Vitamin C intake and breast cancer mortality in a cohort of Swedish women

H R Harris*,1,2, L Bergkvist3 and A Wolk1

1Division of Nutritional Epidemiology, The National Institute for Environmental Medicine, Karolinska Institutet, Box 210, 171 77 Stockholm, Sweden; 2Obstetrics and Gynecology Epidemiology Center, Brigham and Women’s Hospital, 221 Longwood Avenue, Boston, MA 02115, USA and 3Department of Surgery and Centre for Clinical Research, Central Hospital, 721 89 Västerås, Sweden

Background: Vitamin C may influence cancer progression through its antioxidant properties. However, the evidence from observational epidemiologic studies on vitamin C intake and survival following breast cancer diagnosis is not consistent, and the safety of vitamin C supplements following breast cancer diagnosis has not been extensively studied.

Methods: Using a food-frequency questionnaire we investigated whether vitamin C intake was associated with survival among 3405 women diagnosed with invasive breast cancer in the Swedish Mammography Cohort.

Results: From 1987–2010, there were 1055 total deaths with 416 deaths from breast cancer. Women in the highest quartile of pre-diagnosis vitamin C intake had an adjusted HR (95% CI) of breast cancer death of 0.75 (0.57–0.99) compared with those in the lowest quartile ($P_{\text{trend}} = 0.03$). There was a borderline significant association between vitamin C intake and total mortality (HR = 0.84; 95% CI = 0.71–1.00; $P_{\text{trend}} = 0.08$). Among 717 breast cancer cases for whom post-diagnosis supplement use was available, there was no association between vitamin C supplement use ($\geq 1000 \text{mg}$) and breast cancer-specific mortality (HR = 1.06; 95% CI = 0.52–2.17).

Conclusion: Our findings suggest that dietary vitamin C intake before breast cancer diagnosis may be associated with breast cancer survival. In addition, post-diagnosis vitamin C supplementation at the level observed in our population was not associated with survival.

Vitamin C is a water-soluble nutrient that has been hypothesised to influence cancer initiation and promotion through its antioxidant properties including the neutralization of free radicals (Frei, 1994; Willcox et al, 2004). In addition, in vitro experiments have shown cytotoxic action of vitamin C against cancer cells without subsequent toxicity to normal cells (Chen et al, 2008; Ullah et al, 2011). However, the evidence from observational epidemiologic studies on vitamin C intake and survival following breast cancer diagnosis is not consistent, with dietary vitamin C intake reported to reduce the risk of mortality in some studies (Rohan et al, 1993; Ingram, 1994; Jain et al, 1994; Fleischauer et al, 2003; McEligot et al, 2006) and no association in other studies (Zhang et al, 1995; Hebert et al, 1998; Holmes et al, 1999; Saxe et al, 1999; Saquib et al, 2011). In addition, the safety of oral vitamin C supplements following cancer diagnosis is not clear (Lavenda et al, 2008) and few studies have examined vitamin C supplements in relation to breast cancer survival (Greenlee et al, 2009; Nechuta et al, 2011; Greenlee et al, 2012).

The aim of this study was to investigate whether pre-diagnosis dietary vitamin C intake was associated with total and breast cancer-specific mortality among women diagnosed with invasive breast cancer in the Swedish Mammography Cohort (SMC). We also examined whether the association between vitamin C and survival differed by hormone receptor status, disease stage at diagnosis, age, body mass index (BMI) and smoking. In a subset of women, we examined whether vitamin C supplement intake following breast cancer diagnosis was associated with survival.

*Correspondence: Dr H Harris, E-mail: holly.harris@ki.se

Received 4 March 2013; revised 6 May 2013; accepted 12 May 2013; published online 4 June 2013

© 2013 Cancer Research UK. All rights reserved 0007 – 0920/13

www.bjcancer.com | DOI:10.1038/bjc.2013.269
MATERIALS AND METHODS

Study population. This study included 3405 participants in the SMC with invasive breast cancer diagnosed from 1987–2010. Recruitment and characteristics of this cohort have been previously described (Wolk et al, 2006). In brief, the SMC is a population-based cohort of 66651 women born between 1914 and 1948 that were recruited between 1987 and 1990 in Västmanland and Uppsala counties in central Sweden. Participants completed a baseline questionnaire with questions regarding diet, reproductive and other factors. In 1997, a second questionnaire was extended to include dietary supplements, physical activity and smoking status, and was sent to participants who were still alive and residing in the study area; 39227 (70%) women returned this questionnaire. Those with an incorrect or missing national registration number, previous cancer diagnosis (except non-melanoma skin cancer) and implausible total energy intake (3 standard deviations (s.d) from the mean value for log transformed energy intake) were excluded from the baseline cohort. Completion and return of the self-administered questionnaire was treated as informed consent of study participants. The study was approved by the ethics committee at the Karolinska Institutet.

Histologically confirmed incident invasive breast cancer cases were ascertained by linkage of the cohort with the Swedish Cancer Registry. (Mattsson and Wallgren, 1984). Oestrogen receptor (ER) and progestrone receptor (PR) status, menopausal status at diagnosis, tumour size, grade, lymph node involvement and type of treatment were available for ~77% of the cases. More detailed information on the evaluation of hormone receptor status in this cohort has been described previously (Larsson et al, 2009).

