

Lipid Peroxidation Serum Levels in Breast Cancer Patients

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Abstract

This prevalence study was conducted to compare lipid peroxide serum levels of breast cancer patients to a control group. Sixteen cases and 12 controls were interviewed for socio-demographic characteristics, medical, cancer and lifestyle history anthropometrical measurements and dietary assessment. A blood sample was collected for quantitative determination of serum lipid peroxides using a colorimetric endpoint assay. The prevalence odds ratio showed a strong and statistically significant inverse association (OR= 6.60; p= 0.02) between breast cancer and lipid peroxide levels. Lipid peroxide levels in serum were different between cases and controls although a level of statistical significance was not attained. The confounding effect of vitamin C, total fat intake, saturated and monounsaturated fatty acid intake was evaluated using a stratified analysis. It revealed that the association between breast cancer and lower lipid peroxidation levels remained strong even after controlling for the variables. Vitamin supplementation and polyunsaturated fatty acid intake appear to modify the levels of serum lipid peroxides. These findings are consistent with previous studies that showed possible inhibitory role of lipid peroxidation products on breast cancer growth.

Introduction

Constituents in foods may be cancer causing, cancer promoting or protective against cancer [1]. Currently, prevention may be our best weapon in the battle against the devastating effects of the disease [2]. Also for the cancer patient, diet can make a crucial difference in the road to recovery [1]. Disparate levels of dietary fat consumption have been a major focus in the attempt to explain international and geographical differences in breast cancer incidence [3]. The reason for recommending low dietary intake of polyunsaturated fats (PUFA) is that they may form free radicals. Free radicals interact with other fats and form other products of oxidation such as hydroperoxides. Hydroperoxides may convert to other radical species that can damage cell membrane [4]. A high concentration of PUFA's in the cell membrane make them very susceptible to peroxidative damage [5, 6] and ultimately the damage can be of such an extent that it may result in cell death due to the interaction of peroxidation products with biomolecules of important and/or regulatory functions [7]. The peroxidation of membrane lipids may result in inactivation of membrane bound enzymes, cross-linking of membrane lipids and proteins, damage to membrane receptors and ultimately cell death [8-10]. It has been reported that when PUFA's undergo lipid peroxidation, they generate large amounts of secondary products of oxidation that restrict cell proliferation and/or increase cell loss which consequentially diminish tumor growth [7, 11]. Dietary fat is thought to be especially important in cancer promotion [1]. High fat consumption has been associated to the high incidence of breast cancer [12]. Nevertheless, the type of fat may be determinant on this action. This study aims to describe the relationship between breast cancer (promotion and progression)

with the levels of lipid peroxide in serum. This is the first study to describe this association in Puerto Rican women.

Methods

Twenty-eight women serve as sample for this study. Sixteen breast cancer cases from the surgery clinics of the Oncology Hospital with a positive diagnosis of breast cancer were selected. The control group was composed of 12 women without breast cancer that worked at the Medical Sciences Campus. This was a prevalence case-control study. All participants were interviewed to collect information about socio-demographic characteristic, medical, cancer and lifestyle history, anthropometrical measurements and dietary assessment. Also, a blood sample (1mL) was collected from each subject for quantitative determination of lipid peroxide in serum using a colorimetric endpoint assay kit (Kamiya Biomedical Co, CA). This specialized kit was used to estimate lipid peroxides levels in serum [13]. All reaction tubes were shielded from light during incubation and then measured. Data was entered into an electronic data bank. Summary tables and frequency analysis were done to compare cases and controls. Non-parametric statistical analyses were performed to evaluate the association between lipid peroxides levels and breast cancer status.

Results

The median age of the study samples was 52.5 years. The minimum age was 26 and the maximums 88. More than half (15/28) of the participants were married. A larger proportion of controls (84%) has a higher education level (high school or more) than breast cancer cases (50%). The age of menarch distribution

had a mode at 14 years. The inferior and superior limits were 8 and 17 years, respectively. The mean age at menopause was 46 for breast cancer cases and 47 for controls. Seventeen participants already had reached menopause status, but the proportion of breast cancer cases (64%) that had reached menopause was larger than that of controls (50%). Twenty three participants that have had children reported the medium age at first birth to be 20 (in both groups) most breast cancer cases (62%) and controls (84%) had their last birth before 36. The most frequent number of pregnancies was 2. Nineteen participants had reached menopause and only 6 were using prescription hormones to treat symptoms (4 controls, 2 cases). Three women in each group (cases and controls) had a family history of breast cancer. More than half (58%) of the participants had the first mammogram before age 50. The most frequent breast cancer type was intraductal cell carcinoma (grade II) with 12 diagnosed cases. Two cases (13%) were diagnosed 'In situ' breast carcinoma; one case had intraductal cell breast carcinoma (grade III). The most advanced breast cancer reported in this study was one case of an inflammatory type (grade III). In the control group 4 controls reported benign breast conditions but with negative breast cancer diagnosis.

