Risk Factors of Osteoporosis in Females: A Hospital-Based Case-Control Study, Yazd, Iran

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Abstract

Background: The aim of this study was to evaluate the predictors of osteoporosis in women in Yazd, Iran.

Methods: This hospital-based case-control study was performed on 270 women 35-65 yr old (135 case and 135 control) from Mar 2016 to Mar 2017. Case and control were matched in terms of age ± 2 (year) as a group matching. Osteoporosis is defined as a T-score of bone mineral density (BMD) below –2.5 SD. Data were analyzed using SPSS 20 software. Statistical tests included chi-square, student t test and Mann Whitney test. Multiple logistic regression (MLR) which forward method was used for modeling.

Results: Odds ratio (OR) of osteoporosis were menarche age>12 yr (OR=3.37, CI:2.29-15.89), history of hysterectomy (OR=13, CI:3.81-44.82), rheumatoid arthritis (RA) (OR=6.58, CI:2.29-18.91) calcium supplements (OR=0.14, CI:0.04-0.41), menopause age <40 (OR=11.84, CI:1.54-90), second smoking (OR=3.38, CI:1.16-9.81) and increase of weight (OR=0.86, CI:0.80-0.94).

Conclusion: Predictors of osteoporosis was menarche age >12 yr (3 times), history of hysterectomy (13 times), RA (6.5 times), menopause age <40 (12 times), second smoking (more than 3 times), calcium supplements and weight (protective).

Keywords: Osteoporosis; Women; Case-control study; Iran

Introduction

The trend of developing non-communicable diseases has changed along with changes in people's lifestyles and aging (1). Osteoporosis is one of the chronic diseases; associated with a decrease in bone mineral density (BMD)(2).

Osteoporosis causes a decrease in bone mass, microstructural changes in bone tissue, and finally bone fractures. The reason for the importance of this disease is bone fractures (3).

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BMD is measured by dual-energy x-ray absorptiometry (DXA) (4). Osteoporosis is defined as a T-score of BMD below −2.5 SD (5). The prevalence of osteoporosis in women in both Europe and the United States is 30% (6). The prevalence of osteoporosis in different countries has different variation value. Its value varies from 9% in the UK to 38% in Japan (7). Based on DALY scale osteoporosis is responsible for more than 36,000 years of loss of life for Iranian men and women. Approximately, 85% of the global burden of osteoporosis and 12.4% of the burden of osteoporotic fractures in the Middle East were related to Iran in 2015 (8-10).

Osteoporosis is a multifactorial disease. Age, sex, body mass index (BMI), smoking, physical activity, glucocorticoid intake, and diseases such as rheumatoid arthritis (RA) are most important risk factors of osteoporosis (11-13). The reason for osteoporosis in women is a deficiency of steroid hormone in menopause (14). Osteoporosis can increase the fragility of the skeleton and the risk of fracture by accelerating bone turnover and decreased bone mass (13). The best way to prevent the complications of osteoporosis is to educate people to change their lifestyle (change their eating habits and intake calcium and vitamin D) (15).

Although osteoporosis is a silent disease and less attention is paid to; while bringing high costs to families. Therefore, it can be very important. However, the risk factors for osteoporosis have been studied in various studies in the world and in Iran; however, by the odds ratio in the case-control study, the strength of the association between two events can be calculated. The strength of the association can also vary in geographical areas and different people for environmental and cultural reasons. We aimed to evaluate the predictors of osteoporosis in women in Yazd, Iran.

Methods

**Study Design and Participants**

This hospital-based case-control study was performed on 270 women 35-65 yr old. The sample size for the study was calculated using the sample size formula of the case-control studies and based on the physical activity Odds Ratio (OR=2.2)(16), according to the following formula \( P_2 = \frac{P_1 \times OR}{1 + P_1 \times (OR-1)} \). Then, with the following formula and type I error of 0.05 and type II error of 0.20 and 20% attrition, 135 individuals were calculated in each group. Thus, a total of 270 patients (135 cases and 135 controls) were randomly selected. \( P_1 = 0.40 \) and \( P_2 = 0.57 \).

\[
 n = \frac{(z_{1-a/2} + z_{1-\beta})^2 |P_1(1 - P_2)|}{(P_1 - P_2)^2}
\]

The sampling method was also simple random using the random number table. Among 3000 patients referred to the Yazd Khatam Al Anbia Clinic, 270 patients (135 case and 135 control) were randomly selected from Mar 2016 to Mar 2017. Case and control were matched in terms of age ± 2 (year) as a group matching. The definition of cases and controls were as follows: Cases are defined as patients with a T-score of BMD below −2.5 SD. Controls are defined as participants with a T-score of BMD −1 SD or more (5).

