

BMJ Open Levels of immunoglobulin isotypes in serum and respiratory samples of patients with chronic obstructive pulmonary disease: protocol for a systematic review and meta-analysis

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To cite: Unninayar D, Shea B, Cameron DW, *et al.* Levels of immunoglobulin isotypes in serum and respiratory samples of patients with chronic obstructive pulmonary disease: protocol for a systematic review and meta-analysis. *BMJ Open* 2023;**13**:e064307. doi:10.1136/bmjopen-2022-064307

► Prepublication history and additional supplemental material for this paper are available online. To view these files, please visit the journal online (<http://dx.doi.org/10.1136/bmjopen-2022-064307>).

Received 03 May 2022
Accepted 27 January 2023



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ABSTRACT

Introduction Chronic obstructive pulmonary disease (COPD) is an inflammatory respiratory disorder characterised by the progressive worsening of lung function. Acute exacerbation of COPD (AECOPD) is a leading contributor to patient morbidity, mortality and hospitalisations. The clinical significance of immunoglobulin (Ig) levels in COPD patients is not well established and is in need of further investigation.

Methods and analysis We will conduct a systematic review to describe levels of different Ig isotypes (IgG, IgA and IgM) in various samples (serum, sputum and bronchoalveolar lavage) of patients with COPD. IgE levels in COPD patients have been researched and reviewed extensively and hence will be excluded from this review. IgD levels will also be excluded from the review as there is a paucity of data on IgD levels in COPD patients. The primary outcome of interest in this systematic review is assessing Ig isotype levels in patients with COPD. Secondary outcomes that will be assessed include the differences between Ig isotype levels in COPD patients compared with healthy controls, as well as the relationships between Ig isotype levels and key clinical variables, including COPD severity, incidence of AECOPD and AECOPD severity. Embase and Ovid MEDLINE will be used to search for non-randomised studies published from 1946 to October 2022 that report our prespecified primary and secondary outcomes. As per the Preferred Reporting Items for Systematic Reviews and Meta-Analyses protocol, retrieved studies will undergo a two-phase screening process conducted by two independent reviewers. Prespecified primary and secondary outcomes will be extracted from eligible studies, and descriptive statistics will be used to analyse extracted outcomes. The risk of bias will be assessed using the Risk Of Bias In Non-randomised Studies - of Interventions (ROBINS-I) tool.

Ethics and dissemination Ethics approval is not required as this is a protocol for a systematic review and meta-analysis. Findings will be disseminated through peer-reviewed publications and other formats including conference presentations.

PROSPERO registration number CRD42020192220.

STRENGTHS AND LIMITATIONS OF THIS STUDY

- ⇒ The search strategy for this review has been designed by a medical librarian experienced in systematic reviews, and is not limited by study language, publication format or publication status.
- ⇒ This systematic review will be the first to summarise and analyse the levels of immunoglobulin isotypes in patients with chronic obstructive pulmonary disease, and will offer the highest level of evidence available for each outcome studied.
- ⇒ This review is limited to evidence from clinical studies, as in vitro studies and in vivo animal studies have been excluded.
- ⇒ We anticipate some heterogeneity between study design and results reporting. This will be addressed with sensitivity analyses to assess robustness and increase the credibility of our findings.

INTRODUCTION

Chronic obstructive pulmonary disease (COPD) is a disorder of the lower respiratory system characterised by the progressive worsening of pulmonary function. As the disease progresses, patients often experience an increase in the frequency and/or severity of acute exacerbation of COPD (AECOPD).^{1 2} AECOPD episodes accelerate loss of lung function and contribute significantly to the increase in morbidity and mortality.^{3 4} Therefore, reducing the frequency and severity of AECOPD is essential in improving health-related quality of life.⁵

From a pathophysiological perspective, COPD is characterised by a chronic inflammatory process of the airways. Respiratory infections may trigger AECOPD episodes, which are characterised by an acute worsening of respiratory symptoms, often secondary to increased airway inflammation.⁶ In healthy airways, a delicate balance is maintained between inflammatory and anti-inflammatory

processes to prevent diseases that are infectious or inflammatory in nature.^{7,8} A subset of individuals with humoral immunodeficiency characterised by low immunoglobulin (Ig) levels have recurrent airway infections and inflammation.^{9–12} Therefore, Igs likely play a protective role in preventing inflammatory processes.

