Nebulized and oral thiol derivatives for pulmonary disease in cystic fibrosis (Review)
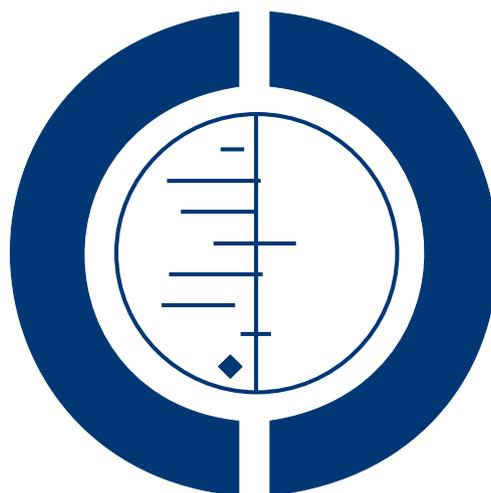
Nash EF, Stephenson A, Ratjen F, Tullis E

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[Intervention Review]

Nebulized and oral thiol derivatives for pulmonary disease in cystic fibrosis

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ABSTRACT

Background

Cystic fibrosis is an inherited condition resulting in thickened, sticky respiratory secretions. Respiratory failure, due to recurrent pulmonary infection and inflammation, is the most common cause of mortality. Muco-active therapies (e.g. dornase alfa and nebulized hypertonic saline) may decrease sputum viscosity, increase airway clearance of sputum, reduce infection and inflammation and improve lung function. Thiol derivatives, either oral or nebulized, have shown benefit in other respiratory diseases. Their mode of action is likely to differ according to the route of administration. There are several thiol derivatives, and it is unclear which of these may be beneficial in cystic fibrosis.

Objectives

To evaluate the efficacy and safety of nebulized and oral thiol derivatives in people with cystic fibrosis.

Search methods

We searched the Cochrane Cystic Fibrosis and Genetic Disorders Group Trials Register, comprising references identified from comprehensive electronic database searches, hand searches of relevant journals, abstract books and conference proceedings.

Most recent search: 08 October 2010.

Selection criteria

Randomized and quasi-randomized controlled trials comparing nebulized or oral thiol derivatives to placebo or another thiol derivative in people with cystic fibrosis.

Data collection and analysis

The authors independently assessed trials for inclusion, analysed methodological quality and extracted data.

Main results

Searches identified 18 trials; eight (seven older than 10 years) (234 participants) are included. Three trials of nebulized thiol derivatives were identified (one compared 20% n-acetylcysteine to 2% n-acetylcysteine; another compared sodium-2-mercaptoethane sulphonate to 7% hypertonic saline; and another compared glutathione to 4% hypertonic saline). Although generally well-tolerated with no significant adverse effects, there was no evidence of significant clinical benefit in our primary outcomes in participants receiving these treatments.

Five studies of oral thiol derivatives were identified. Three studies compared n-acetylcysteine to placebo; one compared n-acetylcysteine, ambroxol and placebo; and one compared carbocysteine to ambroxol. Oral thiol derivatives were generally well-tolerated with no significant adverse effects, however there was no evidence of significant clinical benefit in our primary outcomes in participants receiving these treatments.

Authors' conclusions

We found no evidence to recommend the use of either nebulized or oral thiol derivatives in people with cystic fibrosis. There are very few good quality trials investigating the effect of these medications in cystic fibrosis, and further research is required to investigate the potential role of these medications in improving the outcomes of people with cystic fibrosis.

PLAIN LANGUAGE SUMMARY

Compounds which can break down the structure of mucus for lung disease in cystic fibrosis

Cystic fibrosis is a genetic disorder which mainly affects the lungs. Chest infections recur in people with cystic fibrosis due to a build up of thick sputum (phlegm) in the air passages. Several treatments, including thiol derivatives, aim to loosen this sputum and so improve lung function and reduce the frequency of chest infections. Thiol derivatives may be either nebulized (breathed in) or oral (by mouth). They have been shown to help in other lung conditions, such as chronic obstructive pulmonary disease. This review aims to find out if there is enough evidence to recommend the nebulized or oral thiol derivatives for people with cystic fibrosis. We included eight trials; three assessed the effect of nebulized thiol derivatives. Of the nebulized studies, one compared 20% n-acetylcysteine to 2% n-acetylcysteine; another compared sodium-2-mercaptoethane sulphonate to 7% hypertonic saline; and the other compared glutathione to 4% hypertonic saline. Nebulized thiol derivatives were generally well-tolerated with no major adverse effects. However they showed no significant improvements in any of our outcome measures.

Five included studies assessed the effects of oral thiol derivatives. Three of these studies compared oral n-acetylcysteine to placebo; one compared oral n-acetylcysteine, oral ambroxol and placebo; and one compared oral carbocysteine and oral ambroxol (no placebo). None of the studies showed an overall significant benefit in any of the outcome measures of this review. Oral thiol derivatives were generally well-tolerated with no major adverse effects.

In summary, the studies included in the review did not provide any evidence that nebulized or oral thiol derivatives were either beneficial or harmful to people with cystic fibrosis. Further research investigating the effects of thiol derivatives in people with cystic fibrosis is required before their use can be recommended.

BACKGROUND

Description of the condition

Cystic fibrosis (CF) is a genetic multisystem disorder characterized by thick, tenacious secretions resulting in organ damage, primarily involving the lungs and gastrointestinal system. Over the past 30 years, survival in CF has increased dramatically due to the development of new therapies and aggressive nutritional supplementation (CFF 2007; Corey 1996). Despite these improvements, the most common cause of death is end-stage pulmonary disease and respiratory failure (Davis 2006). Lung damage occurs secondary

to the excessive inflammatory response to bacteria that reside in the airways. The influx of neutrophils and stimulation of mucus secretion within the airways results in thick secretions which are difficult to clear. Furthermore, tenacious secretions adhere to the airways worsening mucociliary clearance and ciliary function. This creates an environment of increased bacterial burden, further inflammation, and thicker secretions.

Description of the intervention

Mucolytic therapy has been proposed as a method to decrease sputum viscosity, increase airway clearance of sputum, reduce bacterial load and inflammation, improve lung function, and ultimately increase survival (Henke 2007). It is typically initiated as part of the ongoing treatment regimen and the effects of such therapies are typically seen after regular use for a minimum of four weeks of treatment. Several mucolytic therapies are available for use in CF including dornase alfa (also known as rhDNase (Pulmozyme®)) (Fuchs 1994), hypertonic saline (Elkins 2006) and thiol derivatives such as n-acetylcysteine (NAC) (Duijvestijn 1999). Macrolides have been shown to affect mucus hypersecretion (Shimizu 2003) but are considered mucoregulatory rather than mucolytic agents. The Cochrane systematic reviews of both dornase alfa (Jones 2003) and hypertonic saline (Wark 2005) have demonstrated an improvement in lung function and a reduction in the need for antibiotic therapy for pulmonary exacerbations of CF. It is unclear whether thiol derivatives result in the same beneficial effects.

Thiol derivatives are compounds which contain a functional group of a sulfur atom and a hydrogen atom. This sulfhydryl group enables thiol derivatives to break down the gel structure of mucus, by substituting sulfhydryl groups for the disulfide bonds connecting mucin proteins (Dasgupta 1996). NAC is the classic thiol derivative, but several other compounds are included under this heading, including glutathione (GSH), cysteine, nacystelyn (NAL), sodium-2-mercaptoethane sulphonate, carbocysteine, erdosteine and mecysteine. In vitro, NAC has been demonstrated to reduce the viscosity and elasticity of mucus when directly in contact with airway secretions (Sheffner 1964). This may make sputum easier to clear; however, thinner secretions could potentially be harder to expectorate due to this reduced viscosity. NAC is a very acidic compound (pH 2.2), and when inhaled results in airway irritation, induction of cough and bronchospasm. Manufacturers therefore suggest that individuals receive pre-treatment with a bronchodilator prior to inhalation. It has been suggested that induction of cough by inhaled NAC, rather than mucolysis, may explain any beneficial effect of NAC on expectoration. This mechanism may reduce airway inflammation by improving clearance of the pro-inflammatory neutrophil breakdown products of which CF sputum largely consists. NAL, a lysine salt of NAC, has an approximately neutral pH and is well-tolerated when inhaled (App 2002). When studied in vitro, NAL has been demonstrated to have a more potent mucolytic activity than NAC, and, also in vitro, has an ad-

ditional inhibitory effect on human neutrophil elastase (Marriott 1993).

How the intervention might work

Nebulized thiol derivatives

NAC is a very acidic compound (pH 2.2), and when inhaled results in airway irritation, induction of cough and bronchospasm. Manufacturers therefore suggest that individuals receive pre-treatment with a bronchodilator prior to inhalation. It has been suggested that induction of cough by inhaled NAC, rather than mucolysis, may explain any beneficial effect of NAC on expectoration. This mechanism may reduce airway inflammation by improving clearance of the pro-inflammatory neutrophil breakdown products of which CF sputum largely consists. NAL, a lysine salt of NAC, has an approximately neutral pH and is well-tolerated when inhaled (App 2002). When studied in vitro, NAL has been demonstrated to have a more potent mucolytic activity than NAC, and, also in vitro, has an additional inhibitory effect on human neutrophil elastase (Marriott 1993).

Oral thiol derivatives

Orally-administered thiol derivatives are proposed to work by a different mechanism to inhaled thiol derivatives. Inhalation of thiol derivatives aims to deliver the compound to the lower airway in an attempt to act directly on airway secretions as a mucolytic. However, when administered orally there is no detectable NAC in bronchoalveolar lavage (Cotgreave 1987). Therefore orally-administered thiol derivatives are unlikely to have a direct effect as a mucolytic. Oral NAC is broken down (deacetylated) to cysteine, whose thiol group has reducing and antioxidant properties (Bonanomi 1980). Neutrophils in the CF airway cause damage by releasing oxidants, and are deficient in GSH, an important endogenous cellular antioxidant. High-dose oral NAC increases neutrophil GSH levels, decreases airway neutrophil recruitment and reduces neutrophilic release of airway elastase (Tirouvanziam 2006a). The sulfhydryl group also allows NAC to interact directly with oxidants, functioning as an oxidant scavenger (Ventresca 1989). Therefore, orally-administered thiol derivatives most likely act by reducing pulmonary oxidative stress and inflammation, and subsequently attenuating airway and parenchymal destruction. NAC also prevents inactivation of alpha-1-antitrypsin by neutrophil elastase in vitro, and could also prevent lung damage in CF by this mechanism (Borregaard 1987). In addition, oral thiol derivatives may have a mechanism of action in reducing airway inflammation by regulating redox signalling (Rahman 2006).

