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RESEARCH ARTICLE

Predictive and prognostic value of SALL4 and Tau protein in serous ovarian cancer patients treated with chemotherapy.

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Abstract

Background: Serous ovarian carcinoma (SOC) forms high percentage of epithelial ovarian cancer with frequent relapse and drug resistance. Spalt-like transcription factor SALL4 is a zinc finger transcriptional factor that regulates multiple downstream targeted genes included in normal development, essential in maintaining pluripotency and self-renewal of embryonic stem cells (escs). The role of SALL4 in serous ovarian carcinoma (SOC) is still controversial although it is involved in some tumor progression. Tau protein (50–64 kd), a product of gene located in chromosome 17 (17q21) shows the ability of combining to beta-tubulin. It may bind to the exterior as well as to the interior microtubules surface and its function has been extensively associated with neurodegenerative diseases.

Aim of the work: To explore and assess the Predictive and prognostic role of SALL4 and Tau protein immunohistochemical expressions in serous ovarian carcinoma patients treated with paclitaxel/ carboplatin first-line chemotherapy.

Methods: Immunohistochemical expressions of SALL4 and Tau protein were evaluated in sections from 50 paraffin blocks of serous ovarian carcinoma patients. The relationship between their level of expressions, prognosis and predictive value in serous ovarian carcinoma patients treated with paclitaxel/platinum first-line chemotherapy was analyzed.

Results: The expression of SALL4 in serous ovarian carcinoma was significantly positively correlated with grade, stage, lymph node metastasis ($p < 0.001$), local recurrence of the tumor ($p = 0.002$) and distant metastasis ($p = 0.005$). The expression of Tau protein was significantly positively correlated with grade, stage, lymph node, distant metastasis ($p < 0.001$) and local recurrence of the tumor ($p = 0.002$). High SALL4 expression was significantly positively correlated with response to treatment, performance status worse 3-year overall survival (OS) and local recurrence free survival rate ($P = 0.018, 0.01, 0.008$ and < 0.001 respectively). High Tau protein expression were significantly positively correlated with response to treatment, performance status, worse 3-year overall survival (OS) rate, local recurrence free survival rate ($p < 0.001$) and resistance to the paclitaxel/platinum first-line chemotherapy ($p = 0.021$).

Conclusion: SALL4 and Tau protein may be considered as poor prognostic and predictive markers in serous ovarian carcinoma.

Introduction:

Epithelial ovarian cancer (EOC) is the 4th commonest malignancy in female [1], and it is one of the most common causes of postmenopausal women cancer mortality worldwide, serous ovarian carcinoma (SOC) forms high percentage of its types [1]. Despite the advancement in surgical intervention for management of SOC, its overall survival and progression-free survival rates are still poor as it is discovered at advanced stage especially in developing countries [3]. The primary goal of surgery is debulking due to the diffuse nature of SOC and the invasion of adjacent organs or peritoneal cavity via peritoneal fluid [4] followed by chemotherapy and molecular targeted therapies that play an important role in the postoperative treatment of SOC. The currently used chemotherapeutic agents are platinum plus taxane therapy [2]; they appeared with more resistance and less efficacy dealing with cancer patients. So a novel therapeutic agent that decrease the resistance to the current chemotherapeutic agents and that also specifically target the serous ovarian cancer is the main target for clinical cancer research [5-6]. Spalt-like transcription factor (SALL4) is a zinc finger transcriptional factor that regulates multiple downstream targeted genes involved in normal development, important in the control of pluripotency and self-renewal of embryonic stem cells (escs) [7], also recent research demonstrated that SALL4 is plays an important role in hematopoiesis and leukemogenesis, the abnormal expression of SALL4 was found in acute myeloid leukemia [8], and it functions as an oncogene in many cancers [9]. Tau protein (50–64 kd), a product of gene located in chromosome 17 (17q21) shows the ability of combining to beta-tubulin. It may bind to the exterior and to the interior microtubules surface [10]. Tau was first isolated from brain and its function has been associated with neurodegenerative diseases [11]. There is a real need to adequately understand the mechanisms of SOC invasion, progression and development of more effective therapeutic molecular targets and prognostic biomarkers. The predictive or prognostic value of Tau protein and SALL4 in SOC is still points under research, and no previous studies had explored the relation between immunohistochemical expression of SALL4 and its relation to chemotherapy resistance in SOC patients or detect the relation between both SALL4 and Tau protein immunoexpression.

To explore and assess the predictive and prognostic role of SALL4 and Tau protein immunohistochemical expressions in serous ovarian carcinoma patients treated with paclitaxel/platinum first-line chemotherapy.

Patients and Methods:-

This retrospective study was carried out at departments of Medical Oncology, Pathology, Clinical Oncology and nuclear medicine, and Gynecology and Obstetrics, Faculty of Medicine, Zagazig University in the period from March 2013 to March 2016.

We included 50 histologically confirmed serous ovarian cancer (SOC) International Federation of Gynaecology and Obstetrics (FIGO) stage IC-IV. All patients had history of debulking surgery followed by first-line

Chemotherapy regimen: paclitaxel (175 mg/m²) with carboplatin (AUC 6), administered every 3 weeks for 6 cycles. We used the tumor-node-metastasis (TNM) and International Federation of Gynecology and Obstetrics (FIGO) classifications for staging of SOC [12] and the WHO grading system, for pathologic grading [13]. We identified sex, age, tumor size, grade, stage, lymph node, distant metastasis, response to first-line chemotherapy according to RECIST criteria of the patients by retrospective examination of the patients' records at the involved departments. Local Research Ethics Committee approval of the study was obtained.

