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# Circulatory biochemical markers and Pulmonary Function in Patients with Idiopathic Pulmonary Fibrosis: A systematic review and Meta-Analysis

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## ABSTRACT

**Background**: A significant part of the pathogenesis of cystic fibrosis (CF) may be played by oxidative stress.

**Objective:** To measure redox abnormalities associated with CF.

**Methodology:** The information databases 'CINAHL, PsycINFO, CENTRAL, and Medline' have been thoroughly searched. Standardized mean difference (SMD) and 95% confidence intervals (95% CI) were computed using the mean concentrations of biological markers in the blood from individuals with 'clinically-stable cystic fibrosis' (CF) and 'non-CF controls'.

**Results:** This systematic review analyzed 842 citations, ultimately including 44 studies conducted between 1998 and 2020 across 54 global centers. Most investigations had a 'cross-sectional design', with a few controlled trials, and were generally of moderate quality. Key findings indicated that individuals with clinically stable cystic fibrosis (CF) had significantly lower levels of plasma or serum vitamin A (SMD -0.67), β-carotene (SMD -2.18), and vitamin E (SMD -0.68) compared to non-CF controls, with no significant differences in vitamin C (SMD -0.04), 25(OH)D (SMD -0.24), or the vitamin E ratio (SMD -0.36). Trace element levels showed no significant differences for zinc (SMD -0.50) or copper (SMD 0.54), while selenium levels were lower in CF patients in one study. No significant differences were observed in erythrocyte CAT activity (SMD -0.41), erythrocyte SOD activity (SMD 0.03), or plasma ceruloplasmin content (SMD 0.62). The analysis highlights specific biochemical markers that are significantly altered in CF patients compared to non-CF controls.

**Conclusions**: In patients with clinically stable cystic fibrosis, there is strong evidence of decreased antioxidant capacity and increased oxidative stress, according to this comprehensive review and meta-analysis.

**Keywords:** Cystic Fibrosis; Biomarkers; Pulmonary Tests; Meta-analysis

## Introduction

Cystic fibrosis (CF) is an autosomal recessive disease that affects more than 70,000 individuals worldwide and shortens life span. Mutations in the gene encoding the CF transmembrane conductance regulator (CFTR), which mainly aids in the efflux of bicarbonate and chloride anions, cause the condition. The cells of the epithelium that line the airways mucous membranes and submucosal glands are primarily where CFTR is expressed.[1,2] Pulmonary infection, inflammation, and a gradual decrease in lung function are all caused by dysfunctional CFTR, which ultimately results in respiratory failure and early mortality.[3,4] The 'gastrointestinal tract, pancreas, sweat ducts, skeletal muscle, cardiovascular system, and reproductive organs' are among the other body systems that contain CFTR.[5] As a result, CF is a complicated multisystem illness that requires ongoing, thorough evaluations in order to track and address health issues.

Variable physiological pathways of signaling that are advantageous for cell functioning and communications are activated by brief increases in free radicals produced from oxygen reactive oxygen species and nitrogen reactive nitrogen species.[6] Nevertheless, a number of complications associated with cystic fibrosis (CF), such as lung disease,[7] inflammation,[8] systemic hypoxemia,[9,10] and dysglycemia,[11] cause an overabundance of reactive oxygen species to be produced, above what is necessary for the best possible physiological performance. Reduced glutathione (GSH), the main non-enzymatic antioxidant, has also been linked to CFTR's efflux.[12] Hence, in vitro impairment of CFTR function increases intracellular GSH buildup, which attenuates the reaction to redox-sensitive pathways of signaling that regulate adaptations to hypoxia[13] and cigarette smoke,[14] thereby increasing the susceptibility of extracellular compartments to oxidative stress.[15] Furthermore, it is probable that variations in the genes encoding glutathione S-transferase (GST) and glutamate-cysteine ligase would further reduce extracellular production and the detoxifying capacity of GSH.[12,16,17] These variables lead to a disparity between oxidants and antioxidants to the benefit of oxidants (i.e., oxidative stress), which disrupts the process of redox signaling and causes damage to molecules.[6] This disparity is compounded by the inadequate absorption of micronutrient with antioxidant capabilities in cystic fibrosis patients.[18]

While CF-related redox discrepancies have been studied since 1967.[19] and have been the focus of multiple 'narrative reviews,[15,20,21] a recent meta-analysis brought attention to the possible importance of these discrepancies in the development and progression of CF.[22] Van et al.[22] discovered that CF had higher levels of F2-isoprostane 8-iso-prostaglandin F2α (8-iso-PGF2α), a biomarker of lipid peroxidation, than other illnesses, which include those linked to oxidative stress-causing environmental triggers and lifestyle variables like cardiovascular illness and cigarette smoking. Although there was some proof of elevated oxidative stress in CF from this research, the number of biomarkers that were taken into consideration was small.[23]

Recent developments in analytical techniques have increased the accessibility of hitherto unobtainable biomarkers for physiological and medical research, such as those utilized to measure oxidative damage in biological samples.[24] Measuring protein carbonyl groups and 8-hydroxy-20-deoxyguanosine (8-OHdG)' can be used to evaluate oxidative changes to proteins and DNA, similar to how elevated levels of 8-iso-PGF2α and malondialdehyde (MDA) indicate lipid peroxidation.[25] Apart from examining the byproducts of oxidative damage, it is now feasible to identify antioxidant deficits that could serve as appropriate targets for therapy.

While vitamins and trace element level monitoring is advised in CF hospitals,[26] quantifying thiols—like cysteine (Cys) and GSH—is also helpful from an antioxidant standpoint, as these substances are critical to comprehending the redox potential (Eh) of biological fluids.[6] Oxidoreductases, like glutathione peroxidase (GPx), catalase (CAT), and superoxide dismutase (SOD), are interesting therapeutic targets because they provide extra functional information about the maintenance of thiol:disulfide pairs.[27] There is a wealth of biomarkers to investigate redox imbalances, however there isn't any solid proof that decreased blood antioxidant levels lead to a state of oxidative stress.

Therefore, in order to offer thorough and up-to-date suggestions for future clinical practice and research trials, a systematic examination of the body of information from studies of redox abnormalities related to cystic fibrosis is required. The purpose of this study was to provide an overview of the research on the possibility of redox anomalies in the blood of individuals with clinically stable cystic fibrosis. Our hypothesis was that individuals with clinically stable cystic fibrosis (CF) would have lower blood markers of antioxidant status compared to non-CF controls, and that their heightened blood markers of oxidative stress would be associated with CF.

## Objective

This review attempted to measure redox abnormalities associated with CF.

## Methodology

Publications that participated defined blood indicators of antioxidant status and/or oxidative stress in CF patients relative to a control group without the disease. We incorporated case-control comparisons from population-based research and baseline case-control comparisons

