Dietary folate, vitamin B₆, vitamin B₁₂ and methionine intake and the risk of breast cancer by oestrogen and progesterone receptor status

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Abstract
Few studies have evaluated the relationship between the consumption of dietary folate and one-carbon metabolism-related nutrients and breast cancer risk defined by oestrogen receptor (ER) and progesterone receptor (PR) status. The objective of the present study was to examine the associations between dietary folate, vitamin B₆, vitamin B₁₂, and methionine intake and the risk of breast cancer by ER and PR status among Chinese women in Guangdong. A hospital-based case–control study was conducted from June 2007 to August 2008, with 438 cases and 438 age (5-year interval)- and residence (rural/urban)-matched controls. Dietary intake information was assessed using a validated FFQ administered through a face-to-face interview. Unconditional logistic regression models were used to calculate multi-variate-adjusted OR and 95% CI. A significant inverse association was found between dietary folate and vitamin B₆ intake and breast cancer risk. The adjusted OR of the highest vs. the lowest quartile were 0·32 (95% CI 0·21, 0·49; P_trend, 0·001) for dietary folate and 0·46 (95% CI 0·30, 0·69; P_trend < 0·001) for vitamin B₆. No associations were observed for vitamin B₁₂ and methionine intake. A significant inverse association between dietary folate intake and breast cancer risk was observed in all subtypes of ER and PR status. These findings suggest that dietary folate and vitamin B₆ intakes were inversely associated with breast cancer risk. The inverse association did not differ by ER and/or PR status.

Key words: Folate; Vitamin B₆; Vitamin B₁₂; Breast cancer; Hormone receptor status

One-carbon metabolism is a complex metabolic network. Dietary factors involved in this pathway include folate, methionine, vitamin B₆ and vitamin B₁₂. Folate is thought to play an important role in cancer prevention (1). As a carbon donor in one-carbon metabolism, folate is essential for normal DNA synthesis and repair. Adequate folate status is also important for the production of S-adenosylmethionine for DNA methylation. Moreover, methionine is the precursor for the endogenous production of S-adenosylmethionine. Additionally, one-carbon metabolism may involve as many as twenty-five enzymes, some of which require the presence of vitamin B₆ and vitamin B₁₂ as co-enzymes (2). For example, vitamin B₁₂ acts as a cofactor for the enzyme methionine synthase, and vitamin B₆ serves as a cofactor for folate-dependent enzymes in the one-carbon metabolism pathway.

Folate digestion and absorption are affected by many factors such as diseases or medications. Alcohol is a known folate antagonist. It can impair folate absorption and metabolism (3), and plausibly increase an individual’s requirement for folate intake. Some studies, but not all, have found that alcohol consumption modifies the effect of folate intake on breast cancer risk (4).

Several epidemiological studies have examined the association of dietary folate with breast cancer, but the results are inconsistent. While some studies have reported a negative association between dietary folate intake and breast cancer risk (5–13), other studies have not observed a significant relationship (14–24). In addition to folate, a few studies have investigated the relationships between other one-carbon metabolism-related nutrients (such as vitamin B₆, vitamin B₁₂ and methionine) and breast cancer risk (8,9,11,16,23,25–31). But the results remain inconclusive. Therefore, further investigation of these issues is warranted.

Abbreviations: ER, oestrogen receptor; PR, progesterone receptor.

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Oestrogen receptor (ER) and progesterone receptor (PR) are the most widely studied markers in breast tissue. Recent studies have raised the hypothesis that the association of dietary factors with breast cancer may differ by ER or PR status\(^{(32–34)}\). Folate may influence the methylation of ER or PR genes and thereby affecting the silencing of these genes\(^{(35,36)}\). The associations between folate intake and breast cancer risk may therefore differ according to ER or PR status of tumours. So far, only a few studies have examined folate intake in relation to breast cancer risk according to ER and/or PR status\(^{(32,33,26,34,37,38)}\) and the results are inconsistent.

