Original Article

Sputum Culture and Antibiogram in Infective Acute Exacerbation of Chronic Obstructive Pulmonary Disease in a Tertiary Care Hospital in India

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Abstract

Background. Infections are the common cause of death in patients with acute exacerbation of chronic obstructive pulmonary disease (AECOPD).

Methods. A prospective cohort study was conducted on 100 patients with AECOPD in a tertiary care hospital in Hyderabad city in South India. Sputum culture of 100 patients was evaluated for the presence of Gram-positive and Gram-negative micro-organisms. Sensitivity and resistance patterns of the micro-organisms against commonly used antibiotics were also investigated.

Results. During the period October 2015 to April 2016, 67% of the patients had sputum culture positive for the presence of pathogenic micro-organisms. Pathogens, most commonly, isolated were Gram-negative organisms like *Klebsiella oxytoca* (13%), followed by *Klebsiella species* (11%) and *Klebsiella pneumoniae* (10%). Amikacin was found to be the most effective antibiotic against all micro-organisms. A significantly higher proportion of organisms were pathogenic compared to non-pathogenic organisms (p=0.0014). Among pathogenic organisms, a significantly higher proportion of Gram-positive organisms were found compared to Gram-negative organisms (p=0.0180). Pathogenic micro-organisms showed a high resistance rate to commonly used antibiotics. Except few strains of *Klebsiella*, *Pseudomonas* and *Streptococcus*, overall 62.9% strains were sensitive to doxycycline.

Conclusions. Our observations suggest that doxycycline can be used as an empirical antibiotic in the treatment of AECOPD. **[Indian J Chest Dis Allied Sci 2018;60:13-18]**

Key words: COPD, Exacerbations, Infection, Sensitivity.

Introduction

Chronic obstructive pulmonary disease (COPD) is a progressive disease characterised by airflow limitation and gradual loss of lung functions. Acute exacerbation of COPD (AECOPD) is an acute, worsening respiratory condition of the patient that is beyond usual day to day variation and necessitates a change in regular medication.[1] Exacerbations are the prime cause of hospitalisations and ultimately morbidity and mortality in COPD patients. It has been reported that patients with moderate to severe COPD suffered an average of two exacerbation episodes per year and infection was the common cause of death in patients with AECOPD.[2,3] Thus, there is an enhanced research need in the effectiveness of interventions used both to treat and prevent exacerbations.

Antibiotics are usually prescribed on an empiric basis in AECOPD patients. Nevertheless, the role of bacteria in causation of AECOPD is increasing in recent years,[4-7] the indication of antibiotics in its treatment is still debatable. The most common bacteria associated with exacerbations are *Haemophilus influenzae*, *Streptococcus pneumoniae* and *Moraxella catarrhalis*.[8] As COPD presents with bacterial colonisation,[9,10] it is difficult to differentiate between colonisation and infection. Furthermore, emergence of resistant strains of bacteria leads to wide variability in the effectiveness of antibiotics used. Thus, considering the above mentioned points, it would be helpful to find out patient characteristics and factors associated with AECOPD that may be useful in deciding the right antibiotic for the treatment and reducing the failure rates.

Hence, the present study was done to find out the bacteriological profile and antibiotic sensitivity patterns that may give the directions to the effective treatment of infective AECOPD patients.

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Material and Methods

A prospective, observational study was conducted in patients with AECOPD, presenting to out-patient department as well as those admitted to the wards of department of pulmonary medicine at our tertiary care hospital in Hyderabad, Telangana, South India.

Previously diagnosed patients with COPD on the basis of exposure to risk factors, clinical history and examination supported by spirometry presenting with acute exacerbation with (i) increased dyspnoea; (ii) increased sputum volume; and (iii) increased sputum purulence, requiring respiratory general ward admission or out-patient management were included.

Patients with bronchiectasis, tuberculosis, pneumonia, malignancy and other evident disease on chest radiography; patients previously admitted within 21 days and taken antibiotics; patients having sputum positive for acid-fast bacilli (AFB); pregnant and lactating women and those not willing to give or not able to give verbal informed consent were excluded from the study.

The study was carried out for seven months from October 2015 to April 2016. The study was approved by the Institutional Ethics Committee of Owaisi Hospital and Research Centre, Deccan College of Medical Sciences Hyderabad. All subjects gave informed consent to participate in the study.