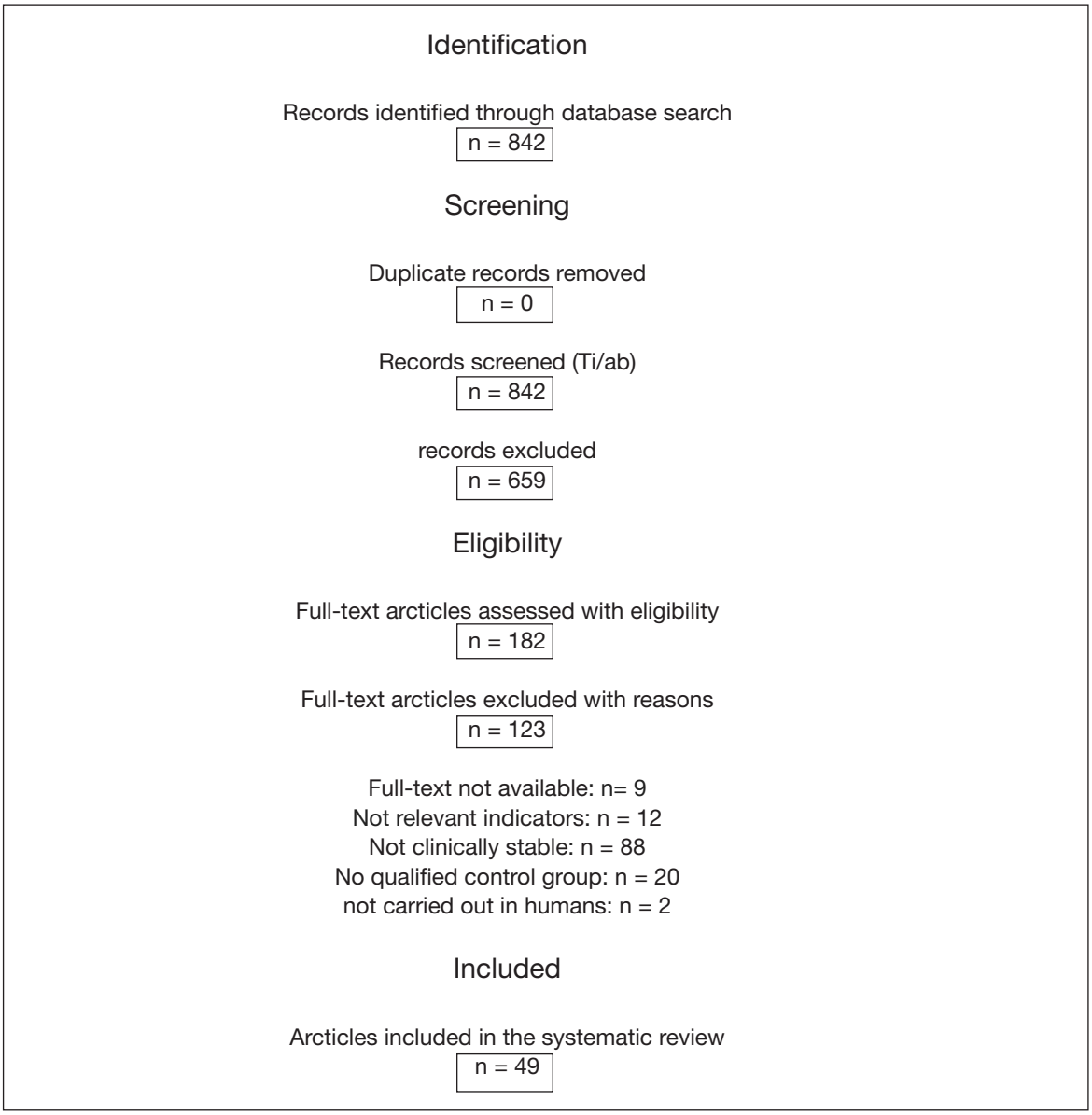


Figure 1. A flow diagram representing the CONSORT method for study identification, screening, and selection.

from intervention trials. The presence of an abnormal sweat test diagnostic genotyping and/or enrolment through a CF clinic is required to establish a clinical diagnosis of CF. Since acute pulmonary exacerbations are a widely recognized cause of antioxidant deficiency and oxidative stress, the study subjects had to be deemed clinically stable and devoid of complaints at the moment of screening.[28] Research that was letters to the editor, case reports, or reviews were not included. Excluded from consideration were studies that contrasted CF patient outcomes with industry-standard values. No constraints were placed on the sort of redox-related biomarker that may be used; however, as CF is linked to renal illness[29] and pelvic floor incontinence,[30] both conditions may change the urine's redox status, the biomarker had to be examined in blood. The duration of publishing was unrestricted. Studies that did not have a full text available in either French or English were disqualified.

The Cochrane Central Register of Controlled Trials (CENTRAL), PsycINFO, Current Nursing and Allied Health Literature (CINAHL), Medline, and PsycINFO databases were thoroughly searched in order to find relevant papers through November 1, 2019.

To find papers that met the criteria for being eligible, this review used a three-part searching procedure, in keeping

with other comparable systematic investigations.[31] The first technique employed the following, Medical topic Heading (MeSH) or topic terms lipid peroxidation MeSH or 'oxidative stress' "MeSH" or 'cystic fibrosis' "MeSH" and 'antioxidants' "MeSH" or 'oxidoreductase' "MeSH" or 'vitamins' "Pharmacological Action" or 'oxidants' "MeSH" and 'blood' "subheading". The second part of the literature search was guided by the set of sixteen prominent markers found in plasma, serum, erythrocyte, or whole blood samples, which was found in eleven independent and qualifying investigations.

The second tactic used specific terms "ti/ab" from the title and/or abstract to identify references. The biological materials and biomarkers found in the "MeSH" search were and-linked to 'cystic fibrosis' in this search in the following ways: 'cystic fibrosis' "tiab" and 'tocopherol' "tiab" or 'carotene' "tiab" or 'malondialdehyde' "tiab" or 'MDA' "tiab" or 'thiobarbituric acid reactive substances' "tiab" or 'TBARS' "tiab" or 'ascorb' "tiab" or 'lycopene' "tiab" or 'protein carbonyl' "tiab" or 'copper' "tiab" or 'isoprostane' "tiab" or 'prostaglandin' "tiab" or 'glutathione' "tiab" or 'hydroperoxide' "tiab" or 'superoxide' "tiab" or 'vitamin "tiab" or 'zinc' "tiab" and 'blood' "tiab" or 'plasma' "tiab" or 'serum' "tiab" or 'erythrocyte' "tiab" or 'whole blood' "tiab". Additionally, the "MeSH search" was run again to provide a thorough, precise and specific search.

Both writers independently executed the 2-step evaluation and selection procedure "AJC and ZLS". The "ti/ab" of all references were evaluated to see if the study fulfilled the eligibility requirements after duplicate publications were eliminated, and papers that weren't suitable were omitted. After that, the complete texts of the relevant citations were obtained for comparison to the qualifying standards. The systematic review contained all relevant references. Meta-analyses were performed using results from a minimum of two separate investigations. Comparable meta-analyses have used these techniques.[31,33]

Using an application designed specifically for reviews, two review authors "AJC and ZLS" separately extracted pertinent information about the research's approach population, drugs, requirements for eligibility, biomarkers and biological fluids examined, and site. When feasible, data about the participants—both case and control—was documented, including their age, sex, genotype, lung function, comorbidities, and medical interventions. We obtained subgroup data (i.e., sorted by severity, genotype, or comorbidities) when accessible. Plot digitizer software was used to gather data from graphics. When data and/or variability measures were missing from an article, they were either computed or obtained by contacting the relevant author. The reference was included in the systematic review but not in the meta-analysis if the data was not made accessible.

The Newcastle-Ottawa Quality Assessment Scale (NOS) for Case-Control research was used by two review writers, "AJC and ZLS," to independently rate the quality of the research [34]. While evaluating the incorporated research, disagreements over assessments were resolved and a compromise was established in order to reduce skew in the scale's perception. This process involved the two writers individually evaluating ten unrelated studies that were left out of the current study. A total of nine points might be obtained for every investigation based on its selection, comparison, and exposure. A study's quality was rated as low if it received a score of 0–2, moderate if it received a score of 3–5, and good if it received a score of 6–9.[33]

The RevMan v5.3 software (Cochrane, UK) was used to calculate group effect measures.[36] Mean differences and '95% confidence intervals (CI) were computed for every research.[36] Standardized mean differences (SMD) were computed since biomarkers were regularly produced in dissimilar units or assessed using dissimilar techniques.

RevMan v5.3, Cochrane UK was used for calculating evaluations of heterogeneity.[36] The I2 statistic was employed in this review to evaluate heterogeneity. The variability in estimates of effect resulting from heterogeneity is expressed as a percentage value by the I2 statistic. Significant heterogeneity is indicated by an I2 of ≥75%.[36] The DerSimonian-Laird method was used to incorporate a random-effects model in the presence of significant heterogeneity, which might be explained by methodological or population-based concerns.[37] Dropped from the meta-analysis was information with significant heterogeneity that was unable to be accounted for by these variables. The information was provided inside the content if variability was unable to be eliminated. Similar meta-analyses have employed these methods.[31,33]

## Results

A schematic representation of the testing procedure is shown in Figure 1. Initially, 842 citations were assessed for inclusion for this systematic review after identical citations were eliminated. 659 of these references were eliminated because they were beyond the purview of this review through the title/abstract screening process. Following the full paper's screening (n = 182; 9 were omitted because the full content was not accessible in either English, the remaining 123 references were eliminated due to the following reasons: the research failed to include relevant indicators (n = 12), the cases were not considered clinically stable (n = 88), there was no qualified control group (n = 20), or the investigation was not carried out in human beings (n = 2).

