Article

Selenium Deficiency is Associated with Mortality Risk from COVID-19

Arash Moghaddam[1], Raban Arved Heller[2,3], Qian Sun[3], Julian Seelig[3], Asan Cherkezov[1], Linda Seibert[1], Julian Hackler[3], Petra Seemann[3], Joachim Diegmann[1], Maximilian Pilz[4], Manuel Bachmann[1], Waldemar B. Minich[3], and Lutz Schomburg[3,*]

1 ATORG, Aschaffenburg Trauma and Orthopedic Research Group, Center for Orthopedics, Trauma Surgery and Sports Medicine, Hospital Aschaffenburg-Alzenau, D-63739 Aschaffenburg, Germany
2 HTRG, Heidelberg Trauma Research Group, Center for Orthopedics, Trauma Surgery and Spinal Cord Injury, Heidelberg University Hospital, D-69118 Heidelberg, Germany
3 Institute for Experimental Endocrinology, Charité-Universitätsmedizin Berlin, corporate member of Freie Universität Berlin, Humboldt-Universität zu Berlin, and Berlin Institute of Health, D-13353 Berlin, Germany
4 Institute of Medical Biometry and Informatics, Heidelberg University Hospital, Im Neuenheimer Feld 130.3, D-69120 Heidelberg, Germany
* Correspondence: lutz.schomburg@charite.de , Tel. +49-30-450524289, Fax. +49-30-450922

Received: date; Accepted: date; Published: date

Abstract:

SARS-CoV-2 infections underlie the current Coronavirus disease (COVID-19) pandemic and are causative for a high death toll particularly among elderly subjects and those with comorbidities. Selenium (Se) is an essential trace element of high importance for human health and particularly for a well-balanced immune response. Mortality risk from severe disease like sepsis or polytrauma is inversely related to Se status. We hypothesized that this relation also applies to COVID-19.

Serum samples (n=166) from COVID-19 patients (n=33) were collected consecutively and analysed for total Se by X-ray fluorescence and selenoprotein P (SELENOP) by a validated ELISA. Both biomarkers showed the expected strong correlation (r=0.7896, p<0.001), pointing to an insufficient Se status for optimal selenoprotein expression. In comparison to reference data from a European cross sectional analysis (EPIC, n=1915), the patients showed a pronounced deficit in total serum Se (mean±SD, 50.8±15.7 vs. 84.4±23.4 µg/L) and SELENOP (3.0±1.4 vs. 4.3±1.0 mg/L). A Se status below the 2.5th percentile of the reference population, i.e., [Se] < 45.7 µg/L and [SELENOP] < 2.56 mg/L was present in 43.4% and 39.2% of COVID samples, respectively. The Se status was significantly higher in samples from surviving COVID patients as compared to non-survivors (Se; 53.3±16.2 vs. 40.8±8.1 µg/L, SELENOP; 3.3±1.3 vs. 2.1±0.9 mg/L).

We conclude that Se status analysis in COVID patients provides diagnostic information. However, causality remains unknown due to the observational nature of this study. Nevertheless, the findings strengthen the notion on a relevant role of Se for COVID convalescence, and support the discussion on adjuvant Se supplementation in severely diseased and Se-deficient patients.

Keywords: trace element; inflammation; selenoprotein P; micronutrient; COVID-19.

1. Introduction

Severe Acute Respiratory Syndrome Coronavirus-2 (SARS-CoV-2) infections underlie the current Coronavirus disease (COVID-19) pandemic and are causative for an increasingly high death

toll particularly among elderly subjects and those who have severe comorbidities, e.g., chronic obstructive pulmonary disease, hypertension, diabetes, cancer, or a combination thereof [1,2]. It has been reported that severe disease course often associates with an overreaction of the body's immune system with a massive cytokine and chemokine release ("cytokine storm") [3]. Accordingly, the attempts to controlling the inflammation by immunosuppressive treatment using e.g. high dosages of corticosteroids has shown promising effects in reducing the rate of fatal disease course among the severely diseased COVID patients on mechanical ventilation (medRxiv 2020.06.22.20137273), causing a surge in dexamethasone demand [4]. This treatment success is reminiscent of the positive reports on dexamethasone capable of positively affecting the course of severe acute respiratory distress syndrome [5], or of reducing mortality rate in severely diseased and delirious patients from typhoid fever [6]. The strategy of repurposing common drugs known to positively affect the immune response are now increasingly applied in the current COVID pandemic [7]. The positive effects with tocilizumab and sarilumab are the most recent example (NCT04306705, NCT04322773). An adjuvant supply of certain micronutrients as positive modulators of the immune system may further support these attempts, and some vitamins (A, B6, B12, C, D, and E) and essential trace elements (zinc, iron, selenium (Se), magnesium or copper) are discussed as particularly promising [8]. However, at present the data base on these micronutrients is very limited in relation to disease pathophysiology, and it is unknown whether certain vitamins or trace elements are indeed deficient in patients with COVID-19, and whether the concentrations are related to disease severity or mortality risk.

For several reasons, the essential trace element Se is of particular relevance for viral infections among these nutritional factors. The immune system relies on a set of specific selenoproteins containing selenocysteine in their active sites and known to depend on abundant Se supply for their full expression and enzymatic activities [9,10]. Se deficiency is an established risk factor for viral infections [11]. Pathogens show higher mutation rates in Se-deficient subjects and can decisively contribute to a rapid evolution of pathogenic viral species [12]. Keshan disease is an endemic cardiomyopathy related to Se deficiency, and supplemental Se has proven meaningful for reducing the virus-associated disease incidence [13]. Se deficiency is also a risk factor for death from severe disease, as shown e.g. for sepsis [14] or polytraumatic injury [15]. Notably, the cure rate from COVID-19 was recently associated with basal Se status in different areas of China [16]. Collectively, the available studies support the notion that Se may be of relevance for infection with SARS-CoV-2 and disease course of COVID-19 [17-19]. However, data on Se status of individual patients severely affected by COVID-19 are missing. We hypothesized that severe Se deficiency is prevalent among the patients and associates with poor survival odds in COVID-19.