In this observational, prospective study, demographic, clinical and microbiological data were collected form consecutive patients with AECOPD. The data were collected from patient's treatment chart/case sheets, previously available spirometry, laboratory reports, and patient's attendees. All the sputum samples were examined for the presence of more than 25 squamous epithelial cells at low power magnification examination with the microscope that suggests salivary contamination and were discarded and another sample obtained. After confirming the adequacy of the sample it was Gram stained and plated on Blood agar, Chocolate agar and Mac Conkey agar. If there was no significant growth after 24 hours it was further incubated for 48 hours for growth and all isolates were identified by microbiological standard operating procedures. Bacterial agents were classified into pathogenic micro-organisms or non-pathogenic micro-organisms.[10] Minimum inhibitory concentration (MIC) technique was used to find out the sensitivity of bacteria identified as pathogenic micro-organisms to antimicrobial agents. Antimicrobial sensitivity test was carried out by Kirby-Bauer disc diffusion method using Mueller Hinton agar. The bacteria was classified as sensitive and resistant to antibiotics according to the criteria issued by the National Committee for Clinical Laboratory Standards.[11]

Results

One hundred patients (83 males) were enrolled in the present study. Their mean age was 64.4 years. Among males, 22 were smokers, 54 were ex-smokers and 24 were non-smokers. Co-morbidities like hypertension was present in 72%, diabetes mellitus in 41%, coronary artery disease in 31% and prostatomegaly in 1%. Out of 100 patients, 41% used oxygen and 62% used nebuliser. Triggering factors such as allergy 6%, cold 21%, pollution 12%, weakness disabling expectoration 24% and unknown factors 37% were found in the patients. Majority of the patients were unemployed 43%, labours 18%, mill workers 8%, drivers 14% and females were all housewives 17% (Table 1). Seventy-one percent of patients had taken antibiotics during last three months. The values of lung function and arterial blood gases are listed in table 1.

Table 1. Socio-demographic characteristics.

Variable	Observations
Male:Female	83:17
Age (years),[mean (range)]	64.4 (36-95)
Occupation (%)	
Housewife	17
Non-workers	43
Labours	18
Mill workers	8
Driver	14
Personal habits (%)	
Smokers	22
Ex-smokers	54
Non-smokers	24
Triggerring factors (%)	
Allergy	6
Cold	21
Pollution	12
Weakness, unable to expectorate	24
Antibiotic use in last 3 months (%)	71
Oxygen use (%)	41
Nebulisation (%)	62
Co-morbidity (%)	
Hypertension	72
Diabetes mellitus	41
Coronary artery disease	31
Prostatomegaly	1
Lung function [mean (range)]	
FEV_1 (L)	2.07 (0.31-4.6)
FEV_1 (% predicted)	51.79 (13.0-87.4)
FVC (L)	2.42 (0.54-6.1)
FVC (% predicted)	56.1 (23-87)
FEV_1/FVC	72.31 (37.43-98.4)
Arterial blood gases	
Arterial oxygen tension (mmHg)	65.9 (28-99)
Arterial carbon dioxide tension (mmHg)	40.8 (22-54.05)
Arterial oxygen saturation (%)	86.8 (72-98.4)

Definition of abbreviations: FEV_1=Forced expiratory volume in the first second; FVC=Forced vital capacity

The non-pathogenic micro-organisms constituted 33% of the isolates; *Klebsiella oxytoca* (13%) was the most common isolate (Table 2). The most effective antibiotic against all the organisms was found to be amikacin followed by amoxicillin+clavulanic acid and the least effective antibiotic was clindamycin (Table 3). A significantly higher proportion of organisms were pathogenic compared to non-pathogenic organisms (÷2 = 10.259, 95% CI [Confidence interval] =11.6466 - 52.8514; p=0.0014). Among pathogenic organisms, a significantly higher proportion of Gram-positive organisms were found compared to Gram-negative organisms (÷2 = 5.601; 95% CI=3.5127 to 52.7892; p=0.0180).

Klebsiella oxytoca (11%) and *Klebsiella pneumoniae* (7%) were most frequently isolated from severe and very severe cases whereas methicillin sensitive *Staphylococcus aureus* (MSSA) constituted 3% of very severe cases, *Klebsiella species* 5%, followed by *Escherichia coli* 4%, *Streptococcus pyogenes* 3% and others were isolated from moderately severe cases (Table 4). Frequently used drug in this study against COPD microbe was amoxicillin and clavulanic acid 55%, followed by ceftriaxone 13%, cefotaxime 9% and piperacillin and tazobactum 7%. The least used drugs were norfloxacin, linezolid, cefoperazone, cefuroxime and cotrimoxazole (Table 5). Comparison of observations of present study with other published studies[13,14,19,23-27] is shown in table 5.

