ERJ open research
THE BEST IN OPEN ACCESS BASIC, TRANSLATIONAL & CLINICAL RESPIRATORY RESEARCH

Early View

Original article

MULTI-PHACET - MULTIdimensional clinical phenotyping of hospitalised acute COPD ExacerbaTions

Martin I. MacDonald, Christian R. Osadnik, Lauren Bulfin, Elizabeth Leahy, Paul Leong, Eskandarain Shafuddin, Kais Hamza, Paul T. King, Philip G. Bardin

Please cite this article as: MacDonald MI, Osadnik CR, Bulfin L, *et al*. MULTI-PHACET - MULTIdimensional clinical phenotyping of hospitalised acute COPD ExacerbaTions. *ERJ Open Res* 2021; in press (https://doi.org/10.1183/23120541.00198-2021).

MULTI-PHACET - MULTIdimensional clinical Phenotyping of Hospitalised Acute COPD ExacerbaTions

MacDonald, Martin. I. (PhD) [1,2,3], Osadnik, Christian.R. (PhD)[1,4], Bulfin, Lauren.[1], Leahy, Elizabeth[1], Leong, Paul. (MBBS), Shafuddin, Eskandarain. (PhD) [1,2], Hamza, Kais. (PhD)[5], King, Paul .T. (PhD)[1,2,3], Bardin, Philip .G. (PhD)[1,2,3]

[1]Monash Lung and Sleep, Monash Health, Melbourne Australia; [2]Monash University, Department of Medicine, Melbourne Australia; [3]Hudson Institute, Melbourne, Australia; [4]Monash University, Department of Physiotherapy, Melbourne Australia; [5]Monash University, Statistical Services, Melbourne Australia.

Conception and design: MIM, PTK, PGB;

Data acquisition: MIM, CO, LB, EL

Statistical analysis and interpretation: MIM, CO, PL, KH, PTK, PGB;

Drafting the manuscript for important intellectual content: MIM, CO, PL, ES, PTK, PGB

Final review of manuscript: all authors.

Corresponding author:

Dr. Martin I MacDonald

Monash Lung and Sleep, Monash Medical Centre,

246 Clayton Road, Clayton 3168,

Melbourne, Victoria,

Australia

E-mail: macdonald.martin@gmail.com

Word count text = 2988 (without references, tables or figure legends)

Take home message

Hospitalised AECOPDs present as complex multidimensional clinical phenotypes, often comprising multiple distinct aetiologies. Profiling AECOPDs according to their multifactorial aetiological components has important prognostic and therapeutic implications.

ABSTRACT (WORD COUNT: 250)

Background

The generic term 'exacerbation' does not reflect the heterogeneity of acute exacerbations of COPD (AECOPD). We utilised a novel algorithmic strategy to profile exacerbation phenotypes based on underlying aetiologies.

Methods

Patients hospitalized for AECOPD (n=146) were investigated for aetiological contributors summarised in a mnemonic acronym **ABCDEFGX** (**A**=Airway virus, **B**=Bacterial, **C**=Coinfection, **D**=Depression/anxiety, **E**=Eosinophils, **F**=Failure (cardiac), **G**=General environment, **X**=Unknown). Results from clinical investigations were combined to construct AECOPD phenotypes. Relationships to clinical outcomes were examined for both composite phenotypes and their specific aetiological components. Aetiologies identified at exacerbation were reassessed at outpatient follow-up.

Results

Hospitalised AECOPDs were remarkably diverse, with 26 distinct phenotypes identified. Multiple aetiologies were common (70%) and unidentifiable aetiology rare (4.1%). If viruses were detected (29.5%), patients had longer hospitalisation (7.7±5.6 vs 6.0±3.9 days, p=0.03) despite fewer 'frequent exacerbators' (9.3% vs 37%, p=0.001) and lower mortality at 1 year (p=0.03). If bacterial infection was found (40.4%), patients were commonly 'frequent exacerbators' (44% vs 18.4%, p=0.001). Eosinophilic exacerbations (28%) were associated with lower pH (7.32±0.06 vs 7.36±0.09, p=0.04), higher $PvCO_2$ (53.7±10.5 vs 48.8±12.8, p=0.04), greater NIV usage (34.1% vs 18.1%) but shorter hospitalisation (4[3-5] vs 6[4-9] days, p<0.001) and lower infection rates (41.4% vs 80.9%, p<0.0001). Cardiac dysfunction and severe anxiety/depression were common in both infective and non-infective exacerbations. Characteristics identified at exacerbation often persisted after recovery.

Conclusions

Hospitalised AECOPDs have numerous causes, often in combination, that converge in complex, multi-faceted phenotypes. Clinically important differences in outcomes suggest that a phenotyping strategy based on aetiologies can enhance AECOPD management.

INTRODUCTION

Chronic obstructive pulmonary disease **(**COPD) is a complex and heterogeneous disease. During acute exacerbations of COPD (AECOPDs), additional complexity ensues given diverse exacerbation aetiologies and comorbidities. Stable COPD is increasingly recognised as encompassing diverse disease phenotypes(1) however the term "acute exacerbation of COPD" remains generic with a nonspecific definition(2). The complexity and heterogeneity of AECOPDs is not yet reflected in either clinical practice or clinical research.

We have previously proposed individualised phenotyping of hospitalised AECOPDs according to aetiological contributors(3, 4). Whilst studies have explored phenotyping strategies based on simple microbiological classification(5, 6), a comprehensive personalised approach encompassing additional factors such as cardiac disease, mood disorders or inadequate social support, has not been reported.

We hypothesised that an algorithmic approach using simple investigations would elucidate the multifactorial complexity of AECOPD aetiology, with prognostic and therapeutic implications.

METHODS

Study population

Patients hospitalised for AECOPD were recruited to a prospective observational study approved by our hospital's Human Research Ethics Committee (HREC13134A). Written informed consent was obtained.

Inclusion required a post-bronchodilator forced expiratory ratio <0.7 verified by spirometry performed when clinically stable(7). Exclusion criteria included overt left ventricular failure or acute myocardial infarction. Due to the need for informed consent, patients mechanically ventilated at initial presentation were excluded. Sufficient cognitive capacity to complete questionnaires was required. Infiltrates on chest X-ray (CXR) were permitted.

Clinical outcomes recorded included rates of non-invasive ventilation (NIV), mechanical ventilation, inpatient mortality, length of hospital stay, readmissions and survival for 12 months following hospital discharge. The research team did not influence clinical care, which was at the discretion of the attending physicians. A follow-up assessment when clinically stable was offered to all patients. Hospitalisations in the 12-month period after hospital discharge were identified by review of electronic health records. Survival at 12 months post hospital discharge was determined by review of electronic health records and patient phone calls.

Study design

Patient recruitment and participation are shown in Figure 1.

We assessed attributable causes of hospitalised AECOPDs using the mnemonic acronym **ABCDEFGX: A - Airway virus; B - Bacterial; C - Coinfection; D - Depression/anxiety; E - Eosinophils; F - Failure (cardiac); G - General environment; X - unknown** (3). Since routine CT pulmonary angiography may not be justified in AECOPD, we revised our originally published acronym(3) by substituting 'E - Eosinophils' in place of the former 'E - Embolism (pulmonary)'. Patients were assigned a final composite phenotype by combining all aetiological factors that were identified (e.g. bacterial infection (**B**) and severe depression/anxiety symptoms (**D**) = phenotype '**BD**').

