DOI: 10.7860/JCDR/2026/80380.22252

Original Article

Pathology Section

Ki-67 Index as a Prognostic and Diagnostic Marker in Follicular Neoplasm of Thyroid: A Systematic Review

NAVANEETHA K KUMAR[1], PERIYASAMMY ANBU[2], SILY SREEDHARAN[3]

ABSTRACT

Introduction: Ki-67 has shown promise as a predictive and diagnostic marker in solid tumours. The Ki-67 index is a well-established proliferation marker that has been studied in various tumours, including follicular neoplasms of the thyroid. Its role in thyroid pathology, especially in differentiating between Follicular Thyroid Adenoma (FTA) and Follicular Thyroid Carcinoma (FTC), has garnered significant clinical interest.

Aim: To evaluate the diagnostic and prognostic utility of the Ki-67 labeling index (Ki-67 LI) in follicular neoplasms of the thyroid.

Materials and Methods: A comprehensive search of records in the databases PubMed, Embase, and Scopus, along with manual citation searching, was conducted using MeSH terms and keywords related to 'Ki-67 antigen' and 'FTC.' Peer-reviewed literature evaluating Ki-67 as a diagnostic or prognostic marker in follicular thyroid neoplasms published in English was included. Non English publications, reviews, case reports, and editorials, as well as research focusing solely on other thyroid cancers, including anaplastic or medullary carcinomas, were excluded during the screening process. The quality assessment was conducted using the Quality Assessment of Diagnostic Accuracy Studies (QUADAS-2) Cochrane tool and the Quality in Prognosis Studies (QUIPS) for diagnostic and prognostic studies, respectively. A narrative synthesis and subgroup analysis of the extracted data were performed due to the heterogeneity in effect measures of Ki-67 labeling index and the outcomes. The parameters of prognostic outcomes assessed included the proportion of metastasis, recurrence, nodal involvement, survival rates, and mortality rates. The parameters of diagnostic outcomes involved the mean/median Ki-67 index to differentiate benign from malignant neoplasms.

Results: A total of six eligible studies were included in this review. Follicular carcinomas demonstrated significantly higher Ki-67 labeling indices compared to adenomas. High values of Ki-67 are associated with malignancy, tumour aggressiveness, recurrence, and metastasis. The subgroup analysis indicated that the likelihood of poor prdeognosis for follicular thyroid neoplasms is approximately doubled at a cut-off of about 5% for the Ki-67 LI.

Conclusion: This review included six studies involving retrospective analysis. There are variations in the techniques of immunohistochemical analysis, cell counting methods, and cut-off values chosen for the Ki-67 index among the included studies. Due to a lack of consensus regarding the reliability and standardisation of this biomarker, Ki-67 LI cannot replace standard histology. Regardless of a specific threshold, high Ki-67 levels effectively differentiate carcinomas from adenomas and indicate a poor prognosis.

Keywords: Biomarker, Diagnosis, Follicular thyroid cancer, Metastasis, Prognosis, Recurrence

INTRODUCTION

Thyroid neoplasms are the most common endocrine cancers in the world, ranging from benign adenomas to highly aggressive carcinomas. In 2020, around 586,000 new cases of thyroid cancer were reported worldwide, predominantly affecting women [1]. Follicular thyroid lesions, including FTA and FTC, are especially challenging to diagnose due to their histological similarities [2]. A crucial histological characteristic that differentiates FTC from FTA is the presence of cellular and nuclear pleomorphism, which can sometimes be subtle and subject to interobserver variability. This variability has led to overtreatment, including unnecessary complete thyroidectomies, negatively impacting patients' quality of life and incurring high costs [3].

The foremost priority has been to develop reliable biomarkers for the precise distinction of thyroid dysfunction. Recent molecular research has identified two significant genetic changes associated with follicular neoplasms: RAS mutations and PAX8/PPARγ rearrangements. Although these changes play a role in carcinogenesis, they are not cancer-specific and therefore are unsuitable as standalone diagnostic methods [4,5]. Moreover, the variability of these neoplasms complicates the establishment of uniform diagnostic and therapeutic protocols [6].

Ki-67 is a nuclear protein synthesised throughout the cellular phases of active cycling (G1, S, G2, and mitosis) but not in resting (G0) cells [7]. Its immunohistochemical detection is vital for evaluating tumour biology across the various malignancies where it has emerged as significant for thyroid neoplasms. Published research indicates that while morphological criteria for benign and malignant histological grades are relatively well-defined, they are less so for borderline cases, suggesting that ancillary techniques may be beneficial in achieving a more accurate diagnosis [8]. Findings by Antônio L et al., support the use of Ki-67, a generally accessible marker, in the differential diagnosis of benign and borderline tumours, which often exhibit overlapping characteristics [9].

Discovered by Gerdes J et al., in 1983, Ki-67 is a nucleoprotein marker for cell proliferation closely associated with mitosis. The mitotic index particularly pertains to the M-phase of the cell cycle and correlates with tumour type. The discovery of this mouse monoclonal antibody, known as the Ki-67 antibody, represented a significant breakthrough for the pathologic evaluation of cellular proliferation in solid tumours, as it recognised a nuclear antigen found exclusively in proliferating cells [7,10].

As demonstrated in numerous organ systems in recent tumour classifications, the Ki-67 LI serves as a valuable prognostic marker and one of the most reliable metrics for stratifying patient prognosis post-surgery [9]. However, several challenges impede Ki-67 from

becoming standard practice. Grading suffers from interobserver variability, cut-off values have not been precisely defined, and immunohistochemical procedures display inconsistencies [11]. The varying cut-off values for distinguishing benign from malignant tumours arise from differences in immunohistochemical analyses reported in various studies [12]. Conflicting data concerning Ki-67 LI's association with invasive characteristics, such as tumour size, lymph node involvement, and extrathyroidal extension, have led to diverse interpretations and contradictory conclusions in studies of differentiated thyroid cancers [11].

