Original Article

IMP3 Expression in Ovarian Serous Tumours: Correlation with Epithelial-Mesenchymal Transition-Related Markers (E-Cadherin and Vimentin) and Clinicopathological Factors

SAMAH SAID ELBASATEENY[1], WALID MOHAMED ELNAGAR[2], MAHMOUD ABDOU YASSIN[3], HEBA MOHAMMED RASHAD[4]

ABSTRACT

Introduction: Ovarian epithelial cancer is an aggressive malignancy of which Ovarian Serous Carcinoma (OSC) represents a common type. The insulin-like growth factor mRNA-binding protein 3 (IMP3) is one of mRNA-binding protein family that is overexpressed in a variety of human cancers, including ovarian carcinomas. Being associated with adverse prognostic outcome in these cancers, which makes this protein a reliable biomarker to distinguish some cancers from their benign mimetic lesions.

Aim: To analyse IMP3 expression in serous ovarian tumours and correlates the expression in malignant tumours with Epithelio-Mesenchymal Transition (EMT) related markers (Epithelial(E)-cadherin and vimentin) and clinicopathological factors to assess the clinical application of IMP3 in the diagnosis, prognosis and possible application in targeted therapy of such tumours.

Materials and Methods: This was a cross-sectional research that was done at the Departments of Pathology, Obstetric and Surgery, Zagazig and Banha Universities Hospitals, Egypt. Fifty-nine formalin-fixed paraffin-embedded specimens (including 43 cases OSC, 6 cases border line serous tumour and 10 cases benign serous cystadenoma) were collected from January 2014 to December 2018. Each case was stained immunohistochemically with IMP3, E-cadherin and Vimentin.

Markers expressions were statistically analysed using SPSS software (version 19.0; SPSS, Chicago, IL). The p-values ≤0.05 was regarded statistically significant.

Results: IMP3 was detected in 76.7% (33/43) of the studied OSC. IMP3 was in a significant association with advanced FIGO (International Federation of Gynaecology and Obstetrics) stage and with lymph node metastases (p=0.02 and 0.019, respectively). E-cadherin was predominantly lost in 53.5% (23/43) of the studied OSCs and was significantly associated with advanced FIGO stage (p=0.004). Vimentin was detected in 79.1% (34/43) of the studied OSCs and was in association with high grades and advanced stage (p=0.018 and 0.007, respectively). An inverse significant correlation was detected between IMP3 and E-cadherin expressions (Spearman correlation (r)=-0.480, p-value=0.001), while positive significant correlation was detected between IMP3 and Vimentin expressions (Spearman correlation (r)=0.393, p-value=0.009).

Conclusion: IMP3 is an oncogene unregulated in OSC. It is a prognostic marker associated with tumour aggressiveness. Moreover, IMP3 could promote tumour invasion and metastasis via EMT in OSCs.

INTRODUCTION

Ovarian epithelial cancer is an aggressive malignancy which has a highly invasive and metastatic feature with the reported 5-year survival only 37% [1].

OSC is a common ovarian epithelial malignancy, which now graded as low-grade and high-grade [2]. The evolution of aggressive carcinoma, occurs in a slowly step-wise manner. Low-grade neoplasm are slowly growing and have better prognosis than high-grade neoplasm [3].

The insulin-like growth factor mRNA-binding protein 3 (IMP3) is one of mRNA-binding protein family which regulates mRNA transport, translation and turnover [4]. Previous studies have concluded that IMP3 is an onco-fetal protein that facilitates the invasive abilities of tumour cells [5]. IMP3 was found to be overexpressed in a variety of human cancers, including ovarian, lung and gastrointestinal carcinomas [6,7]. Being associated with advanced tumour stage and adverse prognostic outcome in these cancers and which makes this protein a reliable biomarker to distinguish some cancers from their benign mimetic lesions [8].

Epithelial to Mesenchymal Transition (EMT) is the process by which

epithelial cancer cells lose cell-cell adhesion, apical-basal polarity and acquire spindle cell morphology and gain mesenchymal markers positivity, which help them to metastasise [9]. Hypoxia [10], inflammation [11] and oxidation stress [12], are potent EMT inducers. These factors trigger signals that converge on EMT-inducing transcriptional factors such as Snail, Slug, Twist, with diminished expression of epithelial-related genes such as E-cadherin and, increased expression of mesenchymal-related genes such as vimentin [13]. The objectives of this study were to analyse IMP3 expression in serous ovarian tumours of Egyptian women, and to correlate the expression in malignant tumours with EMT related markers (E-cadherin and vimentin) and clinicopathological factors to assess the clinical application of IMP3 in the diagnosis, prognosis and possible application in targeted therapy of such tumours.