Dietary assessment. Diet was assessed using a 67-item food-frequency questionnaire (FFQ) at baseline and a 96-item FFQ in 1997. Participants were asked how often, on average, they had consumed each item during the previous 6 months (1987) or year (1997). Vitamin C intake was calculated as the frequency of consumption of each food item multiplied by its vitamin C content per age-specific serving (Bergström et al, 1991). Women were asked about dietary supplement use on the 1997 questionnaire including predefined questions about vitamin C. In Swedish populations, 1000 mg has been reported as the most frequently used dose of single nutrient vitamin C supplements (Holmquist et al, 2003; Messerer and Wolk, 2004). The FFQ has been previously validated for vitamin C and for foods that were the main sources of vitamin C. The correlation coefficients between the questionnaire and four 1-week diet records were 0.3 for dietary vitamin C, 0.5 for citrus fruits, boiled potatoes and apples/pears, 0.4 for juice, tomatoes and bananas, and 0.3 for fruit drinks (A Wolk, unpublished data, 1992). The sensitivity and specificity of vitamin C supplement use have been estimated to be 67% and 95%, respectively (Messerer and Wolk, 2004). Nutrient intakes were adjusted for energy using the residual method (Willett and Stampfer, 1986).

Outcome assessment. Date of death was identified through linkage to the Swedish National Death Registry at Statistics Sweden. It is estimated that 93% of all deaths in Sweden are reported within 10 days and 100% are reported within 30 days (Ludvigsson et al, 2009). Cause of death was determined by International Classification of Diseases (ICD) codes (ICD9 and ICD10) through linkage to the Cause of Death Registry at the National Bureau of Health and Welfare.

Statistical analysis. Cox proportional hazard models were used to calculate hazard ratios (HRs) and 95% confidence intervals (95% CIs) for death from any cause. Participants contributed person-time from the date of breast cancer diagnosis until death from any cause, or end of follow-up on 16 October 2010. We also examined death from breast cancer and non-breast cancer death as the end-points with end of follow-up on 31 December 2008 as information on the cause of death was not available after this time. Baseline diet (1987) was considered the exposure in all analyses except when dietary supplement use and dietary change were examined. Dietary vitamin C intake was categorised in quartiles with the lowest quartile as the reference group. Vitamin C supplement use was categorised as supplement user and non-supplement user. Total caloric intake and age at diagnosis were included in all models.

Education, marital status, menopausal status at diagnosis, BMI, alcohol and year of diagnosis were considered potential confounders in all multivariable models. Parity/age at first birth, oral contraceptive use, postmenopausal hormone use, height and family history of breast cancer were not observed to be confounders in the study population and therefore were not included in the final models. Categories were created for missing data. Multivariable models were adjusted for the following clinical characteristics: stage, grade of tumour, radiation treatment and chemotherapy/ hormonal therapy. Additional adjustment for the clinical covariates tumour size and number of positive lymph nodes did not further alter the effect estimates, thus were not included in the final models. Tests for linear trend were performed by assigning the median value of each category to each participant in that group.

We examined whether the association between vitamin C and breast cancer survival differed by hormone receptor status (ER+, ER−, PR+, PR−, ER+/PR+, ER−/PR−), disease stage at diagnosis (I, II, III/IV), age (<65 years, ≥65 years), BMI (<30 kg m−2, ≥30 kg m−2) and smoking status (never, ever), with a likelihood ratio test comparing the model with the cross-product term between vitamin C and each potential effect modifier to the model with main effects only. All tests of statistical significance were two-sided, and analyses were performed using SAS Version 9.2 (SAS Institute Inc., Cary, NC, USA).

RESULTS

During 30 080 person-years of follow-up contributed by 3405 breast cancer cases, there were 1055 deaths with 416 deaths from breast cancer. The mean (± s.d.) age at diagnosis 65 years (±10.3) and the median follow-up time was 7.8 years (range 1 month to 23.5 years). Pre-diagnosis dietary assessment occurred a mean of 11.0 years before breast cancer diagnosis (range 1 month to 23.4 years). Among the subset of women with post-diagnosis dietary information, dietary assessment occurred a mean of 4.6 years after breast cancer diagnosis (range 1 year to 10 years). The mean dietary vitamin C intake was 72.2 mg d−1 (±40.1). The main sources of dietary vitamin C in the study population were citrus fruit (28.0%), boiled potatoes (14.0%), juice (13.5%), tomatoes (8.8%) and apples/pears (7.7%). Women in the highest quartile of vitamin C intake were more likely to have used oral contraceptives and postmenopausal hormones, were younger at cohort enrolment and had a lower mean BMI than women in the lowest quartile of vitamin C intake (Table 1).

Pre-diagnosis dietary vitamin C intake was associated with a decreased risk of breast cancer death (Table 2). Women in the highest quartile of dietary vitamin C intake had a covariate-adjusted HR (95% CI) of death from breast cancer of 0.74 (0.57–0.98) compared with those in the lowest quartile (P trend = 0.04). Further adjustment for clinical characteristics and treatment did not alter the results (HR = 0.75; 95% CI = 0.57–0.99; P trend = 0.03). In a sensitivity analysis that excluded women with stage IV breast cancer, results were not materially different from the main analysis (covariate and clinical characteristics-adjusted...
There was a borderline significant association between vitamin C intake and total mortality (HR = 0.84; 95% CI = 0.71–1.00; Ptrend = 0.08); however, there was no association between vitamin C intake and non-breast cancer deaths (HR = 0.91; 95% CI = 0.73–1.13; Ptrend = 0.65) (Table 2).