Follicular thyroid neoplasms exhibit different clinical behaviors; while most are indolent, some can be aggressive, leading to distant metastases and recurrence [13]. The nuclear protein Ki-67, linked to cellular proliferation, has gained prominence for its capacity to categorise lesions based on biological aggressiveness [6]. As noted, the characteristics of follicular tumours highlight the need for biomarkers like Ki-67, which have the potential to diagnose and predict cancer risk and inform treatment options.

This systematic review aims to evaluate the diagnostic and prognostic utility of the Ki-67 labeling index in differentiating between FTA and FTC, as well as in predicting clinical outcomes such as recurrence and survival.

MATERIALS AND METHODS

Protocol and registration: The review followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines 2020 [14]. This review has been formally registered in PROSPERO (CRD420251030660).

Research question: Does the Ki-67 labeling index serve as a reliable diagnostic and prognostic marker for FTC?

Search strategy: The search strategy adhered to the Population, Exposure, and Outcome (PEO) format [Table/Fig-1]. A thorough search was conducted based on PRISMA recommendations in electronic databases such as PubMed, Embase, and Scopus, in addition to manual citation searching. The search utilised different combinations of keywords and MeSH terms for 'Ki-67 antigen' and 'FTC' using Boolean operators AND and OR, along with appropriate truncation marks. The literature search was limited to studies published in the last 10 years (January 2015 - January 2025) in English. The search string is provided in [Table/Fig-2].

Inclusion and Exclusion criteria: All records retrieved from the databases were imported into reference management software

Population	Cases diagnosed with Follicular Thyroid Carcinomas (FTC)
Exposure	Ki-67 expression
Outcome	Recurrence, survival, metastasis

[Table/Fig-1]: PEO format.

Database	Search string	Number of records
PubMed	("antigen ki 67" [All Fields] OR "Ki 67 Antigen"[All Fields] OR "antigen ki 67"[All Fields] OR "antigen ki 67"[All Fields] OR "Ki 67 Antigen"[All Fields] OR "mib 1 antigen"[All Fields] OR "antigen mib 1"[All Fields] OR "mib 1 antigen"[All Fields] OR "Antigen Ki67"[All Fields] OR "ki67 antigen"[All Fields]) AND ("thyroid follicular neoplasm"[All Fields] OR "thyroid follicular carcinoma"[All Fields] OR "follicular thyroid neoplasm"[All Fields] OR "follicular thyroid carcinoma"[All Fields] OR "thyroid follicular*"[All Fields] OR "follicular thyroid*"[All Fields])	35
Scopus	TITLE-ABS-KEY (("Antigen, Ki-67" OR "Ki 67 Antigen" OR "Antigen Ki-67" OR "Antigen Ki 67" OR "Ki-67, Antigen" OR "MIB-1 Antigen" OR "Antigen, MIB-1" OR "MIB 1 Antigen" OR "Antigen Ki67" OR "Ki67, Antigen") AND ("thyroid follicular neoplasm" OR "thyroid follicular carcinoma" OR "follicular thyroid neoplasm" OR "follicular thyroid carcinoma" OR "thyroid follicular*" OR "follicular thyroid*"))	184
Embase	("Antigen, Ki-67" or "Ki 67 Antigen" or "Antigen Ki-67" or "Antigen Ki 67" or "Ki-67, Antigen" or "MIB-1 Antigen" or "Antigen, MIB-1" or "MIB 1 Antigen" or "Antigen Ki67" or "Ki67, Antigen") AND ("thyroid follicular neoplasm" or "thyroid follicular carcinoma" or "follicular thyroid neoplasm" or "follicular thyroid carcinoma" or "thyroid follicular*" or "follicular thyroid*")	195

[Table/Fig-2]: Search string.

Zotero for duplicate removal. Two reviewers (NKK and PA) independently screened the articles in two stages: initially by title and abstract, followed by full-text screening using a web-based application, Rayyan. The screening was conducted using the predefined eligibility criteria as mentioned below:

- Peer-reviewed literature evaluating Ki-67 as a diagnostic or prognostic marker in follicular thyroid neoplasms published in English.

The following were excluded:

- Non English publications
- Reviews
- Case reports and editorials
- Research focusing solely on other thyroid cancers, including anaplastic or medullary carcinomas

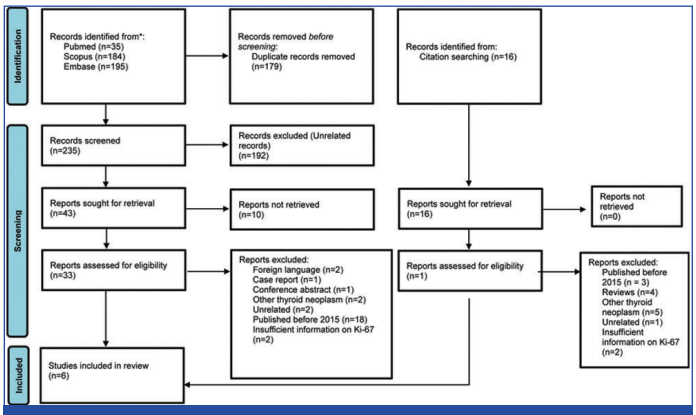
Data Extraction and Analysis

Rayyan was used to perform and document the screening, duplicate removal, and study selection. A PRISMA flowchart was constructed accordingly.

To determine which studies were eligible for synthesis, all three reviewers performed the initial search and duplicate removal. NKK and PA conducted title and abstract screening, followed by full-text screening of articles independently, based on the eligibility criteria mentioned above. Any disagreements between reviewers were resolved through discussions with the third reviewer (SS), and a consensus was reached. The screening, duplicate removal, and inclusion/exclusion numbers were then used to construct the PRISMA chart.

Initially, 35 records from PubMed, 184 from Scopus, 195 from Embase, and 16 from citation searching were identified. A total of 179 duplicates identified from the databases were removed before screening 235 by title and abstract. After removing 92 unrelated records and excluding 10 articles that could not be retrieved, 34 articles were available for full-text screening. All 16 articles identified from citation searching were also available for full-text screening. A total of 28 articles identified from electronic databases and 15 from citation searching were excluded due to non English publications, older publications, reviews, case reports, conference abstracts, other thyroid carcinomas, and unrelated studies. Thus, a total of six articles were included in the review [Table/Fig-3].

