A CORRELATIVE STUDY OF BREAST LESIONS WITH SPECIAL REFERENCE TO FNAC, TRUCUT BIOPSY AND AGNOR SCORING

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"No academic endeavour is single handedly accomplished. This work is no exception" – Anonymous

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CERTIFICATE

This is to certify that the dissertation entitled **"A CORRELATIVE STUDY OF BREAST LESIONS WITH SPECIAL REFERENCE TO FNAC, TRUCUT BIOPSY AND AGNOR SCORING"** presented herewith by ***Dr.VINUTA MALAICHAMY*** to the faculty of pathology, The Tamilnadu Dr. M.G.R. Medical University, Chennai in partial fulfillment of the requirement for the award of M.D. degree in Pathology is a bonafide work carried out by her under my direct supervision and guidance.

Professor and Head
Department of Pathology
Madurai Medical College
Madurai.

Dedicated

To

My parents

my husband and my son

INTRODUCTION

Humans have been plagued by diseases throughout the history of civilization. Medical science has conquered many of the infectious diseases that formerly destroyed large populations. But cancer is still considered as the worse disease of non-epidemic kind. It is notorious in the sense that it can affect anyone without any regard to race or religion, rich or poor, from young to old alike.

In humans and other mammals, the breasts form a secondary sexual feature of females and are the source of nutrition for the neonate, although they are also present in rudimentary form in males. It is both ironic and tragic that malignant neoplasms arising from this organ, readily accessible to self-examination and clinical diagnosis, continue to be a heavy toll among women. The incidence of female breast cancer is rising rapidly between the ages of 35 and 50. Invasive ductal carcinoma comprises the largest group of malignancy ie.65% - 80% of breast carcinoma[73].

Carcinoma breast can be proved by pathological examination of breast tissue only. Fine Needle Aspiration biopsy is a safe procedure that is diagnostically superior to trucut needle biopsy[50]. An accuracy rate of 96.5% was obtained[28]. AgNOR study done in histologic specimen showed high AgNOR count in malignant lesions than benign lesions[41].

"EARLIER THE DIAGNOSIS, BETTER THE PROGNOSIS"

Having this in mind, the present study has been conducted to assess the accuracy of fine needle aspiration biopsy, trucut biopsy and to evaluate and compare AgNOR score in breast neoplasms.

AIM OF THE STUDY

1. **To statistically evaluate the occurrence of breast lesions in and around Madurai.**

2. **To find out the Clinicopathological correlation wherever possible.**

3. **To assess the diagnostic accuracy of the various procedures adapted for pathological evaluation of breast lesions.**

4. **To evaluate and compare the AgNOR score in benign and malignant neoplasms of breast.**

REVIEW OF LITERATURE

EMBRYOLOGY

The breast is a highly modified sudoriferous gland that develops as ingrowths from the ectoderm to form the alveoli and ducts. Supporting vascularised connective tissue is derived solely from mesenchyme. Each mammary gland develops as ingrowths of ectoderm and initiates a primary bud of tissue in underlying mesenchyme. Each primary bud initiates the development of 15 to 20 secondary buds or outgrowths. In the fetus, epithelial cords develop from the secondary buds and extend of into the surrounding connective tissues of the chest wall. Lumina develop in the out growths to from lactiferous ducts with prominent branches[77].

ANATOMY AND DEVELOPMENT

Located within the superficial fascia of the anterior thoracic wall, the breast is composed of 15 to 20 lobes of glandular tissue of the tubuloalveolar type. Fibrous connective tissues connect the lobes; adipose tissue is abundantly interposed between the lobules. The mature breast of the female extends from the level of the second or third rib to the inframammary fold at approximately the sixth or seventh rib. Transversely, it extends from the lateral border of the sternum to the anterior axillary or mid axillary line[77].

HISTOLOGY

The mammary gland consists of 15 to 20 lobes, each of which is an individual compound tubulo-alveolar type of gland. The lobule contains groups of small tubules that are lined with cuboidal or low columnar epithelium. These tubules resemble the ducts. More defined tubules are also seen such as a small intralobular duct or a large intralobular excretory duct that emerges from the lobule to join the interlobular duct. The lobules are surrounded by a loose, fine connective tissue the intralobular connective tissue, which contains fibroblasts, lymphocytes, plasma cells and eosinophils[77].

BREAST – TUMOR AND TUMOR LIKE CONDITIONS

The Edwin smith surgical papyrus (3000-25000 B.C) was the first document that referred to carcinoma of the breast. The author of the papyrus concluded that there was no treatment for cancer of the breast.

In the second century A.D., Galen inscribed one of the classic clinical observations, as "The breast tumour exactly resembles the animal crab. In this disease, the veins extending out from the unnatural growth take the shape of a crab's legs[82].

FIBROADENOMA

Deschenes[14] found a prevalence of 8.3/1000 in a population of Canadian women aged 40-59 in the first round of mammographic screening.

Hughes[90] and colleagues have defined four clinical subgroups of fibroadenoma based upon the size of the lump:

1. **Small palpable superficial lesion, which measures less than 5mm in diameter and may remain, unchanged for many years.**

2. **The most common type, making up approx 80% of clinical cases, which grow to 1-3cm in diameter before becoming static.**

3. **Juvenile or Giant fibroadenoma which is rare, undergo very rapid growth to 15-20 cm.**

4. **An uncommon lesion, 10%, measuring 4-5cm, found at any age in perimenarchal and perimenopausal age group.**

Apocrine metaplasia is found in a significant minority of cases with recorded frequency of between 11 and 35%. Scleorsing adenosis has been reported in <10% of fibroadenoma. Squamous metaplasia occurs only rarely.

Extensive areas of myxoid change were found in the rare syndrome (Carney syndrome) described by Carney and Toorkey[9] in which myxomas might also be present in other organs. In Juvenile fibroadenoma, the epithelial proliferation is often florid and even atypical.

FIBROCYSTIC CHANGE

Fibrocystic change affects women between 20 and 50 yrs of age but the majority occurring between 40 and 50 yrs.

Page and Dupont[64] have noted that flattened cysts are larger than apocrine cysts suggesting that cysts lined by flattened epithelium represent the late stage of development in which the active secretory element is no longer present.

Wellings and Alpers[99] stated that no apocrine metaplasia was seen in the 13 to 19 year age group whereas this change was identified is over half of those above 30yrs.

CYSTOSARCOMA PHYLLODES

Muller[90] coined the name phyllodes meaning leaf like appearance. Incidence ranges from 0.3 to 1% of primary breast tumors as reported by Dyer et al[90]. Norris and Taylor[61] who assessed the behaviour of 94 cases in relation to tumour size, nature of tumour margin, degree of cellular atypia and mitotic counts. Pietruska and Barnes[69] based on the assessment of 42 cases defined benign tumors as those with predominantly pushing margins, no or minimal stromal cell atypia and 0-4 mitosis per 10 HPF. Malignant tumour had predominantly infiltrating margins, moderate to marked stromal atypia and 10 or more mitosis/10HPF. They also recognised a borderline group with pushing or infiltrative margins, moderate stromal atypia and 5-9 mitoses per 10 HPF.

Mofatt et al[59] studied 32 cases of phyllodes where, benign, borderline and malignant being 22, 5 and 5 cases respectively. They studied one case of recurrence in borderline and malignant phyllodes.

Symmer[90] in his study in Nottingham found that 68% of the tumours were benign, 16% borderline and 16% were malignant.

DUCTAL CARCINOMA IN SITU

DCIS was first recognized by Bloodgood[6] (1983). Lewis and Geschickter[46] described comedo DCIS, although they failed to distinguish invasive from insitu carcinoma.

Azzopardi[3] highlighted lobular 'cancerization' in DCIS. Page[63] described cribriform, micro papillary and other non-comedo DCIS. All these studies stated that the perimenopausal women were commonly affected.

High nuclear grade in DCIS is closely correlated with large cell size, larger tumour size, and increased grades of intraductal necrosis, increased c-erb/b-2 protein expression, high cellular proliferation, p53 protein expression and absence of ER and PR expression. They show increased incidence to develop invasive carcinoma[90].

LOBULAR CARCINOMA IN SITU (LCIS)

Foote and Stewart[25] (1941) described the histological features and assigned the term lobular carcinoma in situ. LCIS requires (1) Involvement of acini by characteristic cells (2) cells, fill, distend and distort the acini. Incidence is greater before menopause (only <10% postmenopausal).

Page[65] (1991) stated that 2/3 of LCIS would advance to invasive carcinoma within the first 15 years after biopsy. However, Rosen[74] (1978) recorded that invasive carcinoma developed even after 15 years, with the median follow up of 5 yrs.

INVASIVE DUCTAL CARCINOMA

Rosen[74] (1979) accounts that 75% of cancer death in breast cancer is due to invasive ductal carcinoma. According to current opinion ductal as well as lobular invasive carcinoma starts in the Terminal duct lobular unit (TDLU).

Azzopardi[3] stressed, the work of Wellings and colleagues on the point of view of the site of origin of most ductal carcinoma is TDLU. The incidence is common in perimenopausal persons.

INFILTRATING LOBULAR CARCINOMA

Tavassoli[91] (1992) was the first to account that the invasive carcinoma of the breast composed of small cells with a linear growth pattern.

In 19th century Comil and Waldeyer[90] traced them as intra lobular carcinoma. Foote and Stewart[25] (1946) introduced the term LCIS and infiltrating lobular carcinoma from a study of 300 cases.

TUBULAR CARCINOMA

Fisher[23] (1977) first found an uncommon variant where the tumour cells form microtubules and some of them were arranged in cords.

Martinez and Azzopardi[51] (1987) recognized a similar variant within infiltrating ductal carcinoma. Tavassoli[91] (1992) did not accept a tumour forming tubules as a variant of ILC. She regarded them as a tumour displaying both ductal and lobular patterns of invasion.

Mc Divit[53] and colleagues (1982) have pointed out the morphological features of tubular carcinoma. This carcinoma is in fact a readily recognizable subtype composed as it is of distinct tubular structures with open lumina lined by a single layer of epithelial cells. They lack myoepithelial investment. It usually presents with ductal carcinoma in majority of cases.

MUCINOUS CARCINOMA

It is also referred as gelatinous, mucoid and colloid carcinoma and characterized by abundant extracellular mucin surrounding nests of carcinoma cells. Such carcinomas are typically circumscribed. This neoplastic growth pattern is associated with a relatively favourable prognosis, provided that the carcinoma consists almost entirely of mucinous growth pattern[73].

MEDULLARY CARCINOMA

Foote[60] (1949) defined it as a specific type of invasive breast cancer apparently having a favourable prognosis. It is characterized by a circumscribed growth pattern, pronounced nuclear pleomorphism and mitotic activity, a syncytial arrangement of the neoplastic cells and a lymphocyte or plasma cellular infiltrate.

PAPILLARY CARCINOMA

In the past the term papillary carcinoma has been used in a broad sense and should be distinguished from invasive and in situ types.

Fisher[22] (1980) pointed out that it is important in terms of prognosis and management to distinguish the two types. They are frequent in elderly and less than 2% are only symptomatic.

Siranukgul and Tavassoli[83] (1993) identified 9 cases of invasive micro papillary carcinoma and concluded that they were different from that of Ductal NST carcinoma.

However, Luna, More[47] and colleagues who found 27 cases which showed micro papillary differentiation out of 986 cases of invasive carcinoma and they considered this tumour types to be aggressive.

ADENOID CYSTIC CARCINOMA

Azzopardi[3] (1979) accepted an incidence of 1% of these tumours. Lamovec[44] (1989) who found 6 examples of adenoid cystic carcinoma in a total of 5994 cases of breast cancer.

At least 50% of adenoid cystic carcinoma arises in sub and periareolar region. This is the only form of invasive carcinoma, which typically demonstrates myoepithelial participation. There is no lymph node metastasis. The prognosis is very good.

Lamovec[44] (1989) observed neither recurrence nor metastasis in his cases. Peters and Wolff[67] (1992) stated that no patients died of disease but two developed local recurrence after five years of surgery.

CARCINOMA WITH METAPLASIA

The term metaplastic carcinoma covers a heterogenous range of uncommon entities and has been used loosely by many authors to refer to a variety of breast malignancies of mixed epithelial and mesenchymal appearances. Metaplastic carcinoma is currently believed to arise from either myoepithelial or epithelial cells[90].

Ease. V et al[20] (1989) by an immunocytochemical study of 14 cases proved sarcomatoid carcinoma of the breast.

Wargotz ES, Nortis HU[98] (1989) said that metaplastic carcinoma of breast were matrix producing carcinoma meaning that carcinomas with a direct transition from the epithelial to a chondroid or osteoid stromal component without interspersed spindled or giant cell areas.

APOCRINE CARCINOMA

Frable and Kay[26] (1968) found that apocrine carcinoma constitute 1% of mammary carcinoma. Azzopardi[3] (1979) accounted a lower incidence of 0.3% only. He accepted those tumours, which has PAS positive granules as apocrine tumours.

Mossler[52] (1980) found that apocrine carcinomas constituted 4% of their prospective series. Histological definition by Eusebi et al[19] (1986) has pointed out that patterns of apocrine cell differentiation can be seen in practically any type of carcinoma of breast, including papillary and medullary types. Apocrine carcinoma comprises of two intermingled cell types. One (type A) has abundant granular eosinophilic cytoplasm (granules are PAS +ve after diastase digestion). The second (type B) displays abundant cytoplasm in which fine empty vacuoles are seen.

SECRETORY JUNVENILE CARCINOMA

Mc Divitt and Stewart[54] (1966) first reported this carcinoma in children and were later seen in 87-year-old patient also. Norris and Taylor[62] (1970) found 135 tumours in women less than 30yrs old.

Three patterns are present in varying combinations – Honey combed, compact and tubular. Tubules are mostly present in the central fibrotic areas, which are angulated and lined by cuboidal to flat cells. Honeycomb pattern is composed of follicular and micro cystic structures, which give a spongy appearance. The neoplastic cells have a large amount of pale staining cytoplasm[90].

This carcinoma has an extremely favourable prognosis in children and adolescents. Sullivan et al[89] reported first case of recurrent secretory carcinoma. In adults it is more aggressive. Meis[56] (1993) reported axillary lymph node metastasis in adults.

FINE NEEDLE ASPIRATION BIOPSY OF THE BREAST

For the last 160 years exfoliated and abraded samples of cells have been collected from accessible anatomical surface especially from areas like bronchus and cervix[50].

Mean while, Heyden and Menetriers[57] (1883) employed needles to obtain cells and tissue fragments to isolate pneumonic microorgansims and diagnose pulmonary carcinoma respectively.

It was in Europe and particularly Scandinavian country that FNAC as the technique flourished in 1950 and 1960[50].

For example, in one study by Russ[75] (1978) about one in eight breast cancers diagnosed with FNA biopsy were initially considered benign on physical examination.

It has been emphasized by Fisher[22] that FNA biopsy should be performed before surgery to identify lesions requiring excision with margins. Since excision biopsy may destroy the margin of tumour.

FNA biopsy in addition can significantly reduce the number of unnecessary surgeries for benign disease, which can result in scarring and disfigurement of the breast [15, 66].

According to Howell[36] (1993) possible contraindications to its use include multicentric cancers, a tumour larger than 4cm, suspicious, widespread, mammographic calcifications, centrally located tumour, large, pendulous breasts and pregnant or elderly patients.

Grant[31] (1986) studied the following statistics regarding FNA biopsy from summary of 18 reported cases.

Sensitivity	**-**	**92.5%**
Specificity	**-**	**99.8%**
Positive predictive value	**-**	**99.7%**
Negative predictive value	**-**	**94.2%**
Accuracy	**-**	**96.5%**

FNA biopsy is an excellent, cost effective diagnostic modality. The use of FNA biopsy may reduce the cost of diagnosis by as much as 90% compared with hospitalization and excisional biopsy, as indicated in studies by Kaninsky[40] (1984).

Johnson TL, Kini SR[39] (1989) studied that irregular nuclear shape, irregular chromatin distribution, loss of cell cohesion, necrosis and the absence of benign epithelial cells and bipolar nuclei are all features in apocrine carcinoma.

E. Wilkinson et al[18] (1989) analysed 276 aspirates and found out some tumours are less likely to be diagnosed like lobular carcinoma, tubular carcinoma, small cell ductal carcinoma and as well as minimal and in situ cancers.

Layfield et al[45] (1989) pointed out that various forms of degeneration like cystic, haemorrhagic, necrosis and fibrosis can also result in the paucity of diagnostic cells.

Greely, Frost[32] (1997) studied that some ductal carcinomas have equally small relatively uniform neoplastic cells; conversely, lobular carcinoma may have larger cell similar to those of ductal carcinoma NOS.

Sethis et al[80] (1997) pointed out that cells with intracytoplasmic lumina, when present as single or dispersed cells, are a significant observation. They are

generally associated with malignancy, particularly lobular carcinoma in situ and atypical lobular hyperplasia.

Stanley et al[88] (1993) pointed out that smears showing abundant hyaline globules, nuclear enlargements and hyperchromasia allow a confident diagnosis of adenoid cystic carcinoma.

Dabbs et al[13] (1994) studied that cells of some lobular carcinoma have the appearance of signet ring cells and the pleomorphic variant show features more like poorly differentiated ductal carcinomas.

Jeffrey and Ljung[38] (1994) pointed out that it is difficult to distinguish intracystic or intraductal papillary carcinoma from intraductal papilloma or florid papillomatosis as it is not possible to predict if a well differentiated papillary carcinoma is invasive or non invasive.

Venegas et al[96] (1994) studied cytologically that high nuclear grade lesions are biologically more aggressive and necrosis appears to impart a worse prognosis in low to intermediate nuclear grade lesions.

Dutta et al[17] (2001) studied 51 cases of breast lumps by fine needle aspiration cytology of which 28 were malignant and benign lesions were mastitis, fibroadenoma and fibrocystic disease. Diagnostic accuracy of FNAC was 90.2%.

Ribeiro – Silva et al[70] (2001) by fine needle aspiration found out 3 cases of metaplastic carcinoma of breast which is often confused with benign and other malignant entities.

Chaiwum et al[11] (2002) studied 2375 cases of breast lesions by FNA from 1994 to 1999, 48% were benign, suspicious for malignancy were 5%, malignant were 15%, unsatisfactory 32%, Showing sensitivity of 84.4% and Specificity of 99.5%.

Gomez et al[30] (2002) studied total of 30 cases of breast tumour with papillary pattern which are characterized by an abundance of cellular material, three dimensional papillary clusters without fibrovascular connective tissue cores, small papillae arranged in cell walls, tall columnar cells and isolated naked nuclei.

