Available online at www.journalijmrr.com

INTERNATIONAL JOURNAL OF MODERN RESEARCH AND REVIEWS

Int. J. Modn. Res. Revs.
Volume 4, Issue 11, pp 1402-1406, November, 2016

ISSN: 2347-8314

ESTROGEN RECEPTORS IN BENIGN BREAST LESIONS

[*1]A.R. Arun sundara Rajan, [2]R.Baskaran and [3]M.Prema

[*1]Post Graduate, Department of General Surgery, Rajah Muthiah Medical College, Chidambaram.
[2]Professor, Department of General Surgery, Rajah Muthiah Medical College, Chidambaram.
[3]Assistant Professor, Department of General Surgery, Rajah Muthiah Medical College, Chidambaram.

Article History: *Received 28th October ,2016, Accepted 15th November,2016, Published 16th November,2016*

ABSTRACT

Background: Hormones accelerate the proliferation of duct epithelium stem cells, increase their sensitivity towards environmental carcinogens. . The level of hormonal receptors in benign breast leisions seems to be a suitable prognostic marker which would enable the determination of breast lesion biological activities and the risk for possible malignant transformation of still benign lesion.**Aims and objectives:**To determine the presence of estrogen receptor in benign breast tissue epithelium. To quantify the estrogen receptors in various types of benign breast lesions and thereby predicting the high risk group for developing breast cancer.**Methods:**Biopsies from fifty five female patients with benign breast disease were analyzed for estrogen receptor positivity and percentage.**Results:**High ER concentrations in benign lesion tissues seem to be a good indicator of the significant risk and besides careful dispensarization, this state requires appropriate hormonal therapy.**Conclusion:**Further investigation of estrogen receptor expression in breast epithelium may identify points in the estrogen response pathway that may be interrupted to avoid the cancer-promoting effects of estrogen on breast epithelium. In that context, strategies that decrease estrogen receptor expression may prove protective against the carcinogenic effects

Keywords: Estrogen Receptors, Breast Lesions.

1.INTRODUCTION

Benign breast lesions may be divided into three categories, according to their proliferative activity. Histopathologic characteristics and the correlation with epidemiological studies showed different lesion relations to the risk of breast cancer. Hormonal microenvironment and the condition of hormonal breast tissue receptors further modify this process. Hormones accelerate the proliferation of duct epithelium stem cells, increase their sensitivity towards environmental carcinogens and inhibit the process of apoptosis. The breast contains hormonal receptors and hormones which participate in proliferation control and cancer development.

Estrogen is known to be etiologically important in the development of breast cancer; estrogen effect on target organs is mediated through its receptor called estrogen receptor (ER), and in the case of breast epithelium, it appears that estrogen effect includes the induction of proliferation, particularly of ductal tissue[(1)]. Benign proliferative lesions of the breast indicate a higher risk for subsequent breast malignancy(Dupont and Page,1985; Kreiger and Hiatt,1992; McDivitt et al., 1992). Proliferative multicell breast epithelial tissues may therefore contain a higher number of cells with hormonal receptors in comparison with normal breast tissues. The level of hormonal receptors seems to be a suitable prognostic marker which would enable the determination of breast lesion biological activities and the risk for possible malignant transformation of still benign lesion. This marker could be both an indicator of proliferation activity and a guide towards proper hormonal therapy capable of inhibiting this proliferation activity.

It is possible, therefore, that the presence of ER in benign breast epithelium is of significance in the prediction of breast cancer risk. Earlier studies of the expression of ER in benign breast tissue were performed using ligand binding assays, and ER levels were found to be extremely low or undetectable(Wittlitt et al., 1971; Feherty et al., 1971). This was attributed to the low epithelial cellularity of breast tissue. Interest in the receptor content of nonmalignant breast tissue lagged until the advent of highly specific monoclonal antibodies which enabled immunohistochemical assays for ER and PgR. It is now possible to detect these receptors on small samples of breast epithelium and to control accurately for the cellularity of the specimen and for the presence of benign breast disease. Breast epithelium has since been studied by several investigators using immunohistochemistry.