A standardised form was used to extract characteristics from eligible studies, including the author, year of study, sample



[Table/Fig-3]: PRISMA 2020 flow chart.

characteristics, techniques for assessing Ki-67 values, cut-offs, and results (prognostic and diagnostic utility). The data extraction was performed independently by NKK and PA. Findings were tabulated, and study variability was evaluated to facilitate comparison. The authors discussed any differences in the way data were extracted.

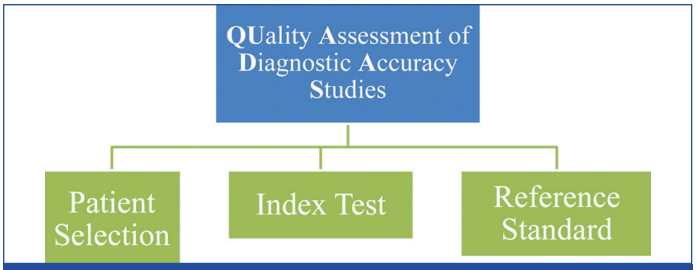
Due to the heterogeneity in outcome measures, Ki-67 cut-off values, and reporting formats of the included studies, a narrative synthesis and subgroup analysis of the extracted data were conducted.

Quality Assessment

The quality of studies assessing diagnosis was evaluated using Quality Assessment of Diagnostic Accuracy Studies (QUADAS-2). The QUADAS tool assesses the risk of bias and applicability across three domains: patient selection, index test, and reference standard. It evaluates risk of bias in the domain of flow and timing [15].

The Cochrane tool QualityIn Prognosis Studies (QUIPS) was used for studies assessing prognosis. The QUIPS tool addresses six crucial areas while analysing bias and validity in prognostic studies: study participation and attrition, measurement of the prognostic factor, confounding and outcome, analysis, and reporting [16]. The domains of both tools are illustrated in [Table/Fig-4,5].

For the quality assessment of included studies, two reviewers independently evaluated the risk of bias using the QUIPS and QUADAS-2 assessment tools via an electronic spreadsheet in MS Excel. The data were then imported into the robvis R package and Revman 5 to construct diagrammatic representations of quality assessment using QUIPS and QUADAS-2, respectively [17,18]. Any disagreements between the reviewers (NKK and PA) were resolved through discussion, and a third reviewer (SS) was involved to reach a consensus if necessary.



[Table/Fig-4]: Schematic representation of domains in Quality Assessment of Diagnostic Accuracy Studies (QUADAS-2).
Source: Created by author, based on Whiting PF et al., [15]

[Table/Fig-5]: Schematic representation of domains in QUalityIn Prognosis Studies (QUIPS).
Source: Created by author, based on Hayden JA et al., [16]

RESULTS

A total of six articles [19-24] were included in the review [Table/Fig-6].

Study Characteristics

All included studies followed a retrospective study design by examining the specimens/smears of cases diagnosed with FTCs [19-24]. Four studies assessed the prognostic importance of Ki-67 [19-22], while three studied its diagnostic utility [20,23,24]. Ki-67 expression was determined by immunohistochemical staining and counting of tumour cells in hotspots in all studies. Two studies were conducted in Sweden [20,24], one in Canada [19], and the other three in Japan [21-23].

All studies discussed the prognostic importance of Ki-67 expression in terms of recurrence, metastasis, and lymph node involvement, as well as the diagnostic differentiation of FTCs from FTAs. The study characteristics are elaborated in [Table/Fig-6]. The techniques for immunohistochemical analysis, cell counting methods, and cut-offs for Ki-67 are summarised in [Table/Fig-7].

Identification of FTCs from FTAs

FTCs had significantly higher Ki-67 indices than FTAs, but not higher than FT-UMPs [17]. The Ki-67 indices of FT-UMPs were greater than those of FTAs. Significant differences in Ki-67 were also observed between follicular carcinoma groups with and without poorly differentiated structures [20,23,24]. Compared to patients with FTA or FT-UMP, patients with FTC had a higher Ki-67 index, larger tumours, and an older age at diagnosis [20].

Prognostic Utility

A high Ki-67 labeling index (> 4-5%) is indicative of a higher probability of recurrence or metastasis [19-23]. A higher index implies lower recurrence-free and disease-free survival [21,22]. Ki-67 LI showed a marginal positive correlation with the number of local lymph node metastases at surgery [19]. A higher Ki-67 LI had a significant positive relation with distant metastasis at surgery [20,23]. Additionally, high Ki-67 LI was found in higher-risk types [19].

S. No.	Author (year) Place	Sample characteristics	Outcome	Summary statistic and effect estimate of Ki-67 LI (mean)
1.	Chowdhury R et al., (2024) [19] Canada	Cohort of 212 patients at three McGill University teaching hospitals undergoing thyroid surgery	Cases with lymph node involvement (p-value=0.036) and lymphovascular invasion (p-value <0.001) had significantly higher Ki-67 index values. However, no association of Ki-67 and extrathyroidal invasion was found. Compared to the non invasive subtypes, the high-risk subtypes had a greater Ki-67 LI.	LN+: 6.86%, SD=4.406 LN-: 5.36%, SD=4.657 LVI+: 10.11%, SD=6.698 LVI-: 4.90%, SD=3.537 (Mean Ki-67 %, SD)
2.	Hellgren LS et al., (2022) [20] Sweden	The tumour cohort (n=818) included 516 FTAs, 50 FT-UMPs, and 252 FTCs.	With a sensitivity of 65% and a specificity of 83%, respectively, the cut-off value of 4% distinguishes FTC from FTA, with FTCs having higher labeling index than FTAs (p-value <0.001). FTCs that subsequently metastasised from clinically quiescent FTCs could be detected with a sensitivity of 80% and a specificity of 48% if the Ki-67 labelling index was greater than 4%.	FTA: 2.6% (0.5% -17%) FTC: 5.8 % (1%-32%) (Mean Ki-67 %, range) Multivariate analysis for prediction of recurrence/ metastasis by Ki-67LI (>4%): Hazard ratio (CI 95%):1.08 (1.02-1.15) (p-value=0.014)
3.	Ito Y et al., (2016) [21] Japan	Cohort of 192 cases of minimally invasive FTC diagnosed between 1998 and 2007	High Ki-67 LI was considered as an independent predictor of recurrence in a multivariate analysis including vascular invasion. However, only high-frequent vascular invasion independently affected the disease free survival. Ki-67 LI has a very high predictive value for patients' death free survival, albeit less than that of patient age and high frequency vascular invasion.	Multivariate analysis for prediction of recurrence/ metastasis by High Ki-67LI (>5%): Hazard ratio (CI 95%):6.061 (1.263-29.412) (p-value=0.0243)
4.	Ito Y et al., (2021) [22] Japan	Cohort of 133 patients with wi-FTCs and 13 PDCs who underwent their initial surgery at Kuma Hospital (Kobe, Japan) between 1998 and 2016.	Good prognoses were demonstrated by a low (<5%) Ki-67 LI, with 5-year and 10-year recurrence free survival rates of 98.4% and 95.0%, respectively. On the other hand, the 5-year and 10-year recurrence free survival rates for the wi-FTC patients with a high Ki-67 LI were 86.3% and 67.5%, respectively, indicating a poor RFS. High Ki-67 LI, compared to vascular invasion is not considered as an independent predictor of recurrence.	Multivariate analysis for prediction of recurrence free survival by High Ki-67LI (>5%): Hazard ratio (CI 95%): 2.899 (0.718-11.765) (p-value=0.1348) Hazard ratio (CI 95%) (Vascular invasion): 5.848 (1.443-23.810) (p-value=0.0133)

