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Identifying a target group for selenium supplementation in high-risk cardiac surgery: a secondary analysis of the SUSTAIN CSX trial

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Abstract

Background Recent data from the randomized SUSTAIN CSX trial could not confirm clinical benefits from perioperative selenium treatment in high-risk cardiac surgery patients. Underlying reasons may involve inadequate biosynthesis of glutathione peroxidase (GPx3), which is a key mediator of selenium's antioxidant effects. This secondary analysis aimed to identify patients with an increase in GPx3 activity following selenium treatment. We hypothesize that these responders might benefit from perioperative selenium treatment.

Methods Patients were selected based on the availability of selenium biomarker information. Four subgroups were defined according to the patient's baseline status, including those with normal kidney function, reduced kidney function, selenium deficiency, and submaximal GPx3 activity.

Results Two hundred and forty-four patients were included in this analysis. Overall, higher serum concentrations of selenium, selenoprotein P (SELENOP) and GPx3 were correlated with less organ injury. GPx3 activity at baseline was predictive of 6-month survival (AUC 0.73; $p = 0.03$). While selenium treatment elevated serum selenium and SELENOP concentrations but not GPx3 activity in the full patient cohort, subgroup analyses revealed that GPx3 activity increased in patients with reduced kidney function, selenium deficiency and low to moderate GPx3 activity. Clinical outcomes did not vary between selenium treatment and placebo in any of these subgroups, though the study was not powered to conclusively detect differences in outcomes.

Conclusions The identification of GPx3 responders encourages further refined investigations into the treatment effects of selenium in high-risk cardiac surgery patients.

Keywords Selenium, Glutathione peroxidase, Cardiac surgery, Critical care, Oxidative stress, SUSTAIN CSX

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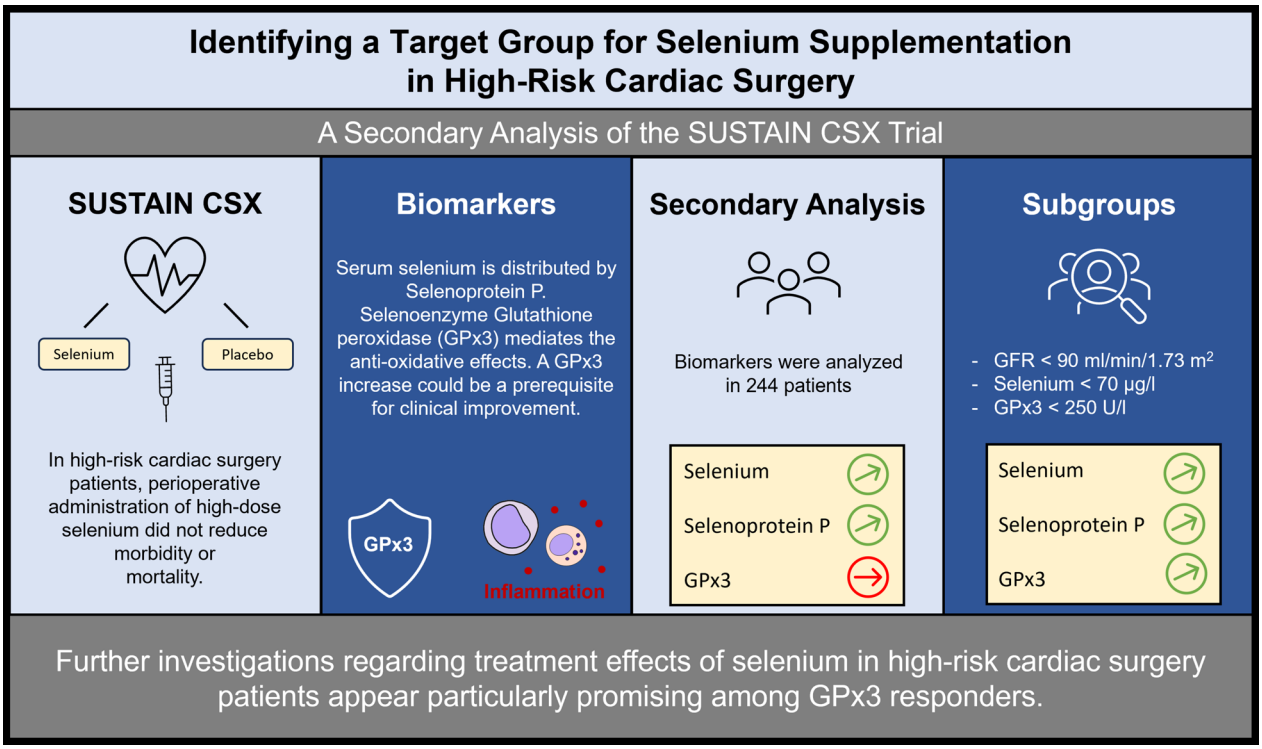
Graphical Abstract



Background

Cardiac surgery provokes a distinct systemic inflammatory response syndrome (SIRS), which has important implications for patients' mid- and long-term outcomes [1]. Perioperative inflammation and oxidative stress arise from iatrogenic tissue trauma, the use of cardiopulmonary bypass (CPB) and ischemia–reperfusion injury and may represent a modifiable risk factor for the development of organ dysfunction [2–4]. Although it has been a target of research for decades, the complex interactions of pro- and anti-inflammatory cytokines, their determinants, and their influence on the development of SIRS and subsequent organ dysfunction are currently not well understood. In the human body, several endogenous mechanisms specifically protect tissues and organs from reactive oxygen species and their sequelae. The essential trace element selenium is a cornerstone in human antioxidant defense mechanisms due to the pleiotropic anti-inflammatory and immunomodulatory properties of selenoproteins [5–7]. This consideration provides a compelling rationale for selenium treatment in high-risk situations to attenuate the inflammatory response following cardiac surgery and ultimately to improve clinical outcomes. Several observational studies demonstrated a significant intraoperative decrease in selenium levels, and subsequent interventional studies suggested benefits for patients' short-term outcomes after perioperative selenium supplementation [8, 9]. However, recent data from the multicenter, randomized, placebo-controlled Sodium Selenite Administration in Cardiac Surgery Trial (SUSTAIN CSX) challenged these findings and could not confirm any clinical benefits from intravenous selenium treatment in a broad population of high-risk patients [10]. Furthermore, blood analysis demonstrated that the perioperative provision of high-dose selenium led to a significant increase in serum selenium and selenoprotein P (SELENOP) levels but did not translate to an adequate downstream response, as selenium-dependent plasma glutathione peroxidase (GPx3) remained unaffected [10]. While SELENOP mainly serves as a selenium transporter, kidney-derived GPx3 catalyzes the neutralization of reactive oxygen species and represents the key mediator of the antioxidant capacities of selenium [11–13]. The missing increase in GPx3 activity might therefore explain the absence of measurable clinical effects. Consequently, the objective of this secondary analysis was to identify and characterize subgroups of patients in whom GPx3 activity increases in response to selenium treatment. We

hypothesize that these responders might benefit from perioperative selenium treatment.

