5

Genomic Landscapes and Tumor Evolution in Metastatic Gynecological Cancers

Erling A. Hoivik[1,2]

[1]Department of Clinical Science, University of Bergen, Bergen, Norway; [2]Department of Obstetrics and Gynecology, Haukeland University Hospital, Bergen, Norway

Author for correspondence: Erling A. Hoivik, Department of Clinical Science, University of Bergen, Bergen, Norway. Email: Erling.Hoivik@uib.no

Cite this chapter as: Hoivik EA. Genomic Landscapes and Tumor Evolution in Metastatic Gynecological Cancers. In: Sergi CM, editor. *Metastasis*. Brisbane (AU): Exon Publications. Online first 2022 Mar 18.

Doi: https://doi.org/10.36255/exon-publications.metastasis.gynecological-cancers

Abstract: Gynecological cancers, such as endometrial cancer, ovarian cancer, and cervical cancers affect women's health worldwide. Metastatic and recurrent cancers are associated with poor survival, and effective treatment is lacking. A deeper understanding of the molecular mechanisms at the genomic level may help decipher the metastatic process, identify new targets, and develop personalized treatment strategies. Recent tumor evolutionary studies have provided phylogenetic interpretation of gynecological cancer metastasis. This has provided new models of metastatic development and pointed to potential targets for treatment. Moreover, cancer genome analysis of simultaneously detected tumor lesions, initially diagnosed as independent synchronous primary cancers of the endometrium and ovary, suggest that they rather represent a primary tumor-metastasis-pair relationship. This chapter provides an overview of the

In: Consolato M. Sergi, editor. *Metastasis*. Exon Publications, Brisbane, Australia. ISBN: 978-0-6453320-2-5. Doi: https://doi.org/10.36255/exon-publications.metastasis

characterization of cancer genomes, from primary tumors to metastatic lesions of the major gynecological cancers, and how such data are interpreted in an evolutionary context.

Keywords: metastatic gynecological cancers; synchronous cancers; tumor evolution; tumor heterogeneity; tumor phylogeny

INTRODUCTION

Metastasis involves the spread of cancer cells from a primary tumor to a distant site in the body (1). Cells in the metastatic lesion share genomic events such as mutations and copy-number alterations with their primary tumor, but the notable difference is that the metastatic cells have additional features that enable them to spread. Fortunately, metastasis is a relatively rare event as not all cancer cells acquire the capacity to metastasize as demonstrated by calculations on clinical samples (2–4). Unfortunately, reduced survival rates are inevitable in patients with metastatic cancers (5). Gynecologic cancers are cancers that originate from the female reproductive organs (Figure 1A). These include the main gynecological cancers of the cervix, ovary, endometrium (uterine wall), and the lesser frequent vaginal- and vulva cancers (6). The primary treatment for most of these cancers is hysterectomy or localized surgery. Despite surgery, a substantial part of gynecological cancers recurs or metastasize, with rates differing depending on primary cancer type (Figure 1B). Current treatment of metastatic gynecological disease is generally through inefficient systemic approaches, and new effective treatment is urgently needed, with the ultimate goal of improving survival rates (7).

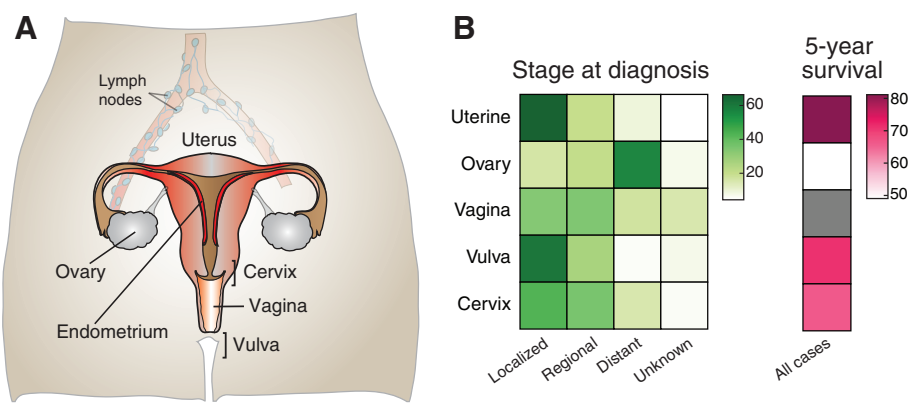

Figure 1. The female reproductive system, cancer stages and 5-year survival for gynecological cancers. A, Schematics of the female reproductive system. **B,** Data from the Surveillance, Epidemiology, and End Results (SEER) Program at the National Cancer Institute (NIH) comparing stages of disease at diagnosis and 5-year survival in gynecological cancers observed in the U.S. population. Relative numbers in percent for cancer stages (green) and 5-year survival (pink) for gynecological cancers. Grey box; missing data.

The era of advanced sequencing has provided opportunities to detail the cancer genome. This new technology is often referred to as *next generation sequencing* (NGS) or alternatively as *massive parallel sequencing*. These methods enable high throughput assays resulting in big datasets assessed by computational and bioinformatical analysis (8). Common types of such sequencing are targeted sequencing (TS), whole-exome sequencing (WES) and whole-genome sequencing (WGS), with covered regions ranging from a few hundred selected genes (sequencing panel) to all protein coding exons, or most of the genome (9). The application of advanced sequencing has proven extremely useful for cancer genomic studies. Until recently, most studies have focused on defining the genomic landscape of primary cancers, but there is an increased attention towards metastatic lesions. Genomic profiling of paired samples consisting of primary tumors and matching metastases allow for comparison of tumors for identifying unique metastatic traits and subclonal compositions (9). Such comparative genomic profiling also provides a picture of the metastatic spread with spatial and longitudinal resolution of the cancer evolution within patients. This chapter takes a closer look at the characterization of cancer genomes, from primary tumors to metastatic lesions of the major gynecological cancers, and how such data are interpreted in an evolutionary context.