Igs are glycoprotein molecules produced by B cells and are an essential component of the immune response to infection.¹³ There are five main Ig isotypes: IgG, IgA, IgM, IgE and IgD. IgG is the predominant isotype found in the body and can be further categorised into IgG1, IgG2, IgG3 and IgG4 subclasses. IgG functions primarily by opsonising bacterial and viral antigens and has also been shown to have an immunosuppressive effect when administered therapeutically for certain diseases.^{14–17} IgA serves a protective role in mucosal secretions in its dimeric form (sIgA), but is also found in the circulation (IgA1 and IgA2).¹⁴ IgM is present in the serum and functions by activating the complement component of the immune system and opsonising antigens for destruction. Relatively low-affinity IgM antibodies, called natural antibodies, have an important role in immunoregulation.^{14,18} IgE is present at the lowest concentration in the serum and is associated with hypersensitivity reactions. Finally, IgD is also found in the circulation but its role in the immune response and immune regulation remains largely unknown.¹⁴ Given the paucity of data on IgD and COPD, IgD will not be included in this systematic review.

A dysregulation in Ig levels in COPD patients has been reported in several studies.^{19–24} While the relationship between serum, sputum and bronchiolar Ig levels and COPD severity, mortality, morbidity and risk of exacerbation has been examined, these studies have yielded heterogeneous results.^{19–25} To date, a clear consensus on the relationship between Ig levels and COPD severity does not exist. Understanding the difference between the systemic and local Ig levels of COPD patients and healthy individuals may further our understanding of COPD pathogenesis and support novel therapeutic approaches for the management of AECOPD. Therefore, the purpose of this systematic review is to evaluate and analyse the current state of evidence on this topic.

Research objectives

We will conduct a systematic review to (a) describe serum, sputum, and bronchoalveolar lavage (BAL) IgG, IgG subclasses, IgA and IgM levels in patients with COPD, (b) determine the differences in serum, sputum and BAL IgG, IgG subclasses, IgA and IgM levels between patients with COPD and healthy controls, (c) evaluate the relationship between Ig levels (all isotypes excluding IgE and IgD) and severity of COPD, incidence rate of AECOPD, severity of AECOPD and mortality, (d) evaluate the correlation among IgG, IgA, IgM measurements from different sources (ie, serum, sputum and BAL).

METHODS AND ANALYSIS

We will follow the Preferred Reporting Items for Systematic Reviews and Meta-Analyses protocol to design our search strategy, conduct our search and report our findings (see online supplemental material 1).²⁶ A copy of the completed checklist and search strategy will be provided as a reference in the published article.

Search strategy

A comprehensive search strategy will be designed with the assistance of a medical librarian and the use of the Preferred Reporting Items for Systematic Review and Meta-Analysis Protocols checklist as guidance.²⁶ Studies will be retrieved using Embase Classic+Embase (1947 to October 2022) and Ovid MEDLINE (1946 to October 2022). The search will be performed using relevant keywords (see online supplemental material 2). In order to optimise generalisability, there will be no restrictions placed on language, publication format or publication status. The reference section of each study will be scrutinised for additional publications that were not included in the search results.

The bibliographic software Covidence will be used to facilitate the management and screening of all articles identified through this process.²⁷

Study screening and inclusion

Two reviewers will perform the screening process independently after completing initial pilot exercises to address any discrepancies in the application of the inclusion and exclusion criteria. The screening process will be conducted in Covidence and will be divided into two phases.²⁷ The first phase will involve the screening of references by study title and abstract. Study titles without an associated abstract will move forward to the full-text screening (second phase) unless it is clear to both reviewers that the study can be excluded. In the second phase, full text will be screened using the inclusion and exclusion criteria. If a decision to include or exclude a study differs between reviewers, Covidence will highlight this study as a conflict.²⁷ On completing each phase of the screening process, the reviewers will document, discuss, and reconcile all conflicts. If the conflict cannot be resolved in the first phase, the study will move forward to the second phase of the screening process. Any discrepancies after the second phase of the screening process will be resolved with the assistance of a third-party reviewer.

