

## A Comparative Bacteriological and Epidemiological Study Of Asthma Patients And Smokers

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### ABSTRACT

Asthma is a chronic respiratory disease that varies in etiology, pathogenesis, severity, and response to treatment. The present study was conducted to detect the bacteriological changes accompanying cases of asthma patients and smokers compared to healthy people. The current study included 88 patients with asthma and smokers who suffer from bacterial infections in the respiratory system, they were compared with 50 healthy people from 15/12/2022 to 28/6/2023 in the Allergy and Asthma Center in Baghdad. Sputum culture results showed that *Staphylococcus aureus* and *Streptococcus pneumoniae* were the dominant bacteria colonizing the respiratory tract of asthma patients and smokers compared to the colonization rates of other types of bacteria. The majority of bacteria isolated from asthma patients showed high resistance to Ampicillin and Gentamycin, at rates of 100% and 96%, respectively. In contrast, it showed the lowest resistance rate against Ciprofloxacin and, Augmentin at rates of 30% and 22%, respectively.

**Keywords:** *Asthma, bacterial infections, Smokers, Antibiotics, Respiratory infections*



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### Introduction

Asthma is one of the chronic respiratory diseases that varies in its causes, pathogenesis, severity, and response to treatment. It is a chronic inflammatory disorder in the airways in which many cells and cellular elements are involved, especially mast cells, eosinophils, T- cells, macrophage cells, neutrophil cells, and epithelial cells(1). Inhaling cigarette smoke stimulates the airways in people with asthma (2). Cigarette smoke is responsible for more than half of asthma cases in People over 40 years old. Cigarette smoking causes chronic bronchitis, a defect in the movement of cilia in the respiratory tract, and inhibits alveolar macrophages, as well as acute inflammation and bronchial irritation (3). Nicotine alters the host's pulmonary defense (4) and suppresses antimicrobial activity (5). Bacteria are among the pathogens responsible for lower respiratory infections after viruses (6). Bacterial infections exacerbate the severity of asthma attacks, whether the bacteria are pathogenic or coexisting in the respiratory tract. They can release many mediators, including histamine, which destroys the epithelial cells of the airways. Which leads to hypersensitivity, which leads to asthma attacks. The importance of this bacteria is also reflected in the role it plays as a source of allergens (7,8).

Bacterial infections participate in modifying the response in asthma patients and play an important role in the occurrence of wheezing which may increase their medical condition (9). Exotoxins and endotoxins secreted by pathogenic bacteria are among the pathogenic factors that cause allergies (10). Many studies have shown the role of bacterial allergens in causing allergic asthma. They participate as allergens that provoke respiratory allergies and hypersensitivity, such as those caused by bacteria: *Streptococcus pyogenes*, *Staphylococcus aureus*, *Haemophilus influenzae*, *Moraxella catarrhalis*, and *Streptococcus pneumoniae* (11, 12). Some researchers have indicated that antibiotics have an important role in eliminating the allergens of bacteria and limit their role by reducing the levels of histamine released into the airways (13). The current study aims to diagnose bacteria in the sputum of a group of patients with asthma, a group of smokers, and a group of apparently healthy people) and their role in causing infections and exacerbating asthma attacks, and making a comparison of the bacterial infections diagnosed between those groups.

### Materials and Methods

**1- Sample collection:** The study included (88) patients with asthma (the number of males was 50 and the number of females was (38), and their ages ranged between (5 - 75) years. It also included (88) smokers (62 male smokers and 26 female smokers) whose ages ranged from 17- 78 years old. Asthma was diagnosed by the specialist doctor at the Allergy

and Asthma Center in Baghdad based on clinical symptoms, as well as information on the history of the disease and family history of allergic diseases, as well as lower respiratory tract infections, according to a research form prepared. The study continued from 15/12/2022 until 28/6/2023.

**2-Culture of samples:** Morning sputum samples were collected in sterile containers and planted on plates of blood agar, chocolate agar, and MacConkey agar. Plates of blood and MacConkey agar were stored in aerobic conditions at 37°C for 24 days- 48 hours. Chocolate agar was incubated in Candle Jars to provide (5-10% CO<sub>2</sub>) and incubated at 37 °C for 24 days- 48 hours.

