Original Article
Middle East Journal of Cancer; July 2015; 6(3): 135-142

Evaluation of p53, PTEN and β-catenin Immunoexpressions in Primary Ovarian Epithelial Tumors

Fatemeh Sari Aslani*♦, Azarmidokht Momeni*, Mozhdeh Momtahan, Mozhgan Akbarzadeh Jahromi*, Amir Reza Dehghanian*, Dorna Motevali***

**Department of Pathology, Shiraz University of Medical Sciences, Shiraz, Iran*
***Department of Obstetrics and Gynecology, Shiraz University of Medical Sciences, Shiraz, Iran*

Abstract

Background: Ovarian cancer comprises a heterogeneous group of neoplasms. The prognosis cannot be predicted by histopathologic examination alone. The aim of this study is to evaluate p53, PTEN, and β-catenin expressions in primary ovarian carcinomas in an attempt to find a possible relationship with morphologic parameters and clinical findings.

Methods: The study included 100 epithelial ovarian tumors (borderline and carcinomas) from affiliated hospitals of Shiraz university of medical sciences during 2007-2013. Immunohistochemical staining for p53, PTEN, and β-catenin was performed on 65 serous, 18 mucinous, 10 endometrioid, 5 clear cell, and 2 mixed tumors.

Results: p53 expression pattern in serous carcinoma significantly differed from endometrioid carcinomas. Strong positivity (2+) in >50% of the tumor cells favored serous carcinoma. PTEN expression significantly differed in mucinous and serous carcinomas as well as in endometrioid carcinoma and borderline endometrioid tumor. There was significantly decreased β-catenin expression in the carcinomas compared with borderline tumors. In all of the different subtypes of ovarian carcinomas, we observed a significant association with decreased β-catenin expression to tumor grade as well as in serous carcinomas with increased nuclear grade, mitosis, and tumor grade. There was no significant relation between expressions of p53, PTEN, and β-catenin in epithelial ovarian tumors to FIGO staging, response to chemotherapy, serum CA-125 marker, and tumor recurrence.

Conclusion: p53 and PTEN are helpful in differentiation of some epithelial ovarian tumor subtypes. In serous carcinomas, diminished expression of β-catenin is associated with higher tumor and nuclear grade. This expression is significantly different in borderline and carcinomas.

Keywords: Epithelial ovarian tumor, Phosphatase and tensin homolog (PTEN), Tumor suppressor p53, Beta-catenin

♦Corresponding Author:
Fatemeh Sari-Aslani, MD
Associate Professor of Pathology, Pathology Department, Shiraz University of Medical Sciences, Shiraz, Iran
Email: ariasf@sums.ac.ir

Middle East Journal of Cancer

Received: October 25, 2014; Accepted: February 24, 2015

Introduction

The incidence of ovarian cancer has continually increased during the past ten years.[1] Serous tumors comprise approximately 70% of ovarian cancers.[2] Ovarian cancer is the most common female genital cancer in Iran. Sometimes, in epithelial ovarian tumors, a definite diagnosis and grading only on the basis of morphology is not possible. In such cases, mutations and gene expression studies can be helpful.[3,4] Each of the four major classes of epithelial tumors has distinct genetic abnormalities, which are not necessarily unique. For instance, in some studies, it has been demonstrated that the p53 mutation is uncommon in low-grade serous tumors. However, in higher grade serous tumors this mutation is common.[2,5] Clear cell and mucinous carcinomas are completely differentiated from serous tumors by their gene expression patterns.[2,6]

β-catenin is a part of the WNT (wingless) signaling pathway; it plays a role in adhesion and polarity of the cells. A previous study has reported that the immunohistochemistry (IHC) staining of β-catenin demonstrated a defect in the WNT signaling pathway in 25% of ovarian endometrioid adenocarcinomas. Although a mutation in this gene is common in this type of adenocarcinoma, it is rare in serous, mucinous, and clear cell carcinomas.[7]