Sneige[86] (2000) studied the cytomorphologic features of in situ proliferative breast lesions and found a spectrum ranging from those of a simple benign lesion to atypical to those indistinguishable from invasive carcinoma.

Bojia et al[7] (2001) studied 120 patients by FNAC and found out 16 patients to have carcinoma and 86 cases were found to have fibroadenoma of the breast. The Sensitivity was 94.3% and Specificity was 78.6%. The high sensitivity and specificity results obtained ascertain that FNAC is the most reliable diagnostic method.

Ustun et al[95] (2002) studied cytological features of FNAC of the breast from 21 patients with proven LCIS which were described and compared with surgical specimens. Aspirates from 8/21 cases had cell groups diagnostic for or compatible with LCIS. Two cases turned out to be invasive lobular carcinoma. Other 11 aspirates were benign.

Mansoor, Jamal[49] (2002) studied the efficacy of breast FNA in 72 cases that were having both FNA cytology and follow up histology diagnosis. The Sensitivity was 98.4% and Specificity was 60%.

Ariga et al[1] (2002) compared the diagnostic accuracy of fine needle aspiration of clinically suspicious palpable breast masses in women younger and older then 40 years of age. A total of 1158 FNA's performed between 1982 and 2002 in women with palpable breast masses. The Sensitivity was 98% and Specificity was 99% overall.

Young et al[102] (2002) pointed out that breast carcinomas were correctly identified as malignant and the values of exact diagnosis were 65% for ductal carcinoma, 20% for lobular adenocarcinoma, 12% for medullary carcinoma and 27% for mucinous variety. This study shows that FNAC of the breast is the reliable method for the diagnosis of breast carcinoma.

Hardisson et al[34] (2003) by FNA reported four cases of solid papillary carcinoma of the breast. Mean age was 66 years. Cytology demonstrated

moderately to highly cellular smears with irregular groups of predominantly monolayered epithelium composed of small, polygonal or cuboidal cells with eosinophilic cytoplasm and rounded eccentrically placed nuclei.

AGNOR TECHNIQUE IN HISTOPATHOLOGY OF BREAST CARCINOMA

Cell kinetics plays an important role in tumour behaviour. Proliferation rates of the tumour can be assessed to determine the behaviour of a particular tumour. The cell cycle can be divided into four phases based on the nuclear chromatin activity. They are S, G1, G2 and Go phases. There is a short resting phase of the cell undergoing replication at the 'S' phase. G2 is the second resting phase before active mitosis. Thus, the DNA content at the end of 'S' phase in an indicator of proliferative activity AgNOR detects the DNA content at this stage[92].

During the phase of active DNA replication, strips of DNA containing RNA genes are seen inside the nucleolus. These DNA fragments are actively transcribing with the help of polymerase I enzyme. They are considered as ribosomal factories, cell cycle in controlled by a few enzymes called M phase promoting factors[92].

This factor has two subunit proteins

1) **34 kd protein, which is the product of the CDC2 gene.**
2) **45 kd subunit consisting of proteins, which accumulate during cell cycle and are destroyed by the end of mitosis.**

These proteins are named as 'cyclins'. The inactivation of these cyclins inactivates M phase promoting factors also and so, this protein is required to end mitosis in a cell cycle.

Nucleolar organizer regions (NOR) are loops of DNA that transcribe ribosomal RNA. They are located in the acrocentric chromosomes 13, 14, 15 and 21. At ultra – structural level, NORs are termed fibrillary centers and can be demonstrated by means of in situ hybridization to localize ribosomal genes. NORs are associated with certain proteins. There NOR Associated proteins (NORAPs) are Polymerase I C_{23} (nucleolin) and B_{23}. These proteins are thought to play a role in RNA transcription[93].

NORs were first visualized in 1975 by means of a simple silver staining technique that recognizes these argyrophilia-associated proteins. AgNORs appear as black dots in the nucleus. The AgNOR content is expressed as mean AgNOR count for 50-100cells. After DNA synthesis, in the G_2 phase of the cell cycle a diploid nucleus might contain 20 AgNORs. In practices the AgNORs are so closely packed that it is difficult to distinguish one AgNOR from another. The argyrophilia of the NOR associated protein (NORAPs) acts as a marker of ribosomal DNA and possibly reflects its transcriptional phase. The number of visible AgNORs therefore indicates the current phase of transcription[92].

Interest has been focused on seeking reliable guide to patient's prognosis by measuring the cell proliferation and correlating these data with the tumour's

potential for metastases and recurrence because the behaviour of a tumour is generally thought to reflect its growth rate. One of the methods of measuring the proliferation rate is the AgNOR technique, which determines Nucleolar Organiser regions (NOR) associated proteins. The nuclei of malignant cells contain NOR of a significantly different number and size from those in normal, reactive or benign neoplastic cells[92].

The NORs attracted lot of attention because of the claims that their frequency within the nuclei is significantly higher in malignant cells than in normal, reactive or benign neoplastic cells. Their potential value in diagnostic histopathology was because they can be easily demonstrated in routinely processed tissue sections.

A quantifiable apparent increase in the mean AgNOR count of a cell population in tissue sections resulted if:

1. **Cell proliferation was so active that nucleolar dissociation was present in many cells, when the AgNOR were seen throughout the nucleus.**
2. **There was a defect of the nuclear association, which resulted in AgNOR dispersion.**
3. **Cell ploidy increased, resulting in a real increase of AgNOR bearing chromosome.**
4. **Transcriptional activity increased resulting in the prominence of otherwise inconspicuous AgNORs.**

Benign cells show only 1-2 AgNOR per nucleus, which was attributed to the difficulty in perceiving the individual AgNORs, when they aggregated within a relatively small nucleus. In malignancy, or with increased cell proliferation, AgNORs get dispersed throughout the nucleus to a varying extent, enabling the histologist to count them more readily. Therefore, the quantification of AgNORs depends on the degree of dispersion or disaggregation of the relatively large number of AgNORs in the nucleus. One great advantage of this technique is that, previously stained cytology slides can be reused for silver staining, thus providing an excellent guide to the diagnosis especially in doubtful cases and when extra-unstained slides are not available.

The major disadvantages are:

1. **The counting procedures adopted are usually manual and hence long and tedious.**
2. **Observer error is the major cause of inaccuracy and inconsistency.**
3. **The dots of AgNOR interphase nuclei need not always correspond actively to the number of such types in the karyotype[93].**
4. **Overlap and coalescence may result in misjudged counts[92].**

Ultra – Structural appearance of Mammalian Nucleoli

a. **Granular component composed of distinct spherical shaped granular each out 15nm in diameter.**

b. **Dense fibrillar component consisting of lightly packed electron dense 3-5nm thick fibrils.**
c. **Fibrillar center – consisting of a loose network of fibrils with a little greater average diameter 4-8nm than the dense fibrillar component.**

Canepa et al[8] (1993) studied nucleolar organizing region as a prognostic factor in infiltrating ductal carcinoma of breast. They analysed 170 cases of infiltrating ductal carcinoma over a period of 138 months and found out that those cases with average score below 9.5 showed favourable prognosis and above 9.5 had unfavourable outcome.

Roller et al[71] (1993) studied nucleolar organizer regions in human breast cancer. They examined the number of AgNOR'S in 56 cases of malignant breast lesions and 20 being cases using routinely fixed material. They could find out a clear-cut difference between benign breast diseases as compared with breast carcinoma by high AgNOR counts.

Yoshida et al[101] (1994) studied a clinicopathological evaluation of NOR proteins in human breast cancer. They pointed out that AgNOR count was low in small sized tumours i.e., less than 2 cms, than large ones, i.e., more than 2 cms. The group with recurrence had a higher count i.e., >9. AgNOR is useful in evaluating cell proliferative activity and helps to predict postoperative recurrence.

Basu et al[5] (1995) studied a total of 72 cases, which were ductal carcinomas. The AgNOR scores showed a tendency to increase with higher grades of malignancy. Their mean AgNOR count was (16.63 ± 7.09).

Simha et al[82] (1996) studied the prognostic value of argyrophilic nucleolar organiser regions (AgNORs) in breast lesion. AgNOR counts correlated with tumour size, mitosis and desmoplasia. ER/PR negative tumours showed a tendency for high NOR counts.

Kumar et al[43] (1997) evaluated in 46 patients that the AgNOR count was significantly higher in breast carcinoma (6.61 ± 1.75) and found better correlation with the increase in the size of the tumour, stage of the cancer, number of lymph nodes and tumour recurrence.

Mehrota A, Chandra T[55], (1998) determined the significance of AgNOR counts in FNAC smears and corresponding paraffin section by using an one step colloidal staining method. AgNOR counts were significantly higher in FNAC smears in malignant neoplasms in comparison to paraffin sections in the same groups of cases.

Sinha SK et al[84] (1998) evaluated the cytomorphological features (nuclear grade and smear pattern) and AgNOR counts in 60 cases of carcinoma breast. AgNOR staining and C-erb B-2 immunostaining were also done in each case. Significantly 23 postmenopausal cases with carcinoma breast, the tumour cells

were positive for c-erb b-2 and an association was found between the nuclear grade and high AgNOR counts.

Ceccarellic et al[10] (2000) found that AgNOR protein was a proliferation related parameter that could be used as a prognostic indicator in breast tumour pathology. They also found that there was significant association between AgNOR protein quality and tumour prognosis.

Hasnan and Jayaram[35] (1996) adopted the argyrophil technique for staining NORs in FNA cytological smears of 56 breast lesions comprising 31 benign and 26 malignant cases. From their study, they concluded that AgNOR score of 5 and less strongly favours benign lesions whereas a score of above 5 could be in favour of a malignant lesion.

Kruger et al[42] (2000) investigated to what extent analysis of silver stained nucleolar organizer regions is cell cycle dependent in breast cancer and to assess the prognostic value of an AgNOR analysis that takes into consideration the cell cycle status of tumour cells. In comparison to the noncycling tumour cells, cycling ones exhibited significantly higher AgNOR numbers (mean values 3.84 ± 1.09 vs 2.40 ± 0.78 per nucleus).

Khanna et al[41] (2001) studied 73 patients (46 malignant and 27 benign) with breast lumps. In all cases FNAC samples and histologic specimen were

studied by conventional and silver staining for AgNOR. AgNOR count was 6.94 ± 2.74 in FNAC and 6.57 ± 2.73 in histology of malignant tumours.

MATERIAL AND METHODS

Of the 2, 11, 201 patients treated in Govt. Rajaji Hospital, Madurai from July 2003 to August 2005 there were 816 cases of breast tumours. Of these 407 were benign and 409 were malignant. The present study evaluates various parameters including incidence of the breast tumours and their diagnostic accuracy by means of FNAC and Trucut biopsy. AgNOR score is calculated for benign and malignant tumours. The following types of breast biopsy materials were received from our hospital.

1. **FNAC**
2. **Trucut biopsy**
3. **Lumpectomy**
4. **Mastectomy**
5. **Re-excisional Biopsy.**

While the FNAC smears were received fixed either in 95% ethyl alcohol, rectified spirit or isopropyl alcohol, the rest of the biopsy materials were received fixed in 10% buffered formalin. Trucut specimens were subjected to histopathological examination in toto. Lumpectomy specimens received were

examined for their size and dimension. Regarding mastectomy specimens, the macroscopic examination included overall size of the specimen, dimensions and appearance of skin with measurements of scars or incisions, appearance of nipple and areola, presence of muscle and axillary tissue and location of any distinct palpable lesion. The specimens were cut into thin slices (about 5mm) and then examined by careful visual inspection and palpation. With breast carcinomas, the size, contour and consistency of the tumour were noted. Also, its location, including the distance from the nearest surgical margin was noted.

Samples for histologic examination were taken from the tumour (especially around its periphery to appreciate in-situ changes and invasion), nipple, skin quadrant (to assess intramammary spread of tumour and multicentricity). The axillary fat was dissected for lymph nodes and all the nodes were subjected to study. The tissue slices were processed in various grades of alcohol and xylol. Paraffin blocks were subsequently prepared and thin sections cut to obtain optimum results. All the biopsy materials were stained routinely with Hematoxylin and Eosin[4].

AGNOR STUDY IN BREAST NEOPLASMS

Tissue sections of benign and malignant tumours were analysed and 55 samples were selected for AgNOR study and comparison. These include both biopsies and mastectomy specimen.

METHODS

The formalin fixed, routinely processed and paraffin embedded tissue blocks were collected.

AgNOR staining[4] was done using the one step silver – colloid technique.

PREPARATION OF STAINING SOLUTION

Solution A: 2% gelatin in 1% formic acid

Solution B: 50% aqueous silver nitrate solution

WORKING SOLUTION

One part of solution A mixed with two parts of solution B.

STAINING METHODS

Sections were cut at 3-5 mm thickness from the blocks. They were dewaxed in xylene and they hydrated to double distilled deionised water.

The sections were exposed to freshly prepared working solution for 60 minutes in the dark at room temperature.

The silver colloid was then washed off with deionised water.

The sections were dehydrated through alcohol, cleared in xylene and mounted using DPX mounting medium.

COUNTING PROCEDURE[92]

Nucleolar organizer regions in silver staining are observed as black dots within the nucleus, which may remain discrete or in aggregates. Rest of the nucleus stains pale yellow.

The standardized approach proposed was followed in counting AgNOR. 100 nuclei were assessed in each slide and mean number of dots per nucleus was determined by light microscope using an oil immersion (100x) objective.

There are two approaches to count AgNORs.

Firstly, all silver stained structures could be counted, but when lying in groups each cluster (almost aggregated or partly disaggregated nucleoli) treated as one structure [Type 1 method].

Secondly, where AgNORs can be separately seen within a nucleolus, each AgNOR could be counted as a unit, together with the smaller AgNORs seen outside the Nucleolus (Type 2 method). Both methods have been followed in our study.

$$\text{AgNOR INDEX} = \frac{\text{Mean NOR count in tumour nuclei}}{\text{Mean NOR count in the nuclei of Non neoplastic breast tissue}}$$

A total of 100 nuclei is randomly chosen from both tumour part and Non-neoplastic breast tissue in each sections, were evaluated and NOR index calculated.

Statistical method used for comparison studies of AgNOR scores is mention below.

TEST OF SIGNIFICANCE[27]

$$Z = \frac{\bar{x}_1 - \bar{x}_2}{S\sqrt{1 + 1}}$$

$\overline{n_1}$ $\overline{n_2}$

FINE NEEDLE ASPIRATION CYTOLOGY

The cytological materials were obtained in the form of smears, which were fixed in 95% alcohol for PAP and H & E staining. The aspiration syringes used were 10-25ml and the needle size between 22-23 gauges. Standard methods of staining were employed[4].

Appropriately labeled specimens were subjected for microscopic examination. The slides from all the specimens were studied and placed into one of the following categories viz. Benign, malignant, tumor like lesions, and inflammatory lesions. Table 1 shows Peterse's criteria of benign and malignant breast cytology. Amongst these, the breast tumours were analysed with FNAC and trucut biopsies for benign and malignant nature and the significance of these diagnostic procedure were evaluated. The sensitivity, specificity and accuracy of these diagnostic procedures were calculated by using the formula shown in Table 2.

Photographs of specimens and photomicrographs of slides are represented at relevant areas in discussion. The classification of mammary neoplasms used in this study is based one the one adopted by the WHO[97].

OBSERVATION AND RESULT

INCIDENCE OF BREAST TUMOURS

In the two-year study period from August 2003 to July 2005, 32, 421 specimens were received from the Govt. Rajaji Hospital, Madurai. Of these, breast tumour accounted for 816 cases. Hence, the overall incidence of breast tumours as per the attendance of cases is 2.51%.

The number of benign and malignant breast tumours was 407 and 409 respectively. Thus, the incidence of benign tumours of the breast 49.8% and the incidence of malignant breast tumours 50.1%.

Diagram 1, shows the year wise distribution of breast tumours during the study period. In the academic year 2003 – 2004 benign cases were 217 and malignant were 189. In the year 2004 – 05 benign cases were 190 cases and malignant cases were 220.

AGE INCIDENCE

The distribution of breast tumours according to age is shown in Diagram 2 while the benign breast tumours had a peak incidence 46.6% in the third decade, the peak incidence (38%) of malignant breast tumour is seen in the fifth decade.

SEX INCIDENCE

Of the total 816-breast tumour, 813 cases occurred in female (incidence of 99.6%) whereas only 3 cases (4%) occurred in males as shown in Table 3. It is clear from the illustration that all the cases seen in male were malignant breast tumour.

BENIGN TUMOUR AND TUMOUR LIKE LESION OF THE BREAST

46.8% of the breast tumours encountered in the study were benign. Table 4 lists the various types of benign breast tumours and their incidence with regard to the total number of breast tumours. It is obvious that fibroadenoma is the most common breast tumour and comprised 30.14% of the benign breast tumours (Fig 1, 2).

MALIGNANT TUMOUR OF THE BREAST

Of the 32,421-biopsy material analysed in the two-year study, there were 409 cases of malignant breast tumour with an overall incidence of 1.26%. During the two years period the overall incidence of breast tumour is 2.51%. The incidence of benign tumours of the breast is 49.8% and the incidence of malignant tumours is 50.1%.

Table 5 depicts the decennial incidence of malignant breast tumours compared to the total number of malignant tumour that was encountered in each decade in this Institution.

SIGNS AND SYMPTOMS

The incidence of various signs and symptoms is shown in Table 6. 90.5% of the cases sought treatment with complaints of lump in the breast, which in many cases was painless. 12 patients (2.9%) noted a bloody nipple discharge 9 cases (2.2%) reported with ulceration. 18 cases (4.4%) reported with painful lump.

CLINICAL STAGING OF CARCINOMA OF BREAST

The clinical staging system jointly adopted by the International union against – cancer (UICC) and American joint commission on cancer staging (AJC) is followed in the study and depicted in Annexure IV. Table 7 shows the percentage of patients in each stage. In the current study 47 cases were in stage I, 70 cases were in stage II, 62 cases were in stage IIIA, 15 cases were in stage IIIB, 5 cases were in stage IV and 73 cases were unstaged.

HISTOLOGIC DISTRIBUTION OF MALIGNANT TUMOUR

Table 8, shows the incidence of the various histological types of malignant breast lesions. In the current study, 2 cases were ductal carcinoma in situ (Fig 3, 4), 389 cases were invasive ductal carcinoma (NOS), 4 cases were invasive lobular carcinoma (Fig 7, 8), one case was invasive ductal carcinoma with predominant intraductal carcinoma (Fig 5, 6), 1 case was medullary carcinoma 10 cases (Fig 9, 10) were colloid carcinoma (Fig 11, 12) and 2 cases were Paget's disease of nipple (Fig 13, 14).

