Lymphovascular space invasion in endometrial carcinoma: A prognostic factor independent from molecular signature

Lymphovascular space invasion in endometrial carcinoma: a prognostic factor independent from molecular signature

Antonio RAFFONE, MD[1,2]; Antonio TRAVAGLINO, MD[3,4]; Diego RAIMONDO, MD, PhD[2]; Daniele NEOLA, MD[1]; Manuela MALETTA, MD[2]; Angela SANTORO, MD, PhD[4]; Luigi INSABATO, MD, PhD[3]; Paolo CASADIO, MD[2]; Francesco FANFANI, MD, PhD[5,6]; Gian Franco ZANNONI, MD, PhD[4]; Fulvio ZULLO, MD, PhD[1]; Renato SERACCHIOLI, MD, PhD[2*] and Antonio MOLLO, MD, PhD[7*]

[1]Gynecology and Obstetrics Unit, Department of Neuroscience, Reproductive Sciences and Dentistry, School of Medicine, University of Naples Federico II, Naples, Italy

[2] Division of Gynaecology and Human Reproduction Physiopathology, Department of Medical and Surgical Sciences (DIMEC). IRCCS Azienda Ospedaliero-Universitaria di Bologna. S. Orsola Hospital. University of Bologna, Via Massarenti 13, Bologna 40138, Italy

[3] Anatomic Pathology Unit, Department of Advanced Biomedical Sciences, School of Medicine, University of Naples Federico II, Naples, Italy

[4] Gynecopathology and Breast Pathology Unit, Department of Woman's Health Science, Agostino Gemelli University Polyclinic, Rome, Italy

[5] Department of Woman and Child Health and Public Health, Division of Gynecologic Oncology, Fondazione Policlinico Universitario A. Gemelli, Istituto di Ricovero e Cura a Carattere Scientifico (IRCCS), Rome, Italy

[6] Department of Women and Child Health and Public Health, Università Cattolica del Sacro Cuore, Rome, Italy.

[7] Gynecology and Obstetrics Unit, Department of Medicine, Surgery and Dentistry "Schola Medica Salernitana", University of Salerno, 84081 Baronissi, Italy

*: The authors equally contributed to the study and shared the last authorship.

CORRESPONDING AUTHORS: Antonio TRAVAGLINO, Anatomic Pathology Unit, Department of Advanced Biomedical Sciences, School of Medicine, University of Naples Federico II, Via Sergio Pansini, 5, Naples, 80131, Italy, Email: antonio.travaglino@unina.it; antonio.travaglino.ap@gmail.com

Paolo CASADIO, Division of Gynaecology and Human Reproduction Physiopathology, Department of Medical and Surgical Sciences (DIMEC). IRCCS Azienda Ospedaliero-Universitaria di Bologna. S. Orsola Hospital. University of Bologna, Via Massarenti 13, Bologna 40138, Italy, Email: p.paolocasadio@gmail.com

ABSTRACT

Background: The 2020 ESGO/ESTRO/ESP guidelines stratify the prognosis of endometrial carcinoma (EC) patients combining The Cancer Genome ATLAS (TCGA) molecular signature and pathological factors, including lymphovascular space invasion (LVSI). However, little is known about the prognostic independence of LVSI from molecular signature.

Aim: To assess whether the prognostic value of LVSI is independent from the TCGA signature.

Material and methods: A systematic review and meta-analysis was performed by searching 5 electronic databases from their inception to March 2021. All peer-reviewed studies reporting assessing LVSI as a prognostic factor independent from the TCGA groups in EC were included. Multivariate HRs with 95% confidence interval (CI) were pooled separately for overall survival (OS), disease-specific survival (DSS) and disease-free survival (DFS). The absence of LVSI was considered as a reference. In DFS analyses, locoregional and distant recurrence were separately considered for one study.

Results: Six studies with 3,331 patients were included in the systematic review and three studies with 2,276 patients in the meta-analysis. LVSI showed a pooled multivariate HR of 1.818 (CI 95%, 1.378-2.399) for OS, 1.849 (CI 95%, 1.194-2.863) for DSS, 1.377 (CI 95%, 1.008-1.880) for DFS excluding one study, 1.651 (CI 95%, 1.044-2.611) for DFS additionally considering locoregional recurrence from one study, and 1.684 (CI 95%, 1.05-2.701) for DFS additionally considering distant recurrence from the same study.