Corresponding author:* ***Dr A.R. Arun sundara Rajan,* *[1]Post Graduate, Department of General Surgery, Rajah Muthiah Medical College, Chidambaram*

This study is an attempt to analyse the ER expression in benign breast tissue of women undergoing breast surgery with the reasoning that the presence of ER in breast epithelium would render the tissue susceptible to the mitogenic stimulus of estrogen, and the presence of ER could thereby function as a risk factor for the development of breast cancer.

2.METHODOLOGY

The present study is an observational study done in the Department of Surgery, Rajah Muthiah Medical College, Annamalai University between September 2014 and August 2016. The study subjects comprised of fifty five women who underwent an excision biopsy of the breast lesion during the study period. The details of the subjects that included demographic data, clinical examination were entered into the proforma. All patients completed a self administered questionnaire regarding breast cancer risk factors at the first clinical visit. All participants signed a document of informed consent.

Breast tissue biopsies were performed due to clinical suspicion and palpable lumps, suspected or uncertain pathological breast formations seen on the mammographic picture.

Inclusion Criteria:

- Women in reproductive age group
- Benign histological findings

Exclusion Criteria:

- Inflammatory breast lesions
- Malignant cases

The surgical specimens were reviewed by a Pathologist. The biopsy samples were embedded in tissue-freezing medium (O.C.T., Miles chemical Co.) and snap-frozen in liquid nitrogen. Cryostat sections were first evaluated by hematoxylin-eosin staining. Histological assessment of benign lesions had been conducted according to Pages classification.

The sections were processed further only if adequate normal epithelium was present. Adequacy was defined as a minimum of 10 ducts or lobular acini. Epithelial samples included in the study showed either morphologically normal epithelium or minimal non profilerative benign change. The tissue was adequate in the 55 subjects and hence included in the study.

Parallel to histological examinations, the estrogen positivity and percentage of receptor content were determined.. Samples for estroge**n** positive estrogen receptor assay was taken to be equal or greater than 10 femtomoles of **(3H)** estradiol binding per mg of cytoplasmic protein. ER concentrations above 10 fmol/mg of cytosol protein were considered as positive. These concentrations approximately correspond with ER concentrations in normal breast tissues.

Statistical Methods

The primary relation of interest was that of estrogen receptor positivity with the occurrence of breast cancer. Analysis of the data involved a two-stage process. A univariable analysis was conducted first to determine the distributions of the study variables. This was followed by a bivariable analysis for investigation of possible confounders, outliers, and collinearity. Covariates in the analyses were defined as age at breast tissue sampling, age at menarche, parity, marital status, and educational level.

3.OBSERVATIONS AND RESULTS

The potential study population comprised of fifty five women in reproductive age group who underwent breast surgery in the institute. Biopsies from fifty five female patients with benign breast disease were analyzed for estrogen receptor positivity and percentage. The mean age of the study population was 35.5 $\pm$ 4.5 years with a minimum age of 15 years and a maximum age of 55 years. The subjects presented with palpable lump in the breast either right or left side. All of them had an insidious onset with slowly progressing lesion. Ten out of fifty five subjects had associated pain in the lump. Nipple discharge was associated in three subjects along with the lump. The mean age at menarche was 12.4 $\pm$ 0.65 years. None of the potential study subjects had a positive history of breast cancer. Five subjects were known case of Diabetes mellitus on regular treatment.

The man age at menarche was 12.4 $\pm$ 1.4 years with the lowest age at menarche being 10 years and the highest being 14 years. The mean duration of the lump in the study population was 8.9 $\pm$ 5.2 months. The least duration of the lump being noticed by the subject was 2 months and the maximum duration was around 24 months. Right sided lump was noticed in 24 (43.6%) and left sided lump in 31 (56.4%) subjects. Among the study population there were 13 nulliparous and 10 multiparous women. Of the remaining 32, primipars were ten and secundipars were twenty two. Forty nine subjects had a regular menstrual cycle and six had a history of irregular menstrual cycle.