Immunohistochemical staining:-

Immunohistochemical staining was carried out using streptavidin-peroxidase system (Zhongshan Goldenbridge Biotechnology, Beijing, China) 5µm thick sections cut from paraffin blocks, put on positively charged slides, then de-paraffinized in xylene and rehydrated in rising grades of ethyl alcohol. Antigen retrieval was done by boiling sections in citrate buffer (ph6.0) for 20 min and then after washing with phosphate buffered saline (PBS), the slides were incubated with rabbit monoclonal anti-Tau antibody [E178] (ab32057) (Abcam, Cambridge, MA, USA) was used at a dilution of 1:500 and Mouse monoclonal anti-Sall4 antibody (ab57577) (Abcam, Cambridge, MA, USA) diluted 1/100 at 4°C overnight. The sections were then washed in PBS and incubated with Poly-peroxidase-anti-mouse/rabbit igg (Zymed Laboratories, San Francisco, CA, USA) for 20 min. 3; 30-Diaminobenzidine was used as the chromogen. At the end, the sections were counterstained with hematoxylin. Normal ovarian epithelium taken from areas near benign ovarian cyst was used as an external positive control for Tau protein [14], human yolk sac

tumor tissue as a positive control for SALL4 [15] and the negative controls by replacing the primary antibodies by the non-immune serum.

Evaluation of immunohistochemical expression of Tau protein:-

Specimens were assessed as follows: IHC score 0 – no staining; 1+ – poor focal staining or very poor diffuse staining (less intense than normal ovarian epithelium); 2+ average diffuse staining (similar to normal ovarian epithelium) or strong staining (more intense than normal ovarian epithelium) in less than 25% cells; 3+ strong staining in 25% of tumors cells or more. Tau expression was interpreted as negative (0 and 1+) or positive (2+ and 3+). The final staining results were determined by using staining intensity of normal epithelial cells as a reference. We consider results as; low (0–1, 5) and high (2– 3) [10].

Evaluation of immunohistochemical expression of SALL4:-

Only nuclear staining was considered positive for SALL4. SALL4 expression was scored according to the percentage of tumor cells stained positive for SALL4, with 0 denoting <5% of tumor cells stained positive, 1, 5-30% of tumor cells stained positive, 2, 31-50% of tumor cells stained positive, 3, 51-80% of tumor cells stained positive, and 4, >80% of tumor cells stained positive. The low expression was defined as 0-1; the high expression was defined as 2-4 [16].

Statistical analysis:-

All statistics were performed using SPSS 22.0 for windows (SPSS Inc., Chicago, IL, USA) & medcalc windows (medcalc Software bvba 13, Ostend, Belgium). Continuous variables were expressed as the mean \pm SD & median (range), and the categorical variables were expressed as a number (percentage). Percent of categorical variables were compared using the Pearson's Chi-square test or Fisher's exact test when appropriate. Strength of relationship between Tau protein and SALL4 & clinicopathological features were determined by computing Spearman's correlation coefficient.

Stratification of OS and RFS were done according to all clinicopathological features and immunohistochemical markers. A p value <0.05 was considered statistically significant.

Results:-

Patients:-

Fifty patients of serous carcinoma of the ovary were included in our study with age ranged from (25-75) years (Mean: 55.48 ± 10.97 years), 18 cases were of low grade and 32 cases were of high grade type.

SALL4 and Tau expressions in ovarian cancer:-

Among 50 patients included in the analysis, 58 % (n=29 patients) had high Tau immunoexpression 42% (n=21) had low Tau immunoexpression. Also, 68 % (n=34) and 32% (n=16) of the patients had high andsall4 immunoexpression, respectively.

Correlation between SALL4 and Tau expressions with clinicopath-ological variables of the 50 SOC patients:-

The expression of SALL4 in serous ovarian carcinoma was significantly positively correlated with grade, stage, lymph node metastasis ($p < 0.001$ for all), ECOG performance status ($p = 0.01$), local recurrence of the tumor ($p = 0.002$) and distant metastasis ($p = 0.005$; Table 1; Figures 1, 2).

As well as, the expression of Tau protein was significantly positively correlated with grade, stage, lymph node, distant metastasis, ECOG performance status ($p < 0.001$ for all), and local recurrence of the tumor ($p = 0.002$; Table 1; Figs 3, 4).

Association between Tau expression and response to chemotherapy in patients with measurable disease:-

Among 35 patients with CR, 11 (100%; 21/ 21) were assessed as low Tau immunoexpression and (48.3%; 14/29; $p = .001$) as high Tau immunoexpression. Proportion of objective response (OR) was higher in low Tau immunoexpression group (100%) compared with high Tau immunoexpression group (65.5%), with statistical significance ($p = 0.003$; Table 2).

The same was for SALL4, Among 35 patients with CR (100%; 16/16) were assessed as low SALL4 immunoexpression and (55.9%; 19/34; 0.01) as high SALL4-expression . Proportion of objective response (OR)

was higher in low SALL4 immunoeexpression group (100%) compared with high SALL4 immunoeexpression (70.6%), with statistical significance ($p=0.02$; Table 2).

Combined High Tau protein and SALL4 immunoeexpression were significantly positively correlated with lower response to treatment ($p=0.02$; Table 2).

Table 1:- Correlation between SALL4 and Tau protein expressions with clinicopathological variables of the 50 serous ovarian carcinoma patients.

Variables	All (N=50)		SALL4				P	Tau protein				P
			Low (N=16)		High (N=34)			Low (N=21)		High (N=29)		
	N	(%)	N	(%)	N	(%)		N	(%)	N	(%)	
FIGO stage												
Stage IC	7	(14)	7	(44)	0	(0)	<0.001	7	(33)	0	(0)	<0.001
Stage II	9	(18)	4	(25)	5	(15)		6	(28.5)	3	(10.3)	
Stage III	20	(44)	5	(31.2)	17	(50)		8	(38)	14	(48.2)	
Stage IV	12	(24)	0	(0)	12	(35)		0	(0)	12	(41.4)	
Lymph node involvement												
Absent	16	(32)	11	(68.8)	5	(14.7)	<0.001	13	(61.9)	3	(10.3)	<0.001
Present	34	(68)	5	(31.3)	29	(85.3)		8	(38.1)	26	(89.7)	
Distant Metastasis												
Absent	38	(76)	16	(100)	22	(64.7)	0.005	21	(100)	17	(58.6)	0.001
Present	12	(24)	0	(0)	12	(35.3)		0	(0)	12	(41.4)	
Performance status												
ECOG<2	39	(78)	16	(100)	23	(67.6)	0.010	21	(100)	18	(62.1)	0.001
ECOG≥2	0	(0)	0	(0)	11	(32.4)		0	(0)	11	(37.9)	
Recurrence												
No	10	(20)	9	(56)	1	(2.9)	0.002	10	(47.6)	0	(0)	0.002
Yes	25	(50)	7	(74)	18	(53)		11	(52.4)	14	(48.3)	
Survival												
Alive	23	(46)	12	(75)	11	(32)	0.005	19	(91)	4	(14)	<0.001
Died	27	(54)	4	(25)	23	(68)		2	(9)	25	(86)	

