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HISTOMORPHOLOGY OF OVARIAN TUMOURS WITH SPECIAL REFERENCE TO IMMUNOHISTOCHEMICAL MARKERS CA-125 AND CEA IN SURFACE EPITHELIAL OVARIAN TUMOURS

Histopathology

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ABSTRACT

Background: The Ovary is the third most common site of primary malignancies of the female genital tract among Indian women. In India, the prevalence of ovarian cancer is 2.5%-3%. Due to its asymptomatic nature and inaccessible site, early diagnosis is difficult, which increases the mortality rate of these neoplasms. Early diagnosis of ovarian tumours is important to decrease the morbidity and mortality of this condition. **Materials and Methods:** All ovarian specimens after oophorectomy, salpingo-oophorectomy, and hysterectomy with salpingo-oophorectomy received from March 2023 to September 2024 were studied. A total of 124 ovarian specimens were examined. Patient details, imaging findings, gross features and microscopic findings of all the cases were noted. Immunohistochemistry was done wherever required for diagnosis. **Results:** Out of 124 ovarian specimens, the majority of the cases were diagnosed as benign (n=114,91.9%), followed by borderline tumours (n=4, 3.2%) and malignant tumours (n=6, 4.8%). Tumour-like lesions (n=55, 44.3%) constituted the majority of the tumours, followed by surface epithelial tumours (n=40,32.2%), germ cell tumours (n=21,16.9%) and sex cord stromal tumours (n=8,6.4%). Tissue expression of CA-125 showed an increasing trend among serous epithelial tumours while CEA tissue expression increased from mucinous cystadenoma to mucinous adenocarcinoma. **Conclusion:** Histopathological examination remains the cornerstone in ovarian tumour evaluation. All ovarian specimens must be thoroughly examined for gross features and microscopy. CA-125 and CEA serve as reliable prognostic markers for serous and mucinous epithelial tumours, respectively.

KEYWORDS

Ovary, histopathology, immunohistochemistry, tumours, malignant

INTRODUCTION

The ovary is the second most common site of primary malignancies of the female genital tract among Indian women[4]. In the world, the incidence of ovarian cancer is 0.73%. In India, the prevalence of ovarian cancer is 2.5%-3%[5]. Due to its asymptomatic nature and inaccessible site, it is difficult to make an early diagnosis. This is responsible for the increase in mortality of these neoplasms[2].

Early diagnosis of ovarian tumours is important to decrease the morbidity and mortality of this condition. Their complex nature, unpredictable behaviour and prognosis make ovarian tumours a difficult problem for gynaecologists all over the world. Despite the availability of advanced technology, the survival rate of ovarian tumours in the early-stage and the advanced stage is poor. Since the histogenesis of many tumours is interrelated, accurate histopathological diagnosis is required for proper treatment[2].

Carcinoembryonic antigen (CEA) is a glycoprotein that belongs to the immunoglobulin family called CEA-related cell adhesion molecules (CEACaMs). CEA is detected in mucinous ovarian tumours. CA-125 is a high molecular weight, membrane-associated mucin, which is expressed in advanced serous epithelial carcinoma of the ovary. CA-125 is the gold standard tumour marker in ovarian cancer[6].

Histopathological examination of the ovarian tumours remains the mainstay of diagnosis, which helps us to determine the prognosis and behaviour of the neoplasms.

Objectives

This study was done at a tertiary care centre in Bangalore,

1) To describe the histopathological spectrum of resected ovarian tumours.
2) To determine the immunohistochemical expression pattern of CA-125 and CEA in surface epithelial tumours.

MATERIALS AND METHODS

After taking informed consent from the patient in his /her language, detailed clinical history and results of relevant investigations were collected from the patient's case files. All ovarian specimens received in the Department of Pathology at Dr. B. R. Ambedkar Medical College and Hospital from March 2023 to September 2024, following oophorectomy, salpingo-oophorectomy, or hysterectomy with salpingo-oophorectomy, were studied. A total of 124 ovarian specimens were received. All the cases studied were obtained after approval from the institutional scientific and ethical committee. Informed consent was obtained in the patient's understandable language for all cases. The present study is a prospective observational study. Autolysed ovarian specimens were excluded from the study.

Specimens were kept for fixation for 24 hours in 10% buffered formalin. After a detailed specimen description, multiple sections were taken as per the standard protocol from the viable sections of the tumour, including surgical margins. After conventional processing, the specimens were embedded in paraffin wax, and sections of 4-5µm thickness were cut using Leica RM 2125 and stained using haematoxylin and eosin for histopathological study. In addition, 4µm sections were cut from paraffin blocks of tissue and taken on 2 poly-L-lysine-coated glass slides for further immunohistochemical analysis to detect CA-125 and CEA expression. The H&E-stained slides were studied for the tumour histology and other features as per the standard reporting protocol. The immune-stained slides were examined for membrane and cytoplasmic staining with CA-125 and CEA.

