Circulating concentrations of insulin-like growth factor I and risk of breast cancer

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Summary

Background Insulin-like growth factor (IGF)-I, a mitogenic and antiapoptotic peptide, can affect the proliferation of breast epithelial cells, and is thought to have a role in breast cancer. We hypothesised that high circulating IGF-I concentrations would be associated with an increased risk of breast cancer.

Methods We carried out a nested case-control study within the prospective Nurses’ Health Study cohort. Plasma concentrations of IGF-I and IGF binding protein 3 (IGFBP-3) were measured in blood samples collected in 1989–90. We identified 397 women who had a diagnosis of breast cancer after this date and 620 age-matched controls. IGF-I concentrations were compared by logistic regression with adjustment for other breast-cancer risk factors.

Findings There was no association between IGF-I concentrations and breast-cancer risk among the whole study group. In postmenopausal women there was no association between IGF-I concentrations and breast-cancer risk (top vs bottom quintile of IGF-I, relative risk 0·85 [95% CI 0·53–1·39]). The relative risk of breast cancer among premenopausal women by IGF-I concentration (top vs bottom tertile) was 2·33 (1·06–5·16; p for trend 0·08). Among premenopausal women less than 50 years old at the time of blood collection, the relative risk was 4·58 (1·75–12·0; p for trend 0·02). After further adjustment for time of blood collection, the relative risk was 4·58 (1·75–12·0; p for trend 0·02). Among premenopausal women less than 50 years old at the time of blood collection, the relative risk was 4·58 (1·75–12·0; p for trend 0·02).

Interpretation A positive relation between circulating IGF-I concentration and risk of breast cancer was found among premenopausal but not postmenopausal women. Plasma IGF-I concentrations may be useful in the identification of women at high risk of breast cancer and in the development of risk reduction strategies. Additional larger studies of this association among premenopausal women are needed to provide more precise estimates of effect.

Introduction

There is substantial experimental evidence that the growth hormone/insulin-like growth factor (IGF)-I axis not only affects the proliferative behaviour of breast cancer, but also stimulates proliferation of normal breast epithelial cells. Transgenic mice that overexpress growth hormone have a high frequency of breast cancer, and overexpression of IGF-I-receptor agonists is associated with cancer and incomplete mammary involution. Similarly, rhesus monkeys treated with growth hormone or IGF-I have mammary gland hyperplasia. By contrast, ductal hypoplasia is seen in animals that overexpress a growth-hormone antagonist. Studies in mice have shown a positive correlation between the strain-specific incidence of breast cancer and the activity of the growth hormone/IGF-I axis.

There is a large variation between individuals in circulating concentrations of IGF-I and IGF binding protein 3 (IGFBP-3)—the principal IGFBP in the circulation. The effect of IGF-I on the proliferation of breast epithelial cells and evidence that increased turnover in certain epithelial-cell populations is associated with a greater risk of neoplastic transformation, led us to investigate the relation of circulating IGF-I concentrations and the risk of breast cancer in women.

Methods We carried out a nested case-control study within the Nurses’ Health Study cohort. The Nurses’ Health Study started in 1976 when 121 700 female registered nurses aged 30–55 years completed and returned a mailed questionnaire. The cohort is updated every 2 years by questionnaire to update exposure status and to identify newly diagnosed disease. Data have been collected on breast-cancer risk factors such as height, weight, age at menarche, menopause, and at the birth of their first child, postmenopausal hormone use, and family history of breast cancer in mother, sister, or both. As of 1994, the follow-up rate was 90%.

In 1989–90, blood samples were collected from 32 826 women who were any age between 43 and 69 years inclusively. Details of the blood collection methods and storage have been previously published. The study was approved by the Committee on the Use of Human Subjects in Research at the Brigham and Women’s Hospital.

There is evidence that a single plasma IGF-I measurement, such as we have, reflects longer term circulating IGF-I concentrations: a correlation coefficient of 0·94 was found among 24 adults who had two blood samples drawn about 6 weeks apart.