CYTODIAGNOSIS

Pre-operatively FNAC was performed on 607 patients of these only 532 yielded cellular aspirates of which 260 were positive for benign lesions (48.8%) (Fig 15, 16, 17), 272 were positive for malignancy (51.1%) (Fig 18, 19, 20, 21), 73 were cases negative for malignancy, 1 was false negative (0.18%) and 1 was false positive (0.18%).

TRUCUT BIOPSIES

Trucut biopsies were performed on 280 cases, which were suspected for malignancy of which 256 had the required yield. All the 256 cases were positive for malignancy (100%) (Fig 23, 24). The sensitivity & specificity obtained for this diagnostic procedure was 95.8% and 100% respectively. The Accuracy of the test was 96%.

AgNOR SCORE

Of the total 816 cases in this study, 35 cases were selected as shown in Table 9. All the selected sections stained by AgNOR staining technique were carefully examined under light microscope with oil immersion objective. The nuclei were stained light yellow and the nucleolar organizer regions were seen as black coloured dots within the nuclei (Fig 25, 26, 27). The numbers of black dots were counted in 100 nuclei in each section and the mean AgNOR value for each case was determined as shown in Table 10.

LYMPHNODE METASTASIS

Out of total 409 cases, axillary node involvement was found clinically in 262 cases (64%) of these 212 cases (81%) were proven positive histologically (Fig 22, 28) and one node showed granulomatous reaction (Fig 29). Of the remainder, sinus histiocytosis was found in 27 cases (10.3%), 23 cases (9%) were found to be reactive nodes.

DISCUSSION

Sidney J.Cutler[81] who analysed data from the Connecticut tumour registry and California tumour registry revealed that there has been an increase in the number of diagnosed cases of breast cancer per lakh women younger than 55 year of age. This was more than balanced by improvement in patient survival, resulting in a decrease in mortality from breast cancer.

During the early 1980s mammographic screening was introduced, and the number of women of appropriate age undergoing screening increased steadily to current reported rates of 60% to 80%. In 1994, after a lag of time of about 10 years, the mortality rate started to decline[90].

Glauco Frizzera[29], advisor American cancer society comments that the incidence of breast cancer over the past 50 years showed 3 distinct phases.

1. **Between 1940 & 1982 – steady annual rate of increase of 1% per year.**
2. **Between 1982 & 1987 – incidence was 4% per year.**
3. **Between 1990 + 1994 – The incidence appear to statistic at approximately 110.2 cases / 10,000 women.**

SEER[79] cancer statistics review 1973 – 1999 shows the breast cancer incidence from 1994 to 2000 as 379 cases / 1,00,000 women.

The current study also shows an increase incidence in this beginning of the millennium compared to the past 3 decades. Technically, the increased incidence has been ascribed to the increased use of mammography. Moreover, the rise in prevalence of breast cancer risk factors may also be responsible for the apparent increase.

CLINICOPATHOLOGICAL CORRELATION

Benign Breast Neoplasm

Fibroadenoma and fibrocystic disease of breast present mostly as a palpable mass. Fibroadenoma is the one of the commonest causes of a lump in the female breast accounts to about 10 cases / 1000 women[90]. The current study has an incidence of 30.14%.

Malik R and Bharadwaj[48] studied the lumps in breast of males and females. Of the lumps in females 89% were benign and of these 49% were reported fibrocystic diseases of breast. Jamal AA et al[37] quoted that the incidence of benign breast disease was 85.3%. In the current study, the incidence of benign breast disease was 49.8%.

FIBROCYSTIC CHANGE

Fibrocystic change affects women between 20 and 50 yrs of age but the majority occurring between 40 and 50 yrs.

Page and Dupont[64] have noted that flattened cysts are larger than apocrine cysts suggesting that cysts lined by flattened epithelium represent the late stage of development in which the active secretory element is no longer present.

Wellings and Alpers[99] stated that apocrine metaplasia was not found in the 13 to 19 year age group whereas this change was identified in over half of those above 30yrs.

CYSTOSARCOMA PHYLLODES

Microscopically the two key features of phyllodes tumour are stromal hypercellularity and presence of benign glandular element as an integral component of the neoplasm[73].

Pietruszka et al[69] have subdivided these tumours into benign borderline and malignant groups based on histologic features. An important diagnostic criterion for malignant overgrowth of the glands by the sarcomatous stroma, so that, low power views of the tumour show only stroma without epithelial elements. Presence of tumour necrosis and heterolgous stromal elements signifies an adverse prognosis[90]. In the current study, the incidence of malignant phyllodes was 0.24%.

Metastasis, which is restricted only to malignant lesion, shows stromal elements only. However, recurrances can occur with both types and simulate

original tumour multiple recurrence of a borderline neoplasm can get differentiated and produce metastasis.

Muller[90] coined the name phyllodes meaning leaf like appearance. Incidence ranges from 0.3 to 1% of primary breast tumors as reported by Dyer[90] et al. Norris and Taylor[61] who assessed the behaviour of 94 cases in relation to tumour size, nature of tumour margin, degree of cellular atypia and mitotic counts. Pietruska and Barnes[69] based on the assessment of 42 cases defined benign tumors as those with predominantly pushing margins, no or minimal stromal cell atypia and 0-4 mitosis per 10 HPF. Malignant tumour had predominantly infiltrating margins, moderate to marked stromal atypia and 10 or more mitosis/10HPF. They also recognised a borderline group with pushing or infiltrative margins, moderate stromal atypia and 5-9 mitoses per 10 HPF. The current study 13 cases of Cystosarcoma phyllodes (1.5%). Of which 1 case was malignant.

Mofatt et al[59] studied 32 case of phyllodes where, benign, borderline and malignant being 22, 5 and 5 cases respectively. They studied one case of recurrence in borderline and malignant phyllodes. Symmer[90] in his study in Nottingham found that 68% of the tumours were benign, 16% borderline and 16% were malignant.

MALGINANT BREAST NEOPLASMS

Non-invasive carcinoma

In the late 1960's Gallagher and Martin published the results of their whole organ section studies and affirmed the transition that established a stepwise evolution of invasive breast cancer from benign epithelium through in-situ and subsequent invasive stages[90].

This recognition allowed them to coin the term 'Minimal Breast cancer' in which was included DCIS, LCIS and minimally invasive cancers smaller than 5 cm. These with proper therapy would translate into a 10-year cure probability of 90% or more[90]. Fisher et al[22] quoted a high incidence of DCIS tumours i.e., 5%. The current study which has an incidence of 5% is lower than the other study performed in India Viz. Usha[94] et al. Table 11 source the percentage of patients in each stage, and illustrates the result of two studies conducted by BCDDP (Breast cancer detection demonstration project) and SEER[79] (surveillance, Epidemiology and End Results-conducted by USA's National Cancer Institute). In all these studies the number of malignant cases with FNAC proof has been taken into account. The result of the BCDDP study fairly correlated with present study, whereas the SEER project reports higher incidence of patients in stage II (57%) compared to 37% in the present study. This can be explained by the fact that breast cancer specific screening programs are a common place in the USA whereas it is not economically feasible in developing countries like India. As a consequence more number of patients, present in higher stages. That the Indian women are more socially inhibited also contributed to the delay in diagnosis. It is therefore clear that the increased awareness and mass-screening program for

breast cancer in our country has aided detection of many pre-invasive malignant tumours. A comparative analysis of incidence encountered by Fisher et al[22] and Usha et al[94] is also furnished (Table 12). While it was satisfying to note that in most cases all the three studies represented a similarity in range of incidence, Fisher et al[22] study depicts a higher incidence of the non-invasive cancer. The screening programs in the west has helped to detect many cancers in their pre-invasive stage thereby accounting for the apparent increase in non-invasive cancers and the corresponding decrease of invasive cancers. The most common histological type in accordance with the results of other studies was invasive ductal carcinoma (N.O.S.).

Based on Scott et al's recent classification of DCIS one case was found in intermediate grade with cribriform pattern and focal necrosis and the other case was low grade with low nuclear grade and absence of necrosis. When the carcinoma is confined to the ductal system without violation of the basement membrane, axillary lymph node involvement is unlikely[90].

Invasive carcinomas

Tumour in this category are all those in which stromal invasion in detectable, whether an in situ component is identifiable or not and regardless of the relative proportion of these two components. The classification of invasive breast carcinoma in corporate a wide range of criteria such as cell type, type and amount of secretion, architectural feature and pattern of spread[90].

Invasive ductal carcinoma – Not otherwise specified (IDC – NOS).

This comprises the largest group of malignant mammary tumour. Consisting 65% - 80% of breast cancer[73]. In the current study it comprises about 95.1%. WHO defines this entity by exclusion as the most frequently encountered malignant lesion of the breast, not falling into any of the other categories of invasive mammary carcinoma. These are also designated generically as the classic or ordinary type. They are seen throughout the age range of breast carcinoma and most common in mid to late fifties[73].

Grossly, most tumours were of the comedo type and had an irregular stellate (Crab-like) outline with yellowish grey, hard, gritty cut surface often exhibiting yellow flecks of elastin. Areas of necrosis, hemorrhage and cystic degeneration were often seen. A minority of tumours had grossly circumscribed margins that are presumed to have a better prognosis. Histologically, the neoplastic cells were seen either in diffuse sheets, well-defined nests, and cords or as individual cells. There was a variable amount of glandular or tubular differentiation. Some showed necrosis within the ducts and micro calcifications. Some tumours showed growth patterns varying from solid sheets devoid of stroma to those with abundant, densely fibrotic to cellular stroma, the so called 'scirrhous type'. Foci of squamous metaplasia and clear cell changes were seen. A lymphoplasmocytic infiltration was present at the interphase between tumour and stroma in many cases. Many tumours showed invasion of perineural spaces,

lymph vessel and blood vessel, which represent unfavourable prognostic findings. Several studies show nipple invasion in 23% to 31% of IDC (NOS) cases[90]. In the current study 120 cases of IDC (NOS) showed invasion of the nipple (29.3%) and in a large majority the tumour was located in a zone less than 2.5 cm from the nipple.

INVASIVE DUCTAL CARCINOMA WITH PREDOMINANT INTRADUCTAL COMPONENT

Azzopardi et al[2] who propounded this concept that was incorporated in the recent WHO classification, describe this as, the carcinoma which were overwhelmingly intraductal and contain only small foci of stromal invasion. It was suggested that only those tumours in which the intraductal carcinoma was at least 4 times greater than the invasive component, should be included. The current study encountered one such tumour with an incidence of 0.2%.

INVASIVE LOBULAR CARCINOMA (ILC)

Foote and Stewart[25] established the term ILC in 1941 with the publication of the classic paper on this carcinoma. The reported incidence of has varied from 1% - 14%[51]. The current study quotes to0.9% and the median age at diagnosis was between 45 and 56 years, which is in accordance with other studies where it was 52 years. Grossly, a classic ILC forms a firm to hard tumour with irregular borders. Another gross manifestation is formation of innumerable fine, hard nodules. A desmoplastic stromal reaction, linear arrangement of carcinoma cells "Indian – File pattern" and their tendency to grow in a circumferential fashion around ducts and lobules (targetoid growth) were the peculiar diagnostic features of classic ILC, emphasised by Foote and Stewart[25]. The tumour cells are small and medium sized, uniform in staining properties and exhibit relatively little irregularity. Clues for diagnosis on FNAC include a sparsely cellular sample, small cells with scanty cytoplasm dispersed singly or linear / Indian file arrays.

MEDULLARY CARCINOMA

Medullary carcinoma constitutes 7% of breast tumour in several studies with a mean age ranging from 46 to 54 years. The current studies show compatible data with an incidence of 0.24% and mean age of 48 years. Grossly, these tumours have well-circumscribed margin firm in consistency with a lobulated or nodular cut surface. The extent of necrosis if present is directly related to tumour size. Foote and Stewart[25] described a constellation of definitive histopathologic features, which include

1. **Lymphoplasmacytic reaction: This must involve at least 75% of the periphery and be present diffusely in the substance of the tumour**
2. **Microscopic circumscription: Edge of the tumour should have a smooth, rounded contour that appears to push side rather than infiltrate the breast**
3. **Syncytial growth pattern of most of the tumour growth (>75%)**
4. **Poorly differentiated nuclear grade**
5. **High mitotic rate.**

These tumours have an overall lower frequency of axillary node involvement and have a good prognosis even in stage II. When the tumour resembles medullary carcinoma with >75% syncytial growth pattern, but lacks all of the necessary features, which has been designated as 'Atypical' medullary carcinoma.

MUCINOUS CARCINOMA

This type, also known as colloid mucinous or gelatinous carcinoma, is characterised by large amount of extracellular epithelial mucin, recognisable microscopically, surrounding and within tumour cells[73].

Pure mucinous carcinomas contain at least 50% of the tumour growing in a mucinous pattern and with extracellular mucin constituting at least 33% of the lesion. This criterion helps to differentiate from focal mucinous differentiation found in other carcinomas in which the prognosis depends on the latter[73]. Pure forms have a very low incidence of local metastasis and a better prognosis. Grossly these tumours exhibit a circumscribed margin often accentuated by red to purple zone of congested parenchyma and a gelatinous cut surface. Synder et al[87] found an overall incidence of pure forms as 2% with a greater mean age at diagnosis then mixed forms. The current study parallels with an incidence of 2.4% an mean age of 53 years.

INFILTRATING DUCTAL CARCINOMA WITH PAPILLARY PATTERN

WHO defines this as a rare carcinoma in which invasive pattern is predominantly in the form of papillary structures. The same architecture is usually displayed in the metastasis. Frequently foci of intraductal papillary growth are recognizable[24]. In the present study 4 cases have been reported with an incidence of 1% and a mean age of 53 years.

PAGET'S DISEASE OF NIPPLE

Sir James Paget described this disease more than a century ago[90]. The incidence in Fisher's[22] study and in the current study in 2.3% and .48%. Clinically it presents as a weeping eczema – like lesion centered in the nipple and later involving the areola. Microscopically, Paget's cells, which are large, clear cells with typical nuclei are seen within the epidermis, usually concentrated along the basal layer. These cells may occur singly or in clusters and typically contain mucin or rarely melanin granules[73]. An associated carcinoma is seen in more than 95% of these cases, which have virtually all been intraductal carcinoma with an invasive component[73]. The current study reveals only 2 cases (0.48%) of which one had an invasive ductal component.

MALE BREAST CANCER

Breast Carcinoma is an uncommon neoplastic condition among men, accounting for not more than 1% of all breast cancers[73] (0.1% in this study). The mean age at diagnosis is 60 yrs (55 years in the current study). The gross features are similar to carcinoma of female breast. Approx 85% of male breast cancer are of the invasive ductal carcinoma (N.O.S) type (100%), one showing carcinoid pattern and other two showed moderately or poorly differentiated carcinoma[73]. Unfavourable Prognostic factors regardless of nodal status are tumour size greater than 2cm and poor histologic differentiation[90].

FNAC AND TRUCUT BIOPSIES STUDY

Romanidis[72] quoted that FNAC done for breast cancer had sensitivity 99% and specificity (96%) in their study. Franzen and Zajicek[50] who have a very long experience at the Karolinska Institute reported that 80% of carcinomas were definitely diagnosed 10% equivocal and another 10% missed in other words, false negatives.

Most studies reveal an average sensitivity of 87% and specificity of 100%. In the current study the sensitivity rate of FNAC was 99.6% & specificity of 98.6% for both benign and malignant cases as shown in Table 13. The accuracy for benign cases was 99.4% & for malignant cases was 95.1%.

In the current study total number of cases analysed for FNAC was 607. 260 cases were reported as benign disease of the breast. False positivity was reported for one case and false negativity for one case. Sensitivity rate obtained was 99.6% and specificity rate as 98.6%. The accuracy of FNAC in benign breast neoplasm accounts to 99.4%. In the current study, the trucut biopsy analysed subsequently for 502 cases of which 221 cases where diagnosed as benign neoplasm, 256 cases diagnosed as carcinoma one case was false negative and one case was false positive. Sensitivity was 95.2% specificity was 99.3% accuracy was 95.1%. As many tumours on which a trucut was performed were greater than 2.5cm in diameter, it accounted for the high index of sensitively. The overall sensitivity rates in various series have ranged from 75% to 90%.

Ariga et al[1] quoted a 99% sensitivity, and 99% specificity in their study of FNAC in palpable breast masses. The accuracy of FNAC was 99%

Bojia et al[7] in his study reported that the sensitivity of the FNAC and excision biopsy of breast lesion was 94.3% and specificity as 78.6%, accuracy rate as 84.3%.

Mansoor et al[49] reported in their study the sensitivity, specificity and accuracy as 98.4%, 60%, 93% respectively. Zhong et al[103] quoted 97.3% as sensitivity, 97.7% as specificity and 97.4% as accuracy in their study of 951 cases of FNAC smear study.

Grant[31] concluded with the following statistics regarding FNAC quoting sensitivity (92.5%) specifying 99.8% accuracy – 96.5%. Dutta et al[17] studied that the diagnostic accuracy of FNAC was 90.2% in their study. Chaiwun et al[11] reported in their study that FNAC had provided a sensitivity of (84.4%) & specificity (99.5%) Yong et al[100] in their study obtained an accuracy of 90% for FNAC and 67% accuracy for Trucut biopsy in surprises breast lumps.

Scopa et al[78] quoted in their study the sensitivity, specificity and accuracy of FNAC as 90%, 100% 94% respectively. For Trucut biopsies the sensitivity rate was 89%, specificity as 100%, and accuracy as 90%.

In Comparison with the above-mentioned studies and the current study, the sensitivity specificity and accuracy of the FNAC, Trucut and excisional biopsy quoted in the current study are higher than that of other studies. In the current study the sensitivity index of FNAC was 99.6%, which in higher than the sensitivity index of trucut biopsy in 95.2%. The diagnostic accuracy of FNAC was 99.4% which in higher than the accuracy of Trucut (95.1%).

This ascertains that the FNAC has a better diagnostic accuracy then the trucut biopsies. The reason behind FNAC being more accurate is as follows.

1. **Adequacy of smeared material**
2. **FNAC done from a more representative area, as the needle is poked in different directions thus providing sampling from different foci.**
3. **Good Fixation and staining preparation of the smears.**
4. **Efficient reporting system.**

False positive diagnoses were avoided, as the interpretive errors were negligible in the hands of experienced cytopathologists. Carcinomas that have the highest potential for false negative diagnosis include those showing extensive fibrosis or extracellular matrix with decreased number of malignant cells. By having these in mind, false negative diagnosis was avoided.

