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# Absorption, excretion, and retention of selenium from a high selenium yeast in men with a high intake of selenium

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## Abstract

**Objective:** The purpose of this study was to evaluate the pharmacokinetics of a single dose of selenium (Se) from yeast given to humans with a habitual long-term daily intake at a supra-nutritional level.

**Methods:** Twelve healthy males with a daily supplemental intake of 300 µg Se as selenised yeast over 10 weeks were supplemented with a single dose of 327 µg as stable <sup>77</sup>Se incorporated into selenised yeast manufactured by the same standardised process (SelenoPrecise®, Pharma Nord, Denmark).

**Results:** Absorption of Se from <sup>77</sup>Se-enriched yeast was 89 ± 4% and the retention was 74 ± 6%. The <sup>77</sup>Se excretion from the single-dose was 47 ± 15 µg in urine and 37 ± 13 µg in faeces. The maximum, enriched <sup>77</sup>Se concentration in plasma was 9.8 ± 1.5 µg/l and the time to maximum was 9.2 hours. The plasma half-time of <sup>77</sup>Se was longer with increasing time; 1.7 days for the initial phase (½-2 days), 3.0 days for the middle phase (2-3 days) and 11.1 days for the later phase (3-14 days).

**Conclusion:** The Se from the standardised Se-enriched yeast was well absorbed and retained in the body.

Keywords: *selenium yeast; stable isotope; humans; pharmacokinetics; absorption; excretion*

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Selenium (Se) intake in many European countries is relatively low (1) due to the low soil Se concentrations or poor bioavailability of soil Se for uptake into the plant roots in a great part of Europe. The recommended daily allowances of Se is 50 µg for men and 40 µg for women in the Nordic countries (2), and is based on saturation of the activity of glutathione peroxidase -3 (GSH-Px-3) in plasma. However, saturation of GSH-Px-3 occurs at a relatively low intake of Se (40 µg/d) compared to the intake of Se needed to saturate GSH-Px-1 in erythrocytes (60–80 µg/d) or GSH-Px-1 in platelets (100–110 µg/d). Dietary Se intake may be sub-optimal with respect to prevention of disease, notably in populations of adults in the UK, parts of Europe and China, New Zealand and in the USA (1). There have been indications that certain forms of cancer might be prevented by intake of Se as selenised yeast (Se-yeast), in doses that are higher than required in order to saturate the selenoenzymes (1,

3). Recently, it has been hypothesised that Se intake in the range of 40–100 µg/d primarily has antioxidant and immune-strengthening effects, while intake in the range 200–500 µg/d has specific cancer-preventive properties (4). An intake of Se of this magnitude is, however, difficult to obtain from the diet in most parts of the world where Se content and bioavailability from soils is moderate or low.

Selenium can be found in a number of different Se-containing molecules (species), which are absorbed from the intestinal tract, but retained in different fractions depending on the species (5). Rat studies have shown that selenomethionine (Se-meth) is absorbed by an active transport mechanism, which is shared with methionine, while other species are absorbed by sharing an active transport system with other amino-acids, by simple diffusion or by Na-dependent transport systems (6). Selenium is primarily excreted via urine and faeces. The

amounts in urine and faeces have been shown to be of similar magnitude when Se intake is low (7). The most important factors that influence Se excretion are probably the amount and form of dietary intake and Se status of the individual. Whole-body retention studies in young women following oral administration of radioactive  $^{75}\text{Se}$ -meth have shown that Se elimination is triphasic (8).

Few studies have made use of the stable isotope technique for measurement of Se absorption, excretion and retention in humans. To our knowledge, no study has previously been published on estimation of the absorption and retention of a single dose of Se-yeast in individuals with a supra-nutritional Se intake at the 300  $\mu\text{g}/\text{day}$  level. The aims of this study were to measure the absorption, excretion and retention of Se contained in intrinsically labelled yeast in humans with a high habitual Se intake. The enriched stable isotope approach using  $^{77}\text{Se}$ -SelenoPrecise yeast was applied for the measurements of absorption, retention, and excretion. Furthermore, the pharmacokinetics in a real-life situation of long-term supplementation with Se was investigated.