Thirty four specimens were classified as fibroadenoma; twelve as fibrocystic disease, six as fibroadenosis, two had ductal ectasia and one specimen was labeled as giant fibroadenoma. Positive estrogen receptor expression was noted in 37 out of 55 cases (67.3%) of benign breast lesions.

In our study we found low ER concentrations in the group of women with nonproliferative lesions. These ER concentrations were found in groups 1, 4, and 7. In groups with more serious proliferative changes and atypia (3 and 6), ER concentrations were statistically significantly higher. ER concentrations changed parallelly according to the lesions proliferation activity. In groups with atypical hyperplasia we noted ER positivity prevalence. A progression in Quick score of ER and PR was observed from fibrocystic disease to proliferative lesions. ER quick score was 2 and 3 in fibrocystic disease and 4, 5, 6 and 7 in hyperplastic lesions.

TABLE 1: DISTRIBUTION OF SUBJECTS ACCORDING TO AGE

Age group (in years)	Number of subjects (n = 55)	Percentage (%)
< 20	11	20
21 – 40	36	65.4
> 40	8	14.6

.

TABLE 2: DURATION OF LUMP AMONG THE STUDY POPULATION.

Duration of lump (in months)	Number of subjects (n=55)	Percentage (%)
< 6	29	52.8
6 – 12	18	32.7
> 12	08	14.5

TABLE 3: DISTRIBUTION OF SIDE OF LUMP AMONG THE STUDY POPULATION.

Side	Number of subjects (n=55)	Percentage (%)
Right	24	43.6
Left	31	56.4

TABLE 4: PARITY AMONG THE STUDY POPULATION.

Parity	Number of subjects (n=55)	Percentage (%)
Nulliparous	13	23.6
Parous	32	58.2
Multiparous	10	18.2

TABLE 6: HISTOLOGICAL DIAGNOSIS IN STUDY POPULATION.

Histological variety	Number (n=55)	Percentage (%)
Fibroadenoma	34	61.8
Fibrocystic disease	12	21.8
Fibroadenosis	06	10.9
Ductal ectasia	02	3.6
Giant fibroadenoma	01	1.8

TABLE 7: ESTROGEN RECEPTOR STATUS AMONG THE STUDY POPULATION.

Estrogen receptor status	Number (n=55)	Percentage (%)
Positive	37	67.3
Negative	18	32.7

TABLE 8: HISTOLOGICAL VARIETY OF BBD AND ESTROGEN RECEPTOR STATUS

Histolgical variety (n=55)	ER positive (n=37)	ER negative (n=18)
Fibroadenoma (n=34)	23	11
Fibrocystic disease (n=12)	06	06
Fibroadenosis (n=06)	05	01
Ductal ectasia (n=02)	02	00
Giant fibroadenoma (n=1)	01	00

TABLE 9: DISTRIBUTION OF STUDY POPULATION ACCORDING TO HISTOLOGIGAL DIAGNOSTIC GROUP.

Group	Diagnostic group	Number (n=55)	Age ± SD (in years)
1	NPL + FA	16	29.8 ± 11.1
2	PLWA + FA	13	32.7 ± 5.9
3	AH + FA	02	28.9 ± 5.3
4	NPL	13	28.2 ± 9.5
5	PLWA	06	24.4 ± 7.7
6	AH	02	32.1 ± 8.2
7	FA	03	23.9 ± 8.0

TABLE 10: DISTRIBUTION OF ESTROGEN RECEPTOR CONCENTRATION AMONG THE HISTOLOGICAL DIAGNOSTIC GROUP.