**3-Diagnosis of bacterial isolates:** Bacteria were identified based on the methods previously mentioned by both (14,15). The phenotypic characteristics of the colonies and biochemical tests were used for diagnosis.

**4- Antibiotics Sensitivity Test:** A susceptibility test was performed for the antibiotics by disk diffusion method according to the Kirby – Bauer method (16) using antibiotic discs (Streptomycin, Vancomycin, Ampicillin, Gentamicin, Augmentin, Ciprofloxacin, Tetracycline, Trimethoprim, Erythromycin, Nalidixic acid). The bacteria were considered sensitive or resistant based on the specifications contained in the Clinical and Laboratory Standards Institute (CLSI) (17).

## Results and discussion

The current study was conducted on 88 people with asthma compared with 50 healthy people (non-asthma and non-smokers) and was extended to include 88 smokers. The study showed that there were no significant differences between males and females. The total number of study groups was 226 people, as shown in Table (1).

**Table (1) :** The gender, number, and percentage of the study groups.

Study groups	Total numbers	Male		Female	
		No.	%	No.	%
Asthma	88	50	57	38	43
Smoker	88	62	70	26	30
Control	50	19	38	31	62

### ► Bacterial species isolated from healthy people:

50 sputum samples were collected from healthy people, and after conducting approved diagnostic tests, 50 isolates belonging to different genera of bacteria were identified, 14 of which (28%) belonged to *Streptococcus viridans*, 10 isolates (20%) *Staphylococcus epidermidis*. Similarly, 10 isolates (20%) belong to the *Staphylococcus albus*, 7 isolates (14%) *Staphylococcus aureus*, 5 isolates (10%) *Streptococcus pneumoniae*, and 4 (8%) isolates belonging to *Haemophilus influenza* bacteria.

The presence of these bacteria is normal in the upper respiratory tract, and these results were consistent with the results of several previous studies that indicated the presence of these bacterial species as commensals in the upper parts of the respiratory system (18,19). Despite the pathogenic properties of pneumococcal bacteria, they have been isolated from healthy people and many studies indicate the possibility of isolating this type of bacteria from healthy people. (20,21) .

**Table (2):** Percentage of bacterial species isolated from the sputum of asthma patients and healthy individuals(Control).

Bacterial species	Asthma patients(88)		Control(50)	
	No.	%	No.	%
<i>Streptococcus pneumoniae</i>	25	28	5	10
<i>Streptococcus pyogenes</i>	17	19	0	0
<i>Staphylococcus aureus</i>	10	11	7	14
<i>Staphylococcus albus</i>	8	9	10	20
<i>Streptococcus viridans</i>	7	8	14	28
<i>Moraxella cattarrhalis</i>	6	7	0	0
<i>Proteus mirabilis</i>	6	7	0	0
<i>Pseudomonas aeruginosa</i>	5	6	0	0
<i>Haemophilus influenza</i>	4	5	4	8
<i>Escherichia coli</i>	0	0	0	0
<i>Staphylococcus epidermidis</i>	0	0	10	20

### ► Bacterial species isolated from asthma patients and smokers:

88 sputum samples were collected from asthma patients and those who did not receive treatment that affected the test results, in addition to 88 sputum samples from smokers. The results of culturing these two groups and conducting