Methods

SUSTAIN CSX overview

This was an a priori defined secondary analysis of the international, double-blind, randomized, placebo-controlled SUSTAIN CSX trial (NCT02002247), which was conducted at 23 centers in Canada and Germany between 2015 and 2021 [10]. Adult patients undergoing elective or urgent cardiac surgery with the use of CPB were eligible if the European System for Cardiac Operative Risk Evaluation II (EuroSCORE II) predicted an operative mortality risk of at least 5% or if combined surgical procedures were scheduled. Patients were randomly assigned to receive either selenium or placebo before surgery and postoperatively throughout the ICU stay [10]. There were no significant differences between the treatment and placebo groups with regard to primary or secondary endpoints, and the results did not identify any clinical benefits of selenium supplementation in high-risk cardiac surgery patients.

Patient selection and subgroup analyses

Participation in this nested substudy of SUSTAIN CSX was optional for the sites and within the framework of the existing ethical approval [14]. Blood samples were collected, and selenium, SELENOP and GPx3 activity were measured as described [10]. Patients were selected for the current secondary analysis if information on their respective biomarkers was available at any time point. Four subgroups were defined to identify patients who responded to high-dose selenium with an increase in GPx3 activity. These subgroups were as follows:

I. Patients with normal renal function (GFR ≥ 90 ml/min/1.73 m^2): As the kidneys are the primary source of GPx3, this subgroup may have optimal conditions for an adequate GPx3 response [15].
II. Patients with reduced kidney function (GFR < 90 ml/min/1.73 m^2): A common finding in organ injury is increased resistance to regulatory stimuli [16]. This resistance might be overcome by high-dose selenium supplementation and result in a GPx3 increase. The threshold of 90 ml/min/1.73 m^2 has been chosen in accordance with KDIGO guidelines.
III. Patients with selenium deficiency (serum selenium < 70 µg/l): Selenium supplementation might be most effective when serum levels are low and tissues are poorly supplied [7].
IV. Patients with submaximal baseline activity of GPx3 (GPx3 < 250 U/l): In the literature, a threshold of 250 U/l has been established as the upper reference limit for GPx3 to distinguish patients with moderate and high activity. This cutoff value also corresponded to the average GPx3 activity of the present patient population at baseline. By excluding all patients with a priori elevated GPx3 activity from this subgroup, the maximum potential for optimization might be ensured [10, 17].

Quantification of selenium biomarkers

Patient samples were analyzed at the Institute for Experimental Endocrinology (Charité Berlin, Germany) as recently described [18–20]. Total reflection X-ray fluorescence spectroscopy (S4 T-STAR, Bruker Nano GmbH, Berlin, Germany) was used to determine selenium concentrations. Selenium deficiency was defined as the presence of baseline serum selenium levels < 70 µg/l. For quantification of SELENOP, a commercial enzyme-linked immunosorbent assay kit (selenOtest ELISA, selenOmed GmbH, Berlin, Germany) was used. The activity of GPx3 was assessed via the consumption of nicotinamide adenine dinucleotide phosphate (NADPH) at 340 nm in a coupled enzymatic assay using hydrogen peroxide as a substrate [21]. Reference ranges were used as published elsewhere [22, 23]

Statistical analyses

The normality of the data was not assumed, and nonparametric testing was applied throughout the manuscript. A p value < 0.05 was considered statistically significant (asterisk *), and a p value < 0.10 was considered a potential trend (wave ≈). Categorical variables were expressed as absolute numbers and percentages, while continuous variables were presented as the median and interquartile range (IQR, 25–75%). To account for the clustering of sites, categorical outcomes were analyzed by a logistic generalized linear mixed effects model, while continuous outcomes were analyzed by generalized estimating equations. Outcome statistics (Additional file 1: S1, S3–S6) were either reported as odds ratios (ORs), hazard ratios (HRs) or mean differences with 95% confidence intervals (CIs). Mann–Whitney U tests were used for further group comparisons.

The correlation coefficient (rho, r) quantified associations between continuous variables according to Spearman. To estimate predictive values with regard to survival, receiver operating characteristic (ROC) analyses were conducted and areas under the curves (AUCs) were computed. Statistical analyses were performed using GraphPad Prism® Version 9.5 (GraphPad Software, San Diego, USA) and SAS® Version 9.4 (SAS Institute, Cary, USA).

Results

Baseline characteristics and outcomes

Selenium biomarkers were available in $n=244$ high-risk cardiac surgery patients who were recruited at 11 participating sites (Fig. 1). Baseline characteristics and outcomes did not differ between the treatment and placebo groups (Table 1; Additional file 1: S1). Five percent of patients died before ICU discharge, and a total of 8% within a 6-month period. Selenium levels at baseline were significantly higher in patients from Canada than in patients from Germany (150, 123–167 µg/l versus 60, 51–74 µg/l; $p<0.0001$; Additional file 1: S2).

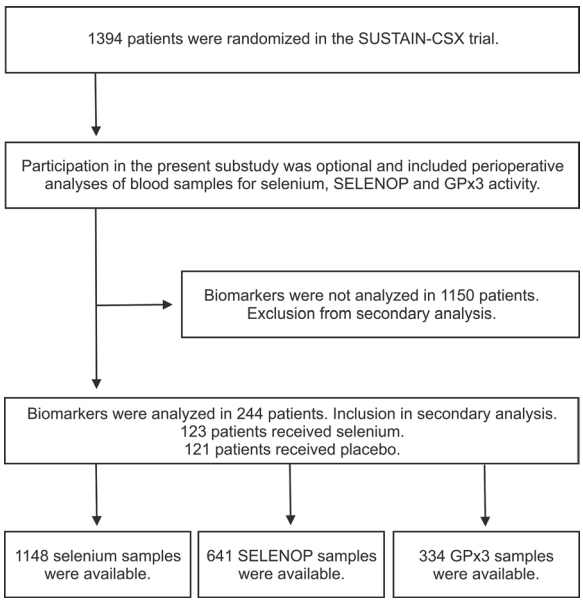


Fig. 1 Flow diagram of patient selection. *SELENOP* selenoprotein P, *GPx3* glutathione peroxidase 3

Perioperative levels of selenium biomarkers

Baseline serum selenium was below the reference range in half of the patients, while preoperative SELENOP was mostly adequate. Both biomarkers were significantly increased in the treatment group and remained unaffected in patients receiving placebo. The median GPx3 activity was not affected by selenium treatment, as values did not differ between the groups throughout the observation period (Fig. 2A). ICU admission and day one after cardiac surgery were characterized by decreased selenium (placebo group only), SELENOP and GPx3 levels in comparison to baseline and a distinct inflammatory reaction, as reflected by a pronounced spike in interleukin-6 (IL-6) (Fig. 2B). Selenium and SELENOP were closely correlated throughout the study period (e.g., baseline: $r=0.70$; $p<0.0001$), whereas GPx3 activity showed