Criteria for evaluation of tissue expression of CA-125 and CEA were done by multiplying the intensity score by the proportion of tumour cells stained. For calculating intensity scoring, 0- negative,1- weak staining; cytoplasm and membrane blue-brown, 2- moderate staining; cytoplasm and membrane brown and 3-strong staining; deep brown or black. Proportion is evaluated by taking the overall percentage of tumour cells positive for the staining[6,22].

Analysis of data was done using SPSS, version 30 (IBM Corp, NY, USA). All the patients were analysed and distributed based on the age group, presenting symptoms, laterality and type of ovarian tumour according to the WHO 2020 classification.

RESULTS

Out of 124 ovarian specimens, the majority of the cases were diagnosed as benign (n=114,91.9%), followed by malignant tumours (n=6, 4.8%) and borderline tumours (n=4, 3.2%) (Figure 1).

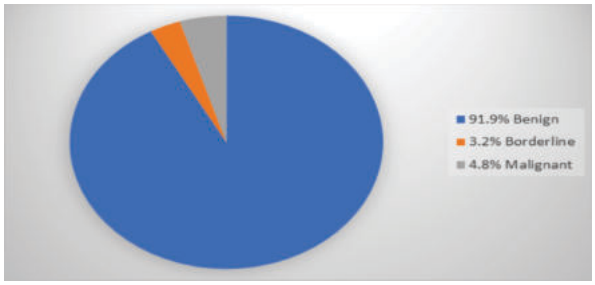

Figure 1: Pie Chart Showing Distribution Of Benign, Borderline And Malignant Ovarian Tumours In Our Study

Tumour-like lesions constituted the majority of the tumours, followed by surface epithelial tumours, germ cell tumours and sex cord stromal tumours (Table 1).

Table 1: Total Number Of Cases Distributed Among The Major Who Classifications

Classification	Number of cases	Percentage
Tumor-like lesions	55	44.3
Surface epithelial tumours	40	32.2
Germ cell tumours	21	16.9
Sex cord stromal tumours	8	6.4
Total	124	100

Among tumour-like lesions, follicular cysts were the most common, with 34 cases, followed by luteal cysts with 21 cases. In surface epithelial tumours, serous cystadenoma constituted the majority of the cases (n=16), followed by 6 cases of mucinous cystadenoma (figure 2) and 5 cases of endometroid cystadenoma. Borderline serous tumour constituted 3 cases (figure 3), and borderline brenner tumour constituted one case each. We also received 2 cases of seromucinous cystadenoma. In our study, the most common malignant case reported was high-grade serous cystadenocarcinoma of the ovary (n=3) (figure 4), followed by one case each of mucinous adenocarcinoma (figure 5) and carcinosarcoma of the ovary (figure 6). Among germ cell tumours of the ovary, we have received 21 cases of benign mature teratoma (figure 7). In sex cord stromal tumours of the ovary, we have received two cases of fibroma, four cases of thecoma and one case each of adult granulosa cell tumour (figure 8) and sertoli cell tumour (Table 2).

Table 2: Distribution Of Ovarian Tumours According To The Who 2020 Classification, Nos: Not Otherwise Specified

Classification	Subclassification	Tumor type	Number of cases	Percentage
Surface epithelial tumours	Serous Tumors	Serous Cystadenoma NOS	16	12.9
		Serous borderline tumour NOS	3	2.4
		Low-grade serous carcinoma	0	0
		High-grade serous carcinoma	3	2.4
	Mucinous tumors	Mucinous cystadenoma NOS	6	4.8
		Mucinous borderline tumour	0	0
		Mucinous adenocarcinoma	1	0.8
	Endometrioid tumors	Endometrioid cystadenoma NOS	5	4.03
		Endometrioid tumour, borderline	0	0
		Endometrioid adenocarcinoma NOS	0	0
	Clear cell tumours	Clear cell cystadenoma	0	0
		Clear cell borderline tumour	0	0
		Clear cell adenocarcinoma NOS	0	0
	Seromucinous Tumors	Seromucinous cystadenoma	2	1.6
		Seromucinous adenofibroma	0	0
		Seromucinous borderline tumour	0	0
	Brenner Tumors	Brenner tumor NOS	2	1.6
		Brenner tumour borderline, malignancy	1	0.8
		Brenner tumor, malignant	0	0
	Other Carcinomas	Carcinosarcoma NOS	1	0.8
		Fibroma NOS	2	1.6
Sex cord stromal tumours	Pure Stromal Tumours	Thecoma NOS	4	3.2
		Sclerosing stromal tumour	0	0
		Fibrosarcoma NOS	0	0
	Pure Sex-Cord Tumours	Adult granulosa cell tumour	1	0.8
		Granulosa cell tumour, juvenile	0	0
		Sertoli cell tumor NOS	1	0.8
	Mixed Sex-Cord-Stromal tumors	Sertoli-Leydig cell tumor NOS	0	0
Germ cell tumours		Teratoma, benign	21	16.9
		Immature teratoma NOS	0	0
		Dysgerminoma	0	0
		Yolk sac tumour NOS	0	0
		Embryonal carcinoma NOS	0	0
		Mixed germ cell tumour	0	0
		Struma ovarii NOS	0	0
Germ cell-sex cord-stromal tumours		Gonado-blastoma	0	0
		Mixed germ cell-sex cord-stromal tumour NOS	0	0
Tumor-like lesions		Follicle cyst	34	27.4
		Corpus luteum cyst	21	16.9
Metastases to the ovary		Signet ring cell adenocarcinoma	0	0
		Metastatic adenocarcinoma	0	0
Total			124	100