The phosphatase and tensin homolog (PTEN) is a tumor suppressor that plays an important role in apoptosis and sensitization of ovarian cancers to cisplatin, which is the treatment of choice in ovarian cancers. This is to some extent fulfilled via p53-dependent mechanisms. Loss of PTEN in K-ras mutant mice leads to changes in their ovarian epithelial cells and rapid development of low-grade and invasive serous adenocarcinoma.[3,8,9] Low-grade tumors are usually associated with mutations, deletions, or gene amplifications in K-ras and PTEN, while high-grade tumors are associated with p53 mutations or deletions.[2,5]

Major prognostic factors in ovarian cancer consist of histological subtype, clinical staging according to the FIGO staging system, malignancy level, and residual tumor. Nevertheless, these items only provide an incomplete image of the tumor biology in ovarian cancers. Clinical staging on the basis of the FIGO staging system is a helpful prognostic factor in ovarian cancers. However, there is a remarkable heterogeneity in each FIGO stage.[10] Genotypic and phenotypic characteristics of ovarian cancers not only determine their metastatic potential, but also are responsible for their resistance against conventional treatments of cancer, recurrence, and poor prognosis. In this regard, identification of novel prognostic markers, which may provide a better understanding of the biology of primary epithelial ovarian tumors, is of great value. This study aims to investigate the expressions of p53, PTEN, and β-catenin markers by IHC analyses in carcinomas as well as ordinary and micropapillary borderline epithelial ovarian tumors. We have sought to determine the presence of any possible

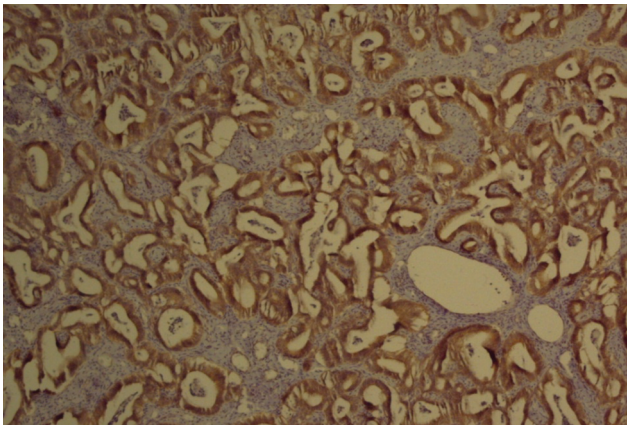

Figure 1A. Borderline endometrioid tumor shows 2+ phosphatase and tensin homolog (PTEN) cytoplasmic staining (100×).

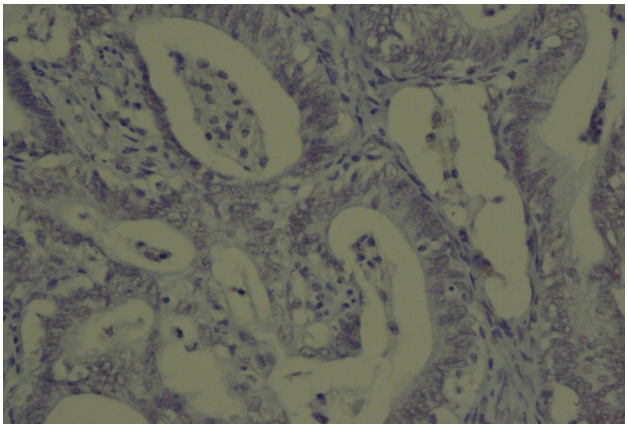

Figure 1B. Endometrioid carcinoma shows negative cytoplasmic staining of PTEN (400×).

relationship with clinicopathological findings. Two grading systems for serous carcinoma were also compared. The correlation of IHC results of these markers with parameters of a universal grading system was evaluated for the first time in this study.

