An estimation of selenium requirements for New Zealanders[1–3]

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ABSTRACT

Background: Current US dietary recommendations for selenium are based on maximization of plasma glutathione peroxidase (GSHPx) activity according to data from one study of Chinese men.

Objective: The effect of various amounts of supplemental selenium on GSHPx activities in blood of New Zealand adults was investigated to calculate a selenium requirement for New Zealanders. The effect on plasma selenoprotein P and thyroid hormones was also investigated.

Design: Fifty-two adults with low blood selenium concentrations ingested a placebo or 10, 20, 30, or 40 μg Se as L-selenomethionine daily for 20 wk.

Results: Plasma and whole-blood GSHPx activities increased in all supplemented groups but reached a plateau only in the group receiving 40 μg Se, as determined by statistical analysis. Increases in selenoprotein P were greater than those for selenium and GSHPx at all supplement intakes. Thyroxine concentrations decreased in supplemented groups but the decrease was significantly different from that in the control group only for the 10-μg group and for all supplemented groups combined.

Conclusions: An upper estimated requirement of 90 μg Se/d was calculated as the intake necessary for maximization of plasma GSHPx activity, as used in the derivation of the US recommended daily allowance. Our lower estimated requirement of 39 μg Se/d was the intake necessary to reach two-thirds of maximal GSHPx activity, as was used in calculating the World Health Organization normative requirement. The lower estimate is a realistic goal for New Zealand but the upper estimate could be achieved only with regular inclusion of high-selenium foods. *Am J Clin Nutr* 1999;70:896–903.

KEY WORDS Selenium, glutathione peroxidase, selenoprotein P, thyroxine, supplementation, requirements, New Zealand residents, RDA, recommended dietary allowance

INTRODUCTION

Forty micrograms dietary and supplemental selenium was established by Yang et al (1) in 1987 as the physiologic daily selenium requirement for maximal glutathione peroxidase (GSHPx) activity in plasma of Chinese men. By adjusting for body weight and individual variation, this value was used to calculate the 1989 US recommended dietary allowance (RDA) for selenium of 70 and 55 μg/d for adult men and women, respectively (2, 3).

This recommendation is easily achieved by eating a mixed diet in the United States but residents of countries such as New Zealand have difficulty in reaching this intake (4). New Zealanders typically consume <30 μg Se/d from food and can have intakes as low as 10 μg/d if no eggs or poultry are consumed or as high as 100 μg/d if selenium-rich foods such as kidney, liver, Brazil nuts, or fish are included (5). The low selenium intake is reflected in lower blood selenium concentrations and urinary outputs than those reported in most Western countries (4, 6). GSHPx activities in erythrocytes and plasma are also low and may be enhanced by selenium supplementation, which suggests that New Zealanders may have suboptimal GSHPx activities (7). However, there has been little indication of direct effects on health, except for one case of a selenium-responsive muscular syndrome reported in a patient receiving total parenteral nutrition (8). Thus, it appears that selenium intakes lower than the US RDA have had no adverse consequences for the health of New Zealanders, who may have adapted to their low selenium intake (9).

In his critique of the US RDA for selenium, Combs (10) questioned the interpretation of results of the Chinese study (1) in identifying 40 μg Se/d as the intake associated with optimal GSHPx response, and expressed concern that nonreplicated results from a single study were used as the basis for the RDA. Combs also examined the assumptions on which the RDA was derived, in particular, that maximal GSHPx activity was necessary for optimal health. More recently, the WHO/FAO/IAEA Expert Consultation (11) used the selenium intake necessary to achieve two-thirds of maximal GSHPx activity as the criterion for their normative requirements for selenium. Using the Chinese data, the WHO/FAO/IAEA group calculated a normative requirement of 40 μg Se/d (11), considerably less than the US RDA. Combs also discussed the choice of plasma GSHPx activity as the

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key measure of selenium status, pointing out that other measures such as erythrocyte GSHPx activity, selenium concentrations, or activities of other selenoproteins such as selenoprotein P were also potentially useful (10).

Because of the uncertainty about selenium requirements, we investigated the effects of various amounts of supplemental selenium on selenium concentrations and GSHPx activities in blood of New Zealand adults, and used these results to calculate a selenium requirement for New Zealanders. The effects of supplementation on selenoprotein P and thyroid hormone concentrations were also investigated.

SUBJECTS AND METHODS

Fifty-two New Zealand residents (17 men and 35 women) aged 19–59 y were recruited from a group of 122 healthy volunteers in 1995 in Otago, New Zealand. The mean (±SD) and range of whole-blood selenium concentrations for the 122 adults were 1.24 ± 0.25 and 0.66–2.11 μmol/L, respectively. All subjects with whole-blood selenium concentrations <1.26 μmol/L were invited to participate in the study.