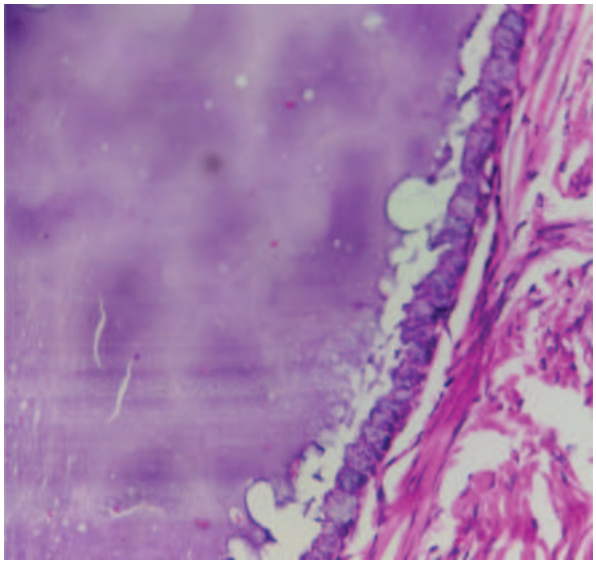

Figure 2: Mucinous Cystadenoma (H&E, 40x)- Cyst Lined By Mucinous Columnar Epithelium

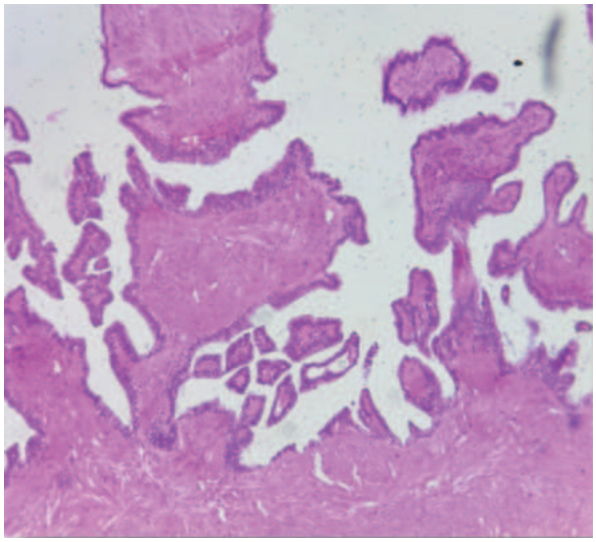

Figure 3: Borderline Serous Cystadenofibroma (H&E,4x)- Papillae Are Lined By Stratified Squamous Epithelium With Detached Clusters Of Tumor Cells.

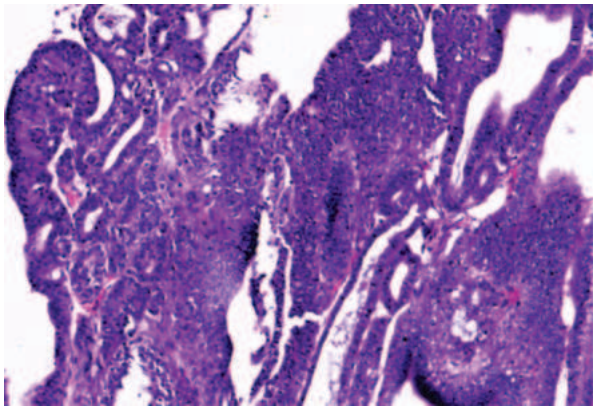

Figure 4: High-grade Serous Carcinoma Of The Ovary (H&E, 10x): Pleomorphic Tumour Cells Arranged In Glandular And Papillary Pattern With Nucleus Showing Loss Of Basal Polarity And Nuclear Crowding.