Cases in this analysis did not have a diagnosis of invasive breast cancer before blood collection but were diagnosed before June 1, 1994. The diagnosis in 398 of 399 women was confirmed by review of the medical records. For one case, the woman confirmed the diagnosis, but the medical record was unavailable. Because of the high confirmation rate (99%) by record review, this case was kept in the analysis.

Menopausal status was defined at the time of blood collection. A woman was defined as premenopausal if she had had at least one natural menstrual cycle in the previous 12 months or was premenopausal for 2 years after blood collection and at menopause at the time of blood collection. A woman was defined as postmenopausal if she either did not have a natural menstrual cycle in the previous 12 months or was postmenopausal for 2 years after blood collection and was menopausal at the time of blood collection. Cases and controls were matched at diagnosis and were required to be 43–69 years old at diagnosis.

Details of the study population, methods of data collection and analysis, and sources of funding were reported in the study protocol and will not be repeated here. The study protocol was approved by the Committee on the Use of Human Subjects in Research at the Brigham and Women’s Hospital.

Results

There was no association between IGF-I concentrations and breast-cancer risk among the whole study group. In postmenopausal women there was no association between IGF-I concentrations and breast-cancer risk (top vs bottom quintile of IGF-I, relative risk 0·85 [95% CI 0·53–1·39]). The relative risk of breast cancer among premenopausal women by IGF-I concentration (top vs bottom tertile) was 2·33 (1·06–5·16; p for trend 0·08). Among premenopausal women less than 50 years old at the time of blood collection, the relative risk was 4·58 (1·75–12·0; p for trend 0·02). After further adjustment for time of blood collection, the relative risk was 4·58 (1·75–12·0; p for trend 0·02). Among premenopausal women less than 50 years old at the time of blood collection, the relative risk was 4·58 (1·75–12·0; p for trend 0·02).
study the ratio between plasma IGF-I and IGFBP-3. In another as a factor since there is evidence that IGFBP-3 may decrease the premenopausal controls.

premenopausal women tertiles were used, based on the values in only, quintiles of IGF-I and IGFBP-3 were based on the values 140 postmenopausal women that increased when tertiles of IGFBP-3 were added to the model (top vs. middle vs. bottom quintile). When IGFBP-3 was added to the statistical model, the relative risk of in-situ breast cancer by IGF-I concentration and breast-cancer risk among the whole study population was not significantly different from 1 (table 3). When IGFBP-3 was added to the statistical model, the relative risks were similar. The relative risk of in-situ breast cancer by IGF-I concentration also was not significant (2·26 top quintile vs. bottom quintile, 95% CI 0·70–7·28).

In premenopausal women there was no relation between IGF-I and breast-cancer risk even after exclusion of current or recent users of hormone-replacement therapy. There was a positive association between IGF-I concentration and breast-cancer risk in premenopausal women that increased when tertiles of IGFBP-3 were added to the model (top vs. middle vs. bottom quintile of IGF-I, relative risk 2·88 [1·21–6·85]). The relative risk was 2·65 (1·04–6·75) among only the breast-cancer cases that occurred premenopausally (ie, excluding cases who were premenopausal at blood collection but became postmenopausal before the diagnosis of breast cancer). A non-significant inverse association between IGFBP-3 and concentrations was highly associated with breast-cancer risk and so this ratio was also assessed.

A linear regression model of IGF-I by case or control status and adjusted for the use of hormone-replacement therapy was used to compare mean IGF-I concentrations between cases and controls. Because of the skewed distribution of the IGF-I values, the square-root was used and we accounted for the correlation between multiple control values per case. Unconditional logistic regression analysis adjusted for the matching factors was used to estimate the relative risks (odds ratios) and 95% CI. Adjustment for other breast-cancer risk factors (age at menarche [less than 12 years, 12 years, 13 years, more than 14 years], age at menopause [premenopausal, less than 44 years, 45–49 years, more than 50 years, unknown], parity [0, 1–2 births, 3–4 births, more than 5 births], age at birth of first child [0, less than 24 years, 25–29 years, 30 years or older], family history of breast cancer, and body-mass index [less than 23 kg/m², 23–28·9 kg/m², 29·0 kg/m² or greater]) did not substantially alter the relative risk, thus estimates adjusted only for the matching factors are presented here. The results of the unconditional analyses were essentially identical to those from conditional analyses but had the advantage of increased statistical power. Tests for trend were done by calculating the Wald statistic for a continuous distribution of IGF-I values.