Inclusion criteria were 1) live in Yazd, 2) aged 35-65 yr and 3) consent to participate in this study. Participants were invited by an epidemiologist to their nearest comprehensive health center by telephone to complete the questionnaire. Finally, questionnaires were completed for them.

BMD densitometry data were obtained by Dual Energy X-Ray Absorptiometry method (DEXA) in femoral neck and lumbar vertebrae (L2-L4) in Yazd Khatam Al Anbia Clinic. BMD densitometry has been done for the case and control groups. In this study, osteoporosis is defined as a T-score of BMD below −2.5 SD (5).

**Data collection**

A checklist consisting of 3 sections (demographic information, lifestyle, and medical history) was completed for case and control groups. The socioeconomic status questionnaire was self-made questionnaire. Result of socioeconomic status published in a separate article (17). BMI was cat-
categorized according to a WHO report (18). Definition of overweight was a BMI greater than or equal to 25; and definition of obesity was a BMI greater than or equal to $30 \frac{kg}{m^2}$.

Daily physical activity was assessed using international physical activity questionnaire (IPAQ). According to a study, IPAQ has good content validity (CVI=0.85 and CVR=0.77), internal consistency (Cronbach’s Alpha coefficient=0.7) and reliability (Spearman Brown correlation coefficient=0.9) (19).

Dietary intakes during the past year were assessed using validated semi-quantitative food frequency questionnaire (FFQ). The validity and reliability of this questionnaire were calculated in the study of Isfahani et al. (Mean of spearman correlation coefficients was 0.44 in men and 0.42 in women) (20).

Statistics analysis
After collecting data and performing the quality control, data were analyzed using SPSS 20 software (IBM Corp., Armonk, NY, USA). According to the research objectives, data were described with ratio, mean, median, standard deviation and also statistical tests such as chi-square (for qualitative variables), student t-test (for quantitative variables) and Mann Whitney test (for non-parametric analysis). Multiple logistic regression (MLR) which backward method was used for modeling. Group matching was conducted in our study. Therefore, modeling was performed twice. First time without age variable and second time with entering age variable. Significance level (95%) was used to interpret the results to determine the predictors of osteoporosis. In the analysis, the coefficient of determination ($R^2$) was calculated to be 87.4%. That is, the predictor variables were able to explain 87.4% of the changes in the dependent variable.

Ethics
This study was approved by the Code of Ethics IR.SSU.SPH.REC.1395.141 of Shahid Sadoughi University of Medical Sciences in Yazd, Iran.

Results
Demographic and behavioral factors
This case-control study was designed on 270 women aged 35-65 years. T-score prevalence of osteoporosis, in terms T-score of total spine, hip neck, and total hip were 69.6%, 56.6% and 49.3%, respectively. Demographic factors are listed in Table 1.

| Group                          | Variable | Case N(%) | Control N(%) | P-value* |
|-------------------------------|----------|-----------|--------------|----------|
| Marital status                | Married  | 111(82.2) | 112(83)      | 0.52     |
|                               | Unmarried| 24(17.8)  | 23(17)       |          |
| Age (yr)                      | 35-45    | 14(10.4)  | 16(11.9)     | 0.85     |
|                               | 46-55    | 64(47.4)  | 66(48.9)     |          |
|                               | 56-65    | 57(42.2)  | 53(39.3)     |          |
| BMI                           | <25      | 35(25.9)  | 11(8.1)      | <0.001   |
|                               | 25-30    | 59(43.7)  | 50(37)       |          |
|                               | >30      | 41(30.4)  | 74(54.8)     |          |
| Socio-economic level          | Low      | 87(64.4)  | 44(32.6)     | <0.001   |
|                               | Moderate | 12(8.9)   | 11(8.1)      |          |
|                               | High     | 36(13.7)  | 30(11.1)     |          |