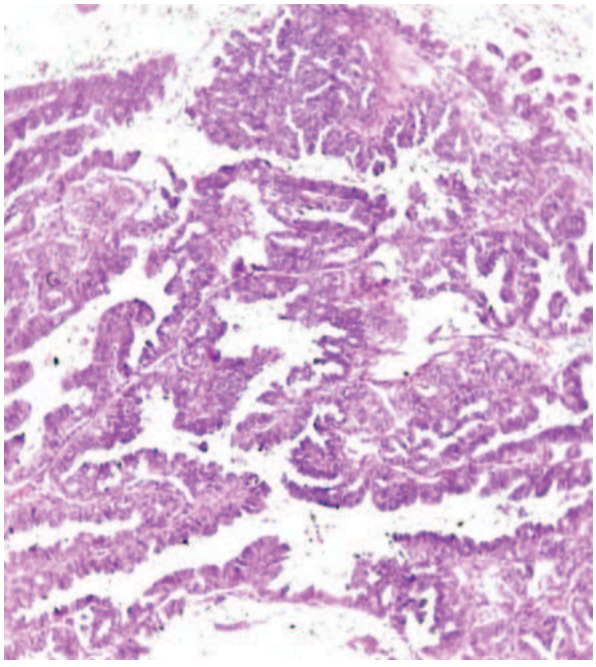

Figure 5: Mucinous Adenocarcinoma Ovary (H&E, 10x)- Pleomorphic Tumour Cells Arranged In Papillary Growth Pattern With Expansile/confluent Invasion.

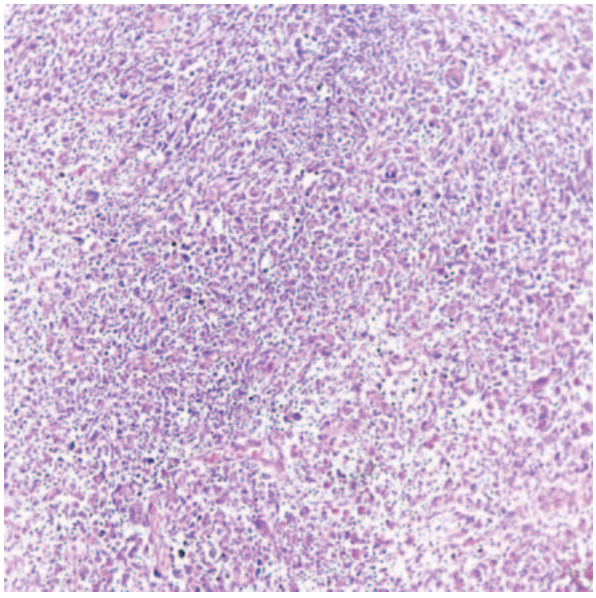

Figure 6: Carcinosarcoma Of The Ovary (H&E, 10x)- Shows A Biphasic Pattern With High-grade Epithelial And Mesenchymal Elements.

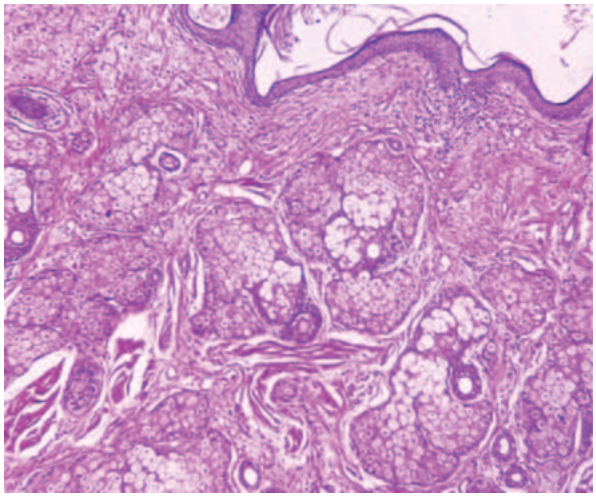

Figure 7: Mature Cystic Teratoma (H&E, 10x) Showing Sebaceous Glands And Hair Shafts.

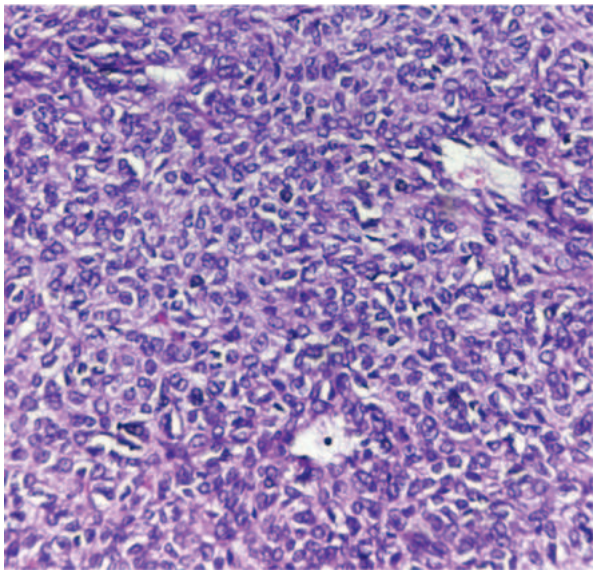

Figure 8: Adult Granulosa Cell Tumour Of The Ovary (H&E, 40x): Showing A Monotonous Population Of Tumour Cells Exhibiting Nuclear Grooving.