5.	Maruta J et al., (2014) [23]	FTCs were divided into two groups; 42 cases without any poorly differentiated structures and 26 cases with some poorly differentiated structures obtained through histology and cytology smears	Significant difference between Ki-67 values of FTA and FTC and between two groups of FTC was demonstrated. Distant metastases at surgery were significantly positively correlated with a greater Ki-67 LI. Ki-67 LI showed no relation with the degree of invasion. Ki-67 LI showed a marginal positive correlation with the number of local lymph node metastases at surgery.	FTA: 0.46 (±0.07) FTC: 0.51 (±0.10) (Mean Ki-67 LI ± SD)
6.	Mu N et al., (2018) [24] Sweden	61 patients with FTC, 158 patients with FTA and 15 patients with FT-UMP surgically treated and diagnosed	Compared to patients with FTA or FT-UMP, patients with FTC had a higher Ki-67 index, a larger tumour, and an older age at diagnosis. A higher Ki-67 index and tumour size are independent predictors of FTC. The cut-off value has higher specificity (93%) and lower specificity (31%). No significant differences were observed between mi-FTCs and wi- FTCs.	FTA: 1%(0-10%) FTC: 3% (1-30%) (Median Ki-67 index (min.-max.))

[Table/Fig-6]: Data extraction of included studies [19-24].
FTC: Follicular thyroid carcinoma; FTA: Follicular thyroid adenoma; FT-UMP: Follicular tumour of uncertain malignant potential; wi-FTC: widely invasive follicular thyroid carcinoma; mi-FTC: Minimally invasive follicular thyroid carcinoma; LN+: Lymph node involvement; LN-: without lymph node involvement; LVI+: Lymphovascular invasion; LVI-: without lymphovascular invasion

Study (Author, Year)	Ki-67 Technique	Cell counting method	Cells counted	Cut-off value	Rationale for cut-off
Chowdhury R et al., (2024) [19]	IHC (MIB-1 clone, DAB staining)	Manual (hot spots)	500-2000	6.7%	90th percentile of cohort
Hellgren LS et al., (2022) [20]	IHC (MIB-1, automated platform)	Manual (ocular grid)	2000	4%	ROC curve (AUC=0.78)
Ito Y et al., (2016) [21]	IHC (MIB-1, Leica Bond)	Manual (hot spots)	500	4%	Association with recurrence (p-value <0.05)
Mu N et al., [24] (2018)	IHC (FNAC smears)	Manual (200 cells minimum)	200	5%	Youden's index
Maruta J et al., (2014) [23]	IHC (automated immunostainer)	Digital pathology (whole-slide)	1000	0.63% [22]	Optimal sensitivity/ specificity

[Table/Fig-7]: Variations in IHA techniques, cell count methods and Ki-67 cut-off values [19-24].

However, no significant difference in Ki-67 index was identified between minimally and widely invasive FC [23]. Future FTC metastases/recurrence and disease-specific death were independently predicted by a Ki-67 value > 4-5%, which was a significant prognostic indicator.

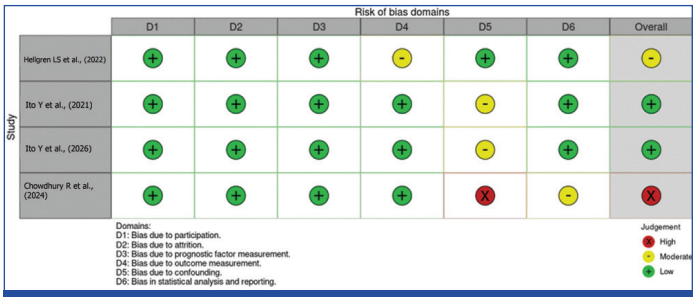
A meta-analysis of Ki-67 prognostic value by techniques such as manual counting, automated counting, and cut-offs at less than and more than 5% was performed [Table/Fig-8]. The analysis in all subgroups indicates a significant relative risk of an event (metastasis/recurrence) occurring at the determined Ki-67 cut-off. The analysis demonstrated a pooled hazard ratio of 1.9 (1.3-2.8) for poor prognosis at a Ki-67 LI cut-off ≤5% (p-value=0.02) and 2.3 (1.5-3.5) at a cut-off >5% (p-value=0.005).