Phenotyping strategy

Demographic variables, comorbidities, exacerbation history and pharmacotherapy were obtained from patient interviews and hospital case records. Study investigators administered the COPD Assessment Tool (CAT)(8), Hospital Anxiety and Depression Scale (HADS)(9) and Medical Research Council Dyspnoea (MRCD) scale(10). Results from nasopharyngeal virus PCR (146/146), spontaneously expectorated sputum culture (136/146), C-reactive protein (CRP) and fever (≥38°C) (both 146/146) were used to identify aetiologies '**A**' (Airway virus), '**B**' (Bacteria) or '**C**' (Co-infection) (Figure 2). Inevitably, many AECOPDs have clinical features of infection without identification of a specific microorganism. We assigned putative '**B**' (Bacterial) aetiology if fever or CRP≥20mg/dL(11) was recorded and virus negative. Identification of '**D**' (Depression/anxiety) was based on HADS scores at hospital admission (HADS A/D ≥15 or combined HADS Total ≥27, successfully completed for 134/146). Although not specifically validated for AECOPD hospitalisations, we chose high threshold values for HADS previously shown to have 95% specificity for verified anxiety/depression in an inpatient population(12). Eosinophils were measured on the first Full Blood Count (FBC) in 146/146 with "**E**" assigned when eosinophils >2% total white cell count(13). Investigation for pulmonary embolism was at the discretion of the treating team and not part of the algorithmic investigational approach. Cardiac biomarker analysis was performed on blood taken at initial hospital presentation where sufficient serum was available (119/146). We identified cardiac dysfunction '**F**' (Failure) when high-sensitivity troponin I (hs-TnI and/or N-terminal pro-brain natriuretic peptide (NT-proBNP)(14) were above age and gender adjusted upper limits of normal (ULN)(15, 16). Acute disruption to the patient's physical, social or therapeutic environment was assessed in 146/146 and aetiology '**G**' (General environment) assigned if deemed causative to hospitalization. Exacerbations with no aetiological factor identifiable were categorized '**X**' (unknown).

Analysis

Comparisons between groups were made employing unpaired t-tests and one-way analysis of variance (ANOVA) for normally distributed data or Mann-Whitney and Kruskal-Wallis testing for non-parametric data. Chi-square analyses were used for categorical data. Blood and questionnaire results from acute versus stable disease state were analysed via Pearson correlation coefficients. Time-to-event survival analyses were conducted using Kaplan-Meier methods and log-rank tests. Data are presented as number (percentage), mean±standard deviation (SD) or median [interquartile range, IQR], where appropriate. Statistical significance was accepted at $p<0.05$. Analyses were conducted on Stata MP 14.1 (*Statacorp, Texas, USA*).

RESULTS

Study cohort, aetiologies and phenotypes

Overall 169 AECOPD admissions were enrolled, with 146 patients included (Figure 1). Twenty-three patients were excluded, chiefly because they failed to meet spirometric criteria for a diagnosis of COPD or lacked viral swab results. Demographics, comorbidities and pharmacotherapy are shown (Table 1).

Table 1. Baseline characteristics of 146 patients enrolled during AECOPD

Demographics, n (%), mean±SD, median[IQR]		Comorbidities, n (%)		Medications, n (%)	
Age	71.8±10.4	Bronchiectasis	16 (10.3)	LAMA	133 (85.8)
Male	97 (62.6)	OSA	14 (9.0)	LABA	129 (83.2)
BMI	24.8±6.5	Hypertension	71 (45.8)	ICS	126 (81.3)
FEV_1 (L)	1.1±0.5	AF/flutter	19 (12.3)	OCS¤	13 (8.4)
FEV_1 (%)	45.2±18.6	IHD	43 (27.7)	Antibiotic¤	5 (3.2)
TLCO	38.3±16.2	Cardiac failure	32 (20.6)	Antiplatelet	55 (35.5)
LTOT	19 (12)	CVD	15 (9.7)	Anticoagulant	18 (11.6)
MRC-D	4 [3-5]	Diabetes	29 (18.7)	β-blocker	19 (12.3)
Current smoker	48 (31)	Malignancy*	15 (9.7)	Ivabradine	4 (2.6)
Former smoker	117 (69)	Renal failure°	3 (1.9)	Ca2RA	13 (8.2)
Pack years	44±26	Anxiety	36 (23.2)	ACE-I/ARB	52 (33.5)
AECOPDs in prev. year		Depression	35 (22.6)	Statin	57 (36.8)
Hospital	1.5±2.3	Alcohol misuse	12 (7.7)	Loop diuretic	33 (21.3)
Community	1.7±2.5	Substance misuse	2 (1.3)	Benzodiazepine¤	20 (12.6)
Frequent exacerbators (≥ 2 AECOPD hospitalisations in previous. year)	46 (29.7)	Other psychiatric disorder**	3 (1.9)	Antidepressant/ Antipsychotic	39 (24.5)

Data shown as mean/SD and n (%). BMI = body mass index (kg/m^2), FEV_1 = Forced expiratory volume in 1 second, TLCO = gas transfer, LTOT = long term oxygen therapy, mMRC-D = modified Medical Research Council Dyspnoea score, OSA = obstructive sleep apnoea, AF = atrial fibrillation, IHD = ischaemic heart disease, CVD = cerebrovascular disease, LAMA = long acting muscarinic antagonist, LABA = long acting beta-agonist, ICS = inhaled corticosteroid, OCS = oral corticosteroid, ¤ = maintenance, ACE-I/ARB = Angiotensin converting enzyme inhibitor/Angiotensin receptor blocker, Ca2RA = calcium antagonist, *receiving treatment or palliation, °egfr<30ml/min, **bipolar affective disorder, schizophrenia, post-traumatic stress disorder, ¤(excluding nocte temazepam)

A flow diagram for assigning aetiological components of phenotypes is shown (Figure 2). The process identified a large number of distinct phenotypes (total 26) based on various combinations of 6 underlying aetiologies (Figure 3). Phenotypes consisting of a single aetiology were noted in a minority (38/146, 26.0%). Two (74/146 patients, 50.7%) or three aetiologies (27 patients, 18.5%) were common and no identifiable aetiology was rare (6/146, 4.1%).

Outcomes associated with phenotypes

We first evaluated whether composite phenotypes (rather than individual aetiologies) were associated with clinical outcomes. The large number of phenotypes and resultant small populations in each phenotype precluded meaningful statistical analyses but descriptive summary data are shown for the 10 most common phenotypes (representing 72.6% of study cohort, *Supplementary Table S1*). A higher cumulative number of aetiologies did not show association with clinical outcomes.

Outcomes associated with individual aetiologies

Individual aetiologies (rather than complex phenotypes) are likely to be more informative in a smaller cohort. We therefore compared exacerbations *with* versus *without* individual aetiological components.

Patients with virus infection (43/146, 29.5%) were less commonly frequent hospitalised exacerbators (9.3% vs 37%, p=0.001), had lower baseline MRCD scores (3[2-4] vs 4[3-5], p=0.0007) and less domiciliary oxygen use (7% vs 32%, p=0.001). Despite this favourable profile they had longer hospitalisation (7.7±5.6 vs 6.0±3.9, p=0.02), even after exclusion of those with bacterial co-infection (n=8, 7.9±6.1 vs 6.0±3.9, p=0.03). Their mortality at 12 months post hospital discharge was lower (2/43 (4.7%) vs 20/103 (19.4%), p=0.02). Survival curves over 12 months post-discharge are shown for virus (including coinfection) (n=43) vs bacterial only (n=59) vs non-infective (n=44) AECOPDs (Figure 4, p=0.03).

Bacterial aetiology was assigned in 59/146 patients (40.4%), of whom 24/59 patients (40.7%) had positive sputum culture (Bi, Figure 2). These patients were more likely to be frequent hospitalised exacerbators (44% vs 18.4%, p=0.001), had higher baseline MRCD scores (5[4-5] vs 4[3-5], p=0.002) and higher prevalence of diagnosed ischaemic heart disease (53.8% v 35.6%, p=0.045) and cardiac failure (57.1% vs 36.4%, p=0.045).

Those with combined virus and bacterial infection (coinfection) had higher WCC (14.7±3.7 vs 11.4±4.4, p=0.008), neutrophils (11.7±2.6 vs 8.6±3.4, p=0.01) and CRP (112[65-167] vs 18[4.6-69], p=0.004). Specific viruses and bacteria detected are shown (*Supplementary Table S2*).

