Open Access

Original Article

Histopathological and Immunohistochemical Evaluation of Malignant Ovarian Tumours

Motia Kanwal, Tariq Sarfraz, Hina Tariq

Department of Histopathology, Army Medical College/National University of Medical Sciences (NUMS) Rawalpindi Pakistan

ABSTRACT

Objective: To determine the frequency and histological types of malignant ovarian tumours using morphological features and immunohistochemistry.

Study Design: Cross-sectional study.

Place and Duration of Study: Department of Histopathology, Army Medical College, Rawalpindi Pakistan, Jan 2016 to Dec 2018.

Methodology: Newly diagnosed cases of malignant ovarian tumour who had not received chemotherapy were included. Cases of benign ovarian tumours and those who were treated with pre-surgical chemotherapy were excluded.

Results: In total, 118 cases of malignant ovarian tumours were evaluated. High-grade serous carcinomas were 61(51.7%), which outnumber others, followed by granulosa cell tumours 17(14.4%), germ cell tumours 13(11%), endometrioid carcinoma 9(7.6%), clear cell carcinoma 4(3.4%), mucinous carcinoma 4(3.4%), low-grade serous carcinoma 2(1.7%) and carcinosarcoma in one case (0.8%). Cancer in the ovary was metastatic in 7(5.9%) cases. No Sertoli Leydig cell tumour, malignant Brenner tumour, embryonal carcinoma or immature teratomas were diagnosed.

Conclusion: Surface epithelial tumours were the most common malignancy, followed by granulosa cell tumours and dysgerminoma. An increase in the frequency of ovarian tumours in younger age groups was also noted. Immunohistochemistry was a useful adjuvant diagnostic tool in cases of ovarian malignancy. Metastases to the ovary were mostly gastrointestinal in origin.

Keywords: Histopathology, Granulosa cell tumor, Dysgerminoma Leydig cell tumor, Ovarian neoplasms, Sertoli cell tumor.

How to Cite This Article: *Kanwal M, Sarfraz T, Tariq H. Histopathological and Immunohistochemical Evaluation of Malignant Ovarian Tumours. Pak Armed Forces Med J 2024; 74(1): 26-30. DOI: https://doi.org/10.51253/pafmj.v74i1.5949*

INTRODUCTION

Ovarian cancers are one of the most commonly occurring malignancies in women worldwide. According to Stewart *et al*. the incidence of ovarian cancer in Northern Europe is 11.8 per 100,000 population.[1] Based on the literature available between 2008-2012, the incidence of ovarian cancers in the US is 12.1 per 100,000 women per year. Pakistan has one of the highest rates of ovarian cancer among various Asian countries.[2] Malignant ovarian tumours include primary ovarian tumours and metastatic tumours. Primary tumours include epithelial ovarian carcinomas, which comprise approximately 70% of all ovarian malignancies.[3] Due to the heterogeneity of ovarian carcinoma, diagnosis is not possible only on histopathological evaluation. In such instances, IHC is necessary for further typing. Thus, IHC is important in accurately differentiating between primary ovarian tumour subtypes and identifying metastasis.[4]

Clinically, bilateral presentation, multinodularity, surface involvement, vascular invasion, and involved hilar structures are a few characteristics that raise the possibility of metastatic tumours. Garland-type intraluminal necrosis and cribriform morphology are suggestive of colorectal origin.[5] Majority of ovarian, primary tumours present with unilateral involvement, expansile invasion, larger size and papillary architecture. Mucinous adenocarcinoma mimics primary surface ovarian tumours. In these cases, excluding the possibility of metastasis solely on morphology is tough.[6]

Sometimes, sex cord-stromal tumours also mimic endometrioid carcinoma, which can also show sex cord-like differentiation. The issue can be resolved using an IHC panel including epithelial membrane antigen (EMA), inhibin and calretinin.[7] Interestingly, using IHC markers significantly increases cases initially reported as grade-lll endometrioid carcinoma and later as high-grade serous carcinomas (HGSC).[8,9]

Few studies have been conducted in Pakistan that have evaluated both benign and malignant tumours. However, more studies are required to determine the frequencies of various ovarian malignancies in our population. The current study will focus only on

Correspondence: Dr Hina Tariq, Department of Histopathology, Army Medical College, Rawalpindi Pakistan

Received: 30 Dec 2020, revision received: 28 Aug 2021; accepted: 05 Oct 2021

malignant ovarian tumours and generate more local data to understand this lethal disease.