Results
Baseline characteristics of the cases and controls, by menopausal status, are shown in table 1. The mean time between blood collection and diagnosis was 28 months (SD 16) with a range of 1–57 months. There was a significant relation between IGF-I and age (p=0·12, p=0·01). IGF-I concentrations did not differ between cases and controls in the whole cohort or among postmenopausal women (table 2). Among premenopausal women younger than 50 years, the mean IGF-I concentration was significantly higher in the cases than in the controls (p=0·0004). Concentrations of IGF-I and IGFBP-3 were almost identical and the intraclass correlation between samples analysed by means of current or recent users of hormone-replacement therapy. There was a positive association between IGF-I concentration and breast-cancer risk in premenopausal women that increased when tertiles of IGFBP-3 were added to the model (top vs. middle vs. bottom quintile of IGF-I, relative risk 2·88 [1·21–6·85]). The relative risk was 2·65 (1·04–6·75) among only the breast-cancer cases that occurred premenopausally (ie, excluding cases who were premenopausal at blood collection but became postmenopausal before the diagnosis of breast cancer). A non-significant inverse association between IGFBP-3 and

Table 1: Baseline characteristics

<table>
<thead>
<tr>
<th>Age (years)</th>
<th>Cases (n=397)</th>
<th>Controls (n=620)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Premenopausal women</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age at menopause</td>
<td>48·1 (2·8)</td>
<td>48·1 (2·8)</td>
</tr>
<tr>
<td>Parity</td>
<td>2·7 (1·0)</td>
<td>2·8 (1·0)</td>
</tr>
<tr>
<td>Mean (SD)</td>
<td>50 60 206 (77·6–425)</td>
<td>78 175 (84·9–320)</td>
</tr>
<tr>
<td>Postmenopausal women</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age at menopause</td>
<td>12·6 (3·4)</td>
<td>12·5 (1·4)</td>
</tr>
<tr>
<td>Parity</td>
<td>7 (0·5)</td>
<td>8·6</td>
</tr>
<tr>
<td>Mean (SD)</td>
<td>50 60 206 (77·6–425)</td>
<td>78 175 (84·9–320)</td>
</tr>
</tbody>
</table>

Table 2: Plasma IGF-I concentrations by case or control status

<table>
<thead>
<tr>
<th>Cases</th>
<th>Controls</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>n</td>
<td>Median (range)</td>
<td>n</td>
</tr>
<tr>
<td>All cases or controls</td>
<td>397</td>
<td>154 (20·7–425)</td>
</tr>
<tr>
<td>Postmenopausal</td>
<td>305</td>
<td>142 (20·7–390)</td>
</tr>
<tr>
<td>Premenopausal</td>
<td>105</td>
<td>184 (81·0–320)</td>
</tr>
<tr>
<td>Premenopausal, age &lt;50</td>
<td>60</td>
<td>206 (77·6–425)</td>
</tr>
</tbody>
</table>

IGF-I and IGFBP-3 were assayed by ELISA with reagents from Diagnostic Systems Laboratory (Webster, TX, USA). This method is more reproducible and appropriate for large numbers of samples than the RIA previously employed. There was a high correlation between the IGF-I values from the ELISA and samples analysed by RIA after acid chromatography (Pearson r=0·97). All samples were assayed masked to case or control status. Masked split specimens included within each batch were used to calculate the coefficient of variation within and between batches: for IGF-I these were 8·7% and 15·6%, respectively; for IGFBP-3, 8·7% and 14·6%, respectively. To find out the effect of our blood-collection methods on IGF-I concentrations, we compared IGF-I and IGFBP-3 concentrations in samples that were separated and frozen immediately—the usual collection and processing methods—with samples stored as heparinised whole blood for 24 h and 36 h before separation (similar to our collection conditions). The mean IGF-I and IGFBP-3 values were almost identical and the intraclass correlation between results of the two collection methods was 0·96 for IGF-I and 0·96 for IGFBP-3, which showed that our collection methods did not affect sample integrity.