## Materials and Methods

### Study design

The study was a single dose study with 12 healthy volunteers and was conducted as a pharmacokinetic study. Nine of the subjects were taking Se-supplements regularly and in an attempt to standardise Se intake, subjects were instructed to take 300  $\mu\text{g}$  Se as Se-yeast for ten weeks before entering the study. The stable  $^{77}\text{Se}$  isotope was taken as intrinsically  $^{77}\text{Se}$ -labelled yeast under standardised, controlled conditions. Eleven blood samples were taken after administration of the stable isotope at baseline, after 30 min, 1, 2, 4, 6, 8, 12, 24, 48 and 72 hours. 24 h pooled urines were collected the day before and 3 days after, and faeces was collected the day before and for 5 days after  $^{77}\text{Se}$  administration. Finally, blood, 24 h urine and faeces samples were collected after 14 days for six volunteers randomly selected prior to the beginning of the study.

### Subjects

Twelve healthy males volunteered for the study (Table 1). The average age was 38 years (26–51 years), average body mass index (BMI) was  $25.0 \pm 2.3 \text{ kg}/\text{m}^2$ , and mean fat mass  $19.1 \pm 3.2\%$  (Bio-electrical Impedance Analysis, Omron BF 302, Hoofddorp, Netherlands).

None of the volunteers had any major illness, was treated with medicine, had allergy to Paraaminobenzoic acid (PABA), practised sport at an elite level, or had participated in other clinical studies within a month before inclusion into the present study. The volunteers were examined by a physician, prior to inclusion into the

**Table 1.** Characteristics of the 12 human male volunteers at baseline

	Mean	SD
Age (years)	37.7	8.4
BMI ( $\text{kg}/\text{m}^2$ )	25	2.3
Body fat (%)	19.1	3.2
Hb (g/dl)	15.7	0.8
Ht (%)	49.8	2.5
B-Se ( $\mu\text{g}/\text{L}$ )	212	23
Hg ( $\mu\text{g}/\text{L}$ )	1.6	0.9

BMI = body mass index, Hb = haemoglobin, Ht = haematocrit, B-Se = whole blood Se, Hg = mercury.

study. Eight of the 12 volunteers were smokers. The average duration of smoking was  $18.3 \pm 8.6$  years and the average number of cigarettes per day was  $16.8 \pm 3.7$ . The volunteers were assigned to an intake of 300  $\mu\text{g}$  Se as Se-yeast for 10 weeks before entering the study. Nine of the volunteers had an even longer history of taking organic Se as Se-meth or Se-yeast, usually in amounts of approximately 100  $\mu\text{g}/\text{day}$  (mean intake (min-max) were 119  $\mu\text{g}/\text{d}$  (62.5–225  $\mu\text{g}/\text{d}$ ). Mean total plasma Se was  $182 \pm 14 \mu\text{g}/\text{l}$  and mean whole blood Se was  $213 \pm 23 \mu\text{g}/\text{l}$ . The volunteers continued their habitual intake of food supplements, but were not allowed to take other Se containing products except the 300  $\mu\text{g}$  Se/day as Se-yeast. Table 1 shows the mean baseline whole blood Se and also baseline values of other selected biomarkers, which are all within normal range for a healthy population.

The study was approved by the local ethical committee (Fyn and Vejle County, Denmark, Ref. no 2000-0143) and was conducted in accordance with the Declaration of Helsinki (including amendments of Tokyo 1975, Venice 1983, Hong Kong 1989 and Somerset West 1996). All participants signed an informed consent form.

### Test meal

The stable isotope-labelled yeast was given together with a light meal after an overnight fast. The test meal consisted of one serving of müsli with 200 ml milk (1.5% fat), two slices toast with 10 gram of butter, marmalade, two slices of cheese and one cup of tea or coffee. Nutrient values were calculated using the Dankost 2000<sup>®</sup> dietary assessment software (Danish Catering Center, Herlev, Denmark) (9). The total energy in the meal was 2.9 MJ. Protein content was 16.1%, lipid content was 29.3% and carbohydrate content was 54.6% of the total energy. The calculated content of Se in the meal was 8.4  $\mu\text{g}$ .