2. Materials and Methods

2.1 Study design

A cross-sectional study of patients with COVID-19 was conducted at the nonprofit Public Hospital Klinikum Aschaffenburg-Alzenau, Germany. Diagnosis of COVID-19 was based on positive detection of viral RNA using RT-PCR (Real time PCR - E-Gen according to Corman et al. [20], Medizinisches Versorgungszentrum MVZ Labor PD Dr. Volkmann & Kollegen GbR, Karlsruhe, Germany). The study was conducted in accordance with the declaration of Helsinki. Ethical counselling was provided by the authorities in Bavaria, Germany (Ethik-Kommission der Bayerischen Landesärztekammer, EA No. #20033) and the study was registered at the German Clinical Trial Register (Deutsches Register Klinischer Studien, ID: DRKS00022294). All patients enrolled into the analysis or next of kin have provided written informed consent. The samples were stored at -80°C (Aschaffenburg, Germany) and sent on dry ice to a remote lab from the clinics for analysis (Charité Berlin, Germany). All measurements were conducted by scientists and technicians blinded to the clinical information. Reference values were derived from a comprehensive data set of adult subjects participating in the European Prospective Investigation into Cancer and Nutrition (EPIC) study, analysed by the same technology as published recently [21].

2.2 Trace element analysis

Total reflection X-ray fluorescence (TXRF) was used to determine the concentration of Se in serum samples using a benchtop TXRF spectrometer (S4 T-STAR, Bruker Nano GmbH, Berlin, Germany). Briefly, samples were diluted with a gallium standard, applied to polished quartz glass slides and dried overnight. Seronorm serum standard (Sero AS, Billingstad, Norway) served as control. The concentrations measured were within the specified range of the standard, and inter-assay coefficient of variation (CV) was below 5% at a concentration of 45 µg Se/L serum.

2.3 SELENOP quantification by ELISA

SELENOP concentrations were measured from the serum samples by a sandwich method with monoclonal antibodies against human SELENOP using a validated commercial SELENOP-specific ELISA (selenOtest ELISA™, selenOmed GmbH, Berlin, Germany) as described [22]. Quality of measurements was verified by including two human serum standards in each assay run. The inter assay CV was below 15% during the analyses.

2.4 Assessment of glutathione peroxidase-3 (GPx3) activity

The activity of glutathione peroxidase-3 (GPx3) was assessed by a coupled enzymatic test procedure monitoring nicotinamide adenine dinucleotide phosphate (NADPH) consumption at 340 nm, as described earlier [23,24]. Briefly, serum samples were incubated with enzyme buffer containing 3.4 mM reduced glutathione (GSH), 0.27 mg/mL NADPH, 1 mM NaN_3, and 0.3 U/mL glutathione reductase. The enzymatic reaction was started by hydrogen peroxide, and consumption of NADPH was monitored at 340 nm. Inter- and intra-assay CV were below 20%.

2.5 Statistical analysis

Statistical analysis was performed with GraphPad Prism (Version 7, GraphPad Software Inc., San Diego, CA, USA) and the open software R, version 3.6.0 [25], applying the packages “arsenal”, “precrec”, “tidyr” [26], “dplyr” [27], and “ggplot2” [28]. The Shapiro-Wilk test was used for assessing normal distribution of values. Categorical variables were evaluated by Boschloo’s test [29]. Comparisons were conducted by unpaired Student’s t-test. More than two groups were compared with ANOVA and Dunn's multiple comparisons test. Correlations were tested by Spearman´s correlation test. All statistical tests were two-sided, and P-values < 0.05 were considered significant; * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, and **** $p < 0.0001$.

3. Results

3.1. Patient characteristics

A total of n=33 patients qualified for analysis and were enrolled into this observational study, providing a set of n=166 consecutive serum samples. COVID-19 patients who survived or died showed similar characteristics, except for a lower age range of the survivors (**Table 1**)

Table 1. Characteristics of the COVID-19 patients contributing to this study

	Death	Discharge	Total
Sex			
female	4 (67%)	15 (56%)	19 (58%)
male	2 (33%)	12 (44%)	14 (42%)
Age			
median (IQR)	89 (81, 94)	69 (38, 91)	77 (38, 94)
Comorbidities			
hypertension	4 (67%)	18 (67%)	22 (67%)
diabetes	2 (33%)	4 (15%)	6 (18%)
COPD	0 (0%)	1 (4%)	1 (3%)
CVD	3 (50%)	14 (52%)	17 (52%)
cerebrovascular disease	1 (17%)	5 (19%)	6 (18%)
adipositas	1 (17%)	6 (22%)	7 (21%)
Time to discharge or death* [d]			
median (IQR)	10 (2, 32)	19 (3, 46)	15 (2, 46)

** death in combination with COVID-19 diagnosis, irrespective of final mortality cause.*

3.2. Selenium (Se) Status Analysis

Serum Se status was evaluated from all patient samples as assessed by three complementary biomarkers, i.e., total serum Se and SELENOP concentrations, as well as GPx3 activity. The three Se status biomarkers showed significant and linear correlations over the full range of data, indicating a high quality of the samples (Figure 1). The correlation coefficients were highest for the parameter pair of total serum Se and SELENOP concentration (Figure 1A), followed by the parameter pair GPx3 activity and total serum Se (Figure 1B). GPx3 activity and serum SELENOP concentration showed the least stringent correlation (Figure 1C).

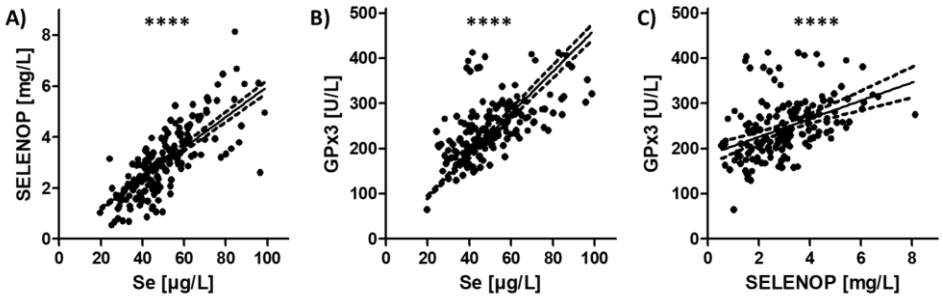

Figure 1. Analysis of Se status from samples of patients suffering from COVID-19 by three complementary serum biomarkers. Serum samples (n=166) were analysed from COVID-19 patients (n=33) by measuring total Se concentration, serum SELENOP level and activity of secreted GPx3. A) The Se transporter SELENOP and total Se concentration showed a tight positive linear correlation (r=0.7896), in agreement with the analysis of B) GPx3 activity and total Se concentration (r=0.6239), as well as with C) GPx3 activity and SELENOP concentration (r=0.4954). r; Spearman correlation coefficient (2-sided, 2-tailed), **** $p < 0.0001$.