When the association between pre-diagnosis dietary vitamin C and breast cancer death was stratified by hormone receptor status of the tumour, the protective association appeared strongest among those with ER-negative/PR-negative tumours (Table 3). Among women with ER-negative/PR-negative tumours, those in the highest quartile of dietary vitamin C intake had a covariate and clinical characteristics-adjusted HR of 0.46 (95% CI = 0.22–0.96; Ptrend = 0.008) compared with women in the lowest quartile. The corresponding HR for ER-positive/PR-positive breast cancer was 0.80 (95% CI = 0.47–1.35; Ptrend = 0.52); however, the interaction was not significant (Ptrend = 0.73). The stronger association observed among women with ER-negative/PR-negative tumours appeared to be driven by the ER-negative receptor status as a significant inverse association was observed among women with ER-negative tumours (HR = 0.50, 95% CI = 0.28–0.89; Ptrend = 0.004), but not among those with PR-negative tumours (HR = 0.76, 95% CI 0.45–1.27; Ptrend = 0.15). A similar association was observed when total mortality was the outcome (data not shown).

We examined whether the association between vitamin C and breast cancer mortality varied by the reactive oxygen species (ROS)-related factors of age, obesity and smoking. Vitamin C intake had a stronger inverse association with breast cancer mortality among women who were aged ≥ 65 (HR = 0.48;
Vitamin C and breast cancer survival

DISCUSSION

In this prospective cohort study among 3405 women with breast cancer, dietary vitamin C intake was inversely associated with breast cancer-specific mortality. In addition, the association appeared to differ by age with a stronger inverse association observed among women aged ≥65 years. There was no association between post-diagnosis vitamin C supplement use and mortality, however these results were based on small numbers.

Results from observational studies on dietary vitamin C intake and survival following breast cancer diagnosis have not been consistent, with dietary vitamin C intake reported to reduce the risk of mortality in some studies (Rohan et al, 1993; Ingram, 1994; Jain et al, 1994; Fleischauer et al, 2003; McEligot et al, 2006) and no association reported in other studies (Zhang et al, 1995; Hebert et al, 1998; Holmes et al, 1999; Saxe et al, 1999; Saquib et al, 2011). In addition, two additional studies have only examined post-diagnosis supplement use reporting inverse associations between vitamin C supplement use and all-cause mortality or recurrence (Nechuta et al, 2011; Greenlee et al, 2012). The varied results may be in part because the measurement of dietary vitamin C has occurred both pre- and post-diagnosis as well as among populations with varying intakes of vitamin C, with not all studies capturing supplement use. Consistent with our results, four (Rohan et al, 1993; Ingram, 1994; Jain et al, 1994; McEligot et al, 2006) of

| Quartile of vitamin C intake (mg d⁻¹) | <42.9 | 42.9–65.5 | 65.6–92.4 | ≥92.5 | \( P_{\text{trend}} \)^a |
|--------------------------------------|-------|----------|----------|-------|-----------------|
| Person-years                         | 7458  | 7523     | 7584     | 7516  | —               |
| Breast cancer deaths                 | 128   | 103      | 97       | 88    | —               |
| Age-adjusted model                   | 1.00  | 0.80 (0.62–1.04) | 0.75 (0.58–0.98) | 0.69 (0.52–0.90) | 0.007        |
| Covariate-adjusted modelb           | 1.00  | 0.84 (0.65–1.09) | 0.81 (0.62–1.06) | 0.74 (0.57–0.98) | 0.04         |
| Covariate-adjusted model + clinical characteristicsc | 1.00  | 0.89 (0.69–1.16) | 0.77 (0.59–1.01) | 0.75 (0.57–0.99) | 0.03         |
| Non-breast cancer deaths             | 192   | 145      | 153      | 149   | —               |
| Age-adjusted model                   | 1.00  | 0.81 (0.65–1.00) | 0.93 (0.75–1.15) | 0.90 (0.72–1.11) | 0.58         |
| Covariate-adjusted modelb           | 1.00  | 0.83 (0.67–1.03) | 0.96 (0.77–1.19) | 0.92 (0.74–1.14) | 0.70         |
| Covariate-adjusted model + clinical characteristicsc | 1.00  | 0.84 (0.68–1.05) | 0.97 (0.78–1.20) | 0.91 (0.73–1.13) | 0.65         |
| Total deaths                         | 320   | 248      | 250      | 237   | —               |
| Age-adjusted model                   | 1.00  | 0.80 (0.68–0.95) | 0.85 (0.72–1.00) | 0.80 (0.68–0.95) | 0.03         |
| Covariate-adjusted modelb           | 1.00  | 0.84 (0.71–0.99) | 0.90 (0.76–1.06) | 0.85 (0.71–1.00) | 0.12         |
| Covariate-adjusted model + clinical characteristicsc | 1.00  | 0.86 (0.72–1.01) | 0.89 (0.75–1.05) | 0.84 (0.71–1.00) | 0.08         |

^aDetermined using category medians
bCox proportional hazard model adjusted for age (continuous), energy intake (continuous), education level (primary, high school, university), marital status (single, married, divorced, widowed, living with partner), menopausal status at diagnosis (premenopausal, postmenopausal, unknown), body mass index (<20, 20–24.9, 25–29.9, ≥30 kg m⁻²), alcohol intake (non-drinker, <3.4, 3.4–9.9, ≥10 g d⁻¹) and calendar year of diagnosis (continuous)
cCox proportional hazard model adjusted for the variables above plus disease stage (I, II, III/IV), grade (I, II, III), radiation treatment (yes/no), and chemotherapy and/or hormonal treatment (no chemotherapy or hormonal treatment, hormonal therapy and no chemotherapy, chemotherapy and no hormonal therapy, and hormonal therapy and chemotherapy).
in cell signalling events (Loo, 2003). Vitamin C may inhibit cancer cell proliferation through the suppression of H₂O₂ and its ROS products (Frei, 1994; Willcox et al, 2003). In addition, neutrophil granulocytes (cells) can generate hydrogen peroxide (H₂O₂) through the transformation of H₂O₂ to hydroxyl radicals as well as through the involvement of H₂O₂ in cell signalling events (Loo, 2003). Vitamin C may inhibit cancer cell proliferation through the suppression of H₂O₂ and its ROS products (Frei, 1994; Willcox et al, 2004). In addition, at high doses vitamin C may also function as a pro-oxidant causing cytotoxicity to cancer cells without similar effects on normal cells (Chen et al, 1994).

