British Journal of Nutrition (2001), **85**, 157–163
DOI: 10.1079/BJN2000227

Selenium absorption and retention from a selenite- or selenate-fortified milk-based formula in men measured by a stable-isotope technique

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(Received 11 January 2000 – Revised 10 July 2000 – Accepted 04 August 2000)

The present study was designed to determine the apparent absorption and retention of the inorganic Se compounds SeO_3^{2-} and SeO_4^{2-}, which are commonly used for Se fortification of clinical nutrition products and infant formulas. Ten healthy men were fed a milk-based formula labelled with 40 μg Se as $^{74}SeO_3^{2-}$ or $^{76}SeO_4^{2-}$ on two consecutive days using a randomised crossover design. Se stable-isotope analysis of 9 d complete collections of urine and faeces was used to calculate apparent Se absorption and retention. Se retention from $^{74}SeO_3^{2-}$ (41·0 (SD 8·4) %) and from $^{76}SeO_4^{2-}$ (46·0 (SD 7·9) %) was not significantly different ($P > 0·05$). However, Se absorption was significantly higher from SeO_4^{2-} than from SeO_3^{2-} (91·3 (SD 1·4) % *v.* 50·2 (SD 7·8) %, $P < 0·05$). Urinary excretion of the administered dose was 9·2 (SD 1·8) % for $^{74}SeO_3^{2-}$ and 45·3 (SD 8·2) % for $^{76}SeO_4^{2-}$ ($P < 0·05$). Urinary Se excretion kinetics differed significantly for the two Se compounds; 90 % of the total urinary Se was excreted after 121 h for $^{74}SeO_3^{2-}$ and after 40 h for $^{76}SeO_4^{2-}$ ($P < 0·05$). These results suggest that although Se absorption and urinary excretion differ for SeO_3^{2-} and SeO_4^{2-}, both Se compounds are equally well retained when administered at a relatively low dose (40 μg Se). The nutritional impact of Se fortification of foods would thus be expected to be similar when SeO_4^{2-} or SeO_3^{2-} are used.

Selenium fortification: Selenium absorption: Selenium retention: Inorganic selenium compounds: Stable isotopes

Se is an essential trace element for human subjects (National Research Council, 1989). Se is an integral part of glutathione peroxidase (GSH-Px), an enzyme involved in cellular protection against oxidative damage, and of iodothyronine deiodinase which catalyses the conversion of thyroxine into triiodothyronine (Zachara, 1993). Several other selenoproteins, such as selenoprotein P, have been isolated, but their physiological role is still not fully elucidated (Zachara, 1993). In addition, Se has been reported to play a role in the maintenance of optimal immune response (Chandra, 1997), and the role of Se in the emergence of viral mutations has been documented (Beck, 1996). Recently, the impact of Se supplementation on the incidence of certain cancers (Clark *et al.* 1996) was reported.

Low dietary Se intake has been associated with a number of deficiency symptoms, of which Keshan disease, a fatal cardiomyopathy endemic to low-Se areas in China, is the most extensively documented (Lockitch, 1989). Populations at risk of impaired Se status are patients on long-term parenteral or enteral nutrition without Se supplementation (Levander, 1984) and infants consuming infant formulas with a low Se content (Litov & Combs, 1991). Although no overt clinical symptoms of Se deficiency have been diagnosed in these individuals, cases of cardiac and muscular dystrophies have been reported in patients on long-term total parenteral nutrition (Casey & Hambidge, 1985; Hambidge, 1985; Vinton *et al.* 1987; Kelly *et al.* 1988).

Food fortification with Se is not widely used, except for clinical products such as enteral and parenteral nutrition products and infant formulas. Inorganic Se compounds, SeO_3^{2-} or SeO_4^{2-}, are typically used for food fortification and information about bioavailability, absorption and retention of these Se compounds in human subjects is needed to estimate the nutritional impact of Se fortification.

Abbreviations: GSH-Px, glutathione peroxidase; Hb, haemoglobin; HGAAS, hydride-generation atomic absorption spectrometry.