Table 2. Micro-organisms isolated from sputum culture.

Type of Organism	Gram staining	Organism	%
Pathogenic organisms (n=67)	Gram-negative (n=50)	*Klebsiella oxytoca*	13
		Klebsiella species	11
		Klebsiella pneumoniae	10
		Escherichia coli	6
		Acinetobacter	3
		Citrobacter freundii	2
		Pseudomonas aeruginosa	3
		Non-specific bacillus	2
	Gram-positive (n=17)	*MRSA*	1
		MSSA	7
		Streptococcus pyogenes	6
		Non-specific cocci	2
		Non-specific bacillus	1
Non-pathogenic organisms (n=33)			33

Definition of abbreviations: MRSA=Methicillin resistant *Staphylococcus aureus;* MSSA=Methicillin sensitive *Staphylococcus aureus*

Table 3. Drug Sensitivity pattern of various pathogens isolated from the sputum.

Antibiotics	*Escherichia coli* (N=6)	*Acinetobacter* (N=3)	*Citrobacter freundii* (N=2)	*Klebsiella oxytoca* (N=13)	*Klebsiella pneumoniae* (N=10)	*Klebsiella species* (N=11)	*MRSA* (N=1)	*MSSA* (N=7)	*Pseudomonas aeruginosa* (N=3)	*Streptococcus pyogenes* (N=6)	*Total* (N=62)
	S	S	S	S	S	S	S	S	S	S	S
	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)	n (%)
Amoxicillin+ Clavulanic acid	6 (100)	3 (100)	1 (50)	13 (100)	10 (100)	5 (45.45)	0 (0)	7 (100)	3 (100)	4 (66.67)	52 (83.87)
Piperacillin+ Tazobactam	6 (100)	2 (66.67)	2 (100)	10 (76.9)	8 (80.0)	8 (72.72)	0 (0)	6 (85.71)	2	2 (33.33)	46 (74.19)
Ampicillin+ Salbactam	6(100)	2 (66.67)	1 (50)	3 (23.0)	9 (90.0)	6 (54.54)	0 (0)	7 (100)	2	3 (50)	39 (62.90)
Amikacin	5 (83.33)	3 (100)	1 (50)	13 (100)	10 (100)	11 (100)	1 (100)	7 (100)	3 (100)	6 (100)	60 (96.77)
Cefotaxime	5 (83.33)	1 (33.33)	0 (0)	9 (69.23)	9 (90.0)	8 (72.72)	0 (0)	5 (71.42)	2	5 (83.33)	41 (66.12)
Tigecycline	4 (66.67)	1 (33.33)	2 (100)	13 (100)	7 (70)	11 (100)	1 (100)	0 (0)	3 (100)	6 (100)	48 (77.41)
Cotrimoxazole	3 (50.0)	0 (0)	0 (0)	10 (76.9)	0 (0)	5 (45.45)	0 (0)	0 (0)	3 (100)	6 (100)	27 (43.54)
Linezolid	4 (66.67)	2 (66.67)	0 (0)	5 (38.48)	5 (50)	7 (63.63)	1 (100)	7 (100)	0	4 (66.67)	35 (54.45)
Meropenem	5 (83.33)	2 (66.67)	1 (50)	13 (100)	10 (100)	10 (90.90)	1 (100)	6 (85.71)	3 (100)	6 (100)	58 (93.54)
Ciprofloxacin	6 (100)	3 (100)	2 (100)	4 (30.76)	8 (80)	10 (90.90)	0 (0)	6 (85.71)	3 (100)	4 (66.67)	46 (74.19)
Cefoperazone	5 (83.33)	0 (0)	1 (50)	13 (100)	7 (70)	5 (45.45)	0 (0)	2 (28.57)	3 (100)	3 (50.0)	39 (62.90)
Clindamycin	3 (50.0)	1 (33.33)	0 (0)	0 (0)	1 (10)	2 (18.18)	1 (100)	0 (0)	1 (33.33)	5 (83.33)	14 (22.58)
Clarithromycin	1 (16.67)	2 (66.67)	1 (50)	7 (53,84)	4 (40)	8 (72.72)	1 (100)	7 (100)	2 (66.67)	6 (100)	39 (62.90)
Minocycline	4 (66.67)	2 (66.67)	0 (0)	6 (46.15)	2 (20)	10 (90.90)	1 (100)	0 (0)	3 (100)	6 (100)	34 (54.83)
Colistin	5 (83.33)	3 (100)	1 (50)	13 (100)	10 (100)	11 (100)	0 (0)	0 (0)	3 (100)	4 (66.67)	50 (80.64)
Doxycycline	6 (100)	1 (33.33)	0 (0)	6 (46.15)	10 (100)	1 (9.09)	1 (100)	7 (100)	1 (33.33)	6 (100)	39 (62.90)

