

Expression of PH Domain Leucine-rich Repeat Protein Phosphatase, Forkhead Homeobox Type O 3a and RAD51, and their Relationships with Clinicopathologic Features and Prognosis in Ovarian Serous Adenocarcinoma

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Abstract

Background: Ovarian serous adenocarcinoma can be divided into low- and high-grade tumors, which exhibit substantial differences in pathogenesis, clinicopathology, and prognosis. This study aimed to investigate the differences in the PH domain leucine-rich repeat protein phosphatase (PHLPP), forkhead homeobox type O 3a (FoxO3a), and RAD51 protein expressions, and their associations with prognosis in patients with low- and high-grade ovarian serous adenocarcinomas.

Methods: The PHLPP, FoxO3a, and RAD51 protein expressions were examined in 94 high- and 26 low-grade ovarian serous adenocarcinomas by immunohistochemistry. The differences in expression and their relationships with pathological features and prognosis were analyzed.

Results: In high-grade serous adenocarcinomas, the positive rates of PHLPP and FoxO3a were 24.5% and 26.6%, while in low-grade tumors, they were 23.1% and 26.9%, respectively ($P < 0.05$ vs. the control specimens; low- vs. high-grade: $P > 0.05$). The positive rates of RAD51 were 70.2% and 65.4% in high- and low-grade serous adenocarcinomas, respectively ($P < 0.05$ vs. the control specimens; low- vs. high-grade: $P > 0.05$). Meanwhile, in high-grade tumors, Stage III/IV tumors and lymph node and omental metastases were significantly associated with lower PHLPP and FoxO3a and higher RAD51 expression. The 5-year survival rates of patients with PHLPP- and FoxO3a-positive high-grade tumors (43.5% and 36.0%) were significantly higher than in patients with PHLPP-negative tumors (5.6% and 7.2%, respectively; $P < 0.05$). Similarly, the 5-year survival rate of RAD51-positive patients (3.0%) was significantly lower than in negative patients (42.9%; $P < 0.05$). In low-grade tumors, the PHLPP, FoxO3a, and RAD51 expressions were not significantly correlated with lymph node metastasis, omental metastasis, Federation of Gynecology and Obstetrics stage, or prognosis.

Conclusions: Abnormal PHLPP, FoxO3a, and RAD51 protein expressions may be involved in the development of high- and low-grade ovarian serous adenocarcinomas, suggesting common molecular pathways. Decreased PHLPP and FoxO3a and increased RAD51 protein expression may be important molecular markers for poor prognosis, and RAD51 may be an independent prognosis factor, of high-grade, but not low-grade, ovarian serous adenocarcinomas.

Key words: Forkhead Homeobox Type O 3a; Immunohistochemistry; Ovarian Serous Adenocarcinomas; PH Domain Leucine-rich Repeat Protein Phosphatase; Prognosis; RAD51

INTRODUCTION

Ovarian serous adenocarcinoma is the most common ovarian epithelial malignant tumor. It can be divided into low- and high-grade tumors, which exhibit substantial differences in the pathogenesis, clinicopathological characteristics, and prognosis.^[1] Low-grade serous carcinomas undergo a progressive, gradual developmental process from benign

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serous cystadenoma to borderline serous cystadenoma to malignant serous adenocarcinoma, while high-grade serous adenocarcinomas develop directly from the oviduct epithelium or ovarian inclusion cysts.

However, it remains unclear whether the two tumor types have completely different molecular mechanisms, whether there is a common molecular basis, and whether the genes involved in the development of these tumors affect the patients' prognosis. Recent studies^[2,3] have indicated that the PI3K/Akt signaling pathway is involved in the initiation and development of ovarian serous adenocarcinoma. For example, PH domain leucine-rich repeat protein phosphatase (PHLPP) is known to negatively regulate Akt and its downstream kinase by dephosphorylating the hydrophobic core of Akt (Akt1 Ser473), thereby antagonizing PI3K/Akt signaling and inhibiting tumor growth. Forkhead homeobox type O 3a (FoxO3a), an important downstream signaling molecule of Akt, is involved in the initiation and development of tumor cells and regulation of cell proliferation after being phosphorylated by Akt.^[4,5] Further, RAD51, a DNA double-strand break repair gene,^[6] has also been implicated in the PI3K/Akt signaling pathway: phosphorylated FoxO3a binds to the promoter of the downstream gene RAD51, thereby regulating RAD51 and promoting tumor development and metastasis.

The present study aimed to (1) determine the differences in the protein expressions of PHLPP, FoxO3a, and RAD51 between high- and low-grade ovarian serous adenocarcinomas, (2) explore the roles of these three genes in the initiation of the different grades of ovarian serous adenocarcinoma, and (3) analyze the relationships between their protein expressions and prognosis.