The lifestyle factors revealed that more than half (63%) of all participants reported sleeping less than 7 hours a day. The cases were sedentary and only one half of the control group was involved in some type of physical activity. Comparing breast cancer cases to controls, this study showed that proportionally more controls (76%) than cases (50%) had never smoked. At the time of interview, none of the study participants admitted consuming alcohol. Frying was the preferred cooking method for 50% of the controls and 38% of the breast cancer cases. Fifty percent (50%) of controls and 38% of the breast cancer cases ate at fast food consignments, when eating out of their homes. Twelve cases (75%) reported they continue with their usual diets. Most of the participants (61%) reported to eat 3 meals per day. More than half (54%) of all participants reported never snacking between meals. Half (50%) of all participants were using some nutritional supplement at the time of the interview. Half of the participants were obese, 4 were overweight, 3 had adequate weight and 1 was underweight using the classifications for Body Mass Index by the Health and Welfare of Canada 1988 [14]. Half (50%) of the participants had a triceps skinfold (TSF) between 21 to 29 mm. Three fourth (75%) of the participants had a medium frame. Most (93%) of the participants had mid-arm muscle circumference below the 25th percentile. The food frequency revealed the following data:

- Fruits were consumed daily by 31% of the cases and 33% of the controls.
- Juice was consumed daily by 63% of the study participants.
- Vegetables were consumed daily by 87% of the cases and 25% of the controls.
- Red meat was consumed more frequently by controls (92%) than by cases (75%).
- More than half of the participants (53%) never consumed poultry without removing skin.
- Egg consumption was more frequent among the controls (100%) than among the cases (87%).

- Most participants (72%) consumed whole milk
- All controls reported cheese consumption while 19% of the cases reported never eating cheese.
- Fried foods weekly consumptions were reported by 76% of the controls and 8% of the cases.
- The most frequently reported bakery product that was consumed weekly by the study participants was French bread (50% cases and 59% controls).
- All participants had a higher intake of unsaturated fats. Vegetable oil was used daily by 85% (75% corn oil) of the participants and 15% reported a weekly intake.
- Most of the median values for nutrient intake were higher for the control group. The cases had a lower caloric and fat intake but with higher antioxidant vitamin consumption than control group.

The analyses lipid peroxide levels in serum revealed a median value of 98.96 nmol/mL among control group. Using this value as the "normal" level for women participating in this study, we found that 11 cases and 3 controls had lipid peroxide values below median value. The average lipid peroxide level among women without breast disease was 109.00 nmol/mL. The lipid peroxide value for women with benign breast disease was 147.18 nmol/mL. The average lipid peroxide level among women with carcinoma in situ was 107.09 nmol/mL. The values in women with intraductal carcinoma grade III was 35.43 nmol/mL and for inflammatory carcinoma grade III 34.85 nmol/mL.

Discussion

This study is the first study in Puerto Rico to determine lipid peroxide levels in serum of breast cancer cases and controls. The median value of the serum lipid peroxide level of the participants of this study was substantially higher from the expected standard value (98.96 nmol/mL vs 2 nmol/mL). This difference may be due to variation in the environment (i.e. high temperature, high humidity and high PUFA intake). Also possible are variations in laboratory conditions where the K-assay LPO-CC was done to establish this value. In relation to breast cancer many factors may interfere in tumor growth and development. Lipid peroxidation should be included among the negatively associated factors associated with tumor growth. The estimated relative risk shows a statistically significant inverse association with breast cancer risk (OR= 6.6; p=0.02). This study pointed out that woman with serum lipid peroxide levels below 98.96 nmol/mL were 6.6 times more likely to have breast cancer compared to women with higher levels. This crude odds ratio is strong and statistically significant (p=0.024), but the extremely small sample size imposes an inadequate statistical power and lack of precision as shown wide confidence limits (0.97–51.62).

Nevertheless, the story clinically significant result (OR=6.6) should encourage more research to further pursue this finding. The association between serum lipid peroxide levels below 98.96 nmol/mL and breast cancer remained significant with of ranking from 2.88 to 4.97 after individually controlling for the effect of confounding variables such as vitamin C, total fat intake, saturated

and non-saturated fat intake. Vitamin supplementation appeared to be a strong effect modifier in addition to PUFA intake. The serum lipid peroxide level may be an indicator of a cytostatic/cytolytic action of these oxidative products. Apparently, lipid serum levels are inversely related to breast cancer status. Lipid peroxides play an important role on PUFA cytotoxicity, since peroxides decompose into secondary products of peroxidation such as aldehydes. The highly cytotoxic aldehydes may decrease cell proliferation and/or increase the rate of cell loss of the tumor [7,11,15]. The serum concentration of primary and secondary products of oxidation and the consumption of dietary long chain PUFA's requiring further attention. The findings reported herein are consistent with those reported by other laboratories [9,15-17].

Furthermore, there is a similar report showing lower lipid peroxide levels in breast cancer patients compared to controls [18]. Caygill showed this lower peroxidation trend in women with breast cancer [17]. Also one of us (MJG) showed the same trend utilizing human breast carcinoma cells in athymic nude mice, in which fish oil was associated with protection against the promotional effect of fat on breast cancer growth and development [19]. Gerber mentioned that the serum concentration of secondary products of lipid peroxidation, Malondialdehyde (MDA) decrease with the severity of disease, MDA also decrease with tumor size and vitamin E: total cholesterol concentration [20]. The mean lipid peroxide levels in the control group were lower than those with benign breast disease moreover, the mean serum lipid peroxides levels were very low, cases with confirmed breast cancer diagnosis. In 1990 Horrobin reported that lipid peroxides are important factors in the PUFA-induced cytotoxicity [21]. Nevertheless we must be cautious when interpreting these results since the sample size is small. Small studies with large OR and strong statistical significance should motivate further research. In spite of the limitations in the study, we found lower serum lipid peroxide levels in breast cancer patients, a finding that is supported by previous research [9,15,20,22]. Two important questions arise from these results.

- Are healthy women with low lipid peroxide serum levels at increased risk for breast cancer?
- Could an increase in the lipid peroxide serum levels induced by dietary supplementation decrease the risk for breast cancer in healthy women?
- Could women with breast cancer benefit from increasing lipid peroxide levels in their serum?
- These follow-up questions may provide the basis for further relevant research in the area of lipid peroxidation and tumor growth and advance our quest for effective non toxic cancer therapy.

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