AGNOR SCORE IN BENIGN AND MALIGNANT BREAST TUMOURS

Roller et al[71] in their study found out that malignant breast lesions had an increased AgNOR score than the benign breast diseases. Basu et al[5] found a mean AgNOR score of 16.63 ±7.09 in their study on malignant cases.

Hasnan, Jayram[35] concluded that AgNOR score of 5 or less strongly favours benign lesions whereas a score above 5 could be in favour of a malignant lesion.

The current study showed no significant difference in AgNOR score between ductal epithelial cells of fibroadenomas and fibrocystic disease. There was significant difference in AgNOR score between DCIS and infiltrating ductal carcinoma. It was observed that the AgNOR score progressively increased with infiltrating ductal carcinoma. It has been observed that Type 2 method of counting have higher AgNOR scores than Type 1 method of counting. The average of the AgNOR index of all benign cases was3 and Malignant was 9.

$$\text{AgNOR index} = \frac{\text{Mean AgNOR counts in tumour nuclei}}{\text{Mean AgNOR counts in the nuclei of Non-neoplastic breast tissue}}$$

In the current study, Type 1 counting method showed less value of AgNOR score then the Type 2 method. So the values obtained by Type 2 method have been considered in this discussion. The AgNOR score of 17 cases of benign breast diseases is 3.2 ± 1.2. The AgNOR index was 3.

The AgNOR score for malignant tumours was 7.9 ± 3.1. Ductal in situ stage of the breast cancer showed AgNOR score of 61 ± 1.3. The infiltrating ductal and Lobular carcinoma showed AgNOR score of 9 ± 2.1. The AgNOR index was 6 for DCIS and 9 for invasive carcinomas. It has been observed that AgNOR scores are directly proportional to grading i.e., with increased grades the AgNOR scores also increased and can be used for assessing the clinical outcome and prognosis of the cases. However the AgNOR scores in our study shows a significant high value for malignant than benign.

AgNOR scores revealed that the benign lesions had a value below 3 and the malignant lesions had a score of 7 and above. Statistical significance showed a calculated value of Z (Test of significance-1.781). The average AgNOR score of malignant lesions are certainly higher then those of the benign lesions.

Recalling the work done by Hasnan, Jayaram[35] (1996) it has been found that Crockers type 2 counting method is more useful in grading the tumours than type 1 method. This observation of Jayaram[35] has made out in our study where high-grade tumours have high AgNOR score when compared to low-grade tumours.

SUMMARY AND CONCLUSION

The study indicates that the overall incidence of breast neoplasms is 2.51%.

The incidence of benign and malignant tumours was more or less equal i.e., 49.8% and 50.2% respectively. Benign and malignant breast tumours were most commonly encountered in females. With male patients, malignant breast lesion was commonly encountered.

In females the peak incidence of benign and malignant tumours where at the third and fifth decade respectively. In males the incidence of malignant tumours was in the fifth decade. The complaint for which most patients (96%) sought treatment was a lump in the breast that was painless in most instances.

Among the benign lesions, Fibroadenoma comprised the maximum proportion of cases accounting for 60.4%. In the malignant lesions infiltrating ductal carcinoma (Nos) comprised the maximum accounting for 86%. The non-invasive carcinomas accounted for only 0.5%. Illiteracy and Poverty play a hand-in-glove role in the delay in diagnosis of Pre-invasive breast cancers in Indian women.

Preoperative FNAC and Trucut biopsies have a sensitivity index of 99.6% and 95.8%. The accuracy of these diagnostic tools was 99.6% and 96% for FNAC and Trucut respectively. The accuracy rate of FNA biopsy increases when the cytopathologist performs the FNA biopsy and uses immediate assessment to guide specimen adequacy. This procedure has several advantages.

1. **Provide rapid, accurate diagnosis.**
2. **Serve a cost-effective role in the treatment of breast manes.**
3. **Differentiate cysts from solid tumour and serve as a therapeutic procedure.**
4. **Provide psychological relief of anxiety for patients with benign breast lesions.**

Trucut biopsies and excision biopsy are painful and cosmetically disabling. If done in experienced hands, it avoids false negatives and provides complete histological diagnosis that helps in further management plans. But due to inadequate material and biopsies taken from less representative area, it led to false negative diagnosis. So the accuracy rate of this diagnostic tool is far below that of FNAC in the current study.

AGNOR STUDY

AgNOR scores revealed that the benign lesions had a value below 3 and the malignant lesions had a score of 7 and above. AgNOR study concludes that

the statistical comparison of AgNOR scores is significantly higher malignant lesions than in benign breast lesions.

ANNEXURE I

PROFORMA

Serial No : **Name :**

Biopsy No. : **Age :**

Cyt. No. : **Sex :**

Unit :

Clinical diagnosis

Breast : **Right/Left / Both**

Specimen : **Excisional (Lumpectomy / mastectomy)**

Incisional (Trucut / FNAC)

Macroscopic Examination

Specimen size :

Skin and Nipple :

Tumor Size :

Colour :

Tumor margins : **Circumscribed / Infiltrative**

Posterior margin :

Lymphnode :

Number

Size

C/S

Histological type of lesion: **Fibroadenoma**

Others

DCIS

Invasive ductal carcinoma (Nos)

Invasive lobular carcinoma

Invasive ductal carcinoma with predominant intraductal carcinoma

Medullary carcinoma

Mucinous carcinoma

Paget's disease of nipple

Others

Skin — **Free / Involved**

Nipple — **Free / Involved**

Muscle — **Free / Involved**

Lymphnodes

- **Total number :**
- **No. of nodes involved :**
- **Other Findings :**

F.N.A.C. Findings :

- **Cellularity**
- **Tumour cells**
- **Myoepithelial cells**
- **Inflammatory cells**
- **Others**
  - **Fat cells / Fibrocytes / RBCs**

AgNOR study

- **AgNOR score**

Pathological Staging:

I / II A / II B / III A / III B / IV

ANNEXURE II

W.H.O. CLASSIFICATION OF BREAST TUMORS

I EPITHELIAL TUMORS

A. Benign

1. Intraductal Papilloma

2. Adenoma of the nipple

3. Adenoma

(a) Tubular

(b) Lactating

4. Others:

B. Malignant

1. Non-Invasive:

(a) Ductal carcinoma – in – situ (DCIS)

(B) Lobular carcinoma sin-situ (LCIS)

2. Invasive

(a) Invasive ductal carcinoma (NOS)

(b) Invasive ductal carcinoma with a predominant intraductal component

(c) Invasive lobular carcinoma

(d) Mucinous carcinoma

(e) Medullary carcinoma

(f) Papillary carcinoma

(g) Tubular carcinoma

(h) Adenoid cystic carcinoma

(i) Secretory (Juvenile) carcinoma

(j) Apocrine carcinoma

(k) Carcinoma with Metaplasia

1. Squamous type

2. Spindle cell type

3. Cartilagenous and osseous type

4. Mixed type

(l) Others

3. Paget's disease of the nipple

II MIXED CONECTIVE TISSUE AND EPITHELIAL TUMORS

(a) Fibroadenoma

(b) Cystosarcoma phyllodes (Benign / Borderline / Malignant)

(c) Carcinosarcoma

III MISCELLANEOUS TUMORS

(a) Soft tissue tumors

(b) Skin tumors

(c) Tumors of hematopoietic and lymphoid tissues.

IV UNCLASSIFIED TUMORS

V MAMMARY DYSPLASIA / FIBROCYSTIC DISEASE

a) Duct ectasia

b) Inflammatory pseudotumors

c) Hamartoma

d) Gynecomastia

e) Others

ANNEXURE III

Definition

Sensitivity - **The number of Carcinomas diagnosed and expressed as a percentage of the total number of carcinomas aspirated.**

$$\text{Sensitivity} = \frac{a}{a+c} \times 100$$

Specificity - **The number of correctly identified benign lesions and expressed as a percentage of the total number of benign lesions aspirated.**

$$\text{Specificity} = \frac{d}{b+d} \times 100$$

$$\text{Accuracy} \quad \frac{a + d}{a + b + c + d}$$

TEST OF SIGNIFICANCE[86]

$$Z = \frac{\bar{x}_1 - \bar{x}_2}{S\sqrt{\frac{1}{n_1} + \frac{1}{n_2}}}$$

ANNEXURE IV

TNM CLASSIFICATION OF BREAST CANCER

Primary Tumor (T)

Tx - Primary Tumor cannot be assessed.

To - No evidence of primary tumor

TIS - DCIS / LCIS OR Paget's disease of nipple with no tumor

T1 - Tumor ≤ 2 cm in greatest dimension

T1a - ≤ 0.5 cm in greatest dimension

T1b - > 0.5 - ≤ 1 cm in greatest dimension

T1c - Tumor > 1 cm - ≤ 2 cm in greatest dimension

T2 - Tumor > 2 cm and ≤ 5 cm in greatest dimension

T3 - Tumor > 5 cm

T4 - Tumor any size with direct extension to chest wall or skin

Regional Lymph Nodes (N)

Nx - Regional Lymph nodes cannot be assessed eg: Not removed for study

No - No regional LN metastasis

N1 - Metastasis to movable ipsilateral axillary node (s)

N2 - Metastasis to ipsilateral axillary node (s) fixed to one another or to other structures.

N3 - Metastasis to ipsilateral internal mammary Lymph node (s)

Distant Metastasis

Mx - Distant metastasis cannot be assessed.

Mo - No distant metastasis

M1 - Distant metastasis

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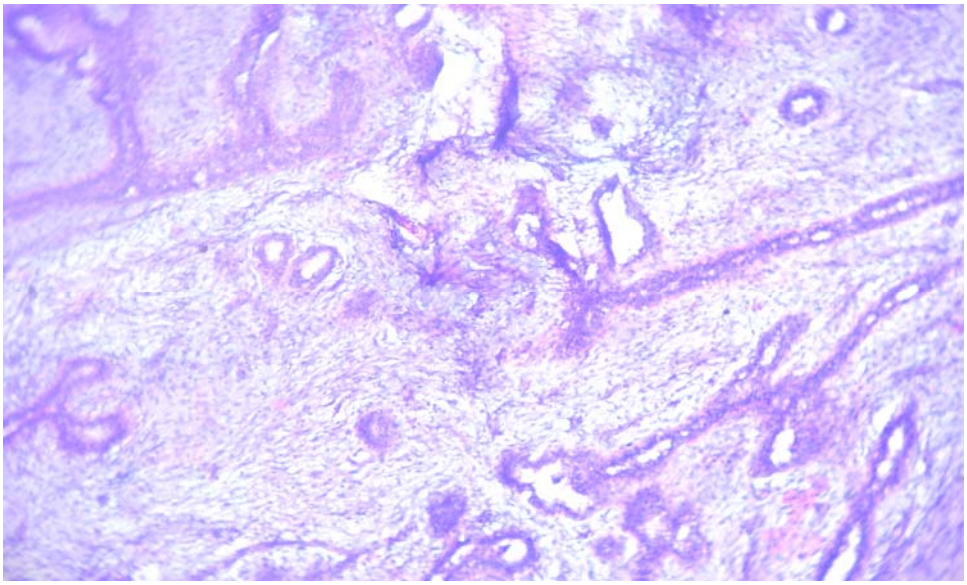


Fig 1 Microscopic appearance of Fibroadenoma intracanalicular type (H&E, x100) Path No.1661/05

Fig 2 Benign Cystosarcoma phyllodes - Gross specimen showing leaf like areas with haemorrhage. Path. No.889/05

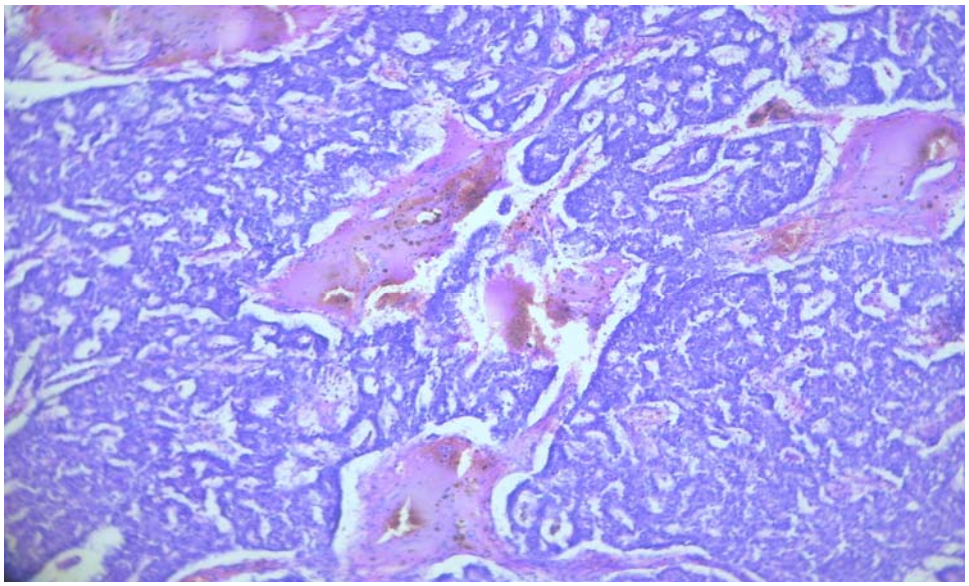


Fig 3 Microscopic appearance of Ductal Carcinoma with cribriform pattern. (H&E, x400) Path No- 505/05

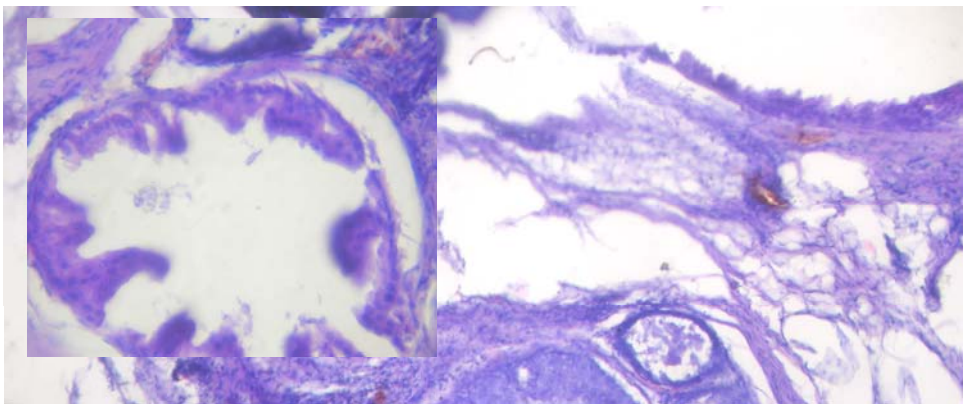


Fig 4 Microscopic appearance of Fibrocystic disease showing few ducts with in situ ductal carcinoma and some ducts showing apocrine changes

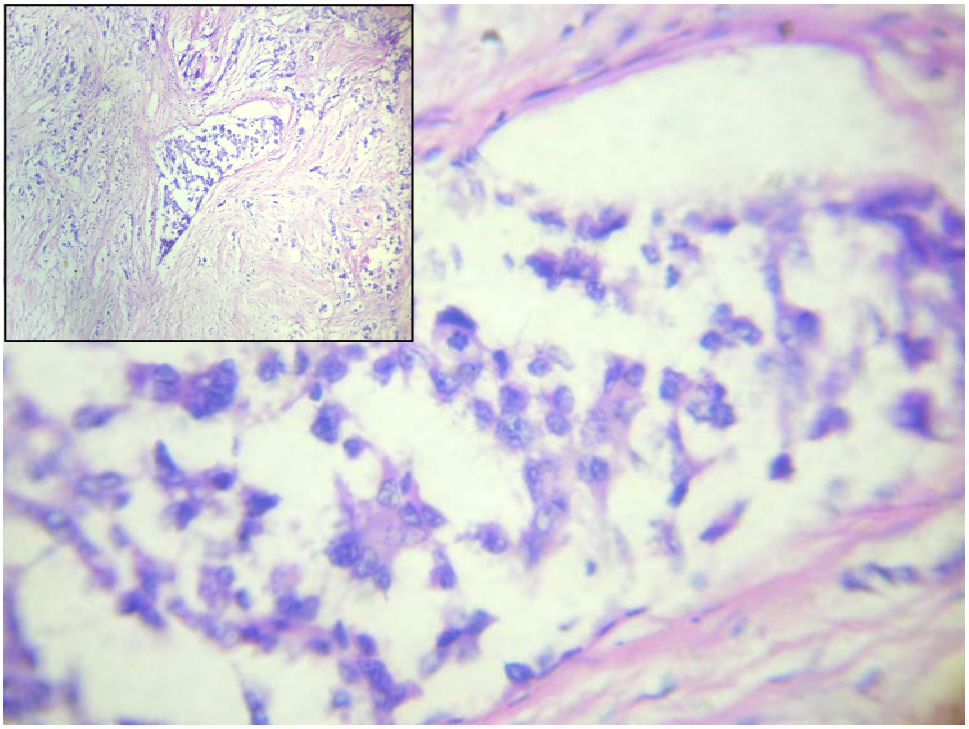


Fig 5 Microscopic appearance of Lymphatic invasion by Carcinomatous cells. Inset shows low power view of involved Lymph vessel. (H&E, x400)Path No.801/04

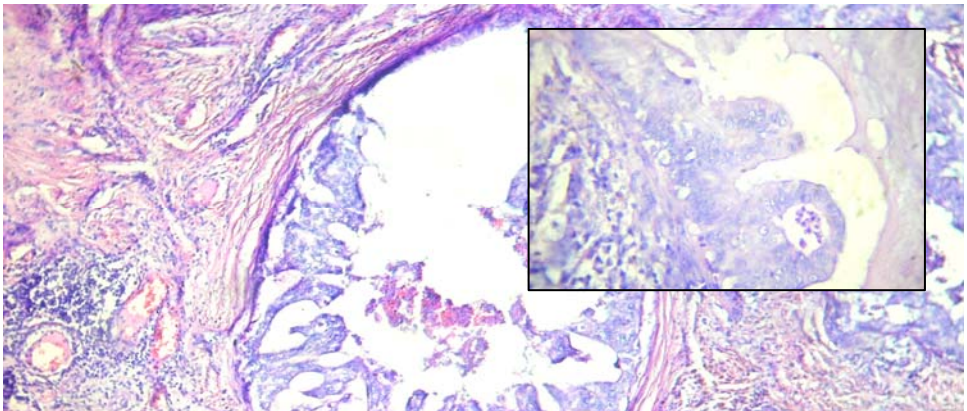


Fig 6 Microscopic appearance of Invasive Ductal Carcinoma showing Predominant Intraductal Papillary Component. Inset shows magnified view of the Papillae. (H&E, x400) Path No- 648/03

