Selenium supplementation, baseline plasma selenium status and incidence of prostate cancer: an analysis of the complete treatment period of the Nutritional Prevention of Cancer Trial

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OBJECTIVE

To present the results (to January 1996, the end of blinded treatment) of the Nutritional Prevention of Cancer (NPC) Trial, a randomized trial of selenium (200 μg daily) designed to test the hypothesis that selenium supplementation (SS) could reduce the risk of recurrent nonmelanoma skin cancer among 1312 residents of the Eastern USA.

MATERIALS AND METHODS

Original secondary analyses of the NPC to 1993 showed striking inverse associations between SS and prostate cancer incidence. A subsequent report revealed that this effect was accentuated among men with the lowest baseline plasma selenium concentrations. The effects of treatment overall and within subgroups of baseline prostate-specific antigen (PSA) and plasma selenium concentrations were examined using incidence rate ratios and Cox proportional hazards models.

RESULTS

SS continued to significantly reduce the overall incidence (relative risk and 95% confidence interval) of prostate cancer (0.51, 0.29–0.87). The protective effect of SS appeared to be confined to those with a baseline PSA level of ≤4 ng/mL (0.35, 0.13–0.87), although the interaction of baseline PSA and treatment was not statistically significant. Participants with baseline plasma selenium concentrations only in the lowest two tertiles (< 123.2 ng/mL) had significant reductions in prostate cancer incidence. A significant interaction between baseline plasma selenium and treatment was detected.

CONCLUSION

To the end of the blinded treatment the NPC trial continued to show a significant protective effect of SS on the overall incidence of prostate cancer, although the effect was restricted to those with lower baseline PSA and plasma selenium concentrations.

KEYWORDS

selenium, prostate cancer, chemoprevention, incidence, risk, PSA

INTRODUCTION

The Nutritional Prevention of Cancer (NPC) trial [1] is the only randomized clinical trial to date to test the effect of selenium supplementation (SS) on cancer in a Western population. Although SS did not reduce the risk of the primary endpoint, recurrence of nonmelanoma skin cancer (NMSC), treatment significantly reduced the secondary endpoints all-cause mortality, total cancer mortality, total cancer incidence (excluding NMSC), lung, prostate and colorectal cancer incidence, in men and women with histories of NMSC. An additional paper by Clark *et al.* [2] showed that the effect of SS on prostate cancer was accentuated among men in the lowest tertile of baseline plasma selenium level.

The results of this landmark trial have not only contributed substantially to the epidemiological evidence supporting selenium as a chemopreventive agent, but have also received considerable public attention. The current report adds considerably to the statistical precision of the trial results by extending the previously reported results through the entire blinded phase of the trial. The primary objective of this report is to evaluate the effect of SS through the complete treatment period on the overall incidence of prostate cancer, and by baseline PSA and plasma selenium concentrations.

MATERIALS AND METHODS

The protocol for the NPC trial was described in the original report [1]. Briefly, the study was a randomized, double-blind, placebo-controlled trial conducted in 1312 participants recruited between 1983 and 1991, from seven dermatology clinics in low-selenium areas of the Eastern USA. Randomization was blocked by time and stratified by clinic. Subjects were eligible if they had confirmed histories of NMSC within the year before randomization, had an estimated 5-year life-expectancy and had had no reported internal cancer within the previous 5 years. Four interviewed patients were unsuccessfully randomized and all but nine participants completed follow-up for the 1983 to January 1996 period. The intervention agent was 200 μg/day of selenium in 0.5-g high-selenium yeast (Nutrition 21, La Jolla, CA to 1995, and Cypress Systems, Fresno, CA thereafter) or a yeast placebo.

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Characteristic	SS	Placebo
Participants randomized, n	457	470
Mean (SD):		
age, years	64.9 (8.8)	63.7 (9.4)*
BMI, kg/m^2	26.0 (3.6)	25.9 (3.7)
Smoking status, %		
Never	25	21
Former	47	46
Current	28	33
Plasma selenium, ng/mL		
Mean (SD)	115.1 (22.1)	115.1 (22.0)
33rd centile	106.8	106.0
50th centile	113.6	114.0
66th centile	123.2	122.4
PSA, %†		
Mean (SD), ng/mL	2.0 (3.4)	1.9 (3.3)
< 4	90.3	89.5
4–7	5.5	6.6
7.1–10	1.4	2.7
> 10	2.8	1.2

TABLE 1
The baseline characteristics of the men participating in the NPC, by treatment group

