# Inhalation Treatment with Glutathione in Patients with Cystic Fibrosis

## A Randomized Clinical Trial

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***Rationale*: Glutathione is the major antioxidant in the extracellular lining fluid of the lungs and depleted in patients with cystic fibrosis (CF).**

***Objectives*: We aimed to assess glutathione delivered by inhalation as a potential treatment for CF lung disease.**

***Methods*: This randomized, double-blind, placebo-controlled trial evaluated inhaled glutathione in subjects with CF 8 years of age and older and $FEV_1$ of 40–90% of predicted. Subjects were randomized to receive 646 mg glutathione in 4 ml ($n = 73$) or placebo ($n = 80$) via an investigational eFlow nebulizer every 12 hours for 6 months.**

***Measurements and Main Results*: $FEV_1$ (absolute values), both as pre–post differences ($P = 0.180$) and as area under the curves ($P = 0.205$), were the primary efficacy endpoints, and were not different between the glutathione group and the placebo group over the 6-month treatment period. Exploratory analysis showed an increase of $FEV_1$ from baseline over placebo of 100 ml or 2.2% predicted; this was significant at 3 months, but not later. Subjects receiving glutathione had neither fewer pulmonary exacerbations, nor better scores for quality of life. Whereas increased glutathione and metabolites in sputum demonstrated significant delivery to the lungs, there was no indication of diminished oxidative stress to proteins or lipids, and no evidence for anti-inflammatory or antiproteolytic actions of glutathione supplemented to the airways. The adverse event incidence was similar between glutathione and placebo.**

***Conclusions*: Inhaled glutathione in the dose administered did not demonstrate clinically relevant improvements in lung function, pulmonary exacerbation frequency, or patient-reported outcomes. Glutathione delivery to the airways was not associated with changes in markers of oxidation, proteolysis, or inflammation.**

**Clinical trial registered with www.clinicaltrials.gov (NCT00506688) and https://eudract.ema.europa.eu/index.html (EudraCT 2005-003870-88).**

### AT A GLANCE COMMENTARY

**Scientific Knowledge on the Subject**

Glutathione is a major antioxidant in the extracellular lining fluid of the lungs and depleted in cystic fibrosis (CF).

**What This Study Adds to the Field**

Glutathione inhalation over 6 months did not demonstrate clinically relevant improvements in lung function, pulmonary exacerbation risk, and patient-reported outcomes. In addition, this treatment did not alter oxidative, proteolytic, or inflammatory balance in CF sputum.

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† Deceased.

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*Author Contributions*: M.G. designed the study protocol, analyzed the data, and wrote the draft of the manuscript, and is the study guarantor, with full responsibility for the finished article, access to any data, and control of the decision to publish. M. Kappler helped to write the manuscript and collected clinical data. C.E., M.B., S.J., E.R., S.v.K.-R., D.S., C.R.-W., U. Mellies, T.K., T.W., S.K., H.T., H.-E.H., M. Kopp, S.H., J.H., D.H., and P.K. coordinated investigator centers, collected clinical data, and contributed to writing of the manuscript, M.H. centrally coordinated, educated, and monitored the centers, U. Mansmann helped with the study design, oversaw statistical analyses, and helped to write the manuscript, I.B.-S. performed all biochemical analyses related to prostanoids, analyzed the data, and wrote corresponding METHODS and RESULTS; due to her recent death, she could not read the whole manuscript. C.F. coordinated the adherence assessments and performed analysis of data, and A.H. collected clinical data, helped with the study design, directed and analyzed the *ex vivo* studies, and contributed to writing the final manuscript.