Conclusion: LVSI has a prognostic value independent of TCGA signature, as well as age and adjuvant treatment, increasing the risk of death of any cause, death due to EC and recurrent or progressive disease by 1.5-2 times.

KEYWORDS: cancer; tumor; endometrium; prognosis; treatment; risk assessment; PROMISE.

INTRODUCTION

In 2020, endometrial carcinoma (EC) was the most common gynecological cancer in Europe, with an incidence of 31.2% and a 5-year prevalence of 34.1% [1]. In the last decades, EC has showed a 300% increase in the number of deaths despite a lower increase in incidence (80%) [1-10]. Such data seems due to an inaccurate risk stratification underlying patient management [2,3,10-16].

In the attempt of improving the risk stratification system, in 2020, the European Society of Gynaecological Oncology (ESGO), the European Society for Radiotherapy and Oncology (ESTRO) and the European Society of Pathology (ESP) drafted joint guidelines for the management of EC, recommending the assessment of molecular signature as additional prognostic factor to be integrated with classic pathological factors, such as histotype, International Federation of Gynaecology and Obstetrics (FIGO) stage, FIGO grade, myometrial invasion, and lymphovascular space invasion (LVSI) [21]. In fact, in 2013, The Cancer Genome Atlas (TCGA) Research Network identified four prognostic groups of ECs with a different molecular signature: POLE-mutated/ultramutated (POLEmt), microsatellite-instable/hypermutated (MSI), copy-number-high/p53-mutated (p53mt), and no specific molecular profile (NSMP) [17]. Afterwards, The Proactive Molecular Risk Classifier for Endometrial Cancer (ProMise), which adopted immunohistochemistry (cheap, fast and widely available) as a surrogate of sequencing techniques, was demonstrated to be prognostically informative and more applicable in clinical practice [2,12,13].

However, how the molecular signature can be integrated with classic pathological factors is still under investigation. In particular, the histotype seems to have a crucial prognostic value independently of the TCGA groups, with non-endometrioid carcinomas having a worse prognosis in each TCGA group [6]. On the other hand, deep myometrial invasion has shown to affect the risk of recurrence independently from the TCGA groups, but not the risk of death of any cause [11]. On the contrary, the prognostic independence of LVSI from the molecular signature has not been enough investigated.

The aim of this study was to evaluate whether the prognostic value of LVSI is independent from the TCGA group, through a systematic review and meta-analysis.

MATERIALS AND METHODS

Study protocol

All study steps were defined according to an *a priori* designed protocol and were independently performed by two authors. In the case of disagreement, a discussion with a third author was adopted as resolution method.

The study was reported according to the Preferred Reporting Item for Systematic Reviews and Meta-analyses (PRISMA) statement and checklist [23].

Search strategy

MEDLINE, Web of Sciences, Google Scholar, Scopus and ClinicalTrial.gov were searched from their inception to May 2021 through several combinations of the following text words: “endometr*”;

“carcinoma”; “cancer”; “tumor”; “neoplasia”; “malignancy”; “LVSI”; lymphovascular space invasion”; “lymph*”; “immunohistochem*”; “marker”; “TCGA”; “ATLAS”; “genome”; “ProMisE”; “Proactive Molecular Risk Classifier”; “TransPORTEC”; “PORTEC”; “prognosis”; “survival”; “hypermutated”; “mismatch repair”; “microsatellite instability”; “MMR”; “MSI”; “MLH1”; “MSH2”; “MSH6”; “PMS2”; “EPCAM”; “ultramutated”; “POLE”; “copy number”; “TP53”; “tumor protein 53”; “p53”; “ESMO”, “ESGO”, “ESTRO”, “ESP”. All references from full-text screened studies were assessed for eligible studies.

Study selection

Peer-reviewed studies with extractable data about LVSI as a prognostic factor independent of the TCGA groups in EC were included. In detail, we selected all studies reporting multivariate hazard ratios (HR) for LVSI and assessing the TCGA group as a variable in the multivariate analysis. Case reports and reviews were excluded. In the cases of overlapping data between two studies (i.e. same period of enrollment, institution and/or results), only the study with higher sample size was considered for the analysis.

Data extraction

Data were extracted without modifications and in accordance with the PICO items [23]. “Population” of the systematic review and meta-analysis consisted of women with EC.