MATERIALS AND METHODS

Type of Study and Specimen Collection

This was a cross-sectional research that was done at the Departments of Pathology, Obstetric and Surgery, Zagazig and Banha Universities Hospitals, Egypt. The research included fifty-nine cases of ovarian serous tumours (43 cases OSC, 6 cases border line tumour and 10

cases benign cystadenoma) that were collected from the archives of Pathology Departments from January 2014 to December 2018. All procedures performed in the current study were ethically approved by Institutional Review Board (ZU -#IRB approval number 4943).

Inclusion criteria: Female patients with primary ovarian tumour debulking operation, the specimens included were diagnosed by two pathologist and met the requirement of the Ethical Committee.

Exclusion criteria: Cases that received chemotherapy before operation or cases with lost clinical data were excluded from the study.

In each case, tissue samples were fixed by formalin and paraffin-embedded blocks were performed. The clinicopathological data were collected from the patients' medical files.

Histopathological Evaluation

Four slides of 4 µm thickness were cut from each Paraffin block. The sections were dewaxed, then one slide was stained with haematoxylin and eosin and examined to confirm the previous diagnosis, asses the tumour grade according to the World Health Organisation classification (WHO) criteria 2016 [14]. Stage was defined according to the FIGO (The International Federation of Gynaecology and Obstetrics staging system) [15].

Immunohistochemical staining: For Immunohistochemical (IHC) assessment, 3 slides were mounted on positively charged slides. They were immune-stained for IMP 3 antibody, E-cadherin antibody and Vimentin antibody obtained from Thermo Fisher Scientific/Lab Vision Corporation, Rockford, and USA. IHC staining was performed, using universal detection kit (Thermo Scientific, USA) according to the manufacturer's data in which 3,3'-Diaminobenzidine (DAB) was utilised as a chromogen. A negative control was used for each marker, by omitting the primary antibody. Antibodies details shown in [Table/Fig-1].

Antibody	Type	State	Positive control	Incubation	Antigen retrieval
IMP3	Rabbit polyclonal	Concentrated (1:100)	Tonsil	1 hour	Citrate buffer
E-cadherin	Mouse monoclonal	Concentrated (1:100)	Skin	1 hour	EDTA
Vimentin	Mouse monoclonal	Concentrated (1:50)	Human bladder tissue	1 hour	EDTA

[Table/Fig-1]: The markers of the study.
EDTA: Ethylenediamine Tetra-acetic Acid; IMP3: Insulin-like growth factor-2 mRNA binding protein 3; E-cadherin: Epithelial cadherin

Immunohistochemical (IHC) Evaluation

IMP3 expression was detected as cytoplasmic dark brown staining of the cellular tumour. The proportion of positively stained cells was assessed by microscopic inspection of the whole stained tumour sections. The intensity of IMP3 staining was noted as weak, moderate, or strong. IMP3 expression was considered negative in absent staining or staining less than 5% of cellular tumour. The expression was considered positive in any definite cytoplasmic staining in more than 5% of cellular tumour, no matter the staining intensity [16].

Membranous E-cadherin and cytoplasmic vimentin expression was evaluated following the methods previously designated by [17] E-cadherin and vimentin was graded into groups from 0 to 3+ as follows: 0 (no staining); weak 1+ (1-25% staining); moderate +2 (26-50% staining); and strong +3 (>50% staining). For E-cadherin, the 2+ and 3+ groups were considered as positive. For vimentin, the 3+ groups were considered as positive [17].

STATISTICAL ANALYSIS

Analysis of the continuous variables was expressed as a means±standard deviation (SD). Analysis of categorical data was achieved using the Chi-square test (χ^2) or Fisher's-Exact test,

Spearman correlation was done to detect the correlation between IMP3 and EMT related markers. The statistical studies were performed with SPSS software (version 19.0; SPSS, Chicago, IL). The p-values ≤0.05 were regarded as statistically significant.

RESULTS

Clinicopathological Results

The mean age of the studied 59 ovarian serous tumours at initial surgery was 53.16±12.03 years (range 28-68 years). Among these 59 cases, 16.95% (10/59) of cases were benign cystadenomas, 10.17% (6/59) were borderline, and 72.88% (43/59) were OSC. Of the OSCs 53.49% (23/43) were low grade while 46.51% (20/43) were high grade. All carcinomas patients' clinicopathological variables are presented in [Table/Fig-2].