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Previous studies of Se bioavailability have focused mainly on the comparison between inorganic Se, in particular SeO_3^{2-}, and organic Se compounds, selenomethionine or Se-rich yeast (Alfthan *et al.* 1991; Xia *et al.* 1992; Nève, 1995). Only very limited data on Se bioavailability from inorganic compounds (SeO_3^{2-} and SeO_4^{2-}) are available. Furthermore, until now, no information about Se absorption and retention from SeO_3^{2-} and SeO_4^{2-} after intake of relatively small amounts of the Se compounds, relevant to food fortification, has been reported. The recent development of stable-isotope techniques offers the possibility of monitoring absorption and retention of trace elements without introducing radiation exposure (Sandström *et al.* 1993).

The aim of the present study was to compare apparent Se absorption and retention from SeO_3^{2-} and SeO_4^{2-} in men using a stable-isotope technique. The inorganic Se compounds were added to a milk-based formula in quantities relevant to food fortification. A milk-based formula was selected as the food vehicle since it represents the most common products fortified with Se (i.e. products used for enteral nutrition or infant formulas).

Materials and methods

Subjects

Ten healthy men, 24–36 (mean 30) years old, were recruited from personnel at the Nestlé Research Centre, Lausanne, Switzerland. The aims and procedures of the study were explained verbally and in writing before enrolment in the study. All subjects gave written informed consent. A medical examination was carried out before enrolment. Medication, dietary supplements and high-Se food items (i.e. offal meat, fish and other seafoods) were excluded during the study. In all other respects subjects maintained their normal lifestyle and dietary habits during the study. None of the subjects consumed dietary supplements containing Se.

The study protocol was reviewed and approved by the external ethical committee of the Nestlé Research Centre and followed the guidelines of the Helsinki Declaration regarding human subjects.

Stable isotope labels

Highly-enriched elemental ^{74}Se (^{74}Se 98·2 %, ^{76}Se 1·8 %, other Se isotopes <0·1 %), ^{76}Se (^{74}Se 1·2 %, ^{76}Se 98·5 %, ^{77}Se 0·2 %, other Se isotopes <0·03 %) and ^{82}Se (^{74}Se 0·06 %, ^{76}Se 0·62 %, ^{77}Se 0·56 %, ^{78}Se 1·76 %, ^{80}Se 4·8 %, ^{82}Se 92·2 %) were purchased from Isotec (St. Quentin, France). Elemental Se was converted into $^{74}SeO_3^{2-}$, $^{76}SeO_4^{2-}$ and $^{82}SeO_3^{2-}$. SeO_3^{2-} was prepared by dissolving a precise amount of elemental Se (approximately 2·5–10 mg; weighed on a high-precision analytical balance) into 1 ml concentrated HNO_3. The solution was heated for 1 h at 60°C under N_2. The clear solution (approximately 0·2 ml) was made up to 20 ml with deionised water, filtred through a 0·22 μm Teflon filter and stored at 4°C. SeO_4^{2-} labels were prepared from the SeO_3^{2-} solution after evaporation to dryness at 100°C under N_2. H_2O_2 (5 ml) was added to the white precipitate to oxidise SeO_3^{2-}. The solution was gently heated to 70°C and 0·2 ml KOH (2 mol/l) was added. The volume was reduced under N_2 to about 1 ml at 70°C and the oxidation step repeated three times. The solution was evaporated to dryness, the white precipitate redissolved in deionised water and 0·1 ml concentrated HNO_3 was added. Finally, the solution was made up to 20 ml, filtered through a 0·22 μm Teflon filter and stored at 4°C.

Speciation of SeO_3^{2-} and SeO_4^{2-} stable-isotope solutions was performed by continuous-flow hydride-generation atomic absorption spectrometry (HGAAS) based on the principle that only SeO_3^{2-}, but not SeO_4^{2-}, can be directly measured by HGAAS on reaction with a $NaBH_4$ reductant solution (Van Dael *et al.* 1995). Determination of SeO_4^{2-} by HGAAS requires an HCl reduction step before HGAAS analysis.

Se concentration of the stable-isotope solutions was verified by continuous-flow HGAAS (Van Dael *et al.* 1995). For each isotope, stock solutions of 100 μg Se/ml in HNO_3 (0·01 mol/l) were prepared.

Test meals

A commercial ready-to-feed milk-based formula was used as the test meal (Carnation Follow-Up®; Carnation, Glendale, CA, USA). The product contained (g/l): protein 32·5, fat 26·6, carbohydrate 85·6. Each test meal was prepared by the addition of 40 μg Se as $^{74}SeO_3^{2-}$ or $^{76}SeO_4^{2-}$ to 500 g formula. Test meals were prepared 15 h before administration and stored at 4°C until consumed.