Diagnostic group	ER in fmol/mg ± SD
NPL + FA	7.0 ± 10.2
PLWA + FA	29.4 ± 25.4
AH + FA	43.4 ± 72.6
NPL	2.5 ± 6.9
PLWA	12.6 ± 9.9
AH	53.5 ± 79.0
FA	14.6 ± 23.1

4.DISCUSSION

From the etiologic point of view, estrogens exhibit the most significant hormonal influence and participate in the process of carcinogenesis. The development of breast carcinoma is the result of the cumulative estrogen exposition of mammal tissue and its mitotic activity, which influences the risk of increased possibility for DNA synthesis defect fixation. Breast ductal epithelium is created from a relatively small population of stem cells with unlimited proliferation activity. These cells, which may be considered as immortal, are sensitive to the influence of carcinogens, and cancer develops by means of cell mutation. The number of stem cells is relatively constant and does not decrease even following menopause, when proliferative indices are very low. Next to the stimulation of proliferative activity, estrogens also influence the activity of Bcl 2 oncogens which modulate

apoptotic processes. The protective effect of estrogens against apoptotic processes in the human breast cancer cell line, MCF-7 has been experimentally proved.

Over expression of the Bcl-2 system prevented apoptosis induced by free radicals[82]. It may be assumed that estrogen exposition of the increasingly sensitive population of breast ductal epithelium stem cells may result in their survival even if DNA is damaged, at the expense of loss of their proliferation control. The acceleration of the cell cycle caused by estrogens may prevent physiological processes of damaged DNA repair in this sensitive cell population. In case of DNA damage, the cell remains in G1 phase until the damage is repaired. Failure of this process initiates the apoptosis mechanism. In a multistage process of breast carcinogenesis, estrogens act as the promoter in the development of already created clone tumor cells. The currently accepted idea is that this failure of equilibrium between proliferation controlling mechanisms and programmed cell death (apoptosis) is responsible for the deregulation of cell growth, the mutation fixation and cancer development. The growth of human epithelial breast cells is controlled by steroid hormones and by growth factor receptors. Both systems are important prognostic markers. The activity of other oncogens, which are stimulated by estrogens (c-erb/B2, fos, ras, myc) increases the creation of their proteins which act both as growth factors and as receptor proteins for growth factors (EGF receptor). For example, Fos is a nuclear transcription factor which may be induced both by steroid hormones and by peptide growth factors. It is an important interaction element between these systems. In the case of a developed neoplasia the Fos overexpression used to be related to the failure of response to hormonal therapy. Oncogen overexpression results in the loss of hormonal control of cell

proliferation which is another manifestation of deregulation in the path to neoplasia development. In a normal breast tissue only a small proportion of cells (5-25%) contain hormonal receptors.

Osborne,(1991) demonstrated a similar HR content in breast tissues of healthy volunteer women during the menstrual cycle and proved the ER down regulation in the luteal phase of the cycle, which was caused by endogenous progesterone secretion. In our study we found similar ER concentrations in the group of women with nonproliferative lesions. These ER concentrations were found in groups 1, 4, and 7.

In groups with more serious proliferative changes and atypia (3 and 6), ER concentrations were statistically significantly higher. ER concentrations changed parallelly according to the lesions proliferation activity. In groups with atypical hyperplasia we noted ER positivity prevalence. Our findings of low ER concentrations in breast nonproliferative lesions were in agreement with other authors results. McDivitt et al., (1992) found only a 10% positivity of both ERs in 100 patients with fibrocystic mastopathy. German authors, Wittlitt et al., (1971)reported ER negativity (i.e. concentrations under 10 fmol/mg of protein) in lesions classified as I and II. Similarly, Feherty et al., (1971) noted very low ER concentrations in six breast fibromas. Thus, we may assume that non-proliferative as well as proliferative breast lesions without atypia are not risk factors of breast cancer.

On the other hand, statistically significant high ER concentrations in groups with serious ductal epithelium hyperproliferation and atypical hyperplasia were markers of strong proliferation activity in our examined group. These findings prove the biological significance of the lesion and its hormonal dependence. In such cases proper hormonal therapy could reduce the proliferative potential of breast lesions.