Moreover, the majority of these studies have been performed in Western populations. Unlike their Western counterparts, most Chinese women consume natural (unfortified and unprocessed) foods, and they seldom take vitamin supplements or drink regularly. Thus, Chinese dietary habits may allow a better assessment of nutrient intake and minimise potential misclassifications in epidemiological studies. The present study aimed to evaluate the association between dietary folate and other one-carbon metabolism-related nutrient intake and the risk of breast cancer characterised by ER and PR and estimate the OR and 95 % CI of each quartile, using the second FFQ and 18 d dietary records were 0·35 for folate, 0·26 for vitamin B\(_6\), 0·50 for vitamin B\(_{12}\) and 0·36 for methionine. These results showed that the reproducibility and validity of our FFQ seem to be comparable with the values reported by others\(^{(44,45)}\).

### Materials and methods

#### Study subjects

A hospital-based case–control study was conducted in Guangdong Province, China, from June 2007 to August 2008. The selection of cases and controls has been described in detail elsewhere\(^{(39,40)}\). In brief, potential case subjects were recruited from patients admitted to the surgical units of two affiliated hospitals of Sun Yat-sen University, Guangzhou, China. Inclusion criteria were female subjects aged 25–70 years and natives of Guangdong Province or having lived in Guangdong for at least 5 years, with incident, primary, histologically confirmed breast cancer diagnosed no more than 3 months before the interview. Women were excluded if they could not understand or speak Mandarin/Cantonese or with a prior history of breast cancer or other cancers. In total, 438 (96 %) cases out of 455 eligible cases were successfully interviewed.

As well as the 438 eligible patients, 438 control subjects with no history of cancer and admitted to the same hospitals during the same time period as the case subjects were interviewed. They were frequency-matched by age (5-year interval) and residence (rural/urban) to the case patients. These patients presented with a wide spectrum of non-neoplastic conditions including eye disorders (glaucoma, uveitis, keratitis, pterygium, dacryocystitis and optic neuritis), ear, nose and throat diseases (sudden deafness, acute bacterial/viral otitis media, sinusitis, deviation of nasal septum, tonsillitis), trisacial neuralgia, varicose veins, traumatic skeletal disorders, osteoarthritis, degenerate joint disease, orthopaedics and acute appendicitis. The present study was conducted according to the guidelines laid down in the Declaration of Helsinki, and all procedures involving human subjects/patients were approved by the Ethical Committee of the Chinese University of Hong Kong. Written informed consent was obtained from all participants.

#### Data collection

Data on sociodemographic characteristics, anthropometrics, menstrual and reproductive history, family history of breast cancer, physical activity, smoking habits, alcohol use and prior disease history were collected from each subject by interviewer-administered FFQ covering the habitual diet of participants during the past year. Food photographs were used to help participants quantify the portions consumed. Daily dietary nutrient intakes were estimated using the China Food Composition Table\(^{(41,42)}\). Information on frequency of intake and portion size was used to calculate the amount of each food item consumed on average (g/d). Total dietary intakes of folate, vitamin B\(_6\), vitamin B\(_{12}\) and methionine were calculated by summing the product of the frequency of consumption, usual portion consumed and micronutrient content of each food item. Information on cooking method was not collected in the present study, as stir-frying and boiling are the two most common methods of cooking green leafy vegetables and animal foods in China.

The validity and reproducibility of the FFQ has been described in detail elsewhere\(^{(43)}\). A total of sixty-one female subjects completed 3 d dietary records at intervals of 2 months during a 12-month period and two FFQ administered 1 year apart. The correlation coefficients comparing the second FFQ and 18 d dietary records were 0·35 for folate, 0·26 for vitamin B\(_6\), 0·50 for vitamin B\(_{12}\) and 0·36 for methionine. The correlation coefficients between the two FFQ were 0·60 for folate, 0·57 for vitamin B\(_6\), 0·60 for vitamin B\(_{12}\) and 0·49 for methionine. These results showed that the reproducibility and validity of our FFQ seem to be comparable with the values reported by others\(^{(44,45)}\).

