RESEARCH COMMUNICATION

Role of Exogenous and Endogenous Sources of Estrogen on the Incidence of Breast Fibroadenoma: Case-control Study in Iran

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Abstract

Breast fibroadenoma (FAD) is the most common benign mammary condition among women but the environmental risk factors have not identified yet. As the role of long term estrogen exposure in the incidence of FAD has been remained controversial; we have decided to investigate the possible role of endogenous and exogenous sources of estrogens in present study. Women less than 45 years old who underwent surgery from June 2009 to June 2010 were matched with controls by age and hospital. From reproductive factors, lack of breast feeding (p<0.001, 8.76 CI95% 3.79-20.24), Nulliparity (p=0.001, OR=8.09, CI95% 3.505-18.67), Lack of parity (p=0.001, OR=6.64, CI 95% 2.56-16.31) and Hormonal dysfunction (p=0.016, OR=4.66, CI 95% 1.26-17.28) were considered as the most important ones. Adiposity and abnormal weight gain after 18 years were considered as major background factor which induce FAD and may be contributed to the level of endogenous estrogen. Out of evaluated exogenous sources of estrogen, lower age at first OCP consumption (20.76_+3.87 vs. 22.85_+3.88, p=0.046) and living near Polycyclic aromatic hydrocarbons (PAHs) producing factories (p<0.001, OR=3.7, CI95%1.61-7.94), were considered as the main sources of exposure to xenestrogens in FAD patients but FAD showed inverse association with cigarette smoking because of antiestrogenic activities of cigarette smoking . This study concludes that the incidence and development of FAD could be associated with the reproductive history of women, activity of ovarian hormones as well as environmental factors.

Keywords: Breast fibroadenoma - estrogen - environmental risk factors - Iran - PAHs

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Introduction

Cancer represents only about 5% of clinical abnormalities of the breast and the rest of abnormalities are benign conditions especially breast fibroadenomas (FAD) which is the most prevalent benign tumor of the breast (Darbre, 2006) .The reason for such a high incidence of mentioned condition is unknown (Coriaty et al.,. 2002) but they are of concern because the chance for developing breast cancer (BC) from history of FAD has remained controversial. Some literatures suggest their presence can be an indicator of future development of BC (Dixon et al., 1999; Dorjgochoo et al., 2008) and some other studies reject this hypothesis and showed how the risk of developing breast cancer is equal to the other women in getting BC (Houssami et al., 2001) moreover the absence of frequent methylation supports a possible non-neoplastic origin for FAD (Huang et al., 2010).

Epidemiological studies show that 90% of BC could be originated from environmental pollutions and the main factor in the development of BC is the cumulative exposure to endogenous and exogenous sources of estrogens (Darbre, 2006) as well as Aryl hydrocarbon receptor (AhR), ligands like dioxin, PCBs, PAHs etc. which mimics estrogenic activities (Van der Heiden et al., 2009).Long-term estrogen-exposed breast cells, in comparison with control cells, showed higher levels of AhR in vitro (Wong et al., 2009) .We showed recently that the higher levels of AhR in young premenopausal BC and FAD for the first time which deregulate the expression of other tumor proliferative genes and increases the risk of tumor growth in both tumor types (Arbabi Bidgoli et al., 2010).

Although the direct role of estrogen on FAD incidence has not clarified and none of the steroid hormone receptors has been expressed in the FAD (Sapino et al., 2006; Arbabi Bidgoli et al., 2010) , an estrogen dependency has been suggested for the growth of FAD. In fact ER-beta is the only hormone receptor expressed by stroma of FAD both at protein and mRNA levels (Vera-Sempere et al., 1997). FAD in young patients with highly ER-beta-positive stroma cells, indicate a hormone-receptor mechanism involved in regulating the growth of FAD (Sapino et al., 2006).

Risk of BC could be increased by increased exposure to endogenous estrogens from early onset of menarche, late onset of menopause, nulliparity, late age of first pregnancy, lack of breastfeeding, taking oral contraceptive

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pill or hormone replacement therapy (Van der Heiden et al., 2009) but the role of these endogenous factors on the incidence of FAD is still controversial. Other than physiological steroidal estrogens, many compounds have now been found to have estrogenic activity which may increase the risk of exposure of human breast cells to environmental estrogens including pharmacological estrogens, plant estrogens (phyto-estrogens) and man-made estrogen-mimicking chemicals (xeno-estrogens) (Pasqualini et al., 1997; Darbre, 2006). We have decided to investigate the possible role of endogenous and exogenous estrogen sources by recording reproductive and lifestyle factors on the incidence of FAD by conducting a case control study in Tehran, the capital city of Iran.