Study design

A randomised crossover design was employed. Labelled test meals ($^{74}SeO_3^{2-}$- or $^{76}SeO_4^{2-}$-fortified formula) were administered after an overnight fast on two consecutive days. No food or drink was allowed for 3 h following intake of the labelled test meals. Complete collection of faeces and urine were made for 10 d, starting immediately after intake of the first test meal. Individual portions of faeces were collected in acid-washed plastic containers. Urine was collected in acid-washed plastic bottles in 4 h portions during the first 4 d followed by 24 h collections for the remaining period of the study. A baseline 24 h urine sample was collected the day preceding the study. All excreta were labelled with the subject's code, date and hour of collection and immediately frozen at −40°C. Faecal material was freeze-dried and homogenised using acid-washed mortars and pestles. A faecal marker, 100 mg Brilliant blue, was used to determine the start of the faecal monitoring. A gelatine capsule containing the dye was ingested on the evening before intake of the first test meal.

All individual faecal and urine samples (4 h and 24 h collections) were analysed for total Se content and Se isotopic ratios.

A venous blood sample (20 ml) was drawn after an overnight fast into heparinised tubes for assessment of Se status. Plasma and erythrocytes were separated by centrifugation (2000 ***g*** at 4°C for 15 min). Erythrocytes were

washed twice with saline (0·9 g NaCl/l). Plasma and erythrocyte samples were stored at −80°C until analysed.

Analytical methods

Total Se and stable-isotope ratios in samples of faeces and urine were determined after acid digestion and derivatisation by GC–MS (Van Dael *et al.* 1998). Briefly, samples of urine and faeces were acid-digested using a mixture of HNO_3–$HClO_4$ (3:1, v/v) followed by the addition of HCl to convert all Se to SeO_3^{2-}, which was then derivatised with 4-nitro-phenylene-diamine to form the volatile piazselenole before the analysis by GC–MS. Total Se in the faecal and urine samples was determined by isotope-dilution GC–MS using $^{82}SeO_3^{2-}$ as a spike. The Se analysis was validated against the standard reference material NIST 2670 toxic metals in freeze-dried urine (National Institute of Standards and Technology, Gaithersburg, MD, USA).

Total Se in the milk-based formula was determined by continuous-flow HGAAS (Van Dael *et al.* 1995) and validated against standard reference material NIST 1549 non-fat milk powder (National Institute of Standards and Technology).

Plasma and erythrocyte Se levels were determined by continuous-flow HGAAS (Van Dael *et al.* 1995) and expressed as μg Se/l and μg Se/g haemoglobin (Hb) respectively. The analysis was validated against a commercial whole-blood and serum quality-control material (Seronorm trace elements whole-blood batch no. 030016 and serum batch no. 010017; Nycomed, Oslo, Norway). Hb was analysed by the cyanomethaemoglobin technique (Makarem, 1974).

Plasma and erythrocyte GSH-Px activities were determined according to Belsten & Wright (1995) and expressed per mg protein and per g Hb respectively. Plasma protein concentrations were determined using a colorimetric method (Pierce BCA, Rockford, IL, USA).

Apparent selenium absorption and retention

Apparent Se absorption and retention were calculated according to Turnlund *et al.* (1993). Apparent absorption of ^{74}Se and ^{76}Se was calculated as the difference between the administered dose and the total amount of stable isotopes excreted in faeces. All faecal material collected from the appearance of the faecal marker in the stools until day 9 after administration of the ^{74}Se and ^{76}Se isotope labels was included in the calculation of apparent ^{74}Se and ^{76}Se absorption respectively. Apparent Se retention was based on apparent absorption and adjusted for the amount of ^{74}Se and ^{76}Se excreted in urine collected over 9 d following the administration of the specific isotope.

In addition, cumulative apparent Se absorption was calculated based on the faecal excretion during consecutive 24 h faecal collections (i.e. 24 h, 48 h, 72 h etc.) after administration of the labelled test meals. Similarly, cumulative urinary Se excretion was calculated based on consecutive 24 h urinary collections. Se retention of the stable-isotope doses was calculated as the difference between the cumulative excretion of ^{74}Se and ^{76}Se in faeces and urine.