### Preparation of selenium supplement

The 10 weeks unlabelled run-in Se preparation used (SelenoPrecise<sup>®</sup>, Pharma Nord, Denmark) was produced

by a standardised procedure using a Pharmacopoeia-controlled growth medium, as a source of carbon, nitrogen, micronutrients, and yeast of *Saccharomyces cerevisiae* (strain PN0056). The yeast was grown aerobically under controlled addition of sodium selenite ( $\text{Na}_2\text{SeO}_3$ ) with a natural Se isotope distribution (abundance of  $^{77}\text{Se} = 7.63\%$ ). The yeast metabolised more than 95% of the added Se, which was incorporated into a range of organic Se-species. The predominant species was Se-meth (81% of all separated and quantified Se species in the enzymatic hydrolysate or corresponding to 60% of the total Se content in the yeast), and the residual selenite ( $\text{SeO}_3^{2-}$ ) in the yeast was less than 1% as determined by enzymolysis and HPLC-ICP-MS (10). The yeast was harvested, washed, pasteurised, spray dried and processed to tablets. The potency of 300  $\mu\text{g}$  per tablet was confirmed by analyses to lie within  $\pm 5\%$  of the target value. The Se yeast, its speciation and long-term bioavailability at dose-levels of 100, 200 and 300  $\mu\text{g}/\text{day}$  has been described in further detail by Larsen et al (11).

The intrinsically labelled single dose of  $^{77}\text{Se}$ -SelenoPrecise<sup>®</sup> (Pharma Nord, Denmark) contained 329.0  $\mu\text{g}$  total Se, which included  $327 \pm 9 \mu\text{g}$  of  $^{77}\text{Se}$  (Chemgas, Boulogne, France) and was produced by the same procedure as indicated above, except that the isotopically enriched  $\text{Na}_2\text{SeO}_3$  used as substrate contained  $^{77}\text{Se}$  at 99.3%. Furthermore, the yeast was freeze-dried instead of spray-dried. The isotopic composition and concentration of Se in the  $^{77}\text{Se}$ -enriched yeast was characterised by mass spectrometric analyses to allow for an accurate administration of the isotope. Speciation analyses showed that Se-meth amounted to 82% of all separated and quantified Se species in the enzymatic hydrolysate, or to 53% of the total Se content in the yeast. The content of  $\text{SeO}_3^{2-}$  was less than 1% (10). The  $^{77}\text{Se}$ -SelenoPrecise yeast was filled into hard gelatine capsules by single weighing of the individual capsules to achieve 327  $\mu\text{g}$   $^{77}\text{Se}$  per dose.

#### Sample collection and preparation

Total faecal samples were collected from the 12 volunteers in acid washed plastic containers for one day before administration of the  $^{77}\text{Se}$ -yeast and for 5 days thereafter, and again for one day after 14 days for 6 of the 12 volunteers. To control the origin in time of the collected faeces samples (12), four different radio-opaque faecal markers (Medi-Fakt, Göteborg, Sweden) were consumed in hard gelatine capsules ( $3 \times 20$  markers per day taken at breakfast, lunch and dinner), one day prior to and during the faecal collection. A total of 60 markers represented one day. One shape of faecal markers were given on the two days prior to the test meal, another shape of faecal markers were given together with the test meal and a third shape of markers were given to the other two main meals

on the test day. A fourth shape of markers was given the following days. This made possible to distinguish between faeces from the day before, from the time of intake, from the first day and the 5 days following administration of the isotope. The faeces samples were pooled accordingly. 24-h urine was collected the day before and 3 days after  $^{77}\text{Se}$  administration and again after 14 days for six of the volunteers. To control complete collection of urine PABA tablets ( $3 \times 80 \text{ mg}$ ) were consumed at the same days as the urine collection (13).

