The Prognostic Value of Ki67 in Phyllodes Tumor of the Breast: A Systematic Review and Meta-Analysis

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ABSTRACT

Many clinicopathological features have been examined as predictive factors for adverse outcomes in patients with phyllodes tumor (PT) of the breast, but there are still no definitive predictive markers to guide management, despite the persistent risk of recurrence, even in benign disease. Whether Ki67 has prognostic value in PT remains uncertain. We therefore conducted a systematic review and meta-analysis to examine whether Ki67 is associated with adverse clinical outcomes, especially recurrence, in patients with PT. The PubMed/MEDLINE, Web of Science, Scopus, Embase, and Cochrane Library databases were searched from inception to July 2024. Study characteristics and outcomes (recurrence and overall survival) according to Ki67 status were extracted from each eligible study, and pooled log odds ratios with 95% CI were derived using a fixed-effects model after testing for homogeneity of effect sizes with Cochran's Q-test. Five studies representing 280 cases were eligible for inclusion. The adverse outcome rate for the $Ki67^{high}$ (Ki67 >10% or >11.2%) population was 28.7% (95% CI 20.1 - 38.6%), while the adverse outcome rate for the $Ki67^{low}$ population was 9.4% (95% CI 5.4 - 13.5%). $Ki67^{high}$ was associated with an increased odds (log OR 1.34 (95% CI 0.65 - 2.02, p<0.001) of an adverse outcome compared with a $Ki67^{low}$ status. All five studies scored eight points on the Newcastle-Ottawa Scale, equivalent to "good" quality by AHRQ standards, and no significant publication bias was noted. This is the first meta-analysis of the predictive value of Ki67 in PT of the breast. A relatively high Ki67 index (greater than about 10%) is associated with recurrence. It is timely to re-evaluate the prognostic value of Ki67 in large retrospective cohorts with long follow-up to firmly establish whether it could contribute to identifying patients at risk of recurrence, especially those with histologically benign disease. Doing so could impact clinical practice by refining follow-up recommendations based on quality evidence.

Keywords: Ki67, phyllodes tumor, prognosis, proliferation, recurrence

INTRODUCTION

Phyllodes tumors (PTs) are rare (0.3 - 1% of all primary breast tumors) fibroepithelial breast neoplasms that carry a risk of recurrence and metastasis [1, 2]. PTs are classified into benign, borderline, and malignant grade disease according to histopathological criteria of stromal cellularity and atypia, mitotic count, stromal overgrowth, and the nature of the tumor border [3, 4]. Histopathological grading is important for prognostication and management: recurrence rates increase with higher tumor grades (10–17%, 14–25%, and 23–30% for benign, borderline, and malignant phyllodes tumors, respectively [3, 4]); benign tumors only very rarely metastasize [3]; and malignant tumors may benefit from adjuvant therapy such as postoperative radiotherapy, although the optimal management remains uncertain [5]. Metastasis of PTs heralds a usually dismal prognosis and death from the disease [6], but in almost all cases metastasis only occurs when the primary tumor is graded as malignant [3, 6].

Although complete excision is the gold standard treatment for PT, and complete excision with an adequate margin probably reduces subsequent recurrence risk, there is still debate about what constitutes an adequate margin to reduce risk of recurrence [5]. Other clinicopathological features such cellular atypia, mitotic rate, and stromal overgrowth have similarly been examined as predictive factors for recurrence, with conflicting results [3, 5]. Other protein biomarkers such as MMP-14, Six-1, PAX3, FoxC2, TWIST, CXCR4, VEGF, stromal Yes-associated protein (YAP), cellular E-cadherin, and CD10 have been associated with recurrent PT, while others have not (e.g., EGFR, HER2, membranous E-cadherin; MMPs 1, 2, 7, 9, 11, and 13; and TIMP 1, 2, and 3) [7]. There are still no definitive markers of recurrence or other outcomes such as survival to guide management, despite the persistent risk of recurrence, even in patients with benign disease. Although benign disease does not usually result in clinically serious sequelae, it would still be helpful to identify all individuals at risk of recurrence, even those with benign disease, to tailor surveillance strategies [8].