Data were gathered from 29 centers in Europe, 16 centers in North America, 4 centers in Asia, 3 centers in Oceania, and 2 centers in South America for the research, which were published between 1998 and 2020. A total of 44 research (88.7%) had a cross-sectional design, while 3

non-randomized controlled trials (5.2%), 3 non-randomized and non-controlled trials (5.2%), 1 randomized controlled trial (3.0%), and 1 open-pilot observation (3.0%) also gathered data at baseline. There were 8–233 cases and 5–178 controls in the included studies, which had sample sizes that were generally moderate.

The generally moderate (n = 32; 64.4%) quality of the research includes 13 good-quality studies (29.5%) and 5 low-quality investigations (9.3%). Data missing from six investigations[38-43] was sought since the mean and/or standard deviation (SD) could not be determined. However, none of the authors provided the additional information.

Twelve applicable studies compared the thiol content of blood neutrophils, plasma, serum, and erythrocytes from clinically stable CF patients to non-CF controls. Among the indicators investigated were blood neutrophil GSH' (n = 2),[40,52] plasma or serum protein thiol groups (n = 2),[53,54] erythrocyte GSH (n = 2),[39,51] and plasma or serum albumin (n = 6).[45-50]

Despite the fact that three studies meeting the criteria for inclusion investigated GSH in bloodstream 'neutrophils (n = 2)[40,52] and erythrocytes (n = 2),[39,51] a meta-analysis for either biomarker could not be finished because the mean ± SD for the two trials that provided medians and interquartile ranges could not be computed.[39,40]

Those with clinically stable cystic fibrosis had significantly lower albumin concentrations in their serum or plasma than non-CF controls (SMD -0.99, 95% CI -1.69 to -0.28, p < 0.001, 85%, 5 trials).[45-50] Between individuals with clinically stable cystic fibrosis (CF) and non-CF controls, there was no discernible difference in the levels of protein thiol groups in plasma or serum (SMD -0.36, 95% CI -0.91 to 0.21, p = 0.23, 12.1%, 2 trials).[53,54]

Vitamin levels found in either the serum or plasma of individuals with clinically stable cystic fibrosis (CF) were compared to non-CF controls in twenty-two investigations. The most researched biomarkers for fat-soluble vitamins are, A (vitamin A, retinol, carotene, β-carotene, lutein, and lycopene n = 13),[7,41,42,48,49,55-62] E (vitamin E, vitamin E, cholesterol, ocopherol, and α-tocopherol, n = 13),[7,38,41,42,52,54-56,59-64] D (vitamin D, 25(OH)D and 1,25(OH)2D, n = 11),[44,45,48,50,58-60,65-68] and C (vitamin C, ascorbic acid, and zeaxanthin; n = 5).[7,41,42,57,61]

The plasma or serum concentrations of vitamin A (which includes retinol; SMD-0.67, 95% CI -1.15 to -0.18, p = 0.03, I2 84%, 7 trials) and β-carotene (which includes carotene; SMD -2.18, 95% CI -3.21 to -1.12, p < 0.01, 91%, 4 trials) were consistently lower in CF patients' plasma or serum than in non-CF controls. These non-provitamin A carotenoids were also examined in plasma or serum samples: lutein (SMD -1.53, 95% CI -1.84 to -1.21, p < 0.01, I2 0%, 2 trials),[7,57] lycopene (2 trials; mean ± SD not computable for 1 trial,[42] hence, meta-analysis was not complete)[7,42] and zeaxanthin (SMD 1.48, 95% CI -5.87 to 8.83, p = 0.68, I2 98%, 2 trials).[7,57]

Between individuals with clinically stable cystic fibrosis (CF) and non-CF controls, there was no discernible difference in the concentration of vitamin C (including "ascorbic acid") in the plasma or serum (SMD -0.04, 95% CI -1.84 to 1.73, p = 0.98, I2 94%, 2 trials) [41,61]. Additionally, there was no discernible difference in the plasma or serum concentration of 25(OH)D, which includes "vitamin D," between individuals with clinically stable cystic fibrosis and non-CF controls (SMD -0.24, 95% CI -0.80 to 0.34, p = 0.51, I2 93%, 8 trials) [45,48,50,58–60,66–68].

The metabolite of 25(OH)D, 1,25(OH)2D, was also investigated; however, there was no statistically significant difference in its plasma or serum levels between CF patients and non-CF patients (SMD -0.47, 95% CI -1.13 to 0.21, p=0.18, I2 91%, 4 trials). Whenever possible, the winter months were used to gather the samples. Vitamin E (tocopherol and α-tocopherol) concentrations in plasma or serum were considerably lower in CF patients than in non-CF patients (SMD -0.68, 95% CI -1.14 to -0.22, p < 0.01, I2 88%, 11 trials). Nonetheless, there was no discernible difference in the plasma or serum vitamin E:cholesterol ratio between

CF patients and non-CF patients (SMD -0.36, 95% CI -0.04 to 0.75, p = 0.08, I2 0%, 2 trials).

Eight investigations examined the amounts of trace elements in the plasma or serum of CF

patients to non-CF patients. Zinc was one of the trace elements examined n = 5. selenium n

= 3 and copper n = 2.

There was no discernible difference in plasma or serum copper levels between CF and non-CF subjects ('SMD 0.54, 95% CI -0.26 to 1.32, p = 0.55, I2 69%, 3 trials'). In one trial, plasma or serum selenium levels were considerably lower in persons with cystic fibrosis (CF); however, the second trial's non-computable mean ± SD precluded doing a meta-analysis. Additionally, there was no discernible difference in plasma or serum zinc levels between CF and non-CF subjects (SMD -0.50, 95% CI -1.22 to 0.21, p = 0.17, I2 90%, 6 trials).

Five investigations compared oxidoreductases in CF patients erythrocytes, plasma, or serum to those in non-CF patients. The oxidoreductases that were examined were plasma or serum ceruloplasmin content (n = 2), erythrocyte CAT activity (n = 2), and erythrocyte SOD activity (n = 5).

Erythrocyte CAT activity was not significantly different between CF and non-CF individuals (SMD -0.41, 95% CI -1.52 to 0.73, p = 0.48, I2 79%, 2 trials). Plasma or serum ceruloplasmin content also showed no significant difference (SMD 0.62, 95% CI -0.39 to 1.59, p = 0.24, I2 59%, 2 trials). Erythrocyte SOD activity was not significantly different between the two groups (SMD 0.03, 95% CI -1.17 to 1.21, p = 0.98, I2 97%, 5 trials).

Total antioxidant capacity (TAC) as well as total

antioxidant status (TAS) and trolox equivalent antioxidant status (TEAS) were evaluated in four investigations comparing the plasma or serum of CF patients to non-CF patients. TAC did not differ statistically between those with CF and those without (SMD -0.83, 95% CI -2.01 to 0.39, p = 0.19, I2 95%, 5 trials).

Eleven investigations compared the levels of lipid peroxidation in CF patients blood or plasma to those in non-CF patients. The biomarkers included total 8-iso-PGF2α (n = 2), hydroperoxides (n = 2), and MDA (measured by the thiobarbituric acid reactive substances [TBARS] assay; n = 6).

MDA levels in CF patients were much greater than in non-CF subjects (SMD 1.34, 95% CI 0.44 to 2.25, p < 0.01, I2 97%, 9 trials). In a similar vein, CF patients had significantly higher total '8-iso-PGF2α levels' ('SMD 0.65, 95% CI 0.24 to 1.06, p < 0.01, I2 0%, 3 trials'). Nonetheless, there was no discernible variation in the levels of hydroperoxide (SMD 2.82, 95% CI -2.69 to 8.23, p = 0.35, I2 98%, 3 trials).