It must be stressed that the characteristics of CF sputum are different to that of sputum seen in other pulmonary diseases, such as chronic obstructive pulmonary disease (COPD). CF sputum

contains low levels of mucin, and consists predominantly of pus derived from neutrophil degradation (Henke 2004). This observation may impact on the ability of inhaled thiol derivatives to reduce sputum viscosity in CF, since mucin is their primary target. In summary, inhaled thiol derivatives are proposed to act primarily by their mucolytic effects, whereas when orally administered these compounds are more likely to act predominantly as antioxidants, with also a possible anti-inflammatory effect. These potential differing mechanisms of action are supported by in vitro evidence. Different pharmacokinetics, mechanisms of action and adverse effects can be expected depending on the thiol derivative utilized, the dose used, and the mode of administration.

Why it is important to do this review

The aim of this review is to collate and analyse the results of randomized controlled trials (RCTs) examining the clinical (in vivo) effects of both inhaled and oral thiol derivatives in the therapy of lung disease in people with CF. We thereby aim to provide clear evidence-based guidance regarding their effectiveness and safety in this group of individuals.

O B J E C T I V E S

The aims of this review are to:

1. evaluate efficacy of both inhaled and oral thiol derivatives in people with CF.

2. evaluate safety of both inhaled and oral thiol derivatives in people with CF.

M E T H O D S

Criteria for considering studies for this review

Types of studies

All published and unpublished randomized and quasi-randomized controlled trials. There was no language restriction for this review.

Types of participants

All individuals diagnosed with CF based on standard diagnostic criteria (sweat testing or genetics and clinical features or family history) were included. There were no restrictions on pulmonary disease severity, gender, or pancreatic status.

Types of interventions

Studies investigating thiol derivatives given at any dose or frequency, via nebulized or oral administration, for a minimum of four weeks duration were eligible for inclusion. We felt four weeks to be the minimum amount of time needed to see an effect from the treatment. Outcomes for participants who received thiol derivatives were compared to participants receiving no treatment or to control groups including placebo or any other medication. Nebulized and oral interventions were considered separately in this review. Thiol derivatives considered for inclusion were acetylcysteine (or N-acetylcysteine), sodium-2-mercaptoethane sulphonate, carbocysteine, erdosteine, nacystelyn, glutathione, cysteine and mecysteine.

Types of outcome measures

The following outcomes were recorded separately for both nebulized and oral thiol derivatives.

Primary outcomes

1. Pulmonary function testing (PFT)
    i) forced expiratory volume in one second (FEV_1) per cent predicted (change from baseline and absolute data)
    ii) forced vital capacity (FVC) per cent predicted (change from baseline and absolute data)

Secondary outcomes

1. Other PFT measurements which reflect airflow obstruction or gas trapping or both, e.g. peak expiratory flow (PEF), vital capacity (VC), FEV_1/VC, mid-expiratory flow 25-75 (MEF_{25-75}), residual volume/total lung capacity (RV/TLC) (change from baseline and absolute data)
2. Inflammatory markers (change from baseline)
    i) serum (white blood cell (WBC), C-reactive protein (CRP), erythrocyte sedimentation rate (ESR))
    ii) sputum (IL-8)
3. Quality of life (QOL) (change from baseline as measured by a validated QOL scale)
4. Need for oral antibiotics for pulmonary exacerbation
    i) number of weeks of treatment
    ii) number of courses
5. Need for intravenous antibiotics for pulmonary exacerbation
    i) number of weeks of treatment
    ii) number of courses
6. Adverse events
7. Number of days in hospital for respiratory exacerbation
8. Adherence
9. Acquisition of new respiratory pathogens (%)
10. Six-minute walk distance

11. Sputum characteristics (including measures of viscosity and elasticity)

Search methods for identification of studies

Electronic searches

We identified relevant trials using the Cystic Fibrosis Trials Register using the terms: n-acetylcysteine OR ((acetylcysteine OR carbocysteine OR erdosteine OR mecysteine OR nacystelyn OR glutathione OR cysteine OR sodium-2-mercaptoethane sulphonate) AND (oral OR nebulised OR unstated)).

The Cystic Fibrosis Trials Register is compiled from electronic searches of the Cochrane Central Register of Controlled Trials (Clinical Trials) (updated each new issue of *The Cochrane Library*), quarterly searches of MEDLINE, a search of EMBASE to 1995 and the prospective handsearching of two journals - *Pediatric Pulmonology* and the *Journal of Cystic Fibrosis*. Unpublished work was identified by searching the abstract books of three major cystic fibrosis conferences: the International Cystic Fibrosis Conference; the European Cystic Fibrosis Conference and the North American Cystic Fibrosis Conference. For full details of all searching activities for the register, please see the relevant sections of the Cystic Fibrosis and Genetic Disorders Group Module.

Date of last search: 08 October 2010.

Data collection and analysis

We used RevMan software to conduct the statistical analysis (RevMan 2008).

Selection of studies

Two authors (EN, AS) assessed the articles identified against the inclusion criteria for this review in an independent fashion. Where possible, authors contacted primary investigators if any details were unclear. They resolved any disagreements by discussion and advice from the other co-authors.

Data extraction and management

Two authors (EN, AS) extracted data from the articles in an independent fashion. Where possible, authors contacted primary investigators if their methodology was unclear. They resolved any disagreements by discussion and advice from the other co-authors. Where possible we grouped outcome data into those measured at one month, up to three months, up to six months, up to twelve months and then annually thereafter. If outcome data were recorded at other time periods we examined these as well.

One of the included trials is a three-arm trial, and we are including it in two comparisons (oral thiols versus placebo and oral thiols

versus other oral agents) (Ratjen 1985). By so doing, we realize that some participants will be counted twice in this review.

Assessment of risk of bias in included studies

The authors assessed the articles for methodological quality to establish the risk of bias in each study using a component approach and recorded details such as method of randomization, concealment of allocation, blinding and whether or not the data were assessed using an intention-to-treat analysis (Jüni 2001). Where possible, authors contacted primary investigators if their methodology was unclear.

Measures of treatment effect

When assessing differences between groups, we recorded the mean difference from baseline for each group as our treatment effect measure for continuous variables. Where applicable, we calculated a pooled estimate of treatment effect by calculating the mean difference (MD) and the 95% confidence intervals (CIs).

With binary outcomes, we planned to use the risk ratio (RR) and the 95% CIs as a measure of treatment effect; however, we were only able to present results using continuous data.

Unit of analysis issues

We included both parallel group trials as well as trials with a cross-over design. Ideally we would have liked to analyse cross-over trials using techniques outlined by Elbourne (Elbourne 2002); however, due to limitations on the data available we were only able to treat these trials as parallel trials and present data for the end of treatment. We realise that this approach is conservative as it ignores within-patient correlation and so does not make use of the advantages of the cross-over design. Also, this approach ignores the fact that patients appear in both arms of the trial and are not independent of each other (Elbourne 2002). We did not combine the data from cross-over trials with parallel trials as we were unable to employ the methods recommended by Curtin (Curtin 2002).

For longitudinal data, we reported the time-points that measurements were taken by the primary investigators and which measures were reported in the papers. In the review we present data from end of treatment. We preferred to report data for the change from baseline at end of treatment, but if the original paper reported only absolute data (means, standard deviations (SD) of groups or raw data), we planned to calculate the mean difference and the variance of the difference imputed using the Follmann method (Follmann 1992). This method allows the use of separate sources of incomplete information to help choose a better variance estimate. This method can be summarised by the formula: Var(change) = Var(pre-test) + Var(post-test) - 2 x SD(pre-test) x SD(post-test) x pre-test-post-test correlation coefficient. We also planned to report these absolute values post-treatment. Where the

correlation coefficient was unknown we estimated it to be 0.4 (a moderate correlation) in order to perform the calculation.

Dealing with missing data

When possible, the authors (EN, AS) contacted primary investigators if missing data were required.

Assessment of heterogeneity

We planned to assess the heterogeneity of the studies using the Q test, with heterogeneity being considered to be present if the Q test was statistically significant at the P < 0.10 level. When assessing the magnitude of any heterogeneity present, we planned to use the I^2 statistic (Higgins 2003).

Assessment of reporting biases

We intended on assessing publication bias using a funnel plot; however, there were insufficient studies (minimum of 10 required) to conduct this analysis.

With regards to selective reporting of outcomes, we were unable to compare the original trial protocols with the final published papers; however, there did not appear to be any obvious omissions to the outcomes reported on by the trial investigators.

Data synthesis

Where heterogeneity was not present, we used a fixed-effect model; however, if in future we establish moderate or high degrees of heterogeneity, we will utilize a random-effects model.

Subgroup analysis and investigation of heterogeneity

Several thiol derivatives are available, and therefore we planned to perform subgroup analysis for each compound. The dose of the thiol derivative used may alter outcomes, and therefore we planned to perform subgroup analysis according to dose. There were insufficient studies to perform either of these subgroup analyses on this occasion, but we plan to perform these analyses in a future update of the review when we are able to combine a sufficient number of trials to allow this. Clinically important outcomes vary depending on gender, age and severity of lung disease (FEV_1 70% to 80% will be considered mild; 60% to 70% moderate; 50% to 60% moderately severe; 34% to 50% severe; and less than 34% very severe (ATS 1991)), and therefore we also planned to perform an analysis using these subgroups. We planned to analyze age and lung function as continuous variables as well as categorical variables in an attempt to identify any high-risk groups. The degree of airway inflammation may also impact on the response to therapy with thiol derivatives, and therefore we also planned to perform subgroup analysis according to whether the study was performed during an acute pulmonary exacerbation or during a period of

disease stability. Again, there were insufficient studies to perform these subgroup analyses in this review, but we plan to perform these analyses in a future update where possible. Lung function will be categorized according to ATS guidelines for disease severity as described above (ATS 1991), and age will be categorized in 10-year blocks where possible.