METHODS

Ethics

The study was approved by the Ethics Committee of the Fourth Hospital of Hebei Medical University and was conducted in accordance with the *Declaration of Helsinki* 1975, as revised in 2000.

Participant selection and description

Patients with ovarian serous adenocarcinoma hospitalized and treated from January 2006 to January 2011 in our hospital were included in the ovarian serous adenocarcinoma groups. No patients received chemotherapy and radiotherapy preoperatively, and their diagnosis was confirmed by pathological examinations postoperatively. All patients had complete clinical data and were graded according to the World Health Organization histological classification for ovarian tumors (2014 edition). Ninety-four and twenty-six cases of high- and low-grade ovarian serous adenocarcinomas were included in this study. Among the high-grade group (mean age, 55 years; range, 36–78 years), there were 26 and 68 patients with International Federation of Gynecology

and Obstetrics (FIGO) Stage (2014 edition) I–II and III–IV disease, respectively. There were thirty and 64 patients with and without lymph node metastasis (LNM), and 41 and 53 patients with and without omental metastasis, respectively. Among the low-grade group (mean age, 44 years; range, 26–70 years), there were 18 and eight patients with FIGO Stage I–II and III–IV disease, respectively. There were 4 and 22 patients with and without LNM, and 5 and 21 patients with and without omental metastasis, respectively. Moreover, 21 cases of borderline serous cystadenoma (mean age, 38 years; range, 18–59 years) and 35 cases of benign serous cystadenoma (mean age, 45 years; range, 22–69 years) were included, along with thirty normal ovaries and thirty oviducts (mean age, 48 years; range, 36–62 years) removed from patients undergoing surgery for uterine fibroids, which were used as controls for the low- and high-grade tumor groups, respectively.

Expression of TP53 protein in high- and low-grade serous carcinoma: Among the 94 cases of high-grade, 92 cases (97.9%) showed strong staining of TP53, while among the 26 cases of low-grade serous adenocarcinoma, there was no case of strong staining.

Technical information

Experimental procedures

The operative specimens were examined, and the patients were diagnosed by experienced pathologists. Paraffin blocks of the specimens were cut, and immunohistochemical staining of the sections was conducted as follows: the samples were sliced, deparaffinized, dehydrated, and subjected to antigen retrieval before blocking with goat serum. Subsequently, the slides were placed in a humidifier chamber and incubated at room temperature for 45 min. The tissues were incubated with primary antibodies (working concentrations of anti-PHLPP, RAD51, and FoxO3a of 1:100, 1:100, and 1:150, respectively, Biogot Biotechnology Co., Ltd., Nanjing, China) in the sealed humidifier chamber at 4°C overnight. After washing of the slides, goat anti-rabbit secondary antibodies were added and the slides were incubated in the humidifier at 37°C for 30 min. Next, horseradish peroxidase-labeled streptavidin was added and the slides were incubated in the humidifier at 37°C for 30 min, after which the tissues were stained by 3,3'-diaminobenzidine, followed by counterstaining, destaining, dehydration, clearing, and mounting. Finally, the slides were air-dried and observed under a microscope.

Evaluation standard for staining

The slides were scored by pathologists according to the below criteria for PHLPP, FoxO3a, and RAD51 under a microscope. Ten high-magnification fields were counted, and the average cell numbers were used.

PH domain leucine-rich repeat protein phosphatase-positive criteria

Cells with cytoplasm stained brown were determined as positive. The staining intensity was scored as follows:

uncolored (0), pale yellow (1 point), brown (2 points), and tan (3 points). The percentages of positive cells were scored as 0, 1, 2, 3, and 4 points for <5%, 5–25%, 26–50%, 51–75%, and >75%, respectively. According to the combined results of the two scores, the staining was divided into four levels: 0–1 point was defined as negative (–), ≥ 2 was defined as positive.^[7]

Forkhead homeobox type O 3a-positive criteria

Cells with nuclei staining brown were defined as positive. No staining was scored as 0, positive cells <10% as 1 point, 11–25% as 2 points, and >26% as 3 points. A total score of >2 points was defined as positive.^[8]

RAD51-positive criteria

Cells with cytoplasm or nuclei staining brown were defined as positive. Positive cells <10% and $\geq 10\%$ were defined as negative and positive expressions, respectively.^[9]

Statistical analyses

SPSS 16.0 software (SPSS Inc., USA) was used for the statistical analysis. Categorical data were analyzed by the Chi-square test, and the survival rates were calculated by Kaplan-Meier univariate analysis. Differences in the survival rates between the different groups were determined by the log-rank test. Cox multivariate survival analysis was used for analysis of independent prognostic factors. The statistical significance level was set as $P < 0.05$.