#### Statistical analysis

All statistical analyses were performed using SPSS 13.0 (SPSS Inc., Chicago, IL, USA). Dietary folate, vitamin B\(_6\), vitamin B\(_{12}\) and methionine intakes were adjusted for total energy intake using the residual method\(^{(46)}\) and then categorised into quartiles based on the distribution among the control subjects. Unconditional logistic regression models were used to estimate the OR and 95 % CI of each quartile, using the lowest quartile group as the reference after adjusting for the various potential confounding factors. Risk factors identified by comparison of baseline characteristics between cases and controls as independently associated with breast cancer risk were adjusted in the multivariate models. These included...
Table 1. Comparison of breast cancer cases and controls on sociodemographic and selected characteristics among Chinese women
(Mean values, standard deviations, number of patients and percentages)

<table>
<thead>
<tr>
<th></th>
<th>Cases (n 438)</th>
<th>Controls (n 438)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n</td>
<td>%</td>
<td>n</td>
</tr>
<tr>
<td>Age (years) Mean</td>
<td>47-04</td>
<td>47-14</td>
<td>0.875</td>
</tr>
<tr>
<td>SD</td>
<td>9-53</td>
<td>9-58</td>
<td></td>
</tr>
<tr>
<td>Marital status</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Married</td>
<td>409</td>
<td>93.4</td>
<td>413</td>
</tr>
<tr>
<td>Unmarried/divorced/widowed</td>
<td>29</td>
<td>6-6</td>
<td>25</td>
</tr>
<tr>
<td>Educational level</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Primary school or below</td>
<td>106</td>
<td>24-2</td>
<td>127</td>
</tr>
<tr>
<td>Junior high school</td>
<td>127</td>
<td>29-0</td>
<td>116</td>
</tr>
<tr>
<td>Senior high school/secondary technical school</td>
<td>109</td>
<td>24-9</td>
<td>109</td>
</tr>
<tr>
<td>College or above</td>
<td>96</td>
<td>21-9</td>
<td>86</td>
</tr>
<tr>
<td>Household income (yuan/month)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt;2000</td>
<td>157</td>
<td>35-8</td>
<td>161</td>
</tr>
<tr>
<td>2001–5000</td>
<td>152</td>
<td>34-7</td>
<td>149</td>
</tr>
<tr>
<td>5001–8000</td>
<td>68</td>
<td>15-5</td>
<td>59</td>
</tr>
<tr>
<td>&gt;8000</td>
<td>61</td>
<td>13-9</td>
<td>69</td>
</tr>
<tr>
<td>BMI (kg/m²) Mean</td>
<td>22-92</td>
<td></td>
<td>22-46</td>
</tr>
<tr>
<td>SD</td>
<td>3-33</td>
<td></td>
<td>3-05</td>
</tr>
<tr>
<td>Regular smoker</td>
<td>7</td>
<td>1-6</td>
<td>2</td>
</tr>
<tr>
<td>Passive smoker from a husband</td>
<td>202</td>
<td>46-1</td>
<td>170</td>
</tr>
<tr>
<td>Regular drinker</td>
<td>12</td>
<td>2-7</td>
<td>10</td>
</tr>
<tr>
<td>Physical activity (exercise for health)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never</td>
<td>170</td>
<td>38-8</td>
<td>139</td>
</tr>
<tr>
<td>Occasional</td>
<td>46</td>
<td>10-5</td>
<td>35</td>
</tr>
<tr>
<td>≥1 time/week</td>
<td>222</td>
<td>50-7</td>
<td>264</td>
</tr>
<tr>
<td>Age at menarche (years) Mean</td>
<td>14-82</td>
<td></td>
<td>15-11</td>
</tr>
<tr>
<td>SD</td>
<td>1-88</td>
<td></td>
<td>1-84</td>
</tr>
<tr>
<td>Nulliparous</td>
<td>17</td>
<td>3-9</td>
<td>19</td>
</tr>
<tr>
<td>Number of live births* Mean</td>
<td>1-98</td>
<td></td>
<td>2-03</td>
</tr>
<tr>
<td>SD</td>
<td>1-12</td>
<td></td>
<td>1-20</td>
</tr>
<tr>
<td>Age at first live birth (years)*</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>≤19</td>
<td>11</td>
<td>2-6</td>
<td>11</td>
</tr>
<tr>
<td>20–24</td>
<td>139</td>
<td>33-0</td>
<td>170</td>
</tr>
<tr>
<td>25–29</td>
<td>227</td>
<td>53-9</td>
<td>201</td>
</tr>
<tr>
<td>≥30</td>
<td>44</td>
<td>10-5</td>
<td>37</td>
</tr>
<tr>
<td>Months of breast-feeding*</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never</td>
<td>49</td>
<td>11-6</td>
<td>26</td>
</tr>
<tr>
<td>1–3</td>
<td>21</td>
<td>5-0</td>
<td>22</td>
</tr>
<tr>
<td>4–11</td>
<td>95</td>
<td>22-6</td>
<td>108</td>
</tr>
<tr>
<td>12–23</td>
<td>118</td>
<td>28-0</td>
<td>102</td>
</tr>
<tr>
<td>≥24</td>
<td>138</td>
<td>32-8</td>
<td>161</td>
</tr>
<tr>
<td>Age at menopause (years)† Mean</td>
<td>49-33</td>
<td></td>
<td>49-06</td>
</tr>
<tr>
<td>SD</td>
<td>3-96</td>
<td></td>
<td>3-93</td>
</tr>
<tr>
<td>Menopausal status</td>
<td></td>
<td></td>
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<tr>
<td>Premenopausal</td>
<td>306</td>
<td>69-9</td>
<td>295</td>
</tr>
<tr>
<td>Postmenopausal</td>
<td>132</td>
<td>30-1</td>
<td>143</td>
</tr>
<tr>
<td>Mother/sister/daughter with breast cancer</td>
<td>17</td>
<td>3-9</td>
<td>4</td>
</tr>
<tr>
<td>Ever had benign breast disease</td>
<td>177</td>
<td>40-4</td>
<td>84</td>
</tr>
<tr>
<td>Ever used oral contraceptives</td>
<td>27</td>
<td>6-2</td>
<td>21</td>
</tr>
<tr>
<td>Ever used nutritional supplements</td>
<td>70</td>
<td>16-0</td>
<td>80</td>
</tr>
</tbody>
</table>