**Two-sample t-test, P < 0.05; †PSA test within 6 months of randomization (694 men).*

Participants visited their respective clinics biannually to provide blood samples and report new illnesses and medications. Plasma selenium concentration was determined by automated electrothermal atomic absorption spectrophotometry (Perkin Elmer 3030, Perkin-Elmer Corp., Norwalk, CT). Retrospective PSA concentrations were obtained on the subsample (694) of men with PSA values from blood samples taken within 6 months of randomization using a microparticle enzyme immunoassay (Abbott Diagnostics, IL, USA). All medical records (pathology reports, medical notes) documenting any cancer screening procedures including PSA tests, DRE, TRUS and prostate biopsies and surgery, including TURP and prostatectomy, were obtained throughout the course of the trial. Incident prostate cancer cases were recently re-reviewed and staged by a urological oncologist according to the TNM system [3].

Statistical analyses were based on data from male participants with selenium concentration values from initial blood samples within 4 days of the randomization date (934 men), and with no history of prostate cancer before randomization (927 men). Excluded participants were evenly distributed between treatment groups (25 in SS and 28 in the placebo groups). Continuous baseline variables were analysed using *t*-tests, while categorical baseline variables were assessed using chi-square tests. Results obtained from the total male cohort of 980 showed no significant differences compared with the subsample of 927.

Person-years (PY) of follow-up were calculated among the subsample of 980 subjects. For subjects with no prostate cancer, PYs were computed using the date of randomization as the start date, and the earlier of 1 February 1996 or the date of death as the closing date. PYs of follow-up for prostate cancer cases were calculated to the date of diagnosis documented in pathology, surgery or medical reports.

Prostate cancer incidence data were statistically analysed by calculating relative risks (RRs), the ratio of the incidence density between treatment groups, and corresponding 95% CI, with *P* values derived from log-rank tests. Supporting analyses included the calculation of hazard ratios (HRs) and 95% CIs, using the Cox proportional hazard model, which allowed adjustment for age (continuous variable) and smoking status (never, former, current) at baseline as covariates. Among the 927 participants with baseline blood samples within 4 days of randomization, the effect of SS on prostate cancer incidence was assessed within subgroups determined by baseline characteristics. Modification of effects by PSA concentration (dichotomized at 4 ng/mL) at randomization was tested using the Mantel-Haenszel test for heterogeneity in the unadjusted models. The statistical significance of the interaction between baseline PSA concentration and treatment group, adjusted for age and smoking status at baseline, was tested in a Cox proportional hazards model that included this interaction and the corresponding main effect terms, in addition to the variables for the adjustment.

The statistical association between prostate cancer incidence and concentrations of baseline plasma selenium was also determined. Based on the distribution among the 927 participants with valid values, baseline plasma selenium concentrations were divided by tertiles (≤ 106.4 ng/mL, 106.8–123.2 ng/mL and >123.2 ng/mL). The effect of SS on prostate cancer incidence was assessed within these subgroups of baseline plasma selenium, using the same techniques as for the analyses within subgroups of baseline PSA concentrations.

No statistically significant differences in incidence data from the total and subsample of NPC participants were detected. A program (STATA 7.0, College Station, TX) was used for all statistical tests.

RESULTS

Selected baseline characteristics of male participants by treatment group are shown in Table 1. The two groups had a slight yet statistically significant difference in age; no other characteristics, including the percentage of men with elevated PSA values, varied appreciably or significantly across treatment groups.

At unblinding, with follow-up to January 1996, the trial had a total of 7263 PYs, with a mean follow-up of 7.6 and 7.3 years ($P < 0.05$) in the SS and placebo groups, respectively. Sixty-four cases of prostate cancer were diagnosed, 22 among the SS and 42 among the placebo groups.

There were no significant differences in clinical stage or incidence of advanced cancer between the groups. The incidence of localized (T1 + T2), T3 and metastatic cancers at presentation was 77%, none and 23%, respectively, in the SS, and 76%, 7% and 17% in the placebo group.