CANCERS OF THE FEMALE REPRODUCTIVE SYSTEM

The female reproductive organs have several commonalities; they originate from the same embryonic structure, their development and normal function is influenced and regulated by female hormones (estrogens), and they act in concert to facilitate the function of female reproduction in reproductive age (10, 11). Endometrial cancer is the most common of the gynecological malignancies in high- and middle-income countries, ranked by the human developmental index (6). Although the prognosis for endometrial cancer is good, about 20% of patients have recurrent- or metastatic disease with poor prognosis (12). Ovarian, fallopian tube, and peritoneal cancer are collectively considered as cancers of the same system by the *International Federation of Gynecology and Obstetrics* (FIGO) committee for gynecologic oncology and typically managed in the same way (13, 14). Ovarian carcinoma is the most common type of ovarian cancer comprising about 95% of cases, with the high-grade serous carcinoma (HGSC) as the most common subtype. Extrauterine serous tumors arising from the ovary, fallopian tube, and the peritoneum have been collectively described as “Mullerian carcinomas” or “pelvic carcinomas”. The epithelial ovarian tumors may arise from endometriosis or cortical inclusions of Mullerian epithelium, forming slow-growing type I tumors (14). Contrasting these, fallopian tube carcinomas are of high grade and considered type II tumors. The cervix is a cylindrical structure at the lowermost part of the uterus. The ectocervix projects into the vagina, lined by squamous epithelium, while the endocervical canal is lined by columnar epithelium (15). The global cervical cancer rate is rapidly declining due to the Human Papilloma Virus (HPV)-vaccination program currently implemented in 130 countries.

THE GENOMIC LANDSCAPES OF GYNECOLOGICAL CANCERS

Comprehensive cancer programs led by large consortiums (The Cancer Genome Atlas, TCGA and the International Cancer Genome Consortium, ICGC), have tremendously helped in the characterization of many cancer types (16, 17). Genomic profiling of clinical samples across cancers has detailed the genomic underpinnings, suggested new cancer subtypes, and identified therapeutic targets that can be treated with available therapies or used to develop new drugs and therapies (17). Additionally, it has revealed high levels of intratumor and intertumor heterogeneity. The TCGA cancer programs have detailed primary cancers of the ovary (18), the cervix (19) and endometrium (20), as well as uterine carcinosarcomas (21).

Gynecological cancers have both shared and distinct features with other cancer genomes. Differences are displayed as unique mutational signatures and structural rearrangements for each cancer type reflected by the cancer "cell-of-origin" (22). Similarities include copy-number alterations shared among the serous types of ovarian- and endometrial cancers (5, 11, 20, 22). Early pan-cancer analysis found the tumor suppressor gene *TP53* to be the most frequently mutated gene with a mutation rate of 43% across 12 cancers. The mutation rate of *TP53* was highest in serous ovarian (95%) and serous endometrial carcinomas (89%) (23). The second most frequently mutated gene in this pan-cancer cohort was the phosphatidylinositol-4,5-bisphosphate 3-kinase signaling gene *PIK3CA* with an overall pan-cancer mutation rate of 17%; it was 52% in endometrial cancer and only 1% in serous ovarian cancer, highlighting differences in mutational profiles (23). *PIK3CA* mutations have been detected in 40% of vulvar cancer, potentially driving HPV-associated squamous cell carcinoma of the vulva (24). Other genomic features are also present in gynecological cancers. Berger and coworkers took a pan-gynecological cancer integrated approach to highlight genomic alterations that distinguished gynecological cancers from other cancer types. They discovered novel genes significantly mutated and enriched in gynecological cancers with prognostic potential and subtype specificity (11).

New molecular classification of endometrial cancer

Comprehensive profiling and data integration from large cancer profiling programs have revealed novel disease subtypes in major gynecological cancers. These data have provided new insights into the etiology and pathogenesis of the diseases, and importantly, provided results with the potential to change clinical practice (17). The use of cancer genome profiling to identify new relevant clinical subgroups of gynecological cancer patients is particularly profound in endometrial cancer. The TCGA study on endometrial cancer enabled a molecular classification, based on molecular and genomic profiling data, that has improved tumor classifications compared to the classical histopathological classification scheme into type I and type II endometrial cancer (20). The new molecular subtypes of endometrial cancers are POLE/ultramutated cancers, microsatellite instable (MSI)/hypermutated cancers, copy-number (CN) low/endometrioid cancers, and copy-number high/serous-like cancers (Figure 2). The phenotypes of these molecular classes are striking. The ultramutated DNA polymerase epsilon