HADS scores were higher in those with diagnosed psychiatric comorbidity (20.4±8.7 vs 14.6±8.1, p=0.0001, *Supplementary Table S3).* HADS scores were above the threshold to assign aetiology '**D**' in 33/136 patients (24.3%), of whom only 14/33 (42.4%) were taking antidepressant or anxiolytic medication. Patients featuring aetiology (**D**) reported higher ("worse") total CAT scores (34[30-37] vs 29[24-33], p=0.0001). Their responses to CAT items 1-5 (physical symptoms) did not differ whereas CAT item responses 6 ("confidence" 4.3±1.2 v 3.3±1.9, p=0.026), 7 ("sleep" 3.9±1.6 vs 2.9±1.6, p=0.015) and 8 ("energy" 3.9±1.6 vs 2.9±1.6, p=0.005) were higher. There were no significant differences in clinical outcomes based on aetiology '**D**'.

Blood eosinophils >2% (aetiology '**E**') was present in 41/146 (28%). Prehospital oral corticosteroid had been prescribed in 9.8% of those >2% and 30.5% with ≤2% eosinophils. Inhaled corticosteroids prescription was similar (75% vs 85%, p=0.34). Infection was less common in AECOPD with eosinophils >2% (41.4% vs 80.9%, p<0.0001). "Eosinophilic exacerbations" were associated with lower blood pH (7.32±0.06 vs 7.36±0.09, p=0.04), higher PvCO2 (53.7±10.5 vs 48.8±12.8, p=0.04) and NIV usage (34.1% vs 18.1%). Despite this, patients with eosinophils >2% had a shorter hospital stay (4[3-5] vs 6[4-9] days, p<0.001). Systemic corticosteroid prescription during hospitalisation was similar in the >/≤2% eosinophil groups (97.6% vs 97.1%).

An elevated cardiac biomarker (either/both, aetiology "**F**') was noted in 85/119 patients (71.2%), NT-proBNP in 83/119 (69.7%) and hs-TnI in 32/119 patients (26.9%). Patients with established diagnoses of cardiovascular disease tended to have higher levels (*Supplementary Table S4*), with NT-proBNP significantly higher in those with a past history of cardiac failure (618ng/L [18.5-2016] versus 321ng/L [117-693], p=0.03). Among patients with an elevated cardiac biomarker, cardiac medication use was notably low: antiplatelets (34.1%), anticoagulants (16.5%), β-blockers (17.7%), ACE-I/ARB (38.8%), statins (32.9%) and loop diuretics (21.1%). Aetiology '**F**' was not associated with significant differences in short term clinical outcomes, survival at 12 months or readmission rates. Using a threshold considered

more definitive for cardiac failure (NT-proBNP>900ng/L) was associated with longer hospital stay (7[5-10] vs 5[4-7] days, p=0.018).

General environmental factors contributing to hospitalisation (aetiology '**G**') were rarely identified (3/146 patients, 2.1%). Factors included running out of medication and failure of home air conditioning during an extreme heatwave.

No aetiology was identified in 6/146 cases (aetiology '**X**', 4.1%).

AECOPDs with versus without evidence of infection

Finally, we compared AECOPDs associated with infection (69.9%) versus no infection (30.1%). No differences in demographics, comorbidities or clinical outcomes were found (Tables 2 and 3). Total CAT scores were similar (30[26-33] vs 30[25-34], p=0.98) with only CAT item 2 ("phlegm") differing in infective exacerbations (4[2-5] vs 3[1-3], p=0.01). Severe anxiety/depression symptoms (23.7% vs 24.5%, p=0.92), hs-TnI (9 [5-32] vs 8 [5-20], p=0.37) and NT-proBNP 395 [164-1221] vs 263 [152-853], p=0.45) did not differ. Non-infective exacerbations featured higher blood eosinophil counts (0.25/uL [0.08-0.46] vs 0.04/uL [0.0-0.14], p<0.001). This observation persisted after excluding patients who had received pre-hospital oral corticosteroids (0.28/uL [0.11-0.46] vs 0.05/uL [0.0-0.2], p<0.001).

Table 2. Characteristics of patients with and without evidence of infection

	Infection	No-infection	p
N (%)	102 (69.9)	44 (30.1)	-
Age	72.3±10.3	69.8±10.7	0.26
Male	65 (63.7)	26 (59.1)	0.6
Freq. exacerbator (hospital)	30 (29.4)	12 (27.3)	0.79
Current smoker	32 (31.4)	15 (34.1)	0.75
Pack year	46.7±28.5	38.7±21.6	0.1
BMI (kg/m2)	25.1±5.8	25.1±5.3	0.97
FEV_1 (L)	1.21±0.49	1.10±0.67	0.42
FEV_1 (% predicted)	51.2±18.0	44.0±20.1	0.12
TLCO (% predicted)	37.3±14.8	42.7±19.7	0.1
MRC-D	4 [3-5]	4 [3-5]	0.81
Days since symptom onset	5 [3-7]	4 [2-14]	0.94
Prior contact with HCP	68 (66.7)	24 (54.5)	0.17
Pre-hospital antibiotics	51 (50)	16 (36.4)	0.13
Pre-hospital OCS	30 (29.4)	6 (13.6)	0.042
CAT total	30 [25-34]	30 [26-33]	0.98
HADS total	16 [10-22]	18.5 [9-24]	0.53
BAP-65 Class	3 [2-5]	2[2-3]	0.13
CXR infiltrate	30.4	20.9	0.24
Fever (≥38°C)	23.5	0	<0.001
WCC	12.2±4.8	10.0±3.0	0.006
Neutrophils	9.5±4.3	7.0±2.4	<0.001
Eosinophils	0.04 [0.0-0.14]	0.25 [0.08-0.46]	<0.001
CRP (mg/dL)	53 [18.7-117]	4 [1.7-7.0]	<0.001
pH	7.36±0.08	7.33±0.08	0.04
P_vCO_2 (mmHg)	48.1±11.2	54.5±13.8	0.006
Bicarbonate (mmol/L)	27.5±4.2	29.3±5.7	0.13
Base excess	2.7±3.8	4.1±5.2	0.11
NT-proBNP (ng/L)	395 [164-1221]	263 [152-853]	0.45
Hs-TnI (ng/L)	9 [5-32]	8 [5-20]	0.38

Table 3. Management and clinical outcomes of patients with and without evidence of infection

	Infective	Non-infective	p
Antibiotics (inpatient), n (%)	102 (100)	40 (90.9)	0.002
Systemic CS (inpatient), n (%)	100 (98.0)	42 (95.5)	0.38
NIV, n (%)	8 (7.8)	10 (22.7)	0.01
HDU/ICU, n (%)	27 (26.4)	18 (40.1)	0.08
Mechanical ventilation, n (%)	4 (3.9)	1 (2.5)	0.62
Length of stay, median [IQR]	5[4-8]	5[3-8]	0.2
Mortality at 6 months, n (%)	10 (9.8)	5 (11.4)	0.89
Mortality at 12 months, n (%)	14 (13.7)	8 (18.2)	0.49
Readmitted within 12 months	40 (39.2)	22 (50.0)	0.39
Days to readmission	44.5[18-195]	78[34-246]	0.39

Repeat evaluation at stable outpatient review

Outpatient review was attended by 68/146 patients (46.6%) at a median of 63[59-98] days. Those who did versus did not attend follow up showed no differences in demographics or spirometry with the only difference in comorbidities being less diagnosed anxiety disorder, 14.9% vs 29.9%, p=0.03, *Supplementary Table 5*). At outpatient review, there was no difference in CRP (3.7[1.3-8] vs 2.9[1.4-5.5], p=0.69), WCC (9.2±2. vs 9.4±2.5, p=0.74) or neutrophils (6.4±2.7 vs 6.3±2.2, p=0.89) between those who had experienced infective versus non-infective exacerbations, including reanalysis based on individual infective exacerbation aetiologies (**A**, **B** or **C**).

In contrast, HADS scores at exacerbation and recovery were correlated (r=0.56, p<0.0001). Patients assigned aetiology '**D'** at exacerbation had significantly higher HADS scores at follow up (22[13-28] vs 9[4-15], p=0.004).

Blood eosinophils at exacerbation correlated with eosinophil counts at recovery (r=0.54, p<0.0001). Patients with eosinophils >2% at exacerbation (aetiology '**E**') had significantly higher blood eosinophils at recovery (0.3[0.2-0.6] vs 0.11[0.02-0.28], p=0.0003).