At the end of the trial, the SS participants had a significantly lower incidence of prostate cancer than those assigned to placebo (Table 2). The HR adjusted for age and

TABLE 2 Prostate cancer incidence by treatment group, follow-up period, baseline PSA and tertile of baseline plasma selenium

Group [n]	Cases, SS/placebo	Incidence† SS/placebo	Unadjusted RR (95% CI) [P]	Adjusted HR (95% CI) [P]
Follow-up				
1983–1993 [974]	13/35	–	0.37 (0.18–0.71) [0.002]*	0.35 (0.16–0.65) [0.001]†
1983–1996 [927]	22/42		0.51 (0.29–0.88) [0.009]*	0.48 (0.28–0.80) [0.005]†
Baseline PSA, ng/mL				
≤ 4 [624]	7/20	0.30/0.86	0.35 (0.13–0.87) [0.01]*	0.33 (0.14–0.79) [0.01]†
> 4 [70]	11/13	5.64/6.40	0.88 (0.36–2.13) [0.86, 0.13]†¶	0.95 (0.42–2.14) [0.90, 0.09]†§
Plasma selenium				
≤ 106.4 [317]	2/15	0.17/1.25	0.14 (0.02–0.59) [0.002]*	0.14 (0.03–0.61) [0.009]†
106.8–123.2 [305]	7/16	0.57/1.48	0.39 (0.14–0.99) [0.03]*	0.33 (0.13–0.82) [0.02]†
> 123.2 [305]	13/11	1.17/0.97	1.20 (0.50–2.97) [0.66,0.02]*¶	1.14 (0.51–2.59) [0.75,0.01]†§

**RR and †HR derived from incidence rate ratios,* P *from log-rank tests and Cox proportional hazard model adjusted for age (continuous) and smoking (never, former, current) at randomization; ‡Annual cumulative incidence per 100 PYs;* P *from log-rank test and ¶Mantel-Haenszel test for heterogeneity; §P for treatment group–characteristic interaction, for the (treatment group × factor) cross-product term in a Cox proportional hazards model.*

smoking status at randomization provided a similar estimate. Table 2 also shows the treatment effect for prostate cancer from the 1983–93 analysis published in the original report [2]. The overall effect of selenium is diminished slightly by the inclusion of 25 months of additional follow-up.

The analysis of SS and prostate cancer incidence within strata of baseline PSA is also shown in Table 2. PSA values within 6 months of randomization were retrospectively available for 694/927 (75%) of randomized men. Among men (624) with PSA values of ≤4 ng/mL, the unadjusted estimate showed a significant 65% reduction in prostate cancer incidence with SS (Table 2). Among 70 men with elevated PSA values no reduction in incidence was evident. The overall RR among the 694 men with PSA data available (0.54, 95% CI 0.29–1.00) is representative of the entire NPC subsample, which suggests that the experience of these men can be generalized to the entire NPC male sample.

Dividing baseline plasma selenium into tertiles based on the distribution among all men yielded thresholds of ≤106.4, 106.8–123.2 and >123.2 ng/mL (Table 2). SS led to significant reductions in prostate cancer incidence among those in both the lowest and middle tertiles. Thus, the effect of SS on prostate cancer incidence was significantly modified by baseline plasma selenium concentration considered in terms of tertiles ($P = 0.02$).

Equal numbers of randomized men had an abnormal PSA value at baseline (9.7% placebo vs 10.5% selenium) and an additional 12% in each group had an abnormal PSA value at some point throughout the trial. However, the follow-up, as per the current clinical standard for a man with an abnormal PSA level, differed significantly between treatment groups; 35% of men with an abnormal PSA in the placebo group underwent biopsy at some point throughout the trial, compared with only 14% in the selenium group ($P < 0.05$; Table 3). This observed difference in biopsy rates could not be accounted for by PSA concentration, age at which the abnormal PSA was detected, nor alternative diagnostic procedures including TURP or TRUS. This discrepancy suggests a potential bias against the detection of prostate cancer in the SS group. This difference was greatest among men with the lowest baseline selenium concentrations; the subgroup that accounted for the overall protective effect of SS (Table 3). To investigate the sensitivity of the results to this inconsistency in biopsy rates, we augmented existing prostate cancer case numbers by assuming that 30% [4–6] of men retrospectively identified with an abnormal PSA level, who were not biopsied external to the trial, would have tested positively for prostate cancer if they had undergone biopsy. For example, 30% of the 22 SS men with baseline plasma selenium concentrations of <106.4 ng/mL who were not biopsied after a PSA level of >4 ng/mL, or seven men, might have been diagnosed while 30% of the 23 unbiopsied, placebo group in the same baseline plasma selenium group, or seven men, might have been found to have prostate cancer. This projected analysis attenuated the incidence of prostate cancer overall, with the protective effect of SS remaining statistically significant *only* among men in the lowest tertile of baseline plasma selenium concentration (nine cases in the SS vs 22 in the placebo group, $P < 0.05$).