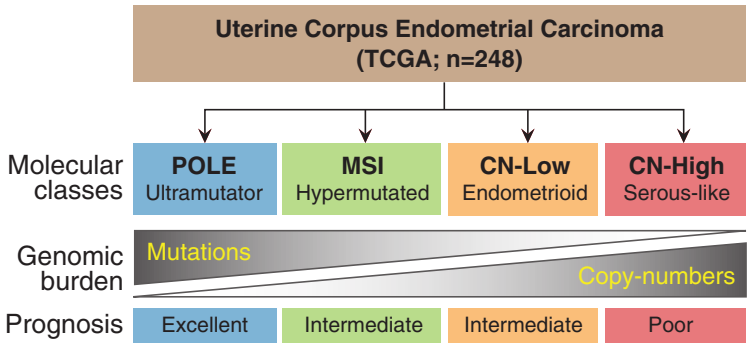

Figure 2. Molecular classes of endometrial cancer based on cancer genome characterization from TCGA. Extensive genomic characterization of cancer genomes from the TCGA endometrial cancer study (20) has suggested a new and improved stratification scheme for endometrial cancer. The four molecular subtypes (POLE/MSI/CN-low/CN-high) groups have distinct genomic and molecular characteristics, including differences in somatic mutations and copy-number burdens, and importantly, prognostic differences.

catalytic subunit (*POLE*) tumors have a very high mutational burden due to mutations in the exonuclease domains coupled to proofreading during replication. (hotspots at p.P286R and p.V411L). The hypermutated MSI tumors also have a high mutation burden, caused by defects in the DNA mismatch repair (MMR) system (MSH6, MSH2, PMS2 or MLH1) leading to uncorrected slippage mutations at mono- and dinucleotide repeats (microsatellites). The copy-number high tumors have a high level of chromosomal alterations that results in gain and loss of copies in focal or chromosomal regions of the DNA and are mostly of serous histologic type and *TP53* mutated. Lastly, the copy-number low class are mostly endometrioid tumors with low levels of amplifications/deletions and with intermediate mutation burden. Targeted therapies directed at these molecular subtypes are in development, including targets of DNA repair systems, the immune system, and signaling pathways (7, 25). Interestingly, the POLE group of patients, which portend an excellent prognosis, might not require additional treatment beyond primary surgery. For selected patients with recurrent, metastatic, or high-risk diseases in gynecological cancer, the US Food and Drug Administration (FDA) has approved Bevacizumab (VEGF inhibitor), Olaparib, Rucaparib, Niraparib (PARP inhibitors), and Pembrolizumab (anti PD-1), and these are currently being tested for clinical use (7).

METASTASES IN GYNECOLOGICAL CANCERS

Currently, treatment of gynecological cancer patients with primary tumors follows established guidelines but for metastatic disease this is generally more diffuse and limited to additional surgery, and systemic neoadjuvant or adjuvant therapies (7, 26). However, these approaches are only able to stagger the disease, that ultimately, will progress after a short period of time. Metastatic progression in

gynecological cancers, as with all cancers, is a complex process occurring in five sequential steps including: invasion, intravasation, circulation, extravasation, and colonization (27). Although the discrete steps in the metastatic cascade have been determined, the driving mechanisms and underlying genetic and molecular processes are lesser understood. Profiling strategies aimed at characterizing tissues of metastases and the corresponding primary gynecological tumor within the same patient is therefore required for both understanding the processes of metastasis and improving patient care.

Patterns of spread

The organotropism of the most frequent gynecological cancers is well known. Gynecological cancers are most likely to spread within the nearby organs, and tissues of the abdominal region and lymph nodes; however, spread to other distant organs such as brain and lung can also occur. Ovarian cancer tends to spread to the abdomen, colon, uterus, liver, and bone. Endometrial cancer also tends to spread locally to nearby organs, including the cervix, ovaries, vagina, and abdomen (5, 28–29). Cervical cancer spread occurs by either growing into nearby areas like vagina, bladder, rectum or tissues near the uterus and vagina. Local tumor growth and infiltration is not uniform but differs greatly among anatomical structures and compartments close to the uterine cervix (30). The spread of distant metastases in cervical cancer happens rarely and occurs in only 2% of cases (15). Recent studies have systematically catalogued genomic alterations as well as sites of metastatic spread of many cancers, including gynecological cancers (5, 30–33). An artificial intelligence (AI) application for determining organotrophic patterns of metastasis spread has been developed (28). Such machine learning algorithms are important as they may predict the metastasis landing-sites based on information from the primary tumor prior to the development of the actual metastasis. Being able to predict localization of metastatic disease may change the therapeutic landscape of both primary and metastatic cancers.

Lymph node metastasis

Lymph node assessment in gynecologic malignancies is important for prognostication and treatment decision. Factors like size and number of positive nodes are important in staging of gynecological cancers (34). Radiologic imaging is crucial for the preoperative detection of lymph node metastasis (35), and positive identification is a significant risk factor for prognosis. The most common pathway of dissemination in gynecologic malignancies are the superficial inguinal, pelvic, and para-aortic pathways, and is dependent on the location of the primary tumor and may follow several pathways depending on the cancer type and location (34). The role of systematic lymph node dissection has become a topic of discussion in gynecological cancers, as severe side effects or reduced quality of life can follow the procedure, with sentinel node (first lymph node) biopsy being a promising alternative (36, 37). Models for predicting lymph node metastasis based on radiologic imaging or molecular data are available (38, 39).

Genomic profiling of metastases

Recently, as an extension to genomic profiling of primary tumors, there has been studies focusing on profiling pan-cancer metastases, including ovarian, endometrial, and cervical cancers (5, 31–33). Nguyen and colleagues recently provided the largest study to date with targeted panel sequencing of 25,000 patients across 50 cancer types, highlighting considerable genomic differences between primary tumors and metastases, and specific metastatic sites (5). Although the cohort consisted of unpaired samples, this study demonstrated that metastases have a higher tumor burden and chromosomal instability compared to primary tumors. Specifically for uterine endometrioid cancer metastases, higher frequency of mutations in *TP53* (tumor suppressor protein p53) and *ESR1* (encoding estrogen receptor alpha), and amplifications of *ERBB2* (also known as *HER2*; human epidermal growth factor receptor 2), were observed but less mutations in the tumor suppressor phosphatase and tensin homolog gene (*PTEN*) compared to the primaries (5). It is disappointing that no real metastasis-specific driver gene has been identified, in any cancer type, despite considerable effort and apparent sufficient analytical detection power. Rather, it seems like the primary tumor and metastases share driver genes as exemplified by endometrial cancer (2, 29).