## Discussion

This is the first systematic review and meta-analysis to investigate if there are differences between non-CF controls and those with clinically stable cystic fibrosis (CF) in terms of circulating indicators of antioxidant status and oxidative stress. There were 49 studies that matched the eligibility criteria; these included 1675 controls and 1792 CF patients, and 25 biomarkers were deemed suitable for meta-analysis. Two primary conclusions were reached: The plasma or serum of clinically stable cystic fibrosis' patients had considerably low levels of vitamins A and E, β-carotene, lutein, and albumin than non-CF controls; and the patients had significantly higher levels of protein carbonyls, total 8-iso-PG2α, and MDA. These results offer comprehensive evidence that oxidative stress, even in clinically stable patients who did not experience a pulmonary exacerbation at the time of sampling, may be involved in the pathogenesis of cystic fibrosis.

The tripeptide glutathione (GSH) is essential for controlling intracellular redox balances. Four studies that measured GSH in CF patients erythrocytes and blood neutrophils relative to controls were included in this analysis. However, the lack of sufficient data in two trials made meta-analyses impractical. There were no discernible variations in erythrocyte GSH levels between CF patients and controls in the included studies. Adults with mild-to-very severe cystic fibrosis (CF) lung disease showed a substantial negative connection between their FEV1 and erythrocyte GSH levels. This finding raises the possibility that high erythrocyte GSH is a marker of past oxidative damage in the CF lung. It's still unclear if oxidative stress from other conditions, like CF-related diabetes (CFRD), affects erythrocyte GSH amounts in people with cystic fibrosis (CF).

Because deficient CFTR does not contain enough extracellular GSH, it can lead to oxidative stress. However, functional CFTR is permeable to GSH. Since GSH in plasma quickly oxidizes to generate glutathione disulfide (GSSG), no research in this review measured plasma GSH. The bulk of the plasma protein thiol pool, plasma or serum albumin, was much smaller in CF patients, despite two investigations finding no differences in plasma protein thiol groups between CF patients and controls. Malnutrition and liver dysfunction are two comorbidities that might affect serum albumin levels in people with cystic fibrosis (CF). There were no trials with a single homozygous genotype group, and only two trials recorded participant genotypes. Future research have to account for genotype and look into variations in thiol concentrations amongst genotype-classified subgroups.

Future studies ought to investigate thiol:disulfide pairs in the blood of CF patients who are in a clinically stable state. For example, total plasma GSH was shown to be approximately 40% lower in CF patients compared to healthy controls by Roum et al. However, this study was eliminated since the clinical stability of the cases was not obvious. In CF patients, oxidative disturbances may have a higher effect on redox potential (Eh) due to lower GSH concentrations. Other thiol:disulfide pairs, including thioredoxin and Cys:cystine (Cyss), should also be taken into account. Although GSH synthesis is regulated by extracellular Cys pools, GSH:GSSG and Cys:Cyss serve separate purposes. Reduced Cys:Cyss ratios can cause mitochondria to produce reactive oxygen species, which can activate Nrf2 and other antioxidant transcription factors.

Vitamins dissolved in fat and water are examples of exogenous antioxidants. Though just two moderate-to-high quality trials addressed this, vitamin C levels were not substantially different between CF patients and controls when it came to water-soluble micronutrients. In one study, vitamin C levels were higher in CF children getting ascorbic acid supplements, but in another, vitamin C levels were lower in CF adolescents who did not take multivitamins. Vitamins A and E, β-carotene, lutein, and 25(OH)D are among the fat-soluble micronutrients that were considerably reduced in CF patients; however, 1,25(OH)2D and the vitamin E:cholesterol ratio did not differ significantly. These analyses' heterogeneity was probably caused by variations in the measurement units, supplementation procedures, and demographics of the populations they included.

Wood et al. discovered noteworthy inverse relationships between lipid peroxidation indicators and vitamins C, E, and β-carotene, implying that heightened generation of free radicals could weaken antioxidant defenses in CF patients who do not take supplements. FEV1 and the Shwachman-Kulzycki score are two clinical outcomes that have a good correlation with vitamins C and E. In CF patients, multivitamin supplementation can increase

antioxidant levels; nevertheless, there are drawbacks, including inadequate dose, poor adherence, and nutrient malabsorption. The development of therapies to reduce oxidative stress in cystic fibrosis is complicated by these circumstances, as indicated by the inconsistent outcomes of research including supplementation with vitamins.

The trace substances in blood have antioxidant activities and affect oxidoreductase activity in alongside vitamins. There were no variations in plasma or serum copper and zinc 'levels between CF patients and controls' in any of the eight studies that looked at trace element levels in CF patients. Although a meta-analysis on selenium was not feasible, no noteworthy variations were documented. Although its involvement in oxidative stress is still unknown, certain studies have revealed links between clinical outcomes and zinc levels, indicating that low zinc may be related to serious lung conditions and starvation.

Antioxidant enzymes found in blood directly combat oxidative stress and preserve thiol:disulfide ratios. Throughout five studies, there were no discernible variations in erythrocyte activity between CF patients and controls for superoxide dismutase (SOD), the most researched enzyme. The findings were not all the same; other research reported reduced, increased, or no change in SOD activity. Technical variations and undocumented dietary consumption or supplementation may be the cause of this variability. Erythrocyte SOD activity may be impacted by iron shortage, which is prevalent in CF patients. In general, it is still unknown how circulatory SOD activity affects the pathogenesis of CF.

There were no appreciable variations observed in CF patients' plasma/serum ceruloplasmin concentration or erythrocyte catalase (CAT) activity, two other oxidoreductases that were reviewed. Further investigations revealed normal levels of glutathione peroxidase (GPx) in whole blood and plasma, reduced levels of plasma diamine oxidase, higher levels of serum glutathione S-transferase (GSTα and GSTα1), higher levels of 'erythrocyte glutathione reductase (GR) activity, and normal levels of serum γ-glutamyl transferase' (GGT). Due to a lack of trials, 'these biomarkers were not included in a meta-analysis'. To ascertain whether circulating oxidoreductases are promising targets for CF therapy, more investigation is required.

As a mediator of the pathogenesis of numerous long-term illnesses, lipid peroxidation is measured with great importance using biomarkers such as 8-iso-PGF2α, which is the end byproduct of arachidonic acid oxidation. Just two investigations that showed increased total 8-iso-PGF2α in CF patients compared to controls were found to be eligible for meta-analysis despite the substantial study on 8-iso-PGF2α in illnesses. Nonetheless, inferences on particular lipid peroxidation increases against general inflammatory activity are limited due to the lack of data on free 8-iso-PGF2α. Future research should examine spirometry and longitudinal changes in 8-iso-PGF2α to determine whether this marker is appropriate for use as therapy in cystic fibrosis.

MDA was the most extensively researched lipid peroxidation biomarker in cystic fibrosis (CF), and a meta-analysis revealed considerably higher levels in cystic fibrosis plasma/serum, but with significant heterogeneity because of varying assay techniques. On the other hand, there was no significant difference in plasma/serum lipid hydroperoxides between the CF and control groups, which highlights the difficulty in determining lipid peroxidation in biological materials.

Lipid peroxidation indicators and immune cell counts correlate, indicating a connection between increased 8-iso-PGF2α and increased inflammatory responses in cystic fibrosis patients. Lipid peroxidation's involvement in non-pulmonary CF sequelae, such dysglycemia connected to CF, is still poorly understood, which calls for more investigation into particular CF subgroups.

When comparing CF plasma/serum to controls, there was a substantial rise in protein carbonylation, a marker of oxidative damage to proteins. Nevertheless, unlike results in other chronic illnesses where protein carbonylation corresponds with skeletal muscle weakness, investigations did not show relationships with clinical results.

Peripheral skeletal muscle impairment in cystic fibrosis (CF) can be caused by oxidative metabolic abnormalities, inflammation, and loss of nutrients. According to recent research, adolescents with cystic fibrosis (CF) produce more alkoxyl radicals when they exercise, which may be a contributing factor to their intolerance to exercise. Oxidative stress control in CF skeletal muscle is further complicated by mitochondrial abnormalities linked to faulty CFTR.