Correlations between exacerbation and recovery measurements were significant for NT-proBNP (r=0.39, p=0.004) but not for hs-TnI (r=0.23, p=0.14). Patients who were aetiology "F" were far more likely to have an elevated cardiac biomarker at recovery (70.6% vs 5.9%, p<0.0001) with both hs-TnI (5 [4-9] vs 3.5 [1.5-5], p=0.01) and NT-proBNP (269 [151-692] vs 67 [25-108], p<0.0001) higher at follow up.

DISCUSSION

The current study demonstrates that hospitalised AECOPDs comprise a remarkably heterogenous group of events, often featuring multiple distinct aetiological contributors. This heterogeneity is a barrier to progress in the field since interventions targeting a specific aetiology or pathology may not show benefit if applied to an unselected group. Focussing attention on exacerbation aetiology and constructing AECOPD phenotypes is a logical approach and may be the most suitable prospective strategy to identify patients eligible for targeted interventions.

AECOPD phenotyping will only gain traction if it can ultimately lead to individualisation of treatment decisions. Aetiologies that are prevalent and responsive to treatment are therefore the most important to target. We based our phenotyping strategy on examining six key putative aetiological factors (virus infection, bacterial infection, depression/anxiety, eosinophil-associated inflammation, cardiac dysfunction, environmental factors). This strategy identified many combinations (26 distinct phenotypes within a cohort of 146 patients) with only around a quarter of AECOPDs associated with a single aetiology. Clearly each AECOPD event may have multiple aetiological "ingredients", each with distinct implications for individualised management and prognosis. The current study provides proof-of-concept evidence for a practical phenotyping strategy and demonstrates that using a relatively small number of commonly available investigations makes it possible to unravel some of the complexity of AECOPD.

Identifying viral infection appears to have prognostic implications and multiplex virus PCR will likely become standard of care for hospitalised AECOPD due to the SARS-CoV-2 pandemic. Our virus detection rate (30%) was consistent with previous research(17-19) Despite more prolonged hospitalisation, a history of frequent severe exacerbations were less common and 12 month survival was better in the viral group. It may be that virus infections are sporadic events whereas AECOPD linked to bacterial infection or high blood eosinophils exacerbations reflect a more 'committed' phenotype(6).

Given the limitations of sputum culture and frequent prehospital antibiotic use (45.2% overall in our cohort) we assigned bacterial aetiology to exacerbations where infection was evident but virus PCR testing was negative. A more precise methodology is difficult within the limitations of routine clinical investigations. Broad-range 16S rDNA PCR of sputum samples would enhance the sensitivity for detection of bacteria on sputum but is not routinely

available and it's clinical interpretation remains uncertain. We chose a sensitive CRP threshold of >20mg/dL previously suggested as optimal threshold to identify bacteria in sputum at AECOPD(20). Putative bacterial exacerbations were associated with frequent hospitalisation, comorbid cardiac disease and reduced survival at one year. The frequent identification of *Pseudomonas aeruginosa* on sputum culture (*Supplementary Table S2*) may reflect advanced structural lung disease and a propensity to recurrent bacterial infection with airway colonisation. Reduced survival has previously been associated with lung dysbiosis identified on sputum culture at the time of hospitalised exacerbation(21).

Secondary bacterial infection is known to be a frequent sequel of virus infection in AECOPD(22) and our low prevalence of confirmed coinfection is likely an underestimate reflecting prior antibiotic use, limitations of sputum culture and phenotyping based on admission samples only. In keeping with previous studies we found the highest inflammatory markers during co-infections(23).

The impact of non-infective aetiologies to AECOPD has been less extensively studied. Anxiety and depression have been associated with increased hospitalization rates, longer hospitalisation and increased mortality in COPD(24, 25). In our study severe symptoms of anxiety and depression were common and often untreated. Importantly, HADS scores at AECOPD and recovery were strongly correlated. Future studies could explore the role for identification of psychological morbidity and initiation of appropriate interventions prior to hospital discharge.

Blood eosinophils appear to identify an important AECOPD phenotype. Infection was less common with higher eosinophils. Whilst patients with eosinophilic exacerbations had lower blood pH, higher PvCO2 and greater need for NIV they had a shorter hospital length of stay, a finding that may reflect corticosteroid responsiveness(26). Eosinophil counts were still higher after recovery suggesting association between the exacerbation and 'stable' phenotype. Given the key benefit of anti-IL5 therapies is reduction of exacerbations, patients hospitalised with an eosinophilic exacerbation may be the ideal candidates for future trials of anti-IL5 therapies in COPD.

Finally, we observed biochemical evidence of acute cardiac dysfunction in a majority of AECOPDs(27-30). The higher levels observed in patients with established cardiovascular disease suggests cardiac biomarkers reflect underlying cardiovascular health. At the same time, cardiac biomarkers were often high even amongst those without an established

diagnosis of cardiovascular disease. This suggests that cardiac disease may be a crucial underdiagnosed 'treatable trait'(31) which can be fortuitously detected during AECOPD. Delineation of the multitude of cardiac pathologies identifiable in a hospitalised AECOPD population was beyond the scope of this paper. While the relationship between an elevated cardiac biomarker at exacerbation and the likelihood of an identifiable treatable cardiac comorbidity requires further study, the low prescription rates of cardiac therapies observed in our cohort suggests a potential need for increased recognition and treatment of cardiovascular pathology in COPD.

Our study has a number of limitations. Our sample size was not adequate to examine associations between clusters of aetiologies (phenotypes) and pertinent outcomes. This will likely require very large scale multi-centre studies of exacerbation characteristics which have been identified as a priority for future AECOPD research(32, 33). The very low inpatient mortality (1/146, 0.7%) may reflect exclusion of patients mechanically ventilated at the time of admission and the requirement for adequate cognition to complete questionnaires. We employed routine clinical investigations to define phenotypes which limits the precision and reliability of diagnosing bacterial infection. However, this strategy was intentionally applicable to 'real-life' practice. Virus and bacterial detection at AECOPD may potentially be 'false positives' reflecting colonization, and even virus PCR may give 'false negatives'(11). Pre-hospital antibiotic (45.2%) and/or oral corticosteroid (23.2%) use were common and may have influenced phenotypes. We phenotyped AECOPDs only once at hospital admission but 'evolution' of AECOPD phenotypes over the course of an exacerbation is an area for future study. Finally, other important AECOPD aetiologies may not have been captured in our methodology. Whilst our strategy focussed on treatable aetiological components of the acute hospitalisation episode, phenotyping the chronic disease state (e.g. emphysema predominant) is also key to individualisation of care. Understanding relationships between the chronic disease phenotype and the acute exacerbation phenotype is an area for future research.

In conclusion, better prevention and management of AECOPD will be challenging since there are numerous causes, often in combination, that converge as complex, multi-faceted phenotypes. Identifying the individual contributory aetiologies is feasible and relates to important clinical outcomes. Large prospective studies employing phenotyping can enhance understanding of disease mechanisms and ultimately drive 'personalised medicine' in AECOPD.

Figure 1. CONSORT diagram of patient recruitment and participation.

Figure 2. Aetiologies identified based on ABCDEFGX acronym in patients hospitalised with AECOPD

Figure 3. Distribution and combinations of identified phenotypes among 146 hospitalised AECOPDs.