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In cystic fibrosis (CF), the most common lethal genetic disease in whites, progressive lung disease is the leading cause of death. Mutations in the CF transmembrane conductance regulator (CFTR) facilitate a chronic pulmonary infection and severe inflammation with large amounts of proinflammatory chemokines,

cytokines, and activated cells in the airways (1, 2). These processes also generate huge excesses of oxidants in the airways, rapidly overwhelming the antioxidant screens, and this oxidative stress may contribute to lung injury (3, 4). The major extracellular antioxidant, glutathione, normally present in very high concentrations in the epithelial lining fluid (5–7), is believed to represent a central element in CF antioxidant defense, and its deficiency to contribute to the progressive lung tissue damage (8). Glutathione, which is a naturally occurring tripeptide, has been linked to CF not only by the repetitively observed pronounced depletion of glutathione in the extracellular epithelial lining fluid of the lung (6, 7, 9), but also from the direct involvement of CFTR in its transport into the extracellular space (10). In accordance, a CFTR-defective cell line secreted significantly less glutathione into the apical fluid than cells after CFTR repletion (11). Similar observations were made in *Cftr* knockout mice (12). In severely affected patients with CF, glutathione levels in bronchoalveolar lavage fluid were as low as 10% of healthy control subjects (5–7). It is notable that, in sputum supernatants from patients with CF, the levels of glutathione were increased when compared with healthy control subjects and control subjects with asthma (13).

The pivotal short-term inhaled glutathione phase 1 study by Roum and colleagues (6) demonstrated not only the feasibility of replete alveolar glutathione levels, but also showed *ex vivo* and *in vitro* suppressed superoxide anion release by alveolar inflammatory cells after glutathione therapy. These results were reproduced in another phase 1 study that also showed improved lung function and dose-dependent increase in alveolar glutathione levels, but no antioxidant effects (7). Improved lung function after inhalation of glutathione was reported in several case reports and in a pilot study of inhaled glutathione (14, 15). Therefore, this investigator-initiated, randomized, multicenter trial was conducted to assess the hypothesis that inhaled glutathione will improve $FEV_1$ in adult and pediatric patients with CF.

## METHODS

This was a phase 2b, randomized, double-blind, placebo-controlled, national, multicenter study of glutathione administered by inhalation (ClinicalTrials.gov identifier: NCT00506688; EudraCT no.: 2005-003870-88). The protocol was reviewed and approved by the institutional ethics committee at each participating center, and all subjects or their parents provided written informed consent. Inclusion criteria were: patients with CF 8 years of age or older (CF defined by positive [≥60 mM $Cl^-$] sweat chloride test and/or two disease-causing mutations), and an $FEV_1$ of 40–90% of predicted for age, sex, and height. Patients on concomitant inhaled thiol-containing medications (e.g., inhaled *N*-acetylcysteine) were excluded. Oral *N*-acetylcysteine was allowed to be continued. Subjects were randomized at a 1:1 ratio by central telephone block randomization within each age group to receive study medication or placebo by inhalation from an investigational eFlow nebulizer system (PARI Pharma GmbH, Graefelfing, Germany) after and in addition to the routine morning and evening chest physiotherapy and routine inhalations ("add on"; i.e., twice daily for 6 mo). For each inhalation, a solution was prepared by dissolving the 646 mg glutathione-Na powder (TAD 600; Biomedica Foscama, Ferentino, Italy) from the provided vial in 4 ml of water for injection and, in the case of placebo, by the addition of 4 ml of 0.9% NaCl for injection to an empty vial that was appropriately covered (Haupt Pharma, Wolfratshausen, Germany). To ensure a reliable blinding of the study medication, both the test product and the placebo were provided in appropriately covered and identical glass containers to obscure the contents. In addition, identical-looking ampoules for reconstitution of verum and placebo were provided. Smell or tastes were not masked due to unresolved toxicology issues of trace agents in long-term usage added to inhalation solutions. The primary efficacy endpoints were the pre–post difference between end of trial and baseline value of $FEV_1$ absolute values, and the time-weighted area under the curve of $FEV_1$ absolute values over the course of the treatment period. Secondary endpoints included change from baseline in percent predicted $FEV_1$ through Week 24, time to first pulmonary exacerbation (16), and patient-reported outcomes, as assessed by the CF Questionnaire for quality of life (17). Changes in laboratory markers were assessed, including free and total glutathione in serum and sputum, inflammatory cells, cytokines, and sputum weight. The study also evaluated safety. The study design consisted of a 2-week run-in period for determining baseline $FEV_1$, defined as the mean of measurements at the beginning and end, and parallel treatment groups with assessments after 1, 3, and 6 months.