Blood for plasma analyses was drawn at  $-\frac{1}{2}$  (baseline),  $\frac{1}{2}$ , 1, 2, 4, 6, 8, 12, 24, 48, 72 hours and for half of the group additionally after 14 days after intake of the stable isotope yeast preparation. Blood was drawn into heparin tubes (Becton-Dickinson, NJ, USA). The blood was centrifuged at room temperature for 10 minutes at 3000 rpm, plasma was pipetted into plastic vials and kept at  $-20^\circ\text{C}$  until the time of analysis of plasma Se. At baseline ( $-\frac{1}{2} \text{ h}$ ) blood was also drawn into a heparin tube and kept at  $-20^\circ\text{C}$  until analysis of trace elements and creatinine.

#### Analytical methods

Faeces samples were freeze-dried, homogenised, microwave digested using nitric acid (14) and then pooled according to the content of faecal markers (15). Prior to Se analysis, the plasma and urine samples and the digested faeces samples were diluted using an aqueous diluent containing 0.5% Triton X-100, 2% nitric acid and 3% methanol. An ELAN 6100 ICP-MS instrument (Perkin-Elmer SCIEX, Concord, Ont., Canada), which was equipped with a dynamic reaction cell (DRC), was used for the interference-free Se measurements as the DRC removed the argon dimer interferences (16). Selenium was detected in all samples as  $^{76}\text{Se}$ ,  $^{77}\text{Se}$  and as  $^{80}\text{Se}$  and quantified by the method of standard additions (17). The degree of enrichment in  $^{77}\text{Se}$ , was estimated from the total amount of  $^{77}\text{Se}$  (supplemented plus endogenous) minus the endogenous amount of  $^{77}\text{Se}$ . The total amount of endogenous Se with a natural isotopic composition was quantified from measurement of  $^{80}\text{Se}$ , the abundance of which was not altered by the administered single dose of 99.3% pure  $^{77}\text{Se}$  isotope. The total Se content in the samples was calculated as the sum of the  $^{77}\text{Se}$ -enriched and endogenous concentrations of Se. The minimum detectable increase in  $^{77}\text{Se}$  was 0.38  $\mu\text{g}/\text{l}$ , 0.58  $\mu\text{g}/\text{l}$  and 15  $\mu\text{g}/\text{kg}$  for plasma, urine and faeces, respectively. The reproducibility was estimated by random duplicate analyses and the accuracy by analysis of the reference materials Seronorm Serum (Nycomed Pharma Diagnostics, Oslo, Norway) and BCR CRM 185 Bovine Liver (BCR, Institute for Reference Materials and Measurements, Geel, Belgium) in parallel with samples (17). Urine was also analysed for PABA (13) and

creatinine (Jaffé Method, Roche Diagnostics GmbH, Mannheim, Germany) was measured in urine and plasma.

#### Statistics and calculation

The enriched concentrations of  $^{77}\text{Se}$  in the individual samples of plasma, urine and faeces were integrated to yield the amount of the isotope that appeared in these biological samples from each human volunteer (17). Only faecal samples, which contained radio-opaque markers, originated from the meal with the  $^{77}\text{Se}$ -yeast administration, or with markers taken later than this, were included.  $^{77}\text{Se}$  mass balances were established and based on all days of faecal and urinary collections. The total  $^{77}\text{Se}$  content of the faecal and urinary samples was analysed and excretion of faecal radio-opaque markers was used to calculate average daily excretion. The apparent absorption of  $^{77}\text{Se}$  was calculated as the difference between the intake of  $^{77}\text{Se}$  tracer and the amount of tracer from this intake found in faeces during the collection period. The retention of  $^{77}\text{Se}$  was calculated from the difference between the administered amount of  $^{77}\text{Se}$  and the sum of faecal and urinary excretions during the collection period.

One-way ANOVA was conducted for the data at different time-points. The significance level was predefined as 95%. Correlation between creatinine clearance and total urine  $^{77}\text{Se}$  was examined in a scatterplot.