“Intervention” (or risk factor) consisted of the presence of LVSI. For included studies that substratified LVSI, “substantial” LVSI was considered as presence of LVSI.

“Comparator” consisted of the absence of LVSI. For included studies that substratified LVSI, “absent” and “mild” LVSI were considered as absence of LVSI.

“Outcomes” consisted of overall survival (OS) as primary outcome, and disease-specific survival (DSS) and disease-free survival (DFS) as secondary outcomes. In particular, OS (or time to death) was defined as time from surgery until death of any cause. DSS (or time to death from disease) was defined as time from surgery until death due to EC. DFS (or time to recurrence or progression) was defined as time from surgery until there is evidence of recurrent or progressive disease (this was based on either clinical evidence of recurrence or imaging confirmation of recurrence) or if death from the disease occurred prior to the censoring date.

Risk of bias within studies evaluation

The risk of bias within studies was evaluated following The Methodological Index for Non-Randomized Studies (MINORS) [24]. Each included study was judged for 7 applicable domains related to risk of bias: 1) Aim (was the aim clear?); 2) Patient selection (were patients randomly or consecutively selected?); 3) Prospective collection of data (were data prospectively collected?); 4) Endpoints appropriate to the study (were OS, DSS and DFS assessed?); 5) Unbiased evaluation of the study endpoints (were endpoints assessed without bias?); 6) Follow-up time (was the follow-up at least 2 years?); 7) Lost to follow-up (were patients lost to follow-up less than 5% of the study population?).

Judgments for each domain were the following: “low risk”, “unclear risk” or “high risk” of bias based on information was “reported and adequate”, “not reported” and “reported but inadequate”, respectively.

Data synthesis

For our analyses, we considered multivariable OS, DSS and DFS analyses that assessed the prognostic value of LVSI and considered the TCGA group as a variable from the included studies. In particular, all included studies used cox proportional hazard models.

Pooled HRs between EC women with and without LVSI were calculated based on the above-mentioned multivariable analyses, separately for death of any cause, death due to EC, and recurrence. The absence of LVSI was considered as a reference. HRs with 95% confidence interval (CI) were graphically reported as individual and pooled estimates on forest plots.

The random effect model of DerSimonian and Laird was used in the analyses.

Statistical heterogeneity among studies was evaluated by using the inconsistency index I^2, as previously described [25-27]. In particular, heterogeneity was judged as: null for $I^2=0\%$, minimal for $0<I^2<25\%$, low for $25\leq I^2<50\%$, moderate for $50\leq I^2<75\%$ and high for $I^2\geq 75\%$.

Comprehensive Meta-Analysis (Biostat,14 North Dean Street, Englewood, NJ 07631, USA) and Review Manager v. 5.4 (Copenhagen: The Nordic Cochrane Centre, Cochrane Collaboration, 2014) were used as software.

RESULTS

Study selection

Electronic databases searches led to 12,558 studies. Of them, 4,186 remained after duplicates removal and 72 after title screening. Abstract screening led to 17 studies, which were evaluated for eligibility [3,13-15,28-40]. Of them, 11 were excluded because of the absence of multivariable survival analysis for LVSI [13,30,32,34-36] or the absence of LVSI as a prognostic factor [29,31,33,37,38]. Therefore, 6 studies with 3,331 EC women were included in the systematic review [3,14,15,28,39,40]. Lastly, 2 studies were excluded from the meta-analysis due to overlapping patient data [3,40] with another included study [39], while one study was excluded because multivariate survival analysis was reported only for the NSMP group [14]. In conclusion, three studies with 2,276 EC women were included in the meta-analysis [15,28,39] (Figure S1).

Included studies and patients

Each included study performed *ad hoc* analyses on samples from previous cohorts of EC patients. In particular, the cohort was retrospective for 4 studies [3,14,39,40], prospective for one study [28], and provided from a randomized controlled trial for the latter one [15].

No included study adopted pathological criteria for patient selection, except for two studies [15,28] which only assessed endometrioid ECs (Table S1).

Our study population had a age which ranged from 41 to 90 years, and a mean follow-up which ranged from 60.6 to 131 months. ECs had endometrioid histotype in 85.7% of cases, International Federation of Gynecology and Obstetrics (FIGO) grade 3 in 27.3%, LVSI in 22.3%, and FIGO stage I in 73.8%. Women underwent adjuvant treatment in 57.7% of cases (Table S2). Regarding TCGA groups, 6.8% of ECs were POLE-mt, 29.3% were MMR-d, 50% were NSMP, and 13.8% were p53-abn (Table S3).