Ki67 is a nuclear antigen that is expressed in all phases of the cell cycle (G1, S, G2), and it is a reliable and widely used immunohistochemical biomarker of proliferation in routine histopathological practice, including in breast cancer [9]. While the mitotic index only reflects cells in M-phase, Ki67 detects other cells at different stages of proliferation (or arrest) and therefore provides different information about proliferation in the histopathological snapshot of tumor biology captured in a tumor section [9]. In PT, stromal expression of Ki67 has been reported to be 5-25% in benign tumors and 15-100% in malignant tumors, and this association with grade has been reported in several studies [10]. However, whether Ki67 has prognostic significance in PT remains uncertain.

We therefore conducted a systematic review and meta-analysis to examine whether Ki67 is associated with adverse clinical outcomes, especially recurrence, in patients with PT.

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METHODS

This meta-analysis is reported according to the 2020 Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) statement [11].

Eligibility criteria: The PICOS criteria for inclusion and exclusion were as follows: P (participants): studies of uni- or bilateral phyllodes tumors of the breast; I and C (intervention and control): studies in which Ki67/MIB1 was measured in phyllodes tumors by immunohistochemistry; O (outcome): studies that included the local recurrence rate or overall survival rate were included; S (study type): research articles published prior to July 11, 2024 were included. All review papers, conference abstracts, meta-analyses, editorial/comment papers, and case reports were excluded from the study.

Information sources and search strategy: The PubMed/MEDLINE, Web of Science, Scopus, Embase, and Cochrane Library databases were searched for reports meeting the inclusion criteria published before July 11, 2024. The following search strategies were used for each database: PubMed/MEDLINE: (phyllodes) AND (breast) AND ((ki67) OR (proliferation)) AND ((survival) OR (recurrence) OR (outcome)); Web of Science: (phyllodes) AND (breast) AND ((ki67) OR (proliferation)) AND (outcome); Scopus: TITLE-ABS-KEY (phyllodes) AND TITLE-ABS-KEY (breast) AND (TITLE-ABS-KEY (outcome) OR TITLE-ABS-KEY (survival) OR TITLE-ABS-KEY (recurrence)) AND (TITLE-ABS-KEY (ki67) OR TITLE-ABS-KEY (proliferation)); Cochrane: phyllodes tumor; Embase (1974 to 2024 July 11): (phyllodes and breast and (ki67 or proliferation) and (survival or recurrence or outcome)).mp. [mp=title, abstract, heading word, drug trade name, original title, device manufacturer, drug manufacturer, device trade name, keyword heading word, floating subheading word, candidate term word].

Selection, data collection, and data items: Two reviewers worked independently to screen the titles and abstracts of all literature retrieved from the database searches. After screening, two reviewers independently collected data from each report and input the data items (study country, outcome types, follow-up period, number of recurrences according to Ki67 status, clinicopathological characteristics, and total number of cases) into a data entry sheet. These sheets were then compared and combined, with discrepancies resolved by discussion with a third reviewer.

Risk of bias assessment: The Newcastle Ottawa Scale (NOS) [12] was used to assess the quality of prognostic/predictive studies with scores converted to AHRQ standards, i.e., good quality: 3 or 4 stars in the selection domain AND 1 or 2 stars in the comparability domain AND 2 or 3 stars in the outcome/exposure domain; fair quality: 2 stars in the selection domain AND 1 or 2 stars in the comparability domain AND 2 or 3 stars in the outcome/exposure domain; and poor quality: 0 or 1 star in the selection domain OR 0 stars in the comparability domain OR 0 or 1 stars in the outcome/exposure domain.

Statistical analysis : Analyses were performed in JASP v0.19 for Apple Silicon [13]. The log odds ratio (OR) with 95% confidence intervals (CI) was used to compare dichotomous variables, i.e., proportions of recurrences/deaths in patients with PTs with a Ki67 index >10% or >11.2%. Thresholds of 10% and 11.2% were selected as these were the thresholds used in the selected studies and were deemed sufficiently close to not unduly affect the analysis. Homogeneity of effect sizes were assessed with the test of residual heterogeneity (Cochran's Q) test, the results of which indicated that a fixed effects (Mantel-Haenszel) model was most appropriate for meta-analysis. Publication bias was assessed using a funnel plot and Egger's test. A p-value of 0.05 was deemed statistically significant.