3.3. Se status of COVID-19 patients in relation to reference range of healthy control subjects

An average population-wide Se status was deduced from n=1915 data sets obtained earlier from healthy adult subjects participating in the cross-sectional EPIC study [21]. Reference ranges for total serum Se and SELENOP concentrations were deduced by determination of the 2.5th-97.5th percentile

of the data. According to this large cross-sectional study, SELENOP concentrations are unrelated to age [21]. The chosen criterion of 95% of data constituting the reference ranges classifies a normal Se status when residing in the range of 45.7 – 131.6 µg/L for serum Se, and 2.56 – 6.63 mg/L for serum SELENOP concentration, respectively. According to these reference ranges, 43.4% of samples from COVID-19 patients were deficient in Se, and 39.2% were deficient in SELENOP, respectively.

3.4. Se status of COVID-19 patients in relation to survival

Separating patient samples from surviving versus deceased COVID-19 patients, the difference becomes more obvious. In the samples of deceased COVID-19 patients, 64.7% and 70.6% showed Se- and SELENOP-deficiency, respectively, whereas 37.9% and 31.1% of the samples from the survivors had to be classified as Se- and SELENOP-deficient, respectively. Accordingly, a significantly lower Se status was identified in the non-survivors in comparison to the survivors with respect to all three biomarkers of Se status analyzed (Table 2).

Table 2. Comparison of Se status biomarkers in COVID-19 samples in relation to survival

	all samples	**discharge**	**death**	**p-value***
	n = 166	**n = 132**	**n = 34**	
serum Se [µg/L]	50.8±15.7	**53.3±16.2**	**40.8±8.1**	**P<0.001**
serum SELENOP [mg/L]	3.0±1.4	**3.3±1.3**	**2.1±0.9**	**P<0.001**
serum GPx3 [U/L]	246.1±64.4	**251.6±69.6**	**224.8±30.3**	**P<0.001**

**Student's t-test, 2-tailed, 2-sided, comparison of discharge versus death*

A comparison of the median values and inter quartile ranges (IQR) of the samples from the COVID-19 patients who did not survive in relation to the reference cohort of healthy adult European subjects indicates that the groups differ strongly, i.e., the IQR do not overlap. This means that the ranges encompassing 75% of all samples are separated from each other, irrespective of biomarker used, i.e., both in relation to total serum Se and serum SELENOP concentrations (Figure 2A, B). Notably, the bottom 75% of values from the deceased patients are below the median values of the surviving COVID-19 patients, suggesting that both parameters of Se status are of value for the identification of patients with severe disease course and high mortality risk.

With regard to the choice of biomarker, both total serum Se and SELENOP concentrations appear similarly suitable for providing information on survival chances of COVID-19 patients. Importantly, Se and SELENOP showed the known positive linear correlation in both the group of non-survivors and of discharged patients, even though the correlation line is less steep and on a lower level in the non-survivors due to their repressed Se status (Figure 2 C, D).