Although increased protein carbonylation in CF is highlighted by this meta-analysis, next research should elucidate its clinical relevance by prospectively examining the relationships between protein carbonylation biomarkers and pertinent outcomes. Knowing about redox imbalances may help with understanding mitochondrial function and oxidative capability in skeletal muscle, two areas that may be targeted for CF therapy.

## Limitations

Numerous intrinsic limitations were faced by this systematic review and meta-analysis. There are still gaps in the literature regarding the influence of genotype, age, and sex on redox biomarkers. Longitudinal research is required to corroborate these findings, even though correlations from included studies indicate a negative link between antioxidants and age and a positive relationship between oxidative stress indicators and age. The high level of biomarker heterogeneity was a major drawback, partially

because so few trials matched controls and patients according to age and other variables. To minimize heterogeneity, controls and cases in subsequent trials should be matched based on age and, ideally, sex. Heterogeneity was also influenced by differences in assay and unit of measurement methodologies.

The age of some included studies poses another limitation. Since the oldest study dates back to 1977, advancements in medicine and CF treatments like CFTR modulators may influence redox imbalances differently today. Future research should investigate whether CFTR modulator treatments alter observed redox imbalances, representing a crucial area for exploration.

## Future Research Directions

This review focused on antioxidant and oxidative stress biomarkers sampled from clinically-stable CF cases, excluding hospitalized individuals. Pulmonary exacerbations in CF are associated with increased lung infection, inflammation, acute dysglycemia, muscle weakness, and weight loss, potentially increasing biomarkers of oxidative damage. Despite treatments for pulmonary exacerbations potentially enhancing antioxidant vitamin levels in blood plasma, elevated lipid peroxidation and protein carbonylation persist despite improved lung function. Future trials should explore whether post-exacerbation redox imbalances correlate with clinical outcomes such as time to subsequent hospitalization.

Several vital biomarkers crucial for understanding in vivo oxidative stress in CF have not been systematically studied on a case-control basis in blood samples from clinically-stable CF individuals. For instance, DNA oxidation can be quantified by circulating 8-OHdG, which has shown elevation in various diseases but remains understudied in CF. Additionally, CF-associated comorbidities like renal disease may limit the use of urinary samples for systemic oxidative stress indices, underscoring the need for confirmation in blood samples.

Endothelial dysfunction, increasingly recognized in CF, may exacerbate with dysglycemia and involve oxidative stress pathways. CFTR expression in endothelial cells suggests a role in modulating pro-inflammatory and pro-oxidant pathways, impacting nitric oxide availability and potentially elevating nitrotyrosine levels, a marker of nitrosative stress. Future studies should quantify nitrotyrosine levels in blood from clinically-stable CF individuals versus controls, building on prior findings from exhaled breath condensate and sputum analyses.

## Conclusion

This analysis methodically shows that, in comparison to controls, clinically-stable CF patients have lower levels of some circulating biomarkers of antioxidant capacity and higher levels of oxidative stress. To clarify the effects of oxidative stress on the pathogenesis of CF-related illnesses like lung disease, diabetes mellitus, and anomalies in skeletal muscle, more research is required.

## References


1. Elborn JS. Adult care in cystic fibrosis. InSeminars in respiratory and critical care medicine. 2019; 40(06): 857-68. Thieme Medical Publishers.
2. Montgomery ST, Mall MA, Kicic A, Stick SM. Hypoxia and sterile inflammation in cystic fibrosis airways: mechanisms and potential therapies. Eur Respir J. 2017;49:1-13. DOI 10.1183/13993003.00903-2016.
3. Pillarisetti N, Williamson E, Linnane B, Skoric B, Robertson CF, Robinson P, et al. Infection, inflammation, and lung function decline in infants with cystic fibrosis. Am J Respir Crit Care Med. 2011; 184:75–81.
4. Foundation CF, Patient Registry: Annual Data Report. 2016.
5. Wang Y, Wrennall JA, Cai Z, Li H, Sheppard DN. Sheppard, Understanding how cystic fibrosis mutations disrupt CFTR function: from single molecules to animal models. Int J Biochem Cell Biol. 2014;52:47–57. DOI: 10.1016/j.biocel. 2014.04.001.
6. Halliwell B, Gutteridge JM. Free radicals in biology and medicine. Oxford university press, USA; 2015.
7. Lagrange-Puget M, Durieu I, Ecochard RR, Abbas-Chorfa F, Drai J, Steghens JPP, et al. Longitudinal study of oxidative status in 312 cystic fibrosis patients in stable state and during bronchial exacerbation. Pediatr Pulmonol. 2004;38:43–49. DOI: 10.1002/ppul.20041.
8. Lee I-T, Yang C-M. Role of NADPH oxidase/ROS in pro-inflammatory mediators-induced airway and pulmonary diseases. Biochem Pharmacol. 2012; 84:581–590. DOI: 10.1016/J.BCP.2012.05.005.
9. Urquhart DS, Montgomery H, Jaffe A. Assessment of hypoxia in children with cystic fibrosis. Arch Dis Child. 2005;90:1138–1143. DOI: 10.1136/adc. 2005.071795.
10. Yuhai GU, Zhen Z. Significance of the changes occurring in the levels of interleukins, SOD and MDA in rat pulmonary tissue following exposure to different altitudes and exposure times. Exp Ther Med. 2015;10:915–920. DOI: 10.3892/etm. 2015.2604.
11. Ntimbane T, Krishnamoorthy P, Huot C, Legault L, Jacob SV, Brunet S, et al. Oxidative stress and cystic fibrosis-related diabetes: a pilot study in children. J Cyst Fibros. 2008;7:373–384. DOI: 10.1016/j. jcf.2008.01.004.
12. Linsdell P, Hanrahan JW. Glutathione permeability of