RESULTS

Study selection: The flow chart of the study selection process is shown in Figure 1. Of 287 records identified in five databases, 145 were screened after removal of duplicates. Of these, 108 were excluded for not meeting the inclusion criteria. One report could not be retrieved from a source in China. Of 36 full reports assessed for eligibility, markers of proliferation (but not Ki67) were recorded, and in seven papers Ki67 was not associated with the outcome of interest. Nine papers did not record outcome data. Two studies [14, 15] were authored by the same research team and published in the same year and, on closer examination of the cohorts, they were found to be nearly identical apart from a few more benign PTs in one of the cohorts. We therefore selected the study with the larger cohort for inclusion [14]. After review, five reports were included in the final analysis [14, 16-19].

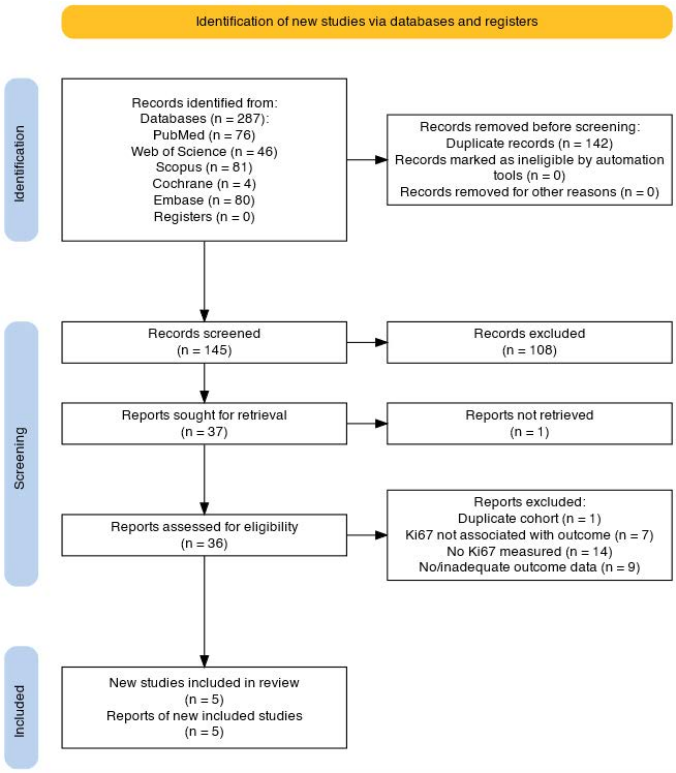

Figure 1. PRISMA flow cart of the study selection process.

Study characteristics and risk of bias/quality assessment: The characteristics of the included studies are shown in Table 1. All five studies were retrospective observational studies, and all five scored eight points on the Newcastle-Ottawa Scale, equivalent to "good" quality by AHRQ standards. Although follow-up varied, in all cases it was long enough to capture the most common period of recurrence (average 13.8 months) [20]. While all five studies reported recurrence (local and/or distant) data and two studies reported OS data [17, 19], one study only reported Ki67 associations with the OS (and not recurrence) outcome [17]. Three studies [14, 16, 18] applied a Ki67 threshold of >10%, while two studies [17, 19] applied a Ki67 threshold of 11.2%.

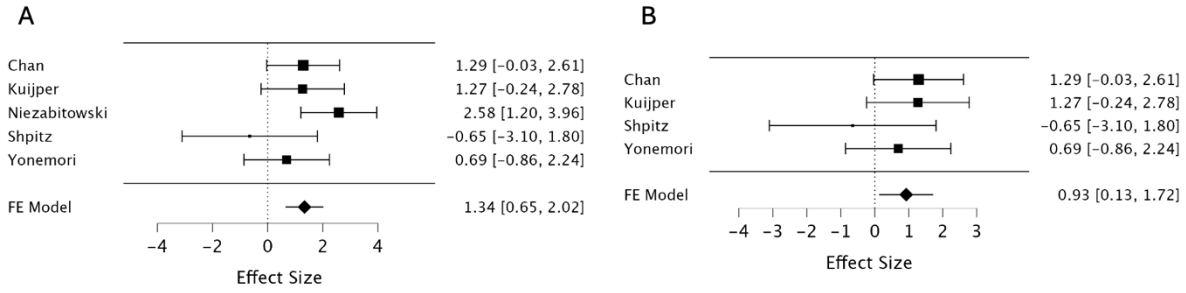

Figure 2. Forest plot showing the results of the meta-analysis of the relationship between Ki67 status and (A) any adverse outcome (local recurrence or OS) and (B) local recurrence alone.