Weight and height were measured at the beginning of the study and all subjects completed a brief questionnaire on dietary habits, nutrient supplementation, and other factors that might influence selenium status. Subjects gave their informed consent and the protocol was approved by the Southern Regional Health Authority Ethics Committee, Otago, New Zealand.

Subjects were randomly assigned to 1 of 5 groups (**Table 1**) and, in a double-blind intervention, consumed tablets containing a placebo (<1 μg Se) or 10, 20, 30, or 40 μg Se as L-selenomethionine (Westar Nutrition Inc, Costa Mesa, CA) daily for 20 wk. Compliance was assessed by the number of tablets returned at the end of each month.

Experimental design

Subjects were encouraged to maintain their usual daily routines and dietary habits and were requested to not eat selenium-rich foods such as fish, liver, kidney, or Brazil nuts. Foods consumed on 3 nonconsecutive days were weighed, recorded, and collected by most subjects (n = 43). Analysis of the duplicate diets and 3-d diet records gave mean (±SD) daily selenium intakes of 28 ± 15 and 29 ± 13 μg, respectively (12). Blood was drawn into evacuated tubes containing EDTA every 2 wk for 2 mo, then every 4 wk for a further 12 wk. Aliquots of whole blood and plasma were stored at −80°C until analyzed.

Analytic methods

Whole-blood selenium concentrations were determined by flow injection hydride generation atomic absorption spectrometry with acid digestion of the sample before analysis (13). Plasma samples were digested by using nitric and perchloric acids (14). Samples were digested in duplicate and each was then analyzed in triplicate by using a Perkin-Elmer atomic absorption spectrometer (model 3100; Norwalk, CT) in combination with an MHS-FIAS-200 flow injection hydride generation system and an AS-90 autosampler (Perkin-Elmer) (13).

Pooled aliquots of the corresponding sample matrix were used for internal quality control in all selenium analyses. Interassay analysis of these controls gave a mean (±SD) of 1.00 ± 0.06 μmol Se/L with a CV of 6.1% for 28 assays of whole blood and a mean (±SD) of 0.69 ± 0.05 μmol Se/L with a CV of 7.9% for 19 assays of plasma. The intraassay CVs were 6.1% (n = 28) and 4.7% (n = 5) for whole blood and plasma, respectively.

Analysis of Seronorm Reference Whole Blood (batch no. 2030566, product no. 210611; Nycomed Pharma Diagnostics, Olso) with a certified selenium value of 1.18 μmol/L gave a mean of 1.18 ± 0.07 μmol/L with a CV of 6.3% (n = 28). Analysis of Seronorm Reference Plasma (lot 605113; Nycomed Pharma Diagnostics) with a certified value of 0.987 μmol Se/L gave a value of 0.988 ± 0.048 μmol Se/L with a CV of 4.8% (n = 5).

GSHPx activity was assayed in whole blood and plasma by using an automated modification (7) of the coupled assay of Paglia and Valentine (15). Whole-blood GSHPx activity was used to represent erythrocyte activity because previous studies that used our assay showed that because plasma GSHPx activity accounts for only 5% of the total activity in whole blood (16), whole-blood GSHPx activity mirrors erythrocyte activity. A stock sample of whole blood or plasma assayed with each series of samples gave a mean (±SD) of 328 ± 22 nkat/g hemoglobin with a CV of 6.9% for 40 assays of whole blood and a mean (±SD) of 57.8 ± 3.4 nkat/g protein with a CV of 5.4% for 20 assays of plasma. Plasma protein was measured by the standard Biuret method (17).

TABLE 1
Baseline selenium concentrations, glutathione peroxidase (GSHPx) activities, and selenoprotein P concentrations in blood of 52 New Zealand residents[1]

	Selenium dose					
	0 μg/d (n = 10)	10 μg/d (n = 10)	20 μg/d (n = 11)	30 μg/d (n = 10)	40 μg/d (n = 11)	ERMS
Age (y)	41 ± 10[2]	37 ± 9	45 ± 13	38 ± 13	30 ± 10[3]	—
Weight (kg)[4]	78.5	66.4	76.6	79.4	67.8	13.5
Plasma selenium (μmol/L)	0.783	0.806	0.846	0.869	0.809	0.025
Whole-blood selenium (μmol/L)	0.939	0.912	0.951	1.017	0.969	0.177
Plasma GSHPx (nkat/g protein)	41.5	45.2	52.4	45.1	46.1	10.9
Whole-blood GSHPx (nkat/g hemoglobin)	330	331	339	335	352	16
Selenoprotein P (U/L)	610	580	610	630	530	150

[1]ERMS, error root mean square, an unbiased estimate of SD.
[2]$\bar{x}$ ± SD.
[3]Significantly different from control group, $P < 0.05$.
[4]Adjusted for sex.