Statistical evaluation

Differences in Se absorption, retention and urinary excretion between SeO_3^{2-} and SeO_4^{2-} were evaluated using the *t* test procedure for crossover studies according to Hills & Armitage (1979). Results are expressed as means and standard deviations, and statistical significance is defined as $P < 0{\cdot}05$.

Results

Plasma Se concentration was 81·5 (SD 14·4; range 60·5–104·0) μg Se/l, erythrocyte Se was 0·304 (SD 0·047; range 0·248–0·385) μg Se/g Hb, plasma GSH-Px was 6·4 (SD 0·8; range 5·3–7·7) units/g protein and erythrocyte GSH-Px activity was 52·4 (SD 6·6; range 41·9–62·8) units/g Hb. Baseline urine Se concentration was 27·8 (SD 7·5; range 15·7–36·4) μg Se/l and 24 h urinary Se excretion was 32·1 (SD 6·1; range 23·8–39·4) μg Se. The Se level of the milk-based formula was 12·6 (SD 0·4) μg Se/l (6·0 μg Se/500 g formula). At least 97·5 % of the elemental stable-isotope labels were transformed into $^{74}SeO_3^{2-}$, $^{76}SeO_4^{2-}$ and $^{82}SeO_3^{2-}$.

Apparent Se retention from $^{74}SeO_3^{2-}$ (41·0 (SD 8·4 %; range 21·9–52·4) %) and $^{76}SeO_4^{2-}$ (46·0 (SD 7·9; range 33·3–56·1) %) was not significantly different. However, apparent Se absorption and urinary excretion from the two Se compounds differed considerably. Se absorption was significantly ($P < 0{\cdot}05$) higher from SeO_4^{2-} (91·3 (SD 1·4; range 89·5–93·4) %) than from SeO_3^{2-} (50·2 (SD 7·8; range 31·9–59·2) %). Urinary excretion of the administered dose represented 9·2 (SD 1·8) % $^{74}SeO_3^{2-}$ and 45·3 (SD 8·2) % $^{76}SeO_4^{2-}$, corresponding to 18 and 50 % Se absorbed from SeO_3^{2-} and SeO_4^{2-} respectively.

Cumulative faecal Se excretion (Fig. 1) differed significantly ($P < 0{\cdot}05$) between the two Se compounds; 90 % of the total faecal Se (^{74}Se and ^{76}Se) was excreted after 152 (SD 18) h and 65 (SD 21) h for $^{76}SeO_4^{2-}$ and $^{74}SeO^{2-}$ respectively. The corresponding times for 95 % excretion were 177 (SD 14) h and 99 (SD 21) h for $^{76}SeO_4^{2-}$ and $^{74}SeO_3^{2-}$ respectively ($P < 0{\cdot}05$). Cumulative urinary Se excretion (Fig. 2) also differed significantly ($P < 0{\cdot}05$) between the two Se compounds; 90 % of the total urinary Se (^{74}Se and ^{76}Se) was excreted after 40 (SD 30) and 121 (SD 51) h for $^{76}SeO_4^{2-}$ and $^{74}SeO_3^{2-}$ respectively. The corresponding times for 95 % excretion were 84 (SD 52) and 144 (SD 55) h for $^{76}SeO_4^{2-}$ and $^{74}SeO_3^{2-}$ respectively ($P < 0{\cdot}05$). Under the present experimental conditions, apparent Se retention based on cumulative faecal and urinary excretion of ^{74}Se and ^{76}Se showed that Se retention was not significantly different between SeO_3^{2-} and SeO_4^{2-} ($P > 0{\cdot}05$) based on 120 h ($^{74}SeO_3^{2-}$ 44·7 (SD 8·0) %, $^{76}SeO_4^{2-}$ 50·0 (SD 7·2) %) or 168 h ($^{74}SeO_3^{2-}$ 42·3 (SD 8·2) %, $^{76}SeO_4^{2-}$ 47·6 (SD 7·6) %). Apparent Se retention based on 120 or 168 h cumulative faecal and urinary excretion did not differ significantly from data based on 9 d collections ($P > 0{\cdot}05$).