## Results

#### Compliance

Compliance was assessed directly by supervising the intake of the  $^{77}\text{Se}$ -preparation. On the day of isotope administration the diet was standardised. The rest of the time participants were asked about compliance with the protocol when returning specimens. Compliance for the  $^{77}\text{Se}$  preparation was 100%. Compliance for the 10 weeks daily intake of 300 µg Se as SelenoPrecise yeast

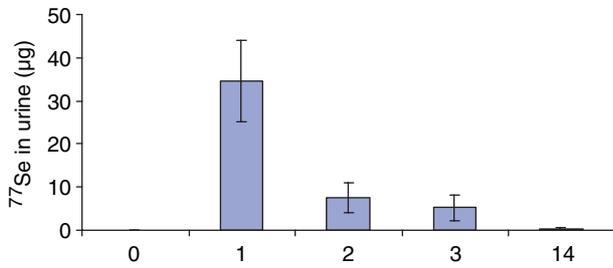
before the study start was 90% or better, which was controlled by interviewing each participant. One of the participants reported that he forgot to take one capsule of the radio-opaque faecal markers (day five in the collection period), the rest of the participants had 100% compliance. One of the participants missed one blood drawing because of illness (diarrhoea and vomiting at day 4). The completeness of urine collection was controlled by analysis of PABA, which showed a full recovery of (mean and SD)  $108\% \pm 6.6\%$ . Radio-opaque faecal markers controlled the complete collection of faeces originating from the day of  $^{77}\text{Se}$  administration. The  $^{77}\text{Se}$  preparation was taken at the same time as the 20 identifiable radio-opaque faecal markers. Retrieval of at least 18 of 20 markers was considered as a full sample collection. All the participants were found to have complete faeces collection. As an extra control of the urine and faeces collection the participants were asked every day for any irregularities in their collection. Three subjects did not collect faeces on day 4 and 5 due to either irregular defecation or constipation. Other reported irregularities were negligible.

#### Selenium pharmacodynamics

The apparent absorption of Se from a single dose of 327 µg  $^{77}\text{Se}$  as Se yeast was found to be  $88.7 \pm 3.9\%$  in the Danish men with a high habitual Se intake (Table 2). The mean  $^{77}\text{Se}$  excretion from the single dose over the collection days in urine and faeces was  $47.4 \pm 14.8$  µg and  $37.1 \pm 12.7$  µg, respectively. The  $^{77}\text{Se}$  excretion in urine is shown in Fig. 1. The  $^{77}\text{Se}$  tracer percentage in faeces over the period is shown in Fig. 2. The mean  $^{77}\text{Se}$  retention at day 4 was  $242 \pm 19$  µg/d, which corresponds to  $74.2 \pm 5.9\%$  of the  $^{77}\text{Se}$  intake. The results for the mean  $^{77}\text{Se}$  plasma concentrations during the 72 h following administration of  $^{77}\text{Se}$  are shown in Fig. 3. The mean  $^{77}\text{Se}$  plasma concentration at day 14 was  $2.28 \pm 0.18$  µg/l (n = 6) (not shown). The maximum  $^{77}\text{Se}$  concentration in plasma was reached after six hours (n = 1), eight hours

**Table 2.** Intake, absorption, excretion and retention of 327 µg  $^{77}\text{Se}$  administered as a single dose to 12 men with a habitual high intake of Se. The values are based on urinary and faecal excretion from days 1–3 after administration of  $^{77}\text{Se}$ . Values are given as median values with 10–90% percentiles and also as means  $\pm$  standard deviation. Values are further given as % of the ingested dose with 10–90% percentiles and also  $\pm$  standard deviation.