Survival data:

After a median follow up of 30 months (range: 10-36), the median OS and RFS were 12(10-36) and 11(9-36) months, respectively.

Univariate analysis revealed following clinical parameters were correlated with RFS: FIGO stage at diagnosis ($p<0.001$), age ($p=0.022$), type of surgery ($p=0.004$), grade ($p<0.001$), LN involvement ($p<0.001$), Tau expression level ($p= p<0.001$) and SALL4 expression level ($p=0.015$).

Overall survival (OS) associations were identified in univariate analysis and presented in Table 4. Statistical significance was achieved in following factors: FIGO stage at diagnosis ($p<0.001$), age ($p=< 0.001$), tumor grade ($p< 0.001$) tau expression status ($p< 0.001$) and SALL4 expression level ($p=0.008$). The results are presented in tables 3&4 and figure 5.

In multivariate analysis, the Tau protein and SALL4 immunoeexpressions were statistically independent parameter associated with OS (HR-3.4, 8.4, and $P=0.001$, 0.01, respectively).

Table 2:- Correlation between SALL4&Tau protein immunohistochemical expressions and response to treatment in patients with serous ovarian carcinoma

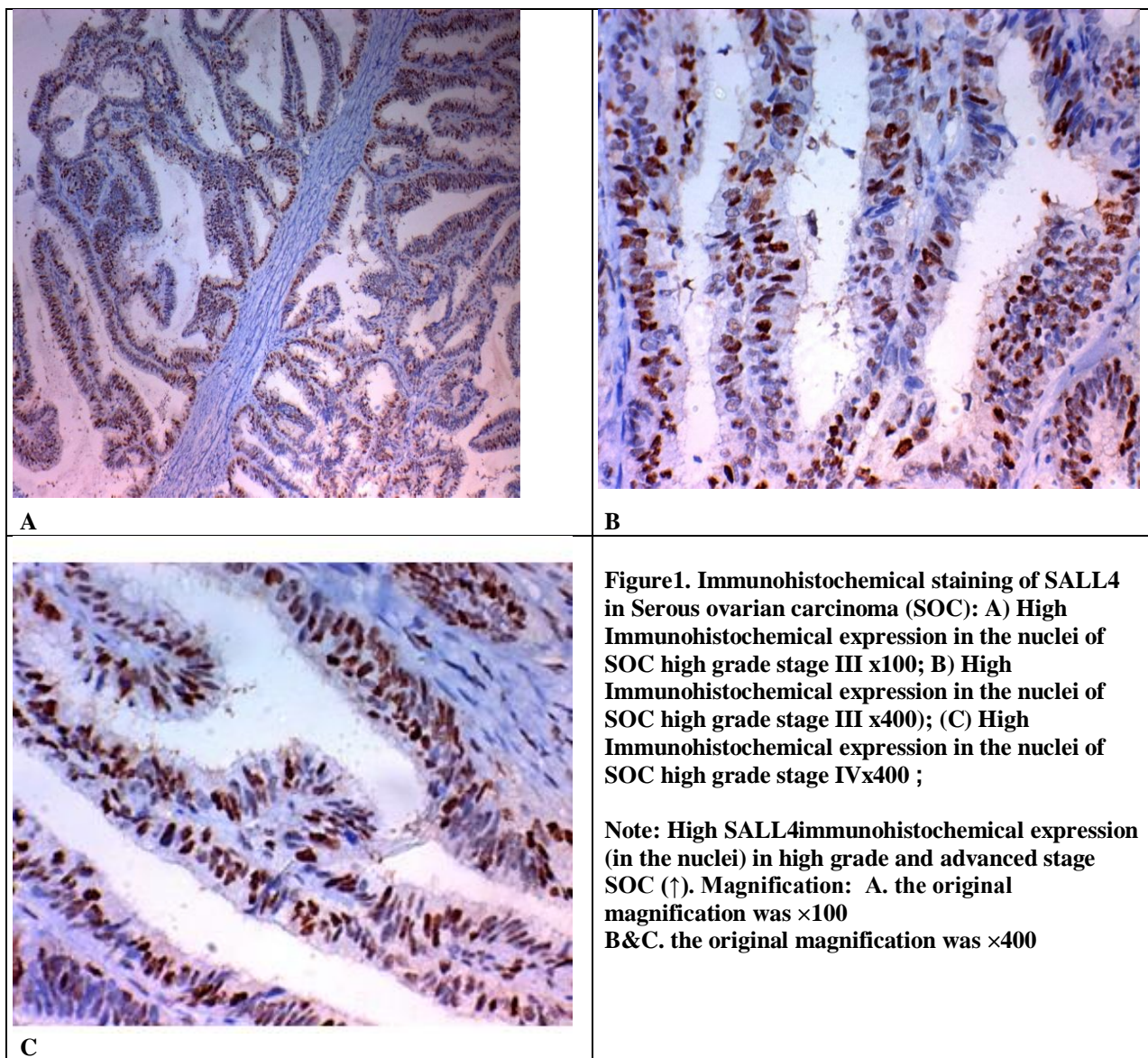
Characteristics	Response to treatment										P
	All (N=50)		PD (N=3)		SD (N=7)		PR (N=5)		CR (N=35)		
	N	(%)	N	(%)	N	(%)	N	(%)	N	(%)	
Surgery											
Suboptimal	16	(32)	2	(12.5)	5	(31.3)	5	(31.3)	4	(25)	<0.001
Optimal debulking	22	(44)	1	(4.5)	2	(9.1)	0	(0)	19	(86.4)	
Radical surgery	12	(24)	0	(0)	0	(0)	0	(0)	12	(100)	
SALL4											
Low	16	(32)	0	(0)	0	(0)	0	(0)	16	(100)	0.01
High	34	(68)	3	(8.8)	7	(20.6)	5	(14.7)	19	(55.9)	
Tau											
Low	21	(42)	0	(0)	0	(0)	0	(0)	21	(100)	0.001
High	29	(58)	3	(10.3)	7	(24.1)	5	(17.2)	14	(48.3)	
SALL4/Tau											
Low/Low	15	(30)	1	(6.7)	2	(13.3)	0	(0)	12	(80)	0.02
Low/High	7	(14)	0	(0)	0	(0)	0	(0)	7	(100)	
High/Low	12	(24)	0	(0)	0	(0)	0	(0)	12	(100)	
High/High	16	(32)	2	(12.5)	5	(31.3)	5	(31.3)	4	(25)	