Patients and Methods

This cross-sectional study collected pathology reports and clinical data of 100 patients with primary epithelial ovarian tumors from affiliated hospitals of Shiraz university of medical sciences from 2007 to 2013. The Medical Research Ethics Committee and Institutional Review Board (IRB) of Shiraz university of medical sciences approved the study protocol. The inclusion criteria were all patients with malignant or borderline primary epithelial ovarian tumors. Exclusion criteria were previous chemotherapy or radiotherapy, lack of sufficient tumor tissue, insufficient fixation, and widespread tumor necrosis.

All hematoxylin and eosin (H&E) slides were re-evaluated for confirmation of the diagnosis. The histological type and grade were assessed according to WHO classification and FIGO grading system, respectively. For serous carcinomas, we used a semi-quantitative grading based on the universal grading system modeled on the Nottingham system of breast cancer grading.[11] Data extracted from patients' tumor clinic follow-up records included: FIGO stage, CA-125 marker before treatment, need for chemotherapy, response to chemotherapy, tumor recurrence, or patient's death in the two-year follow up period. Serum CA-125 marker levels less than 35 before treatment were considered negative, whereas values above 35 were considered positive.

Immunohistochemistry (IHC) study

Immunohistochemical staining for β-catenin, p53 and PTEN were performed manually using a peroxidase kit with mouse monoclonal antibodies M3539 (clone β-catenin-1), M7001 (clone DO-7) and M3627 (clone 6H2.1), all purchased from Dako, Denmark. The slides were evaluated for both tumor cell percentage and intensity of immunoreactivity. p53 expression was nuclear and reported as 0 (absolutely no staining), +1 (< 50% of the cells, patchy weak) and +2 (>50%, strong).[12] For β-catenin, membrane staining was reported as negative (0), weakly positive (<10%), 1+ (10%-50%) and 2+ (>50%, strongly positive). Absence of membrane staining was considered negative.[13] PTEN expression was cytoplasmic, and reported as negative (0), weakly positive (1+, <50% of the cells) and strongly positive (2+, >50%).[11]

Statistical analysis

The data were analyzed with SPSS software, version 18. The relationship between expressions of p53, β-catenin, and PTEN markers to clinico-pathological findings was determined using the chi-square and Fisher's exact tests. For serous carcinomas, these tests were used for determination of the relationship between these

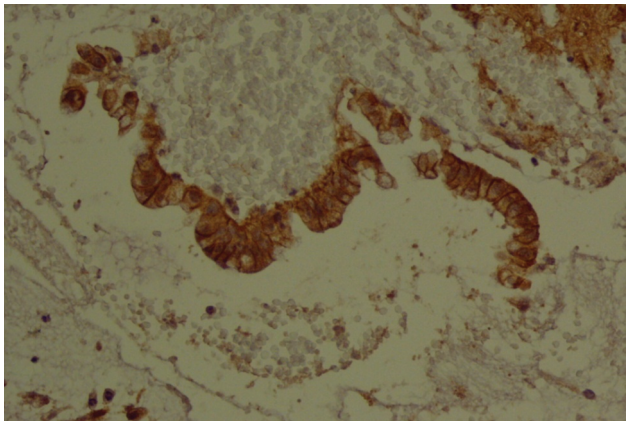

Figure 2A. Borderline serous tumor shows 2+ membranous staining of β-catenin (400×).

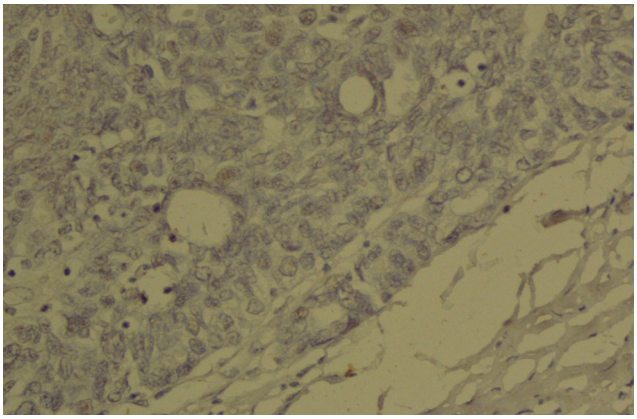

Figure 2B. High grade serous cystadenocarcinoma shows negative β-catenin staining (400×).

