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Antibiotic susceptibility and virulence factors of bacterial species among cancer patients

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ABSTRACT

Antibiotic resistance is one of the most significant challenges of the 20-s century, and the misuse of antibiotics is a driver of antimicrobial resistance. This study aimed to assess the prevalence of multidrug resistance, and detection of its produce virulence factors, including extended-spectrum β-lactamases (ESβLs), biofilm, and siderophores produced by bacterial species isolated from cancer patients. One hundred and seventy-five Gram-negative bacterial isolates were isolated from different samples collected from cancer patients admitted to the National Cancer Institute (NCI), Cairo, Egypt, and processed by standard microbiological methods. One hundred and forty-three bacterial isolates were recovered from adult patients, and 32 were recovered from children. *Escherichia coli* showed the highest frequency (36%), followed by *Klebsiella pneumonia* (30.85%), *Acinetobacter baummannii* (14.28%), and *Pseudomonas* sp. (9.14%). Antibiotic profiles revealed that bacterial isolates are highly resistant to the most commonly available antibiotics. Amikacin and gentamicin were the most effective antibiotics against isolated Gram-negative bacteria. Moreover, the vast majority of bacterial stains produce virulence factors, including EsβLs, biofilm, and siderophores. *E. coli* isolates produced ESβLs with rates of 25.28%, *Klebsiella pneumonia* (11.0%), and *Pseudomonas* sp. (25.0%). Among these collected bacterial isolates, 132 (75.4%) have the ability to form a biofilm to different degrees. Also, the majority of the bacteria isolates generated siderophores, with 133 (75.94%). This study revealed that a significant distribution of multidrug-resistant pathogenic bacteria may increase the burden on healthcare to prevent infections in cancer patients.

1. Introduction

Chronic diseases especially cancer, usually minimize immunity. Patients with cancer are highly susceptible to many types of microbial infections.[1] These infections among cancer patients could happen either endogenously from normal flora at the wound and operative site or exogenously from the hospital staff, inanimate environment, air, and medical equipment.[2] Microbial infection causes a significant problem in cancer patients due to its direct and indirect effects on their immune system. Many factors increase the susceptibility of immunosuppressed cancer patients to infection, such as neutropenia during aggressive therapy, shift of normal flora because of frequent antibiotic administration, disruption of skin, and damage of epithelial surfaces by cytotoxic agents.[3]

Treatment of multidrug-resistant bacteria (MDR) represents a global health challenge that results in increased morbidity and mortality rates worldwide. Infection with (MDR) bacteria is the main cause of death for more than 700.000 people annually worldwide which may rise to ~10 million by 2050.[4] The World Health Organization (WHO) considered multidrug-resistant bacteria from the ESKAPE group (*Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter, Pseudomonas aeruginosa,* and *Enterobacter*) to be highly dangerous infections.[5] Infections due to Gram-negative bacilli are common in cancer patients during aggressive therapy.[6] Data from several large surveillance studies conducted at major cancer centers in both the United States and Europe indicated that *Enterobacteriaceae* cause approximately 65%–80% of documented Gram-negative infections in cancer patients.[7]

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The pathogenic bacterial strains that have been exposed to a wide variety of β-lactam antibiotics over time have induced dynamic and continuous production and mutation of β-lactamases in these bacteria. Extended-spectrum β-lactamases (ESβLs) produced by bacterial strains are one of the major groups of β-lactamases that induce resistance to antibiotics. Genes encoding (ESβLs) are usually discovered on large plasmids that carry antibiotic resistance genes conferring resistance against different antimicrobial agents. Thus, several (ESβLs)-producing bacterial strains are also resistant to non-β-lactam antibiotics.[8] The treatment of bacterial pathogens produced (ESβLs) is very important due to several factors, including the difficulty of identifying (ESβLs) production and inconsistent reporting, it is challenging to determine the incidence of (ESβLs) producing organisms on a larger geographic scale.

