CHAPTER VI

HISTOLOGICAL AND HISTOCHEMICAL OBSERVATIONS AND DISCUSSION ON MUCOSUBSTANCES IN NORMAL AND CANCEROUS OVARY

The ovaries are female reproductive organ symmetrical placed in pelvic region. They are situated on either side of the uterus, below the follipian tubes. There is widest range of cancer in ovary. Ovary showed various types of cancerous condition in it. When ovary showed papillary pattern called papillary adenocarcinoma, some times the cancer arises from the connective tissue. Though the acurnce of ovarian malignancy is more, less attention paid to mucosubstances in it. But major work did on its histological classification and about theraphy.

I) Histological Observations of Normal Ovary. For histological prepration the paraffin sections of 5 μ were taken. Hematoxylene - Esoin stained prepration revealed the histological structure of ovary. The ovary showed germinal epithelium and central stroma. In stroma there were follicles.

Germinal epithelium covers the ovary. The stroma is differentiated into cortex and medulla. The germinal epithelium often spoken as a modified peritoneum. The stroma having primordial follicles. The primordial follicles then develops into graffian follicles. The structural parts in intrest for this investigation are the germinal epithelium as shown in Plate No.7, Figs.1.

II) Histochemical Observations of Normal Ovary.

The histochemical observations of different techniques

are given in table No.6. These reactions about mucosubstances are illustrated photomicrographically in plate No. 7, Figs. 2 to 3.

A) Germinal Epithelium. The germinal epithelium showed intense PAS reactivity (Fig.2). The staining intensity gets reduced after saliva or amylase digestion, showed presence of glycogen. The PAS activity reduced prior to phenylahydraline treatment showed probable presence of neutral and acidic mucosubstances.

Weak alcianophilia was observed with AB pH 1.0 (Fig.3) showed probable presence of sulfomucins. Bluish pink staining was observed with ABpH 1.0 - PAS technique. Metachromasia was observed with all pH levels with azure, A. The critical electrolyte concentration technique showed alciano philia with 0.1 M, 0.2 M Mg^{++} concentration. These reactions showed presence of sulfomucins.

Alcianophilia was observed with AB pH 2.5. The sequential staining method ABpH 2.5 - PAS showed bluish pink staining. Bluish staining observed with CI and bluish pink with CI - PAS method - Alcianophilia was restored after demethylation at 37°C and 60°C. Hence presence of carboxymucin gets confirmed. Alcianophilia was observed after acid hydrolysis showed probable presence of hyaluronic acid. That gets confirmed after hyaluronidase digestion. After sialidase digestion no change in the alcianophilia hence

absence of sialic acid.

So above all reactions showed presence of neutral mucins, sulfomucins, hyaluronic acid and glycogen and absence of sialic acid.

B) Stroma. The stroma showed intense PAS activity (Fig.2) That was reduced to half prior phenylhydrazine treatment Hence probable presence of acidic and neutral mucins. The PAS reactivity was changed after saliva or amylase digestion, hence presence of glycogen in stroma.

Weak alcianophilia was observed with AB pH 1.0 (Fig. 5). The bluish pink staining was observed with sequential staining method AB pH 1.0 - PAS. These primary reactions showed presence of sulfomucins. Pinkish staining was observed with AF. Methachromasia was observed with all pH levels, with azure A, Staining method. The alcianophilia was also observed with critical electrolyte concentration at 0.1 M Mg^{++} and 0.2 M Mg^{++} concentration level. These results confirms the presence of sulfomucins.

Weak alcianophilia was observed with AB pH 2.5. The bluish pink staining observed with AB pH 2.5-PAS. Bluish staining was observed with CI and bluish pink staining with CI - PAS sequential staining method. The bluish pink staining was also observed with AF - AB pH 2.5. These reactions showed probable presence of carboxymucins. After acid hydrolysis the alcianophilia observed hence

probable presence of hyaluronic acid, which was confirmed after hyaluronidase digestion. Alcianopholia gets reduced after sialidase digestion indicates presence of sialic acid. Hence in the stroma presence of glycogen, neutral mucins, sulfoucins and hyaluronic acid, sialic acid.

III) Histological Observations of Cancerous Ovary. For the present investigation the serous papillary adenocarcinoma was identified according to books -

1) Evan's Histological Appearnce of Tumour by David J.B. Ashley.

2) Gynecological And Obstetrical Pathology with clinical and endocrine relations by Emil Novak.

In the serous papillary adenocarcinoma, the cystic ovary gives rise to many papillae like out growths. This serous adenocarcinoma consist of vascular intricate villus like, simple as well as complex papillane. This was observed in HE prepared technique of paraffin sections; as shown in Plate No.7, Fig.4. The papillary cells showed varying number of mitosis. The papillae may be single layered or mulltilayered. The cells showed very scanty cytoplasm, almost whole cell gets occupied by prominant nucleus. Sometimes the section showed some gland like structures, but not gland but area of cystic ovary.