Table 1. Correlation between immunohistochemistry (IHC) expression patterns in p53, phosphatase and tensin homolog (PTEN), and β-catenin with histologic subtypes of ovarian surface epithelial tumors.

	p53			*P*-value	PTEN			*P*-value	β-catenin***			
	0	1+	2+	0	1+	2+			0	Weakly+	1+	2+
Serous tumor	0	16	44	0.044*	16	27	21	0.018**	4	19	14	25
Mucinous tumor	1	7	10		11	6	1		0	4	0	14
Clear cell carcinoma	0	3	2		2	2	1		0	2	1	2
Endometrioid tumor	0	6	4	0.044*	5	4	1		2	0	3	5
Mixed tumor	0	1	1		1	1	0		0	0	1	1

*: Comparison between serous and endometrioid tumors; **: Comparison between serous and mucinous tumors.; ***: *P*-value was not significant for this marker.

tumors as well as between nuclear grade, mitosis, and dominant tumor pattern. A *P*-value <0.05 was considered statistically significant.

Results

A total of 100 patients with epithelial ovarian tumors were selected. These included 30 borderline cases and 70 carcinomas. The tumors comprised 65 serous (19 borderline cases and 46 carcinomas), 18 mucinous (10 borderline cases and 8 carcinomas), 9 endometrioid carcinomas, one borderline endometrioid carcinoma, 5 clear cell carcinomas, and 2 cases of mixed tumor. With regards to histological grading, other than clear cell and mixed tumors, the carcinomas consisted of 34 well-differentiated, 15 moderately differentiated, and 14 poorly differentiated tumors. The patients' age range was 21-80 years, with a mean value of 48.58 years. For borderline and carcinoma cases the mean age was 41.32 and 51.25, respectively. We eliminated 8 cases for p53, 6 cases for β-catenin, and 4 cases for PTEN due to inappropriate IHC staining.

Expression of p53 was observed in 98.9% of the tumors. Severity of p53 expression in serous tumors significantly differed from endometrioid tumors. Severity of PTEN expression in serous tumors significantly differed from mucinous tumors. Table 1 shows two cases of mixed tumor.

Expression of PTEN in endometrioid carcinomas significantly differed from borderline tumors. (Figure 1 A,B) β-catenin expression in borderline tumors (serous, mucinous, and endometrioid) significantly differed from carcinomas (*P*=0.004). Low or lack of β-catenin expression was mostly observed in carcinomas (serous, mucinous, endometrioid and clear cell; Figure 2 A,B). Expressions of PTEN and p53 did not significantly differ in borderline tumors compared to carcinomas.

β-catenin expression in various grades of the above mentioned carcinomas significantly differed. There was an association between low expression and higher tumor grades as well as between higher expression and lower grades (*P*=0.004). Expressions of PTEN and p53 markers were not significantly associated with carcinoma grade. This grading was not indicated for clear cell carcinoma and mixed epithelial tumors (Table 2).

All borderline serous tumors expressed β-catenin. Low expression of β-catenin was associated with higher nuclear grade, higher mitosis rate, and higher tumor grade. Low β-catenin expression was associated with predominantly solid structure and higher tumor grades. The FIGO and universal grading systems for serous ovarian carcinomas showed a statistically significant relationship (Table 3).

We determined FIGO staging in 46 patients, serum CA-125 marker in 22 patients, need for chemotherapy and response in 33 patients, in addition to the one- and two-year follow up periods in 27 patients. Expression patterns of p53, PTEN, and β-catenin in epithelial ovarian tumors were not significantly related to FIGO staging, response to chemotherapy, serum CA-125 marker, and tumor recurrence.