Statistical analyses
We analysed the relation between plasma IGF-I concentration and breast-cancer risk in the whole group and by menopausal status. 16 cases and 32 controls in whom menopausal status was uncertain were excluded from the stratum-specific analyses. For analyses among all women and postmenopausal women only, quintiles of IGF-I and IGFBP-3 were based on the values from all controls. Because of the smaller number of premenopausal women terciles were used, based on the values in the premenopausal controls.

IGF-I was assessed in the models with and without IGFBP-3 as a factor since there is evidence that IGFBP-3 may decrease IGF-I bioactivity by direct or indirect mechanisms. In another study the ratio between plasma IGF-I and IGFBP-3 was almost identical and the intraclass correlation between samples analysed by means of current or recent users of hormone-replacement therapy. There was a positive association between IGF-I concentration and breast-cancer risk in premenopausal women that increased when tertiles of IGFBP-3 were added to the model (top vs. middle vs. bottom quintile of IGF-I, relative risk 2·88 [1·21–6·85]). The relative risk was 2·65 (1·04–6·75) among only the breast-cancer cases that occurred premenopausally (ie, excluding cases who were premenopausal at blood collection but became postmenopausal before the diagnosis of breast cancer). A non-significant inverse association between IGFBP-3 and concentrations was highly associated with breast-cancer risk and so this ratio was also assessed.

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Results
Baseline characteristics of the cases and controls, by menopausal status, are shown in table 1. The mean time between blood collection and diagnosis was 28 months (SD 16) with a range of 1–57 months. There was a significant relation between IGF-I and age (p=0·12, p=0·01). IGF-I concentrations did not differ between cases and controls in the whole cohort or among postmenopausal women (table 2). Among premenopausal women younger than 50 years, the mean IGF-I concentration was significantly higher in the cases than in the controls (p=0·0004). Concentrations of IGF-I and IGFBP-3 were almost identical and the intraclass correlation between samples analysed by means of current or recent users of hormone-replacement therapy. There was a positive association between IGF-I concentration and breast-cancer risk in premenopausal women that increased when tertiles of IGFBP-3 were added to the model (top vs. middle vs. bottom quintile of IGF-I, relative risk 2·88 [1·21–6·85]). The relative risk was 2·65 (1·04–6·75) among only the breast-cancer cases that occurred premenopausally (ie, excluding cases who were premenopausal at blood collection but became postmenopausal before the diagnosis of breast cancer). A non-significant inverse association between IGFBP-3 and
breast-cancer risk also was found (data not shown).

To define our premenopausal group more stringently, we assessed the same correlations in women who were both premenopausal and younger than 50 years at the time of blood collection. The association between plasma IGF-I concentration and breast-cancer risk increased with a relative risk of 2·28 between the top and bottom tertiles (2·40–22·0) when adjusted for tertiles of IGFBP-3; the addition of other breast-cancer risk factors one at a time to these models did not change the results. When the IGF-I/IGFBP-3 ratio was used, the relative risks (top vs bottom tertiles) in premenopausal and premenopausal women younger than 50 years were 2·13 (0·97–4·68) and 2·46 (0·97–6·24), respectively.

The relative risks comparing the top and the bottom tertile of plasma IGF-I adjusted for IGFBP-3 were similar in the analyses that excluded the 89 women diagnosed in the first year after blood collection. Among premenopausal women (13 cases excluded) and premenopausal women younger than 50 years (ten cases excluded), the relative risks after adjustment for IGFBP-3 were 2·35 (0·94–5·85) and 5·18 (1·66–16·1), respectively.

Discussion

To our knowledge this is the first prospective study to investigate the relation between circulating IGF-I concentrations and risk of breast cancer. We observed a strong positive relation among premenopausal women, particularly after adjusting for concentrations of IGFBP-3, but no association among postmenopausal women.