* Among parous women.
† Among menopausal women.
Results

Table 1 presents the sociodemographic and established breast cancer risk factors of the study populations. Compared with controls, cases had an earlier age at menarche, older age at first live birth, fewer months of breast-feeding and higher BMI. Cases were more likely to have a history of breast cancer in a first-degree relative, history of benign breast disease and history of passive smoking from a husband, and were less likely to be physically active than controls. All of the aforementioned variables were considered to be potential confounding factors and controlled for in subsequent analyses. No significant differences were found between the case and control subjects in sociodemographic factors, including educational level, occupational status, marital status and household income, or in reproductive factors, including nulliparity, number of live births, age at menopause and use of an oral contraceptive.

The median intake of folate was 228·3 μg/d, vitamin B₁₂ was 1·54 μg/d and methionine was 1·14 μg/d in the control group (Table 2). Compared with controls, the consumption of dietary folate and vitamin B₁₂ was significantly lower in the case subjects. No significant differences between cases and controls were observed for vitamin B₁₂ and methionine intake.

The associations between the intake of dietary folate, vitamin B₁₂, vitamin B₁₂ and methionine and the risk of breast cancer are shown in Table 3. After adjustment for the various potential confounders, a significant inverse association was observed between the intake of dietary folate and vitamin B₁₂ and the risk of breast cancer. The OR for the highest quartile of intake compared with the lowest were 0·32 (95 % CI 0·21, 0·49; P<0·001) for folate intake and 0·46 (95 % CI 0·30, 0·69; P<0·001) for vitamin B₁₂ intake. Associations for vitamin B₁₂ and methionine were not statistically significant.

The associations between folate intake and breast cancer risk according to sources of folate are shown in Table 4. There was a consistent inverse association between folate intake from different food sources (e.g. vegetables, fruits, soya, grains and animal foods) and breast cancer risk.