METHODOLOGY

The cross-sectional study was carried out at the Department of Histopathology, Army Medical College Rawalpindi, Pakistan from January 2016 to December 2018, after approval of the Institutional Review Board (ERC/FCPS-17). By using the WHO calculator, and the anticipated population proportion of 11.8%, sample size was calculated. The non-probability, consecutive sampling technique was performed.

Inclusion Criteria: All malignant ovarian tumours received with or without neoadjuvant chemotherapy, irrespective of age were included.

Exclusion Criteria: Patients with recurrence of malignant ovarian tumours and those diagnosed as benign ovarian tumours were excluded.

All the specimens of ovarian tumours were fixed in 10% neutral buffered formalin overnight. They were grossed according to the Royal College of Pathologists (RCPath) Datasets. LEICA TP 1020 (Germany) automatic tissue processor was used. The IHC panel was decided after microscopy and applied according to the differential diagnosis of a particular case (Table-I). Results were interpreted by one consultant histopathologist. Final diagnoses were based on histopathological and IHC results. Tumours were classified according to the WHO classification of ovarian tumours.[9,10]

Statistical Package for Social Sciences (SPSS) version 23.0 was used for the data analysis. Quantitative variables were expressed as Mean±SD and qualitative variables were expressed as frequency and percentages.

RESULTS

In total, 118 cases of malignant ovarian tumours were evaluated. The age of patients ranged from 7-75 years. The mean age was 46.4±15.13 years. High grade serous carcinomas were 61(51.7%), which outnumbered other tumours, followed by granulosa cell tumours 17(14.4%), germ cell tumour 13(11%), endometrioid carcinoma 9(7.6%), clear cell carcinoma 4(3.4%), mucinous carcinoma 4(3.4%), low-grade serous carcinoma 2(1.7%) and carcinosarcoma was seen in one case (0.8%). Cancer in the ovary was metastatic in 7(5.9%) cases. No Sertoli Leydig cell tumour, malignant Brenner tumour, embryonal carcinoma or immature teratomas were diagnosed. Further details are mentioned in Table-I.

While evaluating primary malignant ovarian tumours with immunohistochemical markers, surface epithelial tumours WT1 was positive in 81.9% of cases of high-grade serous carcinomas. ER was found to be immunoreactive in 66.7% of cases of endometrioid adenocarcinoma, and 100% of clear cell carcinoma was positive for HNF1b. Inhibin was positive in 82% of granulosa cell tumours, and AFP was positive in 100% of yolk sac tumours. CK 7 was positive in 60% of gastric metastatic carcinoma, while CK 20 was positive in 100% of colorectal metastatic carcinomas to the ovary (Table-II).

Table-I: IHC panel for diagnosis of Ovarian Carcinoma

Primary Ovarian Tumours	IHC markers
Epithelial Tumours	P53, WT 1, CA 125
Serous carcinoma	CK 7, CK20, ER,PR, PAX8,
Mucinous carcinoma	CA 125
Endometrioid carcinoma	ER, PR, CA125
Clear cell carcinoma	HNF-1B, Napsin A
Sex Cord Stromal Tumour	Inhibin, Calretinin
Germ Cell Tumours	PLAP, OCT3/4, SALL4, CD117, AFP, b-h CG, CD 30, CAM5.2
Metastatic Carcinoma	CK7, CK 20, CDX 2, CEA CK 7, WT 1, ER, PR

DISCUSSION

In our stuy we found that, surface epithelial tumours as the most common malignancy, followed by granulosa cell tumours and dysgerminoma. Various studies have mentioned that malignant surface epithelial tumours are the most common malignant ovarian tumours.[11] Lin *et al*. reported 50% of cases, Sheikh *et al*. 59.6%, and Patel *et al*. 68.4%.[8-10] In our study, the largest group was surface epithelial tumours which represented 68.6%. Razi *et al*. mentioned that most malignant surface epithelial tumours were diagnosed above 40 years.[12] In our study, the frequency was rising in younger age groups. Out of 81, 18 patients were diagnosed at or below 40 years of age, representing 22.2% of cases. It can be attributed to possible effects of specific genetic makeup and environmental and lifestyle changes.