the seven studies (Rohan et al, 1993; Ingram, 1994; Jain et al, 1994; Zhang et al, 1995; Saxe et al, 1999; Fleischauer et al, 2003; McElligot et al, 2006) examining pre-diagnosis dietary intake reported a significant inverse association with mortality while only one (Fleischauer et al, 2003) of the four studies (Hebert et al, 1998; Holmes et al, 1999; Fleischauer et al, 2003; Saquib et al, 2011) examining post-diagnosis dietary intake reported a similar association. We had longer follow-up than previous studies (median = 7.8 years) as well as more than twice as many deaths providing us with ample power to examine these associations. In addition, the differences in results between pre- and post-diagnosis intake may indicate that the timing and duration of vitamin C intake may be important. In the FASTCAB study, vitamin C supplement use, including pre- and post-diagnosis intake for > 4 years, was associated with statistically significant decreased risk of breast cancer-related mortality and recurrence, while supplement use for 0–3 years had a non-significant inverse association with breast cancer-related mortality and recurrence (Fleischauer et al, 2003). In addition, Nechuta et al (2011) reported an inverse association between > 3 months of post-diagnosis vitamin C supplement use and total mortality, but no association with ≤ 3 months of use.

Cancer cell proliferation is hypothesised to be stimulated by hydrogen peroxide (H₂O₂) through the transformation of H₂O₂ into hydroxyl radicals as well as through the involvement of H₂O₂ in cell signalling events (Loo, 2003). Vitamin C may inhibit cancer cell proliferation through the suppression of H₂O₂ and its ROS products (Frei, 1994; Willcox et al, 2004). In addition, at high doses vitamin C may also function as a pro-oxidant causing cytotoxicity to cancer cells without similar effects on normal cells (Chen et al, 2006).

Table 3. Hazard ratios (HR) and 95% confidence intervals (95% CI) of breast cancer death across hormone receptor subtypes by quartile of vitamin C intake among 3405 invasive breast cancer cases in the Swedish Mammography Cohort

| Quartile of vitamin C intake (mg d⁻¹) | <42.9 | 42.9–65.5 | 65.6–92.4 | ≥ 92.5 | P_trend | P_heterogeneity |
|--------------------------------------|------|--------|--------|-------|---------|---------------|
| **ER-positive/PR-positive**          |      |        |        |       |         |               |
| Breast cancer deaths                 | 35   | 25     | 29     | 25    | —       | —             |
| Person-years                         | 3113 | 3426   | 3287   | 3410  | —       | 0.73          |
| Covariate-adjusted model             | 1.00 | 0.66 (0.39–1.11) | 0.84 (0.51–1.38) | 0.70 (0.42–1.17) | 0.32 | — |
| Covariate-adjusted model + clinical characteristics | 1.00 | 0.75 (0.44–1.27) | 0.85 (0.51–1.42) | 0.80 (0.47–1.35) | 0.52 | — |
| **ER-negative/PR-negative**          |      |        |        |       |         |               |
| Breast cancer deaths                 | 18   | 23     | 16     | 15    | —       | —             |
| Person-years                         | 749  | 573    | 641    | 802   | —       | —             |
| Covariate-adjusted model             | 1.00 | 1.36 (0.71–2.59) | 0.96 (0.48–1.93) | 0.67 (0.33–1.36) | 0.14 | — |
| Covariate-adjusted model + clinical characteristics | 1.00 | 1.17 (0.60–2.29) | 0.49 (0.23–1.06) | 0.46 (0.22–0.96) | 0.008 | — |
| **ER-positive**                      |      |        |        |       |         |               |
| Breast cancer deaths                 | 52   | 36     | 40     | 39    | —       | —             |
| Person-years                         | 3961 | 4527   | 4239   | 4286  | —       | 0.65          |
| Covariate-adjusted model             | 1.00 | 0.62 (0.41–0.96) | 0.77 (0.51–1.17) | 0.73 (0.48–1.12) | 0.32 | — |
| Covariate-adjusted model + clinical characteristics | 1.00 | 0.73 (0.47–1.13) | 0.82 (0.54–1.26) | 0.88 (0.57–1.35) | 0.72 | — |
| **ER-negative**                      |      |        |        |       |         |               |
| Breast cancer deaths                 | 32   | 29     | 20     | 21    | —       | —             |
| Person-years                         | 1113 | 866    | 1059   | 1039  | —       | —             |
| Covariate-adjusted model             | 1.00 | 1.08 (0.65–1.82) | 0.74 (0.42–1.17) | 0.66 (0.37–1.52) | 0.07 | — |
| Covariate-adjusted model + clinical characteristics | 1.00 | 0.97 (0.57–1.64) | 0.42 (0.23–0.78) | 0.50 (0.28–0.89) | 0.004 | — |
| **PR-positive**                      |      |        |        |       |         |               |
| Breast cancer deaths                 | 49   | 31     | 33     | 31    | —       | —             |
| Person-years                         | 3491 | 3735   | 3715   | 3647  | —       | 0.61          |
| Covariate-adjusted model             | 1.00 | 0.61 (0.39–0.97) | 0.70 (0.45–1.09) | 0.64 (0.41–1.02) | 0.11 | — |
| Covariate-adjusted model + clinical characteristics | 1.00 | 0.69 (0.43–1.10) | 0.65 (0.41–1.03) | 0.72 (0.45–1.15) | 0.18 | — |
| **PR-negative**                      |      |        |        |       |         |               |
| Breast cancer deaths                 | 35   | 34     | 27     | 29    | —       | —             |
| Person-years                         | 1588 | 1659   | 1559   | 1652  | —       | —             |
| Covariate-adjusted model             | 1.00 | 0.91 (0.56–1.48) | 0.78 (0.47–1.31) | 0.79 (0.48–1.31) | 0.32 | — |
| Covariate-adjusted model + clinical characteristics | 1.00 | 0.99 (0.60–1.63) | 0.64 (0.38–1.09) | 0.76 (0.45–1.27) | 0.15 | — |