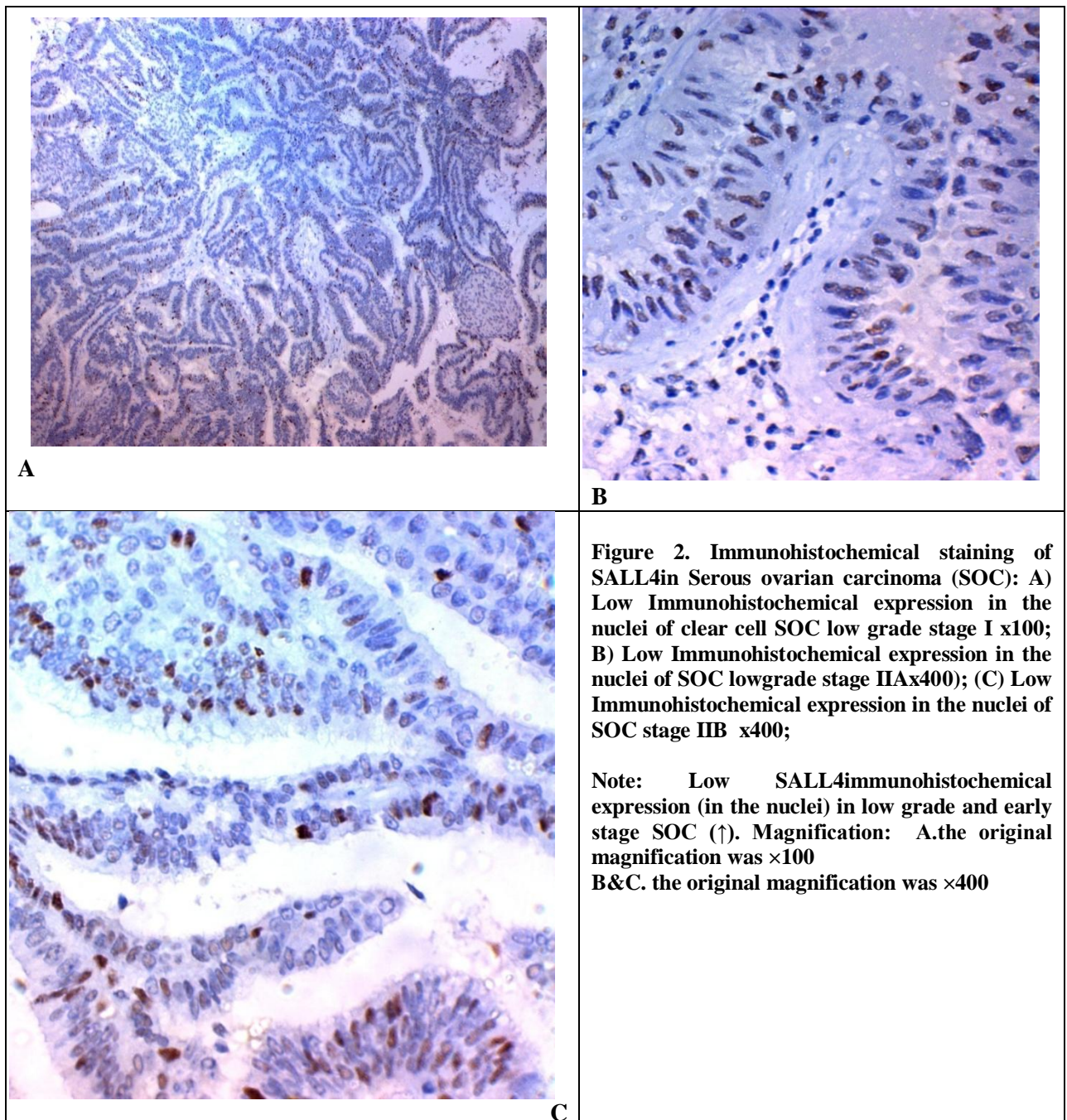
Table 3: Correlation between recurrence free survival in the 35 patients responding and clinicopathological features.