Result of Immunohistochemical (IHC) Expression of IMP3 among the Studied 59 Ovarian Serous Tumours

Brown cytoplasmic IMP3 immunoreactivity was detected in 76.7% (33/43) of the studied OSC. Among the ten negative carcinomas, nine carcinomas showed weak to moderate IMP3 staining but in less than 5% of tumour cells, only one was totally negative. 100% (6/6) of the studied borderline tumours were negative for IMP3 (5 cases were totally negative and only one case showed weak staining in less than 5% of tumour cells and also reported negative). Also, 100% (10/10) of the studied cystadenomas were totally negative [Table/Fig-3,4]. Analysis of IMP3 expression with the clinicopathological criteria of the studied 43 OSCs revealed significant expression with advanced FIGO stage (Stage III-IV) and with lymph node metastases (p=0.02 and 0.019, respectively). No, significant association was found between IMP3 expression and other clinicopathological features (p>0.05) [Table/Fig-2].

Result of Immunohistochemical (IHC) Expression of EMT Related Markers among the Studied 43 OSC

Membranous E-cadherin immunoreactivity was predominantly lost in 53.5% (23/43) of the studied OSCs and immunoreactive in the remaining 46.5% (20/43) cases. Loss of E-cadherin immunoreactivity was significantly detected in advanced FIGO (III/IV) stage (70.4%, 19/27) OSCs compared to early (I-II) stage (25.0%, 4/16) (p=0.004). Although loss of E-cadherin expression was detected more frequently in high grades (65.0%, 13/20) compared to low grades (43.5%, 10/23) OSCs, but the relationship did not reach significant level (p=0.158). No significant relation was detect between E-cadherin expression and other clinicopathological features (p>0.05). Cytoplasmic Vimentin immunoreactivity (detected in tumour cells, stromal cells, and vessels) was detected in 79.1% (34/43) of the studied OSCs (increased intensity of Vimentin staining was frequently detected at the invasive margin of positive tumours). Analysis of the result of Vimentin expression detected significant expression in high grades (95.0%, 19/20) and advanced stage (III-IV) (92.5%, 25/27) OSCs (p=0.018 and 0.007 respectively). No significant relation was detected between Vimentin expression and other clinicopathological features (p>0.05) [Table/Fig-2,5,6].

Result of Correlation Analysis between the Expression of IMP3 and the Expression of EMT Related Markers among the Studied 43 OSC
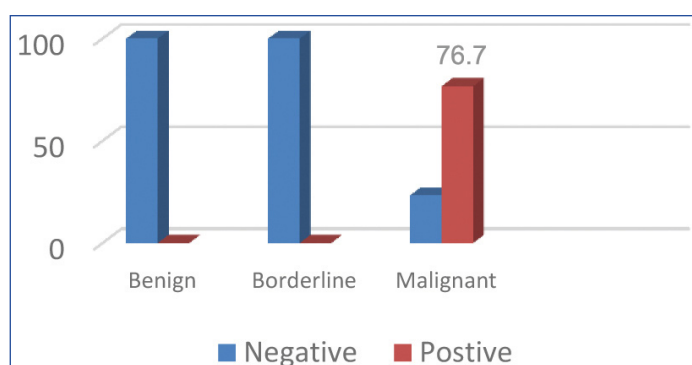
An inverse significant correlation was detected between IMP3 and E-cadherin expressions (Spearman correlation (r)=-0.480, p-value=0.001), while positive significant correlation was detected between IMP3 and Vimentin expressions (Spearman correlation (r)=0.393, p value=0.009) [Table/Fig-7].