a Determined using category medians.
b Value from likelihood ratio test comparing a model with the cross-product term between vitamin C and hormone receptor status to the model with main effects only.
c Cox proportional hazard model adjusted for age (continuous), energy intake (continuous), education level (primary, high school, university), marital status (single, married, divorced, widowed, living with partner), menopausal status at diagnosis, (premenopausal, postmenopausal, unknown), body mass index (<20, 20–24.9, 25–29.9, ≥30 kg m⁻²), alcohol intake (non-drinker, <20, 20–24.9, ≥10 g d⁻¹) calendar year of diagnosis (continuous).
d Covariate-adjusted model adjusted for the variables above plus disease stage (I, II, III/IV), grade (I, II, III), radiation treatment (yes/no), and chemotherapy and/or hormonal treatment (no chemotherapy or hormonal treatment, hormonal therapy and no chemotherapy, chemotheraphy and no hormonal therapy, and hormonal therapy and chemotherapy).

BRITISH JOURNAL OF CANCER
et al 2008; Ullah et al, 2008; Greenlee et al., 2009). While in vitro studies support a role for vitamin C in cancer outcomes, the literature is not clear on the safety of oral supplements containing vitamin C following cancer diagnosis (Lawenda et al, 2008) and few studies have specifically examined breast cancer survival (Greenlee et al, 2009). In addition, it has been hypothesised that use of antioxidant supplements, including vitamin C, during cancer treatment may actually protect cancer cells from treatment agents (D’Andrea, 2005; Lawenda et al, 2008). Two recent observational studies examined post-diagnosis supplement use in women with breast cancer and both observed that vitamin C supplement use was associated with a decreased risk of breast cancer mortality and/or recurrence (Nechuta et al, 2011; Greenlee et al, 2012), while in one study frequent use of carotenoids was associated with increased mortality risk (Greenlee et al, 2012). In contrast, early randomized trials of oral vitamin C supplements in cancer patients demonstrated no benefit of high-dose vitamin C on cancer survival (Creagan et al, 1979; Moertel et al, 1985); however, there have been no randomized trials evaluating oral vitamin C supplementation specifically among breast cancer patients. In the subset of women with information on post-diagnosis diet, we did not observe an association between vitamin C supplement use and survival; however, we had limited power for this analysis. The most frequently reported dose of vitamin C supplement in two Swedish populations has been reported to be 1000 mg (Holmquist et al, 2003; Messerer and Wolk, 2004), which is higher than in the Shanghai Breast Cancer Survival Study, where ~85% of women with dosage information used ≤400 mg per day (Nechuta et al, 2011), but lower than the mega-doses (> 1 g) used in other studies of supplement use and cancer outcomes (Lawenda et al, 2008; Greenlee et al, 2009). Levine et al (1996) have reported that plasma concentrations of vitamin C reach near saturation at doses of 400 mg d⁻¹ and that bioavailability declines at doses of 500 mg d⁻¹ and higher. The amount of vitamin C obtained from dietary sources is considerably lower than what can be obtained through oral supplementation. Thus adequate dietary intake of vitamin C may influence cancer

---

Table 4. Hazard ratios (HR) and 95% confidence intervals (95% CI) of breast cancer death stratified by selected characteristics by quartile of vitamin C intake among 3405 invasive breast cancer cases in the Swedish Mammography Cohort