Risk of bias within studies evaluation

All included studies were at low risk of bias for each domain, except for the “Endpoints appropriate to the study” and “Unbiased evaluation of the study endpoints” domains.

In detail, the study by Stelloo et al. [15] was judged at unclear risk of bias for the "Endpoints appropriate to the study" because it did not consider DSS as a study endpoint and assessed separately locoregional and distant recurrence for DFS. On the other hand, the study by Kommoss et al. was judged at high risk of bias because multivariate survival analysis was reported only for the NSMP group [14].

Data synthesis

Three out of six studies were included in the meta-analysis [15,28,39], as one study was excluded due to the high risk of bias in the "Unbiased evaluation of the study endpoints" domain [14], and two studies [3,40] were excluded due to overlapping data with another study [39]. In particular, all three studies were suitable for OS analysis, while 2 were suitable for DSS and DFS [3,14,28,39,40]. In detail, the study by Stelloo et al [15] was excluded from DSS analysis because it did not consider DSS as a study endpoint, while it was included in additional DFS analysis because it assessed separately locoregional and distant recurrence. Therefore, DFS analyses consisted of 3 separate analyses: one excluding the study by Stelloo et al, one including such study as locoregional recurrence, and one including it as distant recurrence.

Pooled HR of LVSI was:

- 1.818 (CI 95%, 1.378-2.399; p=0.0; I^2=0%) for OS (Figure 1);
- 1.849 (CI 95%, 1.194-2.863; p=0.006; I^2 not calculable) for DSS (Figure 2);
- 1.377 (CI 95%, 1.008-1.880; p=0.044; I^2 not calculable) for DFS excluding the study by Stelloo et al. [15] (Figure 3);
- 1.651 (CI 95%, 1.044-2.611; p=0.032; I^2=45.7%) for DFS including the study by Stelloo et al. [15] as locoregional recurrence (Figure 4);
- 1.684 (CI 95%, 1.05-2.701; p=0.03; I^2=51.5%) for DFS including the study by Stelloo et al. [15] as distant recurrence (Figure 5).

DISCUSSION

This study shows that, in EC patients, LVSI has a prognostic value independent of TCGA groups, as well as age and adjuvant treatment. In particular, the presence of LVSI increased the risk of death of any cause, death due to EC and recurrent or progressive disease by 1.5-2 times. These findings may be the first pooled data in the field.

Since President Obama's 2014 State of the Union address [41,42], Precision Medicine initiative has been essential in oncological studies [42,43]. Precision medicine appears as the most promising approach to cancer, recommending measures tailored to the individual, with the paradigm “the right therapy, at the right time, in the right patient...” [44]. In fact, rather than classifying tumors from the organs in which they originated, they may be classified based on the molecular signature [42]. Among all cancers, this approach has provided the most promising results in EC, where TCGA findings have shown the potential to subvert the negative epidemiological trend observed in the last two decades and due to an inaccurate risk assessment [1-11,13,17].

On these bases, in the attempt to improve and tailor the risk stratification of EC women, the 2020 ESGO/ESTRO/ESP guidelines for the management of patients with EC recommend to integrate classic pathological prognostic factors with TCGA molecular signature [21]. However, the allocation of a patient to a specific risk group based on such an integration should presuppose prognostic independence between factors [11]. To date, while the prognostic independence of molecular signature from pathological factors appears demonstrated [3,13-17,28-40], the opposite has not been well-assessed yet. In detail, while data sustaining the at least partial prognostic independence of histotype and deep myometrial invasion have been recently provided [6,11], little is known about LVSI, with only few studies assessing its impact in a specific TCGA group. In particular, while LVSI was not associated with an increased risk of tumor recurrence or progression [32] and death from disease [45] in POLE-mt ECs, it appeared as an independent predictor of poor outcome in the MSI group [46,47]. Lastly, in the study by Stelloo et al., LVSI was the only pathological prognostic factor independent of TCGA groups for recurrence and death of any cause [15].