Study eligibility criteria

Participants

Studies that evaluated individuals with a confirmed diagnosis of COPD (forced expiratory volume in 1 s (FEV₁)/forced vital capacity <0.7 post-bronchodilator) will be included. Studies that enrolled patients with asthma-COPD overlap or COPD patients with concomitant primary immunodeficiency or receiving Ig replacement therapy will be excluded.

Interventions

Studies that assessed the levels of IgG, IgG subclasses, IgA, IgA subclasses and IgM in the serum, sputum and/or BAL will be included. Studies that limit their analysis to the measurement of free light chain levels or only report levels of IgE or IgD in the serum, sputum or BAL will be excluded.

Comparators

Ig levels of healthy subjects who were included as comparators in studies assessing Ig levels in people with COPD.

Outcomes

Studies that report serum, sputum and BAL Ig levels other than IgE and IgD will be included.

Types of studies

Non-randomised studies including case reports, case series, cross-sectional studies, retrospective analysis and prospective analysis will be included. Review studies and studies that are based solely on in-vitro experiments or animal models will be excluded.

Data extraction

Data will be extracted from eligible studies by two independent reviewers using a predesigned template created on Covidence.²⁷ The data extracted will include information on study identification (authorship, publication year, funding, affiliated centres, journal), methodological approach (type of study, eligibility criteria, sample size), patient characteristics (age, sex, COPD diagnosis, comorbidities, smoking status), Ig levels (serum, sputum and BAL IgG, IgA, IgM; serum, sputum and BAL IgG and IgA subclasses), medication (inhaled corticosteroids, systemic corticosteroids, antimicrobials, bronchodilators) and patient outcomes (see the Data analysis section). Any inconsistencies in data extraction will be recorded and reconciled by a third-party reviewer.

Data analysis

An analysis of the findings and the methodological quality of all included studies will be presented as a narrative synthesis, accompanied by relevant charts and tables. Suitability of the data for meta-analysis will be determined based on the clinical and statistical heterogeneity between the included studies (see the Risk of bias assessment section).

Primary outcome analysis

For primary outcome analysis, we will use descriptive statistics to report the following in COPD patients:

- ▶ Serum IgG, IgG subclasses, IgA, IgA subclasses and IgM levels.
- ▶ Sputum IgG, IgG subclasses, IgA, IgA subclasses, and IgM levels.
- ▶ BAL IgG, IgG subclasses, IgA, IgA subclasses and IgM levels.

This continuous data will be summarised as means and mean differences of Ig levels, with 95% CIs. If

meta-analysis is possible, a random effects model will be used in order to account for possible unobserved heterogeneity between studies.

Secondary outcome analysis

For secondary outcome analysis, we will use descriptive statistics to compare Ig levels between healthy controls and COPD patients. We will also study the association between levels of Ig and COPD severity according to the GOLD criteria,²⁸ severity according to FEV₁ score (very severe if <30%, severe if 30%–49%, moderate if 50%–79%, mild if ≥80%), incidence rate of AECOPD per annum, severity of AECOPD (severe if episode triggered hospital admission, moderate if episode triggered in-patient visit to the emergency department or out-patient use of corticosteroids or prescribed oral antibiotics, mild if episode does not meet moderate or severe criteria), COPD status (stable COPD vs during AECOPD), timing of Ig measurement in relation to recent or current exacerbations, use of inhaled or systemic corticosteroids during exacerbations, and mortality rates.²⁸ We will evaluate the correlation among Ig isotypes from different sources (ie, serum, sputum and BAL measurements).

The secondary outcome analyses will be performed for the levels of each class, subclass and variant of serum, sputum and BAL Ig. If meta-analysis is possible, a random effects model will be applied to account for variability in measurement methods between studies.