Among the case subjects with information on hormone receptor status, 292 (73·2 %) and 348 (87·2 %) were for ER⁺ and PR⁺, respectively; 275 (68·9 %) were ER⁺/PR⁺; seventeen (4·3 %) were ER⁺/PR⁻; seventy-three (18·3 %) were ER⁻/PR⁺; and thirty-four (8·5 %) were ER⁻/PR⁻. Table 5 shows the impact of dietary folate consumption on the risk of breast cancer characterised by ER and PR status. The inverse association between folate intake and breast cancer risk was observed in all subtypes of ER and/or PR status, although the association was statistically non-significant among women with PR⁻, ER⁺/PR⁻ and ER⁻/PR⁺ breast cancer tumours due to the relatively small numbers.

Because pre- and postmenopausal breast cancers probably have a separate disease aetiology, a stratified analysis by menopausal status was conducted. The inverse association of folate intake with breast cancer risk did not vary by menopausal status. The multivariate-adjusted OR were 0·34 (95 % CI 0·21, 0·58) among premenopausal women and 0·28 (95 % CI 0·12, 0·65) among postmenopausal women, comparing the
fourth quartile with the first quartile. No interaction was observed between menopausal status and folate intake ($P_{interaction} = 0.570$, data not shown).

A number of sensitivity analyses were performed to examine the association between dietary folate intake and breast cancer risk. Ductal carcinoma was the most frequent histological type (415, 94.7%), and restricting the analysis to these cases yielded very similar results. It was observed that seventy cases (16.1%) and eighty controls (18.3%) reported ever taking nutritional supplements. Sensitivity analysis that excluded women with nutritional supplement use revealed similar results as compared with the analyses that included nutritional supplement users. In the present study, only twelve (2.7%) cases and ten (2.3%) controls were regular drinkers. The results of sensitivity analyses excluding women with alcohol intake were essentially the same. Further analyses restricted to subjects reporting no dietary change showed no substantial change in the observed folate intake and breast cancer association.

### Table 3. Folate, vitamin B$_6$, vitamin B$_{12}$ and methionine intake and breast cancer risk among Chinese women
(Odds ratios and 95% confidence intervals)

<table>
<thead>
<tr>
<th></th>
<th>Q1*</th>
<th>Q2*</th>
<th>Q3*</th>
<th>Q4*</th>
<th>$P_{trend}$</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Folate</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of cases/controls</td>
<td>174/109</td>
<td>114/110</td>
<td>94/110</td>
<td>56/109</td>
<td></td>
</tr>
<tr>
<td>Crude</td>
<td>1</td>
<td>0.65</td>
<td>0.46, 0.93</td>
<td>0.54</td>
<td>0.37, 0.77</td>
</tr>
<tr>
<td>Adjusted†</td>
<td>1</td>
<td>0.68</td>
<td>0.47, 1.00</td>
<td>0.51</td>
<td>0.34, 0.75</td>
</tr>
<tr>
<td>Vitamin B$_6$</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of cases/controls</td>
<td>159/105</td>
<td>132/112</td>
<td>72/113</td>
<td>75/108</td>
<td></td>
</tr>
<tr>
<td>Crude</td>
<td>1</td>
<td>0.82</td>
<td>0.58, 1.17</td>
<td>0.45</td>
<td>0.31, 0.66</td>
</tr>
<tr>
<td>Adjusted†</td>
<td>1</td>
<td>0.83</td>
<td>0.57, 1.21</td>
<td>0.42</td>
<td>0.28, 0.64</td>
</tr>
<tr>
<td>Vitamin B$_{12}$</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of cases/controls</td>
<td>122/108</td>
<td>106/113</td>
<td>106/106</td>
<td>104/111</td>
<td></td>
</tr>
<tr>
<td>Crude</td>
<td>1</td>
<td>0.86</td>
<td>0.59, 1.25</td>
<td>0.86</td>
<td>0.59, 1.25</td>
</tr>
<tr>
<td>Adjusted†</td>
<td>1</td>
<td>0.74</td>
<td>0.49, 1.09</td>
<td>0.74</td>
<td>0.50, 1.11</td>
</tr>
<tr>
<td>Methionine</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of cases/controls</td>
<td>127/109</td>
<td>103/110</td>
<td>105/110</td>
<td>103/109</td>
<td></td>
</tr>
<tr>
<td>Crude</td>
<td>1</td>
<td>0.80</td>
<td>0.56, 1.16</td>
<td>0.82</td>
<td>0.57, 1.19</td>
</tr>
<tr>
<td>Adjusted†</td>
<td>1</td>
<td>0.80</td>
<td>0.56, 1.24</td>
<td>0.79</td>
<td>0.53, 1.18</td>
</tr>
</tbody>
</table>

* Quartile cut-off points were based on residual energy-adjusted intake among the control subjects.
† OR were adjusted for age at menarche, live births and age at first live birth, months of breast-feeding, BMI, history of benign breast disease, mother/sister/daughter with breast cancer, physical activity, passive smoking and total energy intake.