Table 1 Demographics

	Selenium ($n=123$)	Placebo ($n=121$)	*p*
Baseline characteristics			
Age (years)—median (IQR)	70 (60–76)	71 (64–77)	0.17
Female/male—no. (%)	29 (24)/94 (76)	25 (21)/96 (79)	0.58
Germany/Canada—no. (%)	93 (76)/30 (24)	92 (76)/29 (24)	0.94
Body mass index—median (IQR)	27 (25–30)	28 (25–30)	0.95
Charlson comorbidity index—median (IQR)	1 (0–2)	1 (0–2)	0.41
Clinical frailty score—median (IQR)	3 (2–3)	3 (2–3)	0.34
Baseline chemistry			
GFR (ml/min/1.73 m^2)—median (IQR)	82 (68–97)	81 (69–97)	0.85
Creatinine (µmol/l)—median (IQR)	84 (73–101)	86 (72–103)	0.94
Interleukin-6 (pg/ml)—median (IQR)	44 (32–49)	36 (22–62)	0.92
Operative characteristics			
EuroSCORE II—median (IQR)	9 (6–16)	8 (6–14)	0.45
Elective/urgent surgery—no. (%)	96 (78)/27 (22)	97 (80) / 24 (20)	0.68
Cardiopulmonary bypass (min)—median (IQR)	140 (113–178)	141 (102–190)	0.90
Baseline micronutrient status			
Selenium (µg/l)—median (IQR)	68 (55—114)	65 (52–119)	0.67
Deficiency (< 70 µg/l)—no. (%)	64 (52)	64 (53)	0.73
SELENOP (mg/l)—median (IQR)	4.5 (3.4–5.7)	4.7 (3.5–6.0)	0.75
GPx3 (U/l)—median (IQR)	273 (187–299)	253 (204–289)	0.62

GFR glomerular filtration rate, *GPx3* glutathione peroxidase 3, *IQR* interquartile range, *no.* number of patients, *SELENOP* selenoprotein P

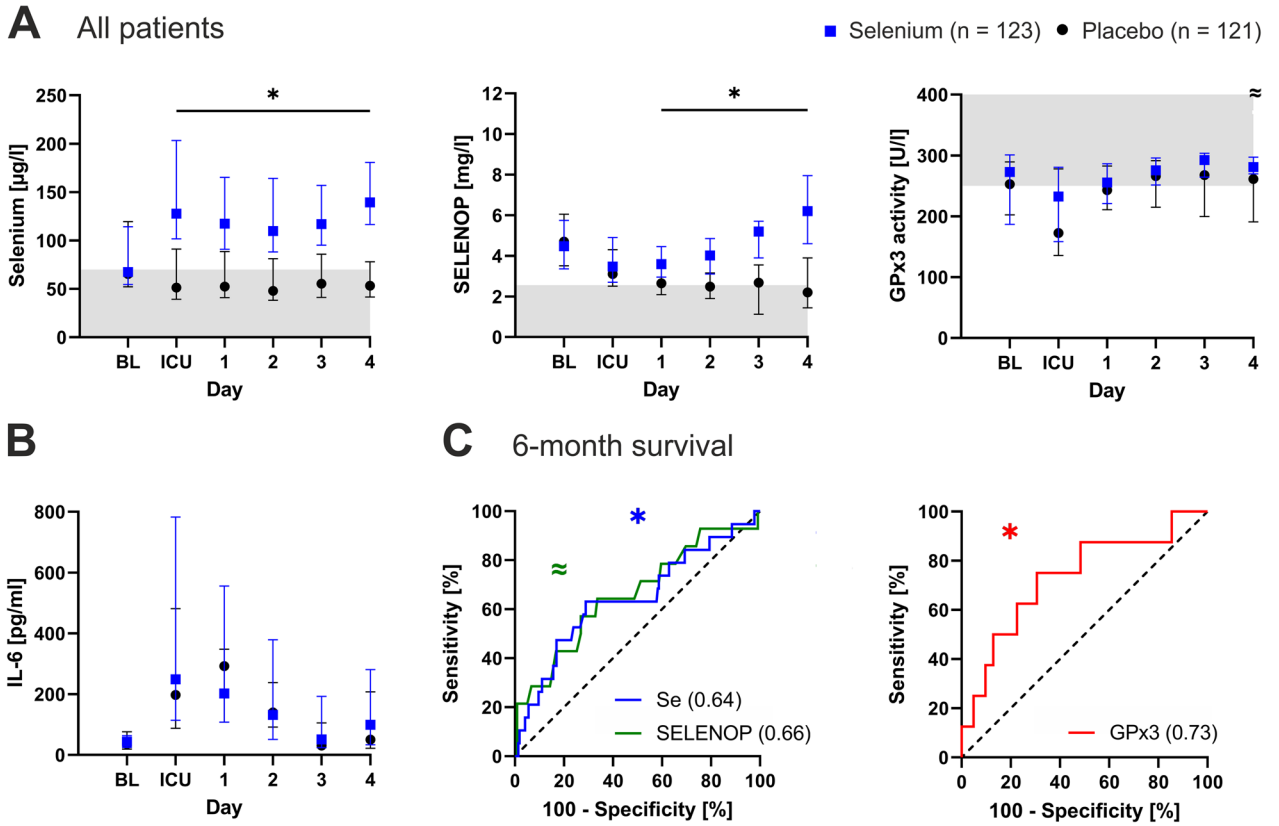


Fig. 2 Perioperative selenium status and inflammation. **A** Selenium treatment (blue) increased serum selenium and selenoprotein P (SELENOP) concentrations compared to placebo (black). However, this increase did not translate to altered glutathione peroxidase 3 (GPx3) activity. Gray coloration indicates values below the lower reference limit (selenium and SELENOP) and above the upper reference limit (GPx3). Reference ranges were used as published elsewhere. **B** Interleukin- (IL-) 6 levels reflect a significant inflammatory reaction following cardiac surgery. **C** Receiver operating characteristic (ROC) analyses on the predictive potential of the three selenium biomarkers at baseline regarding 6-month survival after cardiac surgery. Areas under the curves (AUCs) are listed in brackets. GPx3 outperformed the other two biomarkers of selenium status. Asterisks (*) indicate statistical significance, and waves (≈) indicate a trend. *BL* baseline, *ICU* intensive care unit

only a weak association with selenium ($r=0.23$; $p=0.06$) and was not correlated with SELENOP concentrations ($r=-0.09$; $p=0.45$).

Association of selenium biomarkers and clinical outcomes

An explorative correlation analysis revealed multiple significant associations between perioperative selenium biomarkers, clinical status, ICU course and patient-centered outcomes. Overall, higher selenium, SELENOP and GPx3 levels were correlated with less organ injury and a better patient outcome; however, it is important to note that these correlations were of weak and moderate strength, respectively (Fig. 3).

Low GPx3 activity was correlated with older age and reduced kidney function. Low GPx3 activity was also related to a higher score in the sequential organ failure assessment (SOFA), a longer duration of mechanical ventilation and consequently an extended duration to ICU discharge alive.

Regarding long-term outcomes, adequate pre- and postoperative selenium indicated improved functional outcome measures after 3 months. Six-month survivors had higher selenium (67, 55–119 µg/l versus 54, 45–79 µg/l; $p=0.04$), SELENOP (4.7, 3.5–6.0 mg/l versus 3.7, 2.3–5.3 mg/l; $p=0.05$) and GPx3 levels (273, 208–296 U/l versus 193, 166–256 U/l; $p=0.03$) at baseline than nonsurvivors. To assess the predictive potential of baseline biomarkers for 6-month survival after cardiac surgery, ROC curve analyses were performed. All three biomarkers were related to survival, with GPx3 activity (AUC 0.73; $p=0.03$) outperforming serum selenium (AUC 0.64; $p=0.04$) and SELENOP (AUC 0.66; $p=0.05$) concentrations (Fig. 2C).

