International Journal of Psychosocial Rehabilitation, Vol.24, Issue 01, 2020
ISSN: 1475-7192

Association of Body Mass Index and Age with Positive Receptors Expression and Metastasis Status Subtypes in Iraqi Women with Breast Cancer

[1] Hamzah H. Kzar, [2] Moaed E. Al-Gazally, [3] Moshtak A. Wtwt

Abstract:

Background: *Breast cancer (BC) is a heterogeneous disease witch produce from a number of genetic and epigenetic risks that lead to dysregulation of cell growth, circumvention of apoptosis, and development of the ability to invade the underlining tissue through the basement membrane.* ***Aim:*** *To assessment the association of BMI and age with positive receptors and metastasis status subtypes expression in Iraqi women of BC and determine the ERA and ERB and investigated the impact of those biomarkers alteration on the prognosis of patients with BC.* ***Methods:*** *This case-control study involved on 120 women with BC and same number as control group. Estimation of ERA and ERB were based on sandwich- ELISA.* ***Results:*** *The results examined that age and BMI of women with BC were significant associated with positive expression of ER and PR (p-value <0.05), but there were no association between those parameters and expression of Her-2 receptor subtype (p-value >0.05)**.** The results referred to a significant effects of age and BMI on expression of ER and PR in women with BC (OR=1.25 CI95%=0.49-3.51, OR=1.60 CI=0.61-4.16) (OR=1.4 CI95%=1.4-3.6, OR=1.83 CI95%=0.69-4.87), respectively. The results suggested the significant increase in incidence of IDC type of BC compare to ILC in women with (BMI ≥ 25)* ***Conclusion:*** *Assessment of ERA and ERB are useful in diagnosis and following up of women with BC in different ER, PR, Her-2, and in different histological grades.*

Keywords*:* *Breast cancer, Estrogen receptors alpha and beta (ERA, ERB), BMI, Metastasis status.*

Introduction:

Assessment of the agents that relate with an increase the incidence rates of breast cancer (BC) development is an important factor in general health screening and premature detection for women (1,2). BC is a heterogeneous disease witch results from a chains of genetic and epigenetic events that lead to deregulations of cell growth, circumvention of apoptosis, and development of the capacity to invade the underlining tissue through the basement membrane (3,4). The incidence is much higher for women but BC affects both men and women. Overall rating, women are at 100-fold higher risk of BC than men (5). Besides sex and from the above data suggested that the aging is one of the most important risk factors of the BC, because the incidence of BC is highly related to the increasing age. In 2016, approximately 99.3% and 71.2% of all BC associated deaths in America were reported in women over the age of 40 and 60 years, respectively (6). Therefore, it is necessary to have a mammography screening ahead of time in women aged 40 or older (7). In Iraq, the same study suggested that the mean age of BC incidence was from 50 to 60 years (8). Nearly a quarter of all BC cases are related to family history and women, whose mother or sister has a family history of BC are a higher risk factor to incidence with this cancer (9,10). The aim of this study to assessment the association of BMI and age with positive receptors expression and metastasis status subtypes in Iraqi women of BC and determine the ERA and ERB and investigated the impact of those biomarkers alteration on the prognosis of women with BC.

[1] *Lecturer, Veterinary Medicine collage, Al-Qasim Green University, Iraq. Correspondence E- mail: hamza14shukri72@gmail.com*
[2]*Profesor, Collage of Medicine, Babylon university (works now at University of Al-Ameed), Iraq.*
[3]*profesor, Collage of Medicine, University of Babylon, Iraq.*

Materials and Methods:

Study design:

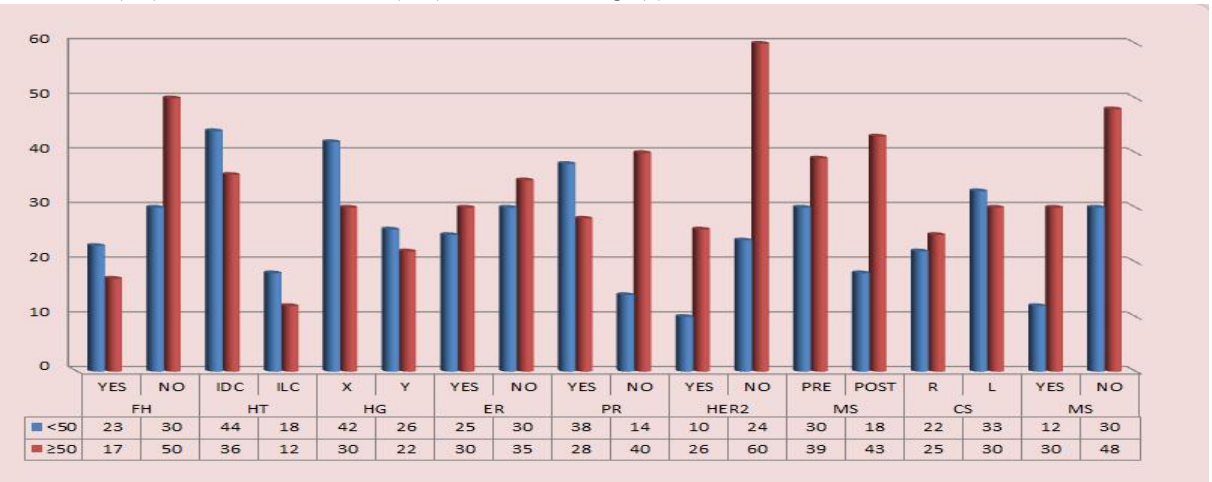
This study was done at the laboratories of chemistry department, College of Medicine , University of Babylon. The patients group recruited in this study were 120 women in the age group ranging from 29 - 72 years , the mean ± SD was (56.3 ± 11.7 y). The diagnosis of BC confirmed by CT- scanning, ultrasound, MRI and biopsy with histo-pathological examination. One hundred and twenty apparently healthy women (without breast diseases) were taken as a control group of the age ranging (27-71 y), the mean ± standard deviation (SD) was (54.9 ± 10.7 y). The ages of this group were matched to ages patient group, where statistical analysis showed no significant differences in ages between patient and control groups ($p>0.05$). Control group underwent full history and physical examination including: address, age, smoking , past history of diseases and medications. Any women whom suffered from mammary glands diseases and whom used contraceptive pills were also excluded from this group.

Estimation of ERA and ERB:

This method was based on Sandwich-enzyme linked immune sorbent assay (ELISA). The micro-Elisa strip plate provided has been pre-coated with an antibody specific to ERA. Standards or samples were added to the appropriate wells and combined with the specific antibody for ERA . Horseradish Peroxidase (HRP)- conjugated antibody specific for ERA was added to each well and then incubated. The TMB substrate solution was added to each well. Only those wells that contain ERA and HRP conjugated ERA antibody will appear blue in color and then turn yellow after the addition of the stop solution. The concentration of ERA can be calculated in the samples by comparing the OD of the samples to the standard curve.

Results:

Women with BC were classified according to the age (≥50 and < 50) into two subgroups depending on family history (FH), histological type (HT), histological grade (HG)(IDC, in situ ductal carcinoma)(ILC, in situ lobular carcinoma), estrogen receptor (ER), progesterone receptor (PR), Her-2 receptor (HER-2), menopause status (MS), cancer site (CS), and metastasis status (MS) , as shown in Fig. (1):



	FH		HT		HG		ER		PR		HER2		MS		CS		MS	
	YES	NO	IDC	ILC	X	Y	YES	NO	YES	NO	YES	NO	PRE	POST	R	L	YES	NO
<50	23	30	44	18	42	26	25	30	38	14	10	24	30	18	22	33	12	30
≥50	17	50	36	12	30	22	30	35	28	40	26	60	39	43	25	30	30	48

Figure (1): Age distribution of patient subgroups

Amongst 120 women with breast cancer who contributed in this study, there were classified according to the BMI(≥25 and < 25) into two subgroups as described in above as shown in Fig. 2:

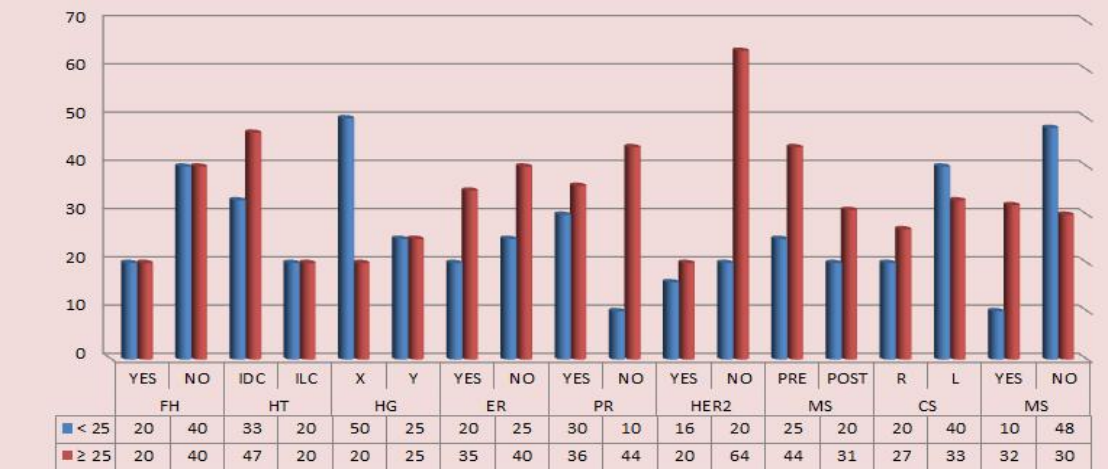


Figure (2) : BMI distribution of patient subgroups

The results of this study revealed that that age and BMI of women with BC were significant associated with positive expression of ER and PR, but there were no association between those parameters and expression of Her-2 receptor subtype.

Table 1 show the levels of ERA (pg/ml) depending on clinic-pathological variables in study subgroups:

Table (1): Levels of ERA (pg/ml) depending on clinic-pathological variables in study subgroups

Clinic-pathological variables	**ERA (mean± SD) pg/ml**			**ERA (mean± SD) pg/ml**		
	Age			**BMI**		
Family history	**≥ 50**	**< 50**	**p-value**	**≥ 25**	**< 25**	**p-value**
Yes	123.7±4	123±3.8	0.055	122.5±3	122±2.9	0.084
No	123.9±3.7	122±3.6	0.088	122.5±2	122±1.5	0.093
Histological type						
IDC	122.7±3.9	122±3.9	0.065	122.6±2	121±2.3	0.094
ILC	123.6±3.8	123±3.5	0.078	122.8±2	121±1.9	0.063
Histological grade						
Grade 1+2	121.7±3.7	121±3.8	0.095	122.3±3	122±2.9	0.084
Grade 3	123.5±3.2	120±3.6	**0.008**	123.5±2	119±1.5	**0.003**
Menopausal status						
Pre	124.7±4	120±3.8	**0.000**	123.6±3	122±1.9	**0.000**
Post	123.9±3.9	121±3.8	**0.000**	122.5±2	124±2.5	**0.000**
Site of cancer						
Right	123.7±4.1	123±3.8	0.075	122.5±2	122±1.9	0.14
Left	123.9±3.7	123±3.6	0.098	122.5±2	122±1.8	0.19
Metastasis status						
Yes	124.8±3.9	120±3.7	**0.000**	124.2±3	122±2.9	**0.000**
No	124.9±3.9	124±3.7	0.054	122.1±2	122±1.5	0.076

The figure 3, showing the serum levels of ERA and ERB in women with BC compare to control groups. The serum levels of ERA were significantly raise (124.9±14 pg/ml, P<0.05) in women with BC compare to control group (99.8±10.2 pg/ml). Serum levels of ERB was showing no significantly differences (12.3±4 pg/ml, P=0.126) in patients with BC compared to control group (11.6±3 pg/ml).