*Chi-square test
Overall, 39.3% of case subjects and 21% of control subjects were exposed to cigarette smoke during the week (Second-smoking). The odds ratio of morbidity of osteoporosis was (OR=3.50, CI =1.96-6.26) in second smokers; which was statistically significant ($P<$0.001). Proportion of moderate physical activity was 91.9% in the case group and this proportion was 76.3% in the control group (Table 2).

### Table 2: Behavioral factors in case and control group

| Group       | Variable     | Case N(%) | Control N(%) | Crude OR(CI)* |
|-------------|--------------|-----------|--------------|---------------|
|             |              | N         | N            |               |
| Hookah con-| Yes          | 3(2.2)    | 2(1.5)       | 1.51(0.24-9.19)|
| sumption    | No           | 132(97.8) | 133(98.5)    | 1             |
| Second      | Yes          | 53(39.3)  | 21(15.6)     | 3.50(1.96-6.26)|
| smoking     | No           | 82(60.7)  | 114(84.4)    | 1             |
| Physical ac-| Low          | 3(2.2)    | 6(4.4)       | 1.62(0.32-8.02)|
| tivity      | Moderate     | 124(91.9) | 103(76.3)    | 6.91(1.69-9.01)|
|             | High         | 8(5.9)    | 26(19.3)     | 1             |

*Univariate analysis (Crude OR)

31.1% of cases and 41.5% of controls had walking in leisure time or Commuting. And the odds of having an osteoporosis was 0.63 for walking ($P=0.07$). The median minutes of walking in the case and control groups were 30 and 60 min per week, respectively. There was a significant difference between the two groups in terms of it ($P=0.001$). The median number of walking days the metabolic equivalent of physical activity (MET) was 198 and 347 kcal/kg/hour in the case and control groups, respectively, which was not statistically significant ($P=0.08$). The median moderate activity of MET were higher in the control group than in the case group (1800 and 1440 kcal/kg/hour, respectively) ($P<0.001$).

**Reproductive factors**

The mean age of menopause in the case group was 44.74 $\pm$ 5.33 and in the control group it was 48.015$\pm$5.20 which showed a significant difference between the two groups ($P<0.001$). Comparison of menopausal status between two groups, was showed 87.4% of the case group and 49.6% of the control group were menopause; which was statistically significant ($P<0.001$). Menopausal age was between 40-50 yr old in the in case group (59.3%), but most participants in the control group (50.4%) were not menopausal. Highest chance of developing osteoporosis was 5.72 (CI: 2-16.35) in those who menopaus before 40 yr old; which was also statistically significant ($P<0.001$).

The mean of duration of breastfeeding in the case group was 34.66$\pm$3.84 (month) and in the control group was 22.05$\pm$3.06; which was significantly different between two groups ($P=0.005$). Mean age of menarche in the case group was 13.57/1$\pm$1.41 and in the control group was 12.19/1 $\pm$1.75 which was significantly different between two groups ($P<0.001$). Percentage of history of 3-5 times pregnancy was calculated 54.5% in the case group and 65.2% in the control group ($P<0.001$). Reproductive factors in case and control group is in Table 3.
Table 3: Reproductive factors in case and control group