IV) Histochemical Observations In Cancerous Ovary. For histochemical observations the cells of the papillae were taken into consideration. The all histochemical observations are given in table 6.

This site showed intense PAS activity (Fig.5) That intensity was not decreased after saliva digestion hence absence of glycogen in it. The staining intensity was slightly decreased prior to phenylhydrazine treatment, hence probable presence of acidic mucins detected.

There were no alcianophilia with AB pH 1.0, hence absence of sulfomucins. No alcianophilia observed with critical electrolyte concentration at 0.1, 0.2, 0.4 M Mg^{++} concentration. Hence absence of sulfomucins in cancerous ovary.

Very poor alcianophilia observed with AB pH 2.5 (Fig. 6) and bluish pink staining with AB pH 2.5 - PAS. These primary reactions showed probable presence of carboxymucins. That also observed with CI and bluish pink staining with CI - PAS. Bluish pink staining observed with AF-AB pH 2.5. After acid hydrolysis no alcianophilia hence absence of hyaluronic acid and presence of sialic acid. The sialic acid presence confirmed after sialidase digestion. No effect of pepsin digestion on alcianophilia.

So in cancerous condition neutral mucins, and sialic

acid present and absence of glycogen, sulfomucins and hyaluronic acid.

DISCUSSION

The present investigation on mucosubstances in normal ovary and cancerous ovary, undertaken with a view to augmenting the mucopolysacchrides. The most of the literature on cancerous ovary and normal ovary did on its histology. There were less investigation did on mucosubstances of ovary normal and that was after carcinoma. This chapter was undertaken to augment the general understanding of secretion and nature of mucosubstances and its probable role in pathological condition.

1. Histochemical Reactivity of Normal Ovary. The mucosubstances observed in the normal ovary under investigation showed mostly the same staining affinities to those exhobited in other organ systems of vertebrates and invertebrates. In the PAS reactivity, modification in PAS activity by phenylhydrazine and diastase digestion; alcianophilia, methachromasia, critical electrolyte concentration methylation, demethylation as well as enzyme digestion. There ware no any special type of mucosubstances.

2. Distribution of Mucosubstances In Normal Ovary. Histologically the normal ovary showed germinal

epithelium and stroma. The germinal epithelium showed intense PAS activity. The acidic mucopolysacchrides were also present which was shown after AB pH 1.0, ABpH 2.5, CI and AF staining methods. Hence in the germinal epthelium there were presence of neutral mucins, sulfomucins, hyaluronic acid, sialic acid and glycogen.

In the stroma of ovary also showed presence of neutral, sialic acid, hyaluronic acid, sulfomucins and glycogen.

3. Comparative Distribution of Mucosubstances In Normal Ovary.

This chapter deals with the past literature and present investigation. Here the point of intrest was only the germinal epithelium as the cancerous condition taken for investigation was arises from germinal epithelium. There was very less attention paid for the mucosubstances. Harter (1948) and Millo et al (1969) demonstrated glycogen in the germinal epithelium of rat. From the present investigation we found presence of glycogen in ovary of woman. Most of the work did on submammalian animals. Konecny (1965) studied past pubertal oogenesis in cats and observed the incorportation of radiosulfate in in superficial layers of the ovarian cortex from this Konecny concluded that the renewal of the cells of the surface (germinal) epithelium takes place in the past pubertal period and

sulfomucopolyscchrides may have an important role in morphogenetic process in the ovary. Present investigation showed presence of neutral mucin, glycogen, sulfomucins, hyaluronic acid and sialic acid in vagina of women.

4. Distribution of Mucosubstances In Cancerous Ovary.

For the present investigation the serous papillary adenocarcinoma was taken. It was purely arises from germinal epithelium of ovary. The papillary cells showed staining reaction with PAS. There was alteration in staining intensity prior to phenlhydrazine treatment, and no decrease after diastase digestion. There was very weak alcianophilia with AB pH 2.5, which gets abolished after sialidase digestion hence there was presence of only neutral and sialic acid and absence of sulformucins,hyaluronic acid

5. Comparative Distribution of Mucosubstances In Cancerous Ovary.

There were very scanty literature available about mucosubstances in malignant ovary. According to Rodriguez Rico et al (1976) there was low amount of mucopolysacchrides. Acid mucosubstances are often totally absent. Faken et al (1981) studied diagnostic method as mucosubstances in pancrease, ovary, and prostatic cancer.' The periodic acid Schiff method is helpful in diagnosis. There was positive reaction in adenocarcinoma for PAS. Histochemical study was done by H.C.Cook (1973). There were neutral mucins in thyroid, gastric tumour and sialomucin in brest and

ovary tumour. In the present investigation we also find presence of neutral and sialic acid in ovarian cancer. According to Kurseladze A.I. (1979) the mucoid substance is rich in sialic acid in ovary carcinoma.