Sensitivity analysis

We intended to conduct a sensitivity analyses based on the methodological quality of the studies in the review; however, there were insufficient studies to proceed with this analysis.

R E S U L T S

Description of studies

See: Characteristics of included studies; Characteristics of excluded studies.

Results of the search

The search process identified 23 published research papers from a variety of international journals. These papers referred to 18 separate studies, of which we were able to include eight studies and we excluded the remaining 10 studies.

Included studies

Eight studies eligible for inclusion were identified; three studies examined the effects of nebulized thiol derivatives (Bishop 2005; Howatt 1966; Weller 1980) and five studies evaluated oral thiol derivatives in people with CF (Caramia 1995; Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989). Clinical details regarding the participants, interventions and outcomes are given in the table Characteristics of included studies.

Nebulized thiol derivatives

Three trials compared nebulized thiol derivatives to other nebulized medications (Bishop 2005; Howatt 1966; Weller 1980). There was considerable heterogeneity between studies and therefore data from only two of these studies could be pooled for analysis (Bishop 2005; Weller 1980); also, the number of outcomes for which data could be pooled was very limited.

Participants

All trials had small numbers of participants (between eight and 29 participants in each trial). Two trials included both adults and children (Bishop 2005; Howatt 1966) and one trial included children only (Weller 1980).

Trial Design

One trial was of parallel design (Bishop 2005) and two of the trials were of cross-over design (Howatt 1966; Weller 1980). The two cross-over trials both compared two treatment arms (Howatt 1966; Weller 1980). Only one of the cross-over trials contained a washout period between treatment blocks (Weller 1980). Duration of treatment blocks ranged from one month to two months; duration of the trials ranged from two to four months.

Interventions

One of the included studies assessed the effects of nebulized NAC (Howatt 1966), another assessed the effects of nebulized sodium-2-mercaptoethane sulphonate (Weller 1980) and the final study assessed the effects of nebulized glutathione (Bishop 2005). Since different thiol derivatives can be expected to have different clinical effects, it is not appropriate to combine the results of studies assessing different compounds.

None of the trials compared the effects of nebulized thiol derivatives against a true placebo. The Howatt study compared the effects of nebulized 20% NAC and nebulized 2% NAC (it is unknown whether nebulized 2% NAC has a clinical effect) (Howatt 1966). The Weller study compared the effects of nebulized sodium-2-mercaptoethane sulphonate and nebulized hypertonic (7%) saline (Weller 1980) and the Bishop study compared nebulized glutathione versus nebulized hypertonic saline (4%) (Bishop 2005). Nebulized hypertonic saline, both at concentrations of 4% and 7%, has been demonstrated to have beneficial effects on the clinical course of CF lung disease, so is not a true placebo (Wark 2005). The number of daily treatments was different in all three trials; in the Bishop trial there were four inhalations daily (Bishop 2005); in the Howatt trial there were three inhalations daily (Howatt 1966); and in the Weller trial there were twice daily inhalations (Weller 1980).

In the Howatt trial, participants either received the drug during positive pressure breathing treatments or while using a Devilbiss nebulizer (Howatt 1966).

Outcome measures

All three trials assessed pulmonary function at the end of treatment, but used a variety of different measures (Bishop 2005; Howatt 1966; Weller 1980). The Howatt paper only provides a table outlining the number of pulmonary function tests which improved or got worse compared to the previous month; however there is insufficient detail in the table to ascertain which specific pulmonary function tests improved (Howatt 1966). All of the trials also reported baseline values (Bishop 2005; Howatt 1966; Weller 1980).

Two trials asked participants to record cough frequency and sputum volume and color (Bishop 2005; Weller 1980). In addition Bishop asked participants to record general wellness and sputum viscosity (Bishop 2005); and Weller asked participants to record any adverse effects (Weller 1980).

One trial additionally measured BMI and six-minute walk distance (Bishop 2005). Howatt evaluated participants clinically based on the method of Shwachman and Kulczycki (Howatt 1966). Weller additionally undertook blood tests (complete blood count, liver function tests, renal function and electrolytes) and chest x-rays (Weller 1980).

Oral thiol derivatives

Five trials reported on the effects of oral thiol derivatives; three of the included trials reported on the effects of oral NAC compared to placebo (Mitchell 1982; Stafanger 1988; Stafanger 1989); one trial compared the effects of oral NAC, oral ambroxol and placebo (Ratjen 1985); and the remaining trial compared the effects of oral ambroxol and oral carbocysteine (Caramia 1995). Since the Ratjen paper reports on a three-arm trial, and we are including it in two comparisons (oral thiols versus placebo and oral thiols versus other oral agents), we realize that some participants will be counted twice in this review.

Participants

The number of participants in the trials ranged from 20 to 52. One trial experienced a high drop out rate (40%) (Stafanger 1989); because of the unusually high dropout rate (which we will discuss further under 'Risk of bias in included studies') and the fact that the results for only 10 participants out of 52 were presented in the paper, we felt that this was unreliable data and therefore it was not included in the analysis for this review.

One trial studied children only (Mitchell 1982), the others recruited a mixture of children and adults. All trials stated how many males and females were recruited and the proportions were approximately equal in all trials. All trials reported on clinical status at randomization. Two trials recorded 'mild to moderate pulmonary disease' (Mitchell 1982; Ratjen 1985); one trial recorded good clinical status (Caramia 1995); two trials reported on colonization with *Pseudomonas aeruginosa* (Stafanger 1988; Stafanger 1989). The earlier trial recruited participants who weren't chronically infected and the later trial recruited participants who were chronically infected with *Pseudomonas aeruginosa*.

Trial Design

Two trials were of parallel design (Caramia 1995; Ratjen 1985). The other three trials were of cross-over design (Mitchell 1982; Stafanger 1988; Stafanger 1989). Two trials described the use of a washout period prior to the start of the intervention and between interventions for cross-over trials (Mitchell 1982; Stafanger 1989). A third cross-over trial does not state any washout period between treatment arms (Stafanger 1988). The duration of each treatment period was three months in four of the trials (Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989) and 80 days in the fifth trial (Caramia 1995).

Interventions

Four trials compared oral NAC to placebo (Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989); one of these studies also compared oral ambroxol to placebo (Ratjen 1985). Of the trials comparing oral NAC to placebo, two trials stated that both active and placebo drugs were given in granular form and could not be distinguished from each other (Mitchell 1982; Ratjen 1985).Two trials compared one oral thiol derivative to another oral thiol derivative (Caramia 1995; Ratjen 1985) The first trial compared oral carbocysteine syrup to oral ambroxol syrup (Caramia 1995) and the second trial compared oral NAC granules to oral ambroxol granules (Ratjen 1985).

Drugs were administered three times daily in all trials with the following exceptions: in the trial of oral carbocysteine syrup versus oral ambroxol syrup, the oral ambroxol syrup was given four times daily to children under 14 years of age (Caramia 1995); in the two trials by Stafanger, oral NAC was given twice daily if the participant's weight was over 30 kg (Stafanger 1988; Stafanger 1989).

Daily dosage of oral NAC was 200 mg three times daily in two studies (Mitchell 1982; Ratjen 1985). The dose of NAC in both Stafanger papers was 200 mg three times daily in participants weighing less than 30 kg and 400 mg twice daily in participants weighing more than 30 kg (Stafanger 1988; Stafanger 1989). The dose of oral ambroxol was 30 mg three times daily in one study (Ratjen 1985), and in the other study participants under 14 years of age received 10 mg four times daily and adults received 33 mg three times daily (Caramia 1995). The dosage of carbocysteine used in the Caramia paper was 270 mg three times daily in participants under 14 years of age and 900 mg three times daily in adults (Caramia 1995).

Outcome measures

All trials measured outcomes at baseline and end of treatment (Caramia 1995; Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989). Four trials also measured outcomes in the interim period: Caramia measured outcomes at 20, 40 and 60 days (Caramia 1995); Ratjen measured outcomes at six weeks (but the study only reports end of treatment data) (Ratjen 1985); both

Stafanger trials measured outcomes on a monthly basis (Stafanger 1988; Stafanger 1989).

All trials measured pulmonary function with a variety of outcome measures, although the two Stafanger papers only reported results for pulmonary function for 23 out of 41 participants (Stafanger 1988) and for 10 participants (whose baseline PEF was less than 70% predicted) out of 52 participants (Stafanger 1989). Four trials assessed sputum characteristics (Caramia 1995; Mitchell 1982; Stafanger 1988; Stafanger 1989). Three trials took blood samples: Caramia performed an arterial blood gas analysis (Caramia 1995); two trials measured white blood cell count, sedimentation rate and antibody titres for *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Hemophilus influenza* (Stafanger 1988; Stafanger 1989). Three trials reported on adverse events (Caramia 1995; Mitchell 1982; Ratjen 1985). Two trials reported on cough frequency (Caramia 1995; Mitchell 1982). Caramia additionally reported participants subjective measurement of dyspnea (Caramia 1995) and Mitchell additionally reported on antibiotic usage (Mitchell 1982).

Excluded studies

Ten trials were excluded because they did not meet the inclusion criteria. Six trials were excluded as the duration of the intervention was less than four weeks (App 2002; Gotz 1980; Griese 2004; Maayan 1989; Tecklin 1976; Snyder 2002); three trials were excluded as they were not randomized (Dasgupta 1996; Tirouvanziam 2006b, Dietzsch 1975); one trial was excluded as the intervention was not relevant (Cezeaux 1967).

Risk of bias in included studies

The details of methodological quality of the reviewed studies are given in the table Characteristics of included studies. The information was extracted from the published papers.

Nebulized thiol derivatives

Three trials looked at nebulized thiol derivatives (Bishop 2005; Howatt 1966; Weller 1980).