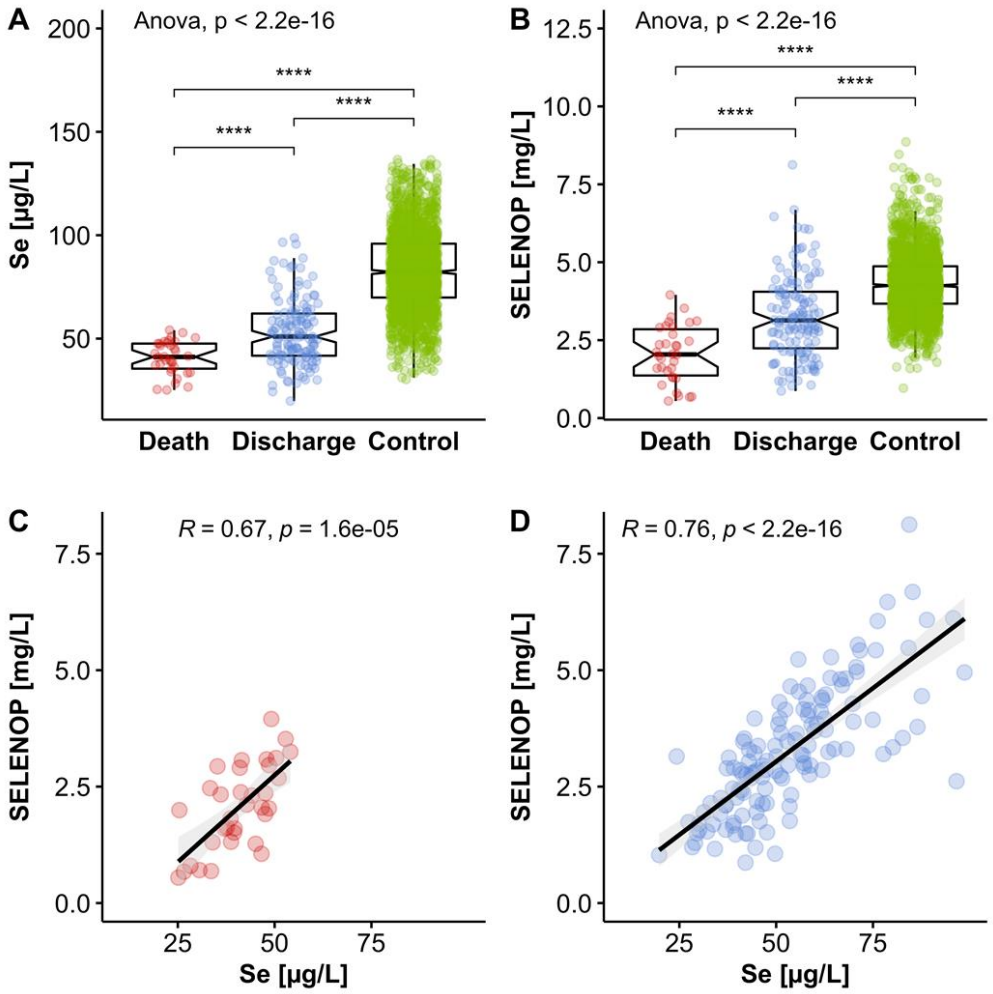

Figure 2. Comparison of Se status in COVID-19 patients who survived or died in relation to healthy controls. A) Total serum Se concentration differed significantly and were most strongly depressed in COVID-19 patients who did not survive. B) SELENOP concentrations differed to a similar extent and were also lowest in non-survivors. C) As observed in the full cohort of samples, Se and SELENOP showed a strong positive correlation albeit with a less steep slope on low concentration levels in the group of non-survivors, and D) with considerable steepness as known from healthy controls in the group of surviving COVID-19 patients. All tests were two-sided and P-values <0.05 were considered statistically significant; *R*, Spearman correlation coefficient (2-sided, 2-tailed), **** indicates p<0.0001.

A direct comparison of Se status in COVID-19 patients to reference values for the activity of GPx3 as biomarker was not possible, as GPx3 had not been determined in the samples of the large reference cohort from the EPIC study [21].

Next, a receiver operating characteristic (ROC) curve analysis was conducted to analyse the diagnostic ability of the Se status biomarkers for survival odds. ROC curve analyses can contribute to decision making in a binary classifier system by testing a discrimination threshold via calculating all possible variations. However, ROC plots alone may be misleading and bear the risk of error when applied in imbalanced classification scenarios [30]. For this reason, a precision recall curve (PRC) was calculated to identify the fraction of true positives among all the positive predictions and thereby providing a more accurate prediction of future classification performance (Figure 3). The available data on SELENOP, Se and GPx3 were suitable to reliably distinguishing between those patients who could be discharged or died. Applying a stepwise AIC selection process revealed that the SELENOP concentration outperformed the other variables as well as combinations thereof. This result is mirrored in both the corresponding ROC and PRC curves (Figure 3A, B).

Peer-reviewed version available at Nutrients 2020, 12, 2098; doi:10.3390/nu12072098

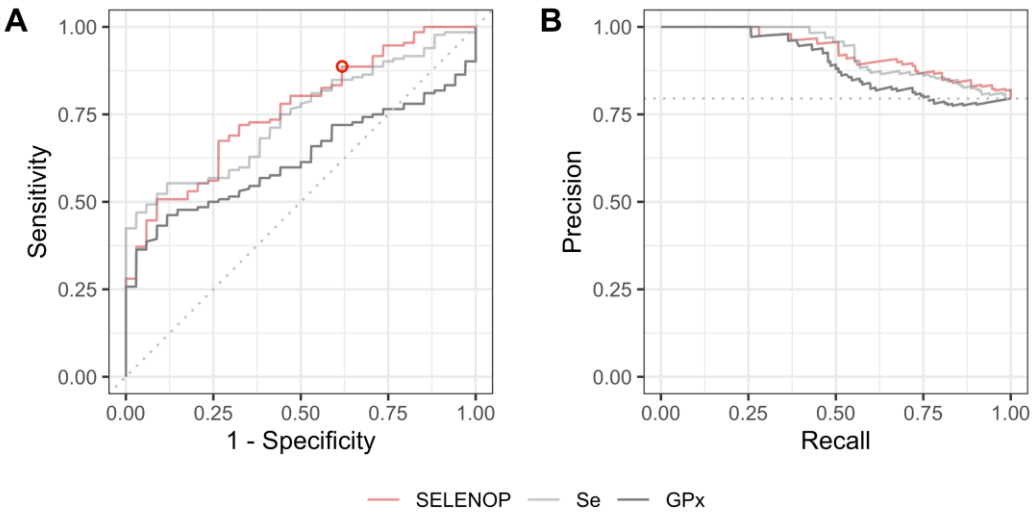

Figure 3. Receiver operating characteristics (ROC) analyses of Se status biomarkers in relation to risk of death from COVID-19. A) A ROC analysis as prediction model based on the serum concentrations of SELENOP, Se and GPx3 (pooled values from admission to the endpoint of the study) is capable of discriminating between patients that died and those that have been discharged. The optimal cutpoint of SELENOP concentrations at 3.1 mg/L according to the Youden's J statistics is indicated by a red circle. B) The corresponding precision recall curve (PRC) indicates the fraction of true positives among all the positive predictions and may serve as a meaningful addition to current risk estimates.

The final model based on SELENOP concentration yielded an AUC of 75.9% when 3.1 mg/L are chosen as optimal cutpoint based on the Youden's J statistic (Figure 3A). This cutpoint is characterized by a sensitivity of 91.18% and a specificity of 50.76%, and may serve as a valuable screening tool to contribute to a better assessment of the mortality risk in patients suffering from COVID-19. This notion is further underlined by the specific characteristics of the predictive models used (Table 3).

Table 3. Specific characteristics of the predictive models used. For each model, the variable estimates included in the calculations are provided with their corresponding confidence interval (CI).

	serum Se	serum SELENOP	GPx3 activity	all
(intercept)	**-1.70*** [-2.20, -1.20]**	**-1.75*** [-2.27, -1.24]**	**-1.42*** [-1.81, -1.02]**	**-1.80*** [-2.34, -1.26]**
Se	**-1.19*** [-1.79, -0.60]**			**-0.55 [-1.39, 0.30]**
SELENOP		**-1.28*** [-1.86, -0.70]**		**-0.94* [-1.72, -0.16]**
GPx3			**-0.46* [-0.89, -0.04]**	**0.09 [-0.37, 0.54]**
N	**166**	**166**	**166**	**166**
AIC	**150.5**	**146.3**	**167.3**	**148.5**
Pseudo R^2	**0.19**	**0.23**	**0.05**	**0.24**

*All continuous predictors are mean-centered and scaled by 1 standard deviation. *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$.*

A precision-recall-plot that is recommended in such analyses [30] is depicted in Figure 3B, and confirms the findings of the superiority of SELENOP and Se over GPx3 as predictive parameter. Interestingly, all three parameters show a precision value uniformly above 0.75, supporting the general suitability of all three parameters with relatively high positive predictive values.