	µg		%	
	Median (10–90 pct)	Mean (SD)	Median (10–90 pct)	Mean (SD)
Intake	–	327	–	100
Absorption	284 (281–308)	290 (13)	87 (86–94)	89 (4)
Retention	248 (220–258)	242 (19)	76 (67–79)	74 (6)
Faecal excretion	43 (19–46)	37 (13)	13 (6–14)	11 (4)
Urinary excretion	44 (36–61)	47 (15)	14 (11–19)	15 (5)



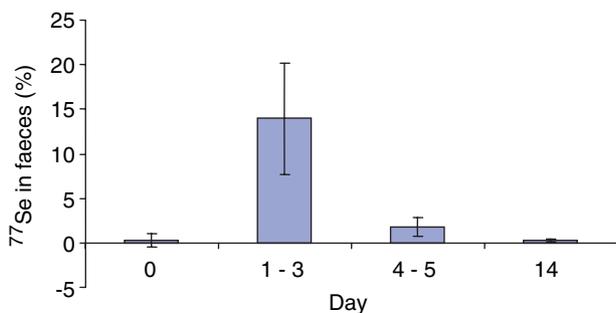
**Fig. 1.** Mean tracer in urine from the single  $^{77}\text{Se}$  dose detectable before administration (day 0) of a single oral dose and at day 1, 2, 3, and 14 after tracer administration (Mean  $\pm$ SD; Day 0 to 3: n = 12; Day 14: n = 6).

(n = 7), and 12 hours (n = 4). The mean  $T_{\max}$  was 9.17 hours (median  $T_{\max}$  = 8 h) and the mean  $^{77}\text{Se}$  plasma  $C_{\max}$  from the single dose was  $9.78 \pm 1.53$  µg/l. The half-time of  $^{77}\text{Se}$  in plasma (n = 6) was longer with increasing time; 1.7 days for the initial phase ( $\frac{1}{2}$ -2 days), 3.0 days for the middle phase (2–3 days) and 11.1 days for the later phase (3–14 days). The mean plasma Se concentration (not shown) varied slightly, but statistically non-significant, between days 0–14.

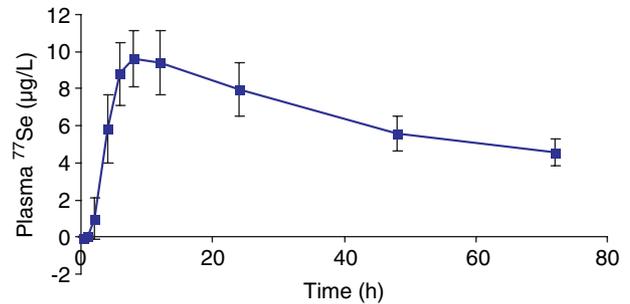
Creatinine clearance was calculated based on serum creatinine, 24 h urine creatinine excretion and on diuresis within 24 h. The urinary creatinine clearance was within the normal range for the age group (0.8 – 2.8 ml/s). No relation was found between creatinine clearance and total urine  $^{77}\text{Se}$  excretion (data not shown).

#### Side effects

One subject reported pharyngitis for three days, one had hay fever for one day, and one had diarrhoea and vomiting for one day. None of these reactions were associated with the yeast-based Se supplements. No other adverse effects were observed. Two of the subjects took



**Fig. 2.** Mean tracer in faeces from the single  $^{77}\text{Se}$  dose as percentage of the total selenium detectable before marker excretion (day 0) and in samples of day 1–3, 4–5, and day 14 after tracer excretion. The periods were pooled according to the markers taken simultaneous with the single dose. The data are presented as mean values  $\pm$ SD with 12 specimens at day 0 and day 1–3. The data of day 4–5 and day 14 included 9 and 6 specimens, respectively.



**Fig. 3.** Mean plasma  $^{77}\text{Se}$  tracer concentration (µg/L) in 12 human volunteers given a single oral dose of 327 µg  $^{77}\text{Se}$  as Selenium yeast together with a light meal after an overnight fast (Mean  $\pm$ SD; n = 12).

medication during the study. One received treatment with penicillin due to pharyngitis in the beginning of the collection period, and one had a short attack of hay fever and took anti-histamine.