Regarding the integrated risk stratification proposal, the 2020 ESGO/ESTRO/ESP guidelines upgrade the risk group from low or intermediate to high–intermediate for stage I MMRd/NSMP endometrioid ECs based on the presence of LVSI. Therefore, LVSI status would affect the recommendation of adjuvant treatment in these patients. On the contrary, no role has been attributed to LVSI in the risk stratification of POLE-mt and p53-mt ECs [21].

Our results seem to support the use of LVSI as additional prognostic factor to be integrated with TCGA signature. The integration of molecular signature and pathological factors would ensure a more tailored management of patients in accordance with the principles of the precision medicine.

On the one hand, this might reduce the under- and overtreatment of EC women observed in the last years [4,6,8,11]; on the other hand, it might reduce the risk of wrong conclusion from oncological trials as patients with different prognosis would not be lumped together based on histological examination alone.

However, in order to further confirm 2020 ESGO/ESTRO/ESP recommendations and to obtain a more and more tailored management of EC patients, additional studies are needed to separately assess the prognostic value of LVSI in each TCGA group. This is necessary as each TCGA group is differently affected from pathological factors, with the POLEmt group appearing as the group least affected [4]. Indeed, the impossibility to provide pooled data about LVSI prognostic independence separately in each TCGA group may be the major limitation of our study. In addition, there was only a small number of studies which were suitable for our analysis. Another point to be clarified is the criterion to categorize LVSI. In fact, in one of the included studies, as well as in the ESGO/ESTRO/ESP guidelines [21], LVSI was dichotomized as “absent/focal” *vs* “substantial” [15,21]; such system was also showed to work in cervical cancer [48]. By contrast, two of the included studies dichotomized LVSI as “absent” *vs* “present” [28,39]. Lastly, we could not assess the prognostic independence of LVSI from other histological factors, as only TCGA group, age and adjuvant treatment were considered in the multivariate analysis of all individual studies. These limitations highlight the great need for further studies in the field.

CONCLUSION

In EC patients, LVSI has a prognostic value independent of TCGA signature, as well as age and adjuvant treatment, increasing the risk of death of any cause, death due to EC and recurrent or progressive disease by 1.5-2 times.

Further studies are necessary to confirm these findings and to assess the prognostic impact of LVSI separately in each TCGA group.

CONTRIBUTION

AR and AT independently assessed electronic search, eligibility of the studies, inclusion criteria, risk of bias, data extraction and data analysis. DR, DN, MM, PC and AS contributed to the elaboration of methods for risk of bias assessment, data extraction and analysis. AR, AT, DN, LI, FZ, GFZ, AM and RS conceived the study; DR, FR, PC, AS and LI worked on the design of the study; AR, AT, DN, DR, FR and LC worked on the manuscript preparation; PC, LI, FZ, GFZ, AM and RS supervised the whole study.

CONFLICT OF INTEREST STATEMENT

Authors report no conflict of interest.

FUNDING INFORMATION

No financial support was received for this study.