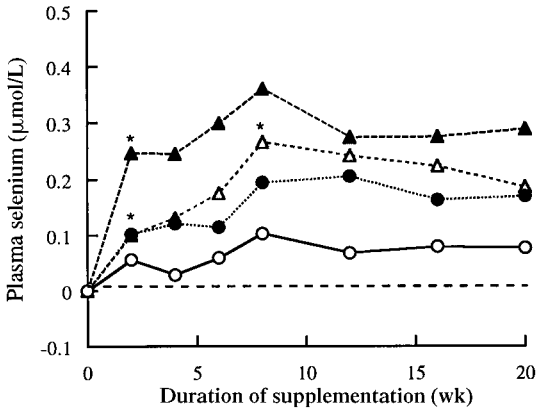

FIGURE 1. Plasma selenium concentrations during supplementation with 10 (○), 20 (●), 30 (△), and 40 (▲) μg Se as selenomethionine daily for 20 wk. Values are differences between each supplemented group and the control group for each week, adjusted for sex and baseline measures. Differences for all treatment groups, except for the 10-μg group, were significant after 2 wk. *Significant increase from the previous week relative to the control group, $P < 0.05$. The SDs of plasma selenium concentrations were between 0.0076 and 0.139 μmol/L.

Selenoprotein P was assayed by a radioimmunoassay using ^{75}Se-labeled human selenoprotein P (18). Total thyroxine (T_4) and thyroglobulin were analyzed by using ^{125}I radioimmunoassays (Count-a-Coat and Double Antibody, respectively; Diagnostic Products Corporation, Los Angeles).

Statistical analysis

The SAS procedure PROC GLM (SAS Institute Inc, Cary, NC) was used to examine differences between the treatments at each time point. These were adjusted for the initial value and for sex by including a factor in the regression analysis. Dunnett's test was used to control for the type I error rate when each group was compared with the control group. We acknowledge, however, that the analysis includes a large number of comparisons. The second part of the analysis examined the relative change between groups by taking into account all the earlier measures rather than just the baseline measure to determine whether a plateau in measurements had occurred. Measures made at 2, 4, 8, 12, and 20 wk were used as dependent variables. Dunnett's test was used to adjust for the significance level for the multiple comparisons among the groups.

The significance of the correlations between selenium, GSHPx activity, and selenoprotein P in whole blood and plasma was adjusted for multiple observations from a single individual by using the appropriate SEs (19). The significance level for all analyses was set at $P < 0.05$.

RESULTS

Baseline selenium concentrations and GSHPx activities in whole blood and plasma and baseline plasma selenoprotein P concentrations are shown in Table 1. Random assignment of the subjects generated groups whose mean baseline values for selenium, GSHPx activity, and selenoprotein P were not significantly different (Dunnett's test). The results are presented as differences between each group and the control group, adjusted for sex and baseline measures.

Selenium concentrations

Mean plasma selenium concentrations increased by 4%, 14%, 23%, 26%, and 41% of baseline in the control, 10-, 20-, 30-, and 40-μg groups, respectively, after 20 wk. Differences between each supplemented group and the control group are shown in **Figure 1**. The differences for all treatment groups, except the 10-μg group, were significant from week 2 onward ($P < 0.05$). The second part of the analysis showed increases in plasma selenium relative to the control group when taking into account all the earlier measures. In this analysis, increases in plasma selenium from baseline to 2 wk were significantly different from the control group in the 30- and 40-μg groups. Plasma selenium continued to show relative increases from the previous week in the 3 highest supplementation groups on the order of 1 SD until week 8, although only the increase from 6 to 8 wk in the 30-μg group was significant. The relative changes between weeks 12 and 20 were <0.45 SD in all groups and were not significant.

Whole-blood selenium concentrations increased by 4%, 9%, 9%, 7%, and 13% in the 5 groups, respectively, although the increase reached 11% in the 30-μg group and 15% in the 40-μg group earlier in the supplementation period than for lower intakes. Differences between the 40-μg group and the control group (**Figure 2**) were significant from week 2 to week 16, and those between the 30-μg group and the control group at weeks 2, 4, 8, and 16. Whole-blood selenium increased significantly relative to the control group in these 2 groups in the first 2 wk. Further increases for both groups between 2 and 4 wk, although equivalent to 1 SD, were not significant. Relative increases of the same magnitude as the SD were also observed between 8 and 12 wk, but again, were not significant. Smaller differences of <0.4 SD were observed between 12 and 20 wk.