Discussion

In ovarian cancers, the patient prognosis cannot be predicted based on precise histopathological evaluations. In epithelial ovarian tumors, the pro-apoptotic molecules (p53 and PTEN) as an index in the genetic pathway have been the target of

Table 2. Phosphatase and tensin homolog (PTEN), p53, and β-catenin expressions in various grades of ovarian carcinomas.

Type and FIGO grade		PTEN*			p53*		β-catenin				
		Negative	1+	2+	Weakly positive	Strongly positive	Negative	Weakly positive	1+	2+	*P*-value
Serous carcinoma											
	Well-differentiated	3	8	9	7	12	1	2	6	10	0.004
	Moderately differentiated	4	7	2	2	10	0	5	4	3	
	Poorly differentiated	5	4	3	4	8	3	7	0	2	
Mucinous carcinoma											
	Well-differentiated	3	3	1	2	5	0	3	0	4	
	Poorly differentiated	0	1	0	0	1	0	0	0	1	
Endometrioid carcinoma											
	I	4	3	0	5	2	0	0	3	4	
	II	1	0	0	0	1	1	0	0	0	
	III	0	1	0	1	0	1	0	0	0	

*: *P*-value was not significant for this marker.

modern therapeutic approaches.[12]

Different reports exist regarding mutation or overexpression of p53 gene in ovarian tumors. A prognostic value of p53 in ovarian cancer was evaluated through a meta-analysis of 62 previously published studies. Out of 62 studies, 25 reported this association with poor survival. When the meta-analysis was restricted to serous tumors, there was a significant association with poor prognosis. The percentage of tumors that expressed p53 ranged from 13.7%-82%.[13] There were not enough studies that reported results for the other histological subtypes to perform an analysis. The IHC results were highly dependent on a variety of methodological factors, choice of primary antibody, and IHC staining protocol. The differences in IHC staining protocols and cut-off values for positive expression (that ranged from >5% to >90% of positively stained cells) might have contributed to the observed heterogeneity.[13] In mucinous tumors, different studies reported the percentages of positive p53 from 6.3%-22%.[14] In the current study, expression of p53 in carcinomas was remarkably higher than previous studies. In some studies the immunoexpression level and pattern of p53 have significantly differed between high- and low-grade carcinomas.[15,16] The molecular genetic profile and behavior of serous carcinomas with grade 2 nuclei are virtually the same as those of serous carcinomas with grade 3 nuclei, hence they are viewed as high-grade tumors.[17] In the current study, p53 expression did not significantly different in low- and high-grade tumors. Chen L et al. concluded that expression of p53 mutants was closely correlated with prognosis of epithelial ovarian carcinoma and it could be considered a prognostic indicator.[18] The current study, as with other recent studies, could not determine a statistically significant relationship between p53 expression and clinical findings. This relationship also could not be detected in serous tumors. This could be explained by the small number of cases that had available information. It has been demonstrated that overexpression of mutant p53 would lead to a remarkable increase in resistance to anticancer agents.[19] Min et al. demonstrated that higher expression of p53 (in more than 50% of cells) was related to the type and grade of ovarian tumors. Higher expression of p53 was observed in ovarian carcinomas, however this was not the case in borderline tumors.[20] In another study, p53 expression was shown to be related to serous carcinoma. This expression was higher in serous types compared to endometrioid carcinomas.[12] However, there was an overlap in molecular and morphological characteristics of high grade endometrioid tumors and high grade serous

Table 3. Association between immunohistochemical (IHC) expressions of p53, phosphatase and tensin homolog (PTEN), and β-catenin and the universal grading system for serous ovarian carcinomas with regards to tumor structures, nuclear grading, and mitosis.