REFERENCES LIST

1. World Health Organization. GLOBOCAN 2018: estimated cancer incidence, mortality and prevalence worldwide in 2018, 2018. Available: http://gco.iarc.fr/today/data/factsheets/cancers/24- Corpus-uteri-fact-sheet.pdf.
2. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2015. CA Cancer J Clin. 2015 Jan-Feb;65(1):5-29. doi: 10.3322/caac.21254.
3. Talhouk A, McConechy MK, Leung S, et al. A clinically applicable molecular-based classification for endometrial cancers. Br J Cancer. 2015 Jul 14;113(2):299-310. doi: 10.1038/bjc.2015.190.
4. Raffone A, Travaglino A, Mascolo M, et al. TCGA molecular groups of endometrial cancer: Pooled data about prognosis. Gynecol Oncol. 2019 Nov;155(2):374-383. doi: 10.1016/j.ygyno.2019.08.019.
5. Travaglino A, Raffone A, Gencarelli A, et al. TCGA Classification of Endometrial Cancer: the Place of Carcinosarcoma. Pathol Oncol Res. 2020 Oct;26(4):2067-2073. doi: 10.1007/s12253-020-00829-9.
6. Travaglino A, Raffone A, Stradella C, et al. Impact of endometrial carcinoma histotype on the prognostic value of the TCGA molecular subgroups. Arch Gynecol Obstet. 2020 Jun;301(6):1355-1363. doi: 10.1007/s00404-020-05542-1.
7. Travaglino A, Raffone A, Mollo A, et al. TCGA molecular subgroups and FIGO grade in endometrial endometrioid carcinoma. Arch Gynecol Obstet. 2020 May;301(5):1117-1125. doi: 10.1007/s00404-020-05531-4.
8. Raffone A, Travaglino A, Mascolo M, et al. Histopathological characterization of ProMisE molecular groups of endometrial cancer. Gynecol Oncol. 2020 Apr;157(1):252-259. doi: 10.1016/j.ygyno.2020.01.008.
9. Travaglino A, Raffone A, Mascolo M, et al. TCGA Molecular Subgroups in Endometrial Undifferentiated/Dedifferentiated Carcinoma. Pathol Oncol Res. 2020 Jul;26(3):1411-1416. doi: 10.1007/s12253-019-00784-0.
10. Travaglino A, Raffone A, Mascolo M, et al. Clear cell endometrial carcinoma and the TCGA classification. Histopathology. 2020 Jan;76(2):336-338. doi: 10.1111/his.13976.
11. Raffone A, Travaglino A, Raimondo D, et al. Prognostic value of myometrial invasion and TCGA groups of endometrial carcinoma. *Gynecol Oncol*. 2021. doi:10.1016/j.ygyno.2021.05.029
12. Hoang LN, McConechy MK, Köbel M, et al. Histotype-genotype correlation in 36 high-grade endometrial carcinomas. Am J Surg Pathol. 2013 Sep;37(9):1421-32. doi: 10.1097/PAS.0b013e31828c63ed.
13. Talhouk A, McConechy MK, Leung S, et al. Confirmation of ProMisE: A simple, genomics-based clinical classifier for endometrial cancer. Cancer. 2017 Mar 1;123(5):802-813. doi: 10.1002/cncr.30496.

14. Kommoss S, McConechy MK, Kommoss F et al. Final validation of the ProMisE molecular classifier for endometrial carcinoma in a large population-based case series. Ann Oncol. 2018 May 1;29(5):1180-1188. doi: 10.1093/annonc/mdy058.
15. Stelloo E, Nout RA, Osse EM, et al. Improved Risk Assessment by Integrating Molecular and Clinicopathological Factors in Early-stage Endometrial Cancer-Combined Analysis of the PORTEC Cohorts. Clin Cancer Res. 2016 Aug 15;22(16):4215-24. doi: 10.1158/1078-0432.CCR-15-2878.
16. Stelloo E, Bosse T, Nout RA, et al. Refining prognosis and identifying targetable pathways for high-risk endometrial cancer; a TransPORTEC initiative. Mod Pathol. 2015 Jun;28(6):836-44. doi: 10.1038/modpathol.2015.43.
17. Getz G, Gabriel SB, Cibulskis K, et al. Integrated genomic characterization of endometrial carcinoma. *Nature*. 2013;497(7447). doi:10.1038/nature12113
18. Raffone A, Travaglino A, D'Antonio A, et al. BAG3 expression correlates with the grade of dysplasia in squamous intraepithelial lesions of the uterine cervix. Acta ObstetGynecol Scand. 2020 Jan;99(1):99-104. doi: 10.1111/aogs.13716.
19. Travaglino A, Raffone A, Saccone G, et al. Nuclear expression of β-catenin in endometrial hyperplasia as marker of premalignancy. APMIS. 2019 Nov;127(11):699-709. doi: 10.1111/apm.12988.
20. Raffone A, Travaglino A, Saccone G, et al. Diagnostic and prognostic value of ARID1A in endometrial hyperplasia: a novel marker of occult cancer. APMIS. 2019 Sep;127(9):597-606. doi: 10.1111/apm.12977.
21. Concin N, Matias-Guiu X, Vergote I, et al. ESGO/ESTRO/ESP guidelines for the management of patients with endometrial carcinoma. *Int J Gynecol Cancer*. 2021;31(1). doi:10.1136/ijgc-2020-002230
22. Singh N, Hirschowitz L, Zaino R, et al. Pathologic Prognostic Factors in Endometrial Carcinoma (Other Than Tumor Type and Grade). *Int J Gynecol Pathol*. 2019;38(1). doi:10.1097/PGP.0000000000000524
23. Moher D, Shamseer L, Clarke M, et al. Preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P) 2015 statement. Systematic Reviews 2015;4:1
24. Slim K, Nini E, Forestier D et al. Methodological index for non-randomized studies (minors): development and validation of a newinstrument. ANZ J Surg. 2003;73(9):712-6
25. Travaglino A, Raffone A, Saccone G, et al. Endometrial hyperplasia and risk of coexistent cancer: WHO vs EIN criteria. Histopathology. 2019 Apr;74(5):676-687.
26. Raffone A, Travaglino A, Saccone G, et al. Endometrial hyperplasia and progression to cancer: which classification system stratifies the risk better? A systematic review and meta-analysis. Arch Gynecol Obstet. 2019 May;299(5):1233-1242.