A biofilm is characterized by a colony of bacteria and an extracellular polymeric substance (EPS) matrix that the bacteria have created on their own. A biofilm shields bacterial cells from adverse environmental factors like extremes in temperature, dehydration, and biocides. Bacterial biofilms pose a significant threat to several applications, including medical equipment such as catheters, stents, and implants. Biofilm formation is often considered to be the underlying reason why treatment with an antimicrobial agent fails, and as an estimated 65–80% of all infections are thought to be biofilm-related, that presents a serious challenge.[9]

Siderophores are organic compounds with low-weight, high-affinity iron-chelating molecules that are produced by both Gram-positive and Gram-negative bacteria in response to an iron shortage and are frequently referred to as essential virulence factors in some bacterial species. More than 500 types of siderophores have been discovered, such as mycobactin, pyoverdine, pyochelin, ferrichrome, enterobactin, parabactin, petrobactin, and staphyloferrin B.[10] Previous studies have shown that a reduction in the pathogenicity of bacteria is associated with the production and/or function of siderophores such as exotoxin A, diphtheria toxin (*C. diphtheriae*), Shiga toxin (*Shigella*) and Shiga-like toxin Enterohemorrhagic (*E. coli*). Also, the production of biofilms is restricted when there is an iron shortage because the hydrophobicity of the microbial surface diminishes, and the makeup of surface proteins alters. Furthermore, the virulence factors are frequently linked to the deletion of genes controlling siderophore expression or another Fe-collecting system.[11] Therefore, the present study aimed to determine the antibiotic susceptibility pattern, the ability of biofilm formation, siderophore production, and the challenges in the treatment of pathogenic gram-negative bacteria isolated from cancer patients at the National Cancer Institute (NCI), Cairo, Egypt.

2. Materials and methods

One hundred and seventy-five samples, including blood, pus, sputum, drain, Central Venous Port (CVP), urine, tongue swab, anal swab, ear swab, throat swab, and tube samples, were collected from cancer patients (adults and children, male and female) during the period from December 2016 to July 2018 at the National Cancer Institute (NCI), Cairo, Egypt. The collected samples were cultivated on MacConkey and blood agar and subjected to conventional microbiological procedures. All media were readily prepared (Oxford, England).

2.1. Assessment, isolation, and purification of bacterial isolates

The plates containing MacConkey and blood agar media were inoculated with samples collected and incubated at 37 °C for 24 and 48 h. The grown colonies were selected, picked up, and transferred to agar slants containing the same medium. The purified isolates were subjected to a scheme of experimental identification.

2.2. Identification of bacterial isolates

The pure culture was identified based on morphology and biochemical tests[12] and was confirmed by the VITEK2 compact automatic system (Biomerieux Inc., Marcy I'Etoile, France). I'Etoile, France).

2.3. Antibiotic susceptibility testing

The susceptibility of bacterial isolates to commonly used antibiotics for the treatment of bacterial infections was performed with the VITEK 2 compact automatic system (Biomerieux Inc., Marcy I'Etoile, France) and disk diffusion method according to the Clinical and Laboratory Standards Institute (CLSI) recommendation in 2018.[13]

2.4. Detection of the virulence factors

2.4. 1. Screening of extended-spectrum β-lactamases (ESβLs) production

Bacterial species that produce ESβLs are a serious global public health concern and a contributing factor to resistance worldwide. Hence, the bacterial isolates were screened for ESβL production by the VITEK2 compact automatic system (Biomerieux Inc., Marcy I'Etoile, France). The VITEK2 automated susceptibility system has introduced an ESβLs test on their system, whereby ceftazidime and cefotaxime are tested alone and in combination with clavulanic acid. The reduction in growth within the well containing clavulanic acid compared to the well not containing it indicates the expression of ESβLs.[14]

2.4. 2. Detection of biofilm formation

For detection of biofilm formation, the bacterial isolates were inoculated in 5 ml of trypticase soy broth and incubated at 37 °C for 24 h. After that, the cultures were diluted at 1:100 with a fresh medium. Individual wells of 96 well–flat-bottom polystyrene Tissue Culture Plates (TCPs) (Thermo Fisher Scientific, Shanghai, China) were filled with 0.2 ml of diluted culture and incubated at 37 °C for 24 h. After the end of the incubation period, the contents were removed from the plates by tapping gently. Plates were washed twice with 0.2 ml of phosphate saline buffer (pH 7.2) and incubated at 37 °C for 1 h. The plates were stained with 0.2 ml of 0.1% crystal violet for 10 min. The excess stain was removed by washing twice with deionized water, and the plates were kept for drying. Following the addition of 200 μl of 33% glacial acetic acid into the wells, the optical density (OD) was measured at 570 nm using an ELISA auto-reader (BIORAD 680). The experiment was performed in triplicate. The interpretation of biofilm production was done according to Stepanovic et al., OD < OD control = no biofilm formation; OD = 0.17–0.34 weak positive; OD = 0.35–0.68 moderate positive; and OD = 0.68 strong positive.[15]