Peer-reviewed version available at Nutrients 2020, 12, 2098; doi:10.3390/nu12072098

4. Discussion

In this manuscript, we report that patients suffering from COVID-19 display a deficiency in the essential trace element Se in blood, along with low concentrations of the Se transporter SELENOP and low enzymatic activity of the secreted GPx3. Notably, the Se deficiency was very strong in comparison to healthy European adults, and it was reflected concordantly in relatively depressed readings of all three different Se status biomarkers determined. The observation that Se deficiency was more severe in the samples obtained from non-survivors as compared to survivor of COVID-19 may suggest some relevance of the trace element for coping with the virus and successful convalescence. Besides the physiological role of Se for supporting biosynthesis of immune system-relevant selenoproteins, the data also highlight that a determination of Se status by any of the biomarkers evaluated is of diagnostic value for a better prediction of disease course and an improved identification of patients at particular risk for losing the battle against this devastating infection.

Although the nature of the analysis as an observational study does not allow the deduction of causal relationships, there are different hypotheses for the underlying biochemical pathways leading to the observations presented in this manuscript.

Firstly, Se status may already have been relatively low in the patients before disease, constituting a risk factor for viral infection as shown previously for other diseases [11,12]. In this respect, the experience with viral-induced Keshan disease [13] or AIDS [31] may serve as paradigmatic examples highlighting the potential relevance of Se for infection risk and disease course [31]. However, the high infection rate of SARS-CoV-2 apparently infecting very many of the directly exposed subjects [32] in combination with the majority of COVID-19 samples exhibiting Se values below 2.5th percentile of the population range argues against a Se-dependent predisposition as explanation for the findings.

Secondly, in disease and upon the growing inflammation, a potentially pre-existing low Se status may decline further. This notion is supported from similar findings in other severe diseases, especially sepsis [14] and polytraumatic injury [15], where low, declining and mortality-relevant Se deficiency has been observed that is unlikely a predisposition. Moreover, the negative acute phase response of hepatic SELENOP biosynthesis [33], together with the suppressive effects of hypoxia [34] or cytokines, e.g. IL-6 [35], argue in favour of this mechanism contributing to the differences.

Thirdly, a longer stay on the ICU under inflammatory and hypoxic conditions may cause an elevated Se requirement due to ongoing Se loss, as erythrocyte Se often remains normal despite declining Se in blood [36]. In human evolution, high quality medical care with supportive ventilation was usually not available, and an infection was followed soon be either remission or death. Under these conditions, safeguarding essential micronutrients for later recovery was no survival advantage. The present care on the ICU over long periods of time constitutes a fundamental different situation, where the constant suppression of hepatic SELENOP biosynthesis may require supplemental measures in the long run [36]. Concordant with this notion, the hypothesized association of low Se status with impaired recovery was reported from an *in silico* analysis of cure rates from COVID-19 in the different areas of China with diverging baseline Se status [16].

Fourthly, an over-shooting immune response may be directly related to Se status as oxidative stress may overrun the capacity of protective selenoenzymes of the GPx and thioredoxin reductase families and low molecular weight antioxidants [37]. This loss of redox balance has been hypothesized before as of etiopathogenic relevance [12,38]. The therapeutic success of dexamethasone or tocilicumab treatment, as well as the perspective of the GPx mimetic ebselen as promising therapeutic measure lend further support for this theory [39,40].

Finally, a declining serum Se status may just constitute a surrogate marker for disease severity and the tone of pathological stressors, like hypoxia and inflammatory cytokines. This notion is supported by a vast body of literature on declining selenoprotein biosynthesis under acute phase conditions, in inflammation and under hypoxia. A declining Se status will further disrupt the redox balance thereby closing a fatal feed-forward loop, again arguing for the potential relevance of some supplemental support to interrupt this vicious cycle during long lasting disease (Figure 4).

Collectively, similar to the proposed interrelation of declining Se status in malignant diseases, the strong deficit in Se and SELENOP observed in COVID-19 may result from a combination of the

Peer-reviewed version available at Nutrients 2020, 12, 2098; doi:10.3390/nu12072098

aforementioned pathways and interactions. Supportive measures aimed at improving selenoprotein biosynthesis in COVID-19 may enable a better redox control and fine-tuned response of the immune system [38]. It appears meaningful, timely and promising to initiate population-wide measures trying to identify subjects with pre-existing Se deficits, not just as preventive measure for viral infections, spread and virulence development [11,12,39], but also to reduce the individual risk for cardiovascular mortality [41-44], cancer [21,45,46], and death from severe disease [10,14,36].