Incidence of ovarian tumours is most common in the age group of 31 to 40 years (n=53, 42.7%), followed by the 42 to 50 years age group (n=36, 29%). Ovarian tumours are least common in both extremes of age, that is less than 10 years age group and in elderly females. In our study, surface epithelial tumours, germ cell tumours and sex cord stromal tumours were most common in the 31-40 years age group. Whereas tumour-like lesions were more common among the 41 to 50-year age group (Table 3).

Table 3: Incidence Of Ovarian Tumours According To Age Distribution In Each Classification

Age in years	Total number of cases	%	SET	SCT	GCT	GCSCT	TLL	M
0-10	1	0.8	0	0	0	0	1	0
11-20	4	3.2	1	0	2	0	1	0
21-30	17	13.7	12	1	3	0	1	0
31-40	53	42.7	13	5	16	0	19	0
41-50	36	29	9	1	0	0	26	0
51-60	10	8	3	0	0	0	7	0
>61	3	2.4	2	1	0	0	0	0
Total	124	100	40	8	21	0	55	0

%- percentage, SET: Surface epithelial tumours, SCT: Sex cord stromal tumours, GCT: Germ cell tumours, GCSCT: Germ cell- sex cord-stromal tumours, TLL: Tumour-like lesions, M- Metastases to the ovary

In our study, the majority of the benign cases were diagnosed in the age group 31-40 years (n=51) and borderline cases in the age group of 21-30 years (n=2). Malignant cases were diagnosed most commonly in the age group of 51-60 years (n=2) (Table 4).

Table 4: Age-wise Distribution Of Benign, Borderline And Malignant Tumours Of The Ovary

Age in years	Benign	Borderline	Malignant	Total number of cases	%
0-10	1	0	0	1	0.8
11-20	4	0	0	4	3.2
21-30	14	2	1	17	13.7
31-40	51	1	1	53	42.7
41-50	34	1	1	36	29
51-60	8	0	2	10	8
>61	2	0	1	3	2.4
Total	114	4	6	124	100

In our study, the most common clinical presentation is mass per abdomen (n=46), followed by abdominal pain (n=33), abdominal distension (n=22) and menstrual irregularity (n=16). Ascites was the least common clinical presentation (n=7). (Figure 9).

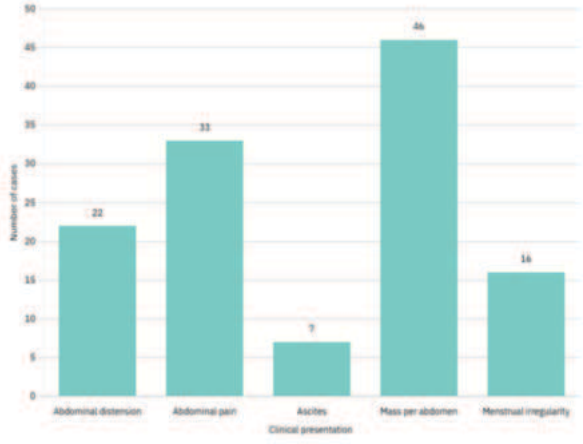

Figure 9: Bar Graph Showing Clinical Presentation Of The Patient On The X-axis And The Number Of Cases On The Y-axis

In our study, the majority of the ovarian tumours were unilateral. In surface epithelial tumours,36 cases were unilateral and 4 were bilateral. Among sex cord stromal tumours, all 8 cases were unilateral and none of them were bilateral. In germ cell tumours, 16 cases were unilateral and 5 cases were bilateral. Among tumour-like lesions, 50 cases were unilateral and 5 cases were bilateral (Figure 10).

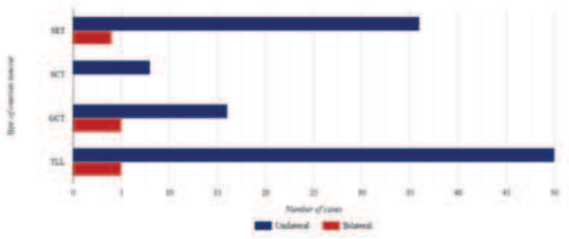

Figure 10: Laterality Of Ovarian Tumours
SET: Surface epithelial tumours, SCT: Sex cord stromal tumours, GCT: Germ cell tumours, TLL: Tumour-like lesions

In immunohistochemical studies, it was found that the CA-125 tissue score shows an increasing trend from benign serous cystadenoma (figure 11) to borderline serous cystadenoma (figure 12) and malignant high-grade serous carcinoma of the ovary (figure 13). In the case of CEA tissue scoring, it was found that the majority of the benign mucinous cystadenomas (Figure 14) showed a score of more than 50. The score was increased in malignant mucinous adenocarcinoma of the ovary (Figure 15) (Table 5).