| Quartile of vitamin C intake (mg d⁻¹) | <42.9 | 42.9–65.5 | 65.6–92.4 | ≥92.5 | \( P_{\text{trend}}^{a} \) |
|---|---|---|---|---|---|
| **Age** | | | | | |
| <65 | 56 | 53 | 54 | 58 | 0.69 |
| Breast cancer deaths | 1.00 | 0.91 (0.62–1.34) | 0.81 (0.55–1.19) | 1.09 (0.75–1.59) | 0.03 |
| Covariate-adjusted model + clinical characteristics \( b \) | | | | | |
| \( P_{\text{interaction}}^{a} = 0.03 \) | | | | | |
| ≥65 | 72 | 50 | 43 | 30 | 0.0007 |
| Breast cancer deaths | 1.00 | 0.88 (0.61–1.28) | 0.76 (0.52–1.12) | 0.48 (0.31–0.74) | 0.30 |
| Covariate-adjusted model + clinical characteristics \( b \) | | | | | |
| \( P_{\text{interaction}}^{a} = 0.30 \) | | | | | |
| **BMI** | | | | | |
| <30 | 102 | 84 | 90 | 74 | 0.17 |
| Breast cancer deaths | 1.00 | 0.86 (0.64–1.15) | 0.84 (0.63–1.12) | 0.80 (0.59–1.09) | 0.17 |
| Covariate-adjusted model + clinical characteristics \( b \) | | | | | |
| \( P_{\text{interaction}}^{a} = 0.30 \) | | | | | |
| ≥30 | 21 | 18 | 4 | 8 | 0.04 |
| Breast cancer deaths | 1.00 | 1.33 (0.64–2.77) | 0.36 (0.12–1.10) | 0.54 (0.22–1.35) | 0.36 |
| Covariate-adjusted model + clinical characteristics \( b \) | | | | | |
| \( P_{\text{interaction}}^{a} = 0.30 \) | | | | | |
| **Smoking** | | | | | |
| Never smoker | 25 | 22 | 17 | 19 | 0.03 |
| Breast cancer deaths | 1.00 | 0.80 (0.44–1.45) | 0.55 (0.29–1.04) | 0.53 (0.28–0.99) | 0.03 |
| Covariate-adjusted model + clinical characteristics \( b \) | | | | | |
| | | | | | |
| Ever smoker | 18 | 12 | 12 | 20 | 0.14 |
| Breast cancer deaths | 1.00 | 0.93 (0.42–2.02) | 0.70 (0.33–1.49) | 1.82 (0.90–3.67) | 0.10 |
| Covariate-adjusted model + clinical characteristics \( b \) | | | | | |
| \( P_{\text{interaction}}^{a} = 0.10 \) | | | | | |

---

\( a \) Determined using category medians.

\( b \) Cox-proportional hazard model adjusted for age (continuous), energy intake (continuous), education level (primary, high school, university), marital status (single, married, divorced, widowed, living with partner), menopausal status at diagnosis (premenopausal, postmenopausal, unknown), body mass index (<20, 20–24.9, 25–29.9, ≥30 kg m⁻²), alcohol intake (non-drinker, < 3.4, 3.4–9.9, ≥10 g d⁻¹), calendar year of diagnosis (continuous), disease stage (I, II, III/IV), grade (I, II, III), radiation treatment (yes/no) and chemotherapy and/or hormonal treatment (no chemotherapy or hormonal treatment, hormonal therapy and no chemotherapy, chemotherapy and no hormonal therapy, and hormonal therapy and chemotherapy). BMI is not adjusted for in models stratified by BMI and alcohol intake is not adjusted for in models stratified by alcohol.

\( P \)-value from likelihood ratio test comparing a model with the cross-product term between vitamin C and potential effect modifier to the model with main effects only.
progression and survival through different mechanisms than supplementation, and a potential U-shaped relation may exist between total vitamin C intake (from diet and supplements) and survival. In addition, the route of administration, oral vs intravenous, of vitamin C intake may also have a role in the efficacy and safety of vitamin C use in cancer patients as these different routes of administration have differing effects on plasma concentrations (Padayatty et al., 2004).

To our knowledge, only one study has examined whether the association between vitamin C intake and mortality among women with breast cancer differs by hormone receptor status. In addition, cohort studies examining dietary vitamin C intake and breast cancer risk have not supported an association that varies by hormone receptor status (Cui et al., 2008). Jain et al. (1994) reported marginally more inverse hazard ratios for mortality for hormone receptor-positive tumours compared with receptor-negative tumours, but the direction and magnitude of the effects were similar between hormone receptor-positive and -negative tumours, no P-values for interaction were reported, and the total number of deaths was only 88. The protective association between dietary vitamin C and breast cancer survival in our population was most evident among women with ER-negative/PR-negative tumours, although the interaction did not reach statistical significance. The potential mechanism behind the stronger association between dietary vitamin C intake and mortality among those with ER-negative/PR-negative tumours is unclear. However, the stronger association observed with these tumours may simply be more apparent, as they are not as susceptible to other factors that are mediated through oestrogen exposure (Huang et al., 2000; Colditz et al., 2004).

We also investigated whether the association between dietary vitamin C intake and breast cancer mortality differed by ROS-related factors. We observed suggestions of stronger inverse associations between dietary vitamin C intake and breast cancer mortality among women aged ≥65 and obese; however, the interaction was only significant for age. The mechanism underlying the interaction between age and dietary vitamin C intake is unclear; however, age is associated with increased ROS production coupled with a decrease in the clearance of ROS that increases oxidative stress (Cannizzo Elvira et al., 2012). Consequently vitamin C may have a greater impact as a scavenger of free radicals with increasing age. Obesity is also associated with oxidative stress (Olusi, 2002; Keaney et al., 2003; Furukawa et al., 2004), and may interact with vitamin C intake in a similar manner.

The limitations of our study need to be considered. Seventy-nine per cent of our participants had only a pre-diagnosis assessment of diet and thus we had limited power to examine post-diagnosis diet. Studies among women with breast cancer have reported dietary changes following diagnosis in 30–40% of women (Salminen and Lagstrom, 2000; Maunsell et al., 2002; Salminen et al., 2004), with increased consumption of fruits and vegetables reported in most (Salminen and Lagstrom, 2000; Maunsell et al., 2002; Salminen et al., 2004; Velentzis et al., 2011) but not all (Wayne et al., 2004) studies. One study examined vitamin C intake and reported a significant increase in vitamin C intake following breast cancer diagnosis (Velentzis et al., 2011). However, younger women were most likely to report these changes and the average age at breast cancer diagnosis in our cohort was 65.1 years. In our study, up to 82% of women who completed a FFQ post-diagnosis remained in the same or adjacent quartile of vitamin C intake following diagnosis. In addition, supplement use was not assessed at baseline and we had limited power to examine the association between post-diagnosis supplement use and mortality. Residual or unmeasured confounding by lifestyle or other dietary factors is also a possibility. However, we adjusted for foods that contributed to vitamin C intake as well as for physical activity in the subset of cases and the associations did not materially change.

