学 位 論 文 の 要 旨

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学 位 論 文 名　Relationship between Microsatellite Instability, Immune Cells Infiltration, and Expression of Immune Checkpoint Molecules in Ovarian Carcinoma: Immunotherapeutic Strategies for the Future

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論 文 内 容 の 要 旨

INTRODUCTION

Ovarian cancer has the poorest prognosis of all gynecological cancers, and therefore new therapies are needed to improve the prognosis. Currently, new drugs such as immune checkpoint inhibitors and poly (ADP-ribose) polymerase inhibitors (PARP inhibitors) are in the spotlight. Anti-programmed cell death-1 (PD-1)/ programmed cell death-ligand 1 (PD-L1) antibody, one of immune checkpoint inhibitors, have been gradually applied in the treatment of various carcinomas, but are not effective in all patients.

More tumor neoantigens are produced from tumors with many genetic mutations such as microsatellite instability (MSI)-High. Then more lymphocytes infiltrate around these tumors. Therefore, it is considered that anti PD-1/PD-L1 antibody is particularly effective for tumors having many genetic mutations. Among epithelial ovarian cancers, endometrioid carcinoma and clear cell carcinoma are reported to have many MSI-High cases. We investigated the correlation between MSI status, immune checkpoint molecules (PD-1 and PD-L1) and tumor-infiltrating lymphocytes (CD8) by immunostaining to investigate the efficiency of anti PD-1/PD-L1 antibody for each histologic type of epithelial ovarian cancer.

MATERIALS AND METHODS

We evaluated the expression of mismatch repair (MMR) proteins (MLH1, MSH2, MSH6, and PMS2), immune checkpoint molecules (PD-L1 and PD-1), and CD8 infiltrations into the tumor by immunohistochemistry. Formalin-fixed, paraffin-embedded tissue samples of 136 ovarian carcinomas (76, 13, 23, and 24 patients with high-grade serous, mucinous, endometrioid, and clear cell carcinoma, respectively) were used in this study. Cases were evaluated as MSI when at least one of the four MMR proteins was negative. Other cases were evaluated as microsatellite stable (MSS). Expression of CD8 lymphocytes infiltrating the tumor was evaluated according to four levels (0, undetectable; 1+, low density; 2+, moderate density; and 3+, high density). Cases which were stained 2+ or 3+ were evaluated as positive. Cases were evaluated as positive for PD-L1 when ≥5% of the tumor cells were stained (membranous and cytoplasmic staining). Cases were evaluated as positive for PD-1 when ≥5% of the tumor-infiltrating lymphocytes were stained.

The cases which were evaluated as MSI by immunohistochemistry were further validated by MSI analysis. The correlations between MSI and the expression of PD-L1, PD-1, CD8 and clinicopathological characteristics were investigated.

This study protocol was approved by the Ethics Committee of Shimane University and written informed consent was obtained from all subjects.

RESULTS AND DISCUSSION

Six cases (4.4%) (2, 1, 2, and 1 in serous, mucinous, endometrioid, and clear cell carcinoma, respectively) were determined to be MSI status. There was no significant difference in PFS or OS between the MSI group and the MSS group. Similarly, there was no significant difference in PFS or OS between the CD8(+) and CD8(-) cases, PD-L1(+) and PD-L1(-) cases, and PD-1(+) and PD-1(-) cases. No significant differences were found between MSI status and expression of CD8 ($p = 0.126$), PD-L1 ($p = 0.432$), and PD-1 ($p = 0.653$). Moreover, there was no correlation between MSI and the expression of CD8, PD-L1, and PD-1 in any histological type. These results may be attributed to the low proportion of MSI in epithelial ovarian cancer. We thought that epithelial ovarian cancer with very few cases could be treated effectively by immune checkpoint inhibitor monotherapy.

Platinum drugs and PARP inhibitors are effective in ovarian cancer with homologous recombination deficiency (HRD). However, ovarian cancer with non-HRD is thought to be less effective to platinum drugs and PARP inhibitors. When ovarian cancer with non-HRD is treated with platinum drugs or PARP inhibitors, both the pathway of activating immune functions (Interferon (IFN) response and neoantigen production due to repair errors) and suppressing immune functions (PD-L1 upregulation via Ataxia Telangiectasia Mutated/Ataxia Telangiectasia and Rad3 Related Protein/Checkpoint kinase1 (ATM/ATR/Chk1) pathway activation) acts during the normal homologous recombination repair mechanism. Therefore, using the combined therapy of immune checkpoint inhibitors with PARP inhibitors will be more effective than each agent

alone in ovarian cancer non-HRD patients. Nowadays, chemotherapy and PARP inhibitors have been reported to upregulate PD-L1 expression. From these results, combined treatment of immune checkpoint inhibitors with chemotherapy or PARP inhibitors is expected to be useful.

Currently, many clinical trials have been performed to evaluate the efficiency of multi-drug therapies together with immune checkpoint inhibitors and various anti-cancer drugs and molecular targeted therapies in ovarian cancer patients. We are examining the effects of multi-drug therapies along with immune checkpoint inhibitors in ovarian cancer using immunocompetent mice. We believe that new treatments for platinum-resistant ovarian cancer might be discovered by administering platinum-resistant cell lines to mice and verifying the potency of combination therapy.

CONCLUSION

Ovarian cancer has only a few cases of MSI. Therefore, there are few ovarian cancer patients who can be expected to benefit from immune checkpoint inhibitor monotherapy. For better prognosis of ovarian cancer, multi-drug therapy together with immune checkpoint inhibitors is necessary. Currently, several clinical trials are being conducted on the efficacy of immune checkpoint inhibitors in ovarian cancer. In addition, clinical trials on combination therapy including anticancer drugs and immune checkpoint inhibitors or molecular targeted therapeutic drugs and immune checkpoint inhibitors are being conducted. Results from these clinical trials may open the door to new therapies for ovarian cancer.