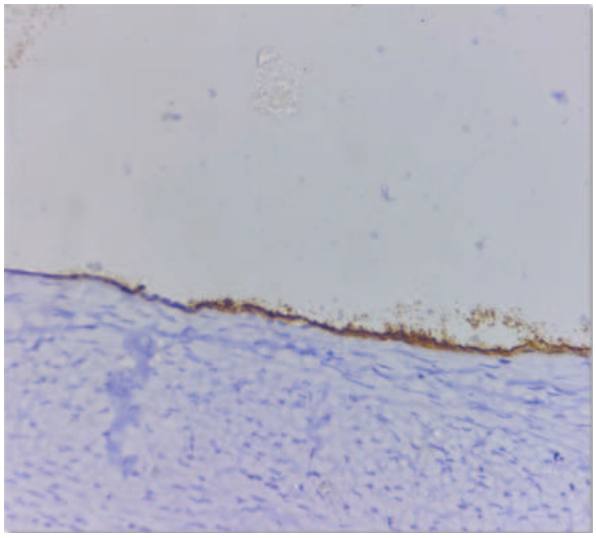

Figure 11: Immunohistochemistry Ca-125 On Serous Cystadenoma (40x) With Score Of 40

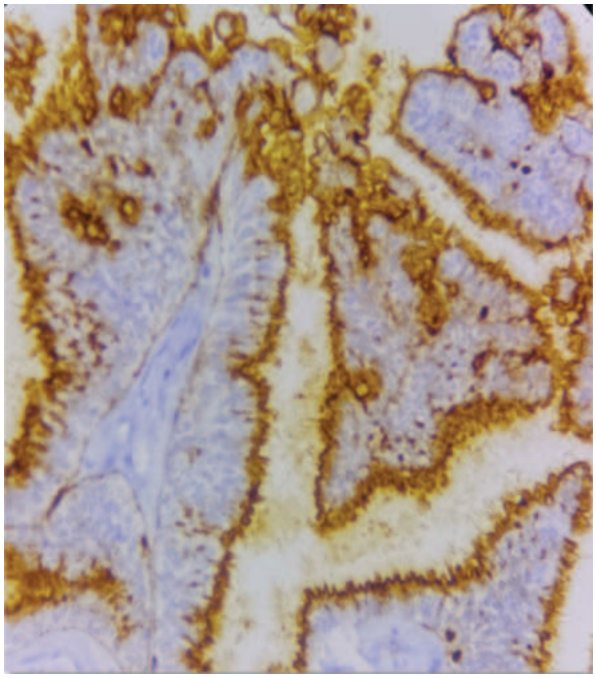

Figure 12: Immunohistochemistry CA-125 On Borderline Serous Cystadenofibroma (40x) With Score- 60

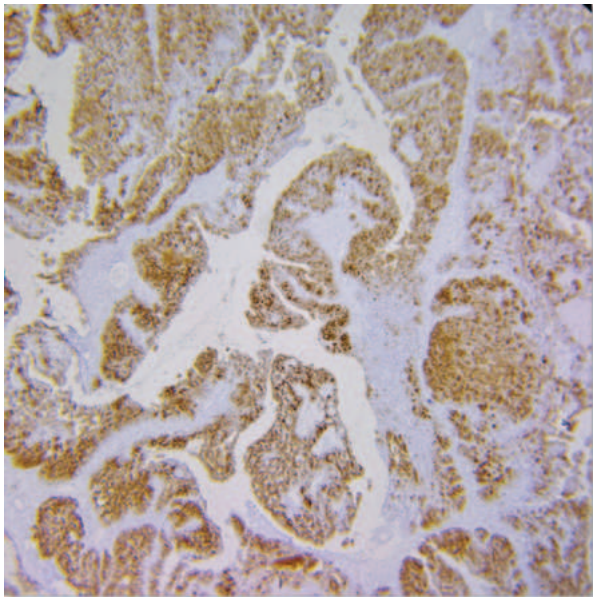

Figure 13: Immunohistochemistry CA-125 On High Grade Serous Carcinoma Ovary (4x) With A Score Of 90.