Definition of abbreviations: MRSA=Methicillin resistant *Staphylococcus aureus;* MSSA=Methicillin sensitive *Staphylococcus aureus;* S=Sensitive

Table 4. Pathogens isolated from sputum in patients with COPD

Micro-organisms	COPD Severity				Total
	Mild	Moderate	Severe	Very severe	
Pathogenic organisms					
Klebsiella oxytoca	0	2	9	2	13
Klebsiella species	2	5	2	2	11
Klebsiella pneumoniae	2	1	6	1	10
MRSA	0	0	1	0	1
MSSA	1	2	1	3	7
Streptococcus pyogenes	2	3	1	0	6
Escherichia coli	0	4	2	0	6
Acinetobacter	0	0	2	1	3
Citrobacter freundii	0	1	1	0	2
Pseudomonas aeruginosa	1	0	1	1	3
Non-specific Gram-positive cocci	0	1	1	0	2
Non-specific Gram-positive bacilli	0	0	1	0	1
Non-specific Gram-negative bacilli	0	1	1	0	2
Non-pathogenic micro-organisms	7	13	11	2	33

Definition of abbreviations: COPD=Chronic obstructive pulmonary diseases; MRSA=Methicillin resistant *Staphylococcus aureus;* MSSA=Methicillin sensitive *Staphylococcus aureus*

Table 5. Comparison of observations of present study with other published studies[13,14,19,23-27]

Organism	Current study	Basu *et al*[13]	Erkan *et al*[14]	Miravitlles *et al*[19]	Ko *et al*[23]	Kumar *et al*[24]	Shashi-bhushan *et al*[25]	Bathoom *et al*[26]	ElFeky *et al*[27]
Klebsiella species	34	33.33	2.6	-	2	-	15	-	-
Escherichia species	6	9.51	-	1.1	-	-	12	-	-
Pseudomonas species	3	19.05	-	15	6.3	30.8	23	20.8	-
Acinetobacter species	3	9.51	1.3	-	<2	-	-	-	3.3
Citrobacter species	2	-	-	1.1	-		1	-	-
Staphylococcus species	8	28.6	-	-	1	30.8	-	8.33	10
Streptococcus species	6	-	5.3	10	4	-	42	-	43.3
Haemophilus influenzae	-	-	30	22	23.1	-	-	41.6	6.7

Note: All figures are given as percentage

Discussion

Our study reveals that patients with AECOPD frequently presents with bacterial infections. Hence, antibiotic should be selected rationally depending upon the type of bacterial infection present. Other factors that may influence the antibiotic prescription include different strains of same bacteria, infection *versus* colonisation, resistance patterns, co-morbid conditions, etc. The most common pathogenic organisms isolated in our study was *Klebsiella oxytoca* whereas the least isolated was *Citrobacter freundii*. Commonly prescribed antibiotic was amoxycillin and clavulanic acid combination as an initial empirical treatment, although, all micro-organisms were found to be mostly sensitive to amikacin as most of the isolates were Gram-negative organisms. More number of severe casses had infection with *Klebsiella oxytoca* (9%) and *Klebsiella pneumoniae* 6% whereas MSSA were present in 3% of very severe cases.