Allocation

All three trials were described as randomized; however, two of these did not describe the randomization process and were therefore deemed to have an unclear risk of bias (Bishop 2005; Weller 1980). The Howatt trial randomized the order of drugs for the four-month period by making up two slips of paper for each of the six possible combinations; the participants then drew a schedule from an envelope (Howatt 1966). The trial was deemed to have a low risk of bias.

One trial does not clearly discuss allocation concealment and therefore it was judged to have an unclear risk of bias (Weller 1980).

The other two trials do give details of adequate allocation concealment and were judged to have a low risk of bias (Bishop 2005; Howatt 1966). Bishop reports that no member of the clinical team was involved in the generation of allocation to treatments (Bishop 2005). In the Howatt trial, the drugs were supplied in 10 ml vials labelled with a letter code and the key to the code was supplied in a sealed envelope, which was not opened until the study was completed (Howatt 1966).

Blinding

All three trials were described as "double-blind" (Bishop 2005; Howatt 1966; Weller 1980). Two of these trials described attempts to mask the characteristics of the different treatments (e.g. taste and odour) (Bishop 2005; Howatt 1966). These trials therefore were deemed to have low risk of bias. The Weller trial made no attempt to mask the characteristics of the different treatments and was therefore judged to have a risk of bias (Weller 1980).

Incomplete outcome data

One trial reported data from all the randomized participants and were therefore judged to have a low risk of bias (Howatt 1966). Both the other two trials reported withdrawals (Bishop 2005; Weller 1980). The numbers of drop outs in the are low and equal across treatment groups, therefore these trials have a low risk of bias.

One trial did not present data for all the participants randomized, but did not discuss the reasons for this (Howatt 1966). This trial is therefore judged to have a high risk of bias.

Weller did not report any data for FEV_1 (Weller 1980). In recently published trials it would be expected that this standard lung function test would be measured at clinic visits and information recorded. The lack of these data in a published paper could signal a potential source of bias; it may have not been reported due to negative results. However, the trial in question is nearly 30 years old and this measure of lung function may not have been employed by the trialists.

Other potential sources of bias

The Howatt trial has a number of potential sources of bias (Howatt 1966). Firstly, there is no washout period between treatments. Secondly, the drug delivery system was not consistent amongst all participants. Two participants received the drug during positive pressure breathing treatments while four participants used a Devilbiss nebulizer. Thirdly, the paper provides a table outlining the number of pulmonary function tests which improved or worsened compared to the previous month; however, there is insufficient detail in the table to ascertain which specific pulmonary function tests improved. This is an important factor since certain pulmonary function tests are clinically relevant and important outcomes to measure, while other aspects of the testing can be highly variable

and difficult to interpret. And finally, the paper only provided lung function data for two of the eight people in the trial (Howatt 1966).

Oral thiol derivatives

Five trials looked at oral thiol derivatives (Caramia 1995; Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989).

Allocation

All five trials were described as randomized; however, four of the trials did not describe the randomization process and were therefore deemed to have an unclear risk of bias (Caramia 1995; Mitchell 1982; Stafanger 1988; Stafanger 1989). In the Ratjen trial randomization was done by computer and the trial was judged to have a low risk of bias (Ratjen 1985).

All five trials do not clearly discuss allocation concealment and therefore were judged to have an unclear risk of bias (Caramia 1995; Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989).

Blinding

The Caramia trial was single-blinded, with investigators being aware of the treatment allocation (Caramia 1995). This trial design introduces a significant potential bias into the interpretation of the results of this trial.

The other four trials were described as "double-blind" (Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989). Two of these described attempts to mask the characteristics of the different treatments (e.g. taste and odour); these trials therefore were deemed to have low risk of bias (Mitchell 1982; Ratjen 1985). Neither of other two trials which were described as double-blind discussed any attempts to mask the characteristics of the different treatments (e.g. taste and odour); these trials therefore have been judged to have a potential risk of bias (Stafanger 1988; Stafanger 1989).

Incomplete outcome data

One trial reported data from all the randomized participants and were therefore judged to have a low risk of bias (Caramia 1995). We have attempted to contact Dr Caramia to obtain further data relevant to his study, but as yet we have not had any reply. If we obtain further data on this paper we plan to include this in a future update of the review.

Two trials reported withdrawals (Mitchell 1982; Ratjen 1985), but one of these does not give any reasons for the drop outs (Mitchell 1982). Mitchell does not give details of why the participants did not complete the trial, nor does he state from which treatment group they withdrew, so this trial has an unclear risk of bias (Mitchell 1982). Also, the four withdrawals were not included in the final analysis (Mitchell 1982). The numbers of drop outs in the

Ratjen trial were low and equal across treatment groups, therefore this trial has a low risk of bias (Ratjen 1985).

Two trials do not present data for all the participants randomized, but do not discuss the reasons for this (Stafanger 1988; Stafanger 1989). There was an unusually high dropout rate in the later trial where data for only 10 participants out of 52 were presented in the paper (Stafanger 1989). These two trials are therefore judged to have a high risk of bias.

Other potential sources of bias

Due to the limited amount of data reported in the Caramia paper there is a risk of bias with regards to selective reporting (Caramia 1995).

One trial has a very serious potential risk of bias as some of the data presented in the tables is incorrect (the data for total and mean values in columns 5 and 6 in Table 2 of the final published paper are not calculated correctly) (Stafanger 1989).

Two cross-over trials have a risk of bias due to either no washout period between treatments (Stafanger 1988) or an unclear duration of the washout period (Stafanger 1989).

Effects of interventions

Nebulized thiol derivatives versus other nebulized medications

Primary outcomes

1. Pulmonary function testing (PFT)

a. forced expiratory volume in one second (FEV$_1$) per cent predicted

Two studies reported on this outcome (Bishop 2005; Howatt 1966), but we were only able to enter data into the analysis for one trial (Bishop 2005). We have reported the results from the second study narratively.

The data from the Bishop study showed no significant difference in the change in FEV$_1$ comparing participants receiving nebulized glutathione and nebulized 4% saline, mean difference (MD) 0.90% (95% CI -6.45% to 8.25%) (Bishop 2005).

The Howatt study reported no statistically significant difference in FEV$_1$ following periods when participants had received nebulized 20% NAC or nebulized 2% NAC (Howatt 1966).

b. forced vital capacity (FVC) per cent predicted

Two studies reported on this outcome (Bishop 2005; Weller 1980) and we were able to enter data from both studies in the analysis but did not combine the results since one trial was parallel and one was cross-over. We found no significant difference between treatment and control groups, in either the parallel trial MD 0.60 (95% CI -6.49 to 7.69) (Bishop 2005) or the cross-over trial MD 4.00 (95% CI -6.18 to 14.18) (Weller 1980).

Secondary outcomes

1. Other pulmonary function test (PFT) measurements

All three studies reported other PFT measurements (Bishop 2005; Howatt 1966; Weller 1980), but only the Bishop and Weller study presented data that we were able to enter into the analysis, again not combined due to differences in trial design (Bishop 2005; Weller 1980). The results for change in PEF were significant in favour of nebulized thiol derivatives, MD 40.20 (95% CI 4.96 to 75.44) for the parallel trial (Bishop 2005), but not for the cross-over trial, MD 9.00 (95% CI -3.66 to 21.66) (Weller 1980). We were not able to combine data from any other PFTs (MEF$_{25-75}$, MEF$_{50}$, Vmax50%VC, RV/TLC, vital capacity, peak inspiratory flow) and none of the trials reported any significant differences (Bishop 2005; Howatt 1966; Weller 1980). We have presented data for: Vmax50%VC, MD 10.00 (95% CI -5.52 to 25.52) (Weller 1980); forced expiratory flow, MD 6.20 (95% CI -6.79 to 19.19) (Bishop 2005); and RV/TLC, MD -4.00 (95% CI -16.11 to 8.11) (Weller 1980).

2. Inflammatory markers

a. serum (WBC, CRP, ESR)

None of the included studies reported on this outcome measure.

b. sputum (IL-8)

None of the included studies reported on this outcome measure.

3. Quality of life (QOL)

None of the included studies reported on this outcome measure.

4. Need for oral antibiotics for pulmonary exacerbation

a. number of weeks of treatment

The Weller study reported that "courses of antibiotics were prescribed as frequently during baseline periods as during either treatment period". There was no indication as to whether antibiotics were oral or intravenous, and no details were supplied as to the number of weeks or number of courses of antibiotics (Weller 1980).

None of the other included studies reported on antibiotic treatment, either oral or intravenous.

b. number of courses

See comments above (in 4a) with regards to antibiotic therapy.

5. Need for intravenous antibiotics for pulmonary exacerbation

a. number of weeks of treatment

See comments above (in 4a) with regards to antibiotic therapy.

b. number of courses

See comments above (in 4a) with regards to antibiotic therapy.

6. Adverse events

The nebulized thiol derivatives investigated in the included studies were generally well-tolerated (Bishop 2005; Howatt 1966; Weller 1980).

The Bishop study reported no increased incidence of adverse effects in participants receiving nebulized glutathione compared to nebulized 4% hypertonic saline (Bishop 2005). The Howatt study reported that participants complained that nebulized NAC "smelled and tasted bad"; and one participant in the same study "complained of severe coughing attacks" while receiving nebulized 20% NAC "which would cause her to discontinue her treatment" (Howatt 1966). Several participants in the Weller study "noted that the inhalations initially made them cough, this occurring equally at the start of both therapies, but usually settling within days", but that "no major adverse effects were noted during either therapy" (Weller 1980). It should be noted that nebulized thiol derivatives are used for their potential mucolytic properties, and therefore increased cough is not necessarily an adverse effect.

7. Number of days in hospital for respiratory exacerbation

Only two studies reported on this outcome, but we were unable to enter any data in the graphs (Bishop 2005; Weller 1980). Bishop reported that "three patients were hospitalized ... due to non-improvement of conditions present at baseline". Two of these participants were receiving nebulized 4% saline and the other was receiving nebulized glutathione. Weller reported that "three patients were admitted to hospital during the course of the trial ... one of these was an inpatient for most of the trial. The other two were admitted electively because of poor growth and persisting respiratory symptoms, one during the initial baseline period and one during the second month of saline treatment" (Weller 1980).