To our knowledge, this is the largest study to examine the relation between dietary vitamin C and mortality among women with breast cancer, giving us the power to examine whether the association varied by hormone receptor status or ROS-generating factors. We also have complete follow-up of all cases, a long follow-up period and data on many important covariates, including clinical and lifestyle characteristics.

In conclusion, we observed that dietary vitamin C intake before breast cancer diagnosis was associated with breast cancer-specific survival. This association was strongest among women aged ≥65. In addition, we did not observe a harmful effect of post-diagnosis vitamin C supplementation with the dosages of ~1000 mg. Future studies examining vitamin C intake from food and supplements are needed to further our understanding of the impact of the timing and dose of vitamin C intake on outcomes in women with breast cancer.

ACKNOWLEDGEMENTS

This work was supported by the Swedish Cancer Foundation, the Swedish Research Council/Committee for Infrastructure, the Swedish Foundation for International Cooperation in Research and Higher Education, and the Regional Research Fund Uppsala-Örebro Region.

REFERENCES

Bergström L, Kylberg E, Hagman U, Erikson H, Bruce Å (1991) The food composition database KOST; the National Food Administration’s Information System for nutritive values of food. Vår Föda 43: 439–447.

Cannizzo Elvira S, Clement Cristina C, Morozova K, Valdor R, Kaushik S, Almeida Larissa N, Follo C, Sahu R, Cuervo Ana M, Macian F, Santambrogio L (2012) Age-related oxidative stress compromises endosomal proteostasis. Cell Rep 2(1): 136–149.

Chen Q, Espey MG, Sun AY, Pooput C, Kirk KL, Krishna MC, Khosh DB, Drisko J, Levine MV (2008) Pharmacologic doses of ascorbate act as a prooxidant and decrease growth of aggressive tumor xenografts in mice. PNAS 105(32): 11105–11109.

Colditz GA, Rosner BA, Chen WY, Holmes MD, Hankinson SE (2004) Risk factors for breast cancer according to estrogen and progesterone receptor status. J Natl Cancer Inst 96(3): 218–228.

Creagan ET, Moertel CG, O’Fallon JR, Schutt AJ, O’Connell MJ, Rubin J, Frytak S (1979) Failure of high-dose vitamin C (ascorbic acid) therapy to benefit patients with advanced cancer. N Engl J Med 301(13): 687–690.

Cui Y, Shikany JM, Liu S, Shaygufta Y, Rohan TE (2008) Selected antioxidants and risk of hormone receptor–defined invasive breast cancers among postmenopausal women in the Women’s Health Initiative Observational Study. Am J Clin Nutr 87(4): 1009–1018.

D’Andrea GM (2005) Use of antioxidants during chemotherapy and radiotherapy should be avoided. CA Cancer J Clin 55(3): 319–321.

Fleischauer AT, Simonsen N, Arab L (2003) Antioxidant supplements and risk of breast cancer recurrence and breast cancer–related mortality among postmenopausal women. Nutr Cancer 46(1): 15–22.

Frei B (1994) Reactive oxygen species and antioxidant vitamins: Mechanisms of action. Am J Med 97(S, Supplement 1): S5–S13.

Furukawa S, Fujita T, Shimabukuro M, Iwaki M, Yamada Y, Nakajima Y, Nakayama O, Makishima M, Matsuda M, Shimomura I (2004) Increased oxidative stress in obesity and its impact on metabolic syndrome. J Clinical Invest 114(12): 1752–1761.

Greenlee H, Hershman D, Jacobson J (2009) Use of antioxidant supplements during breast cancer treatment: a comprehensive review. Breast Cancer Res Treat 115(3): 437–452.

Greenlee H, Kwan ML, Kushi LH, Song J, Castillo A, Weltzien E, Quesenberry CP, Caan BJ (2012) Antioxidant supplement use after breast cancer diagnosis and mortality in the Life After Cancer Epidemiology (LACE) cohort. Cancer 118(8): 2048–2058.
Hebert JR, Hurley TG, Ma Y (1998) The effect of dietary exposures on recurrence and mortality in early stage breast cancer. *Breast Cancer Res Treat* 51(1): 17–28.

Holmqist C, Larsson S, Wolk A, de Faire U (2003) Multivitamin supplements are inversely associated with risk of myocardial infarction in men and women—Stockholm Heart Epidemiology Program (SHEEP). *J Nutr* 133(8): 2650–2654.

Holmes MD, Stampfer MJ, Colditz GA, Rosner B, Hunter DJ, Willett WC (1999) Dietary factors and the survival of women with breast carcinoma. *Cancer* 86(5): 826–835.

Huang W-Y, Newman B, Milikan RC, Schell MJ, Hulka BS, Moorman PG (2000) Hormone-related factors and risk of breast cancer in relation to estrogen receptor and progesterone receptor status. *Am J Epidemiol* 151(7): 703–714.

Ingram D (1994) Diet and subsequent survival in women with breast cancer. *Br J Cancer* 69(3): 592–595.

Jain M, Miller AB, To T (1994) Premorbid diet and the prognosis of women with breast cancer. *J Natl Cancer Inst* 86(18): 1390–1397.