| Group                  | Variable     | Case N(%) | Control N(%) | Crude OR (CI)* |
|------------------------|--------------|-----------|--------------|----------------|
| Menopause              | Yes          | 118(87.4) | 67(49.6)     | 7.04(3.82-12.96) |
|                        | No           | 17(12.6)  | 68(50.4)     | 1              |
| Menopause age (year)   | No menopause | 17(12.6)  | 68(50.4)     | 0.47(0.19-1.7)  |
|                        | <40          | 27(20)    | 9(6.7)       | 5.72(2-16.35)   |
|                        | 41-50        | 80(59.3)  | 37(27.4)     | 4.12(1.80-9.43) |
|                        | >51          | 11(8.1)   | 21(15.6)     | 1              |
| Breastfeeding (Month)  | Yes          | 129(95.6) | 120(88.9)    | 2.68(1.01-7.15) |
|                        | No           | 6(4.4)    | 15(11.1)     | 1              |
| During of breastfeeding (years) | No breastfeeding | 6(4.4) | 16(11.9) | 1 |
|                        | <5           | 102(75.6) | 105(77.8)    | 2.59(0.97-6.88) |
|                        | >5           | 27(20)    | 14(10.4)     | 5.14(1.64-16.06) |
| Menarche age (year)    | <12          | 30(22.2)  | 75(55.6)     | 1              |
|                        | >12          | 105(77.8) | 60(44.4)     | 4.37(2.57-7.42) |
| Number of pregnancy    | ≤ 2          | 11(8.1)   | 27(20)       | 1              |
|                        | ≥ 5          | 73(54.1)  | 88(65.2)     | 2.03(0.94-4.38) |
|                        |              | 51(37.8)  | 20(14.8)     | 6.25(2.61-15)   |
| History of hysterectomy| Yes         | 52(38.5)  | 12(8.9)      | 6.42(3.23-12.76) |
|                        | No           | 83(61.5)  | 123(91.1)    | 1              |
| History of oophorectomy| Yes         | 23(17)    | 22(16.3)     | 1.05(0.55-2)    |
|                        | No           | 112(83)   | 113(83.7)    | 1              |
| History of Menstruation Disorders | Yes | 30(22.2) | 44(32.6) | 0.59(0.34-1.01) |
|                        | No           | 105(77.8) | 91(67.4)     | 1              |

*Univariate MLR analysis (Crude OR)

**History of the disease, fracture, using drugs and supplements, hormone therapy and dietary calcium intake**

Mean duration of rheumatoid arthritis in case group was 6.85±4.21 and in the control group was 4.81±4.16 which was statistically significant (P=0.03). Fifty seven percent of the case group had a history of rheumatoid arthritis. 18.5% of case group and 6.7% of control group had positive history of fracture. There was a significant difference between the two groups in this regard (P=0.005). The odds ratio of osteoporosis was calculated (OR=3.72, CI: 1.74-74.95) in individuals with a history of fracture in the first-degree family. The prevalence of calcium supplements, vitamin D, multi vitamin, glucocorticoid and oral contraceptive pill (OCP) in the case group was 15.60, 15.60, 7.40, 31.9 and 16.30 respectively (Table 4).
Table 4: History of the disease, fracture, using drugs and supplements, hormone therapy and dietary calcium intake

| Group           | Variable          | Case N(%) | Control N(%) | OR(CI)*          |
|-----------------|-------------------|-----------|--------------|------------------|
| Diabetes        | Yes               | 23(17)    | 17(12.26)    | 1.42(0.72-2.80)  |
|                 | No                | 112(83)   | 118(87.40)   |                  |
| Rheumatoid Arthritis | Yes         | 77(57)    | 25(18.70)    | 5.87(3.33-10.05) |
|                 | No                | 58(43)    | 109(81.30)   |                  |
| Hypothyroidism  | Yes               | 13(10.10) | 12(8.90)     | 1.14(0.50-2.62)  |
|                 | No                | 116(89.90)| 123(91.10)   |                  |
| Hypothyroidism  | Yes               | 10(7.40)  | 7(5.60)      | 1.33(0.49-3.62)  |
|                 | No                | 125(92.60)| 117(94.40)   |                  |
| History of fractures | Yes           | 25(18.50) | 9(6.70)      | 3.18(1.42-7.1)   |
|                 | No                | 110(18.50)| 126(93.3)    |                  |
| Family history of fractures | Yes               | 31(23)    | 10(7.40)     | 3.72(1.74-7.95)  |
|                 | No                | 104(77)   | 125(92.60)   |                  |
| Calcium supplements | Yes            | 21(15.60) | 53(39.30)    | 0.28(0.16-0.50)  |
|                 | No                | 114(84.40)| 82(60.70)    |                  |
| Vitamin D       | Yes               | 21(15.60) | 60(44.40)    | 0.23(0.12-0.14)  |
|                 | No                | 114(84.40)| 75(55.60)    |                  |
| Multi vitamin   | Yes               | 10(7.40)  | 16(11.90)    | 0.64(0.27-1.47)  |
|                 | No                | 125(92.60)| 119(88.10)   |                  |
| Glucocortico-   | Yes               | 43(31.9)  | 26(19.30)    | 1.95(1.11-3.43)  |
| coed            | No                | 92(68.10) | 109(80.70)   |                  |
| OCP             | Yes               | 22(16.30) | 18(13.30)    | 1.26(0.64-2.48)  |
|                 | No                | 113(83.70)| 117(86.70)   |                  |
| Hormone Therapy | Yes               | 7(5.20)   | 3(2.20)      | 2.38(0.60-9.43)  |
|                 | No                | 128(94.80)| 131(97.80)   |                  |
| Dietary calcium intake | <500         | 68(50.40) | 18(13.30)    | 4.91(1.85-13.01) |
|                 | 500-1000          | 57(42.20) | 104(77)      | 0.71(0.29-1.72)  |
|                 | >1000             | 10(7.40)  | 13(9.60)     |                  |