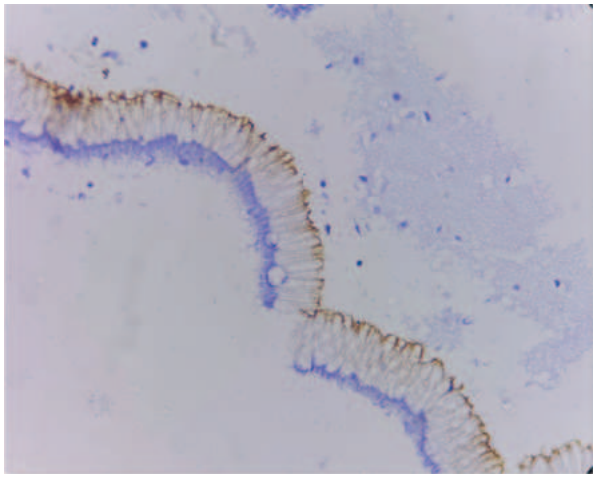

Figure 14: Immunohistochemistry Of CEA (40x) On Mucinous Cystadenoma Showing Apical Staining With Score 55.

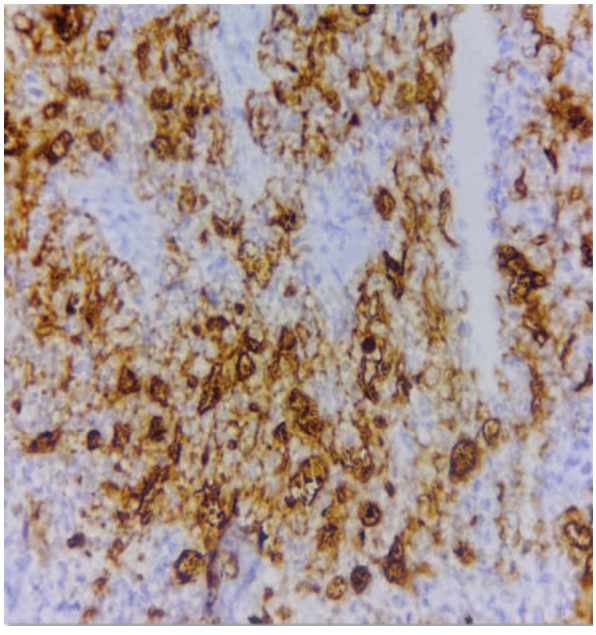

Figure 15: Immunohistochemistry Of CEA (40x) On Mucinous Adenocarcinoma Of The Ovary With A Score Of 80.

Table 5: Immunohistochemical Studies Of CA-125 And CEA In Epithelial Ovarian Tumours

Type of ovarian tumour	Total number of cases	Tissue CA-125 score		Tissue CEA score	
		<50 (%)	>50 (%)	<50 (%)	>50 (%)
Serous cystadenoma NOS	16	11 (68.7)	5 (31.2)	16 (100)	0 (0)
Serous borderline tumour NOS	3	1 (33.3)	2 (66.6)	3 (100)	0 (0)
High-grade serous carcinoma	3	0 (0)	3 (100)	3 (100)	0 (0)
Mucinous cystadenoma NOS	6	5 (83.3)	1(16.6)	1 (16.6)	5 (83.3)
Mucinous adenocarcinoma	1	1(100)	0 (0)	0 (0)	1 (100)
Endometrioid cystadenoma NOS	5	5 (100)	0 (0)	5 (100)	0 (0)
Seromucinous cystadenoma	2	2 (100)	0 (0)	2 (100)	0 (0)
Brenner tumour NOS	2	2 (100)	0 (0)	2 (100)	0 (0)
Borderline Brenner Tumour	1	1(100)	0 (0)	1 (100)	0 (0)
Carcinosarcoma NOS	1	1 (100)	0 (0)	1 (100)	0 (0)

NOS: not otherwise specified

DISCUSSION

In our study, majority of the tumors diagnosed were benign (n=114,91.9%), followed by malignant tumours (n=6, 4.8%) and borderline tumours (n=4, 3.2%) which is in concordance with studies conducted by Pranshu Sharma et al (2020)[2], Priyanka Nimbalkar Jadhav et al (2020)[5], Bagadi V et al (2022)[7], Palak J Modi et al (2018)[9], Mehra P et al (2023)[18]and Batool A et al (2022)[19]. (Table 6)

Table 6: Summary Of Comparison Among Different Studies With Our Study.

Study	Tumour Type	Tumour classifi-cation	Laterality	Most common Clinical presen-tation	Most com-mon age group (years)
Our study	Among 124 cases, majority were Benign (n=114,91.9%) , followed by malignant tumours (n=6, 4.8%) and borderline tumours (n=4, 3.2%)	TLL were most common (n=55,44.3%) followed by SET (n=40,32.2%) , GCT (n=21,16.9%) and SST (n=8,6.4%).	Unilateral (n=110,88. 7%) > Bilateral (n=14,11.2 %)	Mass per abdome n (n=46, 37%)	31-40
Pranshu Sharma et al (2020)2	The majority were benign tumours (66.92%) followed by malignant tumours (28.46%).	SET were most common (76.12%) followed by GCT (18.46%)	-	Pain ab-domen	41-50
Pri-yanka Nim-balkar Jadhav et al (2020)5	Out of 112 cases, 86 were benign and 26 were malignant.	SET were the most common (n=62,55%) followed by GCT (n=36,32%) and SCT (n=14,13%)	Unilateral(89%) > Bilateral (11%)	Mass per ab-domen	21-30