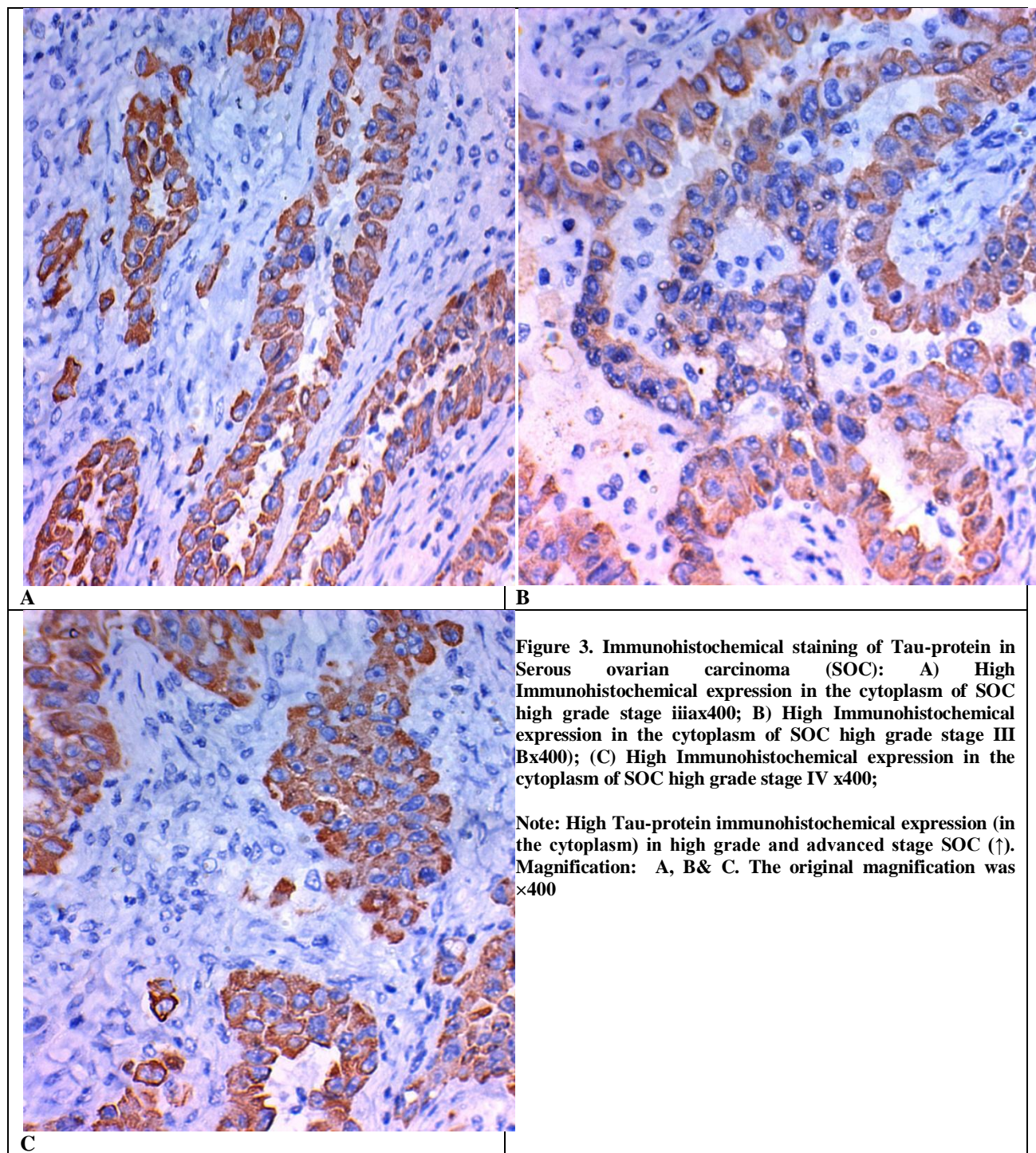
Characteristics	Recurrence						P	Recurrence Free Survival				P
	All (N=35)		No (N=10)		Yes (N=25)			Median (month)	1 ys (%)	2 ys (%)	3 yrs (%)	
	N	(%)	N	(%)	N	(%)						
All patients	35	(100)	10	(28.6)	25	(71.4)		11	48.6%	33.4%	24.3%	
Age (years)												
Mean ± SD	50.74	±8.95	41	±9.39	54.64	±5.02	<0.001					
Median (Range)	55	(25-60)	45	(25-55)	56	(45-60)						
≤40 years	4	(11.4)	4	(100)	0	(0)	0.002	NR	100%	100%	100%	0.022
41-49 years	26	(74.3)	6	(23.1)	20	(76.9)		20	50%	29.2%	10.9%	
> 60 years	5	(14.3)	0	(0)	5	(100)		11	0%	0%	0%	
Surgery												
Suboptimal	4	(11.4)	1	(25)	3	(75)	0.006	21.5	50%	50%	0%	0.004
Optimal debulking	19	(54.3)	1	(5.3)	18	(94.7)		11	31.6%	5.2%	5.2%	
Radical surgery	12	(34.3)	8	(66.7)	4	(33.3)		NR	75%	75%	62.5%	
FIGO stage												
Stage IC	7	(20)	7	(100)	0	(0)	<0.001	NR	100%	100%	100	<0.001
Stage II	9	(26)	2	(25)	7	(75)		21	100%	25%	25%	
Stage III	19	(54)	1	(0)	18	(100)		11	28.6%	0%	0%	
Grade												
Low grade	18	(51.4)	9	(50)	9	(50)	0.007	35	83.3%	55.6%	46.3%	<0.001
High grade	17	(48.6)	1	(5.9)	16	(94.1)		11	11.8%	11.8%	0%	
Lymph node involvement												
Absent	16	(45.7)	9	(56.3)	7	(43.8)	0.002	NR	81.2%	62.5%	52.1%	<0.001
Present	19	(54.3)	1	(5.3)	18	(94.7)		11	21.1%	7%	0%	
SALL4												
Low	16	(45.7)	9	(56.3)	7	(43.8)	0.002	NR	62.5%	62.5%	52.1%	<0.001
High	19	(54.3)	1	(5.3)	18	(94.7)		11	36.8%	6.1%	0%	
Tau												
Low	21	(60)	10	(47.6)	11	(52.4)	0.002	32	76.2%	50.8%	43.5%	<0.001
High	14	(40)	0	(0)	14	(100)		11	7.1%	7.1%	0%	
SALL4/Tau												
Low/Low	12	(34.3)	9	(75)	9	(75)	<0.001	NR	75%	75%	75%	0.015
Low/High	7	(20)	0	(0)	0	(0)		11	14.3%	14.3%	0%	
High/Low	12	(34.3)	1	(8.3)	1	(8.3)		20	58.3%	9.7%	0%	
High/High	4	(11.4)	0	(0)	0	(0)		11	0%	0%	0%	

Table 4: Correlation between overall survival of the 50 patients and clinicopathological features.