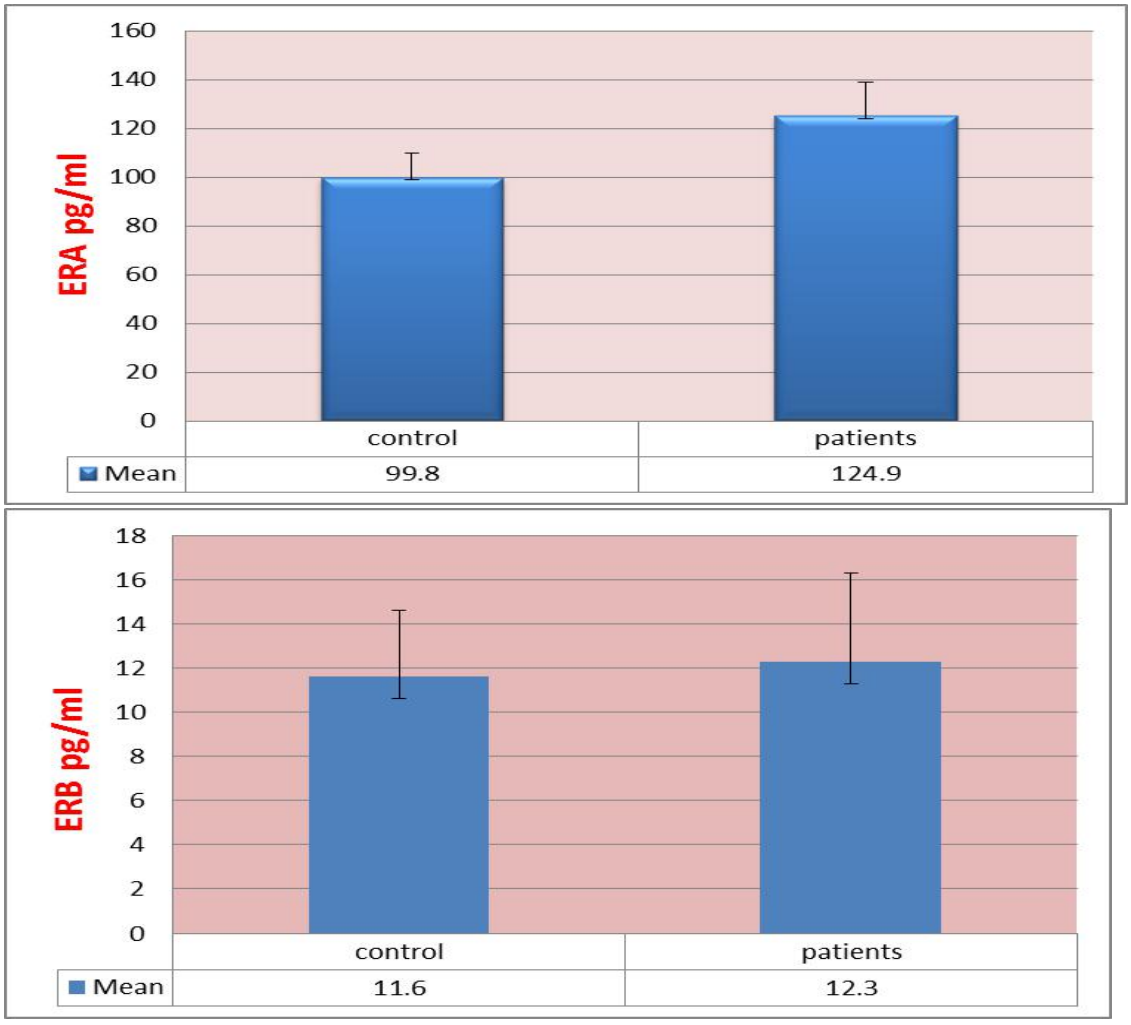


Figure (3): Serum level of ERA and ERB (pg/ml) in patients and control groups

Discussion:

Women with BC were included in this study have an age of 56.3 ± 11.7 years. The minimum and maximum age were 29 and 72 years respectively, in which about 53 % of the patients located in the age ≥50 years of old and 47% were in the age <50 years of old. The current study showing that the highest frequency of BC was in women with age of 51-60 years, followed by those with age of 41-50 years. This trend of frequency rate in relation to age was inconsistent to that reported for Arab Countries and globally. Thakur P *et al., (2017)* were suggested that age is one the most important known risk factor for breast cancer (11). They found that the females with age >50 years were 49% but female with age < 50 were 51%. Furthermore, the fact that the prevalence of BC correlate with age and this finding disagreement with the results of present study that showing the incidence rate increase with age. Other study from Iraq suggested that the age is the main risk factor for BC and the incidence rate increase with age and this results in same line with our finding (12). The present results disagreed with two studies in Iraq performed on women with BC and revealed that the peak of age frequency in the Iraqi BC patients was 44.5 years, and that 76.8% were under 50 years (13,14). For obese and excess body weight (over weight and pre-obese) is generally recognized as a significant risk factor for many common cancers included BC (15). The association between body mass index and BC risk has received much attention in the past few years, but the findings have still been controversial, therefore in this study, we examined the association of BMI and increase risk of Iraqi women with BC. The results of present study suggested the significant increase in incidence of IDC type of BC compare to ILC in women with (BMI ≥ 25) and the IDC is the must type of BC in the world. This results may be due to increase fats and muscles mass in chest of women lead to pressing on milk ducts in breast tissue leading to increased risk of IDC type of BC. The results show increase incidence of BC in women have not expressed Her-2 receptor and this will give a negative correlation between women have (BMI≥ 25) and negative Her-2 receptor expression. This cause–effect relationship between obesity indices and BC has been espoused in various studies including that of Kang L et al., (2018) (16). Obesity is a significant risk factor for BC development, therefore the previous epidemiological study have shown a consistent association between elevated BMI and age with increased BC risk in women (17). In this study the role of age in the