diagnostic tests for them showed that 88 isolates from asthma patients were diagnosed (Table 2), which included the following genera: *Streptococcus pneumoniae* 25 isolates (28%), *Streptococcus pyogenes* 17 (19%), *Staphylococcus aureus* 10 (11%), *Streptococcus viridans* 7 (8%), *Staphylococcus albus* 8 (9%), *Moraxella catarrhalis* 6 (7%), *Proteus mirabilis* 6 (7%), *Pseudomonas aeruginosa* 5 (6%) and *Haemophilus influenza* 4 (5%). The results of the statistical analysis showed that there were significant differences ( $P < 0.01$ ) in the percentages of isolated bacteria of asthma patients compared to healthy people, while *Streptococcus viridans* was the most frequent bacteria in healthy people, *Streptococcus pneumoniae* was also the most common bacteria in asthma patients, and only 5 isolates were isolated from the group of healthy people. As for *Moraxella catarrhalis* bacteria, despite some studies indicating that they are not pathogenic (22), researchers (23) have explained that these bacteria cause respiratory infections and play a latent role in stimulating many mediators that lead to the breakdown of the cells lining the bronchi lead to hypersensitivity in the upper and lower respiratory tract, which is confirmed by the isolation of this bacteria from healthy people in small proportions compared to patients (24,25). The results of this study were consistent with local studies (26, 27), although there are differences in the percentages of isolated bacterial species, this is due to inflammatory events and the accompanying mucus secretions and destruction of many cells in the airways of asthma patients, which follows the inflammatory response as a result of their exposure to inhaled allergens, which significantly changes the microenvironment of microorganisms. Bacterial infections play an important role in the occurrence of wheezing, especially in asthma patients, (28) explained that bacterial infections are responsible for (10.8%) of irritating asthma attacks, while researchers (29) explain that 12% of the triggering of asthma attacks is due to bacterial infections. It is clear from the results of culturing the sputum of smokers and comparing them to a group of healthy people (Table 3) that the frequency of *Streptococcus pneumoniae* in smokers at a rate of 31% compared to the healthy group (10%), with a significant difference of ( $P < 0.01$ ). *Escherichia coli*, *Pseudomonas aeruginosa*, and *Streptococcus pyogenes*, the frequency of bacteria in the smoker's group is observed at the following rates of 3%, 6%, and 8%, respectively, and they are not isolated from the group of healthy people. *Streptococcus viridans*, *Staphylococcus albus*, *Staphylococcus aureus*, and *Haemophilus influenza* are also more frequent in the group of smokers with a rate of 14%, 8%, 20%, and 10%, respectively, and the absence of isolation of both *Moraxella catarrhalis* and *Proteus mirabilis* bacteria from both groups, while *Staphylococcus epidermidis* is observed at a rate of 20% the group of healthy people and they are not isolated from the group of smokers. It is worth noting that most of the bacterial isolates that were isolated during the study are found naturally in the upper respiratory tract, and some of them become active when exposed to the respiratory system (30) for viruses and bacteria, especially opportunistic bacteria, it should be noted that bacterial infections have an effect in causing allergic asthma, as they act as carcinogens that provoke respiratory allergies and cause hypersensitivity, in addition to participating in modulating response in asthma patients, which may worsen their disease condition (31).

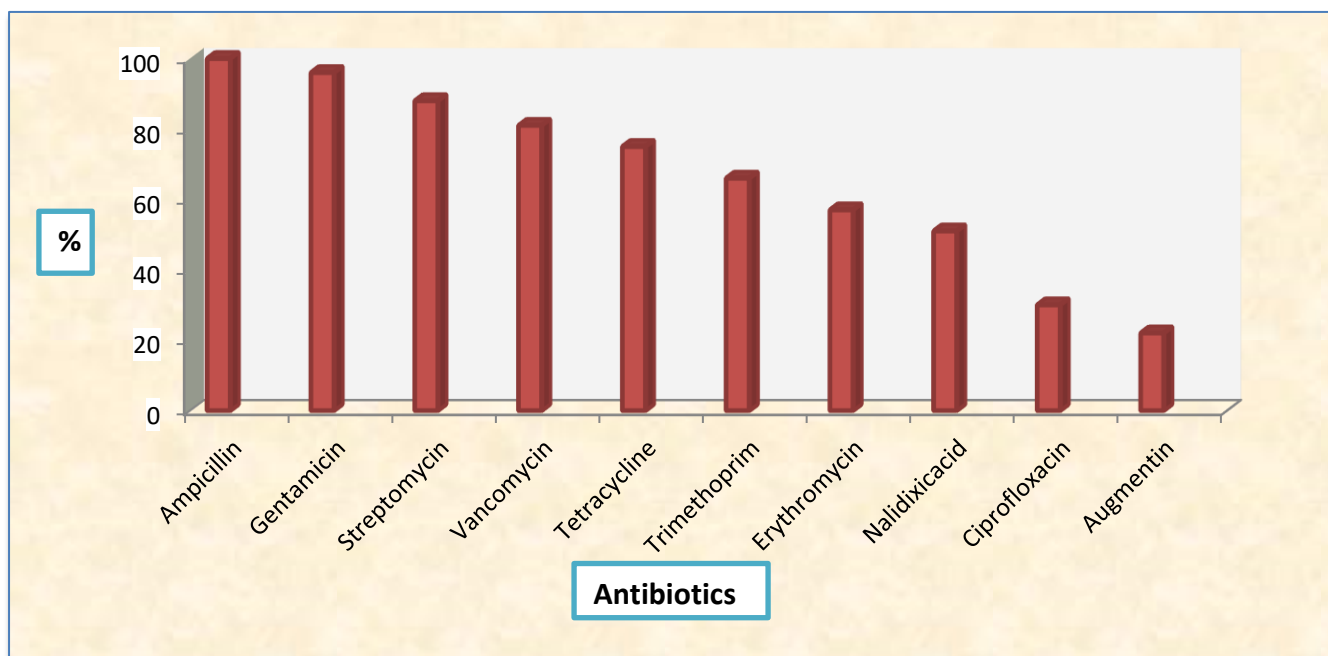
**Table (3):** Percentage of bacterial species isolated from the sputum of smokers and healthy individuals (Control).