27. Raffone A, Travaglino A, Saccone G, et al. Management of women with atypical polypoid adenomyoma of the uterus: A quantitative systematic review. Acta ObstetGynecol Scand. 2019 Feb 3. doi: 10.1111/aogs.13553. [Epub ahead of print]
28. Cosgrove CM, Tritchler DL, Cohn DE, et al. An NRG Oncology/GOG study of molecular classification for risk prediction in endometrioid endometrial cancer. Gynecol Oncol. 2018 Jan;148(1):174-180. doi: 10.1016/j.ygyno.2017.10.037. Epub 2017 Nov 11
29. Eriksson LSE, Nastic D, Lindqvist PG, et al. Sonographic, demographic characteristics, and the Proactive Molecular Risk Classifier for Endometrial cancer (ProMisE) in the prediction of tumor recurrence or progression . *Ultrasound Obstet Gynecol*. 2020. doi:10.1002/uog.23573
30. Britton H, Huang L, Lum A, et al. Molecular classification defines outcomes and opportunities in young women with endometrial carcinoma. *Gynecol Oncol*. 2019;153(3):487-495. doi:10.1016/j.ygyno.2019.03.098
31. Conlon N, Da Cruz Paula A, Ashley CW, et al. Endometrial carcinomas with a “serous” Component in young women are enriched for DNA mismatch repair deficiency, lynch syndrome, and POLE exonuclease domain mutations. *Am J Surg Pathol*. 2020;44(5):641-648. doi:10.1097/PAS.0000000000001461
32. He D, Wang H, Dong Y, et al. POLE mutation combined with microcystic, elongated and fragmented (MELF) pattern invasion in endometrial carcinomas might be associated with poor survival in Chinese women. *Gynecol Oncol*. 2020;159(1):36-42. doi:10.1016/j.ygyno.2020.07.102
33. Dubil EA, Tian C, Wang G, et al. Racial disparities in molecular subtypes of endometrial cancer. *Gynecol Oncol*. 2018;149(1):106-116. doi:10.1016/j.ygyno.2017.12.009
34. Haraga J, Nakamura K, Haruma T, et al. Molecular characterization of second primary endometrial cancer. *Anticancer Res*. 2020;40(7). doi:10.21873/ANTICANRES.14370
35. León-Castillo A, Gilvazquez E, Nout R, et al. Clinicopathological and molecular characterisation of ‘multiple-classifier’ endometrial carcinomas. *J Pathol*. 2020;250(3):312-322. doi:10.1002/path.5373
36. Rau TT, Bettschen E, Büchi C, et al. Prognostic impact of tumor budding in endometrial carcinoma within distinct molecular subgroups. *Mod Pathol*. 2021;34(1):222-232. doi:10.1038/s41379-020-0626-9
37. DeLair DF, Burke KA, Selenica P, et al. The genetic landscape of endometrial clear cell carcinomas. J Pathol 2017;243:230-41. doi:10.1002/path.4947
38. Bosse T, Nout RA, McAlpine JN, et al. Molecular Classification of Grade 3 Endometrioid Endometrial Cancers Identifies Distinct Prognostic Subgroups. *Am J Surg Pathol*. 2018;42(5). doi:10.1097/PAS.0000000000001020