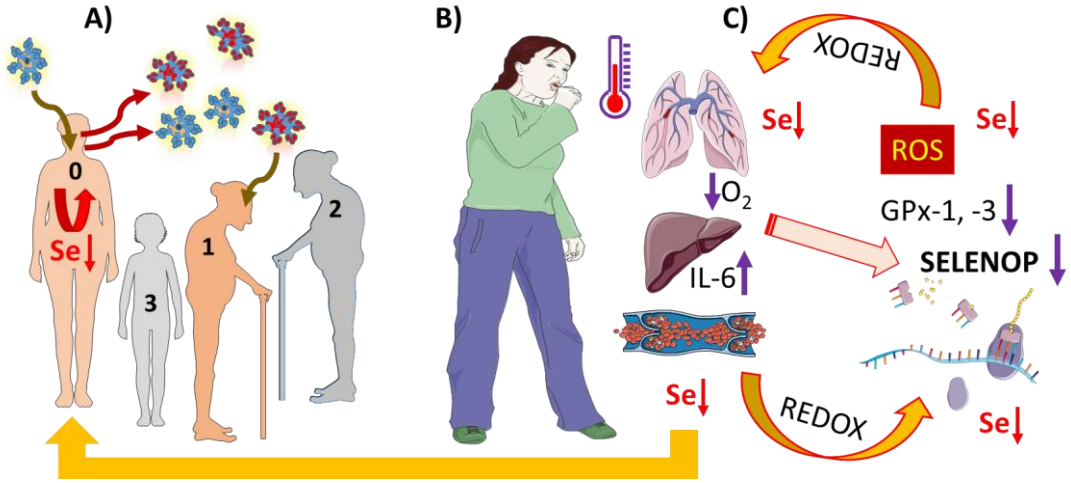

Figure 4. Pathophysiological mechanisms potentially underlying low Se status in severe COVID-19. Infections by SARS-CoV-2 occur largely independent from baseline Se status. A) Some individuals with poor immune system and low baseline Se status (0) may spread the virus efficiently and allow viral replication and rapid evolution of particular pathogenic viral species due to low expression of protective selenoenzymes. Subjects with better Se status (1-3) may be less prone to severe disease course. B) COVID-19 is characterized by inflammation, hypoxia and high cytokine concentrations (e.g. IL-6). The combination of hypoxia and IL-6 suppresses selenoprotein expression. C) Biosynthesis of the Se transporter SELENOP in hepatocytes is particular sensitive, causing whole body Se status decline and insufficient expression of protective selenoenzymes, e.g. cytosolic GPx1 and plasma GPx3. Insufficient inactivation of peroxides as precursors of reactive oxygen species (ROS) results, causing a serious disturbance of redox balance, closing a vicious cycle both with respect to selenoprotein expression, Se status and COVID-19 progression. It is hypothesized that supplemental Se may interrupt this series of detrimental events and contribute to better odds for convalescence. This figure was created by using some Servier Medical Art templates, which are licensed under a Creative Commons Attribution 3.0 Unported License; https://smart.servier.com.

The particular strengths of the current study are the parallel assessment of different and coherent biomarkers of Se status by standardised methodology, and the blinded set-up of the analyses. Among the limitations are, as usual in explorative pilot studies, the relatively limited number of patients and samples, and the lack of clinical data on inflammatory parameters.

5. Conclusions

COVID-19 constitutes a universal threat to human health, necessitating fast, promising and safe measures for reducing infection risk, suppressing virulence development, strengthening the immune system and supporting recovery. The essential trace element Se may be most relevant for these issues. Subjects residing in areas with poor baseline Se supply or on restricted nutrition, and COVID patients with pre-existing comorbidities or long disease course are at particularly elevated risk for severe Se deficiency, and may profit from improving the Se supply by dietary or supplemental measures. The observed association of mortality risk with Se deficit and the likely underlying feed-forward mechanism argues for initiating intervention studies under highest quality standards, in order not to miss a universally available, inexpensive and safe preventive measure and adjuvant treatment option.

Author contributions: Conceptualization, AM, RAH, MB and LS (Lutz Schomburg); Methodology, RAH, QS, JS, AC, LS (Linda Seibert), JH, PS, JD, and WBM; Software, RAH, AC, LS (Linda Seibert), MP and MB; Validation, RAH, QS, JS, AC, LS (Linda Seibert), JH and PS; Formal Analysis, AM, RAH, AC, LS (Linda Seibert), JD, MP, MB, WBM and LS (Lutz Schomburg); Resources, AM, JD, MB and LS (Lutz Schomburg); Data Curation, RAH, QS, JS, MP and LS (Lutz Schomburg); Writing – Original Draft Preparation, AM, RAH and LS (Lutz Schomburg); Writing – Review & Editing, AM, RAH, QS, JS, AC, LS (Linda Seibert), JH, PS, JD, MP, MB, WBM and LS (Lutz Schomburg); Visualization, RAH, JS, AC, LS (Linda Seibert), and MP; Supervision, AM and LS (Lutz Schomburg); Funding Acquisition AM and LS (Lutz Schomburg). All authors have read and agreed to the published version of the manuscript.

Funding: The research has been funded by the Deutsche Forschungsgemeinschaft (DFG), Research Unit FOR-2558 "TraceAge" (Scho 849/6-1). We acknowledge financial support by the Open Access Publication Fund of Charité – Universitätsmedizin Berlin, and the funding received towards the doctoral thesis of RH from the Oskar-Helene-Heim foundation, Berlin, Germany.

Acknowledgments: We thank Vartitér Seher, Gabriele Boehm and Anja Fischbach for excellent technical support, and Prof. Volker Daniel, Heidelberg, and Helena L. Crowell, ETH Zürich, for constructive discussions. Intellectual support and essential motivation was provided by valuable and inspiring colleagues from the International Society for Selenium Research (ISSR).

Conflicts of Interest. L.S. holds shares and P.S. serves as CEO of selenOmed GmbH, a company involved in Se status assessment and supplementation. The other authors declare no competing interest.

