutani S, *et al.* A new method for evaluating tumor-infiltrating lymphocytes

(TILs) in colorectal cancer using hematoxylin and eosin (H-E)-stained tumor sections. PLoS ONE. 2018; 13(4): e0192744. doi: 10.1371/journal.pone.0192744.

27. Hu M, Zhang S. Expression and clinical significance of FGFR1 and FGFR2 in laryngeal squamous cell carcinoma. Transl Cancer Res TCR. 2022; 11(9): 3222–34.
28. Idos GE, Kwok J, Bonthala N, Kysh L, Gruber SB, Qu C. The prognostic implications of tumor infiltrating lymphocytes in colorectal cancer: A systematic review and meta-analysis. Sci Rep. 2020; 10(1): 3360. doi: 10.1038/s41598-020-60255-4.
29. Paijens ST, Vledder A, De Bruyn M, Nijman HW. Tumor-infiltrating lymphocytes in the immunotherapy era. Cell Mol Immunol. 2021; 18(4): 842–59.
30. Ruan R, Li L, Li X, Huang C, Zhang Z, Zhong H, *et al*. Unleashing the potential of combining FGFR inhibitor and immune checkpoint blockade for FGF/FGFR signaling in tumor microenvironment. Mol Cancer. 2023; 22(1): 60. doi: 10.1186/s12943-023-01761-7.
31. Keir ME, Butte MJ, Freeman GJ, Sharpe AH. PD-1 and its ligands in tolerance and immunity. Annu Rev Immunol. 2008; 26(1): 677–704.
32. Wherry EJ, Kurachi M. Molecular and cellular insights into T cell exhaustion. Nat Rev Immunol. 2015; 15(8): 486–99.
33. Pauken KE, Wherry EJ. Overcoming T cell exhaustion in infection and cancer. Trends in Immunology. 2015; 36(4): 265–76.
34. Lin X, Kang K, Chen P, Zeng Z, Li G, Xiong W, *et al*. Regulatory mechanisms of PD-1/PD-L1 in cancers. Mol Cancer. 2024; 23(1): 108. doi: 10.1186/s12943-024-02023-w.
35. Han Y, Liu D, Li L. PD-1/PD-L1 pathway: current researches in cancer. Am J Cancer Res. 2020; 10(3): 727–42.
36. Song Z, Lin Y. Innovative immunotherapy targeting at PD-1/PD-L1 signaling pathway: Mechanism, efficacy and safety analysis of monotherapy and combination therapies in non-small cell lung cancer (NSCLC) treatment. In: Tahir MS, Xing L, editors. E3S Web Conf. 7th International Conference on Energy Science and Chemical Engineering (ICESCE 2021). 2021; 267: 02026. doi: 10.1051/e3sconf/202126702026.