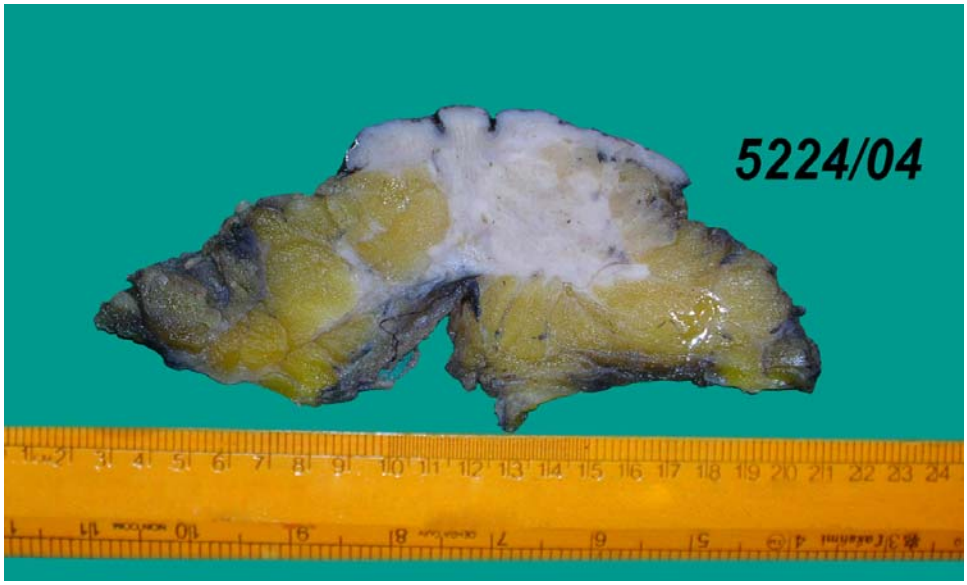


Fig 7 Infiltrating Lobular Carcinoma – Gross showing Grayish white tumour mass and nodules with infiltrating margins. Path. No.5224/04

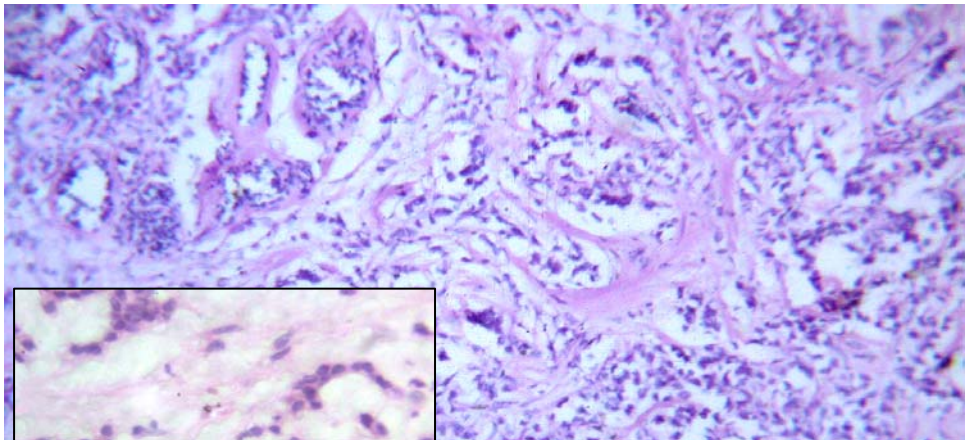


Fig 8 Microscopic appearance of Infiltrating Lobular Carcinoma showing "Indian File Pattern". Inset showing the same. (H&E, x400) Path No-5224/04

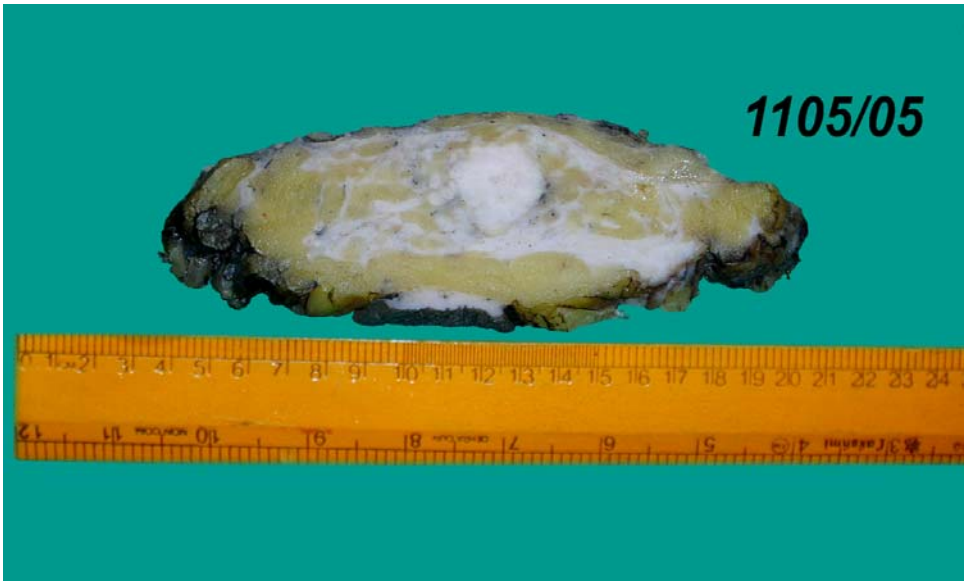


Fig 9 Medullary Carcinoma - Gross showing well circumscribed tumour mass. Path. No.1105/05

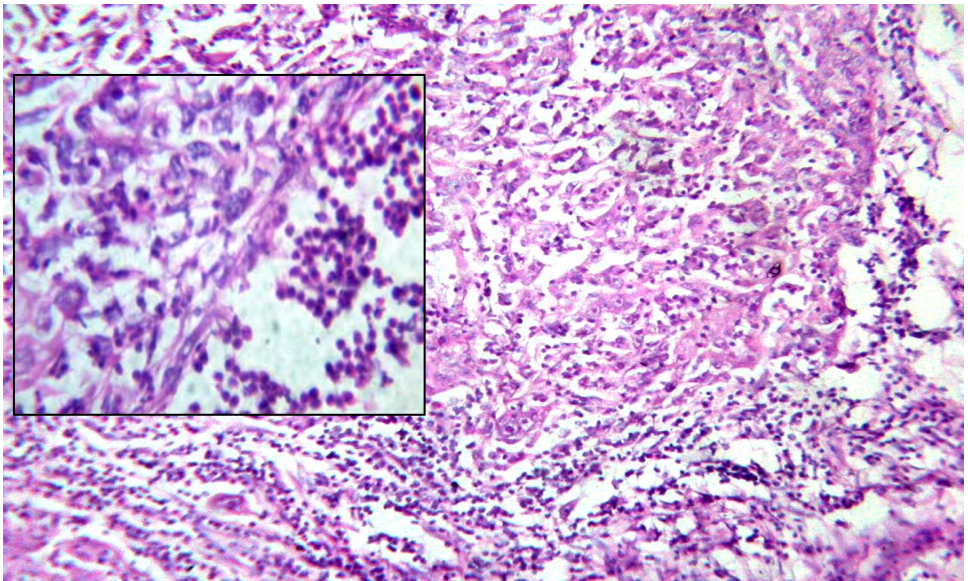


Fig 10 Microscopic appearance of Medullary Carcinoma showing poorly differentiated tumour cells with pushing margins. Inset shows the magnified view of Lymphoplasmacytic reaction. (H&E, x100) Path No:1105/05

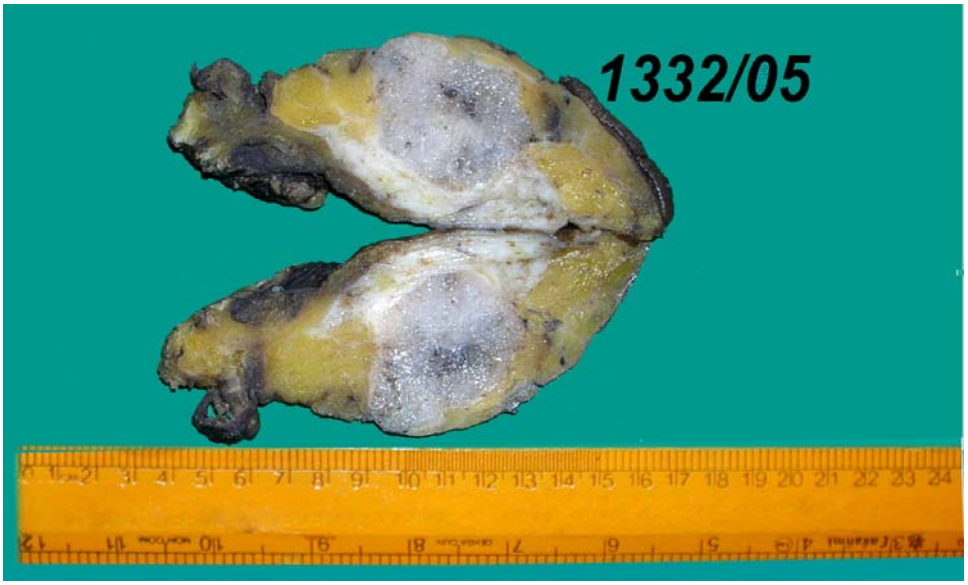


Fig 11 Mucinous Carcinoma - Gross showing tumour tissue exhibiting a circumscribed margin and a gelatinous cut surface. Path. No.1332/05

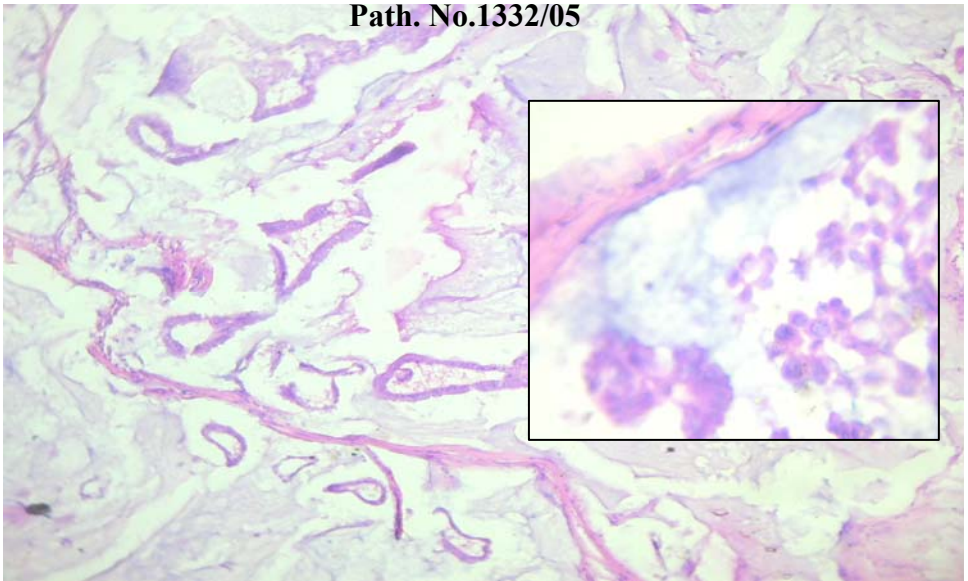


Fig 12 Microscopic appearance of Mucinous Carcinoma showing tumour cells in lakes of mucin. Inset shows the magnified view of tumour cells. (H&E, x100) Path No: 1332/05

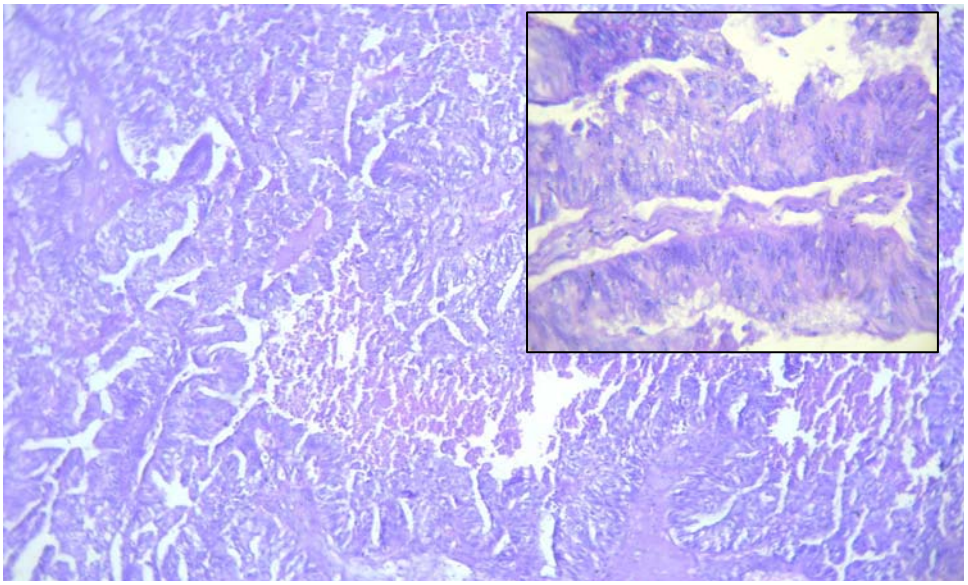


Fig 13 Microscopic appearance of Papillary Carcinoma showing Papillary Structures with Fibrovascular core. Inset shows magnified

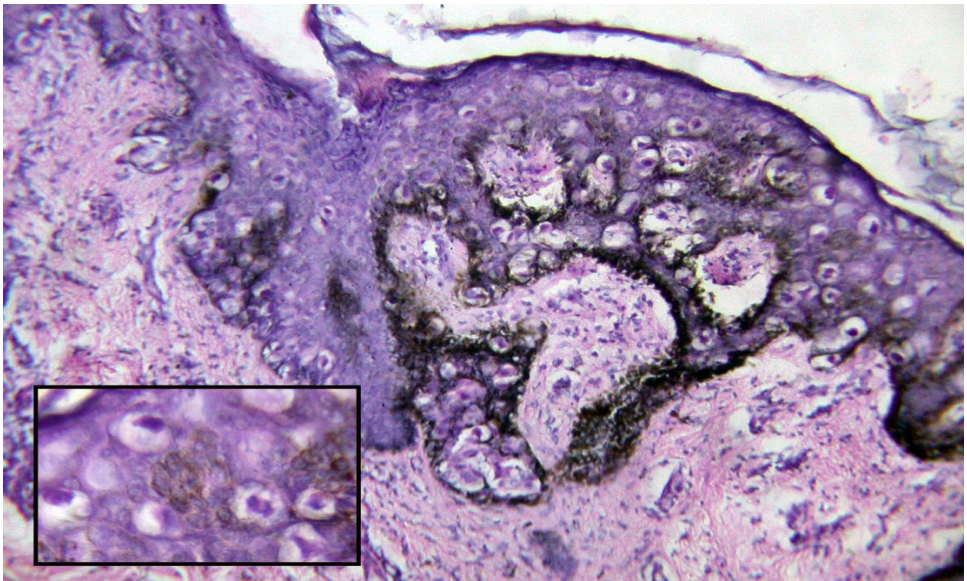


Fig 14 Microscopic appearance of Shows Pagets Disease of Nipple with Intraepidermal location of Pagets cells. Inset shows the magnified view of Pagets Cells. (H&E, x400) Path No: 621/05

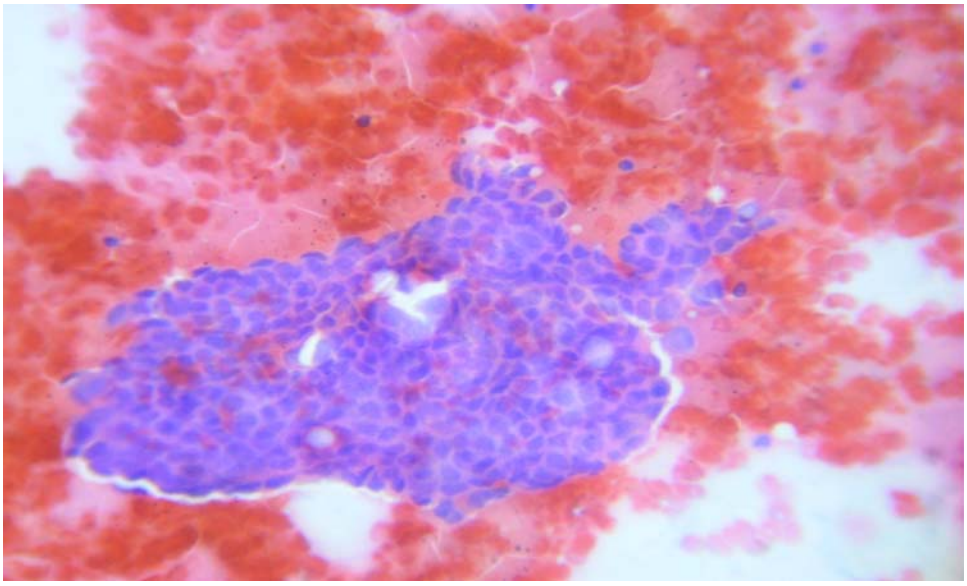


Fig 17 Fibroadenoma - Smear showing tight clusters of uniform ductal epithelial cell in a haemorrhagic background. (H&E, x100) Cyt No: 947/04

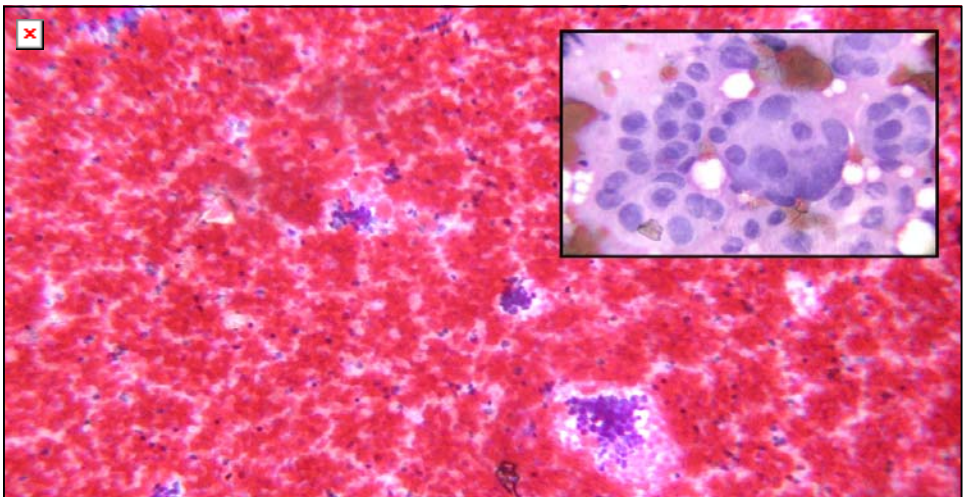


Fig 18 Infiltrating Ductal Carcinoma - Smear showing groups of malignant epithelial cells in a haemorrhagic background. Inset shows magnified view of the same. (H&E, x100) Cyt No. 1541/04