TUMOR EVOLUTION IN GYNECOLOGIC CANCERS

Tumors accumulate a range of somatic alterations, described as mutations, copy-number alterations, and structural variants across the entire genome. In general, we may consider these genomic alterations as informative events of the tumor's evolutionary trajectory informing us on how the tumors develop and how they spread (2, 40). Studying tumor evolution can help us to predict disease behavior, monitor disease progression and personalize treatment. However, the basis for the cancer evolution profiling is dependent on available patient biopsies, and quick turnaround of sequencing data if the patient is monitored, or in treatment.

Phylogenies to evaluate tumor evolution

To evaluate tumor evolution in metastases, samples need to be paired. In most cases, when matching primaries and metastases are sequenced from the same patient, unique genomic alterations can be detected in the metastatic lesion, but this is also true for the primary tumor. These unique, or private mutations (or copy-number alterations) are in addition to the mutations that are shared between the lesions from the same patients. A popular way to visualize comprehensive information included in tumor evolution analysis is phylogenetic trees (9, 41). These tumor-trees enable a description of the diversity of somatic mutations and copy-number alterations detailing the metastatic process. The complexity of phylogenetic analysis has led to new models for tumor evolution and metastasis. These models concern tumor evolution, modes of metastatic dissemination and seeding patterns (1, 42–43). The rationale behind these models is explained by stochastic progression, mutator phenotype and clonal evolution (9, 44). On this basis, studies have described the effect of evolutionary context of the tumor in

different models including clonal progression, neutral evolution, and selection. However, debates and disagreements on how to interpret and apply computational methods for inferring phylogenies from sequencing data also exists (42, 45). Some of these disagreements should be attributed to tumor heterogeneity that might be missed by bulk biopsy sequencing, interfering with the downstream analyses. Diving deeper into the tumor composition with multi-region sequencing and single-cell sequencing approaches in clinical specimens might unravel this issue in future studies.

Monophyly is the dominating evolutionary model in endometrial cancer

Only a few studies have investigated the evolution of gynecological cancers into metastases. Gibson *et al*. were the first to determine the phylogenetic relationships between endometrial tumor biopsies using whole exome sequencing (WES) data. (29). In their cohort of 26 cases, seven patients had multiple metastases available for sequencing as required for phylogenetic interpretations (visualized as phylogenetic trees). They observed that all the metastases were more closely related to each other, than to the primary tumor of same patient, suggesting that all metastases had a common ancestor. This suggested a model that metastases from endometrial cancer are the result of one branched subclone of the cancer. In relation to cancer evolution, this model was interpretated in a phylogenetic context as monophyly (Figure 3A). However, in the seventh case, one of the metastases was more closely related to the primary tumor than to the other metastases, a case of polyphyly, because of the presence of multiple origins (Figure 3B). The high proportion of monophyletic evolution in endometrial cancer has recently been confirmed though whole exome sequencing analysis of metastases with corresponding primary tumor in a similar cohort consisting of nine endometrial cancer patients, where the majority of cases fitted into to the evolutionary model of monophyly (46). These studies found no evidence of reseeding (29, 46).

Interestingly, a study on metastatic prostate cancer, with survival probabilities resembling endometrial cancer, was also determined to be mainly monophyletic, while that of the more aggressive pancreatic cancer was polyphyletic. Future studies may provide information if cancer types considered more aggressive are more likely to have multiple clones giving rise to metastases (47, 48). Indeed, in high-grade serous ovarian carcinoma (HGSOC), considered more aggressive than endometrial cancer, Masoodi and team found 2/6 of cancer phylogenies as polyphyly, although more cases are needed to clarify this (49). In terms of treatment, the presence of only one subclone (monophyly) might be beneficial, if targetable, as multiple subclones (polyphyly architecture) may be more prone to escape treatment and thereby develop drug-resistance.

SYNCHRONOUS ENDOMETRIAL AND OVARIAN LESIONS

The introduction of genomic sequencing in gynecological cancers has also provided a step forward in accessing the relationship of concurrent tumors involving multiple sites of the reproductive system, in particular tumors of the ovary and the