increasing of expression of ER (OR= 1.02, CI95% =0.54-1.93) and PR(OR= 1.02, CI95%= 0.58-1.86) were very clear and supported the hormonally disorders associated factors that effect on tumors progression and increase of BC risk. Therefore, the increasing in the BMI was also found to increase the expression of ER and PR and this may be because increase mass of adipose tissue that help to raise the levels of ER and PR. In contracts the Her-2 receptor expression was found to be not effected by age and BMI and this may be due to the hormones not bind with this receptor and its target of many growth factors and specific drugs (18). The incidence rates of hormone receptors positive BC are increasing in the United States, likely due in part to increasing prevalence of excess body weight and declining fertility rates. Carol E et al., (2019) showing that the decreasing in incidence rates for hormone receptors negative BC, which are associated with poorer survival, may have contributed to the declines in BC mortality (19). Metastatic status of BC frequently differs from the preceding primary BC in properties such as receptors type. BC often may be metastasize anywhere in the body but primarily distribution to the many organs such as bone, lungs, lymph nodes, liver, and brain, with the most common site being the bone (20). The results shows not effects of age and BMI on expression of Her-2 receptors in metastasis status. In metastasis BC , the overexpression of ER and PR may be correlated with cancer stages and metastasis organ status. The results show that expression of Her-2 is not a prerequisite for development of BC metastases. The expression of Her-2, ER, and PR receptors can change during the course of the disease in BC (21). The results of present study supported the conclusions by other studies that demonstrate the molecular detection of Her-2 overexpression in tumor cells is predictive of the Her-2 status on metastases (22,23). The present results provided data that showing of the menopausal, metastasis status, and histological grade 3 were highly significantly differences of ERA (ng/ml)(p-value<0.05) depending on age and BMI between two subgroups (pre ,post) , (yes, no) , and (grade 3) respectively. After menopause, the estrogen levels goes down to 10 folds, whereas androgens levels decrease only 1.5-fold (24). These findings confirm that young age at menopause (less than 50 years) and old age at menopause (more than 50 years) increase BC risk through increasing of ERA levels. Although metastatic BC is unlikely to be cured, there have been meaningful improvements in survival due to the availability of more effective systemic therapies, including endocrine therapy in the treatment of metastasis BC (25). This disturbance in hormones level in pre and post menopause, metastasis, high histological grade of BC may be the main cause of unbalanced ERA levels. Expression of ERB is not predominantly in sex organs except prostate; it is found in skin, bone, brain, lung, urinary bladder, blood vessels, lymphocytes, and fat tissues and appears to be widely distributed throughout the whole body. ERA is expressed mainly in sex organs such as breast, uterus, ovary, and testis (26). For this reason, the ERB levels may be not changed between women with BC and control group and this useful in estimation of ERA/ERB as prognostic biomarker in BC. In conclusion, assessment of ERA and ERB are useful in diagnosis and following up of women with BC in different ER, PR, ,Her-2, and in different histological grades.

Conflicts of interest

No conflict of interest related to this manuscript.

Acknowledgments:

Our gratitude to all of the women for their cooperation in achieving this study. We also thanks all staff of biochemistry department /collage of medicine/ Babylon university for their supported to complete this work.

References:

1- PDQ Screening and Prevention Editorial Board. PDQ Cancer Information Summaries. National Cancer Institute (US); Bethesda (MD) (2018). Breast Cancer Screening (PDQ®): Health Professional Version.

2- Doren A, Vecchiola A, Aguirre B, Villaseca P. Gynecological-endocrinological aspects in women carriers of BRCA1/2 gene mutations. Climacteric;2018,21(6):529-535.

3- Van Keymeulen A, Lee M, Ousset M, Brohee S, Rorive S, Giraddi R et al . Reactivation of multipotency by oncogenic PIK3CA induces breast tumour heterogeneity. Nature. 2015,525:119–23.

4- Wooster R and Weber B. Breast and onvarian cancer. N Engl J Med; 2013,348(23):2339.

5- Cancer Genome Atlas N. Comprehensive molecular portraits of human breast tumours. Nature. 2012,490:61–70.

6- Siegel R, Miller K and Jemal A. Cancer Statistics. CA Cancer J Clin. ;2017, 67:7–30.

7- DeSantis C, Fedewa S, Goding Sauer A et al. Breast cancer statistics. Convergence of incidence rates between black and white women. CA Cancer J Clin. 2016;66:31–42.