Materials and Methods

Study population

We conducted a case-control study to search for any relationship between exogenous or endogenous sources of estrogens and development of FAD. Eligible participants for this study were young (premenopausal) women who underwent surgery in Imam Khomeini medical complex from June 2009 to June 2010. They matched with controls by age and hospital. They had completed study questionnaires by home interviews elicited information on reproductive and other host and environmental factors. Participants of this study were not currently taking hormonal contraceptives (including oral contraceptives, contraceptives implanted subdermally, and progestin releasing intrauterine devices), had not had a hysterectomy, and were not currently pregnant or breast feeding. The study population comprised 150 women, including 50 with FAP and 100 normal women who were Muslim, Iranian and lived in Tehran since their birth. Exact tumor types in benign breast tumors were identified from both self-reports and hospital registration.

Exclusion criteria

Validation of the diagnoses of FAD was performed in the first step. A reference pathologist reviewed slides from selected cases .Of 54 FAD diagnosed by a local pathologist in Tehran, 50 were also confirmed by the reference pathologist as fibroadenoma. Exclusion criteria for cases and controls were menopausal evidence, recent evidence of pregnancy and lactation. Menopausal status at the time of recruitment was defined according to information on ovariectomy, hysterectomy, mensturation status (still mensturating, number of menses over the past 12 months) .Women were considered postmenopausal if they had undergone a bilateral ovariectomy or if their menses had stopped since 12 months or more (unless due to hysterectomy). Women who were still mensturating by using exogenous hormones and women with no information on the number of menses over the past 12 months were also excluded from this study. The control group was matched with cases for age +/- 5 years.

Endogenous estrogen assessment

Status of background endogenous estrogen levels were evaluated by recording the following variables: A) Delivery related factors including patient's age, mother's age, father's age, mother's weight and birth weigh at delivery; B) Mensturation related factors including age and weight at menarche and irregular mensturation; C) Marriage related factors including age at first parity, age at marriage, present marital status; D) Pregnancy related factors including nulliparity, age at first full term pregnancy, age at second full term pregnancy, maximum weight gain at each pregnancy, weight changes after first delivery, weight changes after second delivery, duration of oral contraceptive use ,type and dosage of OCP ,age at first Oral Contraceptives Pills (OCP) consumption, other methods of child control (IUD, tubectomy, progestins), number of live births ,age at first breast feeding period , frequency of breast feeding, duration of breast feeding and history of infertility, history of reproductive disorders covered questions on years of infertility (>2years without birth controlling methods), history of benign breast disease in patient of first degree family ,history of malignant breast disease in first degree family, ovarian cycst and hyrsutism.

Exogenous estrogens

Sources of environmental estrogens were recorded on the basis of the administration of pharmacological estrogens e.g. OCP, Hormone Replacement Therapy (HRT) and use of ovulation stimulating drugs.

Other lifestyle factors

Personal history of endocrine disorders ,background diseases ,pattern of physical activity, occupations, history of psychological disorders, insomnia, severe stressful conditions(e.g death of first degree relative, severe trauma etc) and anxiety, smoking (active vs. passive) ,alcohol consumption, using psychoactive drugs, use of oral supplements including calcium ,Vitamins ,Omega radiation exposure, exact BMI and weight gain profile after 18 were recorded by pretested specific questionnaire.

Statistical methods

Values were expressed as percent per population or as the mean± standard deviation (SD).To assess the association between expressions of markers and clinicopathological data nonparametric chi square test was used. Relative risks and Odds ratios were calculated by Cochran's and Mantel Haenszel statistics using SPSS 16. Probability values of <0.05 and odds ratios >1 were considered significant

Results

Endogenous estrogens

At the first stage possible sources of elevating the endogenous levels of estrogens were assessed by recording the reproductive history of FAD and compared the variables with control group. Following factors were considered as possible reproductive risk factors of FAD.