8. Adherence

Only one study reported on this outcome and stated that one participant receiving nebulized glutathione "was grossly noncompliant, ie, stopped the treatment after the first 5 days of the trial". No reason was given for this non-adherence, but the participant was included in the modified ITT analysis (Bishop 2005).

9. Acquisition of new respiratory pathogens (%)

Only one study reported on this outcome (Weller 1980). It was reported that there "was no change in sputum microbiology - for example, appearance of *Pseudomonas aeruginosa*" between the intervention groups (Weller 1980).

10. Six-minute walk distance

Only the Bishop study reported on this outcome (Bishop 2005). There was no significant difference in 6-minute walk distance between participants receiving nebulized glutathione and nebulized 4% saline, MD 26.90m (95% CI -115.40m to 169.20m) (Bishop 2005).

11. Sputum characteristics

All three studies reported on this outcome (Bishop 2005; Howatt 1966; Weller 1980); but we were only able to enter data from one study for sputum viscosity in the analysis (Bishop 2005).

Bishop reported no significant differences in sputum amount, viscosity or color between the participants receiving nebulized glutathione or nebulized 4% saline; we present data for sputum viscosity, MD -0.40 (95% CI -1.53 to 0.73) (Bishop 2005).

The Howatt study reported that several participants noticed their "sputum was thinner and it seemed easier to bring up" in both groups (Howatt 1966).

The Weller reported "no significant differences in cough frequency, sputum volume, or sputum colour on analysis" in either group (Weller 1980).

Oral thiol derivatives versus placebo

Primary outcomes

1. Pulmonary function testing (PFT)

a. forced expiratory volume in one second (FEV$_1$) per cent predicted

Three studies reported on this outcome (Ratjen 1985; Stafanger 1988; Stafanger 1989); however, we were only able to enter data in the analysis from one of these studies (Ratjen 1985).

When entered into the analysis, data from the Ratjen study showed no significant difference in FEV$_1$, MD 5.00 per cent predicted (95% CI -15.22 to 25.22) between those participants who received oral NAC compared to placebo.

In the earlier study, Stafanger reported no overall significant difference in FEV$_1$ comparing participants receiving oral NAC and those receiving placebo (Stafanger 1988).

In the later study, Stafanger reported no overall significant difference in FEV$_1$ comparing periods when participants received oral NAC and those when they received placebo (Stafanger 1989).

b. forced vital capacity (FVC) per cent predicted

Two studies reported on this outcome, but we were unable to enter data into the analysis for either study (Stafanger 1988; Stafanger 1989).

In the 1988 study, Stafanger reported no overall significant difference in FVC comparing participants receiving oral NAC and those receiving placebo (Stafanger 1988).

In the 1989 study, Stafanger also reported no overall significant difference in FVC comparing periods when participants received oral NAC and periods when they received placebo (Stafanger 1989).

Secondary outcomes

1. Other PFT measurements

All four studies reported a range of other PFT measurements (Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989). Two studies presented data that we were able to enter into the analysis, but we were not able to combine these as they were used different outcome measures (Mitchell 1982; Ratjen 1985).

Data from the Mitchell study show no significant difference in PEF between the periods when participants received oral NAC

compared to when they received placebo, MD -3.00 (95% CI -60.52 to 54.52) (Mitchell 1982).

Ratjen showed no significant difference in any measurements of pulmonary function testing when participants received oral NAC and placebo: PEF, MD 5.90 (95% CI -17.75 to 29.55); TLC, MD -1.70 (95% CI -14.60 to 11.20); FVC$_{75\%}$, MD 15.90 (95% CI -10.55 to 42.35); FVC$_{50\%}$, MD -7.90 (95% CI -36.87 to 21.07); FVC$_{25\%}$, MD -4.00 (95% CI -34.92 to 26.92); FEV$_1$/VC, MD 7.20 (95% CI -10.47 to 24.87); TGV, MD 4.20 (95% CI -29.08 to 37.48); VC, MD -1.00 (95% CI -12.53 to 10.53); and RV/TLC, MD -17.20 (95% CI -67.98 to 33.58)) (Ratjen 1985). Ratjen also reported no significant difference in airway resistance, but we were not able to enter data into the analysis for this outcome (Ratjen 1985).

In the 1988 study, Stafanger reported no significant difference in PEF between periods when participants received oral NAC compared to periods when they received placebo (Stafanger 1988).

In the 1989 study, Stafanger reported no overall significant difference in PEF comparing periods when participants received oral NAC and when they received placebo (Stafanger 1989).

Inflammatory markers

a. serum (WBC, CRP, ESR)

Only two studies reported on this outcome (Stafanger 1988; Stafanger 1989). Both studies reported no difference in white blood cell count or erythrocyte sedimentation rate between those who received NAC and those who received placebo.

b. sputum (IL-8)

None of the included studies reported on this outcome measure.

3. Quality of life (QOL)

None of the included studies reported on this outcome measure.

4. Need for oral antibiotics for pulmonary exacerbation

a. number of weeks of treatment

Four studies reported on this outcome (Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989); but only one study presented data which we could enter into our analysis (Mitchell 1982). There is a high possibility that these data may be skewed since people with CF who require antibiotics usually receive them for at

least two weeks, but a small group of people who are more severely infected will receive them for much longer.

Mitchell does not differentiate between the use of oral or intravenous antibiotic therapy, but states there was no significant difference between antibiotic treatment between participants in the NAC group versus those in the placebo group, MD -0.40 weeks (95% CI -2.98 to 2.18) (Mitchell 1982).

In the paper, Ratjen reported that "no patients needed intermittent antibiotic therapy during the course of the trial" (Ratjen 1985). Neither of the studies by Stafanger reported any difference in antibiotic treatment between periods when the participants received oral NAC or placebo (Stafanger 1988; Stafanger 1989). There was no indication as to whether antibiotics were oral or intravenous, and no details were supplied as to the number of weeks or number of courses of antibiotics. It should be noted that all participants in later study by Stafanger entered the study having recently completed a course of intravenous antibiotics (Stafanger 1989).

b. number of courses

See comments above (in 4a) with regards to antibiotic therapy.

5. Need for intravenous antibiotics for pulmonary exacerbation

a. number of weeks of treatment

See comments above (in 4a) with regards to antibiotic therapy.

b. number of course

See comments above (in 4a) with regards to antibiotic therapy.

6. Adverse events

Three studies reported on this outcome (Mitchell 1982; Ratjen 1985; Stafanger 1989). Two of these studies did not report any adverse effects in either treatment or control groups (Mitchell 1982; Ratjen 1985). In the 1989 study, Stafanger reported that one individual developed Quincke's oedema and one developed a rash while receiving oral NAC (Stafanger 1989). In both participants these adverse effects settled once oral NAC was stopped. Two participants also developed abdominal pain, one while receiving oral NAC and one receiving placebo (Stafanger 1989). A further participant in this same study felt that "she coughed more frequently and less productively with NAC and stopped the treatment". All of these participants were excluded from the study due to these adverse effects (Stafanger 1989).

7. Number of days in hospital for respiratory exacerbation

Only one study reported on this outcome (Stafanger 1988). This study reported that none of the participants receiving either oral NAC or placebo were hospitalized during the study (Stafanger 1988).

8. Adherence

Three studies reported on this outcome (Ratjen 1985; Stafanger 1988; Stafanger 1989).

Ratjen reported that two participants dropped out of the study due to "irregular drug intake" (Ratjen 1985). In the earlier study, Stafanger reported that two participants were excluded due to "poor co-operation", but did not specify whether this reflected poor adherence (Stafanger 1988). Similarly, in the 1989 study, Stafanger reported that 10 participants were excluded due to "poor co-operation", but did not elaborate (Stafanger 1989).

9. Acquisition of new respiratory pathogens (%)

None of the included studies reported on this outcome measure.

10. Six-minute walk distance

None of the included studies reported on this outcome measure.

11. Sputum characteristics

All four studies required participants to complete subjective scores of sputum characteristics whilst receiving the different interventions (Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989). One of the included studies presented data for this outcome measure (Stafanger 1988) and the other three studies reported these results narratively (Mitchell 1982; Ratjen 1985; Stafanger 1989). None of the four studies reported significant differences in these subjective scores of sputum characteristics between participants receiving oral NAC and placebo (Mitchell 1982; Ratjen 1985; Stafanger 1988; Stafanger 1989). Ratjen also reported narratively that there were no significant differences in a subjective score of sputum characteristics between participants receiving oral ambroxol and placebo (Ratjen 1985).

Oral thiols derivatives versus other oral thiol derivatives

Primary outcomes

1. Pulmonary function testing (PFT)

a. forced expiratory volume in one second (FEV$_1$) per cent predicted

Two trials reported on this outcome (Caramia 1995; Ratjen 1985), but only data from one trial was included in the analysis (Ratjen 1985). Ratjen showed no significant difference in FEV$_1$ between groups receiving oral NAC and oral ambroxol -4.20% (95% CI -26.19% to 17.79%) (Ratjen 1985). Caramia presents data showing the change from baseline, but not from both the treatment and control groups, so we are unable to enter the data into the analysis (Caramia 1995). Caramia also reported that there was no significant difference in FEV$_1$ between groups receiving oral carbocysteine and oral ambroxol (Caramia 1995).

b. forced vital capacity (FVC) per cent predicted

None of the included studies reported on this outcome measure

Secondary outcomes

1. Other PFT measurements

Two trials reported on a range of other PFT measurements and but only data from one trial could be included in the analysis (Ratjen 1985).