Biochemical measurements were made in serum and sputum, obtained as described previously (18), in subgroup of subjects from the centers in Munich, Hannover, Cologne, Berlin, Frankfurt, and Bochum, and as detailed in the online supplement.

A total sample size of at least 138 subjects was calculated as adequate to detect an absolute difference in $FEV_1$ of 45 ml (SD = 90 ml) and, as hierarchical coprimary, an absolute $FEV_1$ increase of 5% predicted (area under the curve from baseline to the end of the trial between the two groups) based on the results of similar studies recently published (19, 20) at a power of 80% (nQuery Advisor Release 6.0, Statistical Solutions Ltd, Cork, Ireland). All subjects who received at least one dose of study drug were included in the analyses.

For the analysis of the primary endpoints, the GLM procedure, a method of least squares to fit general linear models, was used for analysis of covariance. For exploratory analysis of all other clinical and laboratory endpoints, the absolute changes from baseline were analyzed by Mann-Whitney nonparametric tests. Results are given in tables as means and SD and in figures as means and SE. Additional methodological details are provided in the online supplement. Prism version 4.00 (GraphPad Software, San Diego, CA) was used for graphics. Analyses were performed using SPSS version 12.0 for Windows (SPSS Inc., Chicago, IL).

## RESULTS

### Subjects

The study was conducted between May 2007 and May 2010. Subject disposition is shown in Figure 1. The study population consisted of 153 subjects who were enrolled, randomized, and received at least one dose of inhaled glutathione ($n = 73$) or placebo ($n = 80$). The study population had a mean age of 23 years, mean $FEV_1$ % predicted of 65%, and 48% were female (Table 1). A total of 64% of the subjects carried at least one delta-F508 mutation, and 53% had at least one positive airway culture for *Pseudomonas aeruginosa* in the previous year. The chronic medications used before and during the study were similar for both groups; of note, glutathione-treated subjects received somewhat more anti-inflammatory treatments (*see* Table E1 in the online supplement).

### Primary Clinical Efficacy Endpoints

Over the 6-month treatment period, the primary efficacy endpoints—changes of absolute $FEV_1$—neither the pre–post differences nor the area under the curves were different between the glutathione group and the placebo group (Table 2). They were measured for all 153 patients enrolled and analyzed in this sample (intention to treat).

### Secondary Clinical Efficacy Endpoints

All secondary analyses were exploratory, and the exact *P* values below 0.1 are given just as orientation for the magnitude of differences at certain time points.

***Lung function.*** Absolute change of $FEV_1$ in glutathione-treated subjects was, on average, slightly higher than in the placebo group, and did reach statistical significance at 3 months (Figure 2A), but not when expressed as % predicted (Figure 2C). For the absolute change of FVC and of forced expiratory flow, midexpiratory phase ($FEF_{25–75}$), expressed as % predicted, significant differences were also found at 3 months (Figure E1).