Clinico-pathological variables	n (%) 43 (100%)	IMP3 expression		E-cadherin expression		Vimentin expression	
		Negative n (%)	Positive n (%)	Negative n (%)	Positive n (%)	Negative n (%)	Positive n (%)
Age							
≤50	15 (34.88%)	4 (26.7%)	11 (73.3%)	10 (66.7%)	5 (33.3%)	4 (26.7%)	11 (73.3%)
>50	28 (65.11%)	6 (21.4%)	22 (78.6%)	13 (46.4%)	15 (53.6%)	5 (17.9%)	23 (82.1%)
*p-value		0.487		0.172		0.380	
Size of the tumour							
≤10	25 (58.14%)	4 (16.0%)	21 (84.0%)	11 (44.0%)	14 (56.0%)	3 (12.0%)	22 (88.0%)
>10	18 (41.86%)	6 (33.3%)	12 (66.7%)	12 (66.7%)	6 (33.3%)	6 (33.3%)	12 (66.7%)
*p-value		0.168		0.122		0.094	
Histological grade							
Low grade	23 (53.49%)	8 (34.8%)	15 (65.2%)	10 (43.5%)	13 (56.5%)	8 (34.8%)	15 (65.2%)
High grade	20 (46.51%)	2 (10.0%)	18 (90.0%)	13 (65.0%)	7 (35.0%)	1 (5.0%)	19 (95.0%)
*p-value		0.07		0.158		**0.018**	
FIGO stage							
Stage I-II	16 (37.20%)	7 (43.7%)	9 (56.3%)	4 (25.0%)	12 (75.0%)	7 (43.8%)	9 (56.2%)
Stage III-IV	27 (62.97%)	3 (11.1%)	24 (88.8%)	19 (70.4%)	8 (29.6%)	2 (7.4%)	25 (92.5%)
*p-value		**0.02**		**0.004**		**0.007**	
Lymph node metastasis							
Absent	28 (65.12%)	10 (35.7%)	18 (64.3%)	14 (50.0%)	14 (50.0%)	5 (17.9%)	23 (82.1%)
Present	15 (34.88%)	0 (0.0%)	15 (100%)	9 (60.0%)	6 (40.0%)	4 (26.7%)	11 (73.3%)
*p-value		**0.019**		0.381		0.380	
Capsular rupture							
Absent	18 (41.86%)	5 (27.8%)	13 (72.2%)	8 (44.4%)	10 (55.6%)	6 (33.3%)	12 (66.7%)
Present	25 (58.14%)	5 (20.0%)	20 (80.0%)	15 (60.0%)	10 (40.0%)	3 (12.0%)	22 (88.0%)
*p-value		0.405		0.242		0.094	
Lymph/Vascular invasion							
Absent	17 (39.53%)	6 (35.3%)	11 (64.7%)	9 (52.9%)	8 (47.1%)	6 (35.3%)	11 (64.7%)
Present	26 (60.47%)	4 (15.4%)	22 (84.6%)	14 (53.8%)	12 (46.2%)	3 (11.5%)	23 (88.5%)
*p-value		0.127		0.599		0.069	

[Table/Fig-2]: Correlation of the clinicopathological parameters of the studied 43 OSC with IMP3 and EMT related markers expression.
*Chi-square test (χ^2) or Fisher's-exact test; p-value <0.05 to be considered as significant
IMP3: Insulin-like growth factor-2 mRNA binding protein 3; E-cadherin: Epithelial cadherin; FIGO: International federation of gynaecology and obstetrics
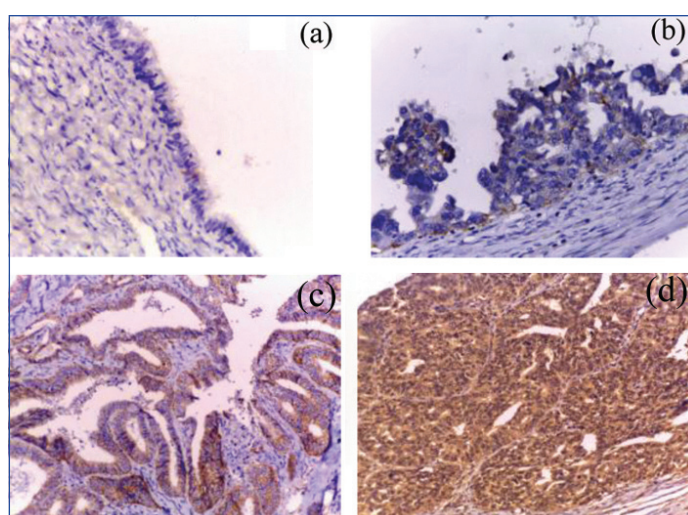

[Table/Fig-3]: IMP3 Immunoexpression in the studied 59 Ovarian Serous Tumours.

[Table/Fig-4]: Representative samples of IMP3 immunoreactivity in ovarian serous tumours: a) Benign serous cystadenoma showing negative staining for IMP3 (x400); b) border line serous tumour showing focal weak staining for IMP3 (x 200); c) low grade serous carcinoma showing weak to moderate diffuse IMP3 staining (x 200); d) high grade serous carcinoma showing strong diffuse IMP3 staining (x 200).

DISCUSSION

Overexpression of IMP3 has been described in several gynaecological and non-gynaecological malignancy [6,7,16]. It is regarded as an essential malignancy specific protein that is linked with aggressive cancers by helping cellular proliferation, invasiveness, and metastasis. [8,16]. Also, EMT have a role in invasiveness and metastatic potential of malignant neoplasms [9,17].