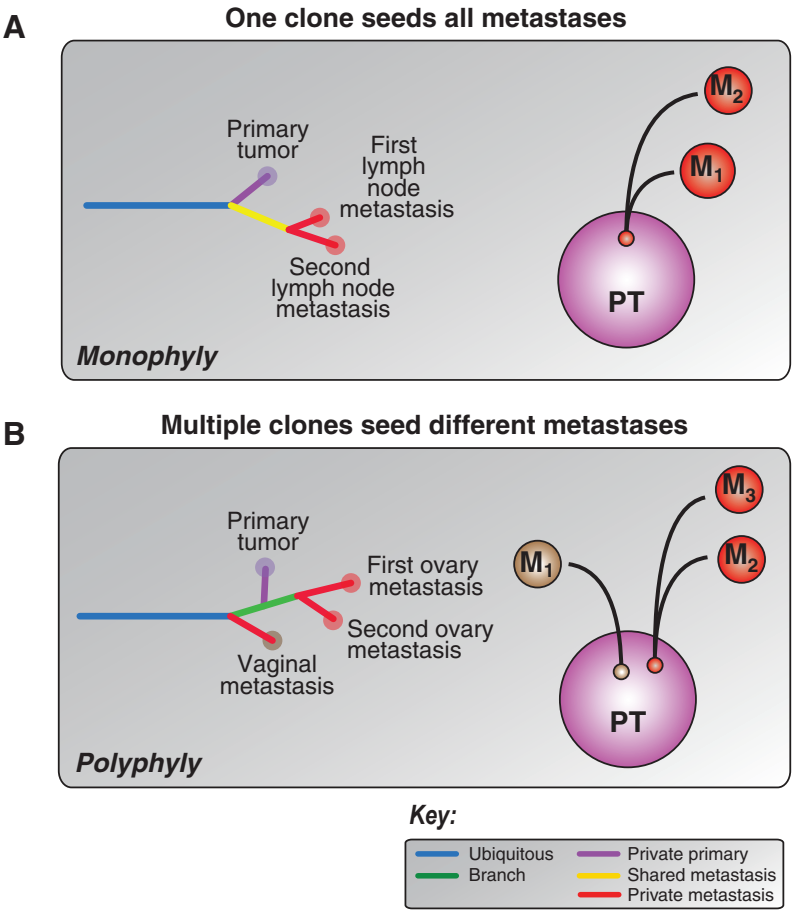

Figure 3. Phylogenetic models of endometrial cancer. Two alternative scenarios describe the origin of multiple metastases in endometrial cancer. **A,** The monophyly model suggest that the origin of metastases (Ms) are from the same ancestral origin, as a subclone or a single cell of the primary tumor (PT). **B,** Alternatively, multiple clones seed the different metastases in the polyphyly model. Current data has demonstrated that the monophyletic model is the dominating and most likely route of metastatic spread (29,46). Tumor evolutionary phylogenetic trees constructed from somatic mutations are displayed to the left, with accompanying models to the right.

endometrium, due to their frequent occurrence. While these cancers often have been evaluated as independent primaries by pathologic examination and diagnosed as synchronous cancers, recent sequencing analysis has demonstrated a clonal relationship suggesting that these tumors rather represent a primary-tumor-metastasis relationship (50). Multiple studies have addressed the relationship of synchronous cancers with endometrial and ovarian lesions through different sequencing approaches, including panel-sequencing and whole exome sequencing approaches (29, 51–55). It is noted that some cases diagnosed with synchronous cancers do not have shared genomic alterations, suggesting either a true

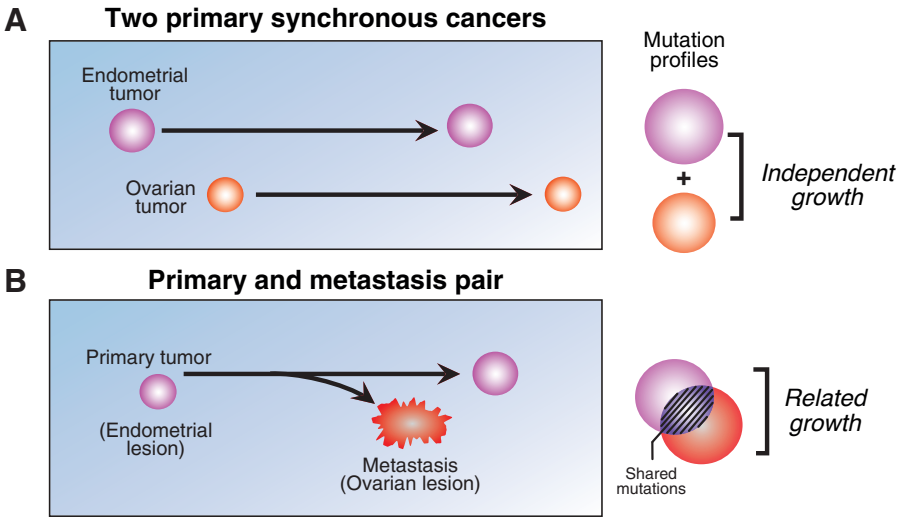

Figure 4. Independent primary synchronous cancers versus metastatic growth. Possible interpretations of simultaneous endometrial and ovarian lesions **A,** Independent growth of two primary tumors of the endometrium and the ovary, suggestive of synchronous cancers (no shared genomic features). **B,** Related growth by endometrial and ovarian tumors with shared mutational profiles, demonstrating relatedness and suggestive of a primary tumor-metastasis relationship. Multiple studies of apparently synchronous cancers are supportive of the latter model (29, 51–55).

synchronous independency, or alternatively, a sequencing approach that are not sufficient to detect rarer shared alterations, such as panel-sequencing (Figure 4). For some of the synchronous cases, for example, Lynch syndrome (hereditary nonpolyposis colorectal cancer), MMR germline deficiency seems to be the underlying cause of multiple lesions, as this disease increases the risk of several cancers including colorectal, endometrial, ovarian, and other cancers (56). The scenario of synchronous versus a primary-metastasis relationship creates a clinical puzzle, as the overall survival for the primary as well as synchronous cancers are good, while that of metastatic cancers generally are not. These conflicting clinical outcomes are important, and distinguishing the two diagnosis is essential to avoid both overtreatment and undertreatment. Advanced sequencing may improve diagnostic accuracy of synchronous cancers when comparing mutational profiles (29, 51–52).