### Table 4. Folate intake from different food sources and breast cancer risk among Chinese women
(Odds ratios and 95% confidence intervals)

<table>
<thead>
<tr>
<th></th>
<th>Q1*</th>
<th>Q2*</th>
<th>Q3*</th>
<th>Q4*</th>
<th>$P_{trend}$</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Folate</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of cases/controls</td>
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<td>141/110</td>
<td>98/110</td>
<td>49/109</td>
<td></td>
</tr>
<tr>
<td>Crude</td>
<td>1</td>
<td>0.93</td>
<td>0.66, 1.32</td>
<td>0.65</td>
<td>0.45, 0.94</td>
</tr>
<tr>
<td>Adjusted†</td>
<td>1</td>
<td>0.98</td>
<td>0.67, 1.42</td>
<td>0.63</td>
<td>0.42, 0.93</td>
</tr>
<tr>
<td><strong>Folate from fruits</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. of cases/controls</td>
<td>130/109</td>
<td>134/110</td>
<td>89/110</td>
<td>83/109</td>
<td></td>
</tr>
<tr>
<td>Crude</td>
<td>1</td>
<td>1.02</td>
<td>0.71, 1.46</td>
<td>0.68</td>
<td>0.46, 0.99</td>
</tr>
<tr>
<td>Adjusted†</td>
<td>1</td>
<td>1.12</td>
<td>0.76, 1.64</td>
<td>0.66</td>
<td>0.44, 0.99</td>
</tr>
<tr>
<td><strong>Folate from soya</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Crude</td>
<td>1</td>
<td>1.16</td>
<td>0.82, 1.66</td>
<td>0.86</td>
<td>0.59, 1.24</td>
</tr>
<tr>
<td>Adjusted†</td>
<td>1</td>
<td>0.64</td>
<td>0.43, 0.95</td>
<td>0.88</td>
<td>0.60, 1.28</td>
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<tr>
<td><strong>Folate from grains</strong></td>
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<td>100/110</td>
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</tr>
<tr>
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<td>1.02</td>
<td>0.71, 1.47</td>
<td>0.78</td>
<td>0.54, 1.13</td>
</tr>
<tr>
<td>Adjusted†</td>
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<td>0.91</td>
<td>0.61, 1.34</td>
<td>0.71</td>
<td>0.48, 1.07</td>
</tr>
<tr>
<td><strong>Folate from other foods</strong></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>No. of cases/controls</td>
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<td>114/110</td>
<td>102/110</td>
<td>91/109</td>
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<tr>
<td>Crude</td>
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<td>0.86</td>
<td>0.60, 1.24</td>
<td>0.78</td>
<td>0.54, 1.13</td>
</tr>
<tr>
<td>Adjusted†</td>
<td>1</td>
<td>0.73</td>
<td>0.49, 1.08</td>
<td>0.70</td>
<td>0.47, 1.04</td>
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* Quartile cut-off points were based on residual energy-adjusted intake among the control subjects.
† OR were adjusted for age at menarche, live births and age at first live birth, months of breast-feeding, BMI, history of benign breast disease, mother/sister/daughter with breast cancer, physical activity, passive smoking and total energy intake.
Folate intake and breast cancer risk

Table 5. Dietary folate intake and breast cancer risk stratified by oestrogen receptor (ER)/progesterone receptor (PR) status (Odds ratios and 95 % confidence intervals)