HGSC was our study's most commonly reported malignant surface epithelial tumour, representing 51.7% of cases. In 2012, Zhao *et al*. reported 67.6% high-grade serous carcinomas.[13] Other studies, including Kommoss *et al*. and Jelovac *et al*. reported 45% and 75% HGSC, respectively.[14,15] In most of these studies, HGSC represented the most common epithelial malignancy except in the former. Reasons for differences could be variations in sample size. Kobel *et*

Table-II: Frequency of various Histologic Subtypes of Ovarian Carcinoma along with their Immunohistochcemical Profile (n=118)

Categories of Ovarian Malignancies	WHO Classification/His-tological type of tumours	No. of cases of each Subtype, n(%)	Immunohistochemical markers applied	No. of cases showing Positive Staining n(%)
Surface Epithelial Tumours		81(59.5)		
A	High Grade Serous Carcinoma	61(51.7)	WT 1	50 (81.9)
			P53	53(86.8)
			ER	40(65.5)
			CA125	48(78.7)
			CK7	38(62.3)
			CK20	00(00)
B	Low Grade Serous Carcinoma	02(1.7)	CK7	02(100)
			WT1	02(100)
C	Mucinous Carcinomas	04(3.4)	CK7	04(100)
			CK20	01(25)
			CDX2	00(00)
			ER	03(75)
			CA125	03(75)
D	Endometrioid Carcinoma	09(7.6)	CK7	09(100)
			CK20	00(00)
			PAX8	08(88.9)
			CA125	07(77.8)
			ER	06(66.7)
			WT1	00(00)
E	Clear Cell Carcinoma	04(3.4)	WT1	00(00)
			HNF1b	04(100)
			p53	01(25)
			CD30	00(00)
			AFP	00(00)
			CEA	01(25)
F	Carcinosarcoma	01(0.8)	CK AE1/AE3	01(100)
			Vimentin	01(100)
			b-hCG	00(00)
			SMA	00(00)
Sex cord stromal tumours		17(14.4)		
	Granulosa Cell Tumor	17(14.4)	Inhibin	14(82)
			Calretinin	13(76.5)
Germ cell tumours		13(12.2)		
A	Mixed Germ Cell tumor	04(3.2)	CD117	02(50)
			CD30	02(50)
			AFP	02(50)
			Beta HCG	00(00)
B	Yolk Sac Tumour	02(1.7)	AFP	02(100)
			Glypican3	02(100)
			SALL4	00(00)
C	Dysgerminoma	07(5.9)	PLAP	05(71)
			OCT3/4	06(71)
			CD 117	04(57.1)
Metastatic Tumours		07(5.9)		
A	Gastric Primary	04(3.4)	CK 7	03(60)
			CK 20	03(60)
			CDX 2	02(40)
B	Colorectal Primary	01(0.8)	CK 7	00(00)
			CK 20	01(100)
			CDX 2	01(100)
			CA 125	00(00)
			PAX 8	00(00)
C	Invasive Lobular Carcinoma	02(1.7)		
			CK7	02(100)
			GATA3	02(100)

al. reported WT1 immunoreactivity in 91.7% of HGSC,[16] Hashmi *et al*. reported 88.8% of CK7, 60% for CA125, and all were negative for CK20.[17] In the current study, 81.9% HGSC were positive for WT1, while ER

positivity was 65.6% and 78.7% were positive for CA125.

Nishal *et al*. analyzed the spectrum of ovarian tumours and pointed out that HGSC were positive for WT1 and p53 while negative for CK7 and CK20.[18] In our study, 62.3% of HGSC were positive for CK7 but all were negative for CK20. In 2017, Kommoss *et al*. reported 3% mucinous carcinoma, Zhao *et al*. found one case, and Modepalli *et al*. reported 2 cases.[14,13,19] The Results of these studies matched the results of our study, in which 4 cases of mucinous carcinomas made up 3.5% of cases. Modepalli *et al*. reported that ovarian mucinous carcinomas showed strong positivity with CK7 and exhibited variable positivity for CK20.[19] In our study, all clear cell carcinoma were WT 1 negative and HNF1 β-positive. Endometrioid carcinoma represented 7.6% in this study, while this entity represented 3.87% by Kobell *et al*. They reported that endometrioid carcinomas strongly co-expressed hormone receptors ER and CA125.[16] In the present study, endometrioid carcinoma showed positivity of ER and CA125 in 66.7% and 76.5%, respectively.

