

Investigating the FIB-4 index and PET/CT metabolic parameters for the differentiation status of hepatocellular carcinoma

FIB-4 index and PET/CT parameters in HCC

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Abstract

Aim: The FIB-4 index is a widely used noninvasive marker of hepatic fibrosis and has prognostic value in hepatocellular carcinoma (HCC). However, its association with tumor differentiation remains unclear. This study aimed to investigate the relationship between the fibrosis-4 (FIB-4) index and histological differentiation of HCC using 18F- fluorodeoxyglucose (FDG) positron emission tomography/computed tomography (PET/CT) metabolic parameters.

Methods: This retrospective, single-center study included 110 patients with newly diagnosed, histologically confirmed HCC who underwent 18F-FDG PET/CT between 2017 and 2024. Patients with moderately differentiated or unclassified tumors, prior liver-directed therapies, recurrent HCC, or other malignancies were excluded. Tumors were categorized as well-differentiated or poorly-differentiated. PET/CT-derived metabolic parameters were analyzed. The FIB-4 index was calculated at the time of imaging. Group comparisons, correlation analyses, and receiver operating characteristic (ROC) curve analyses were performed to evaluate associations among FIB-4, metabolic parameters, and tumor differentiation.

Results: The median FIB-4 index did not differ significantly between well- and poorly differentiated HCC groups (4.2 vs. 4.5, $p = 0.765$). No significant correlation was observed between the FIB-4 index and PET metabolic parameters (all $p > 0.05$). In contrast, poorly-differentiated HCCs showed significantly higher metabolic activity, with markedly elevated SUVmax, SUVmean, SUVpeak, tumor-to-liver and tumor-to-blood ratios (all $p < 0.001$). ROC analysis showed that SUVmax had the highest diagnostic performance for predicting poor differentiation (AUC = 0.981), whereas the FIB-4 index had poor discriminative ability (AUC = 0.517).

Conclusion: The FIB-4 index does not reflect histological differentiation in HCC and has limited utility for tumor grading. 18F-FDG PET/CT metabolic parameters provide superior discrimination of tumor differentiation, although overlap between grades underscores the need for a multimodal diagnostic approach.

Keywords

hepatocellular carcinoma, FIB-4 index, metabolic parameters, HCC differentiation

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Introduction

Hepatocellular carcinoma (HCC) is the most common primary malignant liver tumor and a leading cause of cancer-related death worldwide. Early diagnosis and risk assessment are vital for effective treatment and improved outcomes. Unlike many solid tumors, HCC usually doesn't require histological confirmation; diagnosis relies on characteristic imaging findings on contrast-enhanced computed tomography (CT) and magnetic resonance imaging (MRI), such as arterial-phase hyperenhancement followed by washout in later phases.^{1,2} However, although these modalities are highly sensitive for lesion detection, they provide limited information about tumor biology, aggressiveness, and prognosis. 18F-fluorodeoxyglucose (FDG) PET/CT is a valuable modality in HCC, providing functional insights beyond morphology. Although it is less sensitive for detecting well-differentiated tumors, increased FDG uptake is associated with more aggressive features, including poor differentiation, microvascular invasion, early recurrence, and unfavorable prognosis.^{1,3,4} Lower FDG uptake in well-differentiated HCC is linked to preserved hepatocytic function, including higher glucose-6-phosphatase activity and reduced expression of glucose transporters such as glucose transporter (GLUT)1 and GLUT3.^{5,6} These observations underscore the utility of FDG-PET/CT in preoperative assessment to improve risk stratification and guide treatment for HCC patients. In addition to imaging biomarkers, noninvasive biochemical indices play a pivotal role in managing chronic liver disease and HCC. The fibrosis-4 (FIB-4) index is a simple, cost-effective marker of liver fibrosis with strong prognostic value for HCC development and outcomes in patients with hepatitis and nonalcoholic steatohepatitis (NASH). A study of over 200,000 participants found that those with high FIB-4 scores had a hazard ratio of 58 for HCC compared with those with low scores, underscoring its prognostic value for HCC risk.⁷ Although FIB-4 is a reliable indicator of fibrosis and hepatic reserve, its relationship to tumor-specific characteristics, such as differentiation, remains unclear. Combining noninvasive fibrosis markers with functional imaging could improve risk assessment and personalized treatment in HCC. However, it's unclear whether the FIB-4 index reflects tumor differentiation or aligns with PET/CT metabolic data. This study investigates the association between FIB-4 and histological differentiation of HCC using 18F-FDG PET/CT.