<table>
<thead>
<tr>
<th></th>
<th>Q1*</th>
<th>Q2*</th>
<th>Q3*</th>
<th>Q4*</th>
<th>P_{trend}</th>
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<tr>
<td>ER+</td>
<td>OR</td>
<td>OR</td>
<td>95 % CI</td>
<td>OR</td>
<td>95 % CI</td>
</tr>
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<td>85/110</td>
<td>58/110</td>
<td>34/109</td>
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<tr>
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<td>0·76</td>
<td>0·50, 1·16</td>
<td>0·48</td>
<td>0·31, 0·75</td>
</tr>
<tr>
<td>ER</td>
<td>OR</td>
<td>OR</td>
<td>95 % CI</td>
<td>OR</td>
<td>95 % CI</td>
</tr>
<tr>
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<tr>
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<td>0·50</td>
<td>0·27, 0·94</td>
<td>0·60</td>
<td>0·34, 1·07</td>
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<tr>
<td>PR+</td>
<td>OR</td>
<td>OR</td>
<td>95 % CI</td>
<td>OR</td>
<td>95 % CI</td>
</tr>
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<td>91/110</td>
<td>72/110</td>
<td>42/109</td>
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</tr>
<tr>
<td>Adjusted†</td>
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<td>0·67</td>
<td>0·45, 0·99</td>
<td>0·46</td>
<td>0·31, 0·70</td>
</tr>
<tr>
<td>PR</td>
<td>OR</td>
<td>OR</td>
<td>95 % CI</td>
<td>OR</td>
<td>95 % CI</td>
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<td>0·43, 2·24</td>
<td>0·82</td>
<td>0·35, 1·92</td>
</tr>
<tr>
<td>ER+PR+</td>
<td>OR</td>
<td>OR</td>
<td>95 % CI</td>
<td>OR</td>
<td>95 % CI</td>
</tr>
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<td>No. of cases/controls</td>
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<td>55/110</td>
<td>31/109</td>
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</tr>
<tr>
<td>Adjusted†</td>
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<td>0·74</td>
<td>0·48, 1·13</td>
<td>0·46</td>
<td>0·30, 0·73</td>
</tr>
<tr>
<td>ER+PR−</td>
<td>OR</td>
<td>OR</td>
<td>95 % CI</td>
<td>OR</td>
<td>95 % CI</td>
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<td>No. of cases/controls</td>
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<td>7/110</td>
<td>3/110</td>
<td>3/109</td>
<td></td>
</tr>
<tr>
<td>Adjusted†</td>
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<td>1·67</td>
<td>0·43, 6·56</td>
<td>0·97</td>
<td>0·19, 4·87</td>
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<tr>
<td>ER−PR+</td>
<td>OR</td>
<td>OR</td>
<td>95 % CI</td>
<td>OR</td>
<td>95 % CI</td>
</tr>
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<td>No. of cases/controls</td>
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<td>13/110</td>
<td>17/110</td>
<td>11/109</td>
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</tr>
<tr>
<td>Adjusted†</td>
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<td>0·43</td>
<td>0·21, 0·89</td>
<td>0·51</td>
<td>0·26, 1·00</td>
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<tr>
<td>ER−PR−</td>
<td>OR</td>
<td>OR</td>
<td>95 % CI</td>
<td>OR</td>
<td>95 % CI</td>
</tr>
<tr>
<td>No. of cases/controls</td>
<td>10/109</td>
<td>7/110</td>
<td>10/110</td>
<td>7/109</td>
<td></td>
</tr>
<tr>
<td>Adjusted†</td>
<td>1</td>
<td>0·74</td>
<td>0·26, 2·10</td>
<td>0·84</td>
<td>0·32, 2·23</td>
</tr>
</tbody>
</table>

Q, quartile.
* Quartile cut-off points were based on residual energy-adjusted intake among the control subjects.
† OR were adjusted for age at menarche, live birth and age at first live birth, months of breast-feeding, BMI, history of benign breast disease, mother/sister/daughter with breast cancer, physical activity, passive smoking and total energy intake.

Discussion

In the present hospital-based case–control study, we observed a statistically significant inverse association between dietary folate, vitamin B6 intake and the risk of breast cancer after adjusting for various confounding factors. The inverse association of folate intake with breast cancer risk was observed in all subtypes of ER and/or PR status. No association was found with vitamin B12 and methionine intake.