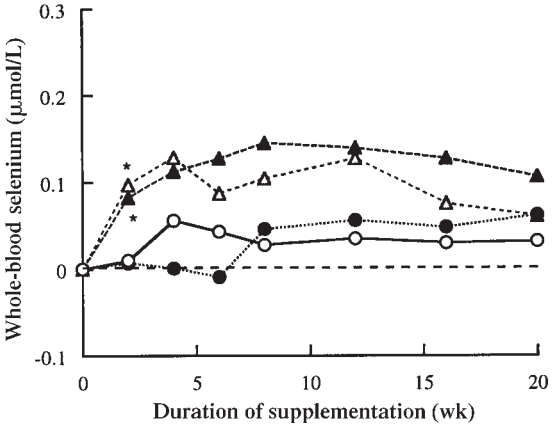

FIGURE 2. Whole-blood selenium concentrations during supplementation with 10 (○), 20 (●), 30 (△), and 40 (▲) μg Se as selenomethionine daily for 20 wk. Values are differences between each supplemented group and the control group at each week, adjusted for sex and baseline measures. Differences between the 40-μg group and the control group were significant from 2 wk to 16 wk, $P < 0.05$. Differences between the 30-μg group and the control group were significant at 2, 4, 8, and 16 wk, $P < 0.05$. *Significant increase from the previous week relative to the control group, $P < 0.05$. The SDs of whole-blood selenium concentrations were between 0.0072 and 0.104 μmol/L.

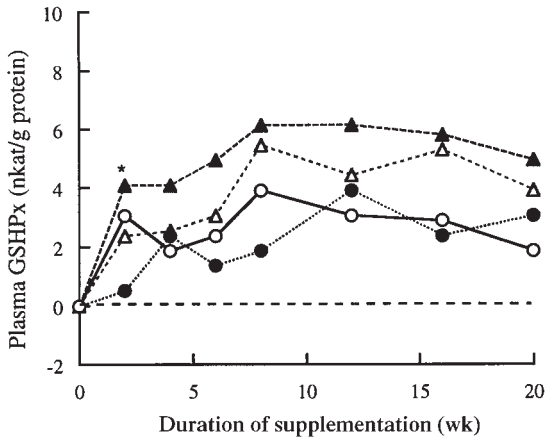

FIGURE 3. Plasma glutathione peroxidase (GSHPx) activities during supplementation with 10 (○), 20 (●), 30 (△), and 40 (▲) μg Se as selenomethionine daily for 20 wk. Values are differences between each supplemented group and the control group at each week, adjusted for sex and baseline measures. Differences between the 40-μg group and the control group were significant at 2, 6, 8, 12, and 14 wk. Differences between the 30-μg group and the control group were significant at 8 and 16 wk. *Significant increase from the previous week relative to the control group, $P < 0.05$. The SDs of plasma GSHPx activities were between 3.4 and 4.9 nkat/g protein.

Glutathione peroxidase activities

Plasma GSHPx activities increased by 13%, 17%, 19%, 21%, and 30% in the 5 groups, respectively. The differences between the treatment groups and the control group are shown in **Figure 3**. Differences for the 40-μg group were significant for all weeks except 2 and 20, and those for the 30-μg group were significant at 8 and 16 wk. Differences from the control group for the other groups were not significant. In the second part of the analysis, there was a significant increase in the 40-μg group relative to the control group between baseline and week 2. Later relative changes were not significant, indicating a plateau in activity. None of the relative changes in the 30-μg group were significant, although the relative change between 6 and 8 wk was almost 0.7 SD. The relative increases for the other groups were not significant.

Whole-blood GSHPx activities reflected more slowly the trends seen in plasma activities and increased by 12%, 10%, 18%, 23%, and 26% in the 5 groups, respectively. The differences between the treatment groups and the control group are illustrated in **Figure 4**. Differences for the 40-μg group were significant for each time at which measurements were made. The differences between the other groups and the control group were not significant. The GSHPx activity of the 40-μg group increased significantly relative to the control group between 0 and 2 wk, whereas relative increases between 2 and 4 wk and 4 and 6 wk were <0.5 SD and not significant. Although the relative increase between 12 and 20 wk was small and not significant, it was of the order of 1 SD and preceded a significant increase between 8 and 12 wk. The increase between 12 and 20 wk was small and not significant, indicating that a plateau had been reached. None of the increases in the 30-μg group were significant; however, those for 6 wk onward were all >0.7 SD. None of the relative increases in the other groups were significant.