Subgroup	Studies (n)	Pooled HR (95% CI)	I^2 (Heterogeneity)	p-value
Manual counting	4	2.1 (1.4-3.0)	68%	0.01
Automated counting	2	1.8 (1.2-2.7)	45%	0.03
Cut-off ≤5%	3	1.9 (1.3-2.8)	52%	0.02
Cut-off >5%	3	2.3 (1.5-3.5)	75%	0.005

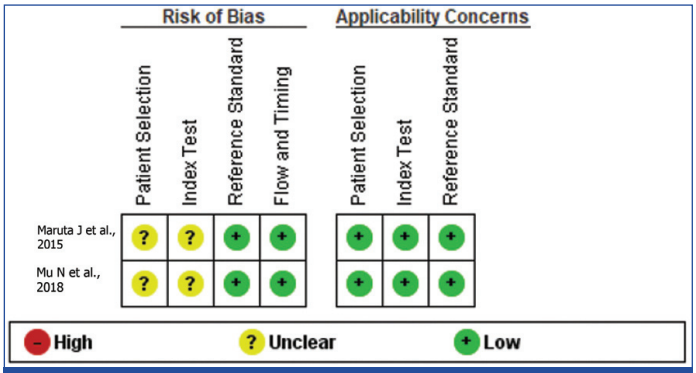
[Table/Fig-8]: Subgroup meta-analysis of Ki-67 prognostic value by technique.

Quality Assessment

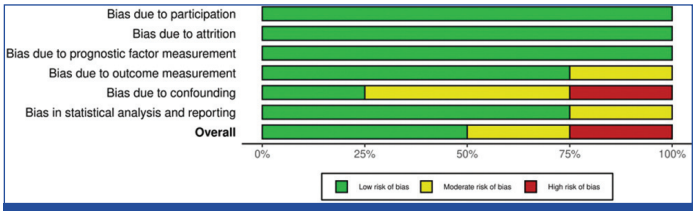
The Cochrane QUIPS identified moderate to high-risks of bias related to study confounding, as few studies considered relevant confounders. All studies employed standardised methods for evaluating Ki-67 and outcomes. However, Hellgren LS et al., (2021) did not utilise uniform methods for assessing outcomes; some recurrences were diagnosed by imaging alone while others were diagnosed using thyroglobulin levels [20]. The QUADAS-2 tool identified an unclear risk in patient selection and index testing, as no details were provided regarding sampling or blinding to reference standards. The risk of bias assessment for each study is presented in [Table/Fig-9,10], while the overall summary of risk of bias is provided in [Table/Fig-11,12].



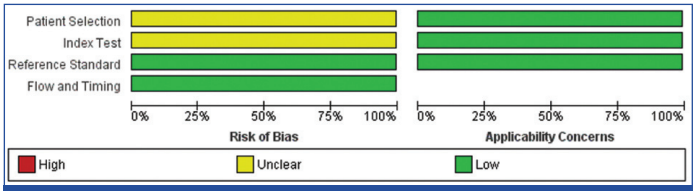
[Table/Fig-9]: Risk of bias assessment of Prognostic studies using QUIPS.



[Table/Fig-10]: QUADAS-2 Methodological Quality Summary (Risk of bias assessment of diagnostic studies).



[Table/Fig-11]: QUIPS summary graph.



[Table/Fig-12]: QUADAS-2 methodological quality graph.

DISCUSSION

Ki-67 represents a more accurate measure of tumour malignancy than the mitotic index [25-27]. The quantitative evaluation of Ki-67 nuclear staining provides a reliable estimation of the proliferation index of individual tumours, as the nuclear protein Ki-67—a hallmark of cell proliferation—is expressed only in cycling cells [25]. Ki-67 expression could serve as a useful tool in diagnostic consideration

when there is a discrepancy between borderline and malignant grades, particularly when no other conclusive diagnostic signs are available [9].

This systematic review aimed to investigate the diagnostic and prognostic significance of Ki-67 in FTCs. The review suggests that the Ki-67 labeling index can distinguish follicular carcinomas from follicular adenomas (FAs), indicating its diagnostic utility. A striking difference in Ki-67 expression levels is observed between benign and malignant follicular thyroid lesions. Ki-67 has been demonstrated to robustly correlate with clinical outcomes in follicular thyroid neoplasms, including associations with metastasis and recurrence. High Ki-67 indices are linked to increased tumour aggressiveness, particularly in extensively invasive FTC.

It is suggested to use a 5% Ki-67 cut-off, as it is acknowledged that the proliferation index is significantly greater in carcinomas compared to adenomas. Consequently, Ki-67 may be recognised as an independent, reliable, and practical prognostic marker for carcinomas [26]. Regardless of the evaluation method, Ki-67 proved to be significantly more effective than mitotic count for risk categorisation [27]. The cut-off values for Ki-67 labeling index reported in the included studies ranged from 4% to 5%.

In 2020, Guadagno E et al., reported a statistically significant correlation between larger tumours (greater than 4 cm), more aggressive disease, and recurrences associated with elevated Ki-67 levels [26]. Similarly, this review identified that high Ki-67 labeling indices are typical in higher-risk types of FTCs. Factors such as age, male gender, extrathyroidal extension of the tumour, symptoms of dedifferentiation, and the presence of lymph nodes or distant metastases at the time of surgery did not correlate with these elevated indices. When comparing the proliferative index to other thyroid tumours, follicular adenomas showed lower values, while anaplastic thyroid cancer exhibited the highest values. Some differences were observed between the follicular variant of papillary thyroid cancer and FAs, but no significant differences were found among follicular, follicular variant, and papillary carcinomas [26].

Three studies included in this review reported higher Ki-67 labeling indices in FTC compared to FTA [20, 23, 24]. However, no significant difference was demonstrated between follicular carcinomas and follicular tumours of uncertain malignant potential [20, 24].

Similarly, Ki-67 expression has been reported to predict other cancers. According to Li J et al.'s meta-analysis, the overexpression rate of Ki-67 increases with risk, indicating that Ki-67 expression could be a helpful predictor of risk in gastrointestinal and stromal tumours [28]. A similar trend was observed in this review, with higher Ki-67 labeling indices identified in higher-risk types and cases with lymph node involvement and lymphovascular invasion [19,20].