8- Sarab K et al. Risk factors of breast cancer among Iraqi women. J Contemp Med Sci **:** 2019,5, (3):149–153

9- Runnak A, Hemin A, Dana N, , Hazha A, Michae D. Breast cancer in Iraq is associated with a unimodally distributed predominance of luminal type B over luminal type A surrogates from young to old age. BMC women's health .2017,17:27.

10- Brewer H, Jones M, Schoemaker M et al. Family history and risk of breast cancer: an analysis accounting for family structure. Breast Cancer Res Treat.2017 ;165:193–200.

11- Thakur P, Seam RK, Gupta MK, Gupta M, Sharma M, Fotedar V. Breast cancer risk factor evaluation in a Western Himalayan state: A case–control study and comparison with the Western World. South Asian J Cancer.2017, 6(3):106–109.

12- Runnak A, Hemin A, Dana N, et al. Breast cancer in Iraq is associated with a unimodally distributed predominance of luminal type B over luminal type A surrogates from young to old age. BMC Women Health.2017; 17: 27.

13- Al-Anbari, S. Correlation of the clinicopathological presentations in Iraqi breast cancer patients with the finding of bio field breast cancer diagnostic system (BDS), HER-2 and Ki-67 immunohistochemical expressions. Ph.D thesis, College of medicine, University of Baghdad, 2009.

14- Al-Sanati, M. PCR Study of BRCA1 BRCA2 in Correlation to Immunohistochemical Expression of p53, Estrogen and Progesterone Receptors in Breast Cancer. Ph.D thesis, College of medicine, University of Baghdad, 2009.

15- Renehan AG, Tyson M, Egger M, Heller RF, Zwahlen M. Body-mass index and incidence of cancer: a systematic review and meta-analysis of prospective observational studies. Lancet.2008;371:569–578.

16- Kang L, Weining Z, Zhiming D et al. Association between body mass index and breast cancer risk: evidence based on a dose–response meta-analysis. Cancer Manag Res.2018; 10: 143–151.

17- Goldberg M et al. Early-Life Growth and Benign Breast Disease. Am J Epidemiol. 2019, 1;188(9):1646-1654.

18- Carol E et al. Breast Cancer Statistics, 2019. CA CANCER J CLIN.2019;69:438–451.

19- Macedo F, Ladeira K, Pinho F, Saraiva N, Bonito N, Pinto L, Goncalves F. Bone Metastases: An Overview. Oncology Reviews. 2017,11 (1): 321.

20- Riethdorf S, Müller V, Zhang L, et al. Detection and HER2 Expression of Circulating Tumor Cells: Prospective Monitoring in Breast Cancer Patients Treated in the Neoadjuvant GeparQuattro Trial. Clin Cancer Res. 2010;16(9):2634–45.

21- Bahriye A, Sabine K, Volkmar M. et al. Comparison of the HER2, estrogen and progesterone receptor expression profile of primary tumor, metastases and circulating tumor cells in metastatic breast cancer patients. BMC Cancer.2016; 16: 522.

22- Linares A et al. Increased expression of the HDAC9 gene is associated with antiestrogen resistance of breast cancers. Mol Oncol.2019;13(7):1534-1547.

23- Rothman MS, Carlson NE, Xu M, Wang C, Swerdloff R, Lee P, Goh VH, Ridgway EC, Wierman ME. Reexamination of testosterone, dihydrotestosterone, estradiol and estrone levels across the menstrual cycle and in postmenopausal women measured by liquid chromatography-tandem mass spectrometry. Steroids.2011;76:177–82.

24- Chia S, Gradishar W, Mauriac L, et al. Double-blind, randomized placebo controlled trial of fulvestrant compared with exemestane after prior nonsteroidal aromatase inhibitor therapy in postmenopausal women with hormone receptor-positive, advanced breast cancer: results from EFECT.J Clin Oncol.2008; 26:1664.

25- Early Breast Cancer Trialists' Collaborative Group (EBCTCG), Davies C, Godwin J, Gray R, Clarke M, Cutter D, et al. Relevance of breast cancer hormone receptors and other factors to the efficacy of adjuvant tamoxifen: Patient-level meta-analysis of randomised trials. Lancet.2011, 378(9793):771–84.

26- Yoko O and Hirotaka I. Clinical significance of estrogen receptor β in breast and prostate cancer from biological aspects. Cancer Sci.2015; 106(4): 337–343.