CONCLUSION

Understanding metastatic disease in order to develop targeted therapies and personalized treatment remains a challenging task in gynecological cancers. The complex nature of the metastatic processes, lack of available metastatic biopsies, and the large tasks associated with tackling tumor heterogeneity and tumor

diversity represent systematic hurdles to understanding sequencing data. Although no specific metastatic driver has been detected, good progress has been made in understanding metastatic disease and cancer evolution in gynecological cancers, supporting new models of metastasis. Retrospective spatial and nonspatial data from patients with gynecological cancers, both from primary tumors and metastatic lesions, are becoming increasingly in focus, as clinicians and basic scientists are becoming aware for the need for such data to fully understand metastatic disease. Biobanking of patient material is required along with the application of the latest sequencing developments to accelerate the development of specific therapeutics for metastasis and pinpoint diagnosis. Future prioritization to improve female cancer therapeutics should include not only the most frequent gynecological cancers (endometrial, ovarian, and cervical cancers), but also vulvar- and vaginal cancers.

Acknowledgements: Data underlying Figure 1B has been accessed through https://seer.cancer.gov.

Conflict of Interest: The author declares no potential conflicts of interest with respect to research, authorship and/or publication of this manuscript.

Copyright and Permission Statement: The author confirm that the materials included in this chapter do not violate copyright laws. Where relevant, appropriate permissions have been obtained from the original copyright holder(s), and all original sources have been appropriately acknowledged or referenced.

REFERENCES

1. Birkbak NJ, McGranahan N. Cancer Genome Evolutionary Trajectories in Metastasis. Cancer Cell. 2020 01 13;37(1):8–19. https://doi.org/10.1016/j.ccell.2019.12.004
2. Patel SA, Rodrigues P, Wesolowski L, Vanharanta S. Genomic control of metastasis. Br J Cancer. 2021 01;124(1):3–12. https://doi.org/10.1038/s41416-020-01127-6
3. Michaelson JS, Cheongsiatmoy JA, Dewey F, Silverstein MJ, Sgroi D, Smith B, et al. Spread of human cancer cells occurs with probabilities indicative of a nongenetic mechanism. Br J Cancer. 2005 28;93(11):1244–9. https://doi.org/10.1038/sj.bjc.6602848
4. Szczurek E, Krüger T, Klink B, Beerenwinkel N. A mathematical model of the metastatic bottleneck predicts patient outcome and response to cancer treatment. PLoS Comput Biol. 2020 10;16(10):e1008056. https://doi.org/10.1371/journal.pcbi.1008056
5. Nguyen B, Fong C, Luthra A, Smith SA, DiNatale RG, Nandakumar S, et al. Genomic characterization of metastatic patterns from prospective clinical sequencing of 25,000 patients. Cell. 2022 02 3;185(3):563–575.e11.
6. 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 05;71(3):209–49. https://doi.org/10.3322/caac.21660
7. Wang Q, Peng H, Qi X, Wu M, Zhao X. Targeted therapies in gynecological cancers: a comprehensive review of clinical evidence. Signal Transduct Target Ther. 2020 07 29;5(1):137. https://doi.org/10.1038/s41392-020-0199-6
8. Cortés-Ciriano I, Gulhan DC, Lee JJ, Melloni GEM, Park PJ. Computational analysis of cancer genome sequencing data. Nat Rev Genet. [published online: December 08, 2021]. 10.1038/s41576-021-00431-y