Keeney JF, Larson MG, Vasan RS, Wilson PWF, Lipinska J, Corey D, Massaro JM, Sutherland P, Vita JA, Benjamin EJ (2003) Obesity and systemic oxidative stress. *Arterioscler Thromb Vasc Biol* 23(3): 434–439.

Larsson SC, Bergqvist L, Wolk A (2009) Long-term meat intake and risk of breast cancer by oestrogen and progesterone receptor status in a cohort of Swedish women. *Eur J Cancer* 45(17): 3042–3046.

Lawenda BD, Kelly KM, Ladas EJ, Sagar SM, Vickers A, Blumberg JB (2008) Should supplemental antioxidant administration be avoided during chemotherapy and radiation therapy? *J Natl Cancer Inst* 100(11): 773–783.

Levine M, Conry-Cantilena C, Wang Y, Welch RW, Washko PW, Dhariwal KR, Park JB, Lazarev A, Graumlich JF, King J, Cantilena LR (1996) Vitamin C pharmacokinetics in healthy volunteers: evidence for a recommended dietary allowance. *Proc Natl Acad Sci USA* 93(8): 3704–3709.

Loo G (2003) Redox-sensitive mechanisms of phytochemical-mediated inhibition of cancer cell proliferation (review). *J Nutr Biochem* 14(2): 64–73.

Ludvigsson J, Otterblad-Olausson P, Pettersson B, Ekholm A (2009) The Swedish personal identity number: possibilities and pitfalls in healthcare and medical research. *Eur J Epidemiol* 24(11): 659–667.

Mattsson B, Wallgren A (1984) Completeness of the Swedish Cancer Register. *Non-notified cancer cases recorded on death certificates in 1978*. Acta Radiol Oncol 23: 305–313.

Maunsell E, Drolet M, Brisson J, Robert J, Deschénes L (2002) Dietary change in overweight and eat less fat. The Iowa women's health study. *Eur J Clin Nutr* 56(11): 844–848.

McEligot A, Largent J, Ziogas A, Peel D, Anton-Culver H (2006) Dietary fat, estrogen receptor and progesterone receptor status. *Nutr Cancer* 58(1): 137–141.

Nechuta S, Lu W, Chen Z, Zheng Y, Gu K, Cai H, Zheng W, Shu XO (2011) Vitamin supplement use during breast cancer treatment and survival: a prospective cohort study. *Cancer Epidemiol Biomarkers Prev* 20(2): 262–271.

Olus I (2002) Obesity is an independent risk factor for plasma lipid peroxidation and depletion of erythrocyte cytoprotective enzymes in humans. *Int J Obes Relat Metab Disord* 26(9): 1159–1164.

Padayatty SJ, Sun H, Wang Y, Riordan HD, Hewitt SM, Katz A, Welsh RA, Levine M (2004) Vitamin C pharmacokinetics: implications for oral and intravenous use. *Ann Int Med* 140(7): 533–537.

Rohan T, Hiller J, McMichael A (1993) Dietary factors and survival from breast cancer. *Nutr Cancer* 20(2): 167–177.

Salminen E, Bishop M, Poussa T, Drummond R, Salminen S (2004) Dietary attitudes and changes as well as use of supplements and complementary therapies by Australian and Finnish women following the diagnosis of breast cancer. *Eur J Clin Nutr* 58(1): 137–144.

Salminen EK, Lagstrom HK (2000) Does breast cancer change patients’ dietary habits? *Eur J Clin Nutr* 54(11): 844–848.

Sanquist B, Rock CL, Natarajan L, Saquiib N, Newman VA, Patterson RE, Thomson CA, Al-Delaimy WK, Pierce JP (2011) Dietary intake, supplement use, and survival among women diagnosed with early-stage breast cancer. *Nutr Cancer* 63(3): 327–333.

Saxe GA, Rock CL, Wicha MS, Schottenfeld D (1999) Diet and risk for breast cancer recurrence and survival. *Breast Cancer Res Treat* 53(3): 241–253.

Ullah M, Khan H, Zubair H, Shamim U, Hadi S (2011) The antioxidant ascorbic acid mobilizes nuclear copper leading to a prooxidant breakage of cellular DNA: implications for chemotherapeutic action against cancer. *Cancer Chemother Pharmacol* 67(1): 103–110.

Valentzis L, Keshgair M, Woodside J, Leathem A, Titcomb A, Perkins K, Mazurowiska M, Anderson V, Wardell K, Cantwell M (2011) Significant changes in dietary intake and supplement use after breast cancer diagnosis in a UK multicentre study. *Breast Cancer Res Treat* 128(2): 473–482.

Wayne SJ, Lopez ST, Butler LM, Baumgartner KR, Baumgartner RN, Ballard-Barbash R (2004) Changes in dietary intake after diagnosis of breast cancer. *J Amer Diet Assoc* 104(10): 1561–1568.

Willcox JK, Ash SL, Catagnani GL (2004) Antioxidants and Prevention of Chronic Disease. *Crit Rev Food Sci Nutr* 44(4): 275–295.

Willett W, Stampfer MJ (1986) Total energy intake: implications for epidemiologic analyses. *Am J Epidemiol* 124(1): 17–27.

Wolk A, Larsson SC, Johansson J-E, Ekman P (2006) Long-term fatty fish consumption and renal cell carcinoma incidence in women. *JAMA* 296(11): 1371–1376.

Zhang S, Folsom AR, Sellers TA, Kushi LH, Potter JD (1995) Better nutrition and medical research. *Crit Rev Food Sci Nutr* 34(4): 275–295.