Delivery related factors

Out of different delivery related factors which were

Table 1. Comparison of Reproductive and Other Factors between Fibroadenoma Cases and Controls

Characteristics	Cases	Controls	P	OR CI 95%
Age	29.2 (8.3)	33.2 (5.6)	NS	
Delivery factors				
Mother's age	26.4 (6.7)	26.5 (6.8)	NS	
Father's age	32.5 (7.7)	33.9 (8.4)	NS	
Weight >4 kg	8 (16%)	11(11%)	NS	
Menarche and menstruation factors				
Age	13.2 (1.5)	13.2 (1.5)	NS	
Overweight	17 (34%)	18(18%)	NS	
Irregular	26 (52%)	32(32%)	0.014	1.34 1.03-1.74
Marriage factors				
Married	32 (64%)	92 (92%)	<0.001	6.46 2.56-16.31
Age	20.5 (3.9)	22.9 (4.1)	0.014	2.84 1.23-6.53
Pregnancy factors				
Nulliparity	25 (50%)	11 (11%)	<0.001	8.09 3.51-18.7
Age[1]	21.8 (4.1)	25.0 (4.2)	0.023	2.61 1.08-6.30
Abortion	6 (18%)	16(16%)	NS	
Infertility	0 (0%)	2(2%)	NS	
Breastfeeding				
Lack	26 (52%)	11 (11%)	<0.001	8.76 3.79-20.2
<1 year	3 (6%)	19 (19%)	0.025	3.67 1.03-13.`
1-2 years	3 (6%)	19 (19%)	0.025*	3.67 1.03-13.1
>2 years	18 (36%)	51 (51%)	NS	
Anthropomorphic factors				
Height	161 (6.7)	163 (5.5)	NS	
Weight	70 (12.5)	64 (10.5)	0.008	5.58 1.49-9.68
25-30 kg/m^2	18 (36%)	32 (32%)	NS	
≥ 30 kg/m^2	11 (22%)	8 (8%)	0.028	2.45 1.04-3.03
Weight gain after 18				
No change	3 (6%)	16 (16%)	NS	
< 10 kg	8 (16%)	26 (26%)		
10-20 kg	21 (42%)	42 (42%)		
20-30 kg	14 (28%)	14 (14%)	0.034	2.0 1.03-3.86
30-40 kg	2 (4%)	2 (2%)	NS	
> 40 kg	2 (4%)	0 (0%)		
Physical activity				
Lack	32 (64%)	60 (60%)	NS	
History of background disease				
Depression	12 (24%)	29 (29%)	NS	
Anemia	4 (8%)	25 (25%)	NS	
Thyroid[2]	7 (14%)	12 (24%)	NS	
CNS[2]	5 (10%)	7 (7%)	NS	
Insomnia	24 (48%)	40 (40%)	NS	
Stress	42 (84%)	59 (59%)	0.001	1.43 1.16-1.76

[1]at first full term pregnancy; [2]dysfunction

compared in Table 1, mother's BMI at delivery was considered significantly higher in FAD than control group. Most of women who got FAD were born from overweight mothers (BMI>25) when compared with normal group (58% vs. 21%, p=0.001, OR= 1.88 , CI95% 1.33-2.645).

Menstruation related factors

From menstural related parameters, history of irregular mensturation was more prevalent in FAD when we compared the same history with the control group (p = 0.014, OR = 1.34, CI 95% 1.031-1.741). In fact 52% of FAD showed clear history of irregular mensturation whereas the same pattern was 32% in normal group.

Parity related factors

Interestingly FAD was more prevalent in single women that mean 8% of normal women were single whereas 36 % of cases were single (p=0.001, OR=6.64, CI 95% 2.56-16.31). Married FAD had significant lower ages at parity) 20.53+3.927 vs. 22.93+4.1, p=0.014, OR=2.84, CI 95% 1.23-6.53) at the same time early marriage (<21 yrs) were considered as another possible risk factor of FAD in married women that means 60% of married women with FAD got married before 21 yrs whereas only 36.9% of control group had early marriage in their reproductive history.