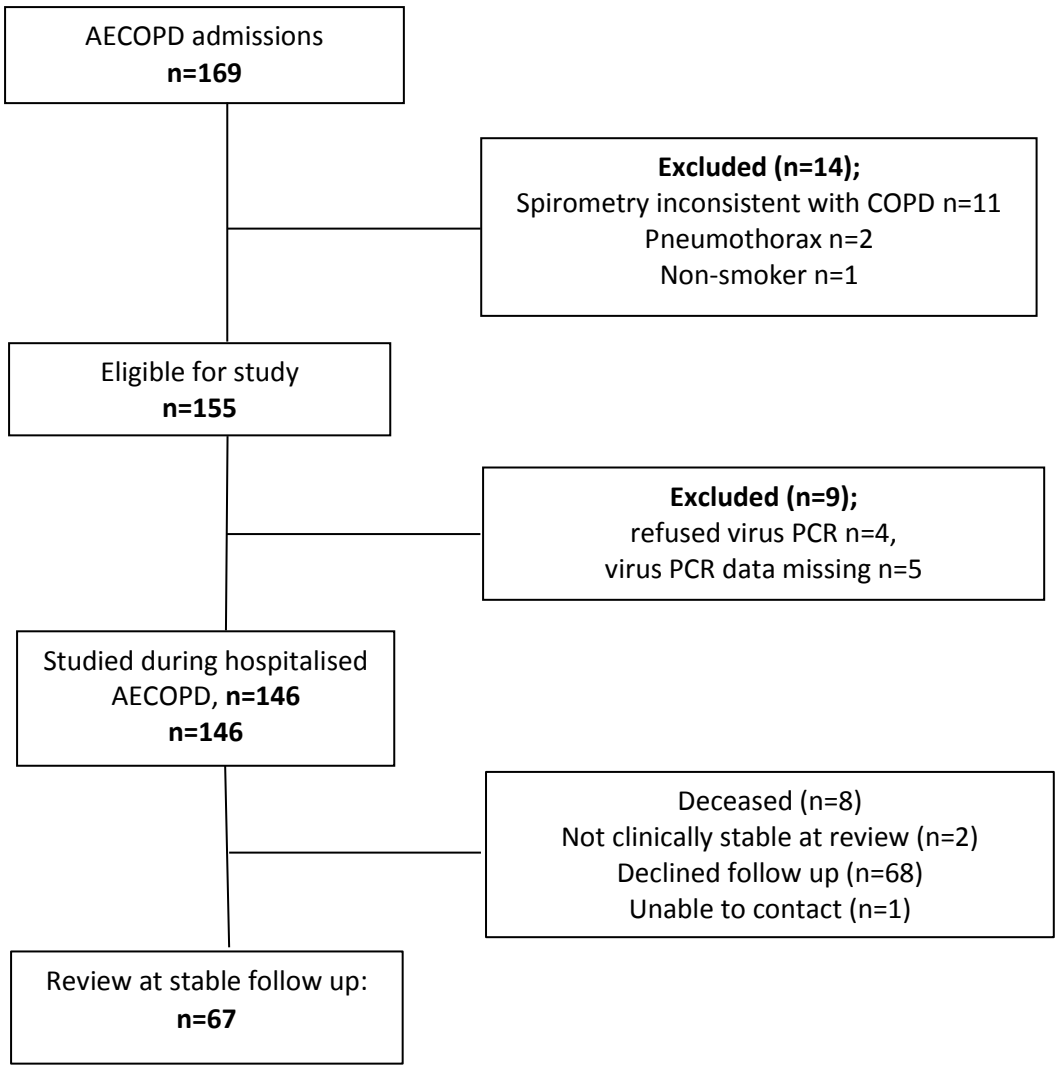
Figure 4. Survival 12 months post-hospital discharge based on presence or absence of infection in AECOPD

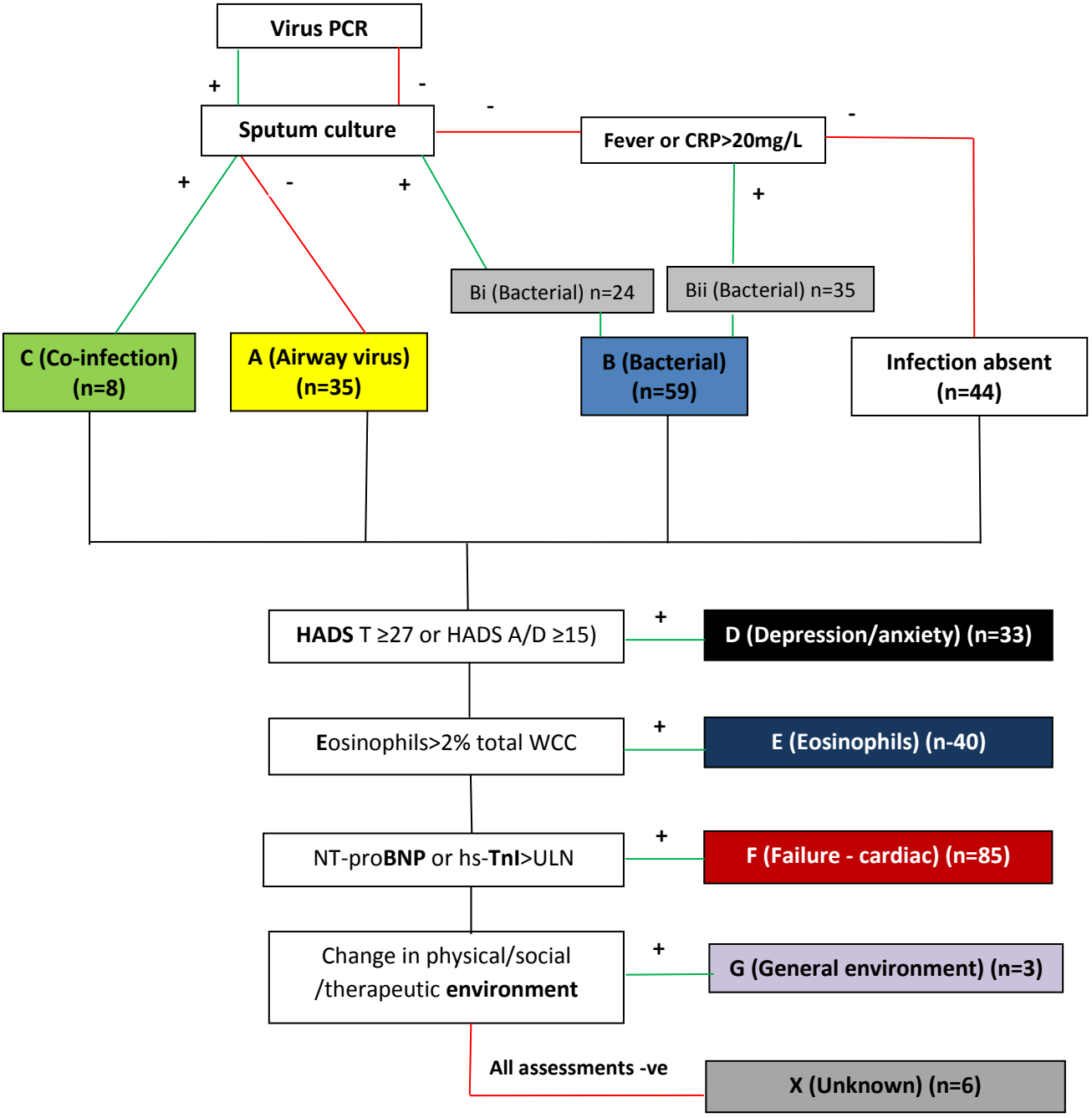
Funding support for this study was provided by an unrestricted educational grant from GlaxoSmithKline. The funder did not play any role in study design, data collection, analysis and interpretation of data or the decision to submit the paper for publication.