Higher ER concentrations were observed in some patients with atypical hyperplasia. According to Khan[91] this state may be considered as precancerous and estrogen receptor prevalence as the marker of possible breast cancer development.

The cell becomes sensitive towards the mitogenous estrogen effect and the gradual loss of proliferation (and apoptosis) controlling mechanisms, results in the fixation of autonomous growth with gene faults and the gradual initiation of cancerogenesis. Bur proved a 91% ER positivity in low-nuclear grade DCIS, whereas in high nuclear grade DCIS he only observed a 57% positivity.

Thus, we may speculate that during precancerosis the high ER content is the marker of possible malignant transformation, whereas its reduction indicates the onset of hormonal insensitivity and tumor dedifferentiation. The estrogen receptor negative tumor is the next stage of dedifferentiation, characterized by the loss of hormonal contribution in proliferation control.

High ER concentrations in benign lesion tissues seem to be a good indicator of the significant risk and besides careful dispensarization, this state requires appropriate hormonal therapy which may inhibit the proliferative potential of this lesion. Our findings are supported by studies which have recently isolated the so called superactive ER variants of 200 times higher estrogen sensitivity. These variants were found in 30% of hyperplastic breast lesions, but never in normal breast tis[92]
One may thus assume that during the routine process d determination we also evaluate variants which may be considered as first markers of breast cancerogenesis initiation.

Due to the division of our examined group into a relatively high number of subgroups, we divided the groups according to the proliferative activity of the leading lesion and did not distinguish whether or not these lesions were accompanied by fibroadenomas. The results of the joined groups, NPL with NPL+FA (groups 1+4), PLWA with PLWA+FA (groups 2+4) and AH with AH+FA (groups 3+6), were similar in all seven groups. This confirmed the original statistical evaluation. The presence of fibroadenomas did not modify these results and therefore one may argue that adenofibromas are not a risk lesion. The second method of joined groups which did not regard different proliferative activities and considered only the presence of fibroadenomas, proved significant PR prevalence in the group of combined lesions with FA as compared with the lesion itself (without FA). However, it did not prove the so called additional risk of fibroadenoma which was in agreement with the previous order.

5.CONCLUSION

In conclusion, further investigation of estrogen receptor expression in breast epithelium may identify points in the estrogen response pathway that may be interrupted to avoid the cancer-promoting effects of estrogen on breast epithelium. In that context, strategies that decrease estrogen receptor expression may prove protective against the carcinogenic effects of estrogen and other compounds collectively called xenoestrogens, which have affinity for estrogen receptor.

6.BIBLIOGRAPHY

Dupont, W. D., and Page, D. L. Risk factors for breast cancer in women with proliferative breast disease. N. Engl. J. Med., 312: 146-151, 1985.

Feherty, P., Farter-Brown, C and Kellie, A. E. 1971.Oestradiol receptors in carcinoma and benign disease of the breast: an in vitro assay. Br. J. Cancer, 25: 697-710.

Kreiger, N., and Hiatt, R. A. Risk of breast cancer after benign breast diseases: variation by histologic type, degree of atypia, age at biopsy, and length of follow-up. Am. J. Epidemiol., 135: 619~631, 1992.

McDivitt, R. W., Stevens, J. A., Lee, N. C., Wingo, P. A., Rubin, G. L., and Gersell, D. 1992.Histologic types of benign breast disease and the risk for breast cancer. Cancer (Phila.), 69: 1408--1414, 1992.

Osborne, M. P. 1991. Breast development and anatomy. In: J. R. Harris, S. Hellman, I. C. Henderson, and D. W. Kinne (eds.), Breast Diseases., pp. 1-13, New York: J. P. Lippincott Company.

Wittlitt, J. L., Hilf, R., Brooks, E, Savior, E. D., Hall, T. C., and Orlando, R. A. 1971.Specific estrogen-binding capacity of the cytoplasmic receptor in normal and neoplastic breast tissues of humans. Cancer Res., 32: 1983-1992.

*****