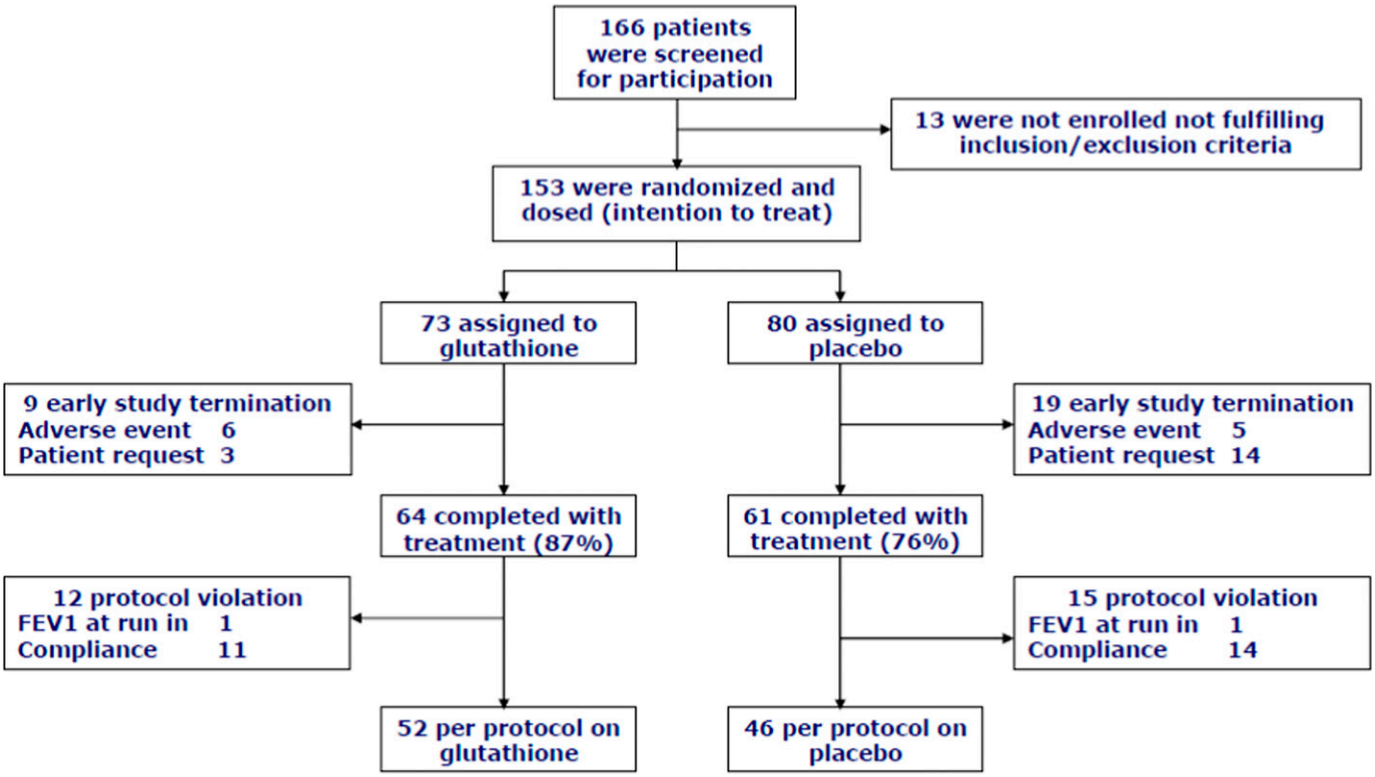


***Figure 1.*** Subject disposition.

Generally, the observed changes were higher in children than in adults (Figure E1).

***Pulmonary exacerbations.*** The time to first pulmonary exacerbation and also the number of pulmonary exacerbations did not differ significantly between the two groups (Table E2). A Kaplan-Meyer plot revealed no difference during the observation period (Figure 2B), although nonsignificant changes had a consistent direction in this and the other parameters assessed. Overall, the rate of exacerbations was (nonsignificantly) reduced by 18% (i.e., from 32 exacerbations in the placebo-treated subjects to 26 exacerbations in the verum-treated subjects).

***Weight.*** Of interest, weight gain was significantly higher in patients treated with glutathione than with placebo during the first 3 months ($P < 0.05$); however, at 6 months, this effect was absent (Table E2).

***Quality of life.*** Subjects treated with inhaled glutathione did not report more improvement in respiratory symptoms compared with placebo using the CFQ-R Respiratory domain (Table E2). Similarly, for the total CFQ-R scores, there was no difference between the two treatment groups.

### Safety and Adverse Event Profile

The incidence of adverse and serious adverse events was similar between the two groups (Table 3). There were two serious adverse events judged as non–CF related; one was a facial palsy in the glutathione group, which resolved, and the other a chronic IgA nephritis in the placebo group, which did not resolve. The number of treatment-emerging adverse events occurring in 10% or more of subjects was expected from other CF studies (Table E3). The magnitude was similar between the two groups, with somewhat higher frequencies of pyrexia, abnormal sputum, and upper respiratory tract infection in the glutathione group. None of these was considered serious or led to discontinuation. Interestingly, the number of patients who requested early study termination was higher in the placebo group than in patients assigned to glutathione (Figure 1).