GPx3 activity in patients with normal versus impaired kidney function (subgroups I and II)

Subgroup I comprised $n=81$ patients who had a glomerular filtration rate (GFR) of 90 ml/min/1.73 m^2 or

	Selenium			SELENOP			GPx3		
	BL	ICU	Day 1	BL	ICU	Day 1	BL	ICU	Day 1
Age							-0.25	-0.21	-0.20
							0.04	0.08	0.09
Glomerular filtration rate							0.28		
							0.03		
Urine output							0.24	0.28	0.21
							0.04	0.02	0.09
PODS-free days	0.16	0.12	0.17			0.20			
	0.01	0.06	0.01			0.02			
SOFA score	-0.31	-0.26	-0.22	-0.23	-0.28	-0.28	-0.22	-0.34	-0.28
	0.0001	0.0001	0.0008	0.009	0.001	0.002	0.06	0.004	0.02
Mechanical ventilation (d)	-0.20	-0.14	-0.15			-0.15	-0.21		
	0.002	0.03	0.03			0.08	0.08		
ICU discharge alive (d)							-0.29	-0.21	
							0.02	0.08	
Activities of daily living	0.38	0.28	0.24	0.28	0.29	0.26			
	0.0001	0.0001	0.0006	0.004	0.003	0.008			
SF 36 mental	0.19	0.18	0.14						
	0.009	0.02	0.05						
SF 36 physical	0.21	0.21	0.19	0.18	0.28	0.23			
	0.003	0.003	0.009	0.07	0.005	0.02			

Fig. 3 Association between selenium biomarkers and organ injury. Correlation matrix of selenium biomarkers and continuous clinical (outcome) parameters at baseline (BL), admission to the intensive care unit (ICU) and postoperative day one. Positive associations are shown in red, inverse associations in blue. The strength of the relationship is depicted by color graduation. Two values specify each correlation: Spearman's rho in the upper row and the p value in the lower row. Only significant p values and trends are shown. No relations were observed in the blank fields. The datasets are different in size for each biomarker, with selenium having the most pairs and GPx3 having the fewest. Overall, the associations were weak to moderate. It is important to note that these correlations do not imply causation. *PODS* persistent organ dysfunction and death, *SOFA* sequential organ failure assessment, *SF 36* short-form 36 questionnaire

above at baseline. Biomarker levels exhibited the same dynamics as described above, and GPx3 activity did not increase in the treatment group compared to the placebo group (Fig. 4A). Furthermore, no significant outcome differences between the groups were observed (Additional file 1: S3).

Subgroup II included $n=137$ patients with impaired kidney function and a GFR below 90 ml/min/1.73 m^2 at baseline. Here, selenium supplementation not only increased serum selenium and SELENOP concentrations, but also GPx3 activity at Day 4 (Fig. 4B). Regarding clinical outcomes in this subgroup, there were no major differences between the treatment and placebo groups. However, the physical domain of the Short Form 36 questionnaire after 3 months tended to be higher with selenium supplementation (46, 41–52 versus 43, 38–51; $p=0.05$; Additional file 1: S4).

Overall, GPx3 activity was higher in patients with normal kidney function than in patients with renal impairment (291, 214–309 U/l versus 224, 185–290 U/l; $p=0.04$).

GPx3 activity in patients with selenium deficiency (subgroup III)

Subgroup III comprised all patients with selenium deficiency (< 70 µg/l, $n=128$) at baseline. Again, selenium treatment elevated serum selenium and SELENOP. In this subgroup, selenium supplementation additionally translated to increased GPx3 activity in comparison to the placebo group starting at postoperative Day 2 (Fig. 5A). However, the evaluated outcomes did not show statistically significant differences between the groups (Additional file 1: S5).

Compared to subgroup III, the patients with preoperative selenium levels ≥ 70 µg/l ($n=109$) also had higher GPx3 activity at baseline (279, 211–297 U/l versus 226, 178–293; $p=0.05$). Here, GPx3 did not differ between patients receiving selenium supplementation and patients receiving placebo throughout the course of intensive care (Fig. 5C).