In this study, Authors investigated IMP3 expression in a group of ovarian serous tumours and correlated the expression in malignant tumours with EMT related markers and clinicopathological factors to assess the clinical application of IMP3 in the diagnosis, prognosis and possible application in targeted therapy of such tumours.

We detected IMP3 expression in 76.7% of the studied OSC, while 100% of the studied borderline and benign serous tumours showed

negative expression. This is similar to finding obtained by previous related study on serous ovarian tumours [16] and primary mucinous ovarian carcinoma [18]. Findings of the present study that IMP3 is expressed in OSC but not in benign and border line tumours, support the role of IMP3 in ovarian serous carcinogenesis, this is also supported by previous related studies on ovarian [16,18,19], on endometrial [20], on cervical [21] and on gastric carcinomas [7].
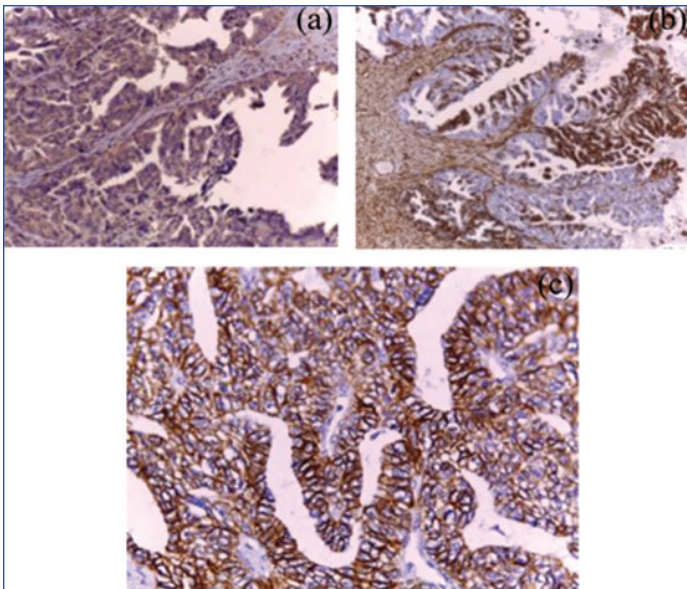
[Table/Fig-5]: IMP3 and EMT immunoreactivity in low grade OSC. a) Diffuse moderate IMP3 reactivity (x200); b) Focal strong cytoplasmic vimentin reactivity at the invasive margin of tumor cells and stromal component (x100); c) The previous low grade serous carcinoma showing diffuse strong memberanous E-cadherin immunostaining (x400).
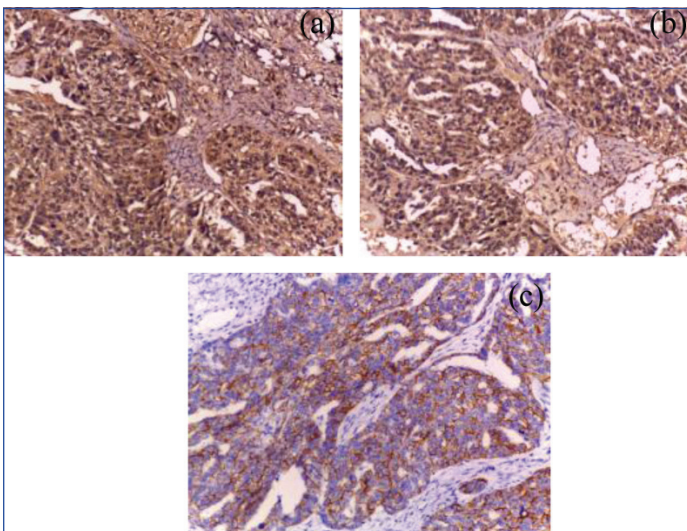
[Table/Fig-6]: IMP3 and EMT immunoreactivity in high grade OSC. a) Diffuse strong cytoplasmic IMP3 reactivity (x400); b) Diffuuse strong cytoplasmic vimentin immunoreactivity (x400); c) The previous high grade serous carcinoma showing focal weak memberanous E-cadherin immunostaining (x200).