2.4. 2. Production of siderophores by bacterial isolates

Siderophores contribute to the colonization of pathogens in the host and increase the severity of disease. The detection of the ability of bacterial isolates to produce a siderophore was conducted using Minimal Media 9 (MM9) (HiMedia, India) supplemented with glucose and casmino acid (2 g/L).The turbidity of the bacterial growth was adjusted to 0.01 McFarland solution, inoculated into MM9, and incubated for 48 h at 37 °C. Appearing growth in the medium indicates positive results.[16]

3. Results

3.1. Bacterial species isolated from cancer patients

One hundred and seventy-five Gram-negative bacterial species isolated from cancer patients were included in the present study. Of the 175 bacterial isolates, 143 (81.71%) were recovered from adult patients and 32 (18.28%) were from children. Bacterial isolates were recovered mainly from blood samples (n = 72), pus (n = 58), sputum (n = 25), drains (n = 7) and urine (n = 4). Among the 175 included Gram-negative bacterial isolates, the predominant bacterial isolates were *E. coli* (36%), followed by *Klebsiella pneumonia* (30.85%), *Acinetobacter baummannii* (14.28%), *Pseudomonas* sp. (9.14%), *Enterobacter* sp. (4.57%), and

Proteus mirabilis (1.71%) as shown in Table 1.

3.2. Antibiotics profile of bacterial species isolated from cancer patients

The antibiotic resistance patterns of the different Gram-negative bacterial species showed high resistance to ampicillin, cefazolin, ceftriaxone, ampicillin-sulbactam, and trimethoprim/sulfamethoxazole with a ratio of (97.07%), (95.35%), (90.21%), (87.93%) and (86.79%) respectively. The most potent antibiotics against Gram-negative bacteria were amikacin and gentamicin with sensitivity ratios of (57.67%) and (38.25%) respectively as shown in Table 2.

3.3. Detection of the virulence factors

3.3. 1. Production of extended-spectrum β-lactamase (ESβLs) by gram-negative bacterial isolates

In the present study, a significant proportion of the bacterial isolates are considered MDR bacteria. Extended-spectrum β-lactamase (ESβLs) producing Gram-negative bacteria recovered from cancer patients were evident, *E. coli* produced (ESβLs) with rates of (25.28%), *Klebsiella pneumonia* (11.10 %) and *Pseudomonas* sp. (25.0 %).

3.3.2. Biofilm formation by bacterial isolated from cancer patients

In this study, 132 bacterial isolates with a ratio of (75.4%) have the ability to produce biofilm in different degrees as follows, *E. coli, Klebsiella pneumonia, Acinetobacter baummannii*, *Pseudomonas* sp., *Enterobacter* sp. *Proteus mirabilis* and *Citrobacter freundii* with 50 (79.35%), 39 (72.15%), 20 (80.0%), 13 (81.25%), 6 (75.0%), 3 (100%), 1 (50.0%) respectively as shown in Table 3. Bacterial isolates biofilm forming were highly resistant to tested antibiotics comparable with a non-biofilm bacterial strain. The most bacterial-forming biofilm with different degrees was isolated from blood, pus, sputum, and urine samples, respectively.

3.3. 3. Production of siderophores by bacterial isolates

Siderophores contribute to the colonization of bacterial infections within the host and help them pull iron from host-bound proteins. The results showed the ability of the bacterial isolates under study to produce siderophore with 147 (83.93). The most bacterial isolates that produce siderophore were found *E. coli, Klebsiella pneumonia*, *Acinetobacter baummannii*, *Pseudomonas* sp., *Enterobacter* sp., *Proteus mirabilis*, and *Citrobacter freundii*, while *Serratia marcescens*, *Burkholderia cepacian*, and *Pantoea agglomerans* did not produce siderophore, as shown in Table 4.

4. Discussion

Cancer patients are highly susceptible to microbial infections. Severe infections due to Gram-negative rods are common in cancer patients. Cancer patients' normal flora is shifted due to frequent antibiotic administration and damage to epithelial surfaces contributes to the development of infection.[1] In the present study, we have analyzed the resistance pattern of pathogenic Gram-negative bacteria isolated from cancer patients admitted from December 2016 to July 2018 at the National Cancer Institute (NCI), Cairo, Egypt. Vahedian-Ardakani et al. demonstrated that the re-emergence of Gram-negative bacteria is a major cause of infection in cancer patients with a percentage of 84.9%.[17] The International American Therapy Cooperative and the European Organization for Research and Cancer Treatment reported that Gram-positive pathogens were consistently rising from 1973 to 1994.[18] In addition, a study by Sirkhazi et al. reported that most bacterial pathogens isolated from cancer patients were Gram-negative bacteria with rates of 65.94% (91/138) and Gram-positive bacteria with 34.06% (47/138).[19] In this study, the most frequent gram-negative bacterial species in cancer patients were *E. coli* (36%), followed by *Klebsiella pneumonia* (30.85%) *Acinetobacter baummannii* (14.28%) *Pseudomonas*

Table (2)
Antibiotics profile of Gram-negative isolates.