References

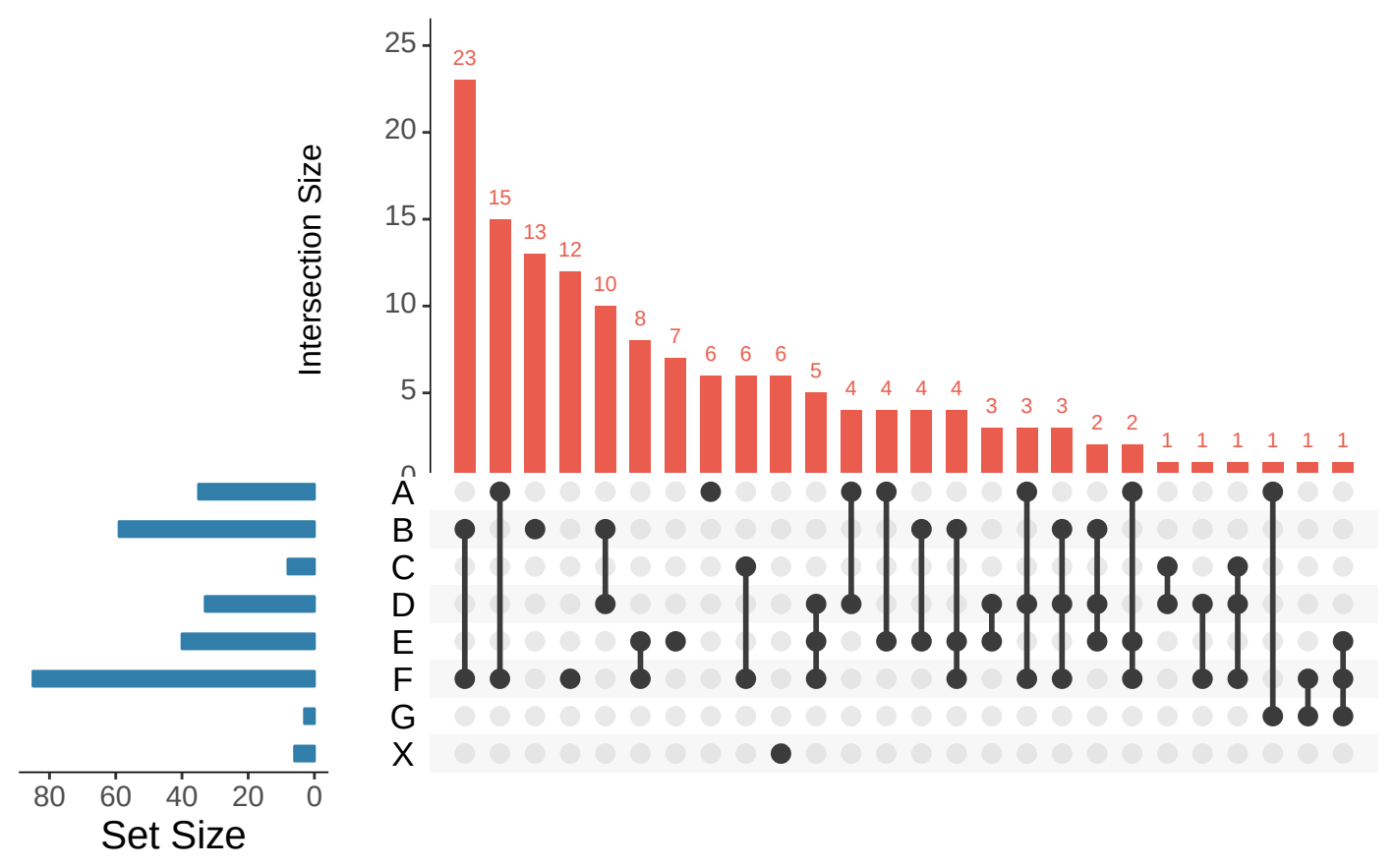
1. Han MK, Agusti A, Calverley PM, Celli BR, Criner G, Curtis JL, et al. Chronic obstructive pulmonary disease phenotypes: the future of COPD. American journal of respiratory and critical care medicine. 2010;182(5):598-604.
2. Vogelmeier CF, Criner GJ, Martinez FJ, Anzueto A, Barnes PJ, Bourbeau J, et al. Global Strategy for the Diagnosis, Management, and Prevention of Chronic Obstructive Lung Disease 2017 Report. GOLD Executive Summary. American journal of respiratory and critical care medicine. 2017;195(5):557-82.
3. MacDONALD M, BEASLEY RW, IRVING L, BARDIN PG. A hypothesis to phenotype COPD exacerbations by aetiology. Respirology. 2011;16(2):264-8.
4. MacDonald M, Korman T, King P, Hamza K, Bardin P. Exacerbation phenotyping in chronic obstructive pulmonary disease. Respirology. 2013;18(8):1280-1.
5. Bafadhel, McKenna S, Terry S, Mistry V, Reid C, Haldar P, et al. Acute exacerbations of chronic obstructive pulmonary disease: identification of biologic clusters and their biomarkers. American journal of respiratory and critical care medicine. 2011;184(6):662-71.
6. Mayhew D, Devos N, Lambert C, Brown JR, Clarke SC, Kim VL, et al. Longitudinal profiling of the lung microbiome in the AERIS study demonstrates repeatability of bacterial and eosinophilic COPD exacerbations. Thorax. 2018;73(5):422-30.
7. Vogelmeier, Criner GJ, Martinez FJ, Anzueto A, Barnes PJ, Bourbeau J, et al. Global Strategy for the Diagnosis, Management, and Prevention of Chronic Obstructive Lung Disease 2017 Report: GOLD Executive Summary. American journal of respiratory and critical care medicine. 2017;195(5):557-82.
8. Jones. COPD assessment test --rationale, development, validation and performance. Copd. 2013;10(2):269-71.
9. Zigmond AS, Snaith RP. The hospital anxiety and depression scale. Acta psychiatrica Scandinavica. 1983;67(6):361-70.
10. Hsu KY, Lin JR, Lin MS, Chen W, Chen YJ, Yan YH. The modified Medical Research Council dyspnoea scale is a good indicator of health-related quality of life in patients with chronic obstructive pulmonary disease. Singapore medical journal. 2013;54(6):321-7.
11. Peng C, Tian C, Zhang Y, Yang X, Feng Y, Fan H. C-reactive protein levels predict bacterial exacerbation in patients with chronic obstructive pulmonary disease. The American journal of the medical sciences. 2013;345(3):190-4.
12. Singer S, Kuhnt S, Gotze H, Hauss J, Hinz A, Liebmann A, et al. Hospital anxiety and depression scale cutoff scores for cancer patients in acute care. British journal of cancer. 2009;100(6):908-12.
13. Bafadhel M, McKenna S, Terry S, Mistry V, Reid C, Haldar P, et al. Acute exacerbations of chronic obstructive pulmonary disease: identification of biologic clusters and their biomarkers. American journal of respiratory and critical care medicine. 2011;184(6):662-71.
14. Chang, Robinson SC, Mills GD, Sullivan GD, Karalus NC, McLachlan JD, et al. Biochemical markers of cardiac dysfunction predict mortality in acute exacerbations of COPD. Thorax. 2011;66(9):764-8.
15. Galasko GI, Lahiri A, Barnes SC, Collinson P, Senior R. What is the normal range for N-terminal pro-brain natriuretic peptide? How well does this normal range screen for cardiovascular disease? European heart journal. 2005;26(21):2269-76.
16. Trambas C, Pickering JW, Than M, Bain C, Nie L, Paul E, et al. Impact of High-Sensitivity Troponin I Testing with Sex-Specific Cutoffs on the Diagnosis of Acute Myocardial Infarction. Clinical chemistry. 2016;62(6):831-8.
17. Sapey E, Stockley RA. COPD exacerbations . 2: aetiology. Thorax. 2006;61(3):250-8.
18. Hewitt R, Farne H, Ritchie A, Luke E, Johnston SL, Mallia P. The role of viral infections in exacerbations of chronic obstructive pulmonary disease and asthma. Therapeutic advances in respiratory disease. 2016;10(2):158-74.