| EMT related markers | n (% of total) 43 (100%) | IMP3 | | Spearman correlation value (r) | *p-value |
		Negative n (% of total) 10 (23.3%)	Positive n (% of total) 33 (76.7%)		
E-cadherin					
Negative	23 (53.5%)	1 (2.3%)	22 (51.2%)	-0.480	**0.001**
Positive	20 (46.5%)	9 (20.9%)	11 (25.6%)		
Vimentin					
Negative	9 (20.9%)	5 (11.6%)	4 (9.3%)	0.393	**0.009**
Positive	34 (79.1%)	5 (11.6%)	29 (67.4%)		

[Table/Fig-7]: Correlation between IMP3 and EMT related markers expression in Ovarian Serous Carcinomas (OSC).
*Chi-square test (χ^2)
EMT: Epithelial-mesenchymal transition; E-cadherin: Epithelial cadherin; IMP3: Insulin-like growth factor-2 mRNA binding protein 3

Analysis of IMP3 immunoreactivity among the studied 43 OSC, detected significant expression in invasive carcinomas with advanced FIGO stage and with lymph node metastases. This is consistent with the study of Zhu Q et al., who reported significant IMP3 expression in mucinous ovarian carcinomas with invasion-related indexes such as LN metastasis and advanced FIGO stage [18]. Moreover, they reported that IMP3 overexpression facilitated

cellular invasion in their in-vitro study. Thus, results of this study support that IMP3 is a prognostic marker associated with advanced stage and tumour invasiveness.

Analysis of EMT related markers expression among the studied 43 OSCs, detected significant loss of E-cadherin expression in advanced FIGO stage carcinomas compared to early stage. Although loss of E-cadherin expression was detected more frequently in high grades than in low grades OSCs, but the relationship did not reach significant level. This is consistent with previous researches that detected loss of E-cadherin immunoreactivity in invasive ovarian carcinomas in comparison with benign, borderline, and well-differentiated ovarian carcinomas [22-25]. Yuecheng Y et al., and Ryabtseva OD et al., detected significant relationship between loss of E-cadherin and clinical stage of epithelial ovarian carcinomas and serous tumours [23,25]. Moreover, they found significant association between E-cadherin loss and tumour grade. In contrast to results of this study, Voutilainen KA et al., and Koensgen D et al., found no significant association between loss of E-cadherin and clinicopathological features of the studied ovarian epithelial cancer [22,24].

The inactivation of E-cadherin gene with the progression of ovarian carcinoma may be attributed to gene mutation, or to two epigenetic mechanisms, including promoter hypermethylation and down regulation transcriptional factors, such as SLUG, SNAI, SIP1, PAK1, and mRNAs [26,27].

Result of present study detected Vimentin expression in 79.1% of the studied OSCs. Analysis of the result of Vimentin expression with clinicopatholgical features detected significant expression in high grades and advanced stage carcinomas. This is consistent with the finding detected by other related researches on ovarian carcinomas [28,29]. Also, Authors detected that vimentin expression with increased intensity was frequently detected at the invasive margin of positive tumours, an observation that was formerly described by Zhi Dong LV et al., [30].

In the present study, correlation analysis between the results of IMP3 and EMT related markers expressions among the studied 43 OSCs, detected an inverse significant correlation between the expressions of IMP3 and E-cadherin and positive significant correlation between IMP3 and vimentin. Therefore, the overexpression of IMP3 contributed to the EMT in OSCs progression. This finding is consistent with the finding of Su P et al., who reported that IHC expression of IMP3 inversely correlated with E-cadherin (r=-0.163, p=0.029) and directly correlated with both SLUG and vimentin expressions (r=0.27, p<0.001 and 0.366, p<0.001, respectively) among the studied breast cancer cases [31]. They concluded that IMP3 could promote invasion and metastasis via EMT in breast carcinoma cells. Jeng YM et al., also reported similar change of E-cadherin expression in IMP3-expressed hepatocellular carcinoma [32]. Thus, results of this study support the evidence that IMP3 is involved in EMT of OSC.

Limitation(s)

The researchers faced some limitations such as financial problems, and limited number of the studied cases due to exclusion of some cases with missed clinicopathological data.

CONCLUSION(S)

Insulin-like growth factor mRNA-binding Protein 3 (IMP3) is an oncogene unregulated in OSC. It is a prognostic marker associated with tumour aggressiveness. Moreover, IMP3 could promote tumour invasion and metastasis via EMT in OSCs.

REFERENCES

[1] Anuradha S, Webb PM, Blomfield P, Brand AH, Friedlander M, Leung Y, et al. Survival of Australian women with invasive epithelial ovarian cancer: A population-based study Med J Aust. 2014;201(5):283-88.
[2] Vang R, Shih Ie M, Kurman RJ. Ovarian low-grade and high-grade serous

carcinoma: Pathogenesis, clinicopathologic and molecular biologic features, and diagnostic problems Adv Anat Pathol. 2009;16(5):267-82.