No effect of the order of administration of labelled test meals was detected for Se absorption, urinary excretion or retention. For $^{74}SeO_3^{2-}$ Se retention was positively correlated with absorption (r 0·98, $P < 0{\cdot}05$), whereas for

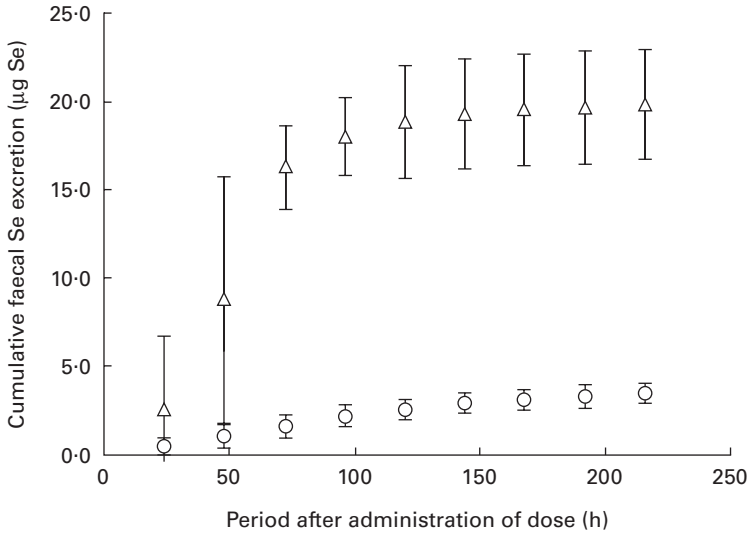

Fig. 1. Cumulative faecal selenium (^{74}Se and ^{76}Se) excretion by healthy men after a single oral dose of 40 μg selenium as $^{74}SeO_3^{2-}$ (△) or $^{76}SeO_4^{2-}$ (○) added to a milk-based formula. Values are means with their standard errors represented by vertical bars for ten subjects. For details of experimental procedures, see p. 159.

$^{76}SeO_4^{2-}$ Se retention and urinary excretion were negatively correlated (r −0·99, $P < 0·05$).

Discussion

The present study demonstrated that in men, mean apparent Se retention was similar for SeO_3^{2-} and SeO_4^{2-} added to a milk-based infant formula (41 % *v.* 46 %), although large differences in absorption and urinary excretion were found. The study design and the dose of 40 μg Se for each isotope were based on nutritional considerations. Preliminary analytical data showed that accurate Se stable-isotope measurements in urine and faeces were possible after intake of 40 μg Se and demonstrated the analytical feasability of the study (Van Dael *et al.* 1998). The labelled test meals were administered in a crossover design on two consecutive days. A milk-based formula was selected as the test meal because milk is the most common raw material for dietetic foods such as enteral nutrition products or infant formulas. These milk-based formulas, often the only dietary source of energy and nutrients, are naturally low in Se and often fortified with Se in order to meet dietary Se requirements.

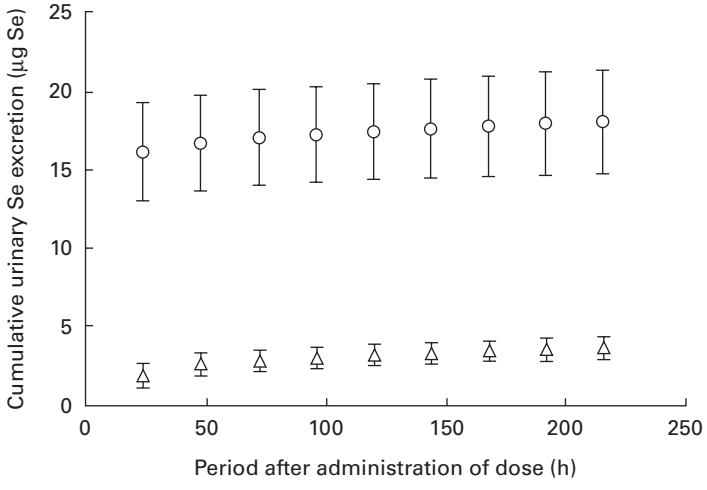

Fig. 2. Cumulative urinary selenium (^{74}Se and ^{76}Se) excretion by healthy men after a single oral dose of 40 μg selenium as $^{74}SeO_3^{2-}$ (△) or $^{76}SeO_4^{2-}$ (○) added to a milk-based formula. Values are means with their standard errors represented by vertical bars for ten subjects. For details of experimental procedures, see p. 159.