Characteristics	Survival						P	Overall Survival					P
	All (N=50)		Alive (N=23)		Died (N=27)			Median (month)	1 Yr (%)	2 ys (%)	3 Ys (%)		
	N	(%)	N	(%)	N	(%)							
All patients	50	(100)	23	(46)	27	(54)		12	49.3%	44.8%	44.8%		
Age (years)													
Mean ± SD	55.48	±10.97	46.73	±8.60	62.92	±6.27	<0.001						
Median (Range)	57	(25-75)	47	(25-56)	60	(56-75)							
≤40 years	4	(8)	4	(100)	0	(0)	<0.001	NR	100%	100%	100%	<0.001	
41-49 years	29	(58)	19	(65)	10	(35)		NR	72.4%	64.4%	64.4%		
> 60 years	17	(34)	0	(0)	17	(100)		11	0%	0%	0%		
Surgery													
Suboptimal	16	(32)	4	(25)	12	(75)	<0.001	11	21.9%	21.9%	21.9%	0.005	
Optimal debulking	22	(44)	7	(32)	15	(68)		11	40.9%	31.8%	31.8%		
Radical surgery	12	(24)	12	(100)	0	(0)		NR	100%	100%	100%		
FIGO stage													
Stage IC	7	(14)	7	(100)	0	(0)	<0.001	NR	100%	100%	100%	<0.001	
Stage II	9	(18)	9	(100)	0	(0)		NR	100%	100%	100%		
Stage III	22	(44)	7	(32)	15	(68)		NR	100%	100%	100%		
Stage IV	12	(24)	0	(0)	12	(100)		11	0%	0%	0%		
Grade													
Low grade	18	(36)	18	(100)	0	(0)	<0.001	NR	100%	100%	100%	<0.001†	
High grade	32	(64)	5	(15.6)	27	(84.4)		11	20.8%	13.9%	13.9%		
Lymph node involvement													
Absent	16	(32)	16	(100)	0	(0)	<0.001	NR	100%	100%	100%	<0.001	
Present	34	(68)	7	(20.6)	27	(79.4)		11	25.7%	19.3%	19.3%		
Distant metastasis													
Absent	38	(76)	23	(60.5)	15	(39.5)	<0.001	NR	65.8%	59.8%	59.8%	<0.001	
Present	12	(24)	0	(0)	12	(100)		11	0%	0%	0%		
SALL4													
Low	16	(32)	12	(75)	4	(25)	0.005	NR	75%	75%	75%	0.008	
High	34	(68)	11	(32.4)	23	(67.6)		11	37.6%	31.4%	31.4%		
Tau													
Low	21	(42)	19	(90.5)	2	(9.5)	<0.001‡	NR	90.5%	90.5%	90.5%	<0.001	
High	29	(58)	4	(13.8)	25	(86.2)		11	20.7%	13.8%	13.8%		
SALL4/Tau													
Low/Low	15	(30)	10	(66.7)	5	(33.3)	0.002§	NR	100%	64%	64%	0.002	
Low/High	7	(14)	3	(42.9)	4	(57.1)		11	42.9%	42.9%	42.9%		
High/Low	12	(24)	10	(83.3)	2	(16.7)		NR	83.3%	83.3%	83.3%		
High/High	16	(32)	0	(0)	16	(100)		11	0%	0%	0%		