Risk of bias assessment

The Risk Of Bias In Non-randomised Studies - of Interventions (ROBINS-I) tool will be used to assess the risk of bias in the non-randomised studies included in the review.²⁹ This risk of bias assessment will be performed by two independent reviewers, and any discrepancies in assessment will be resolved by a third party. As per the ROBINS-I criteria, the studies will be evaluated in the context of seven domains, namely baseline confounding, selection of participants, classification of interventions, deviation from intervention, missing data, measurement of outcomes and selection of the reported result.²⁹ The results from these assessments will be disclosed in the published report. If the number of included studies is greater than or equal to 10, a funnel plot will also be designed to assess the potential existence of publication bias.³⁰ In order to determine the suitability of the data for meta-analysis, we will assess the clinical and statistical heterogeneity between the included studies. Clinical heterogeneity will be evaluated through subgroup analyses, while statistical heterogeneity will be evaluated using the I² statistic and Cochran Q/ χ^2 test.^{31 32} If meta-analysis is possible, we will use a random effects model and perform sensitivity analyses on the data to increase the credibility of our findings.

Subgroup analyses

Preplanned subgroup analyses will be conducted to examine clinical heterogeneity between studies and

will include the following: (a) COPD severity by GOLD criteria; (b) COPD severity by FEV₁ score; (c) incidence of AECOPD; (d) severity of AECOPD; (e) COPD status (stable COPD vs during AECOPD).

Sensitivity analyses

Preplanned sensitivity analyses will be conducted in order to determine the robustness of our assessment and increase the credibility of our findings. The analyses will include the following: (a) studies with low versus high/undetermined risk of bias as evaluated by ROBINS-I; (b) studies where COPD patients have no comorbidities vs studies where COPD patients have comorbidities or their comorbid status is unknown.

Patient and public involvement

This is a protocol for a systematic review and meta-analysis, and therefore the results from this study will be based solely on previously published data. Patients and the public will not be involved in the design, conduct, reporting, or dissemination plans of this research.

DISCUSSION

To our knowledge, this will be the first systematic review to evaluate Ig isotype levels in COPD patients, compare their Ig isotype levels to those of healthy controls, and analyse the relationship between Ig isotype levels, COPD disease severity and AECOPD severity.

We anticipate some limitations in the implementation of this protocol. The measurement of patient Ig levels can be accomplished using several different techniques, and hence we expect some baseline variability in Ig measurements between study centres. We also expect differences in study design between publications. However, this variability will be accounted for in the data analysis. Further, differences in the type and dose of medication, alpha-1 anti-trypsin status, presence of comorbid conditions and time differences in which Ig levels were measured across studies may influence Ig levels. We will conduct multi-variate statistical analyses to recognise and minimise these limitations.

Despite these challenges, our systematic review will fill a knowledge gap in the literature and provide insight into the association between Ig levels and COPD severity as well as AECOPD frequency and severity. The results from this systematic review may serve to enhance our understanding of the humoral immunity relevant to COPD pathophysiology, and may identify Ig levels as a potential 'treatable trait' in this population.

Acknowledgements We would like to thank Risa Shorr, Librarian and Information Specialist at the Ottawa Hospital Research Institute, Ottawa, Ontario, for developing the search strategy and conducting the literature search for this review.

Contributors JC is the guarantor. DU drafted the protocol. JC and BS critically reviewed and revised the protocol. JC and DU will perform the study screening and data extraction. JC will reconcile any discrepancies in all steps of data collection. JC, BS and DU will conduct data analysis and prepare the manuscript. JC, DWC and DU will be consulted for the interpretation of results and the preparation of the manuscript. All authors conceived and designed this review as well as read and approved the final manuscript.

Funding The authors have not declared a specific grant for this research from any funding agency in the public, commercial or not-for-profit sectors.

Competing interests The authors declare that they have no competing interests.

Patient and public involvement Patients and/or the public were not involved in the design, or conduct, or reporting, or dissemination plans of this research.

Patient consent for publication Not applicable.

Provenance and peer review Not commissioned; externally peer reviewed.

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