Univariate analysis (Crude OR)

Results of modeling

By entering all variables into the MLR, many variables were excluded from the model. Significant risk factors were included menarche age >12 yr, history of hysterectomy, RA, menopause age <40, during of breastfeeding >5, second smoking. Significant protective factors were included calcium supplements and weight (Table 5).
Table 5: The odds ratio of the factors in modeling

| Group                | Variable                  | OR (CI)*       | OR Adjusted(CI)** |
|----------------------|---------------------------|---------------|------------------|
| Socio-economic level | Low                       | 2.34(0.95-5.75)| 2.33(0.90-6)     |
|                      | Moderate                  | 0.54(0.14-2.04)| 0.27(0.06-1.20) |
|                      | High                      | 1             | 1                |
| Menarche age (year)  | <12                       | 5.42(2.20-13.36)| 3.37(2.29-15.89) |
|                      | >12                       | 9.33(2.29-95.45)| 13(3.81-44.82)   |
| History of hysterectomy | Yes                    | 5.03(1.93-13.12)| 6.58(2.29-18.91) |
|                      | No                        | 1             | 1                |
| Rheumatoid Arthritis | Yes                      | 1.15(0.21-6.11)| 0.19(0.02-1.57)  |
|                      | No                        | 1             | 1                |
| Diet calcium intake  | <500                      | 3.95(0.79-19.69)| 3.97(0.69-22.73) |
|                      | 500-1000                  | 1.1(0.26-4.65)| 1.28(0.27-5.96)  |
|                      | >1000                     | 1             | 1                |
| Menopause age (year) | <40                       | 1.15(0.21-6.11)| 0.19(0.02-1.57)  |
|                      | 41-50                     | 21.24(3.28-70.13)| 11.84(1.54-90)  |
|                      | >51                       | 7.71(1.53-38.85)| 4.03(0.67-24)   |
|                      | No breastfeeding          | 1             | 1                |
|                      | <5                        | 1.55(0.35-3.75)| 1.15(0.24-5.34)  |
|                      | >5                        | 7.37(1.26-43.04)| 6.49(1.01-41)   |
| Second smoking       | Yes                       | 3.30(1.20-9.02)| 3.38(1.16-9.81)  |
|                      | No                        | 1             | 1                |
| Weight               | ----                      | 0.92(0.88-0.95)| 0.86(0.80-0.94)  |

*:Adjusted OR for All variables except age, **: Adjusted OR for All variables

Discussion

Osteoporosis is the common bone tissue disease and its importance is due to bone fractures that it can lead to death (18). Our case-control study was performed to evaluate the osteoporosis risk factors on 270 (135 cases and 135 controls) women’s aged 35-65 yr who referred to Khatam-al-Anbia clinic for densitometry.

In our study, the odds ratio of people with osteoporosis was more than four times that of people who had their first menstruation before the age of 12. That was statistically significant. In multivariate regression, this value increased by one unit, and the odds ratio increased more than six times after entering age in the model. The age of 11 and lower are associated with a reduced incidence of osteoporosis (21).