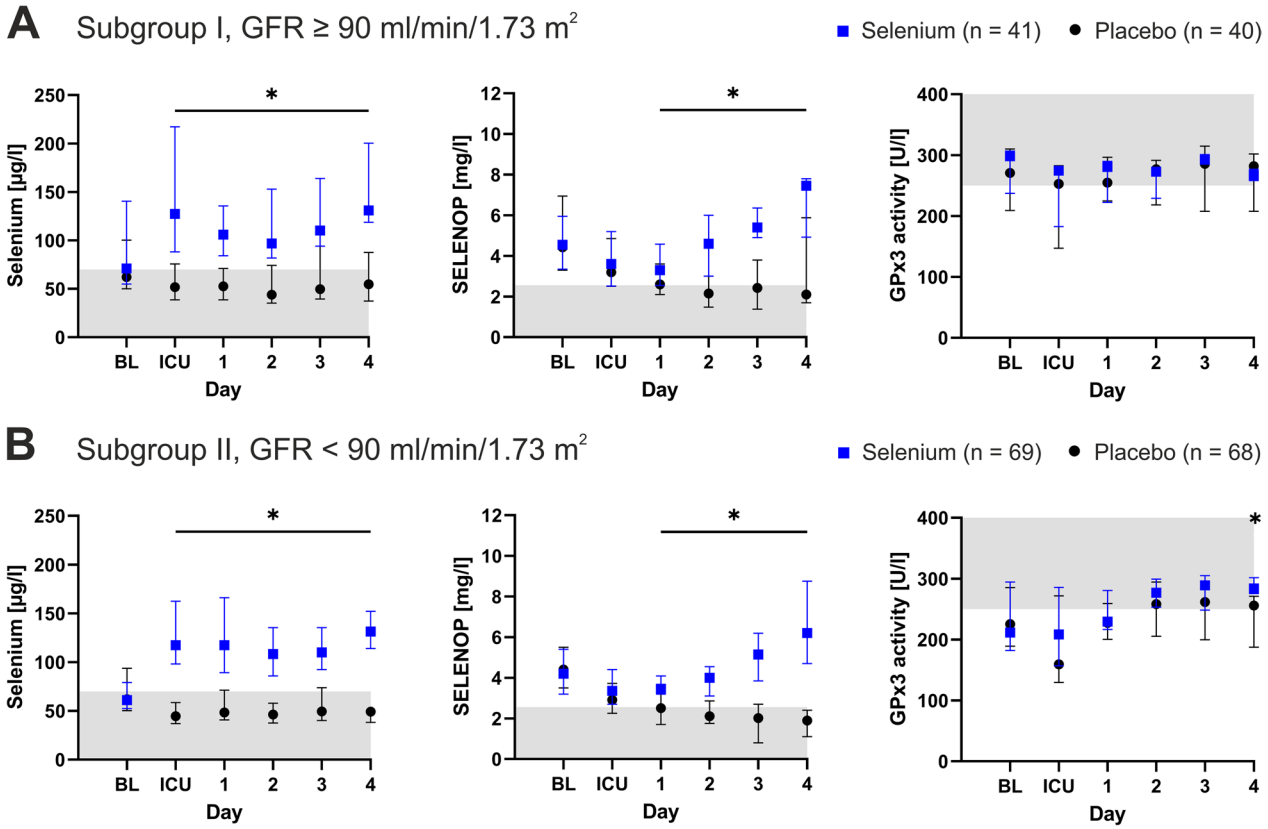


Fig. 4 Subgroup I and II. **A** Selenium treatment (blue) increased serum selenium and selenoprotein P (SELENOP) but not glutathione peroxidase 3 (GPx3) activity in patients with normal kidney function according to the KDIGO definition (I). **B** Selenium supplementation in patients with reduced renal function led to an increase in serum selenium, SELENOP and GPx3 at Day 4 (II). Gray coloration indicates values below the lower reference limit (selenium and SELENOP) and above the upper reference limit (GPx3). Reference ranges were used as published elsewhere. Asterisks (*) indicate statistical significance. *BL* baseline, *GFR* glomerular filtration rate, *ICU* intensive care unit

GPx3 activity in patients with submaximal baseline GPx3 levels (subgroup IV)

Subgroup IV consisted of patients with GPx3 baseline activity below 250 U/l ($n=32$). Selenium treatment triggered an adequate biological response, including an increase in serum selenium, SELENOP and GPx3 (Fig. 5B). GPx3 optimization was achieved until Day 2, when a stable plateau was reached. Similarly, the GPx3 activity of patients receiving placebo remained below 250 U/l throughout the observation period. Again, the outcome analysis did not reveal significant differences between the groups (Additional file 1: S6).

In contrast to subgroup IV, the patients with baseline GPx3 activity ≥ 250 U/l ($n=38$) were not affected by selenium supplementation. Their GPx3 activity constantly remained unchanged at the plateau level (Fig. 5C).

Discussion

The SUSTAIN CSX intervention trial is currently the most comprehensive study on selenium supplementation in high-risk cardiac surgery patients [10].

Consistent with the original report, intravenous selenium supplementation elevated serum selenium and SELENOP concentrations in the present secondary analysis. The increase of SELENOP ensured efficient uptake and metabolism of the supplemental trace element in the liver and further indicated an improved transport and distribution capacity of selenium throughout the body [24, 25]. Surprisingly, GPx3 failed to display a similar positive response to selenium supplementation and enhanced SELENOP status, with clinical outcomes being similar between the intervention and placebo groups. Closer investigation revealed a transient decline in all selenium biomarkers and a

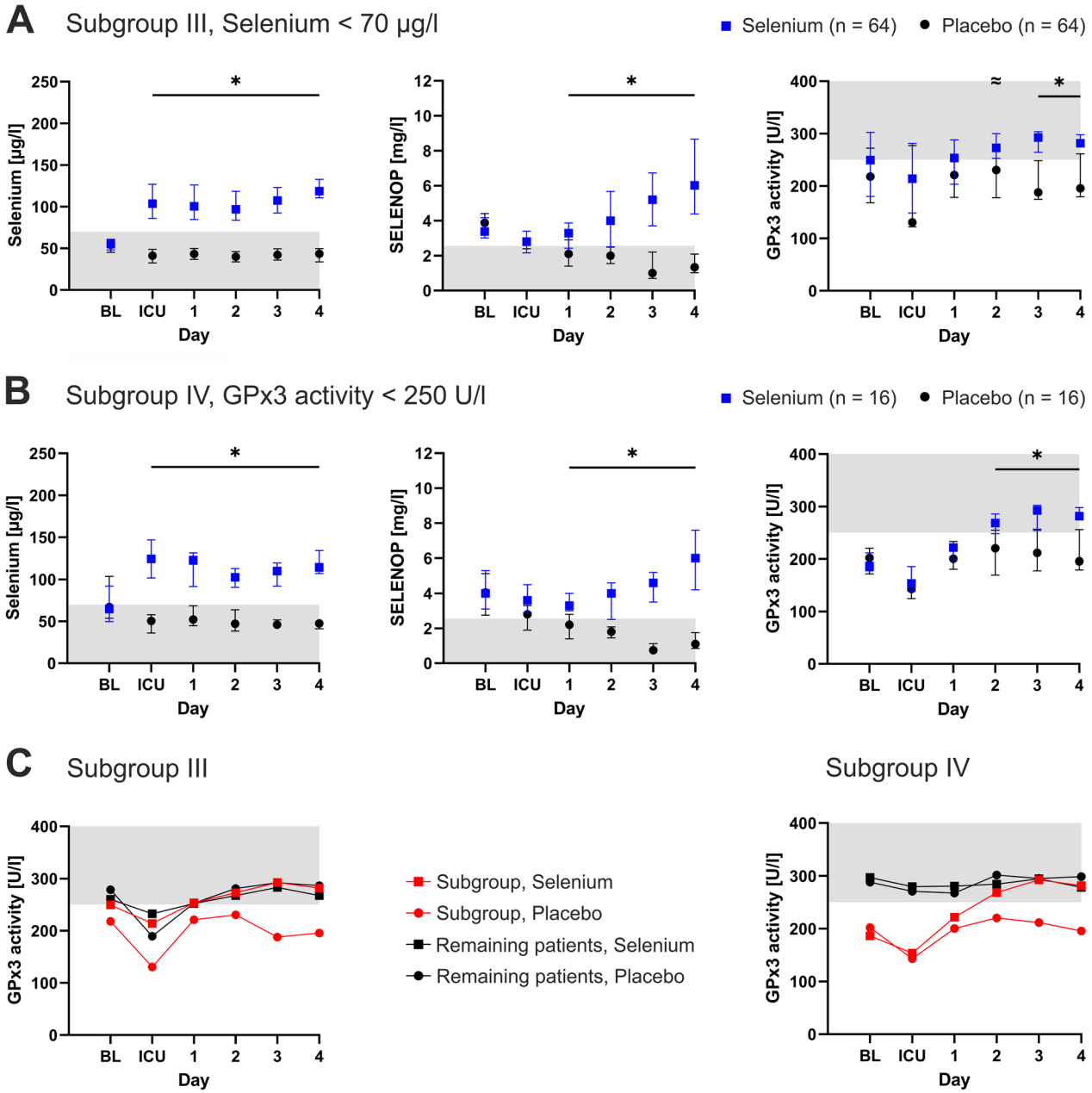


Fig. 5 Subgroup III and IV. **A** In patients with selenium deficiency at baseline (defined as values < 70 µg/l), selenium treatment (blue) increased serum selenium, selenoprotein P (SELENOP) and glutathione peroxidase 3 (GPx3) activity (III). **B** In patients with GPx3 activity < 250 U/l at baseline, selenium treatment again successfully increased all biomarkers, including GPx3 (IV). **C** Subgroups III and IV are contrasted with the respective remainder of each patient population. High GPx3 activity remained unchanged throughout the observation period independent of selenium supplementation. For better presentation, error bars were removed, and connecting lines were inserted. Gray coloration indicates values below the lower reference limit (selenium and SELENOP) and above the upper reference limit (GPx3). Reference ranges were used as published elsewhere. Asterisks (*) indicate statistical significance, and waves (≈) indicate a trend. *BL* baseline, *ICU* intensive care unit

simultaneous peak in inflammatory IL-6 upon surgery, which may indicate the consumption of antioxidant capacity during and after cardiac surgery [9, 26]. Not only did selenium and especially GPx3 activity correlate with kidney function, organ dysfunction and ICU length of stay, but GPx3 activity also emerged as a good predictive marker for long-term survival. This is in line with a recent study, that proposed an early GPx3-based prediction model for acute kidney injury after cardiac surgery [27].