DISCUSSION

This report of the NPC trial extends the original secondary analyses and describes the effect of SS on prostate cancer incidence through to the end of randomized, blinded treatment (2 January 1996). The inverse association of treatment with prostate cancer incidence was slightly attenuated with extended follow-up, although SS continued to reduce the incidence of prostate cancer overall after a mean follow-up of 7.5 years. This result is consistent with many epidemiological [7–13] and *in vitro* studies [14–17] that indicate inverse associations between selenium and prostate cancer.

The overall protective effect of SS was most prominent in men with baseline PSA concentrations of ≤4 ng/mL, corroborating the proposal that men with PSA values of 1–4 ng/mL would be most appropriate for chemoprevention studies [18].

	Baseline plasma selenium, ng/mL			
Group	≤ 106.4	106.8–123.2	> 123.2	Total
SS	138	149	148	435
Placebo	159	144	145	448
PSA > 4 ng/mL, n (%)				
SS	24 (17)	35 (23)	39 (26)	98 (23)
Placebo	33 (21)	31 (22)	28 (19)	92 (21)
Biopsy after PSA > 4 ng/mL, n (%)				
No				
SS	22 (92)	31 (89)	31 (79)	84 (86)
Placebo	23 (70)	19 (61)	18 (64)	60 (65)
Yes				
SS	2 (8)	4 (11)	8 (21)	14 (14)
Placebo	10 (30)*	12 (39)*	10 (36)	32 (35)*

TABLE 3
Biopsy rate after abnormal PSA by tertile of baseline plasma selenium and treatment group

*P < 0.05.

Clark *et al.* [2] reported that, to 12 December 1993, SS had the greatest effect (RR 0.08, $P = 0.002$) on prostate cancer among men in the lowest category of baseline plasma selenium (< 106.4 ng/mL) and this analysis supports that observation. The protective effect of treatment to January 1996 was confined to men with plasma selenium concentrations in the first and second tertiles of the baseline distribution. When corrected for differences in biopsy rates, the protective effect remained only in the lowest tertile. Thus, extended follow-up appears to more firmly mark a threshold on the benefit gained by SS, such that the only men to benefit from supplementation were those with low baseline plasma selenium concentrations. Modification of the association between treatment and prostate cancer incidence by baseline status of a supplemented nutrient was also reported in the Physician's Health Study [19]; beta-carotene supplementation in that study reduced prostate cancer incidence only among those individuals with low baseline plasma beta-carotene concentrations.

There are several limitations to the present trial. First, prostate cancer was not a primary endpoint of the NPC trial [20] and the trial spanned the period of introduction of PSA testing. It was thus prudent to determine whether treatment and control subjects had an equal opportunity to be diagnosed with prostate cancer throughout the trial. They did not, although our attempts to take possible diagnostic differentials into account do not appear to eliminate the protective effect of SS. Second, confounding may potentially explain the association of baseline plasma selenium and prostate cancer incidence. Although randomization should minimize the likelihood of confounding of treatment effects by unmeasured risk factors, genetic predisposition to prostate cancer, physical activity, dietary supplement use and the intake of other nutrients remain possible confounders. Future investigations of this cohort should consider the effect of other antioxidants, including the tocopherols and carotenoids, and the extent to which they may modulate the effect of SS. Finally, there were few cases of prostate cancer (64); chance remains a possible source of the observations we report.

In conclusion, this report continues to provide support for the efficacy of SS in reducing the incidence of prostate cancer. However, this conclusion is tempered by an unexplained difference in biopsy rates between treatment groups among men with an elevated PSA level. With a reasonable correction for the difference, the protective effect of SS is restricted to men in the lowest tertile of baseline selenium concentration.

The ongoing Southwest Oncology Group selenium trial for men with high grade prostatic intraepithelial neoplasia, as well as related chemoprevention trials, will add to these results of the NPC trial [21]. The recently initiated Selenium and Vitamin E Cancer Prevention Trial (SELECT) [22], targeting men with normal PSA concentrations and DREs that are not suspicious, may also provide valuable information. Unlike the NPC trial, prostate cancer detection is a primary study endpoint of the SELECT, such that men will be subject to standardized cancer detection methods. Furthermore, SELECT is powered to stratify and assess any modification of the effect of the association between SS and prostate cancer incidence by baseline plasma selenium.

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Abbreviations: **NPC**, Nutritional Prevention of Cancer (trial); **SS**, selenium supplementation; **NMSC**, nonmelanoma skin cancer; **PY**, person-years; **RR**, relative risk; **HR**, hazard ratio; **SELECT**, Selenium and Vitamin E Cancer Prevention Trial.