RESULTS

Expression of PH domain leucine-rich repeat protein phosphatase in ovarian serous adenocarcinomas and its association with clinicopathological features and prognosis

Among 94 cases of high-grade serous adenocarcinoma, 23 cases (24.5%) were PHLPP-positive, which was significantly lower than the positive rate of 86.7% in the normal oviduct group ($P < 0.05$). Of 26 low-grade serous adenocarcinoma cases, 6 (23.1%) were positive, which was significantly lower than that in the normal ovary, borderline serous cystadenoma, and serous cystadenoma groups ($P < 0.05$). However, there was no difference in the positive expression rates between high- and low-grade tumors [$P > 0.05$; Figure 1 and Table 1].

In high-grade tumors, LNM, FIGO Stage III/IV tumors, and omental metastasis were significantly associated with lower PHLPP protein expression levels [$P < 0.05$; Table 2]. On the other hand, there were no significant correlations between the clinicopathological features and PHLPP protein expression in low-grade tumors [$P > 0.05$].

Among the 94 patients with high-grade serous adenocarcinoma, the 5-year survival rates of PHLPP-negative and positive patients were 5.6% and 43.5%, respectively [$P < 0.05$, Figure 2]. Among the 26 cases of low-grade serous adenocarcinoma, however, the expression of PHLPP did not significantly associate with the 5-year survival rate of the patients [$P > 0.05$].

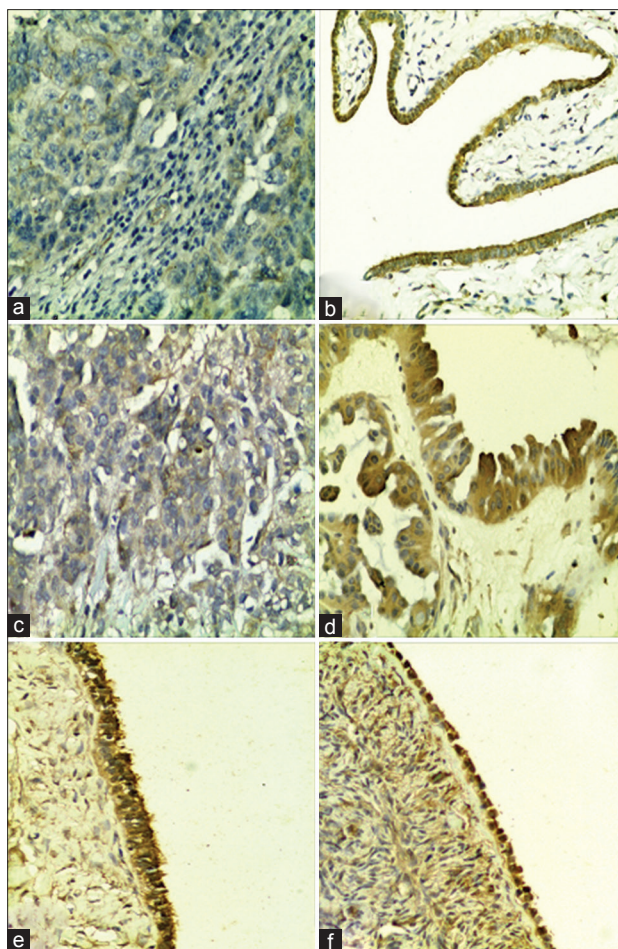


Figure 1: Protein expression of PH domain leucine-rich repeat protein phosphatase in high- and low-grade serous ovarian adenocarcinoma by immunohistochemical analysis (original magnification $\times 200$). (a) Negative high-grade adenocarcinoma, (b) positive normal oviduct, (c) negative low-grade adenocarcinoma, (d) positive borderline serous cystadenoma, (e) positive cystadenoma, (f) positive normal ovary.

Expression of forkhead homeobox type O 3a in ovarian serous adenocarcinoma and its association with clinicopathological features and prognosis

Among the 94 cases of high-grade serous adenocarcinoma, 25 (26.6%) were positive for FoxO3a protein expression, which was significantly lower than the positive rate of 90.0% in the normal oviduct group ($P < 0.05$). Among the 26 cases of low-grade serous carcinoma, 7 (26.9%) were positive, which was significantly lower than in the normal ovary, borderline serous cystadenoma, and serous cystadenoma groups ($P < 0.05$). However, the difference in the positive expression rates between the high- and low-grade groups was not significant [$P > 0.05$; Figure 3 and Table 3].