CFTR. Am J Physiol. 1998;275

13. Duranton C, Rubera I, Cougnon M, Melis N, Chargui A, Mograbi B, Tauc M. CFTR is involved in the fine-tuning of intracellular redox status: physiological implications in cystic fibrosis. Am J Pathol. 2012;181: 1367–1377. DOI: 10.1016/j.ajpath. 2012.06.017.
14. Gould NS, Min E, Martin RJ, Day BJ. CFTR is the primary known apical glutathione transporter involved in cigarette smoke-induced adaptive responses in the lung. Free Radic Biol Med. 2012;52:1201–1206. DOI: 10.1016/j.freeradbiomed. 2012.01.001.
15. Ziady AG, Hansen J. Redox balance in cystic fibrosis. Int J Biochem Cell Biol. 2014;52:113–123. DOI: 10.1016/j.biocel.2014.03.006.
16. de Lima Marson FA, Bertuzzo CS, Secolin R, Ribeiro AF, Ribeiro JD. Genetic interaction of GSH metabolic pathway genes in cystic fibrosis. BMC Med Genet. 2013;14:60. DOI: 10.1186/1471-2350-14-60.
17. de Lima Marson FA, Bertuzzo CS, Ribeiro AF, Ribeiro JD. Polymorphisms in the glutathione pathway modulate cystic fibrosis severity: a cross-sectional study. BMC Med Genet. 2014;15:27. DOI: 10.1186 /1471-2350-15-27.
18. Siwamogsatham O, Dong W, Binongo JN, Chowdhury R, Alvarez JA, Feinman SJ, et al. Relationship between fat-soluble vitamin supplementation and blood concentrations in adolescent and adult patients with cystic fibrosis. Nutr Clin Pract. 2014;29:491–497. DOI: 10.1177/088453361 4530170.
19. Bennett MJ, Medwadowski BF. Vitamin A, vitamin E, and lipids in serum of children with cystic fibrosis or congenital heart defects compared with normal children. Am J Clin Nutr. 1967;20:415–421. DOI: 10.1093/ajcn/20.5.415.
20. Galli F, Battistoni A, Gambari R, Pompella A, Bragonzi A, Pilolli F, et al. Oxidative stress and antioxidant therapy in cystic fibrosis. Biochim Biophys Acta Mol Basis Dis. 2012;1822:690–713. DOI: 10.1016/j. bbadis.2011.12.012.
21. Ntimbane T, Comte B, Mailhot G, Berthiaume Y, Poitout V, Prentki M, et al. Cystic fibrosis-related diabetes: from CFTR dysfunction to oxidative stress. Clin Biochem Rev. 2009;30:153–177.
22. van 't Erve TJ, Kadiiska MB, London SJ, Mason RP. Classifying oxidative stress by F2-isoprostane levels across human diseases: a meta-analysis. Redox Biol. 2017;12:582–599. DOI: 10.1016/j.redox.2017. 03.024.
23. Cobley JN, Close GL, Bailey DM, Davison GW. Exercise redox biochemistry: conceptual, methodological and technical recommendations. Redox Biol. 2017;12:540–548. DOI: 10.1016/j.redox. 2017.03.022.
24. Frijhoff J, Winyard PG, Zarkovic N, Davies SS, Stocker R, Cheng D, et al. Clinical relevance of biomarkers of oxidative stress. Antioxid Redox Signal. 2015;23:1144–1170. DOI: 10.1089/ars.2015. 6317.
25. Halliwell B, Whiteman M. Measuring reactive species and oxidative damage in vivo and in cell culture: how should you do it and what do the results mean? Br J Pharmacol. 2004;142:231–255. DOI: 10.1038/sj.bjp. 0705776.
26. Turck D, Braegger CP, Colombo C, Declercq D, Morton A, Pancheva R, et al. ESPEN-ESPGHAN-ECFS guidelines on nutrition care for infants, children, and adults with cystic fibrosis. Clin Nutr. 2016;35:557–577. DOI: 10.1016/j.clnu.2016.03.004.
27. Younus H. Therapeutic potentials of superoxide dismutase. Int J Health Sci. 2018;12:88–93. Available from URL: http://www.ncbi.nlm.nih.gov/pubmed/ 29896077.
28. McGrath LT, Mallon P, Dowey L, Silke B, McClean E, McDonnell M, et al. Oxidative stress during acute respiratory exacerbations in cystic fibrosis. Thorax. 1999;54:518–523. DOI: 10.1136/thx.54.6.518.
29. Nazareth D, Walshaw M. A review of renal disease in cystic fibrosis. J Cyst Fibros. 2013;12:309–319. DOI: 10.1016/j.jcf.2013.03.005.
30. Neemuchwala F, Ahmed F, Nasr SZ. Prevalence of pelvic incontinence in patients with cystic fibrosis. Glob Pediatr Health. 2017;4:2333794X17743424. DOI: 10.1177/2333794X17743424.
31. Frustaci A, Neri M, Cesario A, Adams JB, Domenici E, Dalla Bernardina B, et al. Oxidative stress-related biomarkers in autism: systematic review and meta-analyses. Free Radic Biol Med. 2012;52:2128–2141. DOI: 10.1016/j.freeradbiomed. 2012.03.011.
32. Ugolini D, Neri M, Casilli C, Bonassi S. Development of search filters for retrieval of literature on the molecular epidemiology of cancer. Mutat Res Genet Toxicol Environ Mutagen. 2010;701:107–110. DOI: 10.1016/j.mrgentox. 2010.05.016.
33. Murri M, Luque-Ramírez M, Insenser M, Ojeda-Ojeda M, Escobar-Morreale HF. Circulating markers of oxidative stress and polycystic ovary syndrome (PCOS): a systematic review and meta-analysis. Hum Reprod Update. 2013;19:268–288. DOI: 10.1093 /humupd/dms059.
34. Wells G, Shea B, O'Connell D, Peterson J, Welch V, Losos M, et al. The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomized studies in

meta-analysis. Proc 3rd Symp Syst Rev Beyond Basics Improv Qual Impact, Oxford, 2000, pp. 3–5.

35. Irwig L, Macaskill P, Berry G, Glasziou P. Bias in meta-analysis detected by a simple, graphical test. Graphical test is itself biased. BMJ. 1998;316. Available from URL: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2127453/pdf/9310563.pdf.
36. Higgins J, Green S. Cochrane Handbook for Systematic Reviews of Interventions. John Wiley & Sons, Chichester, United Kingdom, 2015. Available from URL: http://handbook.cochrane.org/.
37. Dersimonian R, Laird N. Meta-analysis in clinical trials. Stat Med. 1986;188:177–188. DOI: 10.1016/0197-2456(86)90046-2.
38. James DR, Alfaham M, Goodchild MC. Increased susceptibility to peroxide-induced haemolysis with normal vitamin E concentrations in cystic fibrosis. Clin Chim Acta. 1991;204:279–290.
39. Bernardi DM, Ribeiro AF, Mazzola TN, Vilela MMS, Sgarbieri VC. The impact of cystic fibrosis on the immunologic profile of pediatric patients. J Pediatr. 2013;89:40–47. DOI: 10.1016/j.jped.2013.02.007.
40. Tirouvanziam R, Conrad CK, Bottiglieri T, Herzenberg LA, Moss RB, Herzenberg LA. High-dose oral N-acetylcysteine, a glutathione prodrug, modulates inflammation in cystic fibrosis. Proc Natl Acad Sci USA. 2006;103:4628–4633. DOI: 10.1073/pnas.0511304103.
41. Wood LG, Fitzgerald DA, Gibson PG, Cooper DM, Collins CE, Garg ML. Oxidative stress in cystic fibrosis: dietary and metabolic factors. J Am Coll Nutr. 2001;20:157–165. DOI: 10.1080/07315724.2001.10719028.
42. Back EI, Frindt C, Nohr D, Frank J, Ziebach R, Stern M, Ranke M, Biesalski HK. Antioxidant deficiency in cystic fibrosis: when is the right time to take action? Am J Clin Nutr. 2004;80:374–384. DOI: 10.1093/ajcn/80.2.374.
43. Durieu I, Vericel E, Guichardant D, Roth H, Steghens JP, Drai J, Nove Josserand R, Fontaine E, Lagarde M, Bellon G. Fatty acids platelets and oxidative markers following intravenous n-3 fatty acids administration in cystic fibrosis: an open pilot observational study. J Cyst Fibros. 2007;6:320–326. DOI: 10.1016/j.jcf.2006.12.005.
44. Nicolaidou P, Stavrinadis I, Loukou I, Papadopoulou A, Georgouli H, Douros K, et al. The effect of vitamin K supplementation on biochemical markers of bone formation in children and adolescents with cystic fibrosis. Eur J Pediatr. 2006;165:540–545. DOI: 10.1007/s00431-006-0132-1.
45. Lee MJ, Kearns MD, Smith EM, Hao L, Ziegler TR, Alvarez JA, et al. Free 25-hydroxyvitamin D concentrations in cystic fibrosis. Am J Med Sci. 2015;350:374–379. DOI: 10.1097/MAJ.0000000000000592.
46. Kearns GL, Crom WR, Karlson KH, Mallory GB, Evans WE. Hepatic drug clearance in patients with mild cystic fibrosis. Clin Pharmacol Ther. 1996;59:529–540. DOI: 10.1016/S0009-9236(96)90181-2.
47. Mocchegiani E, Provinciali M, Di Stefano G, Nobilini A, Caramia G, Santarelli L, et al. Role of the low zinc bioavailability on cellular immune effectiveness in cystic fibrosis. Clin Immunol Immunopathol. 1995;75:214–224. DOI: 10.1006/clin.1995.1074.
48. Hahn TJ, Squires AE, Halstead LR, Strominger DB. Reduced serum 25-hydroxyvitamin D concentration and disordered mineral metabolism in patients with cystic fibrosis. J Pediatr. 1979;94:38–42.
49. Jacob RA, Sandstead HH, Solomons NW, Rieger C, Rothberg R. Zinc status and vitamin A transport in cystic fibrosis. Am J Clin Nutr. 1978;31:638–644.
50. Stead RJ, Houlder S, Agnew J, Thomas M, Hodson ME, Batten JC, et al. Vitamin D and parathyroid hormone and bone mineralisation in adults with cystic fibrosis. Thorax. 1988;43:190–194.
51. Mangione S, Patel DD, Levin BR, Fiel SB. Erythrocytic glutathione in cystic fibrosis: a possible marker of pulmonary dysfunction. Chest. 1994;105:1470–1473. DOI: 10.1378/chest.105.5.1470.
52. Olveira G, Olveira C, Dorado A, García-Fuentes E, Rubio E, Tinahones F, et al. Cellular and plasma oxidative stress biomarkers are raised in adults with bronchiectasis. Clin Nutr. 2013;32:112–117. DOI: 10.1016/j.clnu.2012.06.002.
53. Sadowska-Bartosz I, Galiniak S, Bartosz G, Rachel M. Oxidative modification of proteins in pediatric cystic fibrosis with bacterial infections. Oxid Med Cell Longev. 2014;389629. DOI: 10.1155/2014/389629.
54. McGrath LT, McCall D, Hanratty CG, Brennan S, Devine A, McCauley DF, et al. Individuals with cystic fibrosis do not display impaired endothelial function or evidence of oxidative damage in endothelial cells exposed to serum. Clin Sci (Lond). 2001;101:507–13. DOI: 10.1042/CS20010114.
55. Hubbard VS, Dunn GD, di Sant'Agnese PA. Abnormal fatty-acid composition of plasma-lipids in cystic fibrosis. A primary or a secondary defect? Lancet. 1977;310:1302–1304. DOI: 10.1016/S0140-6736(77)90359-2.
56. Winklhofer-Roob BM, Puhl H, Khoschsorur GA, van't Hof MA, Esterbauer H, Shmerling DH. Enhanced