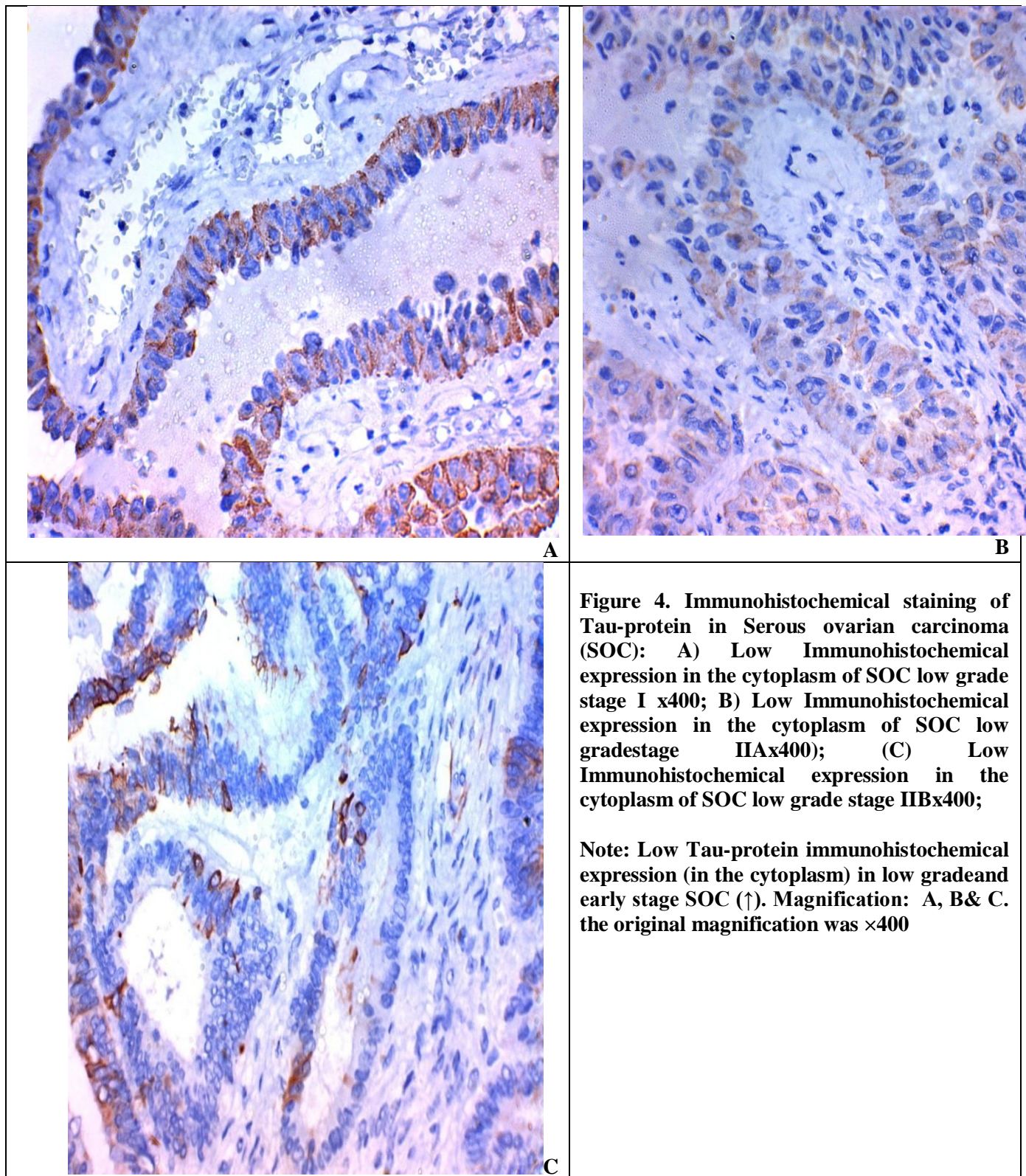
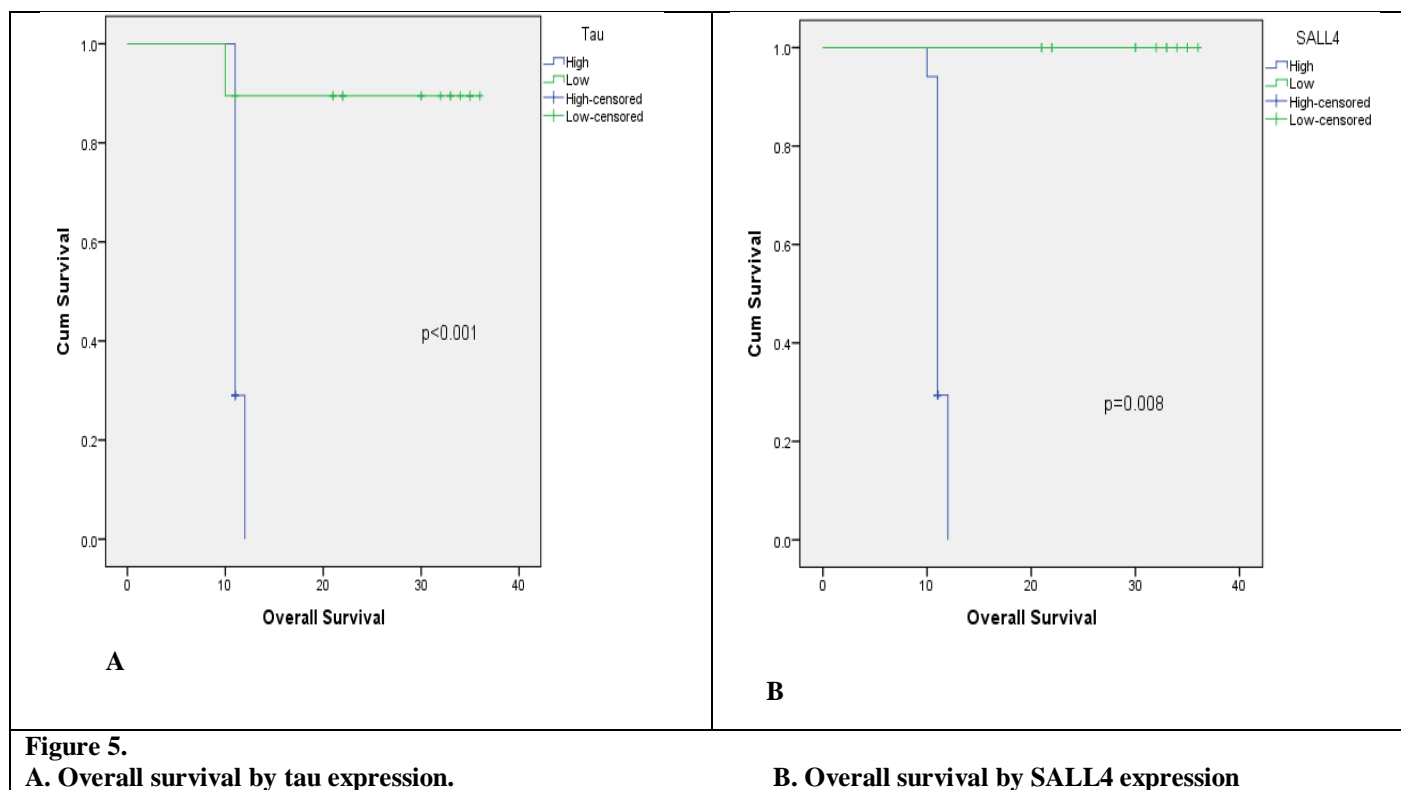


Figure 4. Immunohistochemical staining of Tau-protein in Serous ovarian carcinoma (SOC): A) Low Immunohistochemical expression in the cytoplasm of SOC low grade stage I x400; B) Low Immunohistochemical expression in the cytoplasm of SOC low grade stage IIAx400; (C) Low Immunohistochemical expression in the cytoplasm of SOC low grade stage IIBx400;

Note: Low Tau-protein immunohistochemical expression (in the cytoplasm) in low grade and early stage SOC (↑). Magnification: A, B & C. the original magnification was ×400



Discussion:

Ovarian cancer is the leading cause of morbidity among malignant gynecologic diseases. The most frequent subtype among ovarian cancer is SOC [17]. It can directly broadly invade adjacent organs or peritoneal cavity via peritoneal fluid. Because of the diffuse nature of SOC, the objective of surgery is often cytoreductive, but not radical [18].