### Exploratory Cellular and Biochemical Marker

***Sputum.*** GLUTATHIONE AND ITS METABOLITES. At baseline, all variables assessed, except free glutamyl-cysteine, were not different between the two groups. At all time points after the start of treatment, the pre–post differences of free and total glutathione in sputum (Figure 2D) were significantly higher in patients treated with glutathione (Table E5). In accordance with this, intracellular neutrophil glutathione pre–post differences were higher in the glutathione group at the visits after 3 and 6 months.

Some of the metabolites linked to glutathione (i.e., glutamyl-cysteine and homocysteine after 1 or 3 months; Table E5) were lower in the glutathione treatment group than in the placebo group, whereas cysteinyl-glycine was much higher. Cysteine was not different between treatment groups (Table E5).

PROTEIN CARBONYLS. The change in the amount of proteins that were carbonylated as a sign of oxidative stress was not significantly different in the two groups (Table E5).

SPUTUM WEIGHT, TOTAL CELL COUNT, CELL VIABILITY, AND NEUTROPHIL ELASTASE. Sputum weight was assessed as a measure of sputum removal from the lungs. Compared with placebo, glutathione-induced changes did not differ (Table E5). In addition, the pre–post differences of total numbers of cells in sputum, cell number per gram of sputum, and cell differential counts did not vary between placebo and glutathione. Of interest, cell viability was higher in the presence of glutathione; this effect is compatible with a protection of viability by extracellular glutathione. In accordance with the unchanged absolute neutrophil counts (data not shown) and percentage of neutrophils in cell differentials, neutrophil elastase pre–post differences did not differ between the placebo and the glutathione group (Table E5).

**TABLE 1. BASELINE DATA OF THE STUDY COHORT (INTENTION TO TREAT)**

| | Glutathione (*n* = 73) | | Placebo (*n* = 80) | |
|---|---|---|---|---|
| | Mean (SD) | n (%) | Mean (SD) | n (%) |
| Age, yr | 23.1 (9.8) | — | 23.0 (10.4) | — |
| Height, cm | 166.2 (13.3) | — | 163.9 (16.0) | — |
| Weight, kg | 56.6 (14.4) | — | 54.3 (16.8) | — |
| BMI, kg/m$^2$ | 20.2 (3.5) | — | 19.6 (3.6) | — |
| $FEV_1$, L | 2.2 (0.7) | — | 2.1 (0.7) | — |
| $FEV_1$ % predicted | 65.6 (14.1) | — | 65.2 (14.5) | — |
| FVC % predicted | 78.9 (12.0) | — | 81.6 (14.4) | — |
| $FEF_{25–75}$ % predicted | 39.3 (22.5) | — | 36.0 (20.4) | — |
| Quality of life, total score | 75.0 (10.0) | — | 75.0 (11.8) | — |
| Quality of life, respiratory | 69.5 (14.2) | — | 66.1 (18.0) | — |
| Sex | | | | |
| Male | — | 42 (57.5) | — | 37 (46.3) |
| Female | — | 31 (42.5) | — | 43 (53.8) |
| Ethnic origin | | | | |
| White | — | 72 (98.6) | — | 80 (100.0) |
| Other | — | 1 (1.4) | — | 0 (0.0) |
| Delta-F508 homozygous | — | 31 (42.4) | — | 41 (51.3) |
| Delta-F508 heterozygous | — | 12 (16.4) | — | 14 (17.5) |
| Others | — | 27 (37.0) | — | 21 (26.3) |
| Unknown | — | 3 (4.1) | — | 4 (5.0) |
| *Pseudomonas aeruginosa* | — | 41 (56.2) | — | 40 (50.0) |
| *Staphylococcus aureus* | — | 23 (31.5) | — | 33 (41.3) |
| *Haemophilus influenza* | — | 4 (5.5) | — | 2 (2.5) |
| *Stenotrophomonas maltophilia* | — | 5 (6.8) | — | 7 (8.8) |
| *Mycobacteria* | — | 1 (1.4) | — | 4 (5.0) |
| *Burkholderia cepacia* | — | 0 (0.0) | — | 1 (1.3) |
| *Candida* | — | 25 (34.2) | — | 36 (45.0) |
| *Aspergillus* | — | 15 (20.5) | — | 23 (28.8) |