Table 1. Characteristics of the included studies. Ki67^{high} was defined as >10% for [14, 16, 18] and >11.2% for [17, 19], as stated in the respective manuscripts, while an adverse outcome was either a recurrence (in four studies [14, 16, 18, 19]) or death from the disease (in one study [17]).

Author	Year	Country	Age	PT types	Total cases	Outcome type(s)	Follow-up period	Adverse outcome Ki67^{high}	Adverse outcome Ki67^{low}	Total number of Ki67^{high}	Total number of Ki67^{low}	Log OR (SE)	NOS	Quality
Chan [16]	2004	Taiwan	Benign 42.4 ± 14.3 years, malignant 44.3 ± 9.9 years	50 benign, 13 malignant	63	Recurrence	Mean 36 months (1-15 years)	4	3	19	44	1.29 (0.67)	8	Good
Kuijper [14]	2005	Netherlands	Benign 45.5 ± 16.8 years, borderline 57.9 ± 12.8 years, and malignant 54.3 ± 12.9 years	21 benign, 8 borderline, 11 malignant	40 (37 with follow-up)	Recurrence	Median 93 months (4-215 months)	6	4	14	23	1.27 (0.77)	8	Good
Niezabitowski [17]	2001	Poland	Median 49 (range 16-87)	52 benign, 24 borderline, 42 malignant	117	Overall survival	NS	10	3	31	86	2.58 (0.70)	8	Good
Shpitz [18]	2002	Israel	Median 41 (range 17-69)	16 benign, 4 borderline, 3 malignant	23 (22 with follow-up)	Recurrence	Median 52 months (27-102 months)	1	3	8	14	-0.65 (1.25)	8	Good
Yonemori [19]	2006	Japan	Median 47 (range 22-65)	20 benign, 5 borderline, 16 malignant	41	Recurrence	42 months (1-90 months)	6	3	22	19	0.69 (0.79)	8	Good

Results of individual studies

The pooled data consisted of five studies representing 280 cases. The overall adverse outcome rate was 15.4% (95% CI 11.6 - 20.1%). The adverse outcome rate for the Ki67^{high} population was 28.7% (95% CI 20.1 - 38.6%), while the adverse outcome rate for the Ki67^{low} population was 9.4% (95% CI 5.4 - 13.5%). The effect sizes (log OR) of Ki67^{high} scoring with an adverse outcome, together with their standard errors, are shown in Table 1 and ranged from -0.65 (1.25) to 2.58 (0.70).

Results of meta-analysis

As the effect sizes were homogeneous (test of residual heterogeneity Q-test p = 0.18), a fixed effects (Mantel-Haenszel) model was used. Ki67^{high} was associated with an increased odds (log OR 1.34 (95% CI 0.65 - 2.02, p<0.001) of an adverse outcome compared with a Ki67^{low} status (Figure 2A). Similarly, when examining associations with local recurrences alone (excluding [17]), Ki67^{high} status was still associated with an increased odds (log OR 0.93 (95% CI 0.13 - 1.72, p<0.02) of local recurrence compared with a Ki67^{low} status (Figure 2B).

Publication bias

No significant publication bias was noted in the funnel plot (Egger's test p = 0.06; Figure 3).

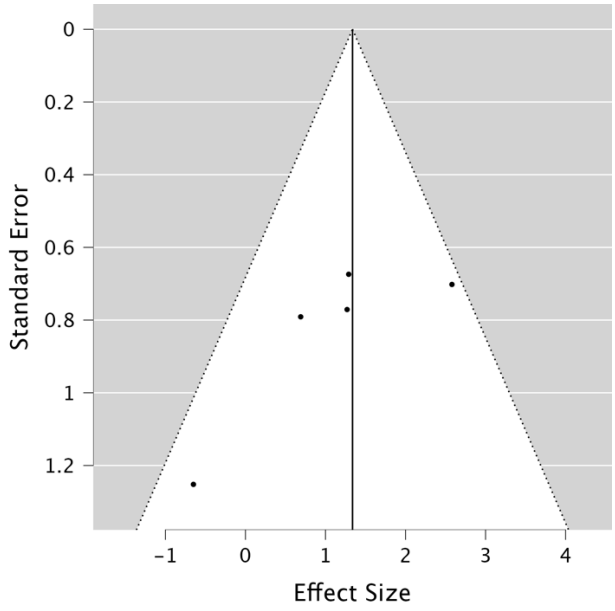

Figure 3. Funnel plot to examine publication bias.