References

1. Guan, W.J.; Liang, W.H.; Zhao, Y.; Liang, H.R.; Chen, Z.S.; Li, Y.M.; Liu, X.Q.; Chen, R.C.; Tang, C.L.; Wang, T., et al. Comorbidity and its impact on 1590 patients with COVID-19 in China: a nationwide analysis. *Eur Respir J* **2020**, *55*, doi:10.1183/13993003.00547-2020.
2. Wang, B.L.; Li, R.B.; Lu, Z.; Huang, Y. Does comorbidity increase the risk of patients with COVID-19: evidence from meta-analysis. *Aging-Us* **2020**, *12*, 6049-6057, doi:10.18632/aging.103000.
3. Song, P.; Li, W.; Xie, J.; Hou, Y.; You, C. Cytokine Storm Induced by SARS-CoV-2. *Clin Chim Acta* **2020**, 10.1016/j.cca.2020.06.017, doi:10.1016/j.cca.2020.06.017.
4. Mahase, E. Covid-19: Demand for dexamethasone surges as RECOVERY trial publishes preprint. *BMJ* **2020**, *369*, m2512, doi:10.1136/bmj.m2512.
5. Villar, J.; Ferrando, C.; Martinez, D.; Ambros, A.; Munoz, T.; Soler, J.A.; Aguilar, G.; Alba, F.; Gonzalez-Higueras, E.; Conesa, L.A., et al. Dexamethasone treatment for the acute respiratory distress syndrome: a multicentre, randomised controlled trial. *Lancet Respir Med* **2020**, *8*, 267-276, doi:10.1016/S2213-2600(19)30417-5.
6. Hoffman, S.L.; Punjabi, N.H.; Kumala, S.; Moechtar, M.A.; Pulungsih, S.P.; Rivai, A.R.; Rockhill, R.C.; Woodward, T.E.; Loedin, A.A. Reduction of mortality in chloramphenicol-treated severe typhoid fever by high-dose dexamethasone. *N Engl J Med* **1984**, *310*, 82-88, doi:10.1056/NEJM198401123100203.
7. Joshi, S.; Joshi, M.; Degani, M.S. Tackling SARS-CoV-2: proposed targets and repurposed drugs. *Future Med Chem* **2020**, 10.4155/fmc-2020-0147, doi:10.4155/fmc-2020-0147.
8. Calder, P.C.; Carr, A.C.; Gombart, A.F.; Eggersdorfer, M. Optimal Nutritional Status for a Well-Functioning Immune System Is an Important Factor to Protect against Viral Infections. *Nutrients* **2020**, *12*, doi:10.3390/nu12041181.
9. Hoffmann, P.R.; Berry, M.J. The influence of selenium on immune responses. *Mol Nutr Food Res* **2008**, *52*, 1273-1280, doi:10.1002/mnfr.200700330.
10. Schomburg, L. The other view: the trace element selenium as a micronutrient in thyroid disease, diabetes, and beyond. *Hormones (Athens)* **2020**, *19*, 15-24, doi:10.1007/s42000-019-00150-4.

11. Guillin, O.M.; Vindry, C.; Ohlmann, T.; Chavatte, L. Selenium, Selenoproteins and Viral Infection. *Nutrients* **2019**, *11*, doi:10.3390/nu11092101.
12. Beck, M.A.; Handy, J.; Levander, O.A. Host nutritional status: the neglected virulence factor. *Trends Microbiol* **2004**, *12*, 417-423, doi:10.1016/j.tim.2004.07.007.
13. Zhou, H.; Wang, T.; Li, Q.; Li, D. Prevention of Keshan Disease by Selenium Supplementation: a Systematic Review and Meta-analysis. *Biol Trace Elem Res* **2018**, *186*, 98-105, doi:10.1007/s12011-018-1302-5.
14. Forceville, X.; Vitoux, D.; Gauzit, R.; Combes, A.; Lahilaire, P.; Chappuis, P. Selenium, systemic immune response syndrome, sepsis, and outcome in critically ill patients. *Crit Care Med* **1998**, *26*, 1536-1544, doi:10.1097/00003246-199809000-00021.
15. Braunstein, M.; Kusmenkov, T.; Zuck, C.; Angstwurm, M.; Becker, N.P.; Bocker, W.; Schomburg, L.; Bogner-Flatz, V. Selenium and Selenoprotein P Deficiency Correlates With Complications and Adverse Outcome After Major Trauma. *Shock* **2020**, *53*, 63-70, doi:10.1097/SHK.0000000000001344.
16. Zhang, J.; Taylor, E.W.; Bennett, K.; Saad, R.; Rayman, M.P. Association between regional selenium status and reported outcome of COVID-19 cases in China. *Am J Clin Nutr* **2020**, *111*, 1297-1299, doi:10.1093/ajcn/nqaa095.
17. Fakhrolmobasheri, M.; Nasr-Esfahany, Z.; Khanahmad, H.; Zeinalian, M. Selenium supplementation can relieve the clinical complications of COVID-19 and other similar viral infections. *Int J Vitam Nutr Res* **2020**, 10.1024/0300-9831/a000663, 1-3, doi:10.1024/0300-9831/a000663.
18. Kieliszek, M.; Lipinski, B. Selenium supplementation in the prevention of coronavirus infections (COVID-19). *Med Hypotheses* **2020**, *143*, 109878, doi:10.1016/j.mehy.2020.109878.
19. Seale, L.A.; Torres, D.J.; Berry, M.J.; Pitts, M.W. A role for selenium-dependent GPX1 in SARS-CoV-2 virulence. *Am J Clin Nutr* **2020**, 10.1093/ajcn/nqaa177, doi:10.1093/ajcn/nqaa177.
20. Corman, V.M.; Landt, O.; Kaiser, M.; Molenkamp, R.; Meijer, A.; Chu, D.K.; Bleicker, T.; Brunink, S.; Schneider, J.; Schmidt, M.L., et al. Detection of 2019 novel coronavirus (2019-nCoV) by real-time RT-PCR. *Euro Surveill* **2020**, *25*, doi:10.2807/1560-7917.ES.2020.25.3.2000045.
21. Hughes, D.J.; Fedirko, V.; Jenab, M.; Schomburg, L.; Meplan, C.; Freisling, H.; Bueno-de-Mesquita, H.B.; Hybsier, S.; Becker, N.P.; Czuban, M., et al. Selenium status is associated with colorectal cancer risk in the European prospective investigation of cancer and nutrition cohort. *Int J Cancer* **2015**, *136*, 1149-1161, doi:10.1002/ijc.29071.
22. Hybsier, S.; Schulz, T.; Wu, Z.; Demuth, I.; Minich, W.B.; Renko, K.; Rijntjes, E.; Kohrle, J.; Strasburger, C.J.; Steinhagen-Thiessen, E., et al. Sex-specific and inter-individual differences in biomarkers of selenium status identified by a calibrated ELISA for selenoprotein P. *Redox Biol* **2017**, *11*, 403-414, doi:10.1016/j.redox.2016.12.025.
23. Schomburg, L.; Schweizer, U.; Holtmann, B.; Flohe, L.; Sendtner, M.; Kohrle, J. Gene disruption discloses role of selenoprotein P in selenium delivery to target tissues. *Biochem J* **2003**, *370*, 397-402, doi:10.1042/BJ20021853.
24. Mehl, S.; Sun, Q.; Gorlich, C.L.; Hackler, J.; Kopp, J.F.; Renko, K.; Mittag, J.; Schwerdtle, T.; Schomburg, L. Cross-sectional analysis of trace element status in thyroid disease. *J Trace Elem Med Biol* **2020**, *58*, 126430, doi:10.1016/j.jtemb.2019.126430.
25. Team, R.D.C. R, A language and environment for statistical computing. *R. Found, Stat. Comput.* **2015**, *1*.
26. Wickham, H.; Henry, L. tidyr: easily tidy data with "spread ()" and "gather ()" functions. R package version 0.8. 0. URL https://CRAN. R-project. org/package= tidyr [accessed 14 May 2018]: 2018.