In univariate regression, the OR of those with a history of hysterectomy was more than six times that of those without history of hysterectomy. That was a significant difference between the two groups. In multivariate regression, the OR of those with a history of hysterectomy increased more than nine times and in model 2, this value has more than 13 times. Hysterectomy had a protective effect and it reduced the risk of osteoporosis by 32% (22). This study was cross-sectional study. And the calculated correlation was not statistically significant.

OR of osteoporosis in participants with history of RA was more than five times higher than participants without history of RA in the univariate and multivariate regression. There was a significant difference between the two groups in this.
regard. The history of RA was significantly associated with the risk of osteoporosis (15). In our study, calcium supplementation remained in the multivariate regression and even after age entry in the model and it retained its protective effect. Various studies have shown the association between calcium supplementation and BMD (23, 24).

There was a significant difference between the two groups regarding the mean age of menopause. In univariate regression, the odds ratio of osteoporosis in postmenopausal women was about seven times that of non-menopausal women, which was statistically significant. The OR of developing osteoporosis in menopausal women before age<40 was more than five times of non-menopausal women, which was statistically significant. In multivariate regression, those who were menopausal before the age<40 had a 11-fold higher risk of osteoporosis than those who did not. Menopause increases the risk of osteoporosis by about 30 times (25). Moreover, the duration of menopause longer than five years increases the chance of developing the disease more than twice (26). Osteoporosis was significantly associated with menopausal age less than 45 years (27).

In our study in multivariate regression, no significant relationship was found between during of breastfeeding and osteoporosis. Breastfeeding for more than two years, increase the OR of developing osteoporosis by 2.6 times (28). In Urmia, a significant relationship was found between the length of breastfeeding and osteoporosis (15). This could be because breastfeeding was high in both groups. While in cities like Tehran, breastfeeding mothers may be less.

In the univariate regression, the OR of developing osteoporosis in women that partner was smoker was more than 5 times greater in the lumbar spine and more than 4 times in the femoral neck (29). Smoking reduces estrogen levels and it leads to bone loss (30). In our study, in multivariate regression, weight had a protective relationship with the risk of osteoporosis. In southern Brazil, BMI ≥25 reduced the risk of osteoporosis by 64% (22). This protective effect may not indicate that being overweight reduces the risk of osteoporosis. People with higher BMD are also more likely to have a higher weight. Aging increases adipocytes and osteoclast activity. As a result, osteoblast activity is reduced and leads to osteoporosis. Therefore, this hypothesis rejects the previous concept, that obesity is protective for osteoporosis (31).

The strengths of our study were: 1. both case and control groups were selected from a single clinic and BMD was measured with a common device. Therefore, we have reduced the selection bias in our study. 2. The data collector was one person in both case and control groups. It therefore reduces the interviewer’s bias. 3. Age-matched and third modeling (adjustment of confounders) have controlled for confounders in our study. 4. The selection of individuals was based on a definitive diagnosis with densitometry.

The limitations of our study include: 1. Hospital-based case-control study. 2. Recall base; to reduce this base, the case group was selected from the new cases.

**Conclusion**

Predictors of osteoporosis at the end of data analysis include: Menarche age >12 yr, History of hysterectomy, RA, Calcium supplements, Menopause age <40, Second smoking and Weight. Menarche age and menopausal age are modifiable factors, but others variable are non-modifiable. By changing people’s lifestyle, these predictors can be reduced or eliminated. Given the long-term trend of osteoporosis, health policy makers pay attention to educational programs, nutrition, and supplementation from childhood and adolescence. Smoking should be reduced at home. Because the family members are exposed to smoke. It is suggested that:

- Conducting a population-based case-control study
• Conducting a cohort studies and follow-up of adolescent girls
• Nutritional interventions, education and introduction of calcium-containing foods in schools and even at younger ages
• Pregnant women training for proper nutrition during pregnancy to receive adequate calcium.

Journalism Ethics considerations

Ethical issues (Including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

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Conflict of interest

The authors declare that there is no conflict of interests.

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1380