In high-grade tumors, LNM, FIGO Stage III/IV tumors, and omental metastasis were significantly associated with lower FoxO3a protein expression [$P < 0.05$, Table 2]. On the other hand, in low-grade tumors, the FoxO3a protein expression showed no association with clinicopathological features ($P > 0.05$).

Among the 94 cases of high-grade serous adenocarcinoma, the 5-year survival rates of FoxO3a-negative and positive patients were 7.2% and 36.0%, respectively [$P < 0.05$, Figure 4]. Among the 26 cases of low-grade serous adenocarcinoma, the expression of FoxO3a showed no significant association with the 5-year survival rate ($P > 0.05$).

Expression of RAD51 in ovarian serous adenocarcinoma and its association with clinicopathological features and prognosis

Among the 94 cases of high-grade serous adenocarcinoma, 66 (70.2%) were positive for RAD51 protein, which was significantly higher than that in the normal oviduct group [$P < 0.05$; Figure 5 and Table 4]. Among the 26 cases of low-grade serous carcinoma, 17 (65.4%) were positive, which was significantly higher than in the normal ovary, borderline serous cystadenoma, and serous cystadenoma groups [$P < 0.05$; Figure 5 and Table 4]. However, the difference in the positive expression rates between the high- and low-grade groups was not significant ($P > 0.05$).

Table 1: Protein expression of PHLPP in high- and low-grade ovarian serous adenocarcinomas

Groups	Cases (n)	Negative (n)	Positive (n)	Positive rate (%)
High-grade serous adenocarcinoma	94	71	23	24.5*
Normal oviduct	30	4	26	86.7
Low-grade serous adenocarcinoma	26	20	6	23.1†
Borderline serous cystadenoma	21	5	16	76.2
Serous cystadenoma	35	3	32	91.4
Normal ovary	30	2	28	93.3

* $P < 0.05$ compared with normal oviducts; † $P < 0.05$ compared with normal ovaries, cystadenomas, and serous cystadenomas. PHLPP: PH domain leucine-rich repeat protein phosphatase.

Table 2: Relationships between PHLPP, FoxO3a, and RAD51 protein expressions and clinicopathological features of high-grade serous ovarian adenocarcinoma

Pathological feature	Cases (n)	PHLPP (n)			FoxO3a (n)			RAD51 (n)		
		Positive	Negative	P	Positive	Negative	P	Positive	Negative	P
Age (years)										
<55	40	10	30	0.918	12	28	0.226	26	14	0.342
≥55	54	13	41		13	41		40	14	
LNM										
Present	30	3	27	0.025	4	26	0.046	26	4	0.017
Absent	64	20	44		21	43		40	24	
FIGO stage										
I-II	26	16	10	<0.001	12	14	0.008	12	14	0.002
III-IV	68	7	61		13	55		54	14	
OM										
Present	41	5	36	0.015	5	36	0.005	35	6	0.005
Absent	53	18	35		20	33		31		

LNM: Lymph node metastasis; FIGO: International Federation of Gynecology and Obstetrics; OM: Omental metastasis; FoxO3a: Forkhead homeobox type O 3a.

In high-grade tumors, LNM, FIGO Stage III/IV tumors, and omental metastasis were significantly associated with higher RAD51 protein expression levels [$P < 0.05$; Table 2]. Conversely, there was no significant correlation between clinicopathological features and RAD51 expression in low-grade tumors ($P > 0.05$).

Finally, among the 94 cases of high-grade serous adenocarcinoma, the 5-year survival rates of RAD51-positive and negative patients were 3.0% and 42.9%, respectively [$P < 0.05$; Figure 6]. Among the 26 cases of low-grade serous adenocarcinoma, the expression of RAD51 showed no significant association with the 5-year survival rate ($P > 0.05$).

Prognostic multivariate survival analyses of high-grade serous ovarian adenocarcinoma

The Cox multivariate analysis results showed that FIGO staging, LNM, and RAD51 protein expression were

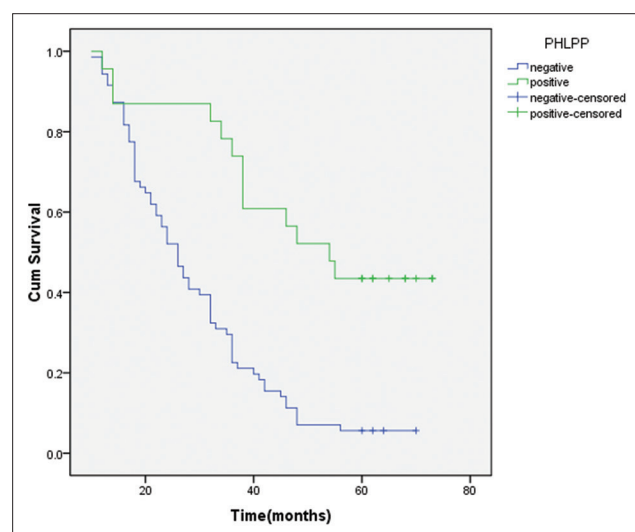


Figure 2: Five-year survival curves of PH domain leucine-rich repeat protein phosphatase-positive and negative patients with high-grade ovarian serous adenocarcinomas.