Materials and Methods

Patient Selection

Three hundred eighty-nine patients were referred with suspicion of hepatocellular carcinoma between January 2017 and July 2024. Patients were excluded if they had metastatic disease from another primary malignancy, cholangiocellular carcinoma, moderately differentiated or unclassified HCC, prior liver transplantation, recurrent HCC, prior locoregional treatments (transarterial chemoembolization [TACE], transarterial radioembolization [TARE], or radiofrequency ablation [RFA]), or significant physiologic FDG uptake interfering with image interpretation (e.g., diffuse brown fat or muscular activity). After applying these criteria, 110 patients with newly diagnosed, histologically confirmed HCC were included in the final analysis. Pathology reports and laboratory data required for the

calculation of the FIB-4 index were obtained from electronic medical records.

FDG PET-CT Study and Interpretation Criteria

18F-FDG PET/CT imaging was performed using an integrated PET/CT scanner (Biograph mCT, Siemens Healthineers, Knoxville, USA) according to European Association of Nuclear Medicine (EANM) guidelines for oncologic imaging.^{8,9} Semi-quantitative analysis was conducted using an automatically generated spherical volume of interest (VOI) placed over the liver lesion with a minimum volume of 20 cm³. Standardized uptake values, including maximum standardized uptake value (SUVmax), mean standardized uptake value (SUVmean), and peak standardized uptake value (SUVpeak), as well as metabolic tumor volume (MTV) and total lesion glycolysis (TLG), were calculated. To minimize variability in background reference selection, both tumor-to-liver and tumor-to-blood pool ratios were analyzed. Tumor-to-liver ratios were calculated using non-tumoral liver SUVmax and SUVmean (T/LmaxR and T/LmeanR), while tumor-to-blood pool ratios were derived from mediastinal blood pool SUVmax and SUVmean (tumor-to-blood pool ratio [T/B]maxR and T/BmeanR). Non-tumoral liver activity was obtained from the average of two automatically placed VOIs (20 cm³ each) in normal liver parenchyma, when feasible, from different hepatic segments.¹⁰ Based on histopathological evaluation, tumors were classified as well-differentiated or poorly differentiated, while moderately differentiated and unclassified tumors were excluded. In addition, metabolic differentiation subgroups were defined using previously reported prognostic cut-off values, with well-differentiated HCC defined as tumor standardized uptake value (SUV) < 4.9 and tumor-to-liver ratio (T/L) ratio < 1.83, and poorly differentiated HCC defined as tumor SUV ≥ 4.9 and T/L ratio ≥ 1.83.¹¹ All PET/CT data were transferred and analyzed using a dedicated workstation via the Digital Imaging and Communications in Medicine (DICOM) protocol (Syngo.via, Siemens).

Calculation of the FIB-4 index

FIB-4 index was calculated at the time of 18F-FDG PET/CT using the following formula: $n(\text{age} \times \text{aspartate aminotransferase [AST]}) / (\text{platelet count} \times \sqrt{\text{alanine aminotransferase [ALT]}})$. Receiver operating characteristic (ROC) curve analysis was performed to evaluate the ability of the FIB-4 index to predict histological differentiation of HCC. Based on previously validated thresholds for advanced liver fibrosis, patients were categorized into three subgroups using established cut-off values: 2.67.¹²

Ethical Approval

This study was approved by the Ethics Committee of Sakarya University (Date: 2025-02-20, No: E-450939/116). Informed consent was obtained.

Statistical Analysis

Patients were grouped by histological differentiation. Baseline characteristics were compared using the chi-square test or Fisher's exact test for categorical variables, as appropriate. Continuous variables were analyzed with the Mann-Whitney

U test. Variables that were statistically significant ($p < 0.05$) were further evaluated with receiver operating characteristic (ROC) curve analysis, including calculation of the area under the curve (AUC) and determination of optimal cutoff values using the Youden index. Univariate analyses assessed associations of the FIB-4 index and PET/CT metabolic parameters with differentiation status, and significant variables were included in multivariate models. One-way analysis of variance and Spearman correlation analyses were used to explore relationships between the FIB-4 index and metabolic parameters. All statistical analyses were performed using IBM SPSS Statistics for Windows, Version 24.0 (IBM Corp., Armonk, NY, USA).