The Ratjen paper showed no significant differences in other PFT measurements between participants receiving oral NAC and oral ambroxol: PEF, MD -0.60 (95% CI -22.21 to 21.01); TLC, MD 1.30 (95% CI -11.18 to 13.78); FVC$_{75\%}$, MD 6.00 (95% CI -18.83 to 30.83); FVC$_{50\%}$, MD 0.00 (95% CI -30.02 to 30.02); FVC$_{25\%}$, MD -4.10 (95% CI -37.74 to 29.54); FEV$_1$/VC, MD -0.90 (95% CI -15.70 to 13.90); TGV, MD 13.50 (95% CI -13.92 to 40.92); VC, MD 2.80 (95% CI -12.75 to 18.35); and RV/TLC, MD -11.50 (95% CI -82.21 to 59.21) (Ratjen 1985). Data from the Caramia study found no significant differences in TV, PEF, MEF$_{25}$ or MEF$_{75}$ between participants receiving oral carbocysteine and oral ambroxol (Caramia 1995).

Inflammatory markers

a. serum (WBC, CRP, ESR)

None of the included studies reported on this outcome measure.

b. sputum (IL-8)

None of the included studies reported on this outcome measure.

3. Quality of life (QOL)

None of the included studies reported on this outcome measure.

4. Need for oral antibiotics for pulmonary exacerbation

a. number of weeks of treatment

None of the included studies reported on this outcome measure.

b. number of courses

None of the included studies reported on this outcome measure.

5. Need for intravenous antibiotics for pulmonary exacerbation

a. number of weeks of treatment

None of the included studies reported on this outcome measure.

b. number of course

None of the included studies reported on this outcome measure.

6. Adverse events

The Caramia and Ratjen studies did not report any adverse effects in participants receiving oral thiol derivatives (Caramia 1995; Ratjen 1985).

7. Number of days in hospital for respiratory exacerbation

None of the included studies reported on this outcome measure.

8. Adherence

None of the included studies reported on this outcome measure.

9. Acquisition of new respiratory pathogens (%)

None of the included studies reported on this outcome measure.

10. Six-minute walk distance

None of the included studies reported on this outcome measure.

11. Sputum characteristics

Ratjen reported no significant differences in a subjective score of sputum characteristics between participants receiving oral NAC and oral ambroxol (Ratjen 1985).

Caramia included a subjective score of sputum characteristics, but also assessed sputum viscosity and elasticity objectively using an oscillometric visco-elastometer (Caramia 1995). There was no difference in the subjective score between participants receiving oral carbocysteine or oral ambroxol (Caramia 1995), and there were also no differences in viscosity or elasticity between participants receiving the two treatments (Caramia 1995).

DISCUSSION

Summary of main results

There have been very few good quality studies investigating the effects of nebulized or oral thiol derivatives in cystic fibrosis. The eight included studies assessed different thiol derivatives and used very different study designs.

For nebulized thiol derivatives, the included studies report no evidence that any of the nebulized thiol derivatives have a significant beneficial effect on the primary outcome measures of this review (FEV_1 per cent predicted and FVC per cent predicted). There were conflicting results with regards to the effect of nebulized thiol derivatives on other pulmonary function tests, but overall no convincing evidence that they are of clinical benefit. There was also no evidence of a beneficial effect of nebulized thiol derivatives on any of the other secondary outcome measures in this review. In summary, we have found no evidence to recommend that nebulized thiol derivatives should be used in routine clinical practice in patients with CF.

For oral thiol derivatives, the five included studies assessed three different drugs, using different preparations (tablets, granules and syrup) and doses (some using a fixed dose and some dosing according to body weight). None of the included studies demonstrated a significant benefit of oral thiol derivatives on any of the primary or secondary outcome measures of this review.

Several methodological challenges were apparent during this review. The vast majority of the studies included very small sample sizes and may have been underpowered. Although pulmonary function testing was typically reported, the type of pulmonary function testing was highly variable with some studies reporting on percent predicted FEV_1 while others reported the peak expiratory flow measurements. This may represent selective reporting whereby only certain measurements were recorded in the final papers and may be a source of potential bias. No studies were powered to look at other clinically important outcomes such as need for hospitalization, new acquisition of bacteria, or changes

in quality of life. Two of the larger studies had unacceptably high dropout rates which raised significant questions as to the reliability of the study results (Stafanger 1988; Stafanger 1989). Because some groups of individuals with CF may respond more than others, we had planned to do subgroup analyses for factors such as severity of lung disease and age. None of the studies provided adequate information on these subgroups, making it impossible to carry out any subgroup analyses.

Overall completeness and applicability of evidence

The included studies were generally complete, although the exclusion criteria of the studies and the age range of the included participants limits their applicability to the general CF population.

Quality of the evidence

The evidence on which this review is based is limited in terms of the quantity and quality of included studies. In all eight studies the generation of allocation sequences was stated as randomized, but in only two of these (Howatt 1966; Ratjen 1985) was the method of randomization adequate, as defined by Jüni (Jüni 2001). The concealment of treatment allocation was unclear in six of the included studies, with only two (Howatt 1966; Ratjen 1985) having an adequate method of allocation concealment (Jüni 2001). Seven of the eight included studies stated that they were double blinded, with one stating that it was single blinded (Caramia 1995). Four trials backed up this statement with an explanation of how they had masked the characteristics of the different interventions (Bishop 2005; Howatt 1966; Mitchell 1982; Ratjen 1985); but three trials made no attempt to disguise the interventions (Stafanger 1988; Stafanger 1989; Weller 1980). Additionally, in the 'placebo-controlled' studies assessing nebulized thiol derivatives, the placebo intervention was either a weaker solution of the active intervention, or was an active intervention in its own right (such as nebulized hypertonic saline). In the case of nebulized hypertonic saline the beneficial effect of this intervention has only been confirmed in recent years and was not known to the investigators in the relevant included studies.

Potential biases in the review process

One of the co-authors of this review, Professor Felix Ratjen, is lead investigator on one of the included trials.

Agreements and disagreements with other studies or reviews

The results of this review are broadly in agreement with the systematic review of NAC in CF prepared by Duijvestijn and Brand (Duijvestijn 1999).

AUTHORS' CONCLUSIONS

Implications for practice

From this review of eligible trials, we have not been able to identify any evidence to recommend the use of nebulized thiol derivatives in people with CF. We have also not been able to find any evidence to recommend the use of oral thiol derivatives in the management of CF lung disease.

Implications for research

Despite the paucity of literature on the effectiveness of thiol derivatives in CF, these are still potentially useful drugs for further study because thickened mucous, leading to chronic infection and inflammation and respiratory failure is still the most common cause of early death in CF. Therapies aimed at improving sputum clearance and therefore reducing pulmonary infection and inflammation are sorely needed. The other effective mucolytic therapies that are available for pulmonary disease in CF are unable to be tolerated by some people due to side effects (mainly bronchospasm) and can also be prohibitively expensive (in the case of dornase alfa). Thiol derivatives are relatively inexpensive, and, especially when administered orally, are well-tolerated. Further studies are required to investigate the potential beneficial effects of both nebulized and oral thiol derivatives in CF lung disease.

Well-designed randomized placebo controlled double-blind trials of people with CF over six years of age are required to adequately assess the potential benefit of these medications. Pulmonary function testing cannot be reliably done in individuals under the age of six years. A wide range of disease severities and age groups should be included in these studies and in sufficient numbers so that data on these subgroups can be analyzed separately. It is not clear from the literature if mucolytic therapy is helpful in early disease to prevent decline in lung function or whether it is more advantageous later in the disease course when mucous production is increased. Enrolling participants with a wide range of disease severity may help to elucidate this. As mucolytic therapy takes time to exert its effect, the duration of the trials should be no less than four months, with six months of follow up to evaluate the long-term effects of therapy with thiol derivatives. Pulmonary function (specifically FEV_1 and FVC), would be reasonable primary endpoints, while other secondary endpoints such as markers of inflammation, need for both IV and oral antibiotics therapy, number of days in hospital, quality of life, acquisition of new respiratory pathogens, and radiologic improvement would be key in assessing the efficacy of these medications. Mortality, although an important hard endpoint, would not be recommended as an outcome measure as it is unlikely changes in mortality would be detected in a 4 to 12 month trial given the median survival in CF at present.

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* *Indicates the major publication for the study*

CHARACTERISTICS OF STUDIES

Characteristics of included studies *[ordered by study ID]*

Bishop 2005

Methods	Randomized, double-blind, placebo-controlled study. Single center in the USA. 8-week study period, with participants receiving the same intervention for the whole 8 weeks A "modified" ITT analysis was carried out whereby all patient outcomes were included, regardless of compliance, except when patients were hospitalized or were missing data
Participants	Nineteen participants were recruited. Mean age 13.1 years (range 6 - 19 years). Stable clinical status with mild pulmonary disease. Participants were paired by age and sex, and then each member of the pair was randomly assigned to the treatment or placebo groups One participant in the placebo group dropped out 'just before the trial began', and was not included in the analysis
Interventions	Nebulized buffered reduced GSH versus nebulized placebo. GSH was buffered with sodium bicarbonate and dosed at 66mg/kg body weight. Placebo was composed of sodium chloride dosed at 15mg/kg body weight, and quinine at 25-30 μg/kg body weight. Participants were instructed to self-administer their intervention medication "across 4 inhalation sessions per day" and asked to "space these sessions 3- to 4-h apart". For the first week of the study, participants "were instructed to use one fourth of the recommended total dosage, and in the second week to use one half of the recommended total dosage. After the second week, patients were instructed to use the full daily total dosage"
Outcomes	Primary outcome measures: FEV_1 (% predicted), FVC (% predicted), FEF_{25-75} (% predicted), PEF. FEV_1, FVC and FEF_{25-75} were measured once prior to starting the intervention period, and once after completing the intervention period. PEF was measured by the participant twice daily throughout the study Secondary outcome measures: Objective - BMI, 6-min walk distance (m). These objective measures were recorded once prior to starting the intervention and once after completing the intervention Subjective: • Sputum color (scale ranging from 1 = clear to 6 = blood streaked) • Sputum amount (1=scant, 2 = <1 teaspoon), 3 = >1 teaspoon) • Sputum viscosity (1 = very thin, 2 = slightly sticky, 3 = very sticky) • Cough frequency (1 = no cough, 2 = infrequent, 3 = several times per day, 4 = every hour) • General wellness (1 = poor, 2 = fair, 3 = good, 4 = excellent) • Usual stamina (1 = poor, 2 = fair, 3 = good, 4 = excellent) • Improvement (1 = significantly worse, 2 = a bit worse, 3 = about the same, 4 = a bit better, 5 = significantly better).