Pregnancy related factors

It seems that history of full term pregnancy and live child birth could reduce the risk of FAD. In fact 50% of FADs were nulliparous whereas the same situation was 11% in control group (p<0.001, OR =8.09, CI 95% 3.505-18.67). The age of the first full term pregnancy was significantly lower in FAD than control (21.84(4.14) vs. 25.03(4.24), p= 0.023 , OR=2.61 CI 95% 1.08-6.03).

Our data suggest the protective role of breast feeding against FAD too. In fact 52% of FADs had no experience of breast feeding while the same pattern was 11% in normal group (p<0.001, OR= 8.76 , CI 95% 3.79-20.24) .

History of reproductive disorders

We considered history of hirsutism, ovarian cysts, habitual abortion and infertility in this part of study. Signs of mentioned abnormalities were considered as hormonal dysfunction which were detected in 14% of cases comparing to 3% in controls (p=0.016, OR=4.66, CI 95% 1.26-17.28).

Pharmacological estrogens

OCP consumption and duration of use were not directly related to the increased risk of FAD, but lower age at first OCP consumption was recorded in FAD group (20.5+3.55 yrs vs. 22.9+ 3.81 yrs, p=0.034, OR= 2.58, CI 95% 1.06-4.11). None of the cases and control groups was exposed to Hormone Replacement Therapy (HRT)

Weight gain and Body Mass Index

Cases were heavier than controls (69.9+12.54 vs. 64.3+ 10.45) in this study (p=0.008, OR=5.58,CI 95%1.49-9.68).The prevalence of adiposity (BMI ≥30 kg/m^2) was 22% in cases while the same pattern was 8% in controls (p=0.028,OR=2.45,CI 95% 1.04-3.03).Risk of FAD was increased in women who had history of massive weight gain (20-30 Kg) after 18 years (28% vs. 14% ,p=0.034,OR=2.00,CI 95% 1.03-3.86).

Background diseases

Out of 6 different evaluated diseases the prevalence of severe stress was significantly higher in cases than control (84% vs. 59%, p=0.001, OR=1.43 CI 95%1.16-1.76).

Environmental factors

Except dietary factors which have been recorded and reported in a parallel study exposure to xenoestrogens as AhR agonists have been summarized in Figure 1. Interestingly 38% of FAD had history of living near PAH producing factories whereas only 5% of controls lived in the same areas(p=0.001,OR=3.7,CI95%1.61-7.94).

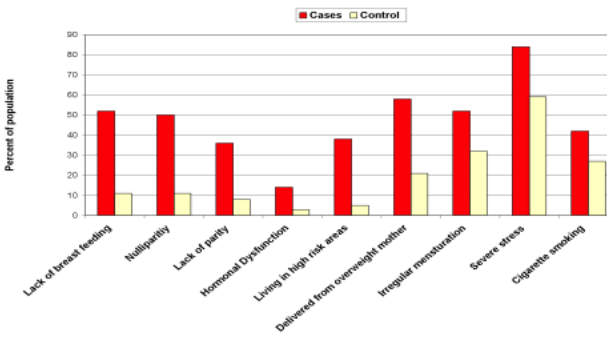

Figure 1. Sources of Endogenous and Exogenous Estrogens as Risk Factors for Breast Fibroadenomas

On the other side 42% of cases were exposed to cigarette smoke as passive smokers whereas the same situation were seen in 27% of control group (p=0.048, OR=0.511, CI95% 0.25-1.43). The prevalence of using plastic dishes in each group was low and didn't show any significant difference.

Other factors

Pattern of physical activity, occupation of women, history of psychological disorders, insomnia, alcohol consumption, using psychoactive drugs and radiation exposure were compared between cases and control group and no significant difference were detected between cases and control group.

Discussion

This study was conducted to estimate the role of exogenous and endogenous estrogens on the incidence of fibroadenoma in Iranian population to identify the risk factors for the most common benign mammary tumor among premenopausal women. To our knowledge, this is the first study which was conducted on more than 100 recordable estrogenic variables. FAD has an inverse association with increased age (Coriaty Nelson et al., 2002; Rego et al., 2009) therefore present study was focused on young premenopausal (<45 yrs) women. The results of this study supports the conclusion that the incidence and development of fibroadenoma is dependent on the reproductive history and presence of ovarian hormones in young premenopausal women but environmental factors may affect the endogenous levels of estrogen and cause hormonal dysfunction, menstural irregulation which may increase the risk of FAD and some lifestyle factors with antiestrogenic effects could decrease the risk when compared with normal group.