Reporting Guidelines

This study was conducted and reported in accordance with the STROBE guidelines for observational studies.

Results

All Characteristics

A total of 110 patients (93 M/17 F; mean age 66.8 ± 10.1 years) underwent 18F-FDG PET/CT. Of these, 56 (51%) had well-differentiated HCC and 54 (49%) had poorly differentiated HCC. Baseline clinical characteristics and PET/CT metabolic parameters by differentiation status are summarized in Supplementary Table 1. No significant association was observed between the FIB-4 index and histological differentiation. In contrast, poorly differentiated HCC was associated with a higher number of liver lesions, increased frequency of peritoneal carcinomatosis, and higher rates of visually positive PET findings ($p < 0.05$). Moreover, poorly differentiated tumors showed significantly higher metabolic activity, with increased SUVmax, SUVmean, SUVpeak, and tumor-to-background ratios compared with well-differentiated HCC ($p < 0.001$). Because a large proportion of well-differentiated tumors (63%) did not show visually highly detectable FDG uptake, it was not possible to reliably define true tumor margins in these cases, which prevented accurate measurement of MTV and TLG.

Comparison of the FIB-4 Index According to Histological Grading

The FIB-4 index did not differ significantly between well- and poorly differentiated HCC groups (median: 4.2 vs. 4.5, $p = 0.765$), indicating limited discriminatory value for histological differentiation. When patients were stratified into three groups using established FIB-4 cut-off values (2.67), no significant associations were observed between FIB-4 categories and histological differentiation or 18F-FDG PET/CT metabolic parameters.

Comparison of Metabolic Parameters According to Histological Grading

Significant differences in PET/CT-derived metabolic parameters were observed between well-differentiated and poorly differentiated HCC. Poorly differentiated tumors showed markedly higher metabolic activity, with increased SUVmax, SUVmean, and tumor-to-liver and tumor-to-blood pool ratios compared with well-differentiated tumors (all $p < 0.001$).

Comparison of FIB-4 Index According to Metabolic Parameters

No significant correlation was observed between the FIB-4 index and any PET/CT metabolic parameters (all $p > 0.05$), indicating that liver fibrosis severity, as assessed by FIB-4, was not associated with tumor metabolic activity in this cohort.

ROC Curve Analysis of the FIB-4 Index and Metabolic Parameters
Receiver operating characteristic (ROC) analysis showed that PET/CT-derived metabolic parameters had higher diagnostic accuracy for predicting poorly differentiated HCC than the FIB-4 index. Among all evaluated variables, lesion SUVmax showed the highest discriminative ability (AUC = 0.981, $p < 0.001$). In contrast, the FIB-4 index showed poor diagnostic performance, with an AUC of 0.517 ($p > 0.05$). When different FIB-4 thresholds were evaluated, a cutoff of ≥ 1.37 had the highest sensitivity (94.4%) but the lowest specificity (14.3%). As the threshold increased, sensitivity decreased (85.2% at ≥ 1.77 and 70.4% at ≥ 2.67), while specificity improved slightly (16% and 34%, respectively). Overall diagnostic accuracy ranged from 50% to 53.6%, and Youden index values remained low across all thresholds, indicating limited discriminative ability ($p > 0.05$). Other PET/CT metabolic parameters, including SUVmean and tumor-to-liver and tumor-to-blood pool ratios, also showed excellent diagnostic performance, with AUC values consistently exceeding 0.960. The detailed data are provided in Supplementary Tables 1 and 2.