Several subgroups were studied in this secondary investigation to facilitate the identification of a particular responder phenotype of high-risk cardiac surgery patients.

First, selenium biomarkers were analyzed with regard to current renal function, as the renal proximal convoluted tubules are the primary source of GPx3 (Subgroups I and II). As expected, GPx3 correlated with GFR and urine output and was low in patients with impaired kidney function. While selenium supplementation did not lead to increased GPx3 production in patients with normal kidney function, it gradually elevated GPx3 in patients with reduced kidney function. In this subgroup, a trend toward a better functional status after 3 months was observed for selenium treatment in comparison to placebo. While the exact reasons for these findings are unclear, chronic disease is often accompanied by increased resistance to finely adjusted regulatory pathways (e.g., treatment-resistant hypertension in kidney disease, insulin-resistance in metabolic syndrome) [28]. It seems conceivable that chronic renal impairment may elevate the threshold to induce a GPx3 response and that high-dose selenium supplementation could sufficiently provide the requisite signal in comparison to placebo.

Next, supplementation was analyzed in the context of selenium deficiency (Subgroup III). For ICU patients, strong correlations between selenium and GPx3, as well as favorable effects of selenium supplementation have been established [29–33]. Those patients were usually characterized by significantly decreased selenium levels due to SIRS and multiorgan failure [34]. In a previous supplementation trial in critical illness, Angstwurm et al. observed a significant GPx3 increase following selenium treatment in patients with sepsis and septic shock [35]. In line with these results, the present subgroup of patients with preoperative selenium deficiency exhibited an adequate treatment response to selenium supplementation. Nevertheless, clinical outcomes did not differ between the groups, despite previous data considering low levels of selenium as a relevant preoperative risk factor [36, 37].

Finally, the potential for GPx3 optimization was considered the largest in patients with low and moderate GPx3 activity at baseline (Subgroup IV). In healthy subjects, circulating SELENOP and GPx3 exhibited saturation kinetics as a function of serum selenium levels [38–40]. Similarly, a plateau of GPx3 activity was observed in the present cardiac surgery population. This might constitute a biological limit under perioperative conditions, where a surplus of selenium could not be translated into more antioxidant activity. Consequently, patients with above-average GPx3 activity at baseline might only have a small biological margin for improvement. In agreement with this hypothesis, selenium supplementation in Subgroup IV led to a significant increase in SELENOP and GPx3 activity. However, outcomes again did not differ between selenium treatment and placebo.

In summary, subgroup analyses revealed that patients with GFR below 90 ml/min/1.73 m^2, selenium below 70 µg/l and GPx3 activity below 250 U/l at baseline were likely to respond to selenium supplementation with an increase in GPx3 activity, which may be a prerequisite for clinical improvement. A common property of all three subgroups was submaximal GPx3 activity at baseline, which can therefore be considered a treatable trait. This concept refers to a clinically important patient characteristic with direct physiological implications and a targeted approach for individualized treatment intervention [41, 42]. In the future, preoperative GPx3 measurement as well as the identification of specific risk constellations, including factors such as advanced age and the knowledge of the above defined subgroups, may help to improve the precision and success of adjuvant high-dose selenium treatment.

Despite these promising findings, the clinical relevance of adequate biological downstream translation could not be shown in the present study. Correlation analyses do not allow conclusions about causation, and outcome differences between GPx3 responders and nonresponders were not found. On the one hand, our dataset was not powered to reliably detect outcome differences. On the other hand, in vitro and clinical data, as well as the present observations, suggest that at least a few days of selenium administration are required to stimulate a significant increase in GPx3 activity [9, 43]. One could therefore argue that outcome-relevant GPx3 optimization could not be achieved due to the dosing regimen of the SUSTAIN CSX trial: the first selenium dose was administered shortly prior to CPB and the inflammatory insult, leaving little time for selenoprotein biosynthesis and tissue protection. In fact, a previous study suggested that early initiation of micronutrient supplementation several weeks before elective cardiac surgery improved redox status and shortened the length of hospital stay [44].

Other limitations of the current secondary analysis include the small number of participating patients and the limited amount of biomarker datapoints, which render our investigation strictly hypothesis generating. For this reason, a subgroup analysis of patients with severe selenium deficiency below 45 µg/l could not be performed, despite being highly relevant. In addition, GPx3 regulatory mechanisms are far more complex than selenium status alone, including peroxisome proliferator-activated receptor signaling and hypoxia-inducible as well as other transcription factors [45]. In this context, a definite GPx3 target for optimal organ protection has

not been established yet. In this study, the hierarchy of selenoproteins was simplified, as GPx3 is only one member of a large family of selenium-dependent enzymes and GPx isoforms [13]. However, it can be argued that SELENOP and GPx3, as the only actively secreted extracellular selenoproteins, account for the majority of circulating selenium in the serum and bear the most clinical relevance [17, 45].

Conclusions

A GPx3 increase in response to selenium treatment was observed in patients with GFR below 90 ml/min/1.73 m^2, selenium below 70 µg/l and GPx3 activity below 250 U/l at baseline. These characteristics might constitute a specific responder phenotype, where further investigations regarding a potential positive treatment effect of selenium supplementation seem promising.

Take-home message

This secondary analysis of the SUSTAIN CSX trial identified different subgroups of GPx3 responders following selenium treatment. The baseline characteristics of these patients included impaired renal function, serum selenium deficiency and submaximal GPx3 activity. Although clinical differences between selenium treatment and placebo could not be observed in any subgroup, further studies may build on these findings to define a specific responder phenotype that may be susceptible to anti-inflammatory treatment strategies with high-dose selenium.

Abbreviations

BL	Baseline
GFR	Glomerular filtration rate
GPx3	Glutathione peroxidase 3
ICU	Intensive care unit
IL	Interleukin
PODS	Persistent organ dysfunction and death
SELENOP	Selenoprotein P
SF 36	Short-form 36 Questionnaire
SOFA	Sequential Organ Failure Assessment

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s40635-023-00574-8.

Additional file 1: **S1**: Outcomes in all patients. **S2**: Baseline selenium levels in Canada and Germany. **S3**: Baseline characteristics and outcomes in patients with baseline glomerular filtration rate (GFR) ≥ 90 ml/min/1.73 m^2. **S4**: Baseline characteristics and outcomes in patients with baseline glomerular filtration rate (GFR) < 90 ml/min/1.73 m^2. **S5**: Baseline characteristics and outcomes in patients with selenium baseline levels < 70 µg/l. **S6**: Baseline characteristics and outcomes in patients with glutathione peroxidase (GPx3) baseline activity < 250 µg/l.