#### Discussion

Intake of Se in the range of 200–500 µg/d has been suggested to be anticarcinogenic (3). However, intakes of this magnitude are difficult to obtain from the diet and therefore there might be a need for safe and efficient supplements. A number of studies have compared the metabolic fate of inorganic Se (selenite and selenate) or SeMeth given in foods or as supplement (18, 19). These previous studies have shown that besides the chemical form of ingested Se and Se status of the individual, other factors e.g. age, alcohol consumption, smoking and hormonal therapy might also affect Se metabolism and status (20, 21).

The high absorption of Se observed in the present study in participants with a high habitual Se intake is in agreement with previous studies (7, 18, 22). The absorbed fraction of Se from different Se yeasts is known to be large, but the absorption and bioavailability of Se vary between different batches of Se-yeast (23), which makes comparisons of results from studies using different Se-yeast difficult. A comparative study showed, that the human absorption of Se from different Se-yeasts, as expressed by plasma Se concentrations, depended on baseline Se status or on Se speciation in the yeast-based preparations used (24). Other research groups have also shown that the whole-body Se content is a critical factor affecting the homeostatic metabolism of newly ingested Se (18, 19, 25, 26), and the reason is probably that some adaptation to different intakes occur. Some studies have shown that the fractional absorption actually increases by increasing the intake of Se (19, 25) and although this study was not designed to compare absorption at high and low intakes of Se, this would suggest that absorption is not a major regulator of homeostasis of Se.

Regarding the Se speciation of the SelenoPrecise yeast, analysis by HPLC-ICPMS following enzymatic hydrolysis showed, that SeMeth constituted 60% of the total Se content in the yeast (10). Selenocysteine was not detected in the yeast, which was in accordance with the fact that the yeast genome does not include the necessary codon for its biosynthesis (27). The remainder of the Se was present as more than 30 unknown Se-species of low abundance whereas selenite and SeMeth Se-oxide were present at less than 1% of the total Se content in the yeast. The identity of Se species, which correspond to the non-extractable fraction (26%), remained unknown (10). A separate study characterised the Se species contained in the  $^{77}\text{Se}$ -SelenoPrecise yeast used in this study (10). The results showed that approximately 55% of the total Se in the yeast was present as SeMeth, which was similar to the value found for the SelenoPrecise yeast.

Selenium excretion occurs primarily via urine and faeces (8). Levels of faecal excretion of Se have been reported to be similar to levels of urinary Se excretion when dietary levels of Se are not excessive. In the present study we observed a relatively low excretion of Se from urine (15%) and faeces (11%) of a single  $^{77}\text{Se}$  dose during the first 4 days. The majority of the urinary Se was excreted within the first 24 hours after intake of  $^{77}\text{Se}$  and thereafter excretion was reduced to low levels. The same tendency was observed for the faecal excretion, although the  $^{77}\text{Se}$  faecal excretion remained high until 48–72 hours after the single  $^{77}\text{Se}$  supplementation (results not shown). This might indicate entero-hepatic recirculation in agreement with previous studies with SeMeth (28) showing that a significant proportion (46%) of the amount leaving the liver was found to re-enter the intestines.

The analytical results of plasma and urine samples from day 1 to day 14 showed that the elimination of  $^{77}\text{Se}$  was not complete and that the plasma concentration was approximately 20% of the initial peak plasma concentration. At day 14 the  $^{77}\text{Se}$  excretion was still on-going, as the  $^{77}\text{Se}$  concentration in urine and faeces had not returned to baseline. The total amount of  $^{77}\text{Se}$  excreted at day 14 was 1.61  $\mu\text{g}$ , 48% of which was excreted via kidneys to urine and 52% was excreted via faeces.