resistance to oxidation of low density lipoproteins and decreased lipid peroxide formation during beta-carotene supplementation in cystic fibrosis. Free Radic Biol Med. 1995;18:849–859. DOI: 10.1016/0891-5849(94)00203-v.

57. Schupp C, Olano-Martin E, Gerth C, Morrissey BM, Cross CE, Werner JS. Lutein, zeaxanthin, macular pigment, and visual function in adult cystic fibrosis patients. Am J Clin Nutr. 2004;79:1045-52. Available from URL: http://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=2603302&tool=pmcentrez&rendertype=abstract.
58. Olveira C, Padilla A, Dorado A, Contreras V, Garcia Fuentes E, Rubio Martin E, et al. Inflammation and oxidation biomarkers in patients with cystic fibrosis: the influence of azithromycin. Eurasian J Med. 2017;49:118–123. DOI: 10.5152/eurasianjmed.2017.17010.
59. Yadav K, Singh M, Angurana SK, Attri SV, Sharma G, Tageja M, et al. Evaluation of micronutrient profile of North Indian children with cystic fibrosis: a case-control study. Pediatr Res. 2014;75:762–766. DOI: 10.1038/pr.2014.30.
60. Ambroszkiewicz J, Sands D, Gajewska J, Chelchowska M, Laskowska-Klita T. Bone turnover markers, osteoprotegerin and RANKL cytokines in children with cystic fibrosis. Adv Med Sci. 2013;58:338–343. DOI: 10.2478/ams-2013-0011.
61. Madarasi A, Lugassi A, Greiner E, Holics K, Biró L, Mozsary E. Antioxidant status in patients with cystic fibrosis. Ann Nutr Metab. 2000;44:207–211. DOI: 10.1159/000046685.
62. Vaisman N, Kerasin E, Hahn T, Trifon S, Voet H, Tabachnik E. Increased neutrophil chemiluminescence production in patients with cystic fibrosis. Metabolism. 1994;43:719–722. DOI: 10.1016/0026-0495(94)90120-1.
63. Oudshoorn JH, Lecluse AL, van den Berg RL, Vaes WHJ, van der Laag J, Houwen RHJ. Decreased coenzyme Q10 concentration in plasma of children with cystic fibrosis. J Pediatr Gastroenterol Nutr. 2006;43:646–650. Available from URL: http://www.ncbi.nlm.nih.gov/pubmed/17130743.
64. Dominguez C, Gartner S, Linan S, Cobos N, Moreno A. Enhanced oxidative damage in cystic fibrosis patients. Biofactors. 1998;8:149–153.
65. Reiter EO, Brugman SM, Pike JW, Pitt S, Dokoh S, Haussler MR, et al. Vitamin D metabolites in adolescents and young adults with cystic fibrosis: effects of sun and season. J Pediatr. 1985;106:21–26. DOI: 10.1016/S0022-3476(85)80458-3.
66. Cobanoglu N, Atasoy H, Ozcelik U, Yalcin E, Dogru D, Kiper N, Gocmen A. Relation of bone mineral density with clinical and laboratory parameters in prepubertal children with cystic fibrosis. Pediatr Pulmonol. 2009;44:706–712. DOI: 10.1002/ppul.21050.
67. Rovner AJ, Stallings VA, Schall JI, Leonard MB, Zemel BS. Vitamin D insufficiency in children, adolescents, and young adults with cystic fibrosis despite routine oral supplementation. Am J Clin Nutr. 2007;86:1694–1699. DOI: 10.1093/ajcn/86.6.1694.
68. Aris RM, Ontjes DA, Buell HE, Blackwood AD, Lark RK, Caminiti M, et al. Abnormal bone turnover in cystic fibrosis adults. Osteoporos Int. 2002;13:151–157. DOI: 10.1007/s001980200007.
69. Wahab A, Shahin A, Allangawi M, Chandra P, Abdel Rahman MO, Soliman A. Serum zinc concentration in cystic fibrosis patients with CFTRI1234V mutation associated with pancreatic sufficiency. Clin Res J. 2017;11:305–310. DOI: 10.1111/crj.12335.
70. Percival SS, Kauwell GPA, Bowser E, Wagner M. Altered copper status in adult men with cystic fibrosis. J Am Coll Nutr. 1999;18:614–619. DOI: 10.1080/07315724.1999.10718896.
71. Best K, McCoy K, Gemma S, Disilvestro RA. Copper enzyme activities in cystic fibrosis before and after copper supplementation plus or minus zinc. Metabolism. 2004;53:37–41. DOI: 10.1016/j.metabol.2003.07.017.
72. Sadowska-Woda I, Rachel M, Pazdan J, Bieszczad-Bedrejczuk E, Pawliszak K. Nutritional supplement attenuates selected oxidative stress markers in pediatric patients with cystic fibrosis. Nutr Res. 2011;31:509–518. DOI: 10.1016/j.nutres.2011.07.002.
73. Lands LC, Grey VL, Grenier C. Total plasma antioxidant capacity in cystic fibrosis. Pediatr Pulmonol. 2000;29:81–87. DOI: 10.1002/(SICI)1099-0496(200002)29:2<81::AID-PPUL1>3.0.CO;2-N.
74. Antus B, Drozdovszky O, Barta I, Kelemen K. Comparison of airway and systemic malondialdehyde levels for assessment of oxidative stress in cystic fibrosis. Lung. 2015;193:597–604. DOI: 10.1007/s00408-015-9739-1.
75. Collins CE, Quaggiotto P, Wood L, O'Loughlin EV, Henry RL, Garg ML. Elevated plasma levels of F2(α) isoprostane in cystic fibrosis. Lipids. 1999;34:551–556. DOI: 10.1007/s11745-999-0397-1.
76. Jones DP, Carlson JL, Mody VC, Cai J, Lynn MJ, Sternberg P. Redox state of glutathione in human plasma. Free Radic Biol Med. 2000;28:625–635. DOI: 10.1016/S0891-5849(99)00275-0.
77. Stonebraker JR, Ooi CY, Pace RG, Corvol H, Knowles MR, Durie PR, et al. Features of severe liver disease

with portal hypertension in patients with cystic fibrosis. Clin Gastroenterol Hepatol. 2016;14: 1207–1215. DOI: 10.1016/j.cgh. 2016.03.041.