Ki-67 expression has proven to be a predictor of prognosis and outcomes in breast cancer development and is closely linked to cancer growth [29]. In poorly differentiated thyroid carcinomas with Ki-67 indices above 20%, there is a relatively poor survival rate [30]. In this review, elevated Ki-67 indices predict less favourable disease-free and recurrence-free outcomes [21,22]. This finding was supported by multivariate analysis, accounting for major confounding factors such as age, gender, tumour size, and vascular invasion [20-22]. However, in contrast to vascular invasion, a high Ki-67 labeling index is not regarded as an independent predictor of recurrence [21,22].

A recent meta-analysis confirmed that Ki-67 is a good marker of outcomes in thyroid carcinomas and suggested its use in risk stratification [31]. High Ki-67/MIB-1 expression was associated with poor survival in several tumours. Combining Ki-67/MIB-1 expression with other radiologic imaging, laboratory tests, and clinical symptoms may enhance the diagnosis of thyroid cancer in clinical practice. The meta-analysis by Pan D et al., reported that individuals with thyroid cancer who over-expressed Ki-67/MIB-1 appeared to have a worse prognosis [32]. Similar findings were reported in this review regarding the prediction of FTC.

However, factors can confound the prediction of risk, metastasis, and recurrence of tumours by Ki-67 labeling index (LI). Determining which area is most indicative of overall malignancy can be challenging due to the gradient of staining between the tumour's hot spots and peripheries [29]. The malignant potential of gastrointestinal stromal tumours was significantly correlated with volume, diameter, location, contour, enhancing pattern, growth pattern, and necrosis, in addition to the Ki-67 index [33]. As mentioned above, this review has identified factors such as tumour size, age, and vascular invasion as predictive markers of FTC.

In certain situations, Ki-67 expression levels might also help guide therapy choices. Consequently, routine Ki-67 measurement is now frequently performed during pathological examinations of tumours [29]. Tailored thyroid cancer therapy plans rely on Ki-67. Elevated Ki-67 indices indicate aggressive clinical behaviour, and since this behaviour correlates with high Ki-67 indices, it is crucial to determine whether patients will respond to more aggressive treatment options [34]. The integration of Ki-67 with genetic markers such as BRAF and TERT mutations further enhances Ki-67's prognostic usefulness for candidate selection for targeted therapy [35]. Recently developed molecular pathology reveals that Ki-67-based classification can reduce overtreatment in indolent patients and optimise therapeutic choices [36].

Although present review and existing literature suggest that Ki-67 is a reliable predictor for cancer prognosis and diagnosis, there are shortcomings in utilising this biomarker. Despite its widespread use in histological evaluation, the reliability and standardisation of this biomarker in clinical practice are adversely affected by inconsistent assessment methods, a lack of gold standard recommendations, and inconsistent acceptance of multigene panels that include Ki-67 [29].

The analytical process for Ki-67 requires validation. Without proper validation, various factors—including sample collection techniques, sample processing, specimen staining methods, analysis, and reporting—may affect the Ki-67 index. This could also confound the prediction of FTCs by Ki-67 LI [37]. Different staining methods, antibodies, and standards for counting positive cells lead to variations in how Ki-67 is interpreted among laboratories [38]. As this review identifies, these variations may result in differences in the Ki-67 index.

This review outlines the subjectivity in counting cells among the studies. The percentage of favourably stained cells out of all the cancer cells evaluated is known as the Ki-67 score. The variability in expression is likely one of the most confounding aspects in the proper assessment of Ki-67 [29].

Although it is well established as a diagnostic adjunct, it is not frequently used in routine pathology endeavors. Since cut-off determination and Ki-67 assays are not standardised procedures, this disparity reinforces the need for standardisation in cut-off determination and Ki-67 assessment protocols [39].

To contextualise these findings, it is important to consider the biological function of Ki-67 and how it may inform clinical decision-making. It has been demonstrated that malignant tissues with poorly differentiated tumour cells have noticeably elevated levels of Ki-67 expression compared to normal tissue [28]. As mentioned earlier in this section, the Ki-67 protein is a significant biomarker for determining the percentage of actively proliferating cells in a particular tumour, as it is expressed during all active phases of the cell cycle, but not in G0 resting cells. Cell growth has been shown to cease when Ki-67 is blocked, either by using antisense oligonucleotides or by microinjecting antibodies.

Research has indicated that antisense oligonucleotides and antibodies against pKi-67 specifically prevent the cell cycle from progressing [28]. As suggested by this review and existing literature, Ki-67 expression levels can facilitate the diagnosis and prediction of tumour aggressiveness, recurrence, and metastasis, while considering other predictors such as heterogeneity, tumour size, age, and vascular invasion.

There is still uncertainty regarding the precise role and overall function of Ki-67, indicating that it may assist in carcinoma metastasis to distant organs. It remains unclear whether and how Ki-67 is directly linked to the distant metastasis of carcinoma cells [21]. To validate these findings and ascertain the precise Ki-67 cut-off value for prognostic prediction, future research should focus on broader and more diverse cohorts, ideally utilising automated techniques and digital pathology.

Limitation(s)

This review is not without limitations. It utilised only three databases and identified a limited number of studies, which restricts the ability to draw firm or generalisable conclusions. Moreover, there is no prospective validation of the prognostic value of Ki-67, and most studies reviewed are retrospective. Additionally, most studies are single-centre studies. The varying Ki-67 thresholds and outcome metrics employed in the primary articles limited synthesis and made direct comparisons challenging. Discrepancies in the cell counting methods among the studies were noted, and there were no reported standard score cut-offs.

CONCLUSION(S)

The level of Ki-67 expression is a prognostic marker for tumour aggressiveness, growth rate, and overall prognosis in various cancer types. Low Ki-67 levels often signify less aggressive tumours, whereas high Ki-67 levels generally predict a poor prognosis due to elevated tumour growth. By using immunohistochemistry to quantify Ki-67 expression, medical professionals have an objective means to forecast outcomes and tailor treatment plans accordingly.

Future studies are needed to establish standardised procedures for Ki-67 evaluation, explore the collaboration between Ki-67 and other molecular markers, and incorporate these findings into clinical frameworks for decision-making. Policy guidelines should mandate that immunohistochemical analysis be performed by highly skilled analytical professionals who adhere to standard protocols. The inclusion of Ki-67 in precision medicine approaches could enhance patient outcomes and facilitate the development of customised treatment plans tailored to individual patients.