From the reproductive background of women, decreasing rate of parity, nulliparity and lack of breast feeding were considered as three major risk factors of FAD. Although the clinical and morphological characteristics of FAD often vary and the endocrine factors have been implicated in their etiology (Rose et al., 2009), the main risk factors of FAD are the level of endogenous estrogens. Although prior studies on the risk of FAD in relation to parity have yielded inconsistent results (Dorjgochoo et al., 2008) , a lower age at parity was considered as another risk factor of FAD when compared with control. Nevertheless the history of early live child birth and breast feeding showed very important protective roles on the incidence of FAD which is in accordance with other studies (Dijkstra et al 2010). It seems that early pregnancy prepares the breast for lactation, in the meanwhile estrogens and other circulating hormones cause the rapid proliferation of epithelial breast tissue, followed by hormonally driven differentiation of the mammary epithelium. It is possible that this early differentiation protect epithelial cells from the development of FAD, which is the outcome of estrogen dependent hyperplasic processes (Bundred 1994 , Pasqualini 1997).

This study suggest the role of higher body weight , BMI>30 and adiposity, and 20-30 kg weight gain after 18 years as three other important origins of FAD. Overweight/obesity can be experimentally induced by environmental factors like benzo [a]pyrene and endocrine disruptive chemicals with estrogenic activities and well characterized chemical pollutants (Irigaray et al., 2007) which are AhR ligands (Van der Heiden et al., 2009). Adipose tissue acts as a storage organ for lipophilic, liposoluble endocrine disruptive chemicals, so that chemical pollution may generate both overweight/obesity and tumor initiation. Many carcinogens, mutagens or promoters can be stored in the adipose tissue, be released at convenient dose in the blood circulation and therefore target peripheral tissues to induce breast tumors (Rose et al., 2009). Some studies suggested high tissue levels of adiponectin levels (a new adipocyte-secreted protein) in breast cancer patients which was associated with an increased risk of breast cancer (Karaduman et al., 2007; Paz-Filho et al., 2011). Although there is no study on the association between adiponectin and breast fibroadenoma ,this study shows increasing evidence that obesity may have a significant role in the pathophysiology of FAD too. Increased tissue levels of adipokines as well as its association with AhR levels should be analyzed in the future studies.

It was concluded from present study that the incidence of fibroadenoma is not greater in women who used oral contraceptives even in longer periods ; even though the protective effects of OCPs containing >50 mcg estrogen (but not for progestogen-only OCPs) was suggested too (Vessey and Yeates. 2007) .

Similar to previous reports (Berkowitz et al 1985., Rohan et al., 1999,), this study showed also of a reduced risk of FAD associated with cigarette smoking. Due to the small numbers of women who were cigarette smokers, which is consistent with the smoking habits of the female Iranian population, the rest of smokers were passive smokers in both groups. Cigarette smoking has been found to alter the risk of several estrogen-related conditions including endometrial cancer, uterine fibroids, and osteoporosis (Spangler et al., 1999), because smokers tend to have lower levels of endogenous estrogen than do nonsmokers (Jensen et al, 1985), and because fibroadenoma is believed to be stimulated by estrogen (Dorjgochoo et al., 2008), an association between smoking and fibroadenoma is biologically plausible. Further investigation on the effect of this exposure on the risk of fibroadenoma seems necessary.

The summary of the roles of endogenous and exogenous estrogens on the incidence of FAD have been summarized in Figure 1 .Although the role of endogenous estrogens are dominant in this phenomenon, several findings of present study support the role of environmental factors on the incidence of FAD too. Higher prevalence of FAD in women who lived in polluted and high risk areas and near PAH producing factories which releases AhR ligands to the environment emphasize our last finding (Arbabi Bidgoli et al., 2010) on the overexpression of AhR in FAD which was correlated with the age of patients. Other than all mentioned factors, severe stress which is able to increase the endogenous levels of estrogen was considered an another possible this regard but further investigation is necessary to find the role of psychological disorders in the incidence of FAD. As we showed in Fig 1 this study concludes that the incidence and development of FAD could be associated with the reproductive history of women, activity of ovarian hormones as well as environmental factors.

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