Bishop 2005 (*Continued*)

	The baseline measure for each of the subjective measures was taken as the 'average of the first 5 days of the trial', and the 'end of trial measure' was taken as the 'average of last 5 days of the trial' Adverse events.
Notes	Paper states that in the analysis, 'differences between post-trial and baseline outcomes were analyzed using GLMM that allowed for correlation between outcomes with the age/sex pair used for randomization.'

Risk of bias

Item	Authors' judgement	Description
Adequate sequence generation?	Unclear	Randomization process not described.
Allocation concealment?	Yes	No member of the clinical team was involved in the generation of the sequence of allocation to treatments
Blinding? Participants	Yes	Trial described as "double-blind" and attempts were made to mask the characteristic odour of nebulized glutathione by adding quinine to the 4% hypertonic saline
Blinding? Clinicians/assessors	Yes	Trial described as "double-blind" and attempts were made to mask the characteristic odour of nebulized glutathione by adding quinine to the 4% hypertonic saline
Incomplete outcome data addressed? All outcomes	Unclear	Three individuals hospitalized during the study were excluded from data analysis, and therefore data from nine participants receiving nebulized glutathione, and seven participants receiving nebulized 4% hypertonic saline were reported in the study
Free of other bias?	Unclear	No other sources of bias identified.

Caramia 1995

Methods	Randomized, single-blind, parallel-group design. Single center in Italy. 80-day intervention period. ITT analysis was carried out, with all participants completing the trial and all their data being analysed
Participants	26 CF participants (12 males). Age range 8 - 26 years (mean (SE) 15.9 (1.7) years in the SCMS-Lys group, 16.2 (1.5)

Caramia 1995 (*Continued*)

years in the ABX group)

Good clinical status (Schwachmann index 76.2 (2.8) in the SCMS-Lys group, 77.3 (3.5) in the ABX group)

"Concomitant administration of antitussives, muco-actives, sedatives, H_1-receptor antagonists and systemic corticosteroids was not allowed during the study"

Interventions	One group received SCMS-Lys at a dose of 900mg tid in adults and 270mg tid in children under 14 years of age. The other group received oral ABX at dose of 33mg tid in adults and 10 mg qid in children under 14 years of age
Outcomes	At baseline, 20, 40, 60 and 80 days of the intervention period, participants had the following assessments performed: • Cough frequency, "intensity of dyspnoea/tachypnoea" and chest sound abnormalities as assessed and rated on a 5-point digital scale (1 = greatest degree of abnormality, 5 = absence of symptom or sign). • Viscosity and elasticity of expectorated sputum as assessed by an "oscillometric visco-elastometer". • Estimated sputum volume over the preceding 24-hour period was also rated (1 = >50 mls/day, 2 = 25-50 mls/day, 3 = <25 mls/day, 4 = 'little or no expectorate'). • Arterial blood gas analysis. • Tidal volume, FEV_1, PEF, MEF_{25}, MEF_{50}, MEF_{75} and 'Tiffeau index'. • Assessment of adverse effects. In addition, the Shwachmann score was assessed at baseline and at 80 days
Notes	

Risk of bias

Item	Authors' judgement	Description
Adequate sequence generation?	Unclear	Randomization process not described.
Allocation concealment?	Unclear	Trial does not clearly discuss allocation concealment.
Blinding? Participants	Yes	Single-blinded, with investigators being aware of the treatment allocation
Blinding? Clinicians/assessors	No	Single-blinded, with investigators being aware of the treatment allocation
Incomplete outcome data addressed? All outcomes	Unclear	All 26 participants completed the study protocol and data from all participants were reported
Free of other bias?	Unclear	No other sources of bias identified.

Howatt 1966

Methods	Randomized, double-blind, controlled, cross-over design.
	Single centre in USA.
	4-month trial period, participants receiving interventions for 1 month at a time. No washout period.
	No dropouts.
	Data was analysed by ITT, however only limited data was presented in the paper

Participants	8 CF participants (3 male).
	Age range 6 - 22 years (mean 12.6).
	Clinical status ranged from "excellent" to "moderate" (based on the method of Shwachman and Kulczycki).
	4 participants stopped using nebulized isoproterenol, antibiotics, 3% saline during the trial period, whereas the other 4 participants did not alter their pre-trial therapy.
	None of the participants had ever received NAC before the trial

Interventions	Nebulized 20% NAC tid, versus nebulized 2% NAC tid.

Outcomes	Subjective improvement in sputum thickness and ability to expectorate, PFTs (VC, PEF, PIF, E_{50}, FEV1, SBO), adverse reactions.

Notes	Participants had a total of 4 months of treatment but could receive it monthly as follows:
	1st combo: 20%NAC then 2%NAC then 20%NAC then 2%NAC
	2nd combo: 2%NAC then 20%NAC then 2%NAC then 20%NAC
	3rd combo: 20%NAC then 20%NAC then 2%NAC then 2%NAC
	4th combo: 2%NAC then 2%NAC then 20%NAC then 20%NAC
	5th combo: 2%NAC then 20%NAC then 20%NAC then 2%NAC
	6th combo: 20%NAC then 2%NAC then 2%NAC then 20%NAC

Risk of bias

Item	Authors' judgement	Description
Adequate sequence generation?	Yes	Randomized order of drugs by making up 2 slips of paper for each of the 6 possible combinations
Allocation concealment?	Yes	Drugs were supplied in 10 ml vials labelled with a letter code and the key to the code was supplied in a sealed envelope, which was not opened until the study was completed
Blinding? Participants	Yes	Trial described as "double-blind" and attempts were made to mask the characteristic odour of the treatments by using different concentrations of nebulized NAC
Blinding? Clinicians/assessors	Yes	Trial described as "double-blind" and attempts were made to mask the characteris-

Howatt 1966 (*Continued*)

		tic odour of the treatments by using different concentrations of nebulized NAC
Incomplete outcome data addressed? All outcomes	Unclear	8 participants were entered into the trial, but pulmonary function data is only reported for 2 out of these 8
Free of other bias?	Unclear	Trial has a risk of bias due to no washout period between treatments. The drug delivery system was not consistent amongst all participants. 2 participants received the drug during positive pressure breathing treatments while 4 participants used a Devilbiss nebulizer. A table is provided in the paper outlining the number of PFTs which improved or got worse compared to the previous month; however there is insufficient detail in the table to ascertain which specific PFTs improved

Mitchell 1982

Methods	Randomized, double-blind, placebo-controlled, cross-over study design. Single centre in New Zealand. Initial 2-week training period where all participants took placebo. Duration 6 months (3 months in each limb, with a 2-week training period and a 2-week wash out period when all participants took placebo) Not ITT as 4 participants withdrew from the trial and were not included in final analysis
Participants	20 children (10 male) with CF. Mean (SD) age 10.8 (5.9) years. Stable mild to moderate pulmonary disease (mean (SD) Schwachman score 76 (10)). Aerosolized mucolytic therapy was stopped during the trial period
Interventions	3 months on oral placebo and 3 months on 200 mg oral NAC tid
Outcomes	Clinical assessment, body weight, CXR score, daily best-of-three PEF, antibiotic usage, cough frequency (scale of 0 - 3), and self-assessed sputum viscosity (scale of 0 - 3). Numerical results only provided for weight change, duration on antibiotics (not stated whether oral or intravenous) and PEF
Notes	

Risk of bias

Item	Authors' judgement	Description
Adequate sequence generation?	Unclear	Randomization process not described.

Mitchell 1982 (*Continued*)

Allocation concealment?	Unclear	Trial does not clearly discuss allocation concealment.
Blinding? Participants	Unclear	The study drugs were similar in appearance and the study was described as being "double-blind" with no specific details on who was blinded. Both the active drug and the placebo were made of orange flavoured granules
Blinding? Clinicians/assessors	Unclear	The study drugs were similar in appearance and the study was described as being "double-blind" with no specific details on who was blinded. Both the active drug and the placebo were made of orange flavoured granules
Incomplete outcome data addressed? All outcomes	Unclear	Four participants withdrew from the trial; therefore, 16 were included in the final analysis
Free of other bias?	Unclear	No other sources of bias identified.

Ratjen 1985

Methods	Randomized, double-blind, placebo-controlled, parallel study design. Single centre in Germany. 2-week washout period prior to starting treatment. Duration 12 weeks. Not ITT (4 participants dropped out and were not included in final analysis)
Participants	36 participants with CF (16 male). Age 6 - 21 years (mean 13.9). Mild to moderate lung disease. Atopic individuals, and those on bronchodilators excluded.
Interventions	3 treatment arms. Oral NAC 200 mg tid, oral ABX 30 mg tid or placebo, each for 12 weeks
Outcomes	PFTs (Raw, TGV, sRaw, REZ, VC, FEV1, FEV1/VC, V 75% FVC, V 50% FVC, V 25% FVC, PEF, TLC, RV/TLC) recorded after washout period, at 6 weeks and 12 weeks. PFTs reported as mean values as percentage of normal, with no absolute values reported. After the study, parents and participants were asked whether they had improved, deteriorated or remained stable during the study, and if they thought that they had received a drug or placebo during the trial
Notes	

Ratjen 1985 (*Continued*)

Risk of bias		
Item	**Authors' judgement**	**Description**
Adequate sequence generation?	Yes	Computer-generated randomization.
Allocation concealment?	Unclear	Trial does not clearly discuss allocation concealment.
Blinding? Participants	Unclear	The study drugs were similar in appearance and the study was described as being "double-blind" with no specific details on who was blinded. Both the active drug and the placebo were of granular appearance and could not be distinguished with regard to taste, colour or odour
Blinding? Clinicians/assessors	Unclear	The study drugs were similar in appearance and the study was described as being "double-blind" with no specific details on who was blinded. Both the active drug and the placebo were of granular appearance and could not be distinguished with regard to taste, colour or odour
Incomplete outcome data addressed? All outcomes	Unclear	36 participants entered the trial with 4 withdrawals; therefore the final analysis was on 32 participants. 4 participants did not complete the study due to irregular drug intake, missed clinic appointments, or clinical deterioration
Free of other bias?	Unclear	No other sources of bias identified.