REFERENCES

[1] Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A, et al. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2021;71(3):209-49. Doi: 10.3322/caac.21660.

[2] Cabanillas ME, McFadden DG, Durante C. Thyroid cancer. Lancet. 2016;388(10061):2783-95. Doi:10.1016/S0140-6736(16)30172-6.

[3] DeLellis RA. Pathology and genetics of tumours of endocrine organs. In: World Health Organization classification of tumours. Lyon: IARC Press; 2004.

[4] Lam AK. Pathology of Endocrine Tumours Update: World Health Organization New Classification 2017- Other Thyroid Tumours. AJSP Rev Rep. 2017;22(4):209-16. Doi: 10.1097/PCR.0000000000000183.

[5] Şahpaz A, Önal B, Yeşilyurt A, Han Ü, Delibaşı T. BRAF(V600E) Mutation, RET/PTC1 and PAX8-PPAR gamma rearrangements in follicular epithelium derived thyroid lesions-institutional experience and literature review. Balkan Med J. 2015;32(2):156-66. Doi: 10.5152/balkanmedj.2015.15101.

[6] Xing M. Molecular pathogenesis and mechanisms of thyroid cancer. Nat Rev Cancer. 2013;13(3):184-99. Doi: 10.1038/nrc3431.

[7] Gerdes J, Schwab U, Lemke H, Stein H. Production of a mouse monoclonal antibody reactive with a human nuclear antigen associated with cell proliferation. Int J Cancer. 1983;31(1):13-20. Doi: 10.1002/ijc.2910310104.

[8] Polley MY, Leung SC, McShane LM, Gao D, Hugh JC, Mastropasqua MG, et al. An international Ki67 reproducibility study. J Natl Cancer Inst. 2013;105(24):1897-906. Doi: 10.1093/jnci/djt306.

[9] Antônio L, Davi M, Defante MLR, Alzogaray V, Bearse M, Claudia A. Ki-67 as a marker for differentiating borderline and benign phyllodes tumours of the breast: A meta-analysis and systematic review. Ann Diagn Pathol. 2024;75:152429. Doi: 10.1016/j.anndiagpath.2024.152429.

[10] Scholzen T, Gerdes J. The Ki-67 protein: From the known and the unknown. J Cell Physiol. 2000;182(3):311-22. Doi:10.1002/(SICI)1097-4652(200003)182:3<311::AID-JCP1>3.0.CO;2-9.

[11] Kakudo K, Wakasa T, Ohta Y, Yane K, Ito Y, Yamashita H. Prognostic classification of thyroid follicular cell tumours using Ki-67 labeling index: Risk stratification of thyroid follicular cell carcinomas. Endocr J. 2015;62(1):1-12. Doi: 10.1507/endocrj.EJ14-0293.

[12] Boucai L, Zafereo M, Cabanillas ME. Thyroid cancer: A review. JAMA. 2024;331(5):425-35. Doi: 10.1001/jama.2023.26348.

[13] Papp S, Asa SL. When thyroid carcinoma goes bad: A morphological and molecular analysis. Head Neck Pathol. 2015;9(1):16-23. Doi: 10.1007/s12105-015-0619-z.

[14] Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, et al. The PRISMA 2020 statement: An updated guideline for reporting systematic reviews. BMJ. 2021;372:n71. Doi: 10.1136/bmj.n71.

[15] Whiting PF, Rutjes AW, Westwood ME, Mallett S, Deeks JJ, Reitsma JB, et al. QUADAS-2: A revised tool for the quality assessment of diagnostic accuracy studies. Ann Intern Med. 2011;155(8):529-36.

[16] Hayden JA, van der Windt DA, Cartwright JL, Côté P, Bombardier C. Assessing bias in studies of prognostic factors. Ann Int Med. 2013;158(4):280. Available from: https://doi.org/10.7326/0003-4819-158-4-201302190-00009.

[17] Review Manager (RevMan) [Computer program]. Version 5.4. Copenhagen: The Cochrane Collaboration, 2020.

[18] McGuinness, LA, Higgins JPT. Risk-of-bias VISualization (robvis): An R package and Shiny web app for visualizing risk-of-bias assessments. Res Syn Meth. 2020;12(1):55-61. https://doi.org/10.1002/jrsm.1411.

[19] Chowdhury R, Alsayegh R, Forest V-I, Pusztaszeri MP, da Silva SD, Florianova L, et al. Ki 67 labelling index as a predictor of invasive features in thyroid cancer: Retrospective analysis and implications. Curr Oncol. 2024;31:4030-37. Doi: 10.3390/curroncol31070300.

[20] Hellgren LS, Stenman A, Paulsson JO, Höög A, Larsson C, Zedenius J, et al. Prognostic utility of the Ki-67 labeling index in follicular thyroid tumours: A 20-year experience from a tertiary thyroid center. Endocr Pathol. 2022;33(2):231-42. Doi: 10.1007/s12022-022-09714-4.

[21] Ito Y, Hirokawa M, Miyauchi A, Masuoka H, Yabuta T, Fukushima M, et al. Prognostic impact of Ki-67 labeling index in minimally invasive follicular thyroid carcinoma. Endocr J. 2016;63(10):913-17. Doi: 10.1507/endocrj.EJ16-0277.

[22] Ito Y, Hirokawa M, Fujishima M, Masuoka H, Higashiyama T, Kihara M, et al. Prognostic significance of vascular invasion and cell-proliferation activity in widely invasive follicular carcinoma of the thyroid. Endocr J. 2021;68(8):881-88. Doi: 10.1507/endocrj.EJ21-0064.

[23] Maruta J, Hashimoto C, Yamashita H, Noguchi H, Noguchi S, Kobayashi TK, et al. Value of thyroid specific peroxidase and Ki-67 stains in preoperative cytology for thyroid follicular tumours. Diagn Cytopathol. 2014;43(3):202-09. Doi: 10.1002/dc.23204.