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Author contributions

All authors read and approved the final manuscript. Conceptualization: QN, DKH, LS, CS; methodology: QN, ZYL, PM, LS, CS; acquisition and analysis: QN, DKH, ZYL, TC, SM, GE, LS, CS; interpretation of data: QN, ZYL, GE, SO, MKB, PM, LS, CS; writing—original draft: QN, LS, CS; writing—review: DKH, ZYL, JM, JH, SF, GE, CDM, AH, MV, SO, MKB, PM.

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Availability of data and materials

The datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

Declarations

Ethics approval and consent to participate

The ethics approval of the SUSTAIN CSX trial applies for this nested substudy. The original protocol was approved by the ethics committees of Queens University, Canada, and RWTH Aachen University, Germany, the German Federal Institute for Drugs and Medical Devices, and by all participating centers. Each patient gave written informed consent to participate in the study before surgery.

Consent for publication

Not applicable.

Competing interests

Outside of this work, the authors disclose the following relationships: SF: payments to the institution (Canadian Institutes of Health Research, National Institute of Health, Medtronic, Boston Scientific, Amgen), participation on advisory board (PolyPid). GE: honoraria and travel support (Fresenius Kabi, Baxter). AH: grants (Deutsche Forschungsgemeinschaft, Fresenius Kabi, Pascoe), honoraria and travel support (Fresenius Kabi, Baxter). LS: grants (Deutsche Forschungsgemeinschaft), pending patent application for selenium status analysis, shareholder (selenOmed GmbH). CS: consulting fees and honoraria (Baxter, Fresenius, BBRAUN). The other authors have nothing to disclose.

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References

1. Laffey JG, Boylan JF, Cheng DC (2002) The systemic inflammatory response to cardiac surgery: implications for the anesthesiologist. Anesthesiology 97:215–252
2. Squiccimarro E, Stasi A, Lorusso R, Paparella D (2022) Narrative review of the systemic inflammatory reaction to cardiac surgery and cardiopulmonary bypass. Artif Organs 46:568–577
3. Corral-Velez V, Lopez-Delgado JC, Betancur-Zambrano NL, Lopez-Suñe N, Rojas-Lora M, Torrado H, Ballus J (2015) The inflammatory response in cardiac surgery: an overview of the pathophysiology and clinical implications. Inflamm Allergy Drug Targets 13:367–370
4. Motoyama T, Okamoto K, Kukita I, Hamaguchi M, Kinoshita Y, Ogawa H (2003) Possible role of increased oxidant stress in multiple organ failure after systemic inflammatory response syndrome. Crit Care Med 31:1048–1052
5. Avery JC, Hoffmann PR (2018) Selenium, selenoproteins, and immunity. Nutrients 10:1203
6. Hariharan S, Dharmaraj S (2020) Selenium and selenoproteins: it's role in regulation of inflammation. Inflammopharmacology 28:667–695
7. Rayman MP (2012) Selenium and human health. Lancet (London, England) 379:1256–1268
8. Stoppe C, Schälte G, Rossaint R, Coburn M, Graf B, Spillner J, Marx G, Rex S (2011) The intraoperative decrease of selenium is associated with the postoperative development of multiorgan dysfunction in cardiac surgical patients. Crit Care Med 39:1879–1885
9. Stoppe C, Spillner J, Rossaint R, Coburn M, Schälte G, Wildenhues A, Marx G, Rex S (2013) Selenium blood concentrations in patients undergoing elective cardiac surgery and receiving perioperative sodium selenite. Nutrition 29:158–165
10. Stoppe C, McDonald B, Meybohm P, Christopher KB, Fremes S, Whitlock R, Mohammadi S, Kalavrouziotis D, Elke G, Rossaint R, Helmer P, Zacharowski K, Günther U, Parotto M, Niemann B, Böning A, Mazer CD, Jones PM, Ferner M, Lamarche Y, Lamontagne F, Liakopoulos OJ, Cameron M, Müller M, Zarbock A, Wittmann M, Goetzenich A, Kilger E, Schomburg L, Day AG, Heyland DK (2023) Effect of high-dose selenium on postoperative organ dysfunction and mortality in cardiac surgery patients: the SUSTAIN CSX randomized clinical trial. JAMA Surg 158:235
11. Manzanares W, Langlois PL, Heyland DK (2015) Pharmaconutrition with selenium in critically ill patients: what do we know? Nutr Clin Pract 30:34–43
12. Stevanovic A, Coburn M, Menon A, Rossaint R, Heyland D, Schälte G, Werker T, Wonisch W, Kiehntopf M, Goetzenich A, Rex S, Stoppe C (2014) The importance of intraoperative selenium blood levels on organ dysfunction in patients undergoing off-pump cardiac surgery: a randomised controlled trial. PLoS ONE 9:e104222
13. Pei J, Pan X, Wei G, Hua Y (2023) Research progress of glutathione peroxidase family (GPX) in redoxidation. Front Pharmacol 14:1147414
14. Stoppe C, McDonald B, Rex S, Manzanares W, Whitlock R, Fremes S, Fowler R, Lamarche Y, Meybohm P, Haberthür C, Rossaint R, Goetzenich A, Elke G, Day A, Heyland DK (2014) SodiUm SeleniTe Administration IN Cardiac Surgery (SUSTAIN CSX-trial): study design of an international multicenter randomized double-blinded controlled trial of high dose sodium-selenite administration in high-risk cardiac surgical patients. Trials 15:339
15. Bierl C, Voetsch B, Jin RC, Handy DE, Loscalzo J (2004) Determinants of human plasma glutathione peroxidase (GPx-3) expression. J Biol Chem 279:26839–26845
16. Artunc F, Schleicher E, Weigert C, Fritsche A, Stefan N, Häring HU (2016) The impact of insulin resistance on the kidney and vasculature. Nat Rev Nephrol 12:721–737
17. Demircan K, Bengtsson Y, Sun Q, Brange A, Vallon-Christersson J, Rijntjes E, Malmberg M, Saal LH, Rydén L, Borg Å, Manjer J, Schomburg L (2021) Serum selenium, selenoprotein P and glutathione peroxidase 3 as predictors of mortality and recurrence following breast cancer diagnosis: a multicentre cohort study. Redox Biol 47:102145
18. Demircan K, Chillon TS, Bracken T, Bulgarelli I, Campi I, Du Laing G, Fafi-Kremer S, Fugazzola L, Garcia AA, Heller R, Hughes DJ, Ide L, Klingenberg GJ, Komarnicki P, Krasinski Z, Lescure A, Mallon P, Moghaddam A, Persani L, Petrovic M, Ruchala M, Solis M, Vandekerckhove L, Schomburg L (2022) Association of COVID-19 mortality with serum selenium, zinc and copper: six observational studies across Europe. Front Immunol 13:1022673
19. Moghaddam A, Heller RA, Sun Q, Seelig J, Cherkezov A, Seibert L, Hackler J, Seemann P, Diegmann J, Pilz M, Bachmann M, Minich WB, Schomburg L (2020) Selenium deficiency is associated with mortality risk from COVID-19. Nutrients 12:2098
20. Heller RA, Sun Q, Hackler J, Seelig J, Seibert L, Cherkezov A, Minich WB, Seemann P, Diegmann J, Pilz M, Bachmann M, Ranjbar A, Moghaddam A, Schomburg L (2021) Prediction of survival odds in COVID-19 by zinc, age and selenoprotein P as composite biomarker. Redox Biol 38:101764
21. Flohé L, Günzler WA (1984) Assays of glutathione peroxidase. Methods Enzymol 105:114–121
22. Bomer N, Grote Beverborg N, Hoes MF, Streng KW, Vermeer M, Dokter MM, IJmker J, Anker SD, Cleland JGF, Hillege HL, Lang CC, Ng LL, Samani NJ, Tromp J, van Veldhuisen DJ, Touw DJ, Voors AA, van der Meer P (2020) Selenium and outcome in heart failure. Eur J Heart Fail 22:1415–1423
23. Du Laing G, Petrovic M, Lachat C, De Boevre M, Klingenberg GJ, Sun Q, De Saeger S, De Clercq J, Ide L, Vandekerckhove L, Schomburg L (2021) Course and survival of COVID-19 patients with comorbidities in relation to the trace element status at hospital admission. Nutrients 13:3304
24. Olson GE, Winfrey VP, Hill KE, Burk RF (2008) Megalin mediates selenoprotein P uptake by kidney proximal tubule epithelial cells. J Biol Chem 283:6854–6860
25. Chiu-Ugalde J, Theilig F, Behrends T, Drebes J, Sieland C, Subbarayal P, Köhrle J, Hammes A, Schomburg L, Schweizer U (2010) Mutation of megalin leads to urinary loss of selenoprotein P and selenium deficiency in serum, liver, kidneys and brain. Biochem J 431:103–111
26. Frass OM, Bühling F, Täger M, Frass H, Ansorge S, Huth C, Welte T (2001) Antioxidant and antiprotease status in peripheral blood and BAL fluid after cardiopulmonary bypass. Chest 120:1599–1608
27. Zou Z, Ren T, Li Y, Zeng Q, Wang X, Teng J, Xu J, Jia P, Ding X (2023) The association between serum glutathione peroxidase-3 concentration and risk of acute kidney injury after cardiac surgery: a nested case–control study. Am J Cardiol 209:29–35
28. Hall JE, do Carmo JM, da Silva AA, Wang Z, Hall ME (2019) Obesity, kidney dysfunction and hypertension: mechanistic links. Nat Rev Nephrol 15:367–385
29. Manzanares W, Biestro A, Galusso F, Torre MH, Mañay N, Pittini G, Facchin G, Hardy G (2009) Serum selenium and glutathione peroxidase-3 activity: biomarkers of systemic inflammation in the critically ill? Intensive Care Med 35:882–889
30. Allingstrup M, Afshari A (2015) Selenium supplementation for critically ill adults. Cochrane Database Syst Rev 2015:003703
31. Forceville X, Vitoux D, Gauzit R, Combes A, Lahilaire P, Chappuis P (1998) Selenium, systemic immune response syndrome, sepsis, and outcome in critically ill patients. Crit Care Med 26:1536–1544
32. Landucci F, Mancinelli P, De Gaudio AR, Virgili G (2014) Selenium supplementation in critically ill patients: a systematic review and meta-analysis. J Crit Care 29:150–156
33. Mahmoodpoor A, Hamishehkar H, Shadvar K, Ostadi Z, Sanaie S, Saghaleini SH, Nader ND (2019) The effect of intravenous selenium on oxidative stress in critically ill patients with acute respiratory distress syndrome. Immunol Invest 48:147–159
34. Manzanares W, Langlois PL, Hardy G (2013) Update on antioxidant micronutrients in the critically ill. Curr Opin Clin Nutr Metab Care 16:719–725
35. Angstwurm MW, Engelmann L, Zimmermann T, Lehmann C, Spes CH, Abel P, Strauss R, Meier-Hellmann A, Insel R, Radke J, Schüttler J, Gärtner R (2007) Selenium in Intensive Care (SIC): results of a prospective randomized, placebo-controlled, multiple-center study in patients with severe systemic inflammatory response syndrome, sepsis, and septic shock. Crit Care Med 35:118–126