Results of the various studies conducted on bacterial infection in AECOPD produce contrasting results.[4-27] This variance may be attributed to the factors, such as different study location, severity of COPD, non ICU *versus* ICU admission and in-patient *versus* out-patient status of the patients. Indian studies[24,25] on infections in patients with AECOPD are limited and yield conflicting results. *Pseudomonas aeruginosa* was the predominant isolate amongst the hospitalised patients followed by *S. pneumoniae* and

Acinetobacter spp, *Klebsiella spp*. and *M. catarrhalis* according to another study.[12] On the other hand, *Klebsiella pneumoniae* was the commonest bacteria isolated followed by *P. aeruginosa* and *Staphylococcus aureus* as per study conducted by Basu *et al*.[13] Four Western studies showed *Haemophilus influenzae* and *Chlamydophila influenzae* as one of the predominant causative organism in AECOPD.[14-18]

Sensitivity and resistance pattern of micro-organisms against antibiotics showed poor efficacy for clindamycin, minocycline, linezolid, clarithromycin, ampicillin, cefoperazone and cefotaxime. In contrast, amikacin, meropenem, colistin, doxicycline, ciprofloxacin and amoxicillin+clavulanic acid were found to be effective against a wide variety of micro-organisms. Amikacin seems to be the most promising drug against all micro-organisms. Gram-negative organisms were generally found in severe cases of AECOPD with *Klebsiella oxytoca*, *Klebsiella pneumoniae* and *MRSA* commonly present in severe cases of COPD. On the other hand, non-pathogenic micro-organisms were usually present in mild cases of AECOPD. Presence of pathogenic micro-organisms was directly proportional to functional impairment in AECOPD patients. In our study, mean forced expiratory volume in one second (FEV_1%) of the study population was 52% with a high predominance of pathogenic micro-organisms. This result was consistent with study conducted by Miravitlles *et al*[19] which showed that 56% of the study population with FEV_1 less than or equal to 50% had Gram-negative organisms as the cause of AECOPD.

Finally, sputum culture results are not an absolute marker for the presence of bacterial infection, as negative sputum culture does not rule out bacterial infection.[22] The plausible reasons could be the culture media used, bacterial load in the sample, self antibiotic medication by patients before visiting the doctor, etc.

There are several limitations to our study. The study sample used was small. There was a lack of retrospective data for this study. We did not stratify the sample based on the age, gender and socio-economic status. The adjusting factor to account for the influence of co morbid conditions was not included.

No information was collected about viral infection. Nevertheless, the availability of better invasive diagnostic techniques, sputum culture remains the choice of diagnostic approach for lower respiratory tract infections in majority of clinical settings.

References

1. Global Initiative for Chronic Obstructive Lung Disease. Global strategy for the diagnosis, management, and prevention of chronic obstructive pulmonary disease: updated 2005: Available at: www.goldcopd.org. Accessed on February 23, 2018.
2. Miravitlles M, Mayordomo C, Artes M, Sanchez-Agudo L, Nicolau F, Segu JL, on behalf of the EOLO Group. Treatment of chronic obstructive pulmonary disease and its exacerbations in general practice. *Respir Med* 1999;93:173–9.
3. Burrows B, Earle RH. Course and prognosis of chronic obstructive lung disease: a prospective study of 200 patients. *N Engl J Med* 1969;280:397–404.
4. Rosell A, Monso E, Soler N, Angrill J, Riise G, Zalacaín R, *et al*. Microbiologic determinants of exacerbation in chronic obstructive pulmonary disease. *Arch Intern Med* 2005;165:891–7.
5. Sheti S, Evans N, Grant BJ, Murphy TF. New strains of bacteria and exacerbations of chronic obstructive pulmonary disease. *N Engl J Med* 2002;347:465–71.
6. Murphy TF, Brauer AL, Eschberger K, Lobbins P, Grove L, Cai X, *et al*. Pseudomonas aeruginosa in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2008;177:853–60.
7. Sheti S, Murphy T. Infection in the pathogenesis and course of chronic obstructive pulmonary disease. *N Engl J Med* 2008;359:2355–65.
8. Ball P. Epidemiology and treatment of chronic bronchitis and its exacerbations. *Chest* 1995;108:43S–52S.
9. Haas H, Morris JF, Samson S, Kilbourn JP, Kim PJ. Bacterial flora of the respiratory tract in chronic bronchitis: comparison of transtracheal, fiberbronchoscopic and oropharyngeal sampling methods. *Am Rev Respir Dis* 1977;116:41–7.
10. Cabello H, Torres A, Celis R, El-Ebiary M, Puig de la Bellacasa J, Xaubet A, *et al*. Bacterial colonization of distal airways in healthy subjects and chronic lung diseases: a bronchoscopic study. *Eur Respir J* 1997;10:1137–44.
11. Performance standards for antimicrobial susceptibility testing. *National Committee for Clinical Laboratory Standards* 2013;33:27.
12. Chawla K, Mukhopadhay C, Majumdar M, Bairy I. Bacteriological profile and their antibiogram from cases of acute exacerbations of chronic obstructive pulmonary disease: a hospital based study. *J Clini Diagn Res* 2008;2:612–6.
13. Basu S, Mukherjee S, Samanta A. Epidemiological study of bacterial microbiology in aecopd patients of Kolkata. *Asian J Pharma Clin Res* 2013;6:112–6 .
14. Erkan L, Uzun O, Findik S, Katar D, Sanic A, Atici AG. Role of bacteria in acute exacerbations of chronic obstructive pulmonary disease. *Int J Chron Obstruct Pulmon Dis* 2008;3:463–7.
15. Beaty CD, Grayston JT, Wang SP, Kuo CC, Reto CS, Martin TR. Chlamydia pneumoniae, strain TWAR, infection in patients with chronic obstructive pulmonary disease. *Am Rev Respir Dis* 1991;144:1408–10.
16. Karnak D, Beng Sun S, Beder S, Kayacan O. Chlamydia pneumoniae infection and acute exacerbation of chronic obstructive pulmonary disease (COPD). *Respir Med* 2001;95:811–6.
17. Mogulkoc N, Karakurt S, Isalska B, Bayindir U, Celikel T, Korten V, *et al*. Acute purulant exacerbations of chronic obstructive pulmonary disease and Chlamydia pneumonia infection. *Am J Respir Crit Care Med* 1999;160:349–53.
18. Verkooyen RP, Van Lent NA, Mousavi Joulandan SA, Snijder RJ, Van Den Bosch JM, Van Heldens HP, *et al*. Diagnosis of