Universal grading system		β-catenin			P53*		PTEN*		
		Negative	Positive	*P*-value	1+	2+	0	1+	2+
Grade	**1**	1	14	<0.001	7	9	4	8	5
	2	8	8		3	13	3	7	6
	3	8	2		3	7	5	3	2
Architecture	**Glandular**	0	3	0.007	1	2	1	2	0
	Papillary	6	19		8	18	5	12	10
	Solid	11	2		4	9	6	4	3
Nuclear grade	**1**	1	8	0.009	4	5	3	3	4
	2	8	14		6	17	6	11	6
	3	8	2		3	7	3	4	3
Mitosis	**1**	4	10	0.029	6	9	3	8	5
	2	3	9		3	9	2	5	5
	3	10	5		4	11	7	5	3

*: *P*-value was not significant for this marker.

carcinomas. Some researchers have suggested that real high-grade endometrioid carcinomas are rare.[6] In the study by Gomes and Andrade, although a relatively limited number of primary ovarian carcinomas were studied, various gene expression patterns of p53 and PTEN were observed mainly in endometrioid and serous variants. This indicated the different pathogenic mechanisms that underlie various subtypes of ovarian carcinomas.[12] In the present study, the p53 expression pattern in serous tumors significantly differed from endometrioid tumors. Another study concluded that p53 and Ki67 were useful markers in differentiating between borderline mucinous and carcinomas. p53 in mucinous carcinomas had high values compared to borderline tumors.[14] In our study the expression of p53 was not significantly different for these tumors. This might be attributed to the lower number of mucinous tumors. All five cases with characteristic features of clear cell carcinoma were also positive for p53. This less common subtype could not be analyzed due to the low number of cases.

PTEN plays an important role in apoptosis and sensitization of ovarian cancers to cisplatin. Chen et al. have demonstrated that the PTEN expression pattern differed at various stages and grades of ovarian tumors. There was no association with different tumor subtypes and patient age.[21] In another study on 70 cases of primary ovarian carcinomas (serous, endometrioid, and mucinous), IHC expressions of p53 and PTEN showed a significantly higher expression of p53 in histologically high grade tumors - mainly of the serous subtype. Considering PTEN, only statistically significant lack of expression was seen in well-differentiated endometrioid carcinomas.[12] In our study, the severity of PTEN expression in serous tumors was significantly different from mucinous tumors. Also expression pattern of PTEN was significantly different in borderline endometrioid and endometrioid carcinomas. Common genetic changes that included the p53 mutation were probably responsible for similarity in gene expression pattern and morphology in high-grade serous carcinomas and high-grade endometrioid carcinomas. This overlap in gene expression and morphology could make precise differentiation of these two categories problematic for pathologists.[20] Brucka investigated the PTEN gene expression profile in endometrial cysts, clear cell carcinoma, and ovarian endometrioid tumors. The highest expression was observed in endometrial cysts, whereas the lowest expression was observed in endometrioid adenocarcinomas. However, the difference was not statistically significant.[22] It is interesting to note that endometrioid adenocarcinoma originated from endometrium also have the same PTEN expression

pattern; lack of cytoplasmic PTEN expression in the well-differentiated group.[15, 23] Martini et al. demonstrated that impaired expression of PTEN could be helpful in the diagnosis of endometriosis cases at high risk for malignancy.[24]

In previous studies, diminished expression of E-cadherin with β-catenin was associated with several markers of tumor progression[10] such as decreased tumor differentiation,[1, 10, 24] peritoneal metastasis, and advanced stages of the disease.[10] In some other studies, expression of these genes was related to the tumor subtype.[1, 10, 24, 25] Bhagat et al. demonstrated that aberrant expression of β-catenin in malignant epithelial tumors was more than seen with borderline epithelial ovarian tumors.[26] Decreased expression of this marker has been reported in advanced stages of serous and clear cell ovarian tumors.[1] In the current study, expression in borderline tumors (serous, mucinous, and endometrioid) significantly differed from carcinomas. Diminished expression in serous carcinomas was associated with higher tumor and nuclear grade. Expression of β-catenin did not show a statistically significant relationship with tumor subtype, staging, or other clinicopathological findings. This could be due to the small number of clinical data in the current study. Considering the relationship between decreased expression of β-catenin and higher grades of serous carcinomas, as an important prognostic factor in these patients, and the very high prevalence of these tumors in the studied population, it is important to perform further studies in this field with more clinical data.