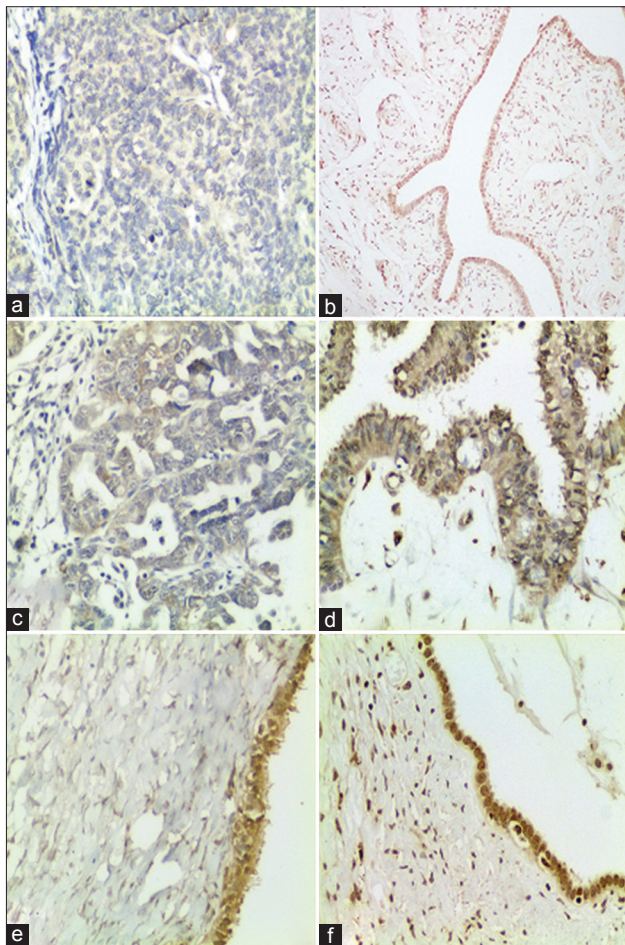


Figure 3: Protein expression of forkhead homeobox type O 3a in high- and low-grade serous ovarian adenocarcinomas by immunohistochemical analysis (original magnification $\times 200$). (a) Negative high-grade adenocarcinoma, (b) positive normal oviduct, (c) negative low-grade adenocarcinoma, (d) positive borderline cystadenoma, (e) positive cystadenoma, (f) positive normal ovary.

independent prognostic factors of patients with high-grade serous ovarian adenocarcinoma [$P < 0.05$; Table 5].

DISCUSSION

Ovarian cancer is one of the most common malignancies of the female reproductive system, and has the highest mortality among all gynecologic malignant tumors, thus seriously affecting women's health and lives. Recent studies have suggested that ovarian serous adenocarcinomas can be divided into high- and low-grade tumors and that the initiation mechanisms, clinical pathological features, and prognoses of the two types greatly differ.^[1,10] Thus, ovarian serous adenocarcinomas have complex biological behaviors. However, although the pathogenesises of the two types involve different molecular pathways, there is some common basis. Recent studies have found that the PI3K/Akt signaling pathway is closely associated with the initiation and progression of ovarian cancer. PHLPP, a known tumor suppressor, can negatively regulate Akt and its downstream kinases, thereby antagonizing the

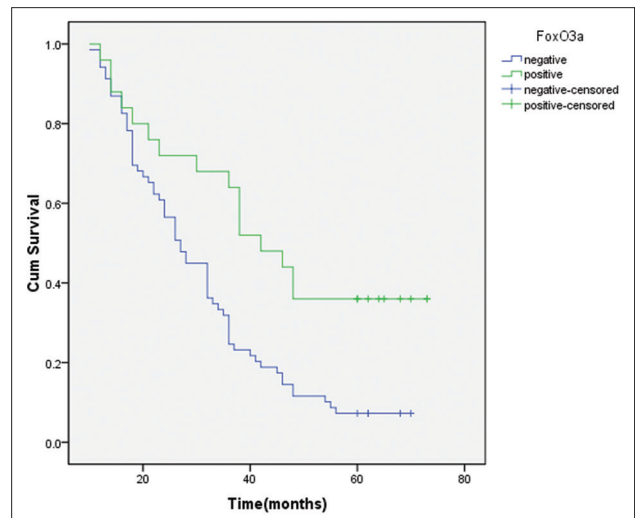


Figure 4: Five-year survival curves of forkhead homeobox type O 3a-positive and negative patients with high-grade ovarian serous adenocarcinomas.