36. Gül-Klein S, Haxhiraj D, Seelig J, Kästner A, Hackler J, Sun Q, Heller RA, Lachmann N, Pratschke J, Schmelzle M, Schomburg L (2021) Serum selenium status as a diagnostic marker for the prognosis of liver transplantation. Nutrients 13:619
37. Koszta G, Kacska Z, Szatmári K, Szerafin T, Fülesdi B (2012) Lower whole blood selenium level is associated with higher operative risk and mortality following cardiac surgery. J Anesth 26:812–821
38. Thomson CD (2004) Assessment of requirements for selenium and adequacy of selenium status: a review. Eur J Clin Nutr 58:391–402
39. Kipp AP, Strohm D, Brigelius-Flohé R, Schomburg L, Bechthold A, Leschik-Bonnet E, Heseker H (2015) Revised reference values for selenium intake. J Trace Elem Med Biol 32:195–199
40. Hurst R, Armah CN, Dainty JR, Hart DJ, Teucher B, Goldson AJ, Broadley MR, Motley AK, Fairweather-Tait SJ (2010) Establishing optimal selenium status: results of a randomized, double-blind, placebo-controlled trial. Am J Clin Nutr 91:923–931
41. McDonald VM, Osadnik CR, Gibson PG (2019) Treatable traits in acute exacerbations of chronic airway diseases. Chron Respir Dis 16:1479973119867954
42. Maslove DM, Tang B, Shankar-Hari M, Lawler PR, Angus DC, Baillie JK, Baron RM, Bauer M, Buchman TG, Calfee CS, Dos Santos CC, Giamarellos-Bourboulis EJ, Gordon AC, Kellum JA, Knight JC, Leligdowicz A, McAuley DF, McLean AS, Menon DK, Meyer NJ, Moldawer LL, Reddy K, Reilly JP, Russell JA, Sevransky JE, Seymour CW, Shapiro NI, Singer M, Summers C, Sweeney TE, Thompson BT, van der Poll T, Venkatesh B, Walley KR, Walsh TS, Ware LB, Wong HR, Zador ZE, Marshall JC (2022) Redefining critical illness. Nat Med 28:1141–1148
43. Lewin MH, Arthur JR, Riemersma RA, Nicol F, Walker SW, Millar EM, Howie AF, Beckett GJ (2002) Selenium supplementation acting through the induction of thioredoxin reductase and glutathione peroxidase protects the human endothelial cell line EAhy926 from damage by lipid hydroperoxides. Biochim Biophys Acta 1593:85–92
44. Leong JY, van der Merwe J, Pepe S, Bailey M, Perkins A, Lymbury R, Esmore D, Marasco S, Rosenfeldt F (2010) Perioperative metabolic therapy improves redox status and outcomes in cardiac surgery patients: a randomised trial. Heart Lung Circ 19:584–591
45. Chang C, Worley BL, Phaëton R, Hempel N (2020) Extracellular glutathione peroxidase GPx3 and its role in cancer. Cancers (Basel) 12:2197

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