Melina-Theoni Gyparaki
Athanasios G. Papavassiliou
Department of Biological Chemistry, Medical School, National and Kapodistrian University of Athens, Athens, Greece

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Epigenetic Pathways Offer Targets for Ovarian Cancer Treatment

Ovarian cancer is the most severe disease of the female reproductive tract, because it is associated with the greatest mortality rates. Ovarian cancer has 3 categories: epithelial, stromal, and germ cell tumors. Of these 3 categories, epithelial (in particular, serous epithelial ovarian cancer) is the most common type.[1] Treatment of the cancer is dependent on its stage. When ovarian cancer is organ-confined, it can be surgically removed.[2] However, in cases in which the cancer has progressed beyond the ovaries, a combination of surgery and chemotherapy is required.[3] An important prognostic factor that could predict for survival is whether the tumor size has decreased after surgery and to what extent.[4] In chemotherapy, the 2 most important drugs used in the treatment of ovarian cancer have been carboplatin and paclitaxel.[5] At present, one of the greatest challenges associated with chemotherapy is that cancers have started developing resistance against such drugs. Consequently, their effectiveness in eradicating tumors has been significantly reduced.[6] Thus, the development of new treatments that would remain unaffected by these mechanisms of resistance is needed.

Epigenetics is a field that focuses on the study of heritable changes that do not result from changes in the DNA sequence. Epigenetic changes include DNA methylation, which is the addition of a methyl group to a cytosine (C) residue with the aid of an enzyme, known as DNA methyltransferase, and histone modifications. These can be divided into histone acetylation, deacetylation, and methylation events performed by histone acetyltransferases, histone deacetylases (HDACs), and histone methyltransferases, respectively.[7] Recently, research has focused on epigenetic changes and their relation to cancer. Aberrant methylation of CpG islands, which are found in close proximity to gene transcription initiation sites and are normally methylated, has been linked to tumor initiation and progression.[8] Because of their involvement in tumorigenesis, the biology of these changes is now being investigated to fully understand its potential in the treatment of ovarian cancer. The present mini-review describes the common molecular mechanisms of ovarian cancer and discusses the prospective of epigenetic remedies in the treatment of ovarian cancer.

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Address for correspondence: Athanasios G. Papavassiliou, MD, PhD, Department of Biological Chemistry, Medical School, National and Kapodistrian University of Athens, Athens 11527, Greece
E-mail contact: papavas@med.uoa.gr

Signaling Pathways in Ovarian Cancer

Epithelial ovarian cancer has been classified according to the differing pathogenesis. More specifically, epithelia ovarian cancer has been classified as type 1 and type 2 ovarian tumors. Type 1 tumors, including endometrioid, mucinous, and low-grade serous tumors develop at a slow pace, and type 2 tumors rapidly progress to high-grade serous carcinoma.[9,10] Expression profiling studies have shown that these 2 types of cancer cluster separately and can therefore be characterized by the different molecular pathways. *P53* mutations are important events in the development of high-grade carcinoma, and mutations in *KRAS*, *BRAF*, phosphatase and tensin homolog, and β-catenin are associated with low-grade tumors[11] (Figure 1). High-grade tumors are associated with genomic instability, an aggressive clinical appearance, and mortality from ovarian cancer.[12]

DNA Methylation in Ovarian Cancer

DNA methylation is a process during which a methyl group is added to a cytosine nucleotide with the aid of a DNA methyltransferase enzyme. DNA methylation events occur normally in cells and appear to be of a repressive nature.[13] Thériault et al[14] suggested that hypomethylation of the promoter region of *KF14*, a gene encoding a mitotic kinase known as an important oncogene in cancer, could explain its overexpression in ovarian cancer. Moreover, they described a variety of approaches for the upregulation of the gene, and these could be important therapeutic targets.[14] CpG island promoter hypermethylation in *BRCA1* and *BRCA2* genes has been found to be associated with increased sensitivity to some chemotherapeutic agents.[15] These genes are frequently mutated in breast, ovarian, and other cancers, and tumors that contain these mutations are more sensitive to chemotherapeutic agents. The sensitivity resulting from epigenetic regulation of the genes has been comparable to that resulting from germline mutations in the genes.[15] Tumor suppressor candidate 3 (*TUSC3*) is another gene that is epigenetically silenced by aberrant methylation of its promoter in ovarian cancer, and it has been suggested that it could act as a prognostic factor for this cancer.[16] Epigenetic regulation of all these genes should be further investigated, because this