Discussion

Early diagnosis and accurate risk stratification are essential for optimizing treatment strategies and improving prognosis in HCC. Because liver biopsy is not routinely performed due to bleeding and tumor seeding risks, HCC diagnosis and management rely largely on imaging and noninvasive biomarkers. However, conventional morphologic features, such as tumor size and number, are insufficient to fully reflect tumor biology, aggressiveness, or recurrence risk. The FIB-4 index is a well-established noninvasive marker of liver fibrosis and has demonstrated prognostic value in patients with chronic liver disease and HCC. In the present study, however, no significant difference in FIB-4 values was observed between well- and poorly differentiated HCC, indicating that FIB-4 does not adequately reflect histological tumor differentiation. Across commonly used cutoff values reported in previous literature¹²⁻¹⁴ (2.67), positive predictive value (PPV) remained around 50% with limited specificity, indicating modest diagnostic ability. Conversely, negative predictive value (NPV) was highest for FIB-4 > 1.37 (72.7%), suggesting a better ability to rule out poor differentiation at this cutoff. These findings suggest that FIB-4 primarily reflects background liver fibrosis and hepatic reserve rather than tumor aggressiveness or differentiation and should therefore be interpreted alongside other diagnostic modalities. Previous studies support the prognostic value of FIB-4 in HCC. Ito et al.¹⁵ demonstrated that lower FIB-4 scores (

Limitations

This study has some limitations. Its retrospective, single-center design limits generalizability. Only well- and poorly differentiated HCC were included, whereas moderately differentiated and unclassified tumors were excluded, potentially introducing

selection bias. Many well-differentiated tumors showed no FDG uptake, preventing measurement of MTV and TLG. Incomplete availability of serum biomarkers, including alpha-fetoprotein (AFP) levels and viral hepatitis status, precluded their inclusion in the analysis. Finally, although PET/CT metabolic parameters demonstrated strong diagnostic performance, histological heterogeneity and molecular variability within HCC may contribute to overlap between differentiation groups.

Conclusion

While the FIB-4 index remains a useful noninvasive marker of hepatic fibrosis and prognosis in HCC patients, it doesn't reflect tumor differentiation. Conversely, 18F-FDG PET/CT metabolic parameters, especially SUV-based measures and tumor-to-background ratios, strongly identify poorly differentiated tumors. These findings underscore FDG PET/CT's value as a functional imaging tool for assessing tumor aggressiveness. However, given the overlap among differentiation grades, FDG PET/CT should be part of a multimodal approach that integrates imaging, biochemical markers, and clinical data to improve risk assessment and treatment planning for HCC.

Ethics Declarations

This study was conducted in accordance with the ethical principles of the Declaration of Helsinki and its subsequent amendments. All procedures were performed in line with institutional and national ethical standards. The study protocol was reviewed and approved by the Ethics Committee of Sakarya University Faculty of Medicine (Date: 20.02.2025, Decision No: E-450939/116).

Animal and Human Rights Statement

All procedures performed in this study were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards.

Informed Consent

Informed consent was waived due to the retrospective design of the study.

Data Availability

The datasets used and/or analyzed during the current study are not publicly available due to patient privacy reasons but are available from the corresponding author on reasonable request.

Conflict of Interest

The authors declare that there is no conflict of interest.

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Scientific Responsibility Statement

The authors declare that they are responsible for the article's scientific content, including study design, data collection, analysis and interpretation, writing, and some of the main line, or all of the preparation and scientific review of the contents, and approval of the final version of the article.

Abbreviations

AFP: Alpha-fetoprotein

ALBI: Albumin-bilirubin

ALT: Alanine aminotransferase

AST: Aspartate aminotransferase

AUC: Area under the curve

CT: Computed tomography

DICOM: Digital Imaging and Communications in Medicine

EANM: European Association of Nuclear Medicine

FDG: Fluorodeoxyglucose

FIB-4: Fibrosis-4 index

GLUT: Glucose transporter

HCC: Hepatocellular carcinoma

MRI: Magnetic resonance imaging

MTV: Metabolic tumor volume

NASH: Nonalcoholic steatohepatitis

NPV: Negative predictive value

PET/CT: Positron emission tomography/computed tomography

PPV: Positive predictive value

ROC: Receiver operating characteristic

RFA: Radiofrequency ablation

SPSS: Statistical Package for the Social Sciences

SUV: Standardized uptake value

SUVmax: Maximum standardized uptake value

SUVmean: Mean standardized uptake value

SUVpeak: Peak standardized uptake value

TACE: Transarterial chemoembolization

TARE: Transarterial radioembolization

TLG: Total lesion glycolysis

T/L: Tumor-to-liver ratio

T/B: Tumor-to-blood pool ratio

VOI: Volume of interest

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