However, disease that initially responds well to this treatment frequently relapses, indicating that the existing therapeutic molecular targets and prognosis markers are not sensitive and efficacious enough. Therefore, it is necessary to make further research on new therapies and treatment targets of SOC. In this study, we investigated the prognostic role of SALL4 and Tau protein in SOC.

We found that SALL4 immunohistochemical expression was significantly positively correlated with grade, stage, lymph node metastasis ($p < 0.001$), local recurrence of the tumor ($p = 0.002$) and distant metastasis ($p = 0.005$). Our follow up data showed that high expression of SALL4 expression was significantly positively correlated with response to treatment, performance status worse 3-year overall survival (OS) and local recurrence free survival rate ($P = 0.018, 0.01, 0.008$ and < 0.001 respectively).

Yang et al. [19], proved the same results in SOC that were near that of the previous studies on the function of SALL4 in solid tumors [20-23].

Our results can be explained by, **Zeng et al. [24]** that explained how SALL4 regulate stemness in embryonic and hematopoietic stem cells and activated in a hepatocellular carcinoma subtype with stem cell features, deletion of SALL4 resulted in the down-regulation of these stem cell markers, together with attenuation of the invasion capacity, also the function of SALL4 in promoting lymph node metastasis and advanced stage might be due to the interaction with b-catenin and subsequent aberrant activation of Wnt/b-catenin signaling pathway [**15, 25, 26**].

Also recent reports showed that SALL4 involved in HOXA9, c-Myc and PTEN-AKT pathway, these genes correlate with epithelial-mesenchymal transition (EMT) [**21, 28**]. EMT plays an essential role in increasing SOC spread [**28-29**].

The aim of adjuvant chemotherapy is prolongation of OS rate that effect is possible to achieve if the cancer is chemo-sensitive. Thus, chemosensitivity is a good prognostic factor and drug resistance is a significant obstacle that affects the overall survival rate for advanced serous ovarian cancer patients.

We tried to find relation between the immunohistochemical expression of our markers and response to chemotherapeutic agents we found that SALL4 can participate in mediating cellular resistance to chemotherapeutic drugs and that low immunohistochemical expression of SALL4 in SOC is associated with better efficacy of chemotherapy ($p=0.626$).

There are no many previous studies studied the relation between SALL4 and drug resistance in SOC, but our results are the same like that of Liu et al., [30] who proved that SALL4 expression is positively correlated with increased resistant to chemotherapeutic agents in endometrial carcinoma, and results of other studies that detected SALL4-induced chemotherapeutic resistance in a variety of cancers [31-32] and our results can be explained by that the majority of drug resistance in human cancer are associated with ATP-binding cassette (ABC) multidrug transporters. SALL4 induces chemotherapeutic drug resistance through the regulation of ABCB1 in endometrial cancer cells. ABC transporters were thought to be closely related to drug resistance ABCB1 is the prototype of this gene superfamily, and its deregulation has been associated with drug resistance in several types of cancers [30].

C-Myc plays an important roles in many oncogenic processes, including tumor drug resistance and metastasis. SALL4 in endometrial cancer cells promoted c-Myc transcriptional activity [27].

In our study Tau protein-immunoexpression in SOC was significantly positively correlated with grade, stage, lymph node, distant metastasis ($p<0.001$) and local recurrence of the tumor ($p=0.002$) and we analyzed the role of its expression as a predictive factor for paclitaxel-containing chemotherapy. High Tau protein expression were significantly positively correlated with response to treatment, performance status, worse 3-year OS rate ($p<0.001$) and resistance to the paclitaxel/ carboplatin first-line chemotherapy ($p=0.021$).

Smoter et al. [14] found similar results that the negative status of Tau protein in SOC is associated with better response to chemotherapy.

Our results were explained by understanding paclitaxel's action which is competitive to Tau protein. Paclitaxel binds beta-tubulin on microtubule's inner surface, in the same point as Tau protein [33], and the presence of Tau protein on the microtubules 'surface creates difficulties in paclitaxel attachment to these structures. Low Tau expression may result in better paclitaxel connection with microtubules and more effective chemotherapy action, expressed in higher objective responses rate and better PFS [13].

Low Tau protein expression was associated with statistically significant more frequent achievement of complete response (CR) to paclitaxel in breast cancer and inhibition of Tau protein may enhance paclitaxel activity [**10**].

Tau protein -negative patients with breast cancer were sensitive to paclitaxel therapy, compared with Tau protein positive patients [**34**].

The predictive value of low Tau expression for paclitaxel therapy was confirmed in gastric cancer [**35**].

Paclitaxel has a more obvious function to cells with low expression of Tau protein; the apoptosis rate is also higher. Paclitaxel may be easily combined with tubulin under low concentrations of Tau protein. The high concentration has

a stabilizing effect to microtubules, reducing the harmful effects of paclitaxel, which leads to drug resistance. However, resistance mechanisms to paclitaxel and their associated prognostic value needs further study [36].

Abd elaziz et al. [37] proved results that are similar to ours. **Steffensen et al.[38]**, results were different from ours they reported that Tau protein was not associated with OS rate or progression free survival.

Our results differ from those obtained in the studies on breast cancer, where co-expression of Tau protein and estrogen receptor was considered as good prognostic factor [39]. This divergence might be caused by Tau significance evaluation in different cancer sites.

Our study was the first to correlate the immunohistochemical expression of SALL4 and Tau protein immunohistochemical expression, prognosis and response to chemotherapy and we found a positive correlation between the expression of both markers, poor prognosis and resistant to chemotherapy (Spearman's $r = +0.198$; $p = 0.161$).

Therefore the targeted therapy against these markers can decrease the resistance to the currently used chemotherapeutic agents and anti SALL4 could be a novel therapeutic drug that specifically target SOC cells decreasing its spread and improving its prognosis.

Conclusion:-

SALL4 and Tau protein may be useful prognostic and predictive markers in ovarian cancer patients receiving chemotherapy. More prospective studies with larger group of patients, may confirm their predictive and prognostic value.

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