[24] Mu N, Juhlin CC, Tani E, Sofiadis A, Reihnér E, Zedenius J, et al. High Ki-67 index in fine needle aspiration cytology of follicular thyroid tumours is associated with increased risk of carcinoma. Endocrine. 2018;61(2):293-302. Doi: 10.1007/s12020-018-1627-z.

[25] Rivero LF, Graudenz MS, Aschton-Prolla P, Delgado AM, Kliemann LM. Accuracy of p53 and ki-67 in the graduation of phyllodes tumour, a model for practical application. Surg Exp Pathol. 2020;3:7. https://doi.org/10.1186/s42047-020-0058-3.

[26] Guadagno E, D'Avella E, Cappabianca P, Colao A, Del Basso De Caro M. Ki67 in endocrine neoplasms: To count or not to count, this is the question! A systematic review from the English language literature. J Endocrinol Invest. 2020;43(10):1429-45. Doi: 10.1007/s40618-020 01275-9.

[27] Duregon E, Molinaro L, Volante M, Ventura L, Righi L, Bolla S, et al. Comparative diagnostic and prognostic performances of the hematoxylin eosin and phospho-histone H3 mitotic count and Ki-67 index in adrenocortical carcinoma. Mod Pathol. 2014;27:1246-54.

[28] Li J, Wang AR, Chen XD, Pan H, Li SQ. Ki67 for evaluating the prognosis of gastrointestinal stromal tumours: A systematic review and meta-analysis. Oncol Lett. 2022;23(6):189. Doi: 10.3892/ol.2022.13309.

[29] Davey MG, Hynes SO, Kerin MJ, Miller N, Lowery AJ. Ki-67 as a prognostic biomarker in invasive breast cancer. Cancers (Basel). 2021;13(17):4455. Published 2021 Sep 3. Doi:10.3390/cancers13174455.

[30] Müssig K, Wehrmann T, Dittmann H, Wehrmann M, Ueberberg B, Schulz S, et al. Expression of the proliferation marker Ki-67 associates with tumour staging and clinical outcome in differentiated thyroid carcinomas. Clin Endocrinol. 2012;77(1):139-45. Doi: 10.1111/j.1365 2265.2012.04343.x.

[31] Martin B, Paesmans M, Mascaux C, Berghmans T, Lothaire P, Meert AP, et al. Ki-67 expression and patient survival in lung cancer: Systematic review of the literature with meta analysis. Br J Cancer. 2004;91(12):2018-25. Doi: 10.1038/sj.bjc.6602233.

[32] Pan D, Wen D, Luo Y, Chen G, Yang H, Chen J, et al. The diagnostic and prognostic values of Ki-67/MIB-1 expression in thyroid cancer: A meta-analysis with 6,051 cases. Onco Targets Ther. 2017;10:3261-66. Doi: 10.2147/ott.s135593.

[33] Tian J, Chen W. Prediction of Ki-67 expression and malignant potential in gastrointestinal stromal tumours: Novel models based on CE-CT and serological indicators. BMC Cancer. 2024;24(1):1412. Doi: 10.1186/s12885-024-13172-y.

[34] Ranjbari N, Rahim F. The Ki-67/MIB-1 index level and recurrence of papillary thyroid carcinoma. Med Hypotheses. 2013;80(3):311-14. Doi: 10.1016/j.mehy.2012.12.015.
[35] Pujani M, Arora B, Singh SK, Tejwani N. Role of Ki-67 as a proliferative marker in lesions of thyroid. Indian J Cancer. 2010;47(3):304-07. Doi: 10.4103/0019-509X.64727.
[36] Nilsson JN, Siikanen J, Hedman C, Juhlin CC, Ihre Lundgren C. Pre-therapeutic measurements of iodine avidity in papillary and poorly differentiated thyroid cancer reveal associations with thyroglobulin expression, histological variants and Ki-67 index. Cancers. 2021;13(14):3627. Doi: 10.3390/cancers13143627.
[37] Louis DM, Nair LM, Vallonthaiel AG, Narmadha MP, Vijaykumar DK. Ki 67: A promising prognostic marker in early breast cancer- A review article. Indian J Surg Oncol. 2023;14(1):122-27. Doi: 10.1007/s13193-022-01631-6.
[38] Khangura N, Bhati S, Bhatia G, Goyal N, Khangura SK. A histomorphological study of meningiomas according to latest CNS 5th Edition WHO Classification 2021 and co-relation of Grading with Ki-67 proliferation index. J Med Sci Health. 2024;10(3):241-46. Doi: 10.46347/jmsh.v10.i3.24.186.
[39] Jin Z, Ye M, Sheng Y, Sun J, Zhang J, Chen Y, et al. Ki-67 in nasopharyngeal papillary adenocarcinoma: Clinicopathologic insights. Am J Surg Pathol. 2024;49(1):35-44. Doi: 10.1097/PAS.0000000000002321.

PARTICULARS OF CONTRIBUTORS:
1. Consultant Pathologist, Department of Pathology, Saveetha Medical College and Hospital, Chennai, Tamil Nadu, India.
2. Professor, Centre for Global Health Research, Saveetha Medical College and Hospital, Chennai, Tamil Nadu, India.
3. Professor, Department of Pathology, Malabar Medical College and Research Centre, Kozhikode, Kerala, India.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Dr. Navaneetha K Kumar,
Consultant Pathologist, Saveetha Medical College and Hospital, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Thandalam, Chennai, Tamil Nadu, India.
E-mail: abumunar05@gmail.com

PLAGIARISM CHECKING METHODS: [Jain H et al.]
- Plagiarism X-checker: Jun 04, 2025
- Manual Googling: Sep 22, 2025
- iThenticate Software: Sep 24, 2025 (9%)

ETYMOLOGY: Author Origin

EMENDATIONS: 7

AUTHOR DECLARATION:
- Financial or Other Competing Interests: None
- Was Ethics Committee Approval obtained for this study? NA
- Was informed consent obtained from the subjects involved in the study? NA
- For any images presented appropriate consent has been obtained from the subjects. NA

Date of Submission: **May 03, 2025**
Date of Peer Review: **Jun 23, 2025**
Date of Acceptance: **Sep 26, 2025**
Date of Publishing: **Jan 01, 2026**