With use of this statistical analysis, the 40-μg group was the only group in which GSHPx activity can be said to have reached a plateau for both plasma and whole blood. These were the only measures for which there was a relative increase during the early weeks of supplementation followed by lack of relative increases in subsequent weeks as well as significant differences from the control group after the initial supplementation period. However, in the 30-μg group, plasma GSHPx activity was significantly different from that of the control group and close to that of the 40-μg group at 8 and 16 wk. In addition, the relative change from 6 to 8 wk, although not significant, was 0.7 SD whereas subsequent changes were small, suggesting that this group was also approaching a plateau.

Selenoprotein P

Selenoprotein P concentrations rose rapidly during the first 4 wk of supplementation, increasing by 24%, 45%, 31%, 52%, and 88% in the 5 groups, respectively. These changes were considerably greater than those for selenium concentrations and GSHPx activities. The drop in selenoprotein P concentration in the 10-, 20-, and 30-μg groups at week 8 cannot be explained. The differences between each of the treatment groups and the control group are shown in **Figure 5**. Differences for the 40-μg group were significant for each time period. The selenoprotein P concentration of the 30-μg group was significantly different from that of the control group at 4 wk and again at 12 wk, and the 20-μg group was different from the control group at 4 wk. Relative increases for the 40-μg group were significant between baseline and 2 wk and between 2 and 4 wk, but not after that, indicating relatively constant differences from 4 wk on. The relative differences between the control group and the 30- and 20-μg groups were significant between 2 and 4 wk but not thereafter. Although there was considerable noise in these data, they do indicate that a plateau of selenoprotein P concentrations occurred at lower supplemental selenium intakes (≈30 μg/d) than for GSHPx activity.

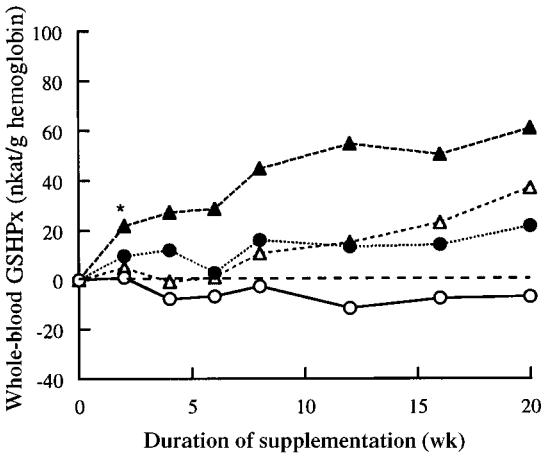

FIGURE 4. Whole-blood glutathione peroxidase (GSHPx) activities during supplementation with 10 (○), 20 (●), 30 (△), and 40 (▲) μg Se as selenomethionine daily for 20 wk. Values are differences between each supplemented group and the control group at each week, adjusted for sex and baseline measures. Differences between the 40-μg group and the control group were significant after 2 wk. None of the other differences were significant. *Significant increase from the previous week relative to the control group, $P < 0.05$. The SDs of whole-blood GSHPx activities were between 16 and 32 nkat/g hemoglobin.

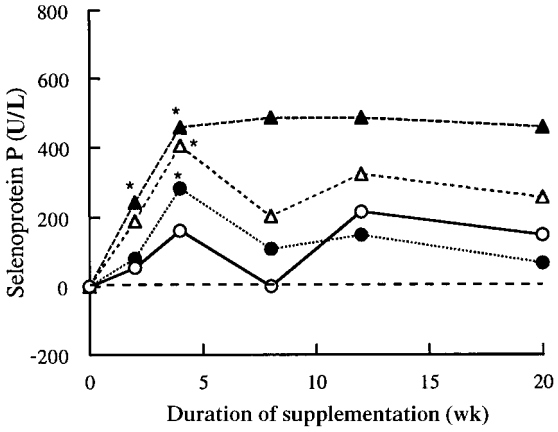

FIGURE 5. Selenoprotein P concentrations in plasma during supplementation with 10 (○), 20 (●), 30 (△), and 40 (▲) μg Se as selenomethionine daily for 20 wk. Values are differences between each supplemented group and the control group, adjusted for sex and baseline measures. Differences between the 40-μg group and the control group were significant after week 2. Differences between the 30-μg group and the control group were significant at 4 and 12 wk, and that between the 20-μg group and the control group at 4 wk. *Significant increase from the previous week relative to the control group, $P < 0.05$. The SDs of selenoprotein P concentrations were between 130 and 210 U/L.