While studying the utility of IHC in undifferentiated ovarian carcinomas, one study found that CEA, CA125, CK7, CK20, and vimentin were useful markers to determine the nature of the tumours in 60-80% of cases.[20] In our study, many markers were used, including CK 7, CK 20, CDX 2, ER, Vimentin, WT1, GATA3, and b-hCG. These markers helped to make the final diagnosis in all difficult cases.

The second most common group in our study was sex cord-stromal tumours. In the current study, sex cord-stromal tumours accounted for 14.9% of all granulosa cell tumours. 82

Germ cell tumours represented 11.3% in our study. Germ cell tumours were slightly underreported in our study. The reason for the differences is the small sample size of the present study compared to previous studies. According to one study dysgerminomas were positive for PLAP, and yolk sac tumours were positive for AFP. Embryonal carcinoma was positive for CD30.[21] In our study, an embryonal component of mixed germ cell tumours was positive for CAM5.2, CD30 and OCT¾, dysgerminomas were positive for OCT ¾, CD117 and PLAP, while yolk sac tumours were positive for AFP. One previous study differentiated primary from metastatic carcinoma and concluded that the frequent primary site was pancreaticobiliary (95%), followed by an appendix (79%), colorectal (73%), endocervical (55%) and intestine (33%).[22] In the current study, 60% were Krukenberg, and 40% were metastatic colorectal carcinoma. In our study, CDX2 with CK20 indicated colorectal origin in two metastatic colorectal tumours.

CONCLUSION

Surface epithelial tumours were the most common malignant ovarian neoplasms. An increase in the frequency of malignant epithelial tumours in younger age groups was also noted, warranting larger population-based studies to verify the above findings. IHC was found to be a useful adjuvant tool in undifferentiated or poorly differentiated tumours and to ascertain the exact histological types of tumours. Sex cord-stromal tumours were the second most common, and granulosa cell tumours were predominant among them. The younger age group was affected mostly by germ cell tumours; among them, dysgerminomas were predominant. Metastasesto ovary was mostly gastrointestinal in origin.

Conflict of Interest: None.

Authors' Contribution

Following authors have made substantial contributions to the manuscript as under:

MK & TS: Study design, drafting the manuscript, approval of the final version to be published.

HT: Data acquisition, data analysis, data interpretation, critical review, approval of the final version to be published.

Authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

REFERENCES

1. Stewart SL. Ovarian Cancer Incidence: Current and Comprehensive Statistics. Ovarian Cancer - Clinical and Therapeutic Perspectives. IntechOpen Limited, London; 2012. http://dx.doi.org/10.5772/29758.
2. Kommoss S, Gilks CB, du Bois A, Kommoss F. Ovarian carcinoma diagnosis: the clinical impact of 15 years of change. Br J Cancer 2016; 115(8): 993-999. http://doi.org/10.1038/bjc.2016.273.
3. Verma N, Kumar M, Sagar M, Babu S, Singhai A, Singh N, et al. Expression of estrogen receptor, progesterone receptor, and human epidermal growth factor receptor type 2/neu in surface epithelial ovarian tumors and its clinichistopathological correlation. Indian J Health Sci Biomed Res 2018; 11(1):19.
4. Duraiyan J, Govindarajan R, Kaliyappan K. Applications of immunohistochemistry. J Pharm Bioallied Sci 2012; 4(Suppl 2): S307-309. http://doi.org/10.4103/0975-7406.100281.
5. Yemelyanova AV, Vang R, Judson K, Wu LS, Ronnett BM. Distinction of primary and metastatic mucinous tumors involving the ovary: analysis of size and laterality data by primary site with reevaluation of an algorithm for tumor classification. Am J Surg Pathol 2008; 32(1): 128-138. http://doi.org/10.1097/PAS.0b013e3180690d2d.
6. Coburn SB, Bray F, Sherman ME, Trabert B. International patterns and trends in ovarian cancer incidence, overall and by histologic subtype. Int J Cancer 2017; 140(11): 2451-2460. http://doi.org/10.1002/ijc.30676.