Thus, information about Se retention and absorption from these products at relevant fortification levels is needed. The daily Se intake in Switzerland is estimated to be 60–70 μg Se/d (Pfannhauser, 1994). The administration of 40 μg Se as SeO_3^{2-} or SeO_4^{2-} on two consecutive days increased the total dietary Se intake of the subjects to levels comparable with the upper range of Se intakes in Europe (van Dokkum, 1995). Previous data on Se retention and absorption were based on Se doses ranging up to 200 μg Se (i.e. considerably higher than the daily dietary Se intake in most countries). Our data are therefore more relevant to Se retention and absorption from Se-fortified foods within the range of habitual dietary Se intakes in Europe. The subjects in the present study were healthy European men with adequate Se status, as judged from blood Se status indicators and 24 h urinary Se excretion (Robberecht & Deelstra, 1984; Van Dael & Deelstra, 1993). Se retention data for SeO_3^{2-} and SeO_4^{2-} found in the present study are in agreement with those of Thomson & Robinson (1986), who reported Se retention from a 1000 μg dietary Se supplement in New Zealand adults using a chemical-balance technique (mean retention (%) SeO_3^{2-} 40, SeO_4^{2-} 32). Percentage Se retention was comparable with that found in the present study despite the much higher doses of Se administered. Our data on Se retention from the test meal labelled with SeO_3^{2-} are within the wide range of reported results (30–75 %) based on stable-isotope techniques (Christensen *et al.* 1983; Sirichakwal *et al.* 1985; Martin *et al.* 1989; Mangels *et al.* 1990; Finley, 1999). The reasons for the wide range in SeO_3^{2-} retention values are not known, although it can be assumed that factors such as differences in study design, stable-isotope dose, dietary regimen and normal Se intake may play a role.

Previous Se retention studies differed considerably in the number of days (5–21) used in the faecal and urinary collection period (Christensen *et al.* 1983; Sirichakwal *et al.* 1985; Martin *et al.* 1989; Mangels *et al.* 1990; Finley, 1999). Our data indicate that Se retention for SeO_3^{2-} and SeO_4^{2-} based on 120 h collections of faeces and urine was not significantly different from 9 d balances. However, although our data suggest that complete faecal and urine collection periods shorter than 9 d may be appropriate for studies of Se retention, it is important to consider carefully the collection time, in relation to factors such as gastrointestinal passage time and dietary habits.

Although retention of the two inorganic forms of Se was found to be similar, a large difference was observed in the percentage absorption of SeO_3^{2-} and SeO_4^{2-}; SeO_4^{2-} was almost completely absorbed (91 %) compared with 50 % for SeO_3^{2-}. The between-subject variation in Se absorption was larger for SeO_3^{2-} (CV 15 %) than for SeO_4^{2-} (CV 1·5 %), and confirmed previously reported data on inter-individual variations in SeO_3^{2-} absorption (Martin *et al.* 1989). A wide range of apparent Se absorption values (35–85 %) based on stable-isotope techniques have been reported for SeO_3^{2-} (Janghorbani *et al.* 1982; Christensen *et al.* 1983; Kasper *et al.* 1984; Sirichakwal *et al.* 1985; Martin *et al.* 1989; Patterson *et al.* 1989; Mangels *et al.* 1990; Ducros *et al.* 1991; Finley, 1999). SeO_3^{2-} has been reported to interact with the lumen content, and therefore it is likely that dietary habits influence Se absorption from SeO_3^{2-} (Vendeland *et al.* 1992). Martin *et al.* (1989) reported that the percentage SeO_3^{2-} absorption in adults fed a Se-restricted diet (18 μg Se/d) was significantly higher than that of a group fed 118 μg Se/d (89 *v.* 58 respectively). Recently, Finley (1999) also reported significant differences in percentage SeO_3^{2-} absorption between subjects at low (32·6 μg Se/d) and high (226·5 μg Se/d) Se intake, but contrary to the data of Martin *et al.* (1989), higher mean Se absorption (38 % *v.* 15 %) was reported at the higher Se intake. As in the present study, Thomson & Robinson (1986) found high Se absorption from SeO_4^{2-} (94 (SD 4) %) based on the classical balance technique. Finley (1999) reported Se absorption from SeO_4^{2-} in the range of 68–76 % based on a stable isotope technique. Our absorption data and those of Thomson & Robinson (1986) for both SeO_4^{2-} and SeO_3^{2-} were substantially higher than those reported by Finley (1999). In agreement with the data in human subjects, higher Se absorption from SeO_4^{2-} *v.* SeO_3^{2-} was also reported in animals (Wolffram *et al.* 1985; Vendeland *et al.* 1992). In those studies SeO_4^{2-} was shown to be rapidly transported across the brush border by a carrier-mediated mechanism, whereas SeO_3^{2-} was absorbed by passive diffusion and showed a strong tendency to bind to the brush border.