Thyroid hormones

Mean values for T_4 at baseline and at week 20 are shown in **Table 2**. T_4 concentrations decreased from week 0 to week 20 in all groups and after baseline concentrations were adjusted for, the differences between the T_4 concentrations of the control group and treatment groups were −19, −7, −9, and −11 nmol/L for the 10-, 20-, 30-, and 40-μg groups, respectively. Only the difference between the control group and the 10-μg group was significant. When results from all supplemented individuals (10–40 μg) were combined, the decrease in T_4 was also significantly different from the change in T_4 of the control group. Thyroglobulin concentrations did not change significantly with supplementation.

DISCUSSION

A response of GSHPx activities and selenoprotein P concentrations to selenium supplementation may reflect marginal selenium status and, thus, the New Zealand population is an ideal group in which to investigate selenium requirements. Previous studies have used higher supplementation doses, eg, 50–200 μg Se (7, 16, 20, 21), whereas this study used a range of intakes from the lowest in New Zealand to the US RDA. This study is also the first to include selenoprotein P and thyroid hormone measurements.

There has been some debate over the choice of selenium compound for supplementation studies. Although selenomethionine and inorganic forms have similar bioavailabilities for GSHPx activity (4, 16), it was suggested that selenomethionine may not adequately represent dietary selenium (10). We chose selenomethionine for this study because it was the form used in the Chinese study (1), allowing direct comparison. Second, seleno-methionine is the major form in cereals (22, 23) and is therefore a major contributor to total selenium intake, whereas the contribution of inorganic selenium is unlikely to be significant. This is particularly relevant for New Zealand, where importation of high-selenium Australian wheat has had a dramatic effect on selenium status (24, 25). It was therefore preferable that total selenium intake (food and supplement) was of a similar form. Although selenomethionine is incorporated nonspecifically into proteins, the accumulation of selenium that occurs at high intakes of selenomethionine would likely be minimal at these relatively low intakes. Furthermore, nonspecific incorporation is minimized when methionine supply is sufficient (26) and it is unlikely that our subjects had insufficient methionine intakes.

Plasma selenium and GSHPx activities increased in all groups, but our statistical analysis indicated that GSHPx activity reached a plateau in the 40-μg group only. With a few more subjects in each group, we might have been able to establish statistically a plateau in the 30-μg group also. As was also observed in other studies (27, 28), there was large individual variation in GSHPx response, which has made establishment of the exact intake necessary for maximal GSHPx activity difficult. Each individual appears to reach his or her own optimal GSHPx activity through a balance between selenium intake and other factors such as oxidant stress, dietary factors, age, sex, and hormones (7, 16).

The plasma selenium concentration needed to achieve full expression of plasma GSHPx activity was ≈1.14 μmol/L, which agrees well with the value of 1.20 μmol/L arrived at in an earlier New Zealand study (16), and with the study of Yang et al (1) in which maximal activity of plasma GSHPx occurred at a whole-blood selenium concentration of 1.13 μmol/L (29). Although the GSHPx enzymes in plasma and erythrocytes are different, in our assay, plasma activity accounts for only 5% of total activity measured in whole blood, and the latter has been shown to mirror erythrocyte GSHPx activity closely (16). Whole-blood GSHPx activity in the 40-μg group also reached a plateau at a selenium concentration of ≈1.15 μmol/L; thus, both extracellular and cellular GSHPx appear to plateau at similar blood selenium concentrations.

Investigations in human subjects suggest that selenoprotein P may also be a suitable indicator of selenium status (18, 30, 31) and may in fact be superior to GSHPx activity because it constitutes a greater proportion of plasma selenium (32). Statistical analysis of our results suggests that selenoprotein P is maximized at selenium concentrations a little lower (1.05–1.10 μmol/L) than those for GSHPx. Marchaluk et al (30) reported that selenoprotein P concentration increased up to a selenium concentration of 1.2–1.5 μmol/L and suggested that 1.2–1.7 μmol/L might be the concentration at which seleno-

TABLE 2
Plasma thyroxine concentrations in subjects at baseline and after 20 wk of daily supplementation with 0, 10, 20, 30, or 40 μg Se as selenomethionine[1]

Selenium dose	Plasma thyroxine	
	Week 0	Week 20
	nmol/L	
0 μg/d (n = 10)	95 ± 32	99 ± 30
10 μg/d (n = 9)	108 ± 21	93 ± 10[2]
20 μg/d (n = 10)	97 ± 15	88 ± 15
30 μg/d (n = 10)	97 ± 18	90 ± 17
40 μg/d (n = 11)	95 ± 23	89 ± 19
All supplemented subjects (n = 40)[3]	98 ± 17	89 ± 15[2]

[1] $\bar{x}$ ± SD.
[2] Significantly different from week 0 after adjustment for baseline values, $P < 0.05$.
[3] 10–40 μg Se/d.