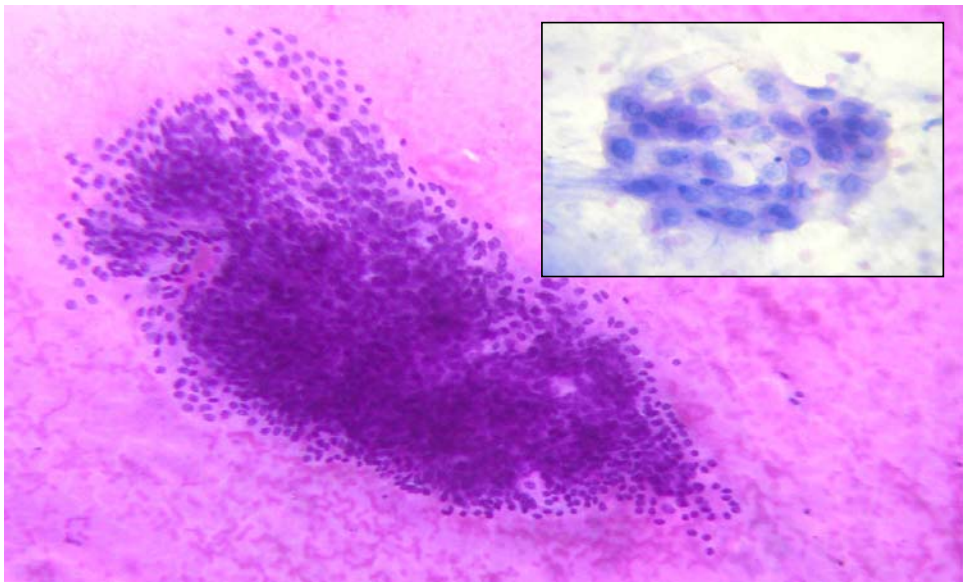


Fig 21 Mucinous Carcinoma - Smear showing aggregates of malignant epithelial cells in a mucinous background. Inset shows magnified tumour cells. (H&E, x100) Cyt No. 101/04

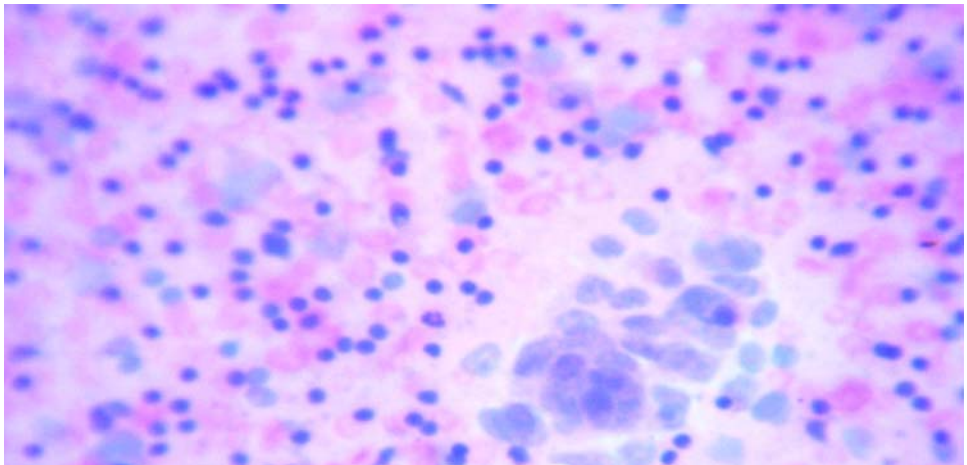


Fig 22 Lymphnode Metastasis - Smear showing Clumps of Malignant epithelial cells admixed lymphocytes and Plasma cells. (PAP, x400)
Cyt No: 1511/04

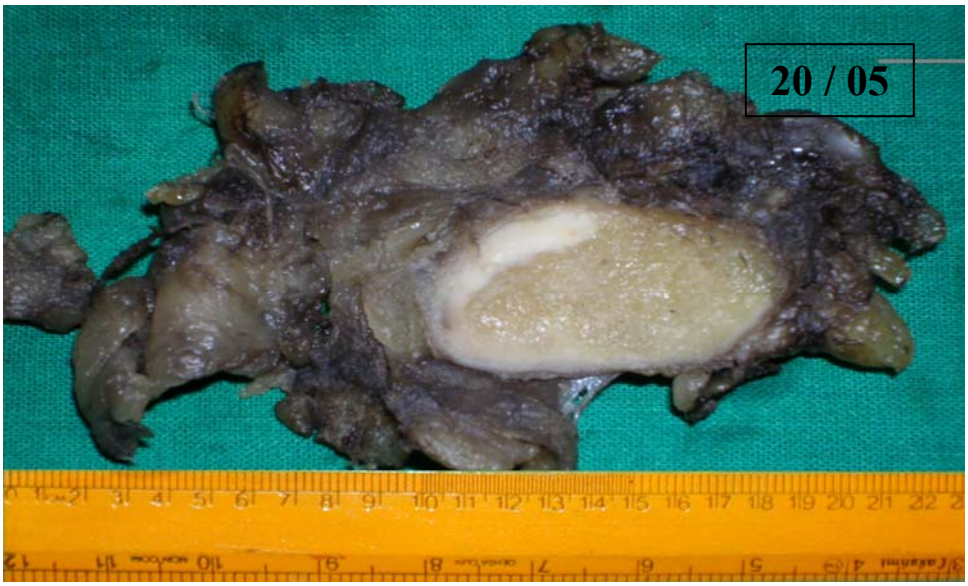


Fig 28 Lymphnode Metastasis - Gross showing subcapsular deposit of Infiltrating Ductal Carcinoma. Path No.20/05

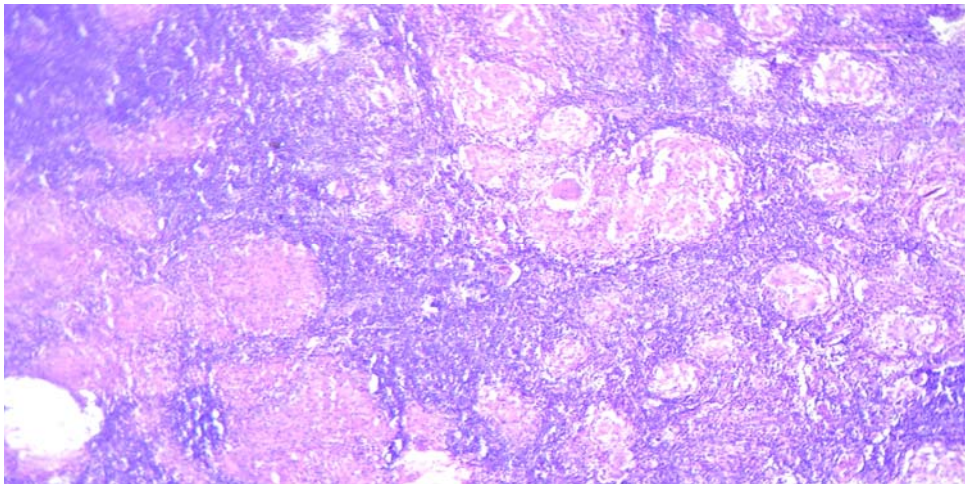


Fig 29 Microscopic appearance of lymphnode with granulomatous reaction to the tumour cells. (H&E, x100) Path No. 1873/04

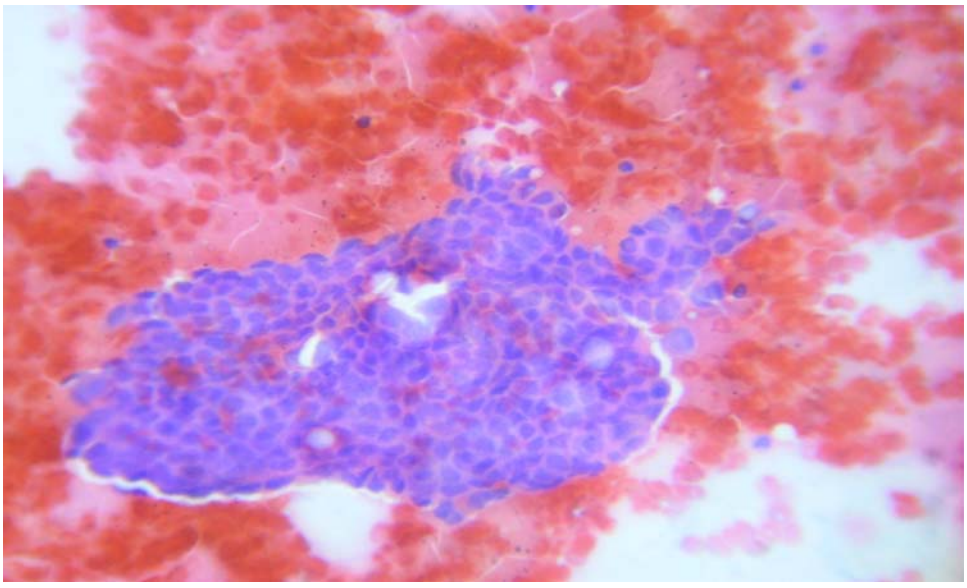


Fig 17 Fibroadenoma - Smear showing tight clusters of uniform ductal epithelial cell in a haemorrhagic background. (H&E, x100) Cyt No: 947/04

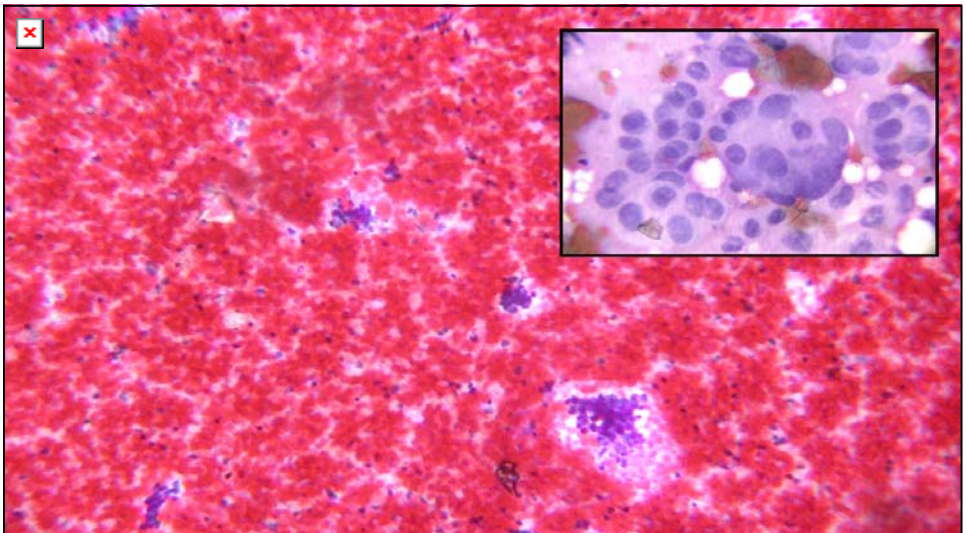


Fig 18 Infiltrating Ductal Carcinoma - Smear showing groups of malignant epithelial cells in a haemorrhagic background. Inset shows magnified view of the same. (H&E, x100) Cyt No. 1541/04

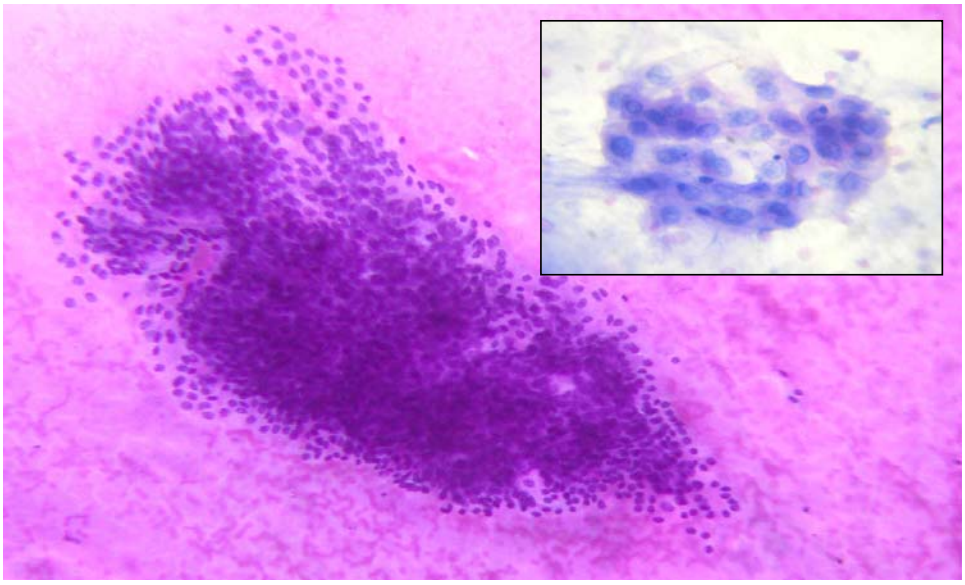


Fig 21 Mucinous Carcinoma - Smear showing aggregates of malignant epithelial cells in a mucinous background. Inset shows magnified tumour cells. (H&E, x100) Cyt No. 101/04

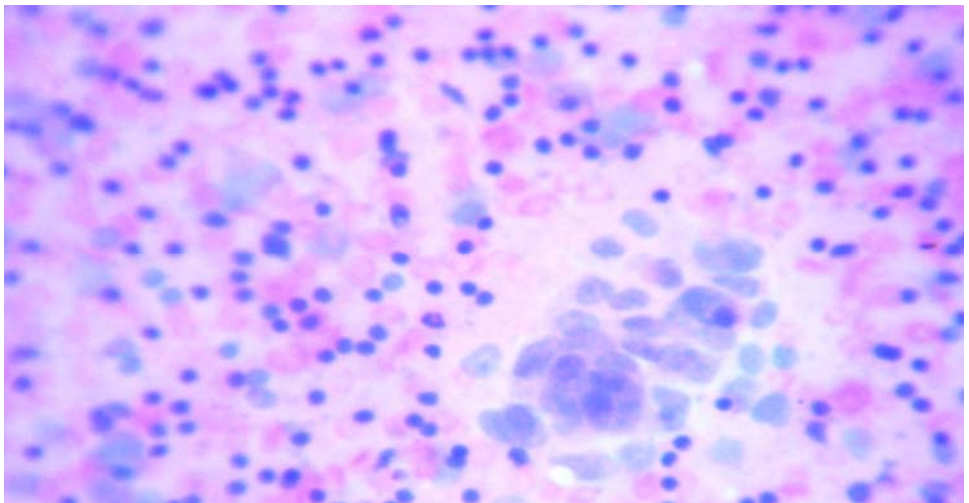


Fig 22 Lymphnode Metastasis - Smear showing Clumps of Malignant epithelial cells admixed lymphocytes and Plasma cells. (PAP, x400) Cyt No: 1511/04

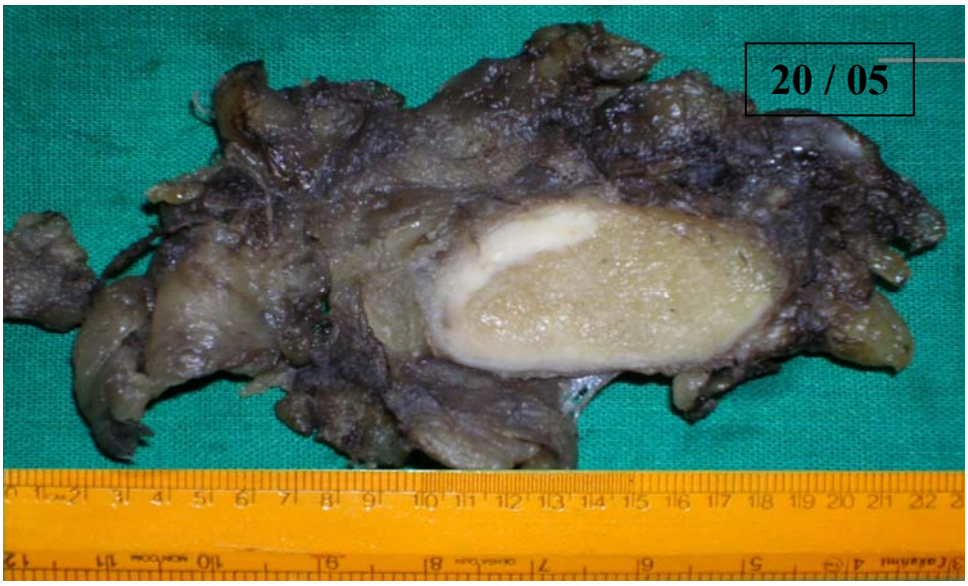


Fig 28 Lymphnode Metastasis - Gross showing subcapsular deposit of Infiltrating Ductal Carcinoma. Path No.20/05

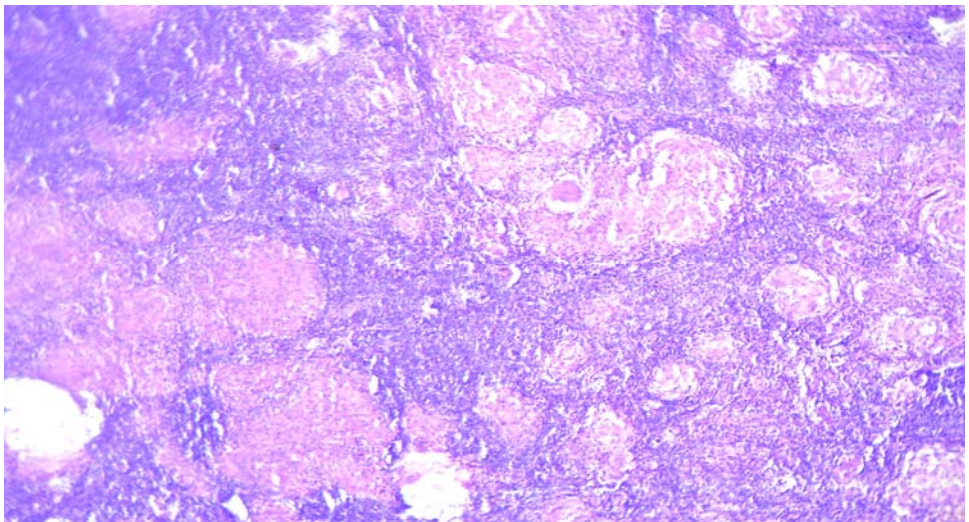


Fig 29 Microscopic appearance of lymphnode with granulomatous reaction to the tumour cells. (H&E, x100) Path No. 1873/04