Conclusion

p53 expression was higher than reported by previous studies; its expression could be useful in differentiating serous from endometrioid tumors. p53 and PTEN are helpful to differentiate some subtypes of epithelial ovarian tumors. PTEN can also help to differentiate serous from mucinous tumors. β-catenin helps to differentiate carcinomas from all borderline subtypes. In general, diminished expression of β-catenin in carcinomas is associated with higher tumor grade. We have observed no association between p53, PTEN, and β-catenin expression patterns and tumor recurrence, response to chemotherapy, and FIGO staging. However, considering the small number of the patients for whom these items were evaluated, and the short follow up period, the results cannot be generalized. Further studies with larger sample sizes and longer follow up periods are recommended. Limitations of the current study included patients' non-compliance, incomplete documents, and low number of certain tumor subtypes with low incidence.

Acknowledgment

This study was supported by grant number 88-4479 from Shiraz university of medical sciences.

Conflict of interest

No conflict of interest is declared.

References

1. Bhagat R, Premalata CS, Shilpa V, Pallavi VR, Ramesh G, Vijay CR, et al. Altered expression of β-catenin, E-cadherin, and E-cadherin promoter methylation in epithelial ovarian carcinoma. *Tumour Biol.* 2013;34(4):2459-68.
2. Mullany LK, Fan HY, Liu Z, White LD, Marshall A, Gunaratne P, et al. Molecular and functional characteristics of ovarian surface epithelial cells transformed by KrasG12D and loss of Pten in a mouse model *in vivo*. *Oncogene.* 2011;30(32):3522-36.
3. Itamochi H. Targeted therapies in epithelial ovarian cancer: Molecular mechanisms of action. *World J Biol Chem.* 2010;1(7):209-20.
4. Jelovac D, Armstrong DK. Recent progress in the diagnosis and treatment of ovarian cancer. *CA Cancer J Clin.* 2011;61(3):183-203.
5. Mullany LK, Liu Z, King ER, Wong KK, Richards JS. Wild-type tumor repressor protein 53 (Trp53) promotes ovarian cancer cell survival. *Endocrinology.* 2012;153(4):1638-48.
6. Cho KR. Ovarian cancer update: lessons from morphology, molecules, and mice. *Arch Pathol Lab Med.* 2009;133(11):1775-81
7. Wu R, Hendrix-Lucas N, Kuick R, Zhai Y, Schwartz DR, Akyol A, et al. Mouse model of human ovarian endometrioid adenocarcinoma based on somatic defects in the Wnt/beta-catenin and PI3K/Pten signaling pathways. *Cancer Cell.* 2007;11(4):321-33.
8. Chen L, Li L, Chen F, He D. Immunoexpression and