TABLE 3
Correlation coefficients for relations among selenium concentration (Se), glutathione peroxidase (GSHPx) activities, and selenoprotein P (SeP) concentrations for all weeks of daily supplementation with 0, 10, 20, 30, or 40 μg Se as selenomethionine[1]

Selenium dose	Se and GSHPx		Se and SeP[2]	GSHPx and SeP[2]
	Whole blood	Plasma		
0 μg/d (n = 10)	0.63[3]	0.88[3]	0.73[3]	0.76[3]
10 μg/d (n = 10)	0.64[3]	0.28	0.48	−0.39
20 μg/d (n = 11)	0.33	0.29	0.17	0.13
30 μg/d (n = 10)	0.26	0.08	0.73[3]	−0.18
40 μg/d (n = 11)	0.35	0.08	0.44	0.34

[1]The probability level for each correlation was adjusted for multiple observations for each person.
[2]In plasma only.
[3]$P < 0.05$.

protein P is maximized in subjects consuming normal diets (33). On the other hand, in Chinese subjects with low selenium status who were supplemented with selenate, maximal activities of selenoprotein P were reached at a plasma selenium concentration of 0.9 μmol/L (18), reflecting the varying responses of blood selenium concentrations to different forms of dietary selenium, and perhaps, in different population groups.

The plateauing effect of GSHPx activity with selenium supplementation is reflected in the striking reduction in correlations between selenium and GSHPx activity in whole blood and plasma with increases in supplemental dose (**Table 3**). This may also reflect in part the form of selenium given, because nonspecific incorporation of selenomethionine into tissue proteins is not all reflected in increases in selenoproteins. This reduction in correlation was also documented in Otago residents between 1985 and 1993 (25) when selenium status rose because of increased use of imported Australian wheat products.

We estimated a daily selenium requirement for New Zealanders from our results by using 2 approaches. An "upper estimated requirement" was calculated from the selenium intake necessary for maximal activity of plasma GSHPx, as used in the derivation of the US RDA (34). A "lower estimated requirement" was based on the intake necessary to reach two-thirds of maximal GSHPx activity, as adopted for the WHO/FAO/IAEA normative requirement (11). Our study design was similar to that of Yang et al (1), whose results were used in the derivation of both the US RDA (2) and the WHO/FAO/IAEA requirement (11). However, we chose to use lower supplemental doses because the baseline dietary intakes of our subjects were greater those of Yang et al's subjects and we had expected from previous studies that plateauing of GSHPx would occur at <40 μg supplemental Se.

Although it was unfortunate that we did not use higher doses of supplemental selenium, our statistical analysis showed that plasma GSHPx activity reached a plateau in the 40-μg supplementation group and was close to a plateau in the 30-μg group. Therefore, with the average daily dietary intake of ≈30 μg Se, the total intake required for maximal GSHPx activity was 70 μg Se/d. As for the US RDA, this requirement was converted to a dietary recommendation by adjusting for a variability factor of 1.3 on the basis of a presumed variation of 15% in individual selenium requirements, and thus should cover the selenium needs of 97.5% of the population (35). It was not necessary to adjust for body weight because weights of our subjects were typical of New Zealand adults, and the response of GSHPx activity to selenium supplementation was not dependent on body weight, age, or sex. The resulting upper estimated requirement was 90 μg Se/d, higher than the US RDA of 55–70 μg Se/d and the value of 60–80 μg Se/d that Alfthan et al (29) estimated as the requirement on the basis of earlier New Zealand studies. If we had been able to establish statistically that the GSHPx activity of the 30-μg group had plateaued, then our estimate would have been closer to 78 μg Se/d. A similar calculation gives the same upper estimated requirement as 90 μg Se/d based on whole-blood GSHPx activity (representing cellular GSHPx), which reached a plateau, as determined by statistical analysis only in the 40-μg group.