*Definition of abbreviation:* $FEF_{25–75\%}$ = forced expiratory flow, midexpiratory phase

No significant differences between the two groups were present at baseline. $P > 0.50$ in all, except sex (0.20), ethnic origin (0.48), delta-F508 homozygous (0.33), *Haemophilus influenza* (0.43), *Mycobacteria* (0.21), *Candida* (0.19), and *Aspergillus* (0.09) (Fisher's exact test) ($n = 153$).

LIPID MEDIATORS. Several lipid mediators were assessed, because we had previously observed changes in alveolar lipid mediator concentration in a study assessing inhaled glutathione by bronchoalveolar lavage. However, the observed pre–post differences were not different between the two groups (Table E5).

INFLAMMATORY AND NEUTROPHIL ACTIVATION MARKERS IN SPUTUM. Lastly, as a measure of inflammatory activity, several chemokines and cytokines and other cellular markers of neutrophil activation were assessed, but we did not observe pre–post differences between placebo and glutathione (Table E5).

***Blood.*** Thiols (including free glutathione and glutathione in blood neutrophils), cytokine receptor expression, and activation markers on neutrophils were not different at baseline (Table E6). In addition, the differences in the levels before and after treatment between the placebo and glutathione study groups (Table E7) did not differ.

## DISCUSSION

In this randomized, placebo-controlled trial in subjects with CF, administration of inhaled glutathione at pharmacological doses did not achieve significant or clinically relevant improvements in primary endpoints (i.e., lung function assessed by $FEV_1$ absolute changes before/after and during the trial). Despite this negative outcome, the results of this study give a comprehensive view of the effect of inhaled glutathione as an "add on" therapy in intensely treated patients with CF. Conclusions can be drawn on the tolerability, side effects, magnitude, and direction of changes in secondary clinical outcomes induced by glutathione, and the previously anticipated role of glutathione for the oxidative and inflammatory balance in the airways.

Overall, the changes in lung function were small, and failed to reach the preset primary endpoint. The absolute change (mean ± SD) of $FEV_1$ from baseline over placebo was 100 ± 140 ml, or 2.2 ± 0.1%, expressed as % predicted. This observation was consistent with changes in other lung function variables (i.e., a significant increase of FVC and $FEF_{25–75}$ % predicted at 3 mo). Such trends were clearly observed in both adults and children. In our previous phase 1 study with a smaller number of subjects, lung function improved by about 5% (7). $FEV_1$ has been established as the pivotal clinical study endpoint for the assessment of novel therapies in CF. When comparing our trial to those others, one has to consider that baseline treatments of patients in previous studies over the past 20 years were

**TABLE 2. RESULTS ON PRIMARY OUTCOME VARIABLE $FEV_1$ IN THE INTENTION TO TREAT COHORT**

| | Glutathione (*n* = 73) | Placebo (*n* = 80) | *P* Value |
|---|---|---|---|
| Pre–post difference of $FEV_1$ absolute values from baseline to EOT, L | 0.10 ± 0.14 | −0.13 ± 0.01 | 0.180 |
| Time-weighted AUC of $FEV_1$ absolute values from baseline to EOT, L | 2.15 ± 0.08 | 2.02 ± 0.08 | 0.205 |

*Definition of abbreviations:* AUC = area under the curve; EOT = end of trial.

Data are results of the analysis of covariance test with treatment, age group, and center used as predictive factors; baseline $FEV_1$ % predicted and time of spirometry as covariates. Given are the least-square means ± SE. The evaluation of the per protocol data set confirmed these results.