Table 3: Protein expression of FoxO3a in high- and low-grade serous adenocarcinomas

Groups	Cases (n)	Negative (n)	Positive (n)	Positive rate (%)
High-grade serous adenocarcinoma	94	69	25	26.6*
Normal oviduct	30	3	27	90.0
Low-grade serous adenocarcinoma	26	19	7	26.9 [†]
Borderline serous cystadenoma	21	6	15	71.4
Serous cystadenoma	35	6	29	82.9
Normal ovary	30	1	29	96.7

* $P < 0.05$ compared with normal oviducts; [†] $P < 0.05$ compared with normal ovaries, cystadenomas, and serous cystadenomas. FoxO3a: Forkhead homeobox type O 3a.

Table 4: Protein expression of RAD51 in high- and low-grade serous adenocarcinomas

Groups	Cases (n)	Negative (n)	Positive (n)	Positive rate (%)
High-grade serous adenocarcinoma	94	28	66	70.2*
Normal oviduct	30	5	25	16.7
Low-grade serous adenocarcinoma	26	9	17	65.4 [†]
Borderline serous cystadenoma	21	11	10	47.6
Serous cystadenoma	35	26	9	25.7
Normal ovary	30	24	6	20.0

* $P < 0.05$ compared with normal oviducts; [†] $P < 0.05$ compared with normal ovaries, cystadenomas, and serous cystadenomas.

PI3K/Akt signaling pathway and inhibit tumor growth. Moreover, as an important downstream signaling molecule of Akt, after being phosphorylated by Akt, FoxO3a is

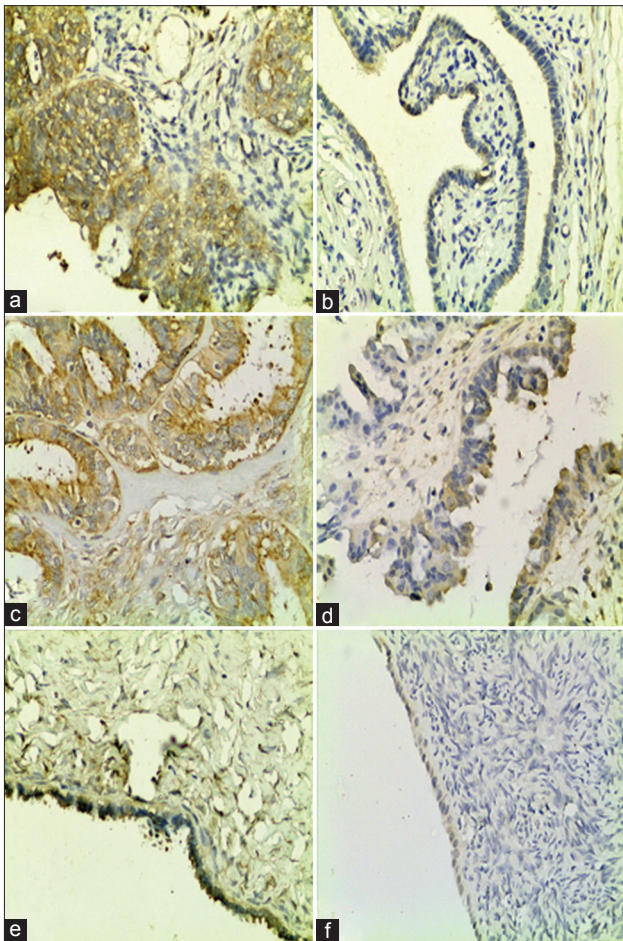


Figure 5: Protein expression of RAD51 in high- and low-grade serous ovarian adenocarcinomas by immunohistochemical analysis (Original magnification $\times 200$). (a) Positive high-grade adenocarcinoma, (b) negative normal oviduct, (c) positive low-grade adenocarcinoma, (d) negative borderline cystadenoma, (e) negative cystadenoma, (f) negative normal ovary.

involved in the initiation and development of tumor cells and regulates cell proliferation. RAD51, a downstream target of FoxO3a, also plays an important role in PI3K/Akt signaling; FoxO3a binds to the promoter of RAD51, consequently regulating RAD51 and promoting tumor development and metastasis. In this study, by examining the differences in the protein expression levels of PHLPP, FoxO3a, and RAD51 between high- and low-grade ovarian serous adenocarcinomas, the roles of the three genes in the development of the different grades of ovarian serous adenocarcinoma were explored, and the relationships between the protein expression levels and the patient prognosis were analyzed.