7. Köbel M, Rahimi K, Rambau PF, Naugler C, Le Page C, Meunier L, et al. An immunohistochemical algorithm for ovarian carcinoma typing. Int J Gynecol Pathol 2016; 35(5): 430-441. http://doi.org/10.1097/PGP.0000000000000274.
8. Lin F, Liu H. Immunohistochemistry in undifferentiated neoplasm/tumor of uncertain origin. Arch Pathol Lab Med 2014; 138(12): 1583-1610. http://doi.org/10.5858/arpa.2014-0061-RA.
9. Sheikh S, Bashir H, Farooq S, Beigh A, Manzoor F, Reshi R, et al. Histopathological spectrum of ovarian tumours from a referral hospital in Kashmir valley, Jammu and Kashmir, India. Int J Res Med Sci 2017; 5(5): 2110–2114. https://doi.org/10.18203/2320-6012.ijrms20171852.
10. Patel AS, Patel JM, Shah KJ. Ovarian tumors-Incidence and histopathological spectrum in tertiary care center, Valsad. Int Arch Integr Med 2018; 5(2): 84-93.
11. Zubair M, Hashmi SN, Afzal S, Muhammad I, Din HU, Hamdani SN. Ovarian Tumours: A study of 2146 Cases at AFIP, Rawalpindi, Pakistan. Aust Asian J Cancer 2015; 14(1): 21-26.
12. Razi S, Ghoncheh M, Mohammadian-Hafshejani A, Aziznejhad H, Mohammadian M, Salehiniya H. The incidence and mortality of ovarian cancer and their relationship with the Human Development Index in Asia. Ecancermedicalscience 2016; 10: 628. https://doi.org/10.3332/ecancer.2016.628.
13. Zhao L, Guo M, Sneige N, Gong Y. Value of PAX8 and WT1 Immunostaining in Confirming the Ovarian Origin of Metastatic Carcinoma in Serous Effusion Specimens. Am J Clin Pathol 2012; 137(2): 304-309. https://doi.org/10.1309/AJCPU0FION3RKKFO.
14. Kommoss F, Gilks CB. Pathology of Ovarian Cancer: Recent Insights Unveiling Opportunities in Prevention. Clin Obstet Gynecol 2017; 60(4): 686-696. https://doi.org/10.1097/GRF.0000000000000314.
15. Jelovac D, Armstrong DK. Recent progress in the diagnosis and treatment of ovarian cancer. CA Cancer J Clin 2011; 61(3): 183-203. https://doi.org/10.3322/caac.20113.
16. Köbel M, Bak J, Bertelsen BI, Carpen O, Grove A, Hansen ES, et al. Ovarian carcinoma histotype determination is highly reproducible, and is improved through the use of immunohistochemistry. Histopathology 2014; 64(7): 1004-1013. https://doi.org/10.1111/his.12349.
17. Hashmi AA, Hussain ZF, Bhagwani AR, Edhi MM, Faridi N, Hussain SD, et al. Clinicopathologic features of ovarian neoplasms with emphasis on borderline ovarian tumors: an institutional perspective. BMC Res Notes 2016; 9: 205. https://doi.org/10.1186/s13104-016-2015-5.
18. Nishal AJ, Naik KS, Modi J. Analysis of spectrum of ovarian tumours: a study of 55 cases. Int J Res Med Sci 2015; 3(10): 2714-2717. http://doi.org/10.18203/2320-6012.ijrms20150820.
19. Modepalli N, Venugopal SB. Clinicopathological Study of Surface Epithelial Tumours of the Ovary: An Institutional Study. J Clin Diagn Res 2016; 10(10): EC01-EC04. http://doi.org/10.7860/JCDR/2016/21741.8716.
20. Li Q, Zeng X, Cheng X, Zhang J, Ji J, Wang J, et al. Diagnostic value of dual detection of hepatocyte nuclear factor 1 beta (HNF-1β) and napsin A for diagnosing ovarian clear cell carcinoma. Int J Clin Exp Pathol 2015; 8(7): 8305-8310.
21. Kao YC, Lin MC, Lin WC, Jeng YM, Mao TL. Utility of hepatocyte nuclear factor-1β as a diagnostic marker in ovarian carcinomas with clear cells. Histopathology 2012; 61(5): 760-768. https://doi.org/10.1111/j.1365-2559.2012.04267.x.
22. DeLair D, Han G, Irving JA, Leung S, Ewanowich CA, Longacre TA, et al. HNF-1β in ovarian carcinomas with serous and clear cell change. Int J Gynecol Pathol 2013; 32(6): 541-546. https://doi.org/10.1097/PGP.0b013e318273fd07.