Chlamydia pneumoniae infection in patients chronic obstructive pulmonary disease by micro-immunofluorescence and ELISA. *J Med Microbiol* 1997;46:959–64.
19. Miravitlles M, Espinosa C, Fernandez-Laso E, Martos JA, Maldonado JA, Gallego M, *et al*. Relationship between bacterial flora in sputum and functional impairment in patients with acute exacerbations of COPD. *Chest* 1999;116:40–6.
20. Papi A, Bellettato CM, Braccioni F, Romagnoli M, Casolari P, Caramori G, *et al*. Infections and airway inflammation in chronic obstructive pulmonary disease severe exacerbations. *Am J Respir Crit Care Med* 2006;173:1114–21.
21. McManus TE, Marley AM, Baxter N, Christie SN, O'Neill HJ, Elborn JS, *et al*. Respiratory viral infection in exacerbations of COPD. *Respir Med* 2008;102:1575–80.
22. Murphy TF, Brauer AL, Schiffmacher AT, Sethi S. Persistent colonization by Haemophilus influenzae in chronic obstructive pulmonary disease. *Am J Respir Crit Care Med* 2004;170:266–72.
23. Ko Fanny WS, Ng Tony KC, Li Thomas ST, Fok Joan PC, Chan Michael CH, Wu Alan KL, *et al*. Sputum bacteriology in patients with acute exacerbations of COPD in Hong Kong. *Respir Med* 2005;99:454–60.
24. Kumar S, Megha V, Singh AV, Mehta S, Jad B, Bareja R. Bacteriological profile of sputum and their antibiogram in cases of acute exacerbation of COPD from a rural tertiary care hospital. *J Adv Res Biol Sci* 2012;4:115–9.
25. Shashibhushan BL, Nagaraja C, Arun BJ, Nagaraj N. Bacteriological profile and antibiotic sensitivity pattern in sputum culture of chronic obstructive pulmonary disease patients. *Int J Adv Med* 2016;3:671–4.
26. Bathoorn E, Groenhof F, Hendrix R, van der Molen T, Sinha B, Kerstjens HAM, *et al*. Real-life data on antibiotic prescription and sputum culture diagnostics in acute exacerbations of COPD in primary care. *Int J Chron Obstruct Pulmon Dis* 2017;12:285–90.
27. ElFeky DS, Elmandory HM, Galal M, Hakim MA. Sputum bacteriology in patients with acute exacerbation of chronic obstructive pulmonary disease. *Int J Curr Microbiol App* 2016;5:289–305.