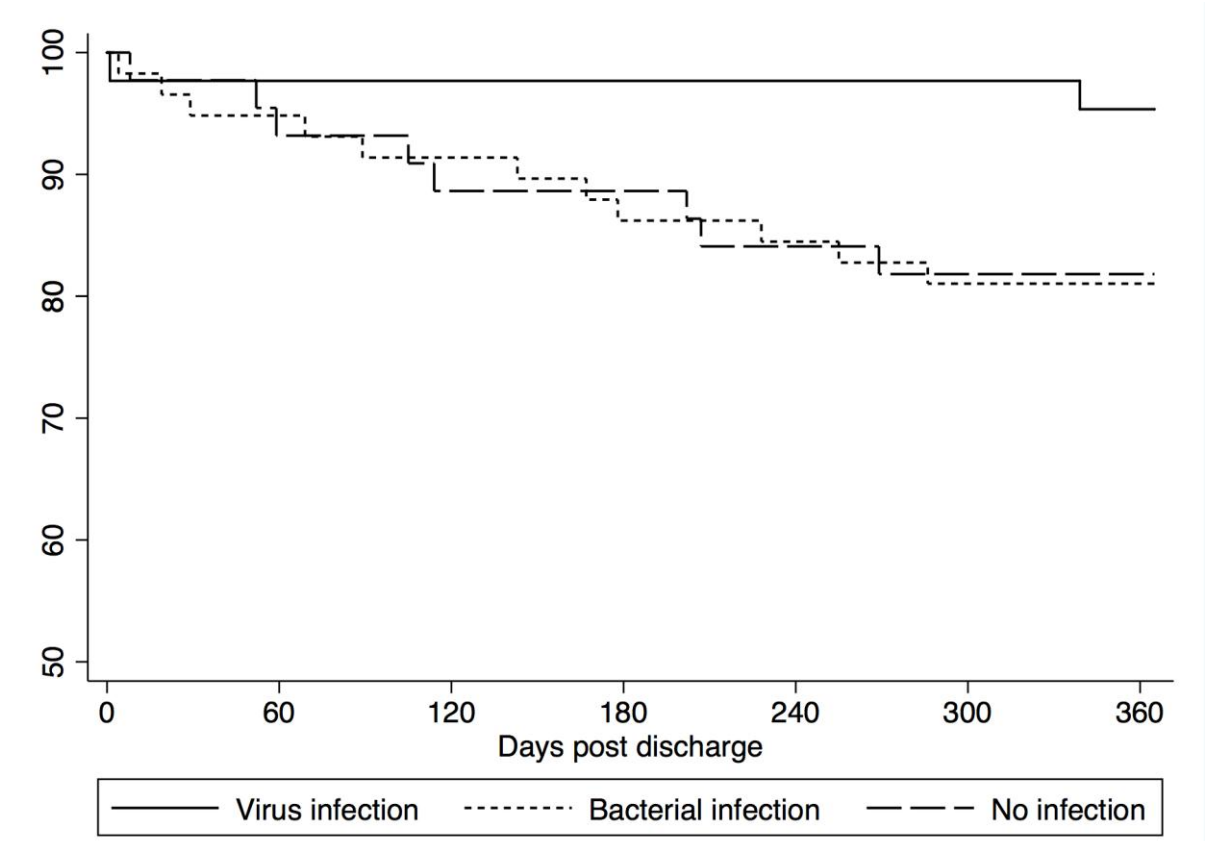
19. Zwaans WA, Mallia P, van Winden ME, Rohde GG. The relevance of respiratory viral infections in the exacerbations of chronic obstructive pulmonary disease-a systematic review. Journal of clinical virology : the official publication of the Pan American Society for Clinical Virology. 2014;61(2):181-8.
20. Peng C, Tian C, Zhang Y, Yang X, Fan H, Feng Y. C-Reactive Protein Levels Predict Bacterial Exacerbation in Patients With Chronic Obstructive Pulmonary Disease. The American journal of the medical sciences. 2012;345.
21. Filho FSL, Alotaibi NM, Ngan D, Tam S, Yang J, Hollander Z, et al. Sputum Microbiome Is Associated with 1-Year Mortality after Chronic Obstructive Pulmonary Disease Hospitalizations. American journal of respiratory and critical care medicine. 2019;199(10):1205-13.
22. Mallia P, Footitt J, Sotero R, Jepson A, Contoli M, Trujillo-Torralbo MB, et al. Rhinovirus infection induces degradation of antimicrobial peptides and secondary bacterial infection in chronic obstructive pulmonary disease. American journal of respiratory and critical care medicine. 2012;186(11):1117-24.
23. Wilkinson TM, Hurst JR, Perera WR, Wilks M, Donaldson GC, Wedzicha JA. Effect of interactions between lower airway bacterial and rhinoviral infection in exacerbations of COPD. Chest. 2006;129.
24. Laurin C, Moullec G, Bacon SL, Lavoie KL. Impact of anxiety and depression on chronic obstructive pulmonary disease exacerbation risk. American journal of respiratory and critical care medicine. 2012;185(9):918-23.
25. Eisner MD, Blanc PD, Yelin EH, Katz PP, Sanchez G, Iribarren C, et al. Influence of anxiety on health outcomes in COPD. Thorax. 2010;65(3):229-34.
26. MacDonald MI, Osadnik CR, Bulfin L, Hamza K, Leong P, Wong A, et al. Low and High Blood Eosinophil Counts as Biomarkers in Hospitalized Acute Exacerbations of COPD. Chest. 2019;156(1):92-100.
27. Aliyali M, Mehravaran H, Abedi S, Sharifpour A, Yazdani Cherati J. Impact of Comorbid Ischemic Heart Disease on Short-Term Outcomes of Patients Hospitalized for Acute Exacerbations of COPD. Tanaffos. 2015;14(3):165-71.
28. Almagro P, Cabrera FJ, Diez J, Boixeda R, Alonso Ortiz MB, Murio C, et al. Comorbidities and short-term prognosis in patients hospitalized for acute exacerbation of COPD: the EPOC en Servicios de medicina interna (ESMI) study. Chest. 2012;142(5):1126-33.
29. Antonelli Incalzi R, Fuso L, De Rosa M, Forastiere F, Rapiti E, Nardecchia B, et al. Co-morbidity contributes to predict mortality of patients with chronic obstructive pulmonary disease. The European respiratory journal. 1997;10(12):2794-800.
30. Marcun R, Sustic A, Brguljan PM, Kadivec S, Farkas J, Kosnik M, et al. Cardiac biomarkers predict outcome after hospitalisation for an acute exacerbation of chronic obstructive pulmonary disease. International journal of cardiology. 2012;161(3):156-9.
31. Agusti A, Bel E, Thomas M, Vogelmeier C, Brusselle G, Holgate S, et al. Treatable traits: toward precision medicine of chronic airway diseases. European Respiratory Journal. 2016;47(2):410-9.
32. Mathioudakis AG, Sivapalan P, Papi A, Vestbo J. The DisEntangling Chronic Obstructive pulmonary Disease Exacerbations clinical trials NETwork (DECODE-NET): rationale and vision. European Respiratory Journal. 2020;56(1):2000627.
33. Janssens W, Bafadhel M. The CICERO (Collaboration In COPD ExaceRbatiOns) Clinical Research Collaboration. European Respiratory Journal. 2020;55(3):2000079.


AECOPD admissions
n=169
Excluded (n=14);
Spirometry inconsistent with COPD n=11
Pneumothorax n=2
Non-smoker n=1
Eligible for study
n=155
Excluded (n=9);
refused virus PCR n=4,
virus PCR data missing n=5
Studied during hospitalised AECOPD, **n=146**
n=146
Deceased (n=8)
Not clinically stable at review (n=2)
Declined follow up (n=68)
Unable to contact (n=1)
Review at stable follow up:
n=67



PCR, polymerase chain reaction, MCS, microscopy and culture of sputum, CRP, C-reactive protein, HADS, Hospital Anxiety and Depression Scale, WCC= white cell count, NT-proBNP, N-terminal pro-brain natriuretic peptide, hs-TnI, high sensitivity troponin I, Bi = sputum culture positive, Bii, fever or CRP>20mg/dL with negative virus PCR.