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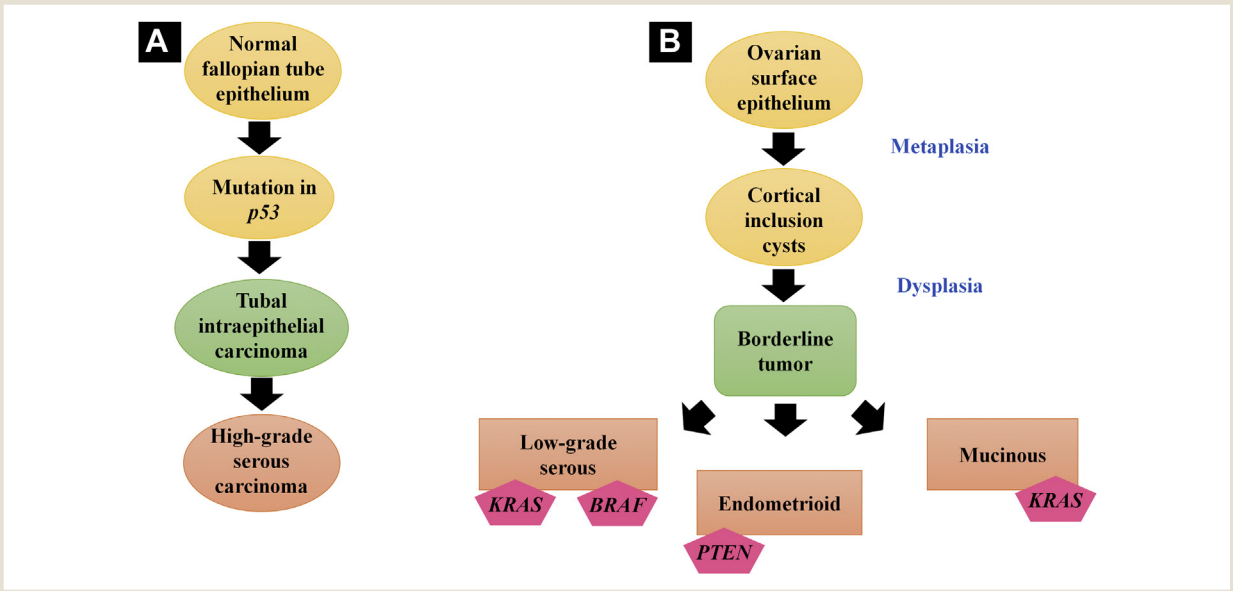
Figure 1 Schematic Diagram Depicting Types of Ovarian Cancer. (A) The Molecular Pathway That Leads to High-grade Serous Carcinoma (Type 2 Tumors). (B) The Molecular Pathway That Leads to Low-grade Serous, Endometrioid, and Mucinous Ovarian Cancer (Type 1 Tumors)



could potentially be exploited to produce artificial targets that silence oncogenes. This was found to be the case for the epithelial cell adhesion molecule, which is overexpressed in ovarian cancer, as well as in other cancers. Nunna et al[17] achieved targeted methylation of the promoter of the gene, which resulted in reduction in the expression of the gene, a mechanism that is very promising for cancer treatment.