Two case-control studies were also found a positive relation between plasma IGF-I concentrations. In the larger study, the relation was strongest among premenopausal women with the ratio of IGF-I to IGFBP-3 most closely related to breast-cancer risk. However, both of these studies were potentially limited by their retrospective design; the blood sample for plasma IGF-I concentration was taken after diagnosis and an effect of the disease on IGF-I concentrations was possible, and the controls may not have been comparable with the cases. Our prospective study reduces these concerns to a minimum and suggests that IGF-I concentrations may be a marker of breast-cancer risk among premenopausal women. Although the relation between plasma IGF-I and IGFBP concentrations and IGF-I bioactivity in the tissues is unclear, these epidemiological data suggest they are correlated.

The up to seven-fold increase in breast-cancer risk among premenopausal women 50 years or younger suggests that the relation between IGF-I and risk of breast cancer may be greater than that of other established breast-cancer risk factors—with the exception of a strong family history of breast cancer or a high-density mammographic profile. However, because we had only a small number of premenopausal women, studies that include large numbers of young women are needed to confirm this finding and provide more precise risk estimates. More research is also needed to find out whether known risk factors for breast cancer are acting as surrogates of IGF-I, or, alternatively, whether IGF-I is a marker of other possible risk factors in premenopausal women.

The difference in relative risk between premenopausal and postmenopausal women may reflect an important effect of the growth hormone/IGF-I axis that occurs early in life after breast development. In our study, circulating IGF-I concentrations did not change acutely with menopause but did decrease with age. If this age-related decrease in IGF-I concentrations varies in a random way, with increasing age IGF-I concentrations may become less and less reflective of those in adolescence and early adulthood. Alternatively, IGF-I concentrations may be particularly relevant to the risk of breast cancer in premenopausal women since oestrogen enhances the action of IGF-I in the breast. Although we did not find a relation between postmenopausal IGF-I concentrations and risk of breast cancer, we cannot rule out an effect of premenopausal IGF-I concentrations on the risk of breast cancer after menopause.

There is substantial indirect evidence of a relation between IGF-I and risk of breast cancer. Energy-restricted diets, which decrease IGF-I, decrease the incidence of breast cancer in rats. Also, there is a positive relation between birthweight and breast-cancer risk, and low birthweight is associated with a lower IGF-I concentration. Similarly, height, which is positively associated with breast-cancer risk in most epidemiological studies, is positively related to IGF-I concentrations. Furthermore, there is evidence that IGF-I concentrations may be related to risk of prostate and colon cancers.
In women who take tamoxifen, circulating IGF-I concentrations decrease and IGF-I bioactivity is reduced because of a decrease in pituitary growth-hormone secretion, IGF-I gene expression and IGF-I-receptor concentrations, and because of upregulation of IGFBP expression. The association between IGF-I concentration and risk of breast cancer provides a novel rationale for the use of tamoxifen as a chemopreventive agent. It will be important to establish whether baseline IGF-I concentrations can predict risk or the potential for risk reduction by antiestrogens and whether IGF-I can be used as an intermediate endpoint in chemoprevention trials. There are several additional agents that lower IGF-I concentrations, such as antagonists of growth-hormone releasing-hormone or growth-hormone, and somatostatin analogues. Some of these are already in clinical breast-cancer trials and may deserve consideration as risk-reducing agents for women who have a high risk related to raised IGF-I concentrations. Interestingly, other proposed agents for breast-cancer prevention, such as retinoids, transforming growth factor-B, and vitamin D analogues, also upregulate IGFBP expression—an action that may be correlated with lower IGF-I bioactivity in certain physiological contexts. Finally, the relation between nutritional or other lifestyle factors and concentrations of IGF-I or IGFBP deserves further study.

References

8 Chen XY, Chen WY, Striker LJ, Striker GE, Kopchick JJ. Co-expression of bovine growth hormone (GH) and human GH antigen genes in transgenic mice. Endocrinology 1997; 138: 851–54.