Our second approach follows that of the WHO/FAO/IAEA Expert Consultation (11), which questioned the assumption that full expression of GSHPx activity is desirable for good health on

TABLE 4
Dietary standards for selenium in selected countries and recommendations by health organizations[1]

Country or organization and dietary standard	Recommended intake	Reference
	μg/d	
Australia, recommended nutrient intake	85 (M), 70 (F)	(43)
United States, recommended dietary allowance	70 (M), 55 (F)	(2)
Norway, reference daily intake	30–60 (M and F)	(44)
United Kingdom		
Reference nutrient intake	75 (M), 60 (F)	(45)
Lower reference nutrient intake	40 (M and F)	(45)
Germany, estimated values for adequate supply	20–100 (M and F)	(46)
Europe, average requirement and reference value for nutrition labeling	40 (M and F)	(47)
US FDA, reference daily intake	70 (M and F)	(48)
WHO, normative requirement	40 (M and F)	(11)

[1]FDA, Food and Drug Administration; WHO, World Health Organization.

the basis of the observation that the ability of blood cells to metabolize hydrogen peroxide is compromised only when GSHPx activity declines to one-quarter or less of normal. Instead, they chose for calculation of their normative requirement an arbitrary value of two-thirds maximal GSHPx activity. In the present study, a plateau in GSHPx activity determined by statistical analysis was reached at ≈56 nkat/g protein in plasma and at 43 nkat/g hemoglobin in whole blood in the 40-μg group. Two-thirds of this maximum value is ≈36 nkat/g protein and 28 nkat/g hemoglobin, respectively, the level of activity already achieved in all groups at baseline and in the placebo group throughout the study. Intake of 30 μg Se/d from the diet was therefore sufficient to achieve this activity, and the lower estimated requirement for adult men and women calculated by using this approach was 39 μg Se/d—almost identical to the WHO/FAO/IAEA normative requirement (11).

The significance of less-than-maximal GSHPx activities and the actual activity required for optimal health are still unclear and need investigation. Sunde et al (36) observed in rats that amounts of GSPHx1 messenger RNA reach a plateau at half the dietary selenium concentration necessary for plateau levels of GSHPx activity. Thus, the normative requirement might be set at a lower intake that gives maximal messenger RNA rather than maximal activity of the enzyme itself. In addition, several selenoproteins other than GSHPx, such as selenoprotein P, the deiodinases, phospholipid hydroperoxide GSHPx, and thioredoxin reductase, have been identified and could be used as endpoints for determining selenium requirements. However, the requirement for maximal activity of these selenoproteins is likely to be less than that for GSHPx (35) because of the hierarchy of importance of selenoproteins in tissues (37). Thus, dietary requirements based on these selenoproteins would presumably lead to recommended intakes lower than current values. Our results showing greater increases in selenoprotein P with every level of supplementation and a plateau reached at lower supplemental intakes (≈30 μg) supports this hierarchy. However, a selenium intake that provides for two-thirds maximal GSHPx activity is not sufficient for maximal concentrations of selenoprotein P. Whether maximal concentrations of selenoprotein P are desirable for optimal health requires further investigation once the function of this protein is established.

Decreased activity of deiodinases in selenium-deficient animals is reflected in reduced deiodination of T_4 to triiodoythyronine (T_3) and therefore increased serum T_4 concentrations (38). A reduction of T_4 with selenium supplementation was observed in several groups with low selenium status (39–42), suggesting that supplementation increased type I deiodinase activity and therefore peripheral conversion of T_4 to T_3. The small decrease in T_4 concentrations in our subjects suggests that a dietary intake a little higher than the baseline selenium intake of 30 μg/d may be desirable for optimal activities of the deiodinases, and that at plasma selenium concentrations < 0.85 μmol/L, there might be changes in the ratio of T_4 to T_3. Our results indicate therefore that our upper estimate is necessary for maximal GSHPx activity, whereas the lower estimate will satisfy requirements for the deiodinases.

International or national nutrition authorities in many countries have proposed recommended dietary intakes for selenium (**Table 4**) that are based on estimates of requirements from Chinese intakes in endemic and nonendemic Keshan disease areas, as well as intakes at which of plasma GSHPx activity is maximized. Recommended intakes range from a high of 85 and 70 μg Se/d for the recommended nutrient intake for Australian men and women, respectively (43), to a low of 40 μg Se/d for the European average requirement (47). Thus, dietary standards throughout the world differ considerably, but have one thing in common: each dietary standard for selenium is available from habitual diets in each country. Our lower estimate of 39 μg/d is a realistic goal for selenium intake in New Zealand, but the upper estimate could be reached only with inclusion of high-selenium foods or supplements.

It is reassuring that our results have produced an upper estimated requirement that is close to the US and Australian recommended intakes and a lower estimated requirement that agrees so well with the WHO/FAO/IAEA recommendations and lends support for the work of Yang et al (1), Levander (34), and the WHO/FAO/IAEA Expert Consultation (11) in deriving dietary selenium recommendations. However, research suggesting that higher selenium intakes may be beneficial for protection against cancer (49, 50) and psychologic function (51) needs to be considered in future deliberations on selenium requirements (35).

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