[3]	Shih I, Kurman RJ. Ovarian tumourigenesis: A proposed model based on morphological and molecular genetic analysis Am J Pathol. 2004;164(5):1511-18.

[4]	Nielsen J, Christiansen J, Lykke-Andersen J, Johnsen AH, Wewer UM, Nielsen FC. A family of insulin-like growth factor II mRNA-binding proteins represses translation in late development Mol Cell Biol. 1999;19 (2):1262-70.

[5]	Sun X, Wei P, Shen C, Yang Y, Wang Y, Li Y, et al. Prognostic value of the IASLC/ATS/ERS classification and IMP3 expression in lung adenocarcinoma of Chinese cases Am J Cancer Res. 2015;5(7):2266-76.

[6]	Hsu KF, Shen MR, Huang YF, Cheng YM, Lin SH, Chow NH, et al. Overexpression of the RNA-binding proteins Lin28B and IGF2BP3 (IMP3) is associated with chemoresistance and poor disease outcome in ovarian cancer Br J Cancer. 2015;113(3):414-24.

[7]	Damasceno EA, Carneiro FP, Magalhaes AV, Carneiro Mde V, Takano GH, Vianna LM, et al. IMP3 expression in gastric cancer: Association with clinicopathological features and HER2 status. J Cancer Res Clin Oncol. 2014;140 (12):2163-68.

[8]	Yan J, Wei Q, Jian W, Qiu B, Wen J, Liu J, et al. IMP3 predicts invasion and prognosis in human lung adenocarcinoma. Lung. 2016;194(1):137-46.

[9]	Kalluri R, Weinberg RA. The basics of epithelial-mesenchymal transition. J Clin Invest. 2009;119(6):1420-28.

[10]	Yang MH, Wu MZ, Chiou SH, Chen PM, Chang SY, Liu CJ, et al. Direct regulation of TWIST by HIF-1alpha promotes metastasis Nature Cell Biology. 2008;10(3):295-305.

[11]	Wu Y, Deng J, Rychahou PG, Qiu S, Evers BM, Zhou BP. Stabilization of snail by NF-kappaB is required for inflammation-induced cell migration and invasion Cancer Cell. 2009;15(5):416-28.

[12]	Prasanphanich AF, Arencibia CA, Kemp ML. Redox processes inform multivariate trans-differentiation trajectories associated with TGFbeta-induced epithelial-mesenchymal transition. Free Radic Biol Med. 2014 76:01-13. doi: 10.1016/j.freeradbiomed.2014.07.032.

[13]	Thiery JP, Acloque H, Huang RY, Nieto MA. Epithelial-mesenchymal transitions in development and disease. Cell. 2009;139(5):871-90.

[14]	Meinhold-Heerlein I, Fotopoulou C, Harter P, Kurzeder C, Mustea A, Wimberger P, et al. The new WHO classification of ovarian, fallopian tube, and primary peritoneal cancer and its clinical implications. Arch Gynecol Obstet. 2016;293(4):695-700.

[15]	Höhn AK, Einenkel J, Wittekind C, Horn LC. New FIGO classification of ovarian, fallopian tube and primary peritoneal cancer. Pathologe. 2014;35(4):322-26.

[16]	Chisté M, Alexis J, Recine M. IMP3 Expression in serous tumours of the ovary. Appl Immunohistochem Mol Morphol. 2014;22(9):658-62.

[17]	Mima K, Hayashi H, Kuroki H, Nakagawa S, Okabe H, Chikamoto A, et al. Epithelial-mesenchymal transition expression profiles as a prognostic factor for disease-free survival in hepatocellular carcinoma: Clinical significance of transforming growth factor-b signaling. Oncol Lett. 2013;5(1):149-54.

[18]	Zhu Q, Qu Y, Zhang Q, Lu L, Weng W, Zhang H, et al. IMP3 is upregulated in primary ovarian mucinous carcinoma and promotes tumour progression. Am J

Transl Res. 2017;9(7):3387-98.

[19]	Wen N, Wang Y, Wen L, Zhao SH, Ai ZH, Wang Y, et al. Overexpression of FOXM1 predicts poor prognosis and promotes cancer cell proliferation, migration and invasion in epithelial ovarian cancer. J Transl Med. 2014;12:134. doi: 10.1186/1479-5876-12-134.