A kinetic model for SeMeth was proposed by Patterson et al (29) who suggested that SeMeth is extensively recirculated and passed through liver, pancreas and peripheral tissues many times before being excreted. This would explain that labelled Se is still found in faeces and urine 2 weeks after administration. The same was observed a year after labelling of body pools by Veillon et al (30). In the present study we also observed a high excretion of total Se in both urine and faeces (data not shown), which reflects the high intake of Se in this group of people and is in accordance with results from other studies feeding high amounts of Se (19). We cannot, however, from our data, conclude if this is caused by

adaptation to the higher intake. In the study by Hawkes (19) it was observed that when the intake of Se was increased from normal (47  $\mu\text{g}/\text{d}$ ) to high (297  $\mu\text{g}/\text{d}$ ), the urinary and faecal excretion of Se almost doubled, while deposition in muscles increased by approximately 25% during the 99 days with high Se intake. It is likely that the high intake in our study has changed the distribution of Se in the body.

According to Clark *et al.* (3) steady state in plasma-Se with a supplemented daily dose of 200  $\mu\text{g}$  Se as Se yeast was reached after 6–9 months, and Clark *et al.* observed a plateau of approximately 190  $\mu\text{g}/\text{l}$  after one year and for the next 9 years of his observational period. When 300  $\mu\text{g}$  Se per day of the same standardised yeast (SelenoPrecise) was given to a UK cohort ( $n = 103$ ) for 6 months the mean plasma-Se was raised from  $92 \pm 20$   $\mu\text{g}/\text{l}$  to  $233 \pm 54$   $\mu\text{g}/\text{l}$  (11). In the present study, the plasma Se concentration was  $182 \pm 14$   $\mu\text{g}/\text{l}$  after 10 weeks supplementation with 300  $\mu\text{g}$  Se/d. When 0, 100, 200 or 300  $\mu\text{g}/\text{day}$  of the same standardised yeast was given to a Danish cohort ( $n = 49$ ) for two years, the Se-whole-blood response was linearly proportional to the total daily intake. The mean blood-Se level in the 300  $\mu\text{g}$  intake group, which was  $441 \pm 132$   $\mu\text{g}/\text{l}$ , was in steady state after 2 years supplementation (11). The blood-Se concentration was  $213 \pm 23$   $\mu\text{g}/\text{l}$ , in the present study, after 10 weeks administration of an identical Se-yeast. The 10 weeks pre-supplementation in the present study with 300  $\mu\text{g}$  Se/day is therefore not sufficiently long to fully attain steady state. This is likely to have affected the validity of the results.

In the present study, the mean  $^{77}\text{Se}$  plasma maximum concentration was  $9.77 \pm 1.53$   $\mu\text{g}/\text{l}$  where the median  $T_{\text{max}}$  was 8 hours and the mean  $T_{\text{max}}$  was found at 9.17 hours. The  $T_{\text{max}}$  results observed in our study were in agreement with a previously reported  $T_{\text{max}}$  at approximately 8 hours after supplementation of 100  $\mu\text{g}$   $^{74}\text{selenate}$  (18). Finley (18) found a plasma maximum concentration of approximately 5.3  $\mu\text{g}/\text{l}$ , which was considerably lower than the plasma maximum observed in the present study, but this is probably a reflection of the chemical form of Se given, as well as the dose. Higher plasma concentrations have been observed in studies feeding SeMeth, compared to other chemical forms of Se (31, 32).

The mean plasma Se concentration observed in this study was considerably higher than values reported in un-supplemented Danish subjects; 75–95  $\mu\text{g}/\text{l}$  (14, 33), but was closer to results reported in other studies, which reported plasma concentrations from 111–190  $\mu\text{g}/\text{l}$  in humans supplemented with 100–200  $\mu\text{g}$  Se yeast (3, 34). This confirms that the subjects did take the supplements.

A number of factors might have affected the results obtained in this study, among these are the fact that although our subjects' year-long high Se intakes, the Se species and amounts of intakes differed among the subjects and the 10 week run-in period obviously was

not sufficient to obtain steady-state. Furthermore, 8 of our subjects were current smokers and this as well might have affected the results as some studies have indicated that smoking might affect Se status (20). We think, however, that for the purpose of this study these factors have only minor effects.

### Conclusion

In conclusion, the absorption and three days retention in Danish men with a high habitual Se intake was found to be high as investigated by the stable enriched isotope approach using a single dose of <sup>77</sup>Se-labelled SelenoPrecise yeast.

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