prognostic role of p53 in different subtypes of epithelial ovarian carcinoma. *J Biomed Res.* 2012;26(4):274-7.
9. Yan X, Fraser M, Qiu Q, Tsang BK. Over-expression of PTEN sensitizes human ovarian cancer cells to cisplatin-induced apoptosis in a p53-dependent manner. *Gynecol Oncol.* 2006;102(2):348-55.
10. Faleiro-Rodrigues C, Macedo-Pinto I, Pereira D, Ferreira VM, Lopes CS. Association of E-cadherin and beta-catenin immunoexpression with clinicopathologic features in primary ovarian carcinomas. *Hum Pathol.* 2004;35(6):663-9.
11. Silverberg SG. Histopathologic grading of ovarian carcinoma: a review and proposal. *Int J Gynecol Pathol.* 2000;19(1):7-15.
12. Gomes CP, Andrade LA. PTEN and p53 expression in primary ovarian carcinomas: immunohistochemical study and discussion of pathogenetic mechanisms. *Int J Gynecol Cancer.* 2006;16 Suppl 1:254-8.
13. de Graeff P, Crijns AP, de Jong S, Boezen M, Post WJ, de Vries EG, et al. Modest effect of p53, EGFR and HER-2/neu on prognosis in epithelial ovarian cancer: a meta-analysis. *Br J Cancer.* 2009;101(1):149-59.
14. Kamal CK, Simionescu CE, Mărgăritescu C, Stepan A. P53 and Ki67 immunoexpression in mucinous malignant ovarian tumors. *Rom J Morphol Embryol.* 2012;53(3 Suppl):799-803.
15. Sundov D, Caric A, Mrklic I, Gugic D, Capkun V, Hofman ID, et al. P53, MAPK, topoisomerase II alpha and Ki67 immunohistochemical expression and KRAS/BRAF mutation in ovarian serous carcinomas. *Diagn Pathol.* 2013;8:21.
16. Mishra SK, Crasta JA. An immunohistochemical comparison of P53 and Bcl-2 as apoptotic and MIB1 as proliferative markers in low-grade and high-grade ovarian serous carcinomas. *Int J Gynecol Cancer.* 2010;20(4):537-41.
17. Ayhan A, Kurman RJ, Yemelyanova A, Vang R, Logani S, Seidman JD, et al. Defining the cut point between low-grade and high-grade ovarian serous carcinomas: a clinicopathologic and molecular genetic analysis. *Am J Surg Pathol.* 2009;33(8):1220-4.
18. Chen L, Li L, Chen F, He D. Immunoexpression and prognostic role of p53 in different subtypes of epithelial ovarian carcinoma. *J Biomed Res.* 2012; 26(4): 274-7.
19. Kang HJ, Chun SM, Kim KR, Sohn I, Sung CO. Clinical relevance of gain-of-function mutations of p53 in high-grade serous ovarian carcinoma. *PLoS One.* 2013;8(8):e72609.
20. Min KW, Park MH. The expression of c-erbB-2, EGFR, p53 and Ki-67 in ovarian borderline tumors and carcinomas of the ovary. *Korean J Pathol.* 2007;41(5):296-306.
21. Che Y, Yao Q, Dai S, Luo B, Wang Y. Study of the mutation and expression of PTEN gene in endometrial carcinoma and epithelial ovarian cancer [Article in Chinese]. *Zhonghua Fu Chan Ke Za Zhi.* 2002;37(10):608-11.
22. Brucka A, Szyłło K. Immunoexpression of the PTEN protein and matrix metalloproteinase-2 in endometrial cysts, endometrioid and clear cell ovarian cancer. *Ginekol Pol.* 2013;84(5):344-51.
23. Kimura F, Watanabe J, Hata H, Fujisawa T, Kamata Y, Nishimura Y, et al. PTEN immunohistochemical expression is suppressed in G1 endometrioid adenocarcinoma of the uterine corpus. *J Cancer Res Clin Oncol.* 2004;130(3):161-8.
24. Martini M, Ciccarone M, Garganese G, Maggiore C, Evangelista A, Rahimi S, et al. Possible involvement of hMLH1, p16(INK4a) and PTEN in the malignant transformation of endometriosis. *Int J Cancer.* 2002;102(4):398-406.
25. Faleiro-Rodrigues C, Macedo-Pinto IM, Maia SS, Vieira RH, Lopes CS. Biological relevance of E-cadherin-catenin complex proteins in primary epithelial ovarian tumours. *Gynecol Obstet Invest.* 2005;60(2):75-83.
26. Bhagat R, Premalata CS, Shilpa V, Pallavi VR, Ramesh G, Vijay CR, et al. Altered expression of β-catenin, E-cadherin, and E-cadherin promoter methylation in epithelial ovarian carcinoma. *Tumour Biol.* 2013;34(4):2459-68.