Renal excretion is the major pathway for elimination of absorbed Se (Oster & Prellwitz, 1990). In the present study Se excretion from SeO_4^{2-} was high; approximately half the Se absorbed from SeO_4^{2-} was re-excreted in urine compared with only 20 % for Se absorbed from SeO_3^{2-}. The urinary excretion of Se absorbed from SeO_4^{2-} was also very rapid; 87 % of the total urinary Se excretion was recovered within 24 h of administration while 90 % of the total Se excretion of SeO_3^{2-} was completed after 3 d. Thomson & Robinson (1986) found similar differences for total urinary Se excretion and excretion kinetics between SeO_4^{2-} and SeO_3^{2-} after administration of a single dose of Se (1 mg). Previous stable-isotope data in adults reported urinary excretion for SeO_3^{2-} which was similar to our findings (Martin *et al.* 1989). The rapid and high Se excretion from SeO_4^{2-} is probably related to its more rapid absorption and metabolism compared with SeO_3^{2-} (Thomson & Robinson, 1986).

As an alternative to inorganic Se, organic Se could be considered for food fortification. Se retention from organic Se compounds such as selenomethionine and Se-rich yeast has been demonstrated to be high at about 80–90 % (Mangels *et al.* 1990; Alfthan *et al.* 1991). Organic Se may not only be incorporated into Se-specific proteins such as GSH-Px and iodothyronine deiodinase but also non-specifically into blood proteins such as albumin and Hb due to homology between selenomethionine and methionine. It has been shown that organic and inorganic Se compounds are equally well utilised for incorporation into GSH-Px but that organic Se gives higher plasma and erythrocyte Se levels (Alfthan *et al.* 1991; Xia *et al.* 1992; Thomson *et al.* 1993; Nève, 1995). The metabolic similarity between selenomethionine and methionine has been suggested as the reason for Se toxicity at high Se intakes (Whanger, 1998).

Se compounds used for fortification of foods for groups

at risk of low Se intake, such as patients with chronic or acute disease or infants, should ensure adequate absorption and retention of Se. Thus, fortification of nutritional products with SeO_4^{2-} could be considered to be more efficient than SeO_3^{2-}. Nevertheless, SeO_4^{2-} may not be the ideal fortificant for individuals with impaired renal function or preterm infants with immature renal function, since SeO_4^{2-} might not be as efficiently excreted as in healthy individuals. For patients with malabsorption syndromes SeO_4^{2-} may be the more appropriate fortification compound, since Se absorption from SeO_3^{2-} has been shown to be markedly reduced in patients with short bowel syndrome (20 %) compared with healthy adults (82 %; Rannem *et al.* 1996). Finally, concern has been raised about the potential pro-oxidative properties of SeO_3^{2-} and its lower stability compared with SeO_4^{2-} when added to foods (Smith *et al.* 1995; Tyrala *et al.* 1996). Although these issues need to be further investigated, they are additional factors potentially in favour of the use of SeO_4^{2-} for Se fortification of foods.

In conclusion, the results from the present study indicate that SeO_3^{2-} and SeO_4^{2-}, when administered within normal dietary intake ranges, were equally well retained in healthy men, although large differences in absorption and urinary excretion were observed. Thus, the nutritional impact of foods fortified with either of these inorganic Se compounds can be assumed to be similar in healthy individuals, at least at the level of Se intake evaluated in the present study.

Acknowledgements

The authors are grateful for the expert technical assistance of Mrs K. Longet, Mrs S. Metairon, Mrs I. Bartholdi and Mrs J. Clough, and for the fruitful discussions with Dr R. Hurrell and Dr P. Kastenmayer.

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