Bacterial species	Smokers (88)		Control(50)	
	No.	%	No.	%
<i>Streptococcus pneumoniae</i>	27	31	5	10
<i>Streptococcus pyogenes</i>	7	8	0	0
<i>Staphylococcus aureus</i>	18	20	7	14
<i>Staphylococcus albus</i>	7	8	10	20
<i>Streptococcus viridans</i>	12	14	14	28
<i>Moraxella catarrhalis</i>	0	0	0	0
<i>Proteus mirabilis</i>	0	0	0	0
<i>Pseudomonas aeruginosa</i>	5	6	0	0
<i>Haemophilus influenza</i>	9	10	4	8
<i>Escherichia coli</i>	3	3	0	0
<i>Staphylococcus epidermidis</i>	0	0	10	20

► **Resistance of bacterial isolates isolated from asthma patients to antibiotics:**

The sensitivity test of bacteria isolated from asthma patients was performed for several antibiotics. From the observation of Figure (1), we find a variation in the resistance of bacterial isolates according to their genera and types. The highest resistance rate was against Ampicillin and Gentamicin, at rates of 100% and 96%, respectively, while it showed the lowest resistance rate against Ciprofloxacin and Augmentin at rates of 30% and 22%, respectively. The rates of resistance to Streptomycin, Vancomycin, Tetracycline, Trimethoprim, Erythromycin, and Nalidixic acid were 88%, 81%, 75%, 66%, 57 and 51%, respectively. We note from these results that despite the variation of the isolates subject to testing in their resistance to antibiotics, they showed a clear similarity in their sensitivity to both Ciprofloxacin and Augmentin. This may be due to the nature of the effect of these antibiotics and the lack of resistance genes encoded on conjugated or transferable plasmids, which prevents the spread of such resistance, or it may be due to the lack of widespread use of these antibiotics. This study came in agreement with what was indicated by (32). As for the high resistance to the antibiotics Ampicillin and Gentamicin, this is undoubtedly due to the repeated and random use of these

antibiotics. This result came in agreement with the results reported by researchers (33). Among the problems and difficulties in treating respiratory tract infections is the presence of more than one bacterial species at the site of infection that vary in their sensitivity to antibiotics, but they may show cross-resistance to a particular antibiotic due to the transfer of resistance plasmids between them (34). Therefore, antibiotic treatment has a specific but clear role in controlling the hypersensitivity that occurs in the airways during bacterial infections of the respiratory system in asthma patients. For this reason, performing a sensitivity test is necessary to determine the appropriate antibiotic for treatment.



**Figure (1):** Resistance of bacterial isolates isolated from asthma patients to different antibiotics

## Conclusion

The results of the study showed that *Staphylococcus aureus* and *Streptococcus pneumoniae* were the dominant bacteria colonizing the respiratory tract of asthma patients and smokers compared to the colonization rates of other types of bacteria. The majority of bacterial species isolated from asthma patients showed high resistance to the antibiotics used in the study. Therefore, it is necessary to conduct a sensitivity test to determine the appropriate antibiotics that can be added to the anti-asthma treatment, which helps in improving the patient's clinical condition.

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