27. Wickham, H.; Francois, R.; Henry, L.; Müller, K. dplyr: A Grammar of Data Manipulation. R package version 0.4. 3. *R Found. Stat. Comput., Vienna. https://CRAN. R-project. org/package= dplyr* **2015**.
28. Wickham, H. *ggplot2: Elegant Graphics for Data Analysis. R package version 2.1.0*; Springer-Verlag New York: 2009; pp. 1-195.
29. Boschloo, R.D. Raised Conditional Level of Significance for 2x2 Table When Testing Equality of 2 Probabilities. *Ann Math Stat* **1968**, *39*, 1094-&.
30. Saito, T.; Rehmsmeier, M. The precision-recall plot is more informative than the ROC plot when evaluating binary classifiers on imbalanced datasets. *PLoS One* **2015**, *10*, e0118432, doi:10.1371/journal.pone.0118432.
31. Harthill, M. Review: micronutrient selenium deficiency influences evolution of some viral infectious diseases. *Biol Trace Elem Res* **2011**, *143*, 1325-1336, doi:10.1007/s12011-011-8977-1.
32. Wu, J.T.; Leung, K.; Bushman, M.; Kishore, N.; Niehus, R.; de Salazar, P.M.; Cowling, B.J.; Lipsitch, M.; Leung, G.M. Estimating clinical severity of COVID-19 from the transmission dynamics in Wuhan, China. *Nat Med* **2020**, *26*, 506-510, doi:10.1038/s41591-020-0822-7.
33. Renko, K.; Hofmann, P.J.; Stoedter, M.; Hollenbach, B.; Behrends, T.; Kohrle, J.; Schweizer, U.; Schomburg, L. Down-regulation of the hepatic selenoprotein biosynthesis machinery impairs selenium metabolism during the acute phase response in mice. *FASEB J* **2009**, *23*, 1758-1765, doi:10.1096/fj.08-119370.
34. Becker, N.P.; Martitz, J.; Renko, K.; Stoedter, M.; Hybsier, S.; Cramer, T.; Schomburg, L. Hypoxia reduces and redirects selenoprotein biosynthesis. *Metallomics* **2014**, *6*, 1079-1086, doi:10.1039/c4mt00004h.
35. Martitz, J.; Becker, N.P.; Renko, K.; Stoedter, M.; Hybsier, S.; Schomburg, L. Gene-specific regulation of hepatic selenoprotein expression by interleukin-6. *Metallomics* **2015**, *7*, 1515-1521, doi:10.1039/c5mt00211g.
36. Manzanares, W.; Langlois, P.L.; Heyland, D.K. Pharmaconutrition with selenium in critically ill patients: what do we know? *Nutr Clin Pract* **2015**, *30*, 34-43, doi:10.1177/0884533614561794.
37. Nordberg, J.; Arner, E.S. Reactive oxygen species, antioxidants, and the mammalian thioredoxin system. *Free Radic Biol Med* **2001**, *31*, 1287-1312, doi:10.1016/s0891-5849(01)00724-9.
38. Steinbrenner, H.; Al-Quraishy, S.; Dkhil, M.A.; Wunderlich, F.; Sies, H. Dietary selenium in adjuvant therapy of viral and bacterial infections. *Adv Nutr* **2015**, *6*, 73-82, doi:10.3945/an.114.007575.
39. Sies, H.; Parnham, M.J. Potential therapeutic use of ebselen for COVID-19 and other respiratory viral infections. *Free Radic Biol Med* **2020**, 10.1016/j.freeradbiomed.2020.06.032, doi:10.1016/j.freeradbiomed.2020.06.032.
40. Jin, Z.; Du, X.; Xu, Y.; Deng, Y.; Liu, M.; Zhao, Y.; Zhang, B.; Li, X.; Zhang, L.; Peng, C., et al. Structure of M(pro) from SARS-CoV-2 and discovery of its inhibitors. *Nature* **2020**, *582*, 289-293, doi:10.1038/s41586-020-2223-y.
41. Alehagen, U.; Johansson, P.; Bjornstedt, M.; Rosen, A.; Post, C.; Aaseth, J. Relatively high mortality risk in elderly Swedish subjects with low selenium status. *Eur J Clin Nutr* **2016**, *70*, 91-96, doi:10.1038/ejcn.2015.92.
42. Schomburg, L.; Orho-Melander, M.; Struck, J.; Bergmann, A.; Melander, O. Selenoprotein-P Deficiency Predicts Cardiovascular Disease and Death. *Nutrients* **2019**, *11*, doi:10.3390/nu11081852.

43. Bomer, N.; Grote Beverborg, N.; Hoes, M.F.; Streng, K.W.; Vermeer, M.; Dokter, M.M.; J, I.J.; Anker, S.D.; Cleland, J.G.F.; Hillege, H.L., et al. Selenium and outcome in heart failure. *Eur J Heart Fail* **2019**, 10.1002/ejhf.1644, doi:10.1002/ejhf.1644.

44. Hu, X.F.; Stranges, S.; Chan, L.H.M. Circulating Selenium Concentration Is Inversely Associated With the Prevalence of Stroke: Results From the Canadian Health Measures Survey and the National Health and Nutrition Examination Survey. *J Am Heart Assoc* **2019**, *8*, e012290, doi:10.1161/JAHA.119.012290.

45. Rayman, M.P. Selenium and human health. *Lancet* **2012**, *379*, 1256-1268, doi:10.1016/S0140-6736(11)61452-9.

46. Schomburg, L.; Hughes, D.J. The missing link? The potential role of selenium in the development of liver cancer and significance for the general population. *Expert Rev Gastroenterol Hepatol* **2017**, *11*, 707-709, doi:10.1080/17474124.2017.1320219.