Intersection Size
25
20
15
10
5
0
23
15
13
12
10
8
7
6
6
6
5
4
4
4
4
3
3
3
2
2
1
1
1
1
1
1
A
B
C
D
E
F
G
X
80
60
40
20
0
Set Size


100
90
80
70
60
50
0
60
120
180
240
300
360
Days post discharge
Virus infection
Bacterial infection
No infection

Supplementary Table 1. Characteristics of 10 most common phenotypes arranged in order of descending frequency

	BF	AF	B	F	BD	EF	E	A	CF	X
N	23	15	13	12	10	8	7	6	6	6
Age	74±10	73±10	72±11	73±10	67±6	70±10	73±13	74±6	74±15	65±11
Male (%)	56.5	60.0	69.2	58.3	60.0	50.0	57.1	50.0	66.7	50.0
Freq. (hospital) exac. (%)	43.5	13.3	38.5	33.3	50.0	25.0	42.9	16.7	16.7	16.7
Current smoker (%)	21.7	20.0	30.8	50.0	30.0	12.5	42.9	66.7	16.7	16.7
FEV_1 (% predicted)	52±21	46±19	42±12	36±13	33±12	39±11	43±22	50±16	51±26	43±18
TLCO (% predicted)	34±13	38±10	37±10	35±15	29±10	41±18	43±22	46±22	35±18	34±19
MRC-D	4[3-5]	3[3-5]	4[3-5]	4[4-5]	5[5-5]	3.5[2.5-5]	5[3-5]	2[2-4]	3.5[3-4]	3.5[3-5]
Days since symptom onset	3[1-7]	7[4-14]	7[4-14]	3[2-4]	5[3-7]	14[7-18]	4[2-7]	4[2-10]	6[2-7]	3.5[2-4]
Prior contact with HCP (%)	43.4	73.3	61.5	41.7	80.0	62.5	71.4	83.3	100	66.7
Pre-hospital antibiotics (%)	21.7	80.0	69.2	25.0	30.0	50.0	42.9	16.7	66.7	16.7
Pre-hospital OCS (%)	26.1	4.7	23.1	25.0	20.0	12.5	14.3	33.3	50.0	16.7
CAT total	25[22-30]	31[25-34]	25[22-33]	29[26-31]	35[33-37]	29[26-33]	32[23-33]	31[26-37]	30[28-37]	31[30-32]
HADS total	12[8-18]	14[11-21]	15[10-18]	16[12-19]	29[19-32]	22[19-23]	10[9-17]	20[14-22]	12.5[7-14]	6.5[6-9]
BAP-65 Class	3[2-3]	3[2-3]	2[2-3]	3[2-3]	3[2-3]	2[1-3]	3[2-3]	3[2-3]	2[2-3]	2[2-3]
Neutrophils	9.4±5	8.7±3	12.4±10	7.6±3.2	10.9±5.7	6.0±1.5	6.6±2	8.0±2.6	11.4±2.5	7.6±2
Eosinophils	0.03 [0.0-0.1]	0.01 [0.0-0.1]	0.01 [0-0.1]	0.1 [0.0-0.15]	0.04 [0.0-0.09]	0.43 [0.35-0.8]	0.33 [0.3-0.8]	0.05 [0.0-0.2]	0.09 [0.0-0.16]	0.07 [0.01-0.1]
CRP (mg/dL)	49 [30-107]	34 [8-64]	108 [52-176]	5 [2-11]	51 [42-54]	3 [2-4]	2 [1-4]	42 [39-45]	113 [67-160]	8 [4-12]
pH	7.38±0.1	7.31±0.1	7.35±0.1	7.33±0.1	7.37±0.1	7.31±0.1	7.29±0.0	7.39±0.0	7.4±0.0	7.36±0.1
P_vCO_2 (mmHg)	48±12	50±10	45±7	54±14	50±10	58±12	59±14	43±6	42±6	51±20
NT-proBNP (ng/L)	618 [387-2242]	1190 [439-1503]	159 [113-195]	1090 [341-2342]	108 [101-127]	357 [263-4820]	142 [111-157]	116 [63-191]	416 [350-2580]	139 [88-162]
Hs-TnI (ng/L)	18[7-38]	23[8-310]	5[3-10]	13[6-23]	7[4-8]	12[6-49]	5[3-5]	6[4-13]	34[4-74]	6[5-10]
NIV (ED) %	8.7	20.0	23.1	41.7	20.0	50.0	42.9	16.7	0.0	16.7
NIV (ward) %	4.3	26.7	7.7	41.7	0.0	12.5	14.3	16.7	0.0	16.7
Mechanical ventilation (%)	8.7	6.7	0.0	0.0	0.0	12.5	0.0	0.0	0.0	0.0
Length of stay	5[4-7]	9[6-13]	5[3-8]	7[5-8]	6[4-11]	4[3-5]	3[3-4]	6[4-9]	6[5-7]	7[4-13]
Mortality at 12 months (%)	21.7	13.3	23.1	33.3	20.0	25.0	0.0	0.0	0.0	16.7

*Data shown as (%), mean±SD, median[IQR]

Supplementary Table S2. Viruses and bacteria identified in 146 patients with AECOPD

Virus only	**n**	**Bacteria only**	**n**	**Virus with Bacteria**	**n**
Rhinovirus	12	Pseudomonas. aeruginosa.	7	Rhinovirus/Strep. pneumoniae	2
HMPV	8	Haemophilus. influenzae	9	Rhinovirus/Pseudomonas	2
Influenza A	7	Strep. pneumoniae	3	RSV/Streptococcus	2
RSV	3	Moraxella catarrhalis	3	Influenza A /Pseudomonas	1
Influenza B	2	MRSA	1	HMPV/Moraxella	1
Adenovirus	0	MSSA	1	**Total**	**8**
Parainfluenza 1	1	Pseudomonas + MRSA	1		
Parainfluenza 2	1	**Total**	**25**		
Rhinovirus/HMPV	1				
Total	**35**				

HMPV= human metapneumovirus, RSV= respiratory syncytial virus, MRSA= methicillin resistant staphylococcus aureus, MSSA= methicillin sensitive staphylococcus aureus

Supplementary Table S3. HADS scores within subgroups with or without psychological comorbidity

	Overall Population (n=136)	**Anxiety* (n=34)**	**Depression* (n=32)**	**Alcohol Misuse (n=11)**	**None (n=83)**
Male, n (%)	85 (62.5)	19 (55.9)	14 (43.8)	8 (72.7)	55 (66.3)
Antidepressant, n (%)	35 (25.7)	23 (67.6)	28 (87.5)	7 (63.6)	1 (1.2)
HADS Anxiety (mean/SD)	8.8/5.1	10.6/5.4	11.8/5.1	10.7/5.7	7.6/4.6
HADS Depression (mean/SD)	8.1/5.0	11/5.1	10.6/4.8	10.1/4.1	6.9/4.7
HADS Total (mean/SD)	16.8/8.8	21.1/8.9	21.8/8.4	19.3/8.4	14.6/8.1

*patients with comorbid anxiety/depression are included in both categories

Supplementary Table S4. Cardiac biomarker measurements in patients with and without a history of cardiovascular disease.

n(%), median [IQR]	Overall Population	HTN	IHD	Heart Failure	No known cardio-vascular disorder*
n	119	52	31	25	47
hs-TnI	9 [5-24]	10 [6-30.5]	9 [5-23]	16.5 [6-36]	7 [4-16]
hs-TnI >ULN,	32 (26.9)	18 (34.0)	8 (25.8)	11 (44.0)	9 (19.1)
NT-proBNP (ng/L)	368 [162-1201]	422 [174-1545]	422 [185-1545]	618 [185-2016]	258 [117-693]
NT-proBNP>ULN,	83 (69.7)	38 (71.7)	23 (74.2)	19 (75.9)	34 (72.3)

HTN= hypertension, IHD= ischaemic heart disease, *no known history of hypertension, ischaemic heart disease, heart failure, cerebrovascular disease or arrhythmia

Supplementary Table 5- Characteristics of patients who did versus did not undergo follow-up outpatient (stable) assessment.

Demographics (mean±SD), median[IQR]	Follow up	No follow up	p
Age	70.7±10.7	72.5±10.3	0.33
Male (%)	58.2	64.9	0.69
BMI kg/m^2	26.1±5.6	24.4±5.7	0.09
FEV_1 (% predicted)	46.2±19.9	50.0±18.5	0.4
TLCO (% predicted)	39.6±16	38.3±16.7	0.67
MRC-D	4 [3-5]	3 [4-5]	0.35
Current smoker (%)	34.3	29.9	0.33
Pack years	43.3±24.9	44.7±28.8	0.75
IHD (%)	19.4	33.8	0.053
Cardiac failure (%)	14.9	23.4	0.2
Diabetes (%)	22.4	16.9	0.4
Malignancy* (%)	10.5	7.8	0.58
Anxiety (%)	14.9	29.9	0.03*
Depression (%)	20.9	26.0	0.47
Frequent exacerbators (%) (≥ 2 AECOPD hospital admissions in prev. year)	23.9	32.5	0.26

*receiving treatment or palliation