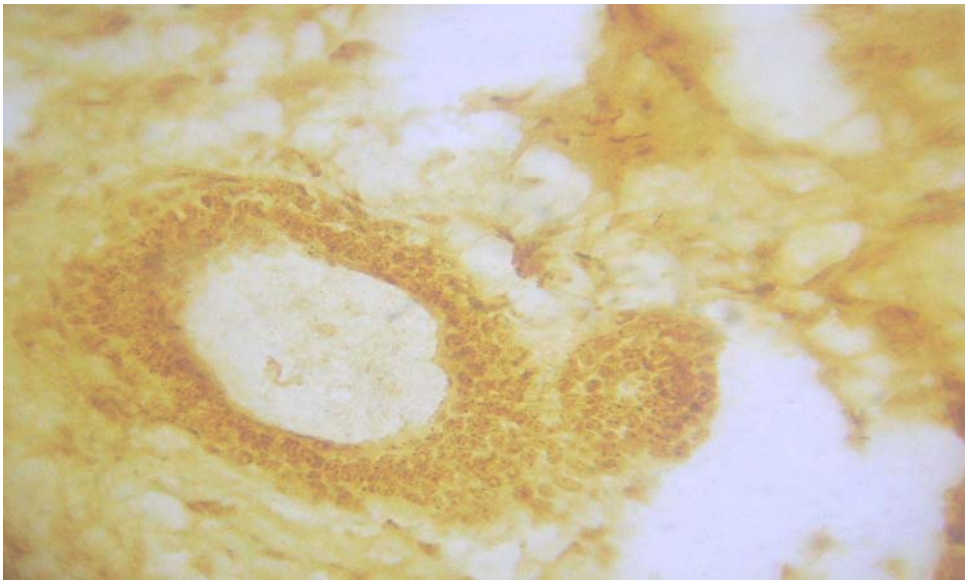


Fig 25 Photograph showing the pattern of AgNOR staining in fibroadenoma. Path No.1662/05

Fig 26 Photographs showing the pattern of AgNOR staining in DCIS with cribriform pattern. Path. No. 505/05

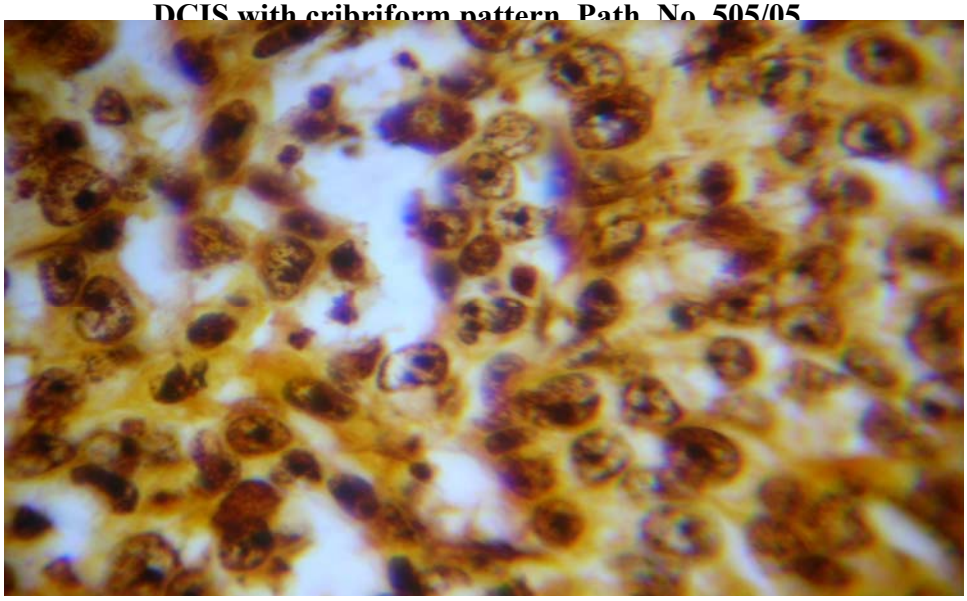


Fig 27 Photograph showing pattern of AgNOR staining in infiltrating ductal carcinoma. Path No. 1016/05

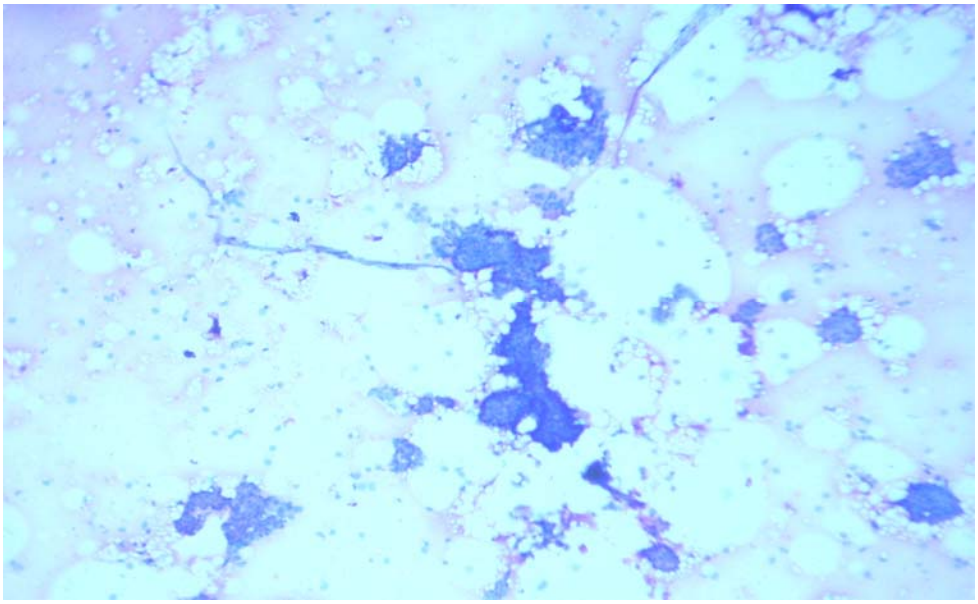


Fig 15 Fibroadenoma – smear showing antler-like papillary fronds. (H&E, x100) Cyt No. 33/05

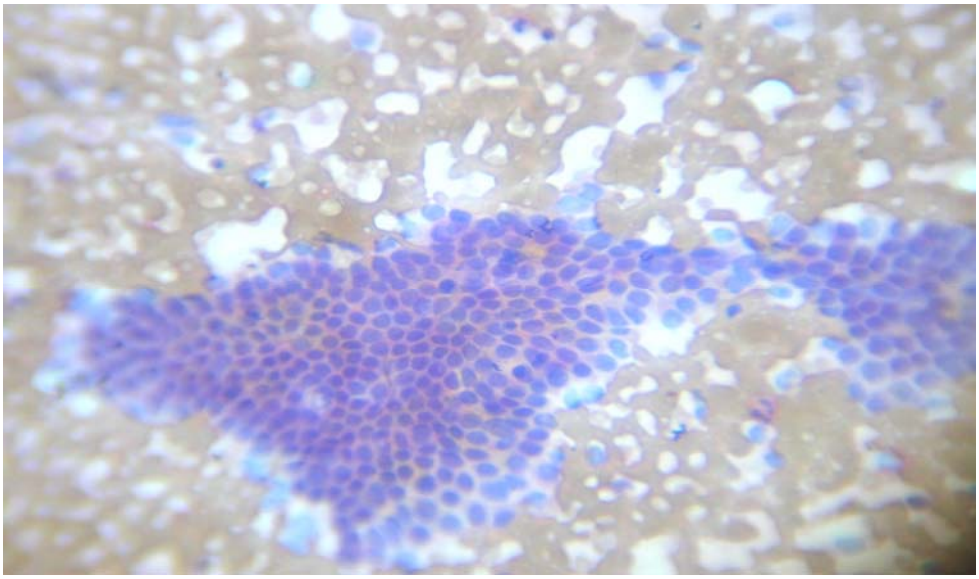


Fig 16 Fibroadenoma – smear shows large honeycombed sheets of tightly packed uniform ductal cells. (H&E, x400) Cyt No. 83/05

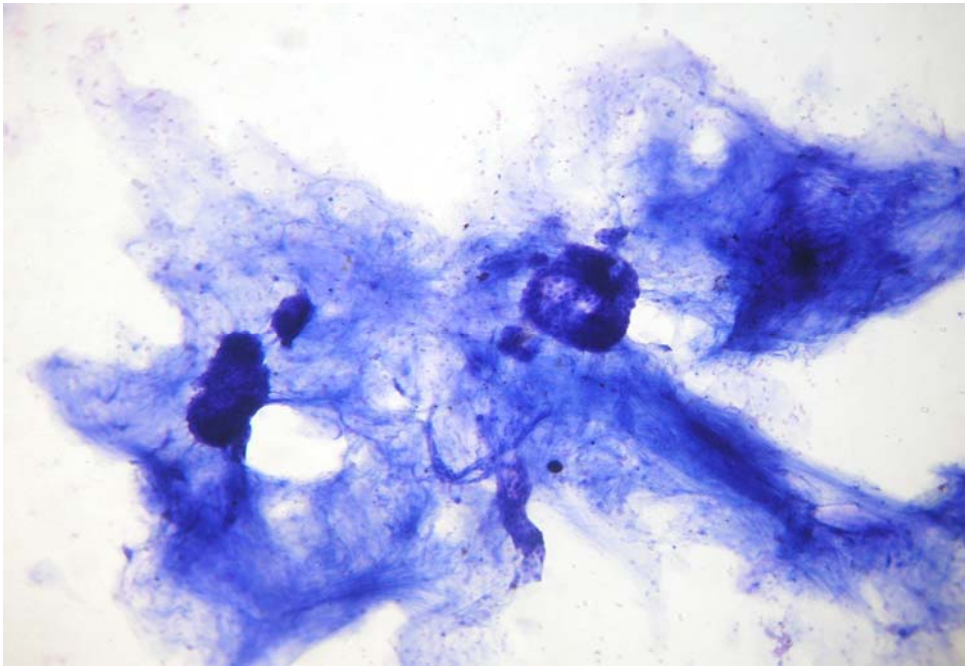


Fig 19 Mucinous carcinoma – smear shows abundant pools of mucin with aggregates and groups of uniform tumor cells. (H&E, x100) Cyt No. 801/05

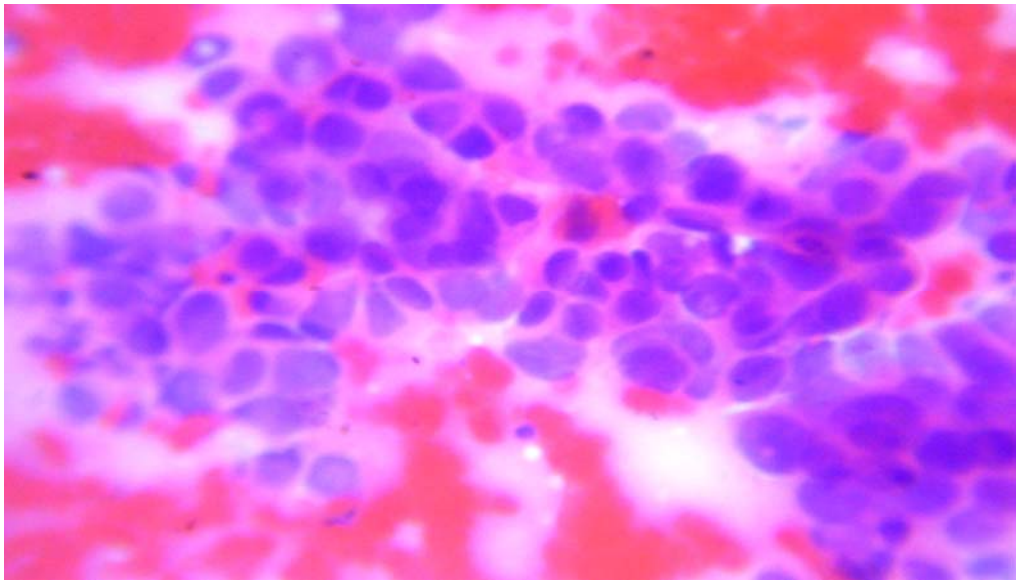


Fig 20 Ductal carcinoma – cellular smear showing loosely cohesive and individually scattered malignant cells. (H&E, x400) Cyt No. 11/05

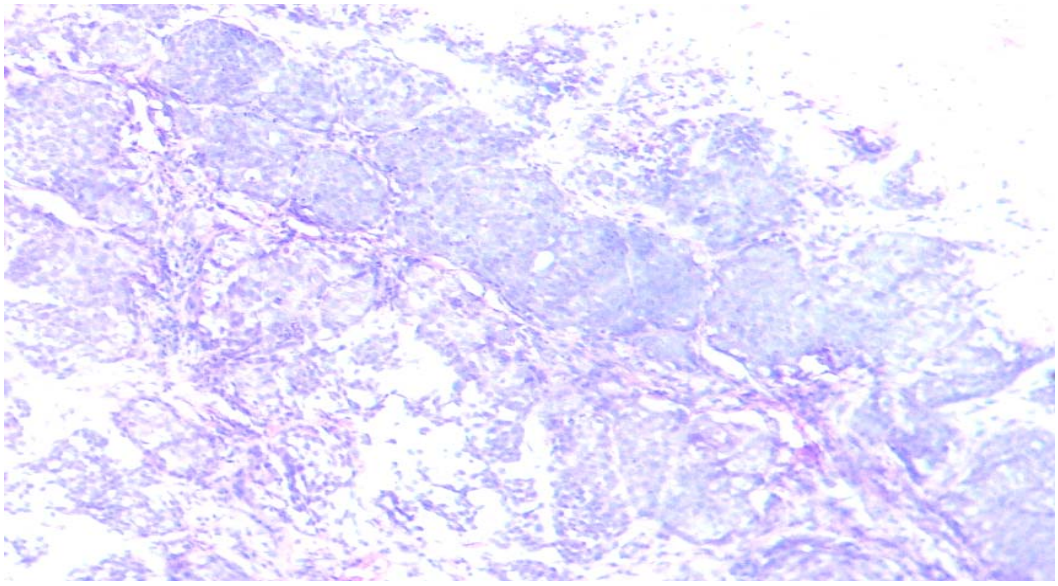


Fig 23 Infiltrating lobular carcinoma – section shows breast tissue infiltrated by small, uniform tumor cells arranged singly, in Indian file in a concentric fashion around the lobules. Trucut biopsy – (H&E, x100) Path. No.1215/05

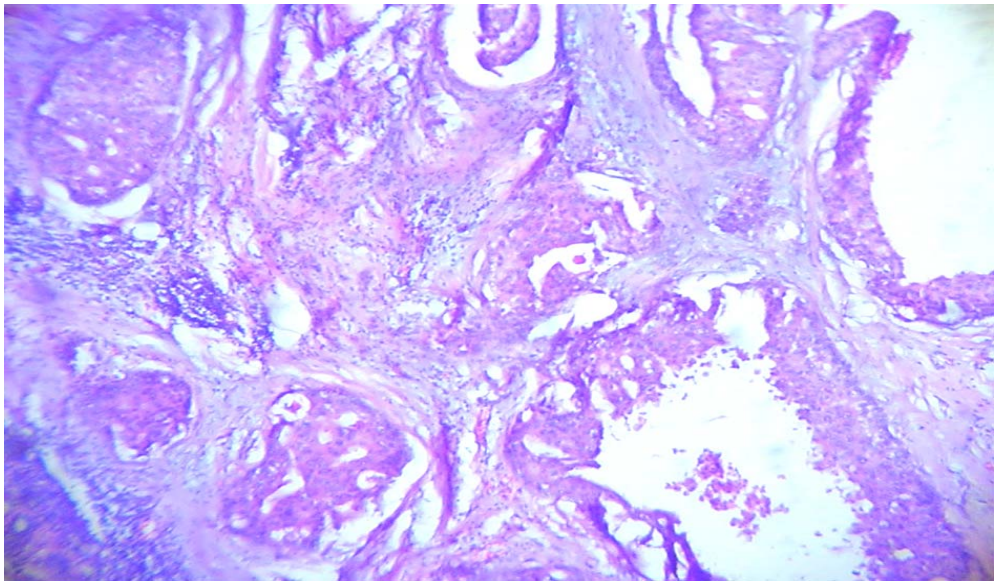


Fig 24 Infiltrating ductal carcinoma – section shows breast tissue infiltrated by tumor cells in well defined nests, and cords showing glandular differentiation. Trucut Biopsy – (H&E, x100) Path. No. 294/04

TABLE 1

PETERSE'S CRITERIA OF BENIGN AND MALIGNANT

BREAST CYTOLOGY

BENIGN	MALIGNANT
Large Monolayer of cells	**Cell dissociation; Arrangement in small clusters**
Nuclei Smaller than 16 μ	**Nuclei Larger than 16 μ**
Uniform nuclear size	**Anisonucleosis; Irregular nuclear borders**
Absence of nucleoli	**Presence of Nucleoli**
Absence of necrosis	**Presence of necrosis**

TABLE 2

EVALUATION OF ACCURACY OF THE DIAGNOSTIC PROCEDURE

Screening Test results	Diagnosis		Total
	Diseased	Not diseased	
Positive	**a (TP)**	**b (FP)**	**a+b**
Negative	**c (FN)**	**d (TN)**	**c+d**
Total	**a+c**	**b+d**	**a+b+c+d**

TP – True positive **FP – False positive**

FN – False negative **TN – True negative**

Formula

$$\textbf{Sensitivity} = \frac{\textbf{a}}{\textbf{a+c}} \times \textbf{100}$$

$$\textbf{Specificity} = \frac{\textbf{d}}{\textbf{b+d}} \times \textbf{100}$$

$$\textbf{Accuracy} = \frac{\textbf{a+d}}{\textbf{a+b+c+d}} \textbf{ x 100}$$

TABLE 3

SEX INCIDENCE OF BREAST TUMOURS

Tumour Type	Females		Males	
	No. of Cases	Incidence %	No. of Cases	Incidence %
Benign	**407**	**100%**	**00**	**0.0**
Malignant	**409**	**99.6%**	**3**	**0.4%**