[20]	Li C, Zota V, Woda BA, Rock KL, Fraire AE, Jiang Z, et al. Expression of a novel oncofetalm RNA-binding protein IMP3 in endometrial carcinomas: Diagnostic significance and clinicopathologic correlations. Mod Pathol. 2007;20(12):1263-68.

[21]	Li C, Rock KL, Woda BA, Jiang Z, Fraire AE, Dresser K. IMP3 is a novel biomarker for adenocarcinomain situ of the uterine cervix: An immunohistochemical study in comparison with p16 (INK4a) expression. Mod Pathol. 2007;20(2):242-47.

[22]	Voutilainen KA, Anttila MA, Sillanpaa SM. Prognostic significance of E-cadherin-catenin complex in epithelial ovarian cancer. J Clin Pathol. 2006;59(5):460-67.

[23]	Yuecheng Y, Hongmei L, Xiaoyan X. Clinical evaluation of E-cadherin expression and its regulation mechanism in epithelial ovarian cancer. Clin Exp Metastasis. 2006;23(1):65-74.

[24]	Koensgen D, Freitag C, Klaman I, Dahl E, Mustea A, Chekerov R. Expression and localization of E-cadherin in epithelial ovarian cancer. Anticancer Res. 2010;30(7):2525-30.

[25]	Ryabtseva OD, Lukianova NY, Shmurakov YA, Polishchuk LZ, Antipova SV. Significance of adhesion molecules expression for estimation of serous ovarian cancer prognosis. Exp Oncol. 2013;35(3):211-18.

[26]	Elloul S, Vaksman O, Stavnes HT, Tropé CG, Davidson B, Reich R. Mesenchymal-to-epithelial transition determinants as characteristics of ovarian carcinoma effusions. Clin Exp Metastasis. 2010;27(3):161-72.

[27]	Davidson B, Tropé CG, Reich R. Epithelial-mesenchymal transition in ovarian carcinoma. Front Oncol. 2012;2:33. doi: 10.3389/fonc.2012.00033.

[28]	Georgescu CV, Florescu M, Simionescu C. Value of immunohistochemistry in confirming undifferentiated ovarian carcinomas. Rom J Morphol Embryol. 2004;45:133-42.

[29]	Wang Y, Wu R, Cho KR, Thomas DG, Gossner G, Liu JR, et al. Differential protein mapping of ovarian serous adenocarcinomas: Identification of potential markers for distinct tumour stage. J Proteome Res. 2009;8(3):1452-63.

[30]	Zhi Dong LV, Kong B, Li JG, Qu HL, Wang XG, Cao WH, et al. Transforming growth factor-b1 enhances the invasiveness of breast cancer cells by inducing a Smad2-dependent epithelial-to-mesenchymal transition. Oncol Rep. 2013;29(1):219-25.

[31]	Su P, Hu J, Zhang H, Li W, Jia M, Zhang X, et al. IMP3 expression is associated with epithelial-mesenchymal transition in breast cancer. Int J Clin Exp Pathol. 2014;7(6):3008-17.

[32]	Jeng YM, Chang CC, Hu FC, Chou HY, Kao HL, Wang TH, et al. RNA-binding protein insulin-like growth factor II mRNA-binding protein 3 expression promotes tumour invasion and predicts early recurrence and poor prognosis in hepatocellular carcinoma. Hepatology. 2008;48(4):1118-27.

PARTICULARS OF CONTRIBUTORS:
1.	Assistant Professor, Department of Pathology, Faculty of Medicine, Zagazig University, Zagazig, EL Sharkia, Egypt.
2.	Assistant Professor, Department of Obstetric and Gynecology, Faculty of Medicine, Zagazig University, Zagazig, EL Sharkia, Egypt.
3.	Lecturer, Department of General Surgery, Faculty of Medicine, Zagazig University, Zagazig, EL Sharkia, Egypt.
4.	Lecturer, Department of Pathology, Faculty of Medicine, Banha University, Banha, Egypt.

NAME, ADDRESS, E-MAIL ID OF THE CORRESPONDING AUTHOR:
Samah Said Elbasateeny,
Assistant Professor, Department of Pathology, Faculty of Medicine,
Zagazig University, Zagazig, EL Sharkia, Egypt.
E-mail: samah_elbasateeny@yahoo.com

AUTHOR DECLARATION:
• Financial or Other Competing Interests:	None
• Was Ethics Committee Approval obtained for this study?	Yes
• Was informed consent obtained from the subjects involved in the study?	NA
• For any images presented appropriate consent has been obtained from the subjects.	NA