The association of dietary folate intake with breast cancer risk has been examined in many epidemiological studies. Of the fifteen case–control studies(5–13,27,29,36,38,47,48), nine have5–13 reported the protective effect of dietary folate intake on breast cancer risk. A case–control study conducted in Shanghai, China, showed that participants in the highest quintile of dietary folate had a 38 % decrease in breast cancer risk compared with those in the lowest quintile(27).

The findings of the present study are consistent with these studies. No differences were observed in the inverse association between folate intake and breast cancer risk according to folate intake from different food sources. However, some prospective cohort studies14–23 did not find a significant inverse association of dietary folate intake with breast cancer risk. A meta-analysis including nine prospective studies and fourteen case–control studies has shown a negative association between dietary folate intake and breast cancer risk in case–control studies but not in prospective studies(48). Methodological differences may partially explain the inconsistent findings.

Relatively few epidemiological studies have evaluated the association of vitamin B6, vitamin B12 and methionine intake with breast cancer. Most of the studies have not found a significant association of methionine intake(11,16,25–28), vitamin B6 or vitamin B12 intake(8,9,11,23,25–28) with breast cancer risk. In agreement with these results, neither vitamin B12 nor methionine was associated with breast cancer in the present study. However, higher vitamin B6 intake was found to be inversely associated with breast cancer risk. Since plant foods are the major sources of vitamin B6 and folate and their intakes are highly correlated, the effect of confounding may exist. Thus, further investigation into the effect of vitamin B6 on breast cancer risk is warranted.

In one case–control study(58) and five prospective cohort studies22,23,26,34,57 that have investigated the association of folate with breast cancer according to the ER and/or PR status of breast tumour, the results remained inconclusive. In the Nurses’ Health Study and the Vitamins and Lifestyle Cohort study25,34, folate intake was inversely associated with the risk of developing ER− but not ER+ tumours. In the Swedish Mammography Cohort Study(22), high folate intake was associated with the decreased risk of developing ER+/PR− breast cancer but not ER+/PR+ and ER−/PR− tumours. However, no overall association was found between folate intake and ER− or ER+ breast cancer tumours in the Nurses’ Health Study(28), the Iowa Women’s Health Study(57) and one case–control study conducted in Japan(58). In the present study, the protective effect of folate intake on breast cancer risk was not statistically significant.
cancer was observed on all subtypes of the ER/PR status of breast cancer. However, in the present study, the power to detect the interactions between dietary folate intake and ER− and PR− status was low, as the number of ER− and PR− tumour subtypes was small. Further studies with larger sample sizes are thus needed to confirm this result.

Some epidemiological studies have found a statistically significant reduction in breast cancer risk for high v. low folate intake among women with high alcohol intake. In the present study, however, the inverse association of dietary folate intake with breast cancer risk remained significant after excluding women with alcohol intake. Therefore, the present study did not support the evidence of the moderating effect of alcohol on folate intake. The reasons for the differences between the present study and others are unclear. The prevalence of alcohol intake is low in Chinese women, and only twelve (2.7%) cases and ten (2.3%) controls were regular drinkers in the present study. Further investigation in populations with a low prevalence of alcohol intake is warranted to help clarify this issue.

Folate intake is strongly influenced by various cooking methods. For example, the method of cooking of green vegetables has been found to have marked effects on folate retention and green vegetables are the major food sources for folate in our population. Therefore, the 68% reduction in risk associated with a high intake of folate that we observed in the present study may be a conservative estimate. It has been suggested that genetic polymorphisms in folate metabolic enzyme genes could influence breast cancer risk. It is biologically plausible that folate-related gene–nutrient interactions might play a role in breast cancer risk. Therefore, further studies with polymorphisms relevant to folate metabolism will help clarify the mechanism of breast cancer risk.

In summary, the present study found that intakes of dietary folate and vitamin B<sub>12</sub> were inversely associated with breast cancer risk. The inverse association was similar for ER and/or PR status. No associations were observed for vitamin B<sub>12</sub> and methionine intake and breast cancer risk.

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References

Folate intake and breast cancer risk