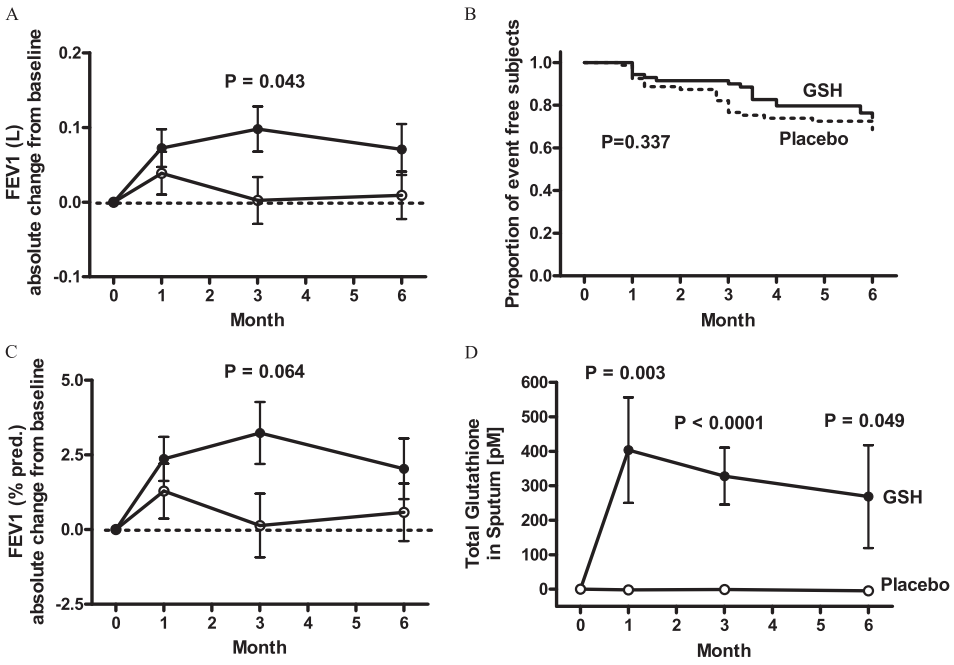


***Figure 2.*** Changes from baseline through Months 1, 3, and 6 in $FEV_1$ absolute value in liters with SE (*A*), time to first pulmonary exacerbation by treatment group (*B*), $FEV_1$ % predicted with SE (*C*), and total sputum glutathione with SE (*D*). Exact *P* values of less than 0.10 of the exploratory analysis are indicated above the corresponding data points at the respective times. *Open symbols*, placebo; *closed symbols*, glutathione. GSH = glutathione.

typically less intense than ours with regard to inhaled antibiotics and other treatments (21). This allowed bigger effects of newly introduced therapies to occur in the past. For inhaled tobramycin, a 12% improvement in $FEV_1$ relative to baseline was observed after 20 weeks of treatment (22); dornase alfa showed a 5.8% improvement in $FEV_1$ in comparison to placebo after 24 weeks (16), hypertonic saline demonstrated a 3.2% improvement after 48 weeks (23), and inhaled mannitol a 3.7% increase (24) after 26 weeks. Before the start of the study, we had set a 5% predicted increase in lung function to be clinically relevant and calculated our sample size based on this assumption. With the results obtained in this study, a sample size of 276 subjects would have been necessary to show a 2.2% predicted change in $FEV_1$ to be statistically significant.

Adherence to therapy is another relevant issue for the interpretation of the study results. Based on vial counts, adherence to study medication was high (90 ± 23%). As generally acknowledged, this commonly used technique to monitor adherence may overestimate adherence (25). In a subset of 35 patients, we electronically monitored adherence to inhalation of the study drugs with a novel eFlow device with monitoring function; mean adherence was 76%, whereas, calculated from vials, it was 88% (26) (see supplemental METHODS and Figure E2). Although this was a double blind study with respect to packaging of the vials and visual appearance of the medication, those subjects treated with verum could recognize glutathione by its smell, which cannot be masked. This may have reassured these subjects of having received active medication, and thus explains the significantly higher dropout rate due to early termination by patient request in the placebo group (3 in the glutathione group, 14 in the placebo group). On the other hand, waning treatment adherence over time may be considered to explain the nonsustained levels of glutathione recovered in sputum at later time points. This may have translated into nonsustained effects on lung function.