9. Kader T, Zethoven M, Gorringe KL. Evaluating statistical approaches to define clonal origin of tumours using bulk DNA sequencing: context is everything. Genome Biol. 2022 02 2;23(1):43. https://doi.org/10.1186/s13059-022-02600-6
10. Mullen RD, Behringer RR. Molecular genetics of Müllerian duct formation, regression and differentiation. Sex Dev. 2014;8(5):281–96. https://doi.org/10.1159/000364935
11. Berger AC, Korkut A, Kanchi RS, Hegde AM, Lenoir W, Liu W, et al. A Comprehensive Pan-Cancer Molecular Study of Gynecologic and Breast Cancers. Cancer Cell. 2018 04 9;33(4):690–705.e9.
12. Koskas M, Amant F, Mirza MR, Creutzberg CL. Cancer of the corpus uteri: 2021 update. Int J Gynaecol Obstet. 2021;155 Suppl 1:45–60. https://doi.org/10.1002/ijgo.13866
13. Prat J. Staging Classification for Cancer of the Ovary, Fallopian Tube, and Peritoneum: Abridged Republication of Guidelines From the International Federation of Gynecology and Obstetrics (FIGO). Obstet Gynecol. 2015;126(1):171–4. https://doi.org/10.1097/AOG.0000000000000917
14. Berek JS, Renz M, Kehoe S, Kumar L, Friedlander M. Cancer of the ovary, fallopian tube, and peritoneum: 2021 update. Int J Gynaecol Obstet. 2021;155 Suppl 1:61–85. https://doi.org/10.1002/ijgo.13878
15. Bhatla N, Aoki D, Sharma DN, Sankaranarayanan R. Cancer of the cervix uteri: 2021 update. Int J Gynaecol Obstet. 2021;155 Suppl 1:28–44. https://doi.org/10.1002/ijgo.13865
16. Campbell PJ, Getz G, Korbel JO, Stuart JM, Jennings JL, Stein LD, et al. Pan-cancer analysis of whole genomes. Nature. 2020 02;578(7793):82–93. https://doi.org/10.1038/s41586-020-1969-6
17. Hutter C, Zenklusen JC. The Cancer Genome Atlas: Creating Lasting Value beyond Its Data. Cell. 2018 04 5;173(2):283–5. https://doi.org/10.1016/j.cell.2018.03.042
18. Bell D, Berchuck A, Birrer M, Chien J, Cramer D, Dao F, et al. Integrated genomic analyses of ovarian carcinoma. Nature. 2011 29;474(7353):609–15. https://doi.org/10.1038/nature10166
19. Burk RD, Chen Z, Saller C, Tarvin K, Carvalho AL, Scapulatempo-Neto C, et al. Integrated genomic and molecular characterization of cervical cancer. Nature. 2017 03 16;543(7645):378–84. https://doi.org/10.1038/nature21386
20. Kandoth C, Schultz N, Cherniack AD, Akbani R, Liu Y, Shen H, et al. Integrated genomic characterization of endometrial carcinoma. Nature. 2013 2;497(7447):67–73. https://doi.org/10.1038/nature12113
21. Cherniack AD, Shen H, Walter V, Stewart C, Murray BA, Bowlby R, et al. Integrated Molecular Characterization of Uterine Carcinosarcoma. Cancer Cell. 2017 03 13;31(3):411–23.
22. Hoadley KA, Yau C, Hinoue T, Wolf DM, Lazar AJ, Drill E, et al. Cell-of-Origin Patterns Dominate the Molecular Classification of 10,000 Tumors from 33 Types of Cancer. Cell. 2018 04 5;173(2):291–304.e6.
23. Kandoth C, McLellan MD, Vandin F, Ye K, Niu B, Lu C, et al. Mutational landscape and significance across 12 major cancer types. Nature. 2013 17;502(7471):333–9. https://doi.org/10.1038/nature12634
24. Han MR, Shin S, Park HC, Kim MS, Lee SH, Jung SH, et al. Mutational signatures and chromosome alteration profiles of squamous cell carcinomas of the vulva. Exp Mol Med. 2018 02 9;50(2):e442. https://doi.org/10.1038/emm.2017.265
25. Yen TT, Wang TL, Fader AN, Shih IM, Gaillard S. Molecular Classification and Emerging Targeted Therapy in Endometrial Cancer. Int J Gynecol Pathol. 2020;39(1):26–35. https://doi.org/10.1097/PGP.0000000000000585
26. Bhatla N, Tomar S, Meena J, Sharma DN, Kumar L. Adjuvant treatment in cervical, vaginal and vulvar cancer. Best Pract Res Clin Obstet Gynaecol. 2022;78:36–51. https://doi.org/10.1016/j.bpobgyn.2021.07.005
27. Hapach LA, Mosier JA, Wang W, Reinhart-King CA. Engineered models to parse apart the metastatic cascade. NPJ Precis Oncol. 2019;3:20. https://doi.org/10.1038/s41698-019-0092-3
28. Jiang B, Mu Q, Qiu F, Li X, Xu W, Yu J, et al. Machine learning of genomic features in organotropic metastases stratifies progression risk of primary tumors. Nat Commun. 2021 11 18;12(1):6692. https://doi.org/10.1038/s41467-021-27017-w
29. Gibson WJ, Hoivik EA, Halle MK, Taylor-Weiner A, Cherniack AD, Berg A, et al. The genomic landscape and evolution of endometrial carcinoma progression and abdominopelvic metastasis. Nat Genet. 2016 08;48(8):848–55. https://doi.org/10.1038/ng.3602