Bagadi V et al (2022)7	Both benign epithelial and malignant tumours were 85.47% and 11.97%, respectively.	SET were the most common (81.96%) followed by GCT (8.54%) , SCT (6.83%) and mixed tumours (3.41%).	Unilateral (n=105,89.7%) > Bilateral (n=12, 10.2%)	Pain abdomen	<50 years
Palak J Modi et al (2018)9	Among 308 ovarian tumours studied, 88% were benign, 8% malignant and 4% borderline ovarian tumours.	SET were the most common (40%) followed by GCT (27%) , SCT (4%) and metastasis (1%)	Unilateral (96%) > Bilateral (4%)	-	20-29 years
Mehra P et al (2023)18	Among 110 ovarian tumours, 69% were benign, 5.4% were borderline and 24.5% were malignant.	SET were the most common (n=77, 70%) followed by GCT (n=23, 21%), metastasis (n=7, 6.3%) and SCT (n=3, 2.7%)	-	Mass per abdomen	21-30
Batool A et al (2022)19	Among 390 ovarian tumours, 320 (82.05%) were benign, 11 (2.82%) were borderline, 57 (14.61%) were malignant ovarian tumours.	SET (n=246 ,63.08%) were the most common, followed by GCT (n=115 ,29.48%), SST(n=27 ,6.92%) and metastasis (n=2,0.52%).	-	-	21-30

SET: Surface epithelial tumours, SCT: Sex cord stromal tumours, GCT: Germ cell tumours, TLL: Tumour-like lesions

In our study most common lesion was tumour-like lesions (n=55, 44.3%), followed by surface epithelial tumours (n=40, 32.2%), germ cell tumours (n=21,16.9%) and sex cord stromal tumours (n=8, 6.4%). In the study conducted by Batool A et al (2022)[19], surface epithelial tumours were the most common followed by germ cell tumours and sex cord stromal tumours. Similar results were seen in the study conducted by Mehra P et al (2023)[18], Palak J Modi et al (2018)[9], Bagadi V et al (2022)[7], Priyanka Nimbalkar Jadhav et al (2020)[5]and Pranshu Sharma et al (2020)[2]. Tumour-like lesions were not included in the above studies.

It was found that majority of the ovarian tumours irrespective of the tumour classification in our study were unilateral which is in concordance with the study conducted by Priyanka Nimbalkar Jadhav et al (2020)[5], Palak J Modi et al (2018)[9] and Bagadi V et al (2022)[7].

In our study, mass per abdomen was the most common clinical presentation of the patient with an ovarian tumour. This is in concordance with the study conducted by Priyanka Nimbalkar Jadhav et al. (2020)[5] and Mehra P et al. (2023)[18]. In contrast, abdominal pain was the most common clinical presentation in the studies conducted by Pranshu Sharma et al. (2020)[2]and Bagadi V et al. (2022)[7].

In our study, the peak incidence of ovarian tumours was found in 31-40 years age group whereas its 21- 30 years in studies conducted by Priyanka Nimbalkar Jadhav et al (2020)[5], Batool A et al (2022)[19], Palak J Modi et al (2018)[9] and Mehra P et al (2023)[18]. 41-50 years age group showed peak incidence of ovarian tumours in the study conducted by Pranshu Sharma et al (2020)[2].

In immunohistochemical studies, tissue expression of CA-125 score was found to have an increasing trend from benign, borderline to malignant serous tumours of the ovary. Score was more than 50 in serous cystadenoma to more than 90 in high grade serous carcinoma. However, among mucinous epithelial tumours and other types, such a trend was not seen. Tissue expression of CEA score was more than 50 in most of the mucinous cystadenomas and less than 50 in mucinous adenocarcinoma. According to the study conducted by Devan SM et al.[22], intensity of CA-125 staining in tissues increased from benign to malignant epithelial ovarian tumours. However, further studies are required on this topic.

CONCLUSION

Histopathological examination remains the cornerstone in ovarian tumour evaluation.All ovarian specimens should be meticulously examined for both gross features and microscopic characteristics. Using WHO classification of ovarian tumours helps in better categorization of these tumours which will aid in the prognosis and treatment of the patients. Tumour-like lesions were the most common lesion, followed by surface epithelial tumours of the ovary. The most common presentation is unilateral with a mass per the abdomen. The peak incidence of ovarian tumours was between 21 to 30 years of age. Tissue expression of CA-125 can be used as a prognostic marker for monitoring the progress of surface epithelial tumours, particularly serous tumours. CEA tissue expression can be used as a prognostic marker for mucinous epithelial tumours.

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