Histone Modifications in Ovarian Cancer

Histone modifications are changes that affect the protruding tails of histone proteins, which are important for the packaging of the DNA in nucleosomes. These modifications can be associated with active or silent chromatin and occur normally in cells.[18] Lysine acetylation of histone 3 and 4 tails is associated with active chromatin (euchromatin), whereas lysine 9 methylation of histone 3 is associated with inactive chromatin (heterochromatin).[19,20] Expression of trimethylation of histone 3 at lysine 27 (H3K27me3) has been suggested to be a useful factor for the prognosis of ovarian cancer, because lower expression has been associated with lower survival. Methylation is performed by the enhancer of zeste 2 (EZH2) complex, which is abundantly expressed in many cancers.[21] Histone hypoacetylation and aberrant promoter methylation are 2 events that have been associated with low levels of the deleted in lung and esophageal cancer 1 (*DLEC1*) gene in ovarian cancer cell lines.[22] *DLEC1* is a gene believed to have tumor suppressor activity in other cancers, including lung and renal cancer.[23] Therefore, it is very important that the gene is studied further, because it could serve as a diagnostic biomarker, not only for ovarian, but also for other cancer types in the future. Furthermore, a family of transcription factor genes, which are silenced by histone modifications, is those encoding members of the GATA family of transcription factors, which—as their name suggests—bind to the nucleotide sequence "GATA."[24] The silencing of these transcription factors is achieved by histone 3 and 4 hypoacetylation and by trimethylation loss at lysine 4 of histone 3, which is in the vicinity of the promoter of the genes. The outcome of GATA silencing is the loss of a tumor suppressor gene, which is known as *Disabled-2*.[24] Therefore, this event promotes ovarian carcinogenesis.

The biology of epigenetic changes is of increasing research interest to many cancer research centers worldwide, because a vast potential exists for genes whose expression has been epigenetically modified to serve as biomarkers or therapeutic targets in cancer. The proteins that perform these modifications are also of great interest. HDAC inhibitors, which, as their name suggests, inhibit the deacetylation of histones, are very promising anticancer agents.[25] Belinostat (also known as PXD101; a small-molecule hydroxamate-type inhibitor of class I, II, and IV HDAC enzymes) is an example of such an inhibitor. It has antitumor activity in ovarian cancer, and its action can be enhanced by the use of carboplatin.[26] Acetylation is associated with active chromatin and, hence, gene expression. Therefore, deacetylase inhibitors in this case maintain the expression of tumor suppressor genes, which are normally silenced in cancer.

Epigenetic Regulation of Noncoding RNAs in Ovarian Cancer

Noncoding RNAs (ncRNAs) are RNA molecules that are transcribed, but not translated; therefore, they play a regulatory role in the cell. Such molecules include microRNAs (miRNAs, miR), small interfering RNAs, small nuclear RNAs, long ncRNAs, and Piwi-interacting RNAs, among others.[27] Their abundance can be

epigenetically altered, and this can influence the expression of certain genes they control. A long ncRNA was found to be epigenetically silenced in type 2 epithelial ovarian cancer. More specifically, it had been previously identified that the CpG island of the promoter of the gene encoding the long ncRNA was methylated and silenced in ovarian cancer tissues. Loss of this long ncRNA results in reduced proliferation and colony formation. This long ncRNA seems to have a role in cell polarity regulation; therefore, these findings have important implications for metastasis in ovarian cancer.[28] In another study, hypermethylation of the promoter region of the gene encoding miRNA-199b-5p resulted in its silencing. This event was associated with chemoresistance in ovarian cancer.[29] Therefore, exploring the biology of this miRNA could produce important findings for the prevention of this chemoresistance. Xiang et al[30] identified 2 miRNAs, miR-152 and miR-185, that are significantly downregulated in ovarian cell lines that are resistant to cisplatin through an epigenetic mechanism. From further experimentation, they concluded that these miRNAs are involved in cisplatin sensitivity.[30] miRNAs should be further investigated, because they could potentially serve as epigenetic markers, not only in ovarian but also in other cancers.

Conclusion

Epigenetic changes play a key role in the regulation of several genes in ovarian cancer. Understanding the mechanisms underpinning these changes is essential, because such understanding could contribute to the development of novel therapeutic targets with improved efficacy. Additionally, it could also contribute to the development of biomarkers and biomarker panels that could serve as prognostic, predictive, and/or diagnostic tools in ovarian cancer.

Disclosure

The authors declare that they have no competing interests.

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