DISCUSSION

As PT is a rare tumor, there are no large, prospective studies of PTs. However, previous retrospective studies have reported that various clinicopathological factors, including adequacy of resection, histopathological features, and tumor protein expression, are associated with clinical outcomes in patients with PT [7, 8]. Although PTs that clearly lie at the extreme ends of the diagnostic spectrum - such as those that are difficult to differentiate from fibroadenomas or those with overt features of malignancy - allow straightforward clinicopathological correlation and management planning, the disease still presents pathological and clinical challenges in practice. The absence of clear prognostic factors means that there are still no evidence-based guidelines for follow-up of patients with PT, meaning that some patients with benign lesions are subjected to unnecessary routine follow-up imaging, which may not be required [8]. Any indictor of an adverse prognostic course, especially in patients with clinically benign lesions, would be useful to tailor follow-up and spare the costs and resources associated with surveillance imaging or, conversely, to divert resources to those at greatest risk of future recurrence.

This prompted us to conduct this systematic review and meta-analysis of the prognostic significance of the Ki67 proliferation marker in patients with PT. Although we identified only five studies that specifically examined the prognostic value of Ki67 in patients with PT, our analysis shows that patients with PT with high expression of Ki67, here defined as >10% or >11.2% expression in tumor stroma cells, are at significantly increased risk of an adverse outcome, especially recurrence. Although the small sizes of the included studies and a lack of relevant data precluded formal subgroup analysis of the prognostic significance of Ki67 in patients with benign and borderline disease alone, the available data seem to suggest that the association between high Ki67 and recurrence is not limited to malignant disease. Indeed, all seven recurrences reported in Chan et al. [16] occurred after resection of benign primary lesions, four of which had Ki67 indices >10%. In the other included reports, even though some initially benign primary lesions went on to recur, their Ki67 status was unclear. Our analysis suggests that a Ki67 index greater than about 10% may be an indicator of the need for extra vigilance during follow-up regardless of other clinical or histopathological features. Even though locally recurrent PT often presents clinically and not through imaging [8, 21], establishing Ki67 as an adverse prognostic indicator could be helpful for identifying the subset of patients who might benefit from focused radiological follow-up.

There have been previous attempts to devise predictive models of the clinical behavior of PT. For instance, Tan et al. developed a nomogram that included atypia, mitoses, stromal overgrowth, and surgical margin status that predicted recurrence-free survival up to 10 years [6], which was subsequently validated in several independent cohorts from around the world [22-24]. Interestingly, stromal mitotic activity was usually associated with recurrence even when the other included parameters were not [22, 23]. Given our findings and that proliferation as measured by mitosis counts are likely to be critical factors related to recurrence in PT, there is a need to re-evaluate the relationship between Ki67 indices and clinical outcomes to establish whether this simple and cost-effective ancillary diagnostic test should be integrated into clinical nomograms, especially with regard to predicting responses in benign and borderline tumors.

This meta-analysis has several limitations. As noted above, only five studies were available for analysis, all of which were retrospective, so selection bias cannot be excluded. The sample size in the meta-analysis was relatively small, limiting the level of evidence. Due to the limited number of available studies, we needed to combine results for similar but slightly different Ki67 thresholds (10% and 11.2%), which may have introduced some error into the results. Furthermore, different Ki67 cut-points were not considered, so the optimal threshold for prognostication remains uncertain. Although the results of the meta-analysis remained the same when the study reporting overall survival outcomes was excluded, we had to combined outcomes to fully evaluate the small number of available studies. Finally, Ki67 measurements were not standardized across laboratories, which may have influenced the results.

Nevertheless, this is the first meta-analysis of the predictive value of Ki67 in PT of the breast, and the results clearly show that a relatively high Ki67 index (greater than about 10%) is associated with recurrence. Given that Ki67 is widely used in clinical practice, it seems timely to re-evaluate the prognostic value of the marker in large retrospective cohorts with long follow-up to firmly establish whether it could contribute to identifying patients at risk of recurrence, especially those with histologically benign disease. Doing so could impact clinical practice by refining follow-up recommendations based on quality evidence and sparing unnecessary imaging in low-risk individuals.

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Competing Interest: None

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