78. Souza dos Santos Simon MI, Drehmer M, de Abreu e Silva FA, Hoffmann A, Druck Ricachinewsky C, de Fonseca Andrade Procianoy E, et al. Association of nutritional status, plasma albumin levels and pulmonary function in cystic fibrosis. Nutr Hosp. 2011;26:1322–1327. DOI: 10.3305/nh.2011. 26.6.4931.
79. Roum JH, Buhl R, McElvaney NG, Borok Z, Crystal RG. Systemic deficiency of glutathione in cystic fibrosis. J Appl Physiol. 1993;75:2419–2424. DOI: 10.1152/jappl.1993.75.6.2419.
80. Jones DP, Go YM, Anderson CL, Ziegler TR, Kinkade JM, Kirklin WG. Cysteine/cystine couple is a newly recognized node in the circuitry for biologic redox signaling and control. Faseb J. 2004;18: 1246–1248. DOI: 10.1096/fj.03-0971fje.
81. Cheng Z, Zhang J, Ballou DP, Williams CH Jr. Reactivity of thioredoxin as a protein thiol-disulfide oxidoreductase. Chem Rev. 2011;111:5768–5783. DOI: 10.1021/cr100006x.
82. Imhoff BR, Hansen JM. Extracellular redox status regulates Nrf2 activation through mitochondrial reactive oxygen species. Biochem J. 2009;424: 491–500. DOI: 10.1042/bj20091286.
83. Chen J, Kinter M, Shank S, Cotton C, Kelley TJ, Ziady AG. Dysfunction of Nrf-2 in CF epithelia leads to excess intracellular H2O2 and inflammatory cytokine production. PLoS One. 2008;3. DOI: 10.1371/journal.pone.0003367.
84. Ciofu O, Smith S, Lykkesfeldt J. Antioxidant supplementation for lung disease in cystic fibrosis. Cochrane Database Syst Rev. 2019. DOI: 10.1002/ 14651858.CD007020.pub4.
85. Madarasi A, Lugassi A, Greiner E, Holics K, Biro L, Mozsary E. Antioxidant status in patients with cystic fibrosis. Ann Nutr Metab. 2000;44:207–211. DOI: 10.1159/000046685.
86. Nebot C, Moutet C, Huet P, Xu JZ, Yadan JC, Chaudiere J. Spectrophotometric assay of superoxide dismutase activity based on the activated autoxidation of a tetracyclic catechol. Anal Biochem. 1993;214:442–451. DOI: 10.1006/abio. 1993.1521.
87. Gifford AH, Miller SD, Jackson BP, Hampton TH, O'Toole GA, Stanton BA, Parker HW. Iron and CF-related anemia: expanding clinical and biochemical relationships. Pediatr Pulmonol. 2011;46:160–165. DOI: 10.1002/ppul.21335.
88. Sidlova K, Skalicka V, Kotaska K, Pechova M, Chada M, Bartosova J, et al. Serum α-glutathione S-transferase as a sensitive marker of hepatocellular damage in patients with cystic fibrosis. Physiol Res. 2003;52:361–365.
89. Hung JC, Howie AF, Beckett GJ, Sood M, Hambleton G, Super M. The use of human glutathione S-transferase A1 in the detection of cystic fibrosis liver disease. J Paediatr Child Health. 1998;34:335–338.
90. Pradas I, Jove M, Huynh K, Puig J, Ingles M, Borras C, et al. Exceptional human longevity is associated with a specific plasma phenotype of ether lipids. Redox Biol. 2019;101127. DOI: 10.1016/J.REDOX. 2019.101127.
91. Milne GL. Classifying oxidative stress by F2-Isoprostane levels in human disease: the re-imagining of a biomarker. Redox Biol. 2017;12:897–898. DOI: 10.1016/j.redox. 2017. 04.028.
92. Milne GL, Musiek ES, Morrow JD. F2-Isoprostanes as markers of oxidative stress in vivo: an overview. Biomarkers. 2005;10:10–23. DOI: 10.1080/ 13547500500216546.
93. van 't Erve TJ, Lih FB, Kadiiska MB, Deterding LJ, Eling TE, Mason RP. Reinterpreting the best biomarker of oxidative stress: the 8-iso-PGF2α/ PGF2α ratio distinguishes chemical from enzymatic lipid peroxidation. Free Radic Biol Med. 2015;83: 245–251. DOI: 10.1016/J.FREERADBIOMED. 2015. 03.004.
94. Forman HJ, Augusto O, Brigelius-Flohe R, Dennery PA, Kalyanaraman B, Ischiropoulos H, Mann GE, et al. Even free radicals should follow some rules: a guide to free radical research terminology and methodology. Free Radic Biol Med. 2015;78: 233–235. DOI: 10.1016/j.freeradbiomed. 2014.10. 504.
95. D'Orazio M, Pacello F, Battistoni A. Extracellular glutathione decreases the ability of Burkholderia cenocepacia to penetrate into epithelial cells and to induce an inflammatory response. PLoS One. 2012;7. DOI: 10.1371/journal.pone.0047550.
96. Montuschi P, Ciabattoni G, Paredi P, Pantelidis P, Bois RM, Kharitonov SA, Barnes PJ. 8-Isoprostane as a biomarker of oxidative stress in interstitial lung diseases. Diseases. 1998;158:1524–1527.
97. Leclercq A, Gauthier B, Rosner V, Weiss L, Moreau F, Constantinescu AA, Kessler R, et al. Early assessment of glucose abnormalities during continuous glucose monitoring associated with lung function impairment in cystic fibrosis patients.

J Cyst Fibros. 2014;13:478–484. DOI: 10.1016/j.jcf. 2013.11.005.

98. Koch C, Rainisio M, Madessani U, Harms HK, Hodson ME, Mastella G, et al. Presence of cystic fibrosis-related diabetes mellitus is tightly linked to poor lung function in patients with cystic fibrosis: data from the European Epidemiologic Registry of Cystic Fibrosis. Pediatr Pulmonol. 2001;32: 343–350. DOI: 10.1002/ppul.1142.
99. Causer AJ, Shute JK, Cummings MH, Shepherd AI, Wallbanks SR, Allenby MI, et al. The implications of dysglycaemia on aerobic exercise and ventilatory function in cystic fibrosis. J Cyst Fibros. 2019. DOI: 10.1016/j.jcf.2019.09.014.
100. Moran A, Dunitz J, Nathan B, Saeed A, Holme B, Thomas W. Cystic fibrosis-related diabetes: current trends in prevalence, incidence, and mortality. Diabetes Care. 2009;32:1626–1631. DOI: 10.2337/ dc09-0586.
101. Ntimbane T, Mailhot G, Spahis S, Rabasa-Lhoret R, Kleme M-LL, Melloul D, et al. CFTR silencing in pancreatic beta-cells reveals a functional impact on glucose-stimulated insulin secretion and oxidative stress response. Am J Physiol Endocrinol Metab. 2016;310–E212. DOI: 10.1152/ajpendo. 00333. 2015.
102. van t Erve TJ. Strategies to decrease oxidative stress biomarker levels in human medical conditions: a meta-analysis on 8-iso-Prostaglandin F2α. Redox Biol. 2018. DOI: 10.1016/j. redox. 2018. 05.003.
103. Barreiro E. Role of protein carbonylation in skeletal muscle mass loss associated with chronic conditions. Proteomes. 2016;4:18. DOI: 10.3390/ proteomes4020018.
104. Rodriguez DA, Kalko S, Puig-Vilanova O, Perez-Olabarría M, Falciani F, Gea J, et al. Muscle and blood redox status after exercise training in severe COPD patients. Free Radic Biol Med. 2012;52: 88–94. DOI: 10.1016/j.freeradbiomed. 2011.09.022.
105. Gruet M, Troosters T, Verges S. Peripheral muscle abnormalities in cystic fibrosis: etiology, clinical implications and response to therapeutic interventions. J Cyst Fibros. 2017;16:538–552. DOI: 10.1016/J.JCF.2017.02.007.
106. Hulzebos HJE, Jeneson JAL, van der Ent CK, Takken T. Cross Talk opposing view: skeletal muscle oxidative capacity is not altered in cystic fibrosis patients. J Physiol. 2017;595:1427–1428. DOI: 10.1113/JP272505.