30. Kubitschke H, Wolf B, Morawetz E, Horn LC, Aktas B, Behn U, et al. Roadmap to Local Tumour Growth: Insights from Cervical Cancer. Sci Rep. 2019 09 4;9(1):12768. https://doi.org/10.1038/s41598-019-49182-1
31. Zehir A, Benayed R, Shah RH, Syed A, Middha S, Kim HR, et al. Mutational landscape of metastatic cancer revealed from prospective clinical sequencing of 10,000 patients. Nat Med. 2017;23(6):703–13. https://doi.org/10.1038/nm.4333
32. Priestley P, Baber J, Lolkema MP, Steeghs N, de Bruijn E, Shale C, et al. Pan-cancer whole-genome analyses of metastatic solid tumours. Nature. 2019 11;575(7781):210–6. https://doi.org/10.1038/s41586-019-1689-y
33. Geistlinger L, Oh S, Ramos M, Schiffer L, LaRue RS, Henzler CM, et al. Multiomic Analysis of Subtype Evolution and Heterogeneity in High-Grade Serous Ovarian Carcinoma. Cancer Res. 2020 10 15;80(20):4335–45. https://doi.org/10.1158/0008-5472.CAN-20-0521
34. Paño B, Sebastià C, Ripoll E, Paredes P, Salvador R, Buñesch L, et al. Pathways of lymphatic spread in gynecologic malignancies. Radiographics. 2015;35(3):916–45. https://doi.org/10.1148/rg.2015140086
35. Ramanathan S, Tirumani SH, Ojili V. Nodal metastasis in gynecologic malignancies: Update on imaging and management. Clin Imaging. 2020 Feb;59(2):157–66. https://doi.org/10.1016/j.clinimag.2019.11.006
36. Brugger S, Hamann M, Mosner M, Beer M, Braun M, Pölcher M. Endometrial cancer-how many patients could benefit from sentinel lymph node dissection. World J Surg Oncol. 2018 17;16(1):95. https://doi.org/10.1186/s12957-018-1392-8
37. Zhai L, Zhang X, Cui M, Wang J. Sentinel Lymph Node Mapping in Endometrial Cancer: A Comprehensive Review. Front Oncol. 2021;11:701758. https://doi.org/10.3389/fonc.2021.701758
38. Fasmer KE, Gulati A, Dybvik JA, Ytre-Hauge S, Salvesen Ø, Trovik J, et al. Preoperative 18F-FDG PET/CT tumor markers outperform MRI-based markers for the prediction of lymph node metastases in primary endometrial cancer. Eur Radiol. 2020;30(5):2443–53. https://doi.org/10.1007/s00330-019-06622-w
39. Berg HF, Ju Z, Myrvold M, Fasmer KE, Halle MK, Hoivik EA, et al. Development of prediction models for lymph node metastasis in endometrioid endometrial carcinoma. Br J Cancer. 2020 03;122(7):1014–22. https://doi.org/10.1038/s41416-020-0745-6
40. Nowell PC. The clonal evolution of tumor cell populations. Science. 1976 Oct 1;194(4260):23–8. https://doi.org/10.1126/science.959840
41. Naxerova K, Jain RK. Using tumour phylogenetics to identify the roots of metastasis in humans. Nat Rev Clin Oncol. 2015;12(5):258–72. https://doi.org/10.1038/nrclinonc.2014.238
42. Davis A, Gao R, Navin N. Tumor evolution: Linear, branching, neutral or punctuated. Biochim Biophys Acta Rev Cancer. 2017;1867(2):151–61. https://doi.org/10.1016/j.bbcan.2017.01.003
43. Gui P, Bivona TG. Evolution of metastasis: new tools and insights. Trends Cancer. 2022;8(2):98–109. https://doi.org/10.1016/j.trecan.2021.11.002
44. Salichos L, Meyerson W, Warrell J, Gerstein M. Estimating growth patterns and driver effects in tumor evolution from individual samples. Nat Commun. 2020 02 5;11(1):732. https://doi.org/10.1038/s41467-020-14407-9
45. Miura S, Vu T, Deng J, Buturla T, Oladeinde O, Choi J, et al. Power and pitfalls of computational methods for inferring clone phylogenies and mutation orders from bulk sequencing data. Sci Rep. 2020 02 26;10(1):3498. https://doi.org/10.1038/s41598-020-59006-2
46. Mota A, Oltra SS, Selenica P, Moiola CP, Casas-Arozamena C, López-Gil C, et al. Intratumor genetic heterogeneity and clonal evolution to decode endometrial cancer progression. Oncogene. [published online: February 10, 2022]. 10.1038/s41388-022-02221-0
47. Gundem G, Van Loo P, Kremeyer B, Alexandrov LB, Tubio JMC, Papaemmanuil E, et al. The evolutionary history of lethal metastatic prostate cancer. Nature. 2015 16;520(7547):353–7. https://doi.org/10.1038/nature14347
48. Campbell PJ, Yachida S, Mudie LJ, Stephens PJ, Pleasance ED, Stebbings LA, et al. The patterns and dynamics of genomic instability in metastatic pancreatic cancer. Nature. 2010 28;467(7319):1109–13. https://doi.org/10.1038/nature09460

49. Masoodi T, Siraj S, Siraj AK, Azam S, Qadri Z, Parvathareddy SK, et al. Genetic heterogeneity and evolutionary history of high-grade ovarian carcinoma and matched distant metastases. Br J Cancer. 2020 04;122(8):1219–30. https://doi.org/10.1038/s41416-020-0763-4
50. Dizon DS, Birrer MJ. Making a Difference: Distinguishing Two Primaries From Metastasis in Synchronous Tumors of the Ovary and Uterus. J Natl Cancer Inst. 2016;108(6):djv442. https://doi.org/10.1093/jnci/djv442
51. Schultheis AM, Ng CK, De Filippo MR, Piscuoglio S, Macedo GS, Gatius S, et al. Massively Parallel Sequencing-Based Clonality Analysis of Synchronous Endometrioid Endometrial and Ovarian Carcinomas. J Natl Cancer Inst. 2016;108(6):djv427. https://doi.org/10.1093/jnci/djv427
52. Anglesio MS, Wang YK, Maassen M, Horlings HM, Bashashati A, Senz J, et al. Synchronous Endometrial and Ovarian Carcinomas: Evidence of Clonality. J Natl Cancer Inst. 2016;108(6):djv428. https://doi.org/10.1093/jnci/djv428
53. Chao A, Wu RC, Jung SM, Lee YS, Chen SJ, Lu YL, et al. Implication of genomic characterization in synchronous endometrial and ovarian cancers of endometrioid histology. Gynecol Oncol. 2016 10;143(1):60–7. https://doi.org/10.26226/morressier.5770e29bd462b80290b4bb94
54. Gilks CB, Kommoss F. Synchronous tumours of the female reproductive tract. Pathology. 2018;50(2):214–21. https://doi.org/10.1016/j.pathol.2017.10.007
55. Moukarzel LA, Da Cruz Paula A, Ferrando L, Hoang T, Sebastiao APM, Pareja F, et al. Clonal relationship and directionality of progression of synchronous endometrial and ovarian carcinomas in patients with DNA mismatch repair-deficiency associated syndromes. Mod Pathol. 2021 05;34(5):994–1007. https://doi.org/10.1038/s41379-020-00721-6
56. Vogt A, Schmid S, Heinimann K, Frick H, Herrmann C, Cerny T, et al. Multiple primary tumours: challenges and approaches, a review. ESMO Open. 2017;2(2):e000172. https://doi.org/10.1136/esmoopen-2017-000172