The role of circulating tumor cells in vulvar carcinoma

Leonie Elisabeth Kleinholz[1]
Harriet Wikman-Kocher[1], Simon Joosse[1], Maggie Banys-Paluchowski[2], Sandra Lenz[1], Jolanthe Kropidlowski[1], Katharina Prieske[3], Anna Jaeger[3], Leticia Oliveira-Ferrer[3], Gerhard Gebauer[4], Barbara Schmalfeldt[3], Linn Woelber[3], Klaus Pantel[1] and Katharina Harms-Effenberger[1]
[1] Institute of Tumor Biology, University Hospital Hamburg-Eppendorf
[2] Department of Gynecology, University Hospital Schleswig-Holstein, Lübeck
[3] Department of Gynecology, University Hospital Hamburg-Eppendorf
[4] Department of Gynecology and Obstetrics, Asklepios Hospital Hamburg-Barmbek

Background & objectives

Vulvar carcinoma is the fifth most common gynecologic cancer entity in Germany with an increasing incidence over the past years, especially in younger women. Nodal status is the strongest prognostic factor for disease outcome. Local relapses are common with moderate treatment success. Circulating tumor cells (CTC) have not been studied here as an additional staging, monitoring or treatment decision tool.

For the very first time we investigated CTCs in vulvar carcinoma including single cell profiling and PD-L1 expression regarding disease characteristics and outcome.

Methods

101 vulvar carcinoma patients were enrolled in this prospective multicenter study and CTCs assessed applying Parsortix® or density gradient centrifugation. Evaluation was based on multiplex-staining (anti-pan-keratin/anti-PD-L1/anti-CD45/DAPI). Cells with strong keratin staining and "typical CTC morphology" were classified as category 1 or 2; other cells with unusual morphology and/or keratin-low staining as category 3 or 4. For verification, 33 different CTC-candidates (cat. 1-4) were picked for single-cell NGS and classified as CTCs based on their CNV profile.

Results

CTC analyses were performed for 75 patients at first diagnosis (51 before, 24 after surgery), for 24 patients at time of recurrent (16 before, 8 after surgery), two were lost from follow-up. 12 patients were excluded (secondary cancers). The mean age at first diagnosis was 67. CTCs were detected in 76.4% (68/89; range 1-933; median 3); 62.9% harbored $^{3}2$ CTCs, 43.8% $^{3}5$ CTCs. PD-L1^{+-}CTCs were present in 21.3% (19/89).

By NGS, 27/31 of category 1&2-cells and 2/2 of category 3&4-cells were proved “true” CTCs.

In the first-diagnosis-group, pre-surgery CTCs showed significant correlations with nodal status (p=0.035), application of adjuvant therapy (p=0.004) and FIGO stage (I/II vs III/IV; p=0.021) for ³5 CTCs (cat.3&4).

Progression-free (PFS) and overall survival (OS) were significantly decreased in the recurrence-group if cat.3&4-CTCs were present (p=0.046 and p=0.023, log-rank).

In this group PD-L1^{+}-CTCs showed a significant correlation with application of adjuvant therapy (p=0.021).

Conclusion

Presence of CTCs in vulvar carcinoma patients has been confirmed. Women at recurrent disease harboring unusual CTCs showed worse PFS and OS compared to women without. Patients harboring CTCs, especially PD-L1^{+}-CTCs, might benefit from adjuvant therapy or new immunotherapy approaches, respectively.