Role of the tumor suppressor PH domain leucine-rich repeat protein phosphatase in the development of ovarian serous adenocarcinoma and its association with patient prognosis

The tumor suppressor PHLPP, located at 18q21.33, can directly dephosphorylate ser473 in the hydrophobic group of Akt, thus negatively regulating Akt and its

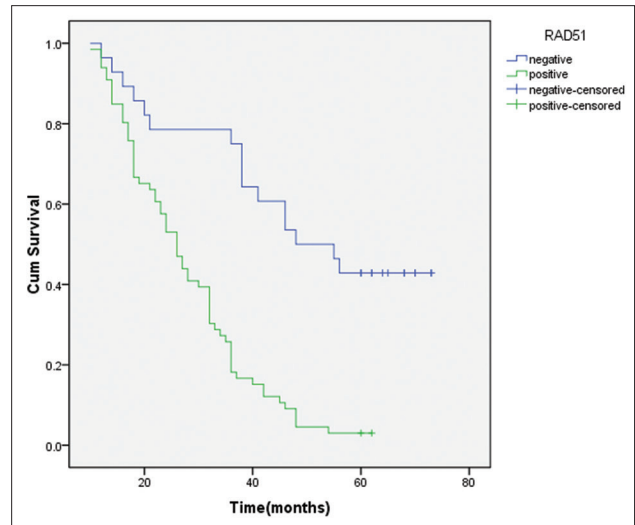


Figure 6: Five-year survival curves of RAD51-positive and negative patients with high-grade ovarian serous adenocarcinomas.

Table 5: Multivariate analysis of independent factors associated with the 5-year overall survival rate in patients with high-grade serous ovarian adenocarcinoma

Factors	β	Wald value	RR	95% CI	P
FIGO	-1.239	12.287	0.290	0.145–0.759	0.000
OM	-0.251	1.062	0.778	0.482–1.254	0.303
LNM	-0.887	10.892	0.412	0.243–0.697	0.001
PHLPP	0.241	0.273	1.272	0.516–3.133	0.601
FoxO3a	0.207	0.471	1.230	0.682–2.219	0.492
RAD51	-0.967	5.131	0.380	0.165–0.878	0.024

RR: Risk ratio; CI: Confidence interval; LNM: Lymph node metastasis; FIGO: International Federation of Gynecology and Obstetrics; OM: Omental metastasis; FoxO3a: Forkhead homeobox type O 3a; PHLPP: PH domain leucine-rich repeat protein phosphatase.

downstream targets. It has been reported that PHLPP can act synergistically with PTEN, another well-known tumor suppressor gene, thereby significantly inhibiting the proliferation of colorectal cancer cells, and negatively regulating PI3K/Akt signaling.^[11] Accordingly, studies have shown that the PHLPP expression is significantly decreased in colon, breast, prostate, and lung cancers.^[5,7,12]

The results of the present study showed that the positive rate of PHLPP protein expression in the high-grade group was significantly lower than that in the oviduct group. The PHLPP-positive expression rate was decreased gradually from normal ovarian tissue to benign tumors to borderline tumors to low-grade adenocarcinomas, indicating that loss of PHLPP expression may be associated with the initiation of ovarian serous adenocarcinoma development, whereas no significant difference was observed between high-grade and low-grade tumors. Further, the PHLPP expression level showed significant associations with LNM, FIGO stages, and omental metastasis in high-grade tumors, but not in low-grade tumors. Similarly, the 5-year survival rates of PHLPP-positive and negative high-grade tumors significantly differed, whereas

no difference was seen in the low-grade tumors. These findings suggest that PHLPP may be a predictor of prognosis of high-grade ovarian serous adenocarcinoma.

Role of the tumor suppressor forkhead homeobox type O 3a in the development of ovarian serous adenocarcinoma and its association with patient prognosis

FoxO3a is an important member of the FOXO family, located on at 6q21, and encodes a 673-amino acid long protein.^[13] FoxO3a, as an important signaling molecule downstream of Akt in the PI3K-Akt signaling pathway, binds to the 14-3-3 chaperone protein after being phosphorylated by Akt, and subsequently translocates from the nucleus to the cytoplasm, thereby activating the apoptotic genes Bim, FasL, and tumor necrosis factor and its related apoptosis-inducing ligand TRAIL, consequently promoting apoptosis.^[14] In addition to its role in apoptosis, FoxO3a plays critical roles in regulating cell proliferation, metabolism, the stress response, and the life span of cancer cells.^[15] Particularly, another target molecule of FoxO3a is the cell cycle-regulating gene P27; overexpression of FoxO3a can arrest the cell cycle in the G0/G1 phase by upregulating P27. In addition, overexpression of FoxO3a can significantly inhibit the proliferation of tumor cells and result in tumor cell G2 arrest, further promoting tumor cell apoptosis.^[16,17]