39. Talhouk A, Derocher H, Schmidt P, et al. Molecular subtype not immune response drives outcomes in endometrial carcinoma. *Clin Cancer Res*. 2019;25(8):2537-2548. doi:10.1158/1078-0432.CCR-18-3241
40. Karnezis AN, Leung S, Magrill J, et al. Evaluation of endometrial carcinoma prognostic immunohistochemistry markers in the context of molecular classification. *J Pathol Clin Res*. 2017;3(4):279-293. doi:10.1002/cjp2.82
41. Collins FS, Varmus H. A New Initiative on Precision Medicine. *N Engl J Med*. 2015;372(9). doi:10.1056/nejmp1500523
42. Barroilhet L, Matulonis U. The NCI-MATCH trial and precision medicine in gynecologic cancers. *Gynecol Oncol*. 2018;148(3). doi:10.1016/j.ygyno.2018.01.008
43. Coyne GOS, Takebe N, Chen AP. Defining precision: The precision medicine initiative trials NCI-MPACT and NCI-MATCH. *Curr Probl Cancer*. 2017;41(3). doi:10.1016/j.currproblcancer.2017.02.001
44. Coleman RL, Matulonis UA. Precision medicine. Gynecol Oncol. 2016 Apr;141(1):1. doi: 10.1016/j.ygyno.2016.03.017. PMID: 27016221.
45. McAlpine JN, Chiu DS, Nout RA, et al. Evaluation of treatment effects in patients with endometrial cancer and POLE mutations: An individual patient data meta-analysis. *Cancer*. 2021:1-14. doi:10.1002/cncr.33516
46. Loukovaara M, Pasanen A, Bützow R. Mismatch repair protein and MLH1 methylation status as predictors of response to adjuvant therapy in endometrial cancer. *Cancer Med*. 2021;(December 2020):1-9. doi:10.1002/cam4.3691
47. Pasanen A, Loukovaara M, Bützow R. Clinicopathological significance of deficient DNA mismatch repair and MLH1 promoter methylation in endometrioid endometrial carcinoma. Mod Pathol. 2020;33(7):1443-1452. doi:10.1038/s41379-020-0501-8
48. Ronsini C, Anchora LP, Restaino S, et al. The role of semiquantitative evaluation of lympho-vascular space invasion in early stage cervical cancer patients. Gynecol Oncol. 2021 Aug;162(2):299-307.

LEGENDS FOR TABLES AND FIGURES

Figure 1. Forest plot of individual and pooled hazard ratios for death of any cause between endometrial carcinoma patients with and without lymphovascular space invasion; multivariable analyses including TCGA group as a variable were considered from included studies. The absence of lymphovascular space invasion was considered as a reference.

Figure 2. Forest plot of individual and pooled hazard ratios for death due to endometrial carcinoma between patients with and without lymphovascular space invasion; multivariable analyses including TCGA group as a variable were considered from included studies. The absence of lymphovascular space invasion was considered as a reference.

Figure 3. Forest plot of individual and pooled hazard ratios for recurrent or progressive disease between endometrial carcinoma patients with and without lymphovascular space invasion; multivariable analyses including TCGA group as a variable were considered from included studies. The absence of lymphovascular space invasion was considered as a reference. The study by Stelloo et al. [15] was not included.

Figure 4. Forest plot of individual and pooled hazard ratios for recurrent or progressive disease between endometrial carcinoma patients with and without lymphovascular space invasion; multivariable analyses including TCGA group as a variable were considered from included studies. The absence of lymphovascular space invasion was considered as a reference. Locoregional recurrence was considered for the study by Stelloo et al. [15].

Supplementary Figure 1. Flow diagram of studies identified in the systematic review (Prisma template [Preferred Reporting Item for Systematic Reviews and Meta-analyses]).

Supplementary Figure 2. a) Assessment of risk of bias. Summary of risk of bias for each study; Plus sign: low risk of bias; minus sign: high risk of bias; question mark: unclear risk of bias; **b)** Risk of bias graph about each risk of bias item presented as percentages across all included studies.

Supplementary Table 1. Characteristics of the studies included in the qualitative synthesis.

FIGO: International Federation of Gynaecology and Obstetrics

RCT: Randomized controlled trial

OS: overall survival

DFS: disease-free survival

DSS: disease-specific survival

***:** DFS was assessed as locoregional recurrence-free survival and distant recurrence-free survival

Supplementary Table 2. Characteristics of patients from studies included in the qualitative synthesis.

FIGO: International Federation of Gynecology and Obstetrics

LVSI: Lymphovascular space invasion

*: In the Stelloo et al. study, LVSI was assessed as absent/mild or substancial

Supplementary Table 3. Distribution of TCGA groups in the studies included in qualitative synthesis.

Supplementary Table 4. List of variables considered at multivariable analyses in the studies included in the qualitative synthesis.

FIGO: International Federation of Gynaecology and Obstetrics

L1CAM: L1 Cell Adhesion Molecule