For a comprehensive judgment of a significant clinical benefit, reduced rates of pulmonary exacerbations and increased scores for quality of life are expected to consistently support the beneficial effect of a treatment. This was clearly not the case, as, for both groups of variables (i.e., quality of life in general and specific categories), as well as rate and time to exacerbations using several definitions, no significant differences between glutathione and placebo treatment were demonstrated. The reduction of the rate of exacerbations over placebo was 18% in this trial, 22% in the large rhDNase trial involving 968 patients (16), 26% in the mannitol trial (not significant) (24), and 66% in the hypertonic saline trial (23).

Due to the intense and early, amplified inflammatory response in CF lungs (1), a large excess of oxidants characterizes these airways (3). Lack of the major extracellular antioxidant, glutathione, usually present in millimolar concentrations in the alveolar space (7, 9, 27), is believed to represent a central event in CF lung pathogenesis, and contributes to the progressive lung tissue damage. Measurements of glutathione and metabolites in sputum during steady state before the next inhalation demonstrated significant delivery to the lungs. This was in good agreement with our previous proof of appropriate delivery, as assessed by bronchoalveolar lavage and increased levels in epithelial lining fluid (7). Inhaled glutathione led to an increase in cysteinyl-glycine, the product generated by cell surface–located γ-glutamyl transpeptidase (28) and to a reduction of its precursors, homocysteine

**TABLE 3. SUMMARY OF ADVERSE EVENTS BY TREATMENT GROUP**

| | Glutathione (*n* = 73) [*n* (%)] | Placebo (*n* = 80) [*n* (%)] |
|---|---|---|
| Subjects with any adverse events | 73 (100) | 77 (96) |
| Subjects with serious adverse events | 8 (11) | 8 (10) |
| Cystic fibrosis lung (pulmonary exacerbation) | 4 (5) | 5 (6) |
| Hemoptysis | 2 (2) | — |
| Abdominal pain, distal intestinal obstruction syndrome | 1 (2) | 2 (2) |
| Facial palsy | 1 (1) | — |
| Nephritis | — | 1 (1) |

and glutamyl-cysteine. However, these significant changes were not associated with diminished oxidative stress to proteins (assessed by their carbonyl content) or to lipids (assessed by 8-isoprostan levels). This is in close agreement with our previous investigation (7), and extends the findings to long-term treatment with inhaled glutathione. In addition, a wide range of cellular and soluble markers was not altered by glutathione treatment compared with placebo, clearly indicating no prominent anti-inflammatory effect, which up to now was ascribed to inhaled glutathione therapy (8, 29). These results on surrogate markers must be interpreted with caution, as the analyses were done in the subset of subjects investigated in centers with appropriate sputum processing facilities. Nevertheless, we did not find any evidence in sputum for significant antioxidative or anti-inflammatory actions of glutathione supplemented to the airways in patients with CF.

Daily inhaled administration of glutathione for 6 months was not associated with an increased safety risk, led to small, but not clinically relevant increases in lung function, did not reduce the rate and time to pulmonary exacerbation, and did not improve quality of live. Despite large increases of extracellular and intracellular glutathione in sputum, surrogate markers of oxidative and inflammatory processes were not altered. The results challenge the concept that the introduction of large doses of the single metabolite, glutathione, produced naturally in the body and having many functions, including antioxidative actions, may be helpful in mitigating oxidative or inflammatory dysbalance in CF. It must be kept in mind that we did not formally show in this study that alveolar glutathione concentrations were elevated to or above normal values. These data support the view that exogenous treatment with glutathione at the dose administered is unlikely to be of clinically relevant benefit in CF.

Glutathione is a major antioxidant in the extracellular lining fluid of the lungs, and is depleted in CF; however, its inhalation over 6 months did not demonstrate clinically relevant improvements in lung function, pulmonary exacerbation risk, or patient-reported outcomes. Furthermore, this treatment did not alter oxidative, proteolytic, or inflammatory balance in CF airways.

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