The results of our analyses showed that the FoxO3a-positive rate in the high-grade tumors was significantly lower than that in the normal oviducts. In the low-grade adenocarcinomas, borderline cystadenomas, cystadenomas, and normal ovaries, the positive expression rates of FoxO3a protein were gradually reduced from normal ovaries to the low-grade adenocarcinomas, and the FoxO3 positive rate of the low-grade tumors was significantly lower than those of the borderline cystadenomas, cystadenomas, and normal ovaries, while the positive rate of borderline cystadenomas was significantly lower than those of cystadenomas and normal ovaries. Moreover, the FoxO3a expression level showed significant associations with LNM, FIGO stage, and omental metastasis in high-grade, but not low-grade tumors. These results suggest that FoxO3a may play a role in the oncogenesis and development of ovarian serous adenocarcinomas. In the high-grade tumors, the 5-year survival rate of FoxO3a-positive patients was significantly higher than that of the negative patients, whereas no difference in survival was seen in low-grade tumors, indicating that positive FoxO3a expression may have a long-term protective effect on cancer patients, and FoxO3a may hence have certain significance in the prediction of the patients' long-term prognosis. Interestingly, it has been reported that tumor cells with low FoxO3a expression can develop drug resistance to paclitaxel and that inhibiting FoxO3a expression with RNAi can reduce E1A-mediated sensitivity to paclitaxel.^[18] Therefore, FoxO3a protein may represent a potential prognostic indicator for high-grade ovarian serous adenocarcinoma.

Role of RAD51 in the development of ovarian serous adenocarcinoma and its association with patient prognosis

RAD51 is a human homologous recombination repair gene. Too strong or too weak homologous recombination will result in genome instability and DNA damage, and the consequent gene loss, loss of heterozygosity, and/or genetic translocation are important causes of tumorigenesis and tumor progression.^[19] Studies have found that RAD51 protein is overexpression in the breast, prostate, pancreatic, and colon cancers. Such overexpression of RAD51 can provide certain advantages for the growth of cancer cells by promoting the initiation and progression of cancer. However, to date, studies on the expression of RAD51 in ovarian cancers are few.

The results of this study showed that the RAD51-positive rate in high-grade tumors was significantly higher than that of normal oviducts, suggesting that RAD51 may play a role in the initiation of high-grade ovarian serous carcinomas. The RAD51-positive rate gradually increased from normal ovaries to cystadenomas to borderline cystadenomas to low-grade serous adenocarcinomas. Particularly, the positive rate of low-grade tumors was significantly higher than that in benign tumors and normal oviducts. The gradual change in the RAD51 expression suggests that the initiation of low-grade serous adenocarcinoma is a gradual procedure from benign to borderline to low-grade adenocarcinoma. However, no significant difference in the expression of RAD51 was found between high- and low-grade tumors. The expression level of RAD51 showed significant associations with LNM, FIGO stage, and omental metastasis in high-grade, but not low-grade tumors.

Herein, the 5-year survival rate of patients with RAD51-positive high-grade adenocarcinomas was significantly lower than that of patients with RAD51-negative tumors, while no difference in survival was found in low-grade tumors. In other words, we found that high-grade tumor patients with high expression of RAD51 had a lower survival rate and poor prognosis, indicating that RAD51 may be a prognostic factor in high-grade ovarian serous adenocarcinoma. Furthermore, multivariate survival analyses showed that RAD51 was an independent prognostic factor, which may be of clinical significance in terms of the development of molecular targeted therapy, in addition to FIGO stage, and LNM. Further investigations on RAD51 are required to confirm these findings and to hopefully be able to improve the quality of life and prognosis of patients using targeted therapy.

In conclusion, the results of the present study indicate that PHLPP, FoxO3a, and RAD51 may play certain roles in the pathogenesis of the different grades of ovarian serous adenocarcinomas. Meanwhile, they also suggest the possible presence of common molecular mechanisms in the pathogenesis of the different grades. Increased protein expression of RAD51 may be an important molecular marker for poor prognosis of high-grade ovarian serous

adenocarcinomas. However, considering the limited number of cases selected, which may have affected the significance of the results, further studies are required to improve and supplement these findings.

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Conflicts of interest

There are no conflicts of interest.

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