TABLE 4

INCIDENCE OF VARIOUS BENIGN BREAST TUMOUR AND

TUMOR LIKE LESION

S. No.	LESION	No of cases	Incidence %
1	**Fibroadenoma**	**246**	**30.14**
2	**Fibrocystic disease**	**27**	**3.3**
3	**Fibroadenosis**	**121**	**14.8**

4	**Benign cystosarcoma phyllodes**	**13**	**1.5**

TABLE 5

DECENNIAL INCIDENCE OF MALIGNANT TUMORS COMPARED TO TOTAL MALIGNANCIES IN THIS INSTITUTION

Decade	Total Malignancies	No. of Malignant Breast Tumors	Incidence %
1960-1969	**11137**	**664**	**5.9**
1970-1979	**14977**	**715**	**4.8**
1980-1989	**15625**	**710**	**4.5**
1990-1999	**14430**	**866**	**6.0**

TABLE 6

INCIDENCE OF SIGNS / SYMPTOMS IN BREAST TUMORS

Sign / Symptom	Benign		Malignant	
	Total	Incidence %	Total	Incidence %

Breast Mass	**398**	**97.7**	**246**	**90.8**
Pain	**6**	**1.5**	**18**	**4.4**
Nipple Discharge	**3**	**0.76**	**12**	**2.9**
Nipple Retraction Puckering / Ulceration of skin	**0**	**0**	**9**	**2.2**

TABLE 7

PERCENTAGE OF BREAST CANCER CASES IN EACH STAGE

Stage	Current Study	
	No.	%
I	**47**	**17.3**
II	**70**	**25.8**
III A	**62**	**22.9**
III B	**15**	**5.5**
IV	**5**	**1.8**
Unstaged	**73**	**26.6**
Total Cases	**272***	

NOTE: *** - Represents those cases with FNAC and trucut biopsy.**

TABLE 8

INCIDENCE OF HISTOLOGICAL TYPES OF MALIGNANT

BREAST TUMOURS

S. No	Histological Type	Current Study	
		Total Cases	%
1	**Ductal Carcinoma in Situ**	**2**	**0.5**
2	**Invasive ductal carcinoma (NOS)**	**389**	**95.1**
3	**Invasive lobular carcinoma**	**4**	**0.9**
4	**Invasive ductal carcinoma with predominant intraductal carcinoma**	**1**	**0.24**
5	**Medullary carcinoma**	**1**	**0.24**
6	**Mucinous / Colloid carcinoma**	**10**	**2.4**
7	**Paget's disease of nipple**	**2**	**0.48**

ABLE 9

Diagnosis	No. of Cases
Normal Control	**1**
Fibroadenoma	**12**
Fibrocystic disease	**4**
Fibrocystic disease with DCIS	**1**
Infiltrating ductal carcinoma	**15**
Infiltrating lobular carcinoma	**2**
Total	**35**

TABLE 10

	No. of. Cases	Mean AGNOR Value counting method	
		Type I	Type II
Normal control	**1**	**1.2/±.2**	**1.2/±.2**
Fibroadenoma	**12**	**1.5/±1.2**	**3.0/±1.1**
		1.0/±.6	**2.5/±.3**
		1.5/±.3	**2.0/±1.1**
		1.8/±.3	**2.3/±1.5**
		1.3/±1.1	**2.1/±1.5**
		1.2/±.5	**2.6/±.3**
		1.7/±.3	**2.3/±1.4**
		1.9/±.25	**2.8/±.5**
		1.4/±.7	**2.3/±1.1**
		1.6/±.3	**2.4/±.7**
		1.1/±.4	**2.7/±.5**

		1.4±.6	**2.3/±.6**
Fibrocystic disease	**4**	**2.1/±.2**	**3.1/±1.2**
		2.3/±1.2	**2.9/±.3**
		2.1/±.4	**2.7/±1.3**
		2.3/±.3	**3.2/±.2**
Fibrocystic disease with DC/S	**1**	**2.5/±2.1**	**3.1/±2.5**
Infiltrating ductal Carcinoma	**15**	**5.2/±1.3**	**9.1/±1.1**
		5.3/±1.1	**9.2/±.2**
		5.2/±.25	**9.4/±1.3**
		5.1/±1.2	**8.3/±.5**
		4.5/±1.5	**8.6/±1.5**
		4.9/±.5	**9.1/±.5**
		5.3/±..2	**8.7/±1.5**
		5.3/±.6	**9.1/±.2**
		4.8/±1.3	**8.9/±1.3**
		5.4/±1.7	**9.2/±.2**
		5.1/±1.6	**9.3/±.5**
		4.9/±1.2	**9.6/±1.2**
		5.3/±1.5	**8.9/±.3**
		5.3/±1.7	**9.1/±.6**
		5.4/±1.2	**9.2/±1.2**
		6.5/±1.2	**8.2/±2.1**
		5.7/±.3	**8.7/±.2**

TABLE 11

PERCENTAGE OF BREAST CANCER CASES IN EACH STAGE A COMPARATIVE STUDY

Stage	Current Study		BCDDP (USA)		SEER (USA)	
	No.	%	No.	%	No.	%
I	**47**	**17.3**	**30**	**30**	**12**	**12**
II	**70**	**25.8**	**37**	**37**	**57**	**57**
III A	**62**	**22.9**	**3**	**3**	**10**	**10**
III B	**15**	**5.5**	**0**	**0**	**3**	**3**
IV	**5**	**1.8**	**NA**	**NA**	**7**	**7**
Unstaged	**73**	**26.6**	**30**	**30**	**11**	**11**
Total Cases	**272**		**100**		**100**	

TABLE 12

COMPARATIVE STUDY OF INCIDENCE OF HISTOLOGICAL TYPES OF MALIGNANT BREAST TUMOURS

S. No	Histological Type	Current Study		Fisher et al %	Usha et al %
		Total Cases	%		
1	**Ductal Carcinoma in Situ**	**2**	**0.5**	**5**	**1.9**
2	**Invasive ductal carcinoma (NOS)**	**389**	**95.1**	**67.5**	**77.8**
3	**Invasive lobular carcinoma**	**4**	**0.9**	**4.9**	**1.9**
4	**Invasive ductal carcinoma with predominant intraductal carcinoma**	**1**	**0.24**	**-**	**-**
5	**Medullary carcinoma**	**1**	**0.24**	**6.2**	**10.89**
6	**Mucinous / Colloid carcinoma**	**10**	**2.4**	**2.4**	**0.62**
7	**Paget's disease of nipple**	**2**	**0.48**	**2.3**	**1.9**

TABLE 13

INCIDENCE OF BENIGN AND MALIGNANT LESION IN FNAC AND TRUCUT SPECIMENS ANALYSED

	FNAC	TRUCUT
No. of Cases analysed	**607**	**502**
No. of Cases (+) Benign	**260**	**221**
No. of Cases (+) Malignant	**272**	**256**
No. of Cases False Positive	**1**	**1**
No. of Cases False Negative	**1**	**11**
No. of Cases (-) for Malignancy	**73**	**13**
Sensitivity (%)	**99.6**	**95.2**
Specificity (%)	**98.6**	**99.6**
Accuracy (%)	**99.4**	**95.1**

DIAGRAM 1

YEAR WISE DISTRIBUTION OF BREAST TUMORS

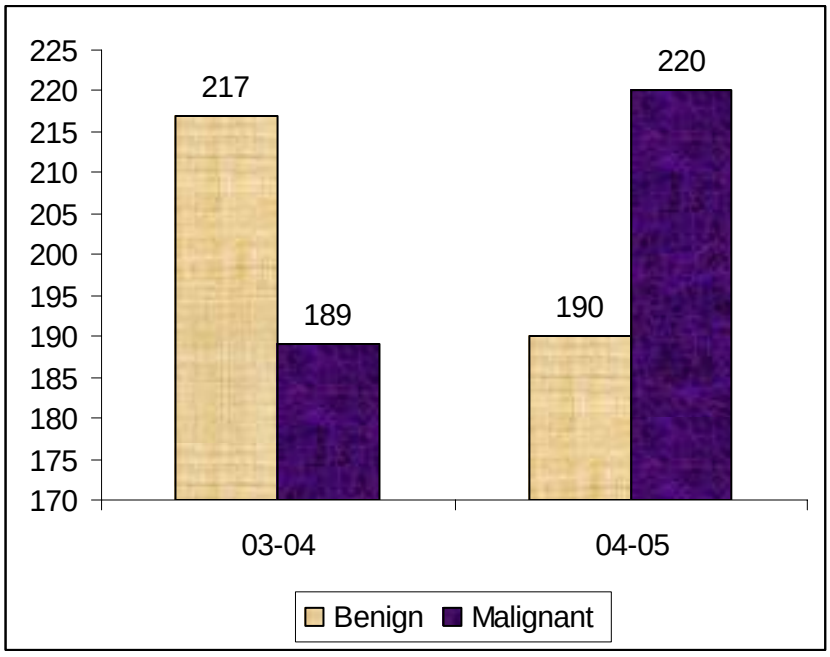
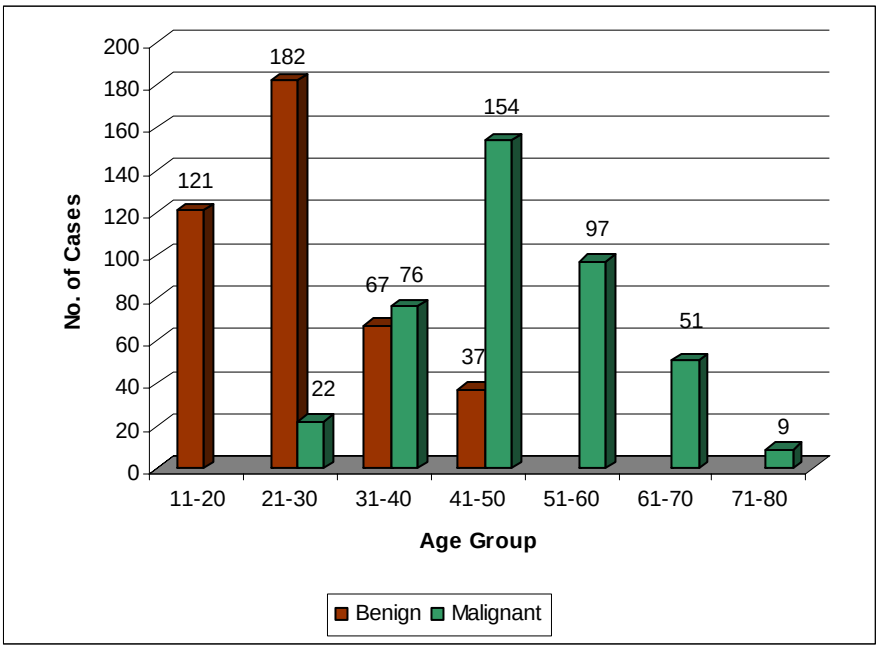
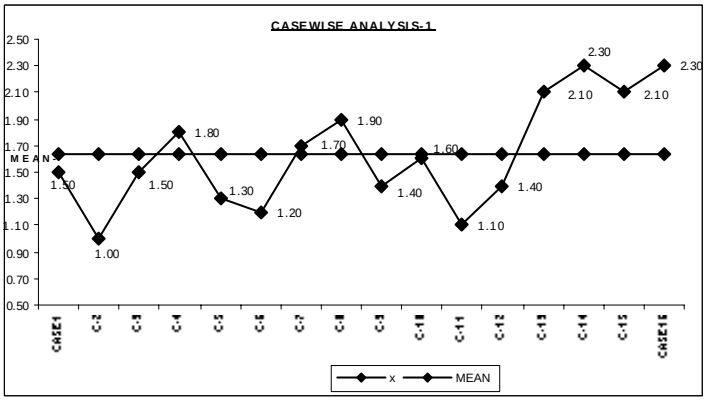


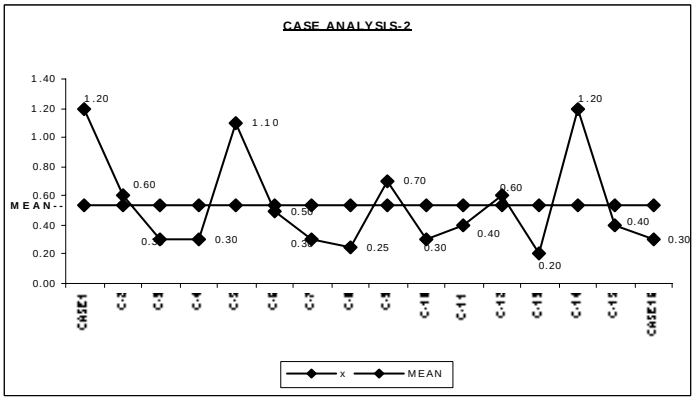
DIAGRAM 2

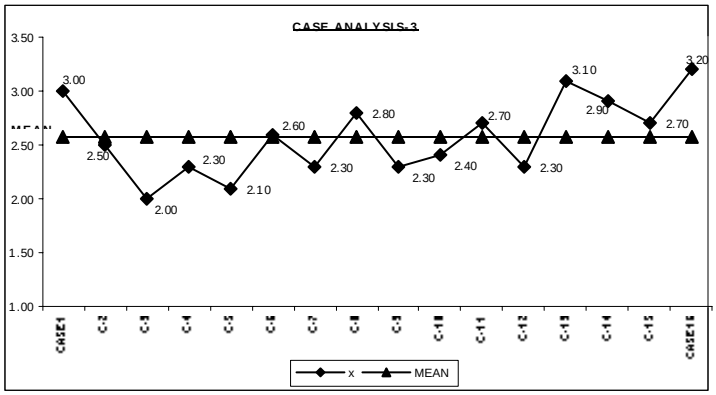
AGE INCIDENCE OF BREAST TUMOURS

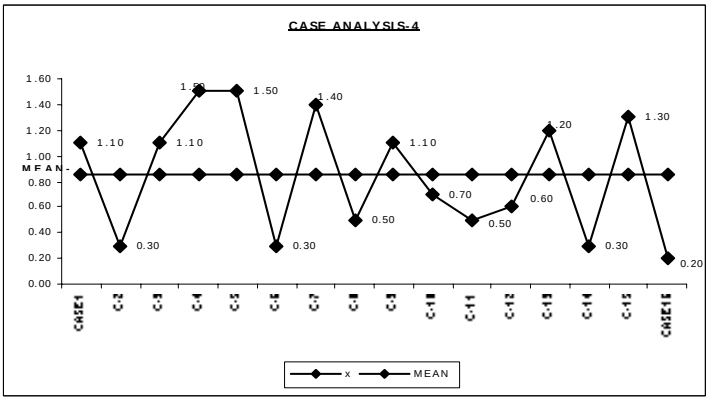


AgNOR score for benign neoplasm

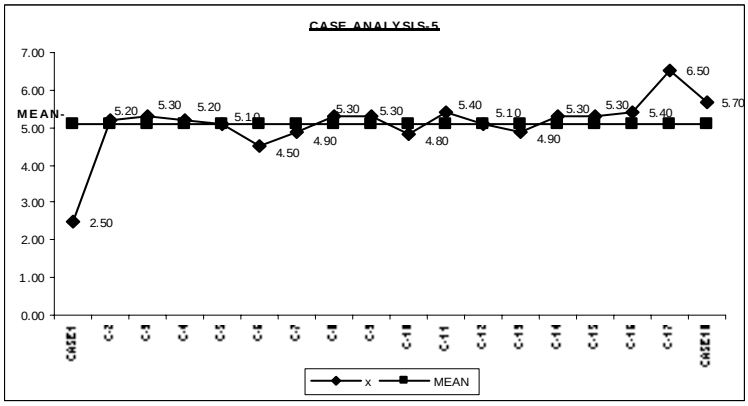


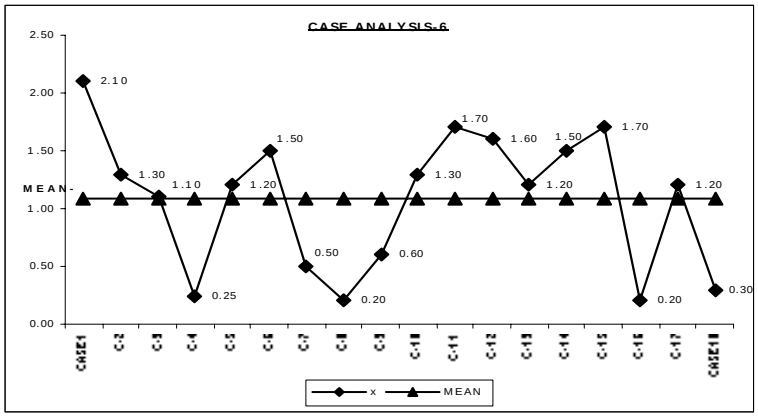


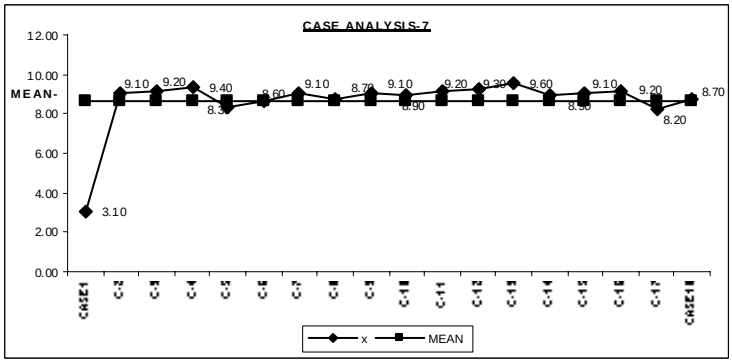


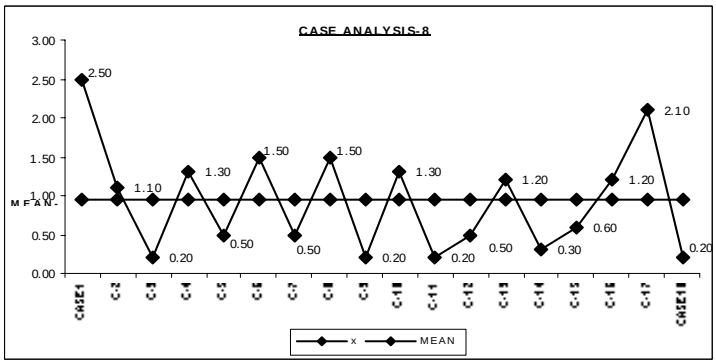


AgNOR score for malignant neoplasm




CASE ANALYSIS-6
MEAN-
x
MEAN


CASE ANALYSIS-7
MEAN-
x
MEAN


CASE ANALYSIS-8
MEAN-
x
MEAN

103.