Stafanger 1988

Methods	Randomized, double-blind, placebo-controlled, cross-over design. Single centre in Denmark. 6 months on each intervention, followed by 3 months follow-up. Not ITT as 3 participants were excluded from the final analysis
Participants	41 participants with CF (23 males). Age 2 - 31 years (mean 9.5). None were infected by *Pseudomonas aeruginosa*. Stable disease, but disease severity not stated. Exclusions: past history of peptic ulcer disease, liver or kidney disease and pregnancy

Stafanger 1988   *(Continued)*

Interventions	3 periods, each 3 months duration. First period oral NAC (200mg tid if <30kg, 400mg bid if >30kg) or placebo, then cross over to the other intervention, then 3 months follow up
Outcomes	Subjective scores of symptoms, body weight, sputum bacteriology and PFTs (FEV1, FVC, PEF) recorded every month. 3-monthly serum WBC, ESR and *Staphylococcal aureus*, *Haemophilus influenzae* and *Pseudomonas aeruginosa* antibody titres. Time on antibiotics also recorded. Ciliary function was also studied (ciliary beat frequency and ciliary beating pattern) in 20 participants
Notes	

Risk of bias

Item	Authors' judgement	Description
Adequate sequence generation?	Unclear	Randomization process not described
Allocation concealment?	Unclear	Trial does not clearly discuss allocation concealment.
Blinding? Participants	Unclear	Trial described as "double-blind" but no details were given on who was blinded
Blinding? Clinicians/assessors	Unclear	Trial described as "double-blind" but no details were given on who was blinded
Incomplete outcome data addressed? All outcomes	Unclear	Studied 44 participants in this trial, and while 41 participants completed the study, data on only 23 participants are presented in the paper
Free of other bias?	Unclear	Trial has a risk of bias due to no washout period between treatments

Stafanger 1989

Methods	Randomized, double-blind, placebo-controlled, cross-over study design. Single centre in Denmark. 2 periods of 3 months receiving either active drug or placebo. All participants received intravenous antibiotics routinely on a 3-monthly basis, once before starting the trial and again at the mid-point of the trial Not ITT as 21 participants were excluded from the final analysis
Participants	52 participants with CF, with 31 (17 males) completing it. Mean age 15 years (range 7 - 33). All were chronically infected with *Pseudomonas aeruginosa*.

Stafanger 1989 (*Continued*)

	Lung function ranged from severely impaired to normal. All pre-trial treatments continued during the trial.
Interventions	Oral NAC 200 mg tid (< 30kg), oral NAC 400 mg BD (> 30kg), or placebo (bicarbonate tablets)
Outcomes	Monthly 'subjective score', body weight, sputum bacteriology and PFTs (FVC, FEV1, PEF). Blood test for WBC, ESR and antibodies to *Staphylococcal aureus*, *Haemophilus influenzae* and *Pseudomonas aeruginosa* at the start of the trial and at the end of each 3-month period. Ciliary function also assessed
Notes	

Risk of bias

Item	Authors' judgement	Description
Adequate sequence generation?	Unclear	Randomization process not described.
Allocation concealment?	Unclear	Trial does not clearly discuss allocation concealment.
Blinding? Participants	Unclear	Trial described as "double-blind" but no details were given on who was blinded
Blinding? Clinicians/assessors	Unclear	Trial described as "double-blind" but no details were given on who was blinded
Incomplete outcome data addressed? All outcomes	Unclear	52 individuals entered the trial, but only 31 completed the trial resulting in a 40% dropout rate The paper presents data only on a subgroup of participants (n = 10) who have baseline PEF < 70%. There is no data provided on the remaining 21 participants
Free of other bias?	Unclear	Trial has a very serious potential risk of bias as some of the data presented in the tables is incorrect (the data for total and mean values in columns 5 and 6 in Table 2 are not calculated correctly). There was also an unclear duration of the washout period. Participants were also excluded from the study due to adverse effects

Weller 1980

Methods	Randomized, double-blind, cross-over study. Single centre in UK. 2-month baseline periods preceded and followed 2 intervention periods each lasting 8 weeks. Not ITT as 2 participants were not included in the final analysis
Participants	29 children with CF. 27 children completed the trial (13 male). Age 6 - 15 years (mean 10.7). Disease severity not reported.
Interventions	Nebulized 3 ml sodium 2-mercaptoethane sulphonate (Mistabron) 20% solution bid or nebulized 3 ml 7% saline bid. Both nebulized from a Wright nebulizer operated by an air compressor (8 litres/minute)
Outcomes	Diary card record of post-physiotherapy sputum volume, sputum color and cough frequency (recorded on a scale from 1 to 3). Sputum culture and PFTs (PEF, FVC, Vmax 50% VC and RV/TLC) taken every month. CXR (Crispin and Norman score), full blood count, liver function tests, electrolytes and creatinine taken at the beginning and end of the study period
Notes	

Risk of bias

Item	Authors' judgement	Description
Adequate sequence generation?	Unclear	Randomization process not described.
Allocation concealment?	Unclear	Trial does not clearly discuss allocation concealment.
Blinding? Participants	Unclear	Trial described as "double-blind" but no details were given on who was blinded. No attempts were made to disguise the taste or odour of the interventions
Blinding? Clinicians/assessors	Unclear	Trial described as "double-blind" but no details were given on who was blinded. No attempts were made to disguise the taste or odour of the interventions
Incomplete outcome data addressed? All outcomes	Unclear	2 children were withdrawn from the study (one due to an acute pulmonary exacerbation while receiving nebulized 7% hypertonic saline and one due to non-compliance), and therefore data for 27 children are reported in the study
Free of other bias?	Unclear	No other sources of bias identified.

ABX: ambroxol hydrochloride
BID: twice daily
CF: cystic fibrosis
CXR: chest X-ray
ESR: erythrocyte sedimentation rate
E_{50}: forced flow rate when 50% of VC has been expired (synonymous with FEF_{50})
FEF_{50}: forced expiratory flow at 50% of VC
FEV_1: forced expiratory volume at one second
FVC: forced vital capacity
GSH: glutathione
ITT: intention to treat
MMEFR: maximum mid-expiratory flow rate
MMV: maximum voluntary ventilation
NAC: n-acetylcysteine
PEF: peak expiratory flow
PFT: pulmonary function test
PIF: peak inspiratory flow
QID: four times daily
Raw: upper airway resistance
REZ: oscillometric determination of airway resistance
RV: residual volume
SBO: small bowel obstruction
SCMS-Lys: oral carbocysteine lysine salt monohydrate
SD: standard deviation
sRaw: specific airways resistance
TGV: thoracic gas volume
TID: three times daily
TLC: total lung capacity
TV: Tidal volume
V 25% FVC: maximal expiratory flow in 25% vital capacity
V 50% FVC: maximal expiratory flow in 50% vital capacity
V 75% FVC: maximal expiratory flow in 75% vital capacity
VC: vital capacity
WBC: white blood cell

Characteristics of excluded studies *[ordered by study ID]*

Study	Reason for exclusion
App 2002	Intervention period less than 4 weeks.
Cezeaux 1967	Intervention not applicable (bronchial lavage under general anaesthetic)
Dasgupta 1996	Non-randomized, in vitro study of the effects of rhDNase and NAL on sputum spinnability and rheology
Dietzsch 1975	Study not randomized.
Gotz 1980	Intervention period less than 4 weeks.

Griese 2004	Intervention period less than 4 weeks.
Maayan 1989	Intervention period less than 4 weeks.
Snyder 2002	Intervention period less than 4 weeks.
Tecklin 1976	Intervention period less than 4 weeks.
Tirouvanziam 2006b	Not a randomized controlled trial.

NAC: n-acetylcysteine
NAL: nacystelyn

DATA AND ANALYSES

Comparison 1.   Nebulized thiols versus other nebulized medications

Outcome or subgroup title	No. of studies	No. of participants	Statistical method	Effect size
1 Change in forced expiratory volume in 1 second	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
1.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
2 Change in forced vital capacity	2		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
2.1 Up to 3 months (parallel)	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
2.2 Up to 3 months (cross-over)	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
3 Peak expiratory flow	2		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
3.1 Up to 3 months (parallel)	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
3.2 Up to 3 months (cross-over)	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
4 Vmax50% Vital capacity	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
4.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
5 Forced expiratory flow 25-75	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
5.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
6 RV/TLC	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
6.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
7 Six-minute walk test [metres]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
7.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
8 Sputum viscosity	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
8.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable

Comparison 2.   Oral thiols versus placebo

Outcome or subgroup title	No. of studies	No. of participants	Statistical method	Effect size
1 Change in forced expiratory volume in 1 second [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
1.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
2 Peak expiratory flow [L/min]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
2.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
3 Change in peak expiratory flow [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
3.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
4 Change in total lung capacity [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
4.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable

5 Change in flow 75% FVC [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
5.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
6 Change in flow 50% FVC [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
6.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
7 Change in flow 25% FVC [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
7.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
8 Change in forced expiratory volume in 1 sec/vital capacity [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
8.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
9 Change in total gas volume [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
9.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
10 Change in vital capacity [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
10.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
11 Change in residual volume/total lung capacity [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
11.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
12 Antibiotic treatment [weeks]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
12.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable

Comparison 3. Oral thiols versus other oral agents

Outcome or subgroup title	No. of studies	No. of participants	Statistical method	Effect size
1 Change in forced expiratory volume in 1 second [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
1.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
2 Change in peak expiratory flow [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
2.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
3 Change in total lung capacity [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
3.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
4 Change in flow 75% FVC [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
4.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
5 Change in flow 50% FVC [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
5.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable
6 Change in flow 25% FVC [% predicted]	1		Mean Difference (IV, Fixed, 95% CI)	Totals not selected
6.1 Up to 3 months	1		Mean Difference (IV, Fixed, 95% CI)	Not estimable