No	Antibiotics	Abbr.	Resistance N (%)	Intermediate N (%)	Sensitive N (%)
1	**Amoxicillin/ Clavulanic acid**	**AMC**	150 (85.65)	8 (4.56)	17 (9.70)
2	**Ampicillin**	**AMP**	170 (97.07)	2 (1.14)	3 (1.71)
3	**Amikacin**	**AK**	69 (39.39)	5 (2.85)	101 (57.67)
4	**Levofloxacin**	**LEV**	122 (69.66)	5 (2.85)	48 (27.40)
5	**Cefotaxime**	**CTX**	136 (77.65)	9 (5.13)	30 (17.13)
6	**Ciprofloxacin**	**CIP**	126 (71.94)	8 (4.56)	41 (23.41)
7	**Ceftriaxone**	**CRO**	158 (90.21)	1 (0.57)	16 (9.13)
8	**Ceftazidime**	**CAZ**	150 (85.65)	6 (3.42)	17 (9.70)
9	**Gentamycin**	**CN**	105 (59.95)	3 (1.71)	67 (38.25)
10	**Cefepime**	**FEP**	147 (83.93)	4 (2.28)	24 (13.70)
11	**Cefoxitin**	**FOX**	128 (73.08)	8 (4.56)	39 (22.26)
12	**Nitrofurantoin**	**F**	99 (56.52)	18 (10.27)	58 (33.11)
13	**Meropenem**	**MEM**	115 (65.66)	0.0 (0.0)	60 (34.26)
14	**Ampicillin/ Sulbactam**	**SAM**	154 (87.93)	7 (3.99)	14 (7.99)
15	**Trimethoprim/ sulfamethoxazole**	**SXT**	152 (86.79)	0 (0.0)	23 (13.13)
16	**Piperacillin/ tazobactam**	**TZB**	116 (66.23)	13 (7.42)	46 (26.26)
17	**Tobramycin**	**TOB**	104 (59.38)	15 (8.56)	56 (31.97)
18	**Cefazolin**	**CE**	167 (95.35)	0.0 (0.0)	8 (4.56)

Table (1)
Isolated Gram-negative bacterial species among cancer patients.

No	Bacteria species	Pus	Oral	Tube	Ear swab	CVP	Anal swab	Sputum	Urine	Drain	Blood	Total (%)
1	***Escherichia coli***	25	1	0	0	0	1	3	2	4	27	63 (36)
2	***Klebsiella pneumonia***	14	1	1	1	1	0	9	2	1	24	54 (30.85)
3	***Acinetobacter baummannii***	3	0	0	0	0	1	9	0	1	11	25 (14.28
4	***Pseudomonas* sp.**	9	0	0	0	0	1	2	0	0	4	16 (9.14)
5	***Enterobacter* sp.**	4	0	0	0	0	0	1	0	0	3	8 (4.57)
6	***Proteus mirabilis***	1	0	0	0	0	0	1	0	1	0	3 (1.71)
7	***Citrobacter freundii***	2	0	0	0	0	0	0	0	0	0	2 (1.14)
8	***Serratia marcescens***	0	0	1	0	0	0	0	0	0	0	1 (0.57)
9	***Cedecea davisae.***	0	0	0	0	0	0	0	0	0	1	1 (0.57)
10	***Burkholderia cepacia***	0	0	0	0	0	0	0	0	0	1	1 (0.57)
11	***Pantoea agglomerans***	0	0	0	0	0	0	0	0	0	1	1 (0.57)
12	**Total**	58	2	2	1	1	3	25	4	7	72	175 (100%)

Table (3)
Detection of biofilm among pathogenic bacterial isolates.

No.	Bacteria species	Number of bacterial isolates (%)	Number of bacterial isolates biofilm formation (%)	Degree (%)			Resistance to tested antibiotics	
				Strong (%)	Moderate (%)	Weak (%)	Biofilm forming (%)	Non biofilm forming (%)
1	***Escherichia coli***	63 (36)	50 (79.0)	22.0	46.0	32.0	94.0	46.14
2	***Klebsiella pneumonia***	54 (30.85)	39 (72.15)	23.03	53.76	23.03	92.16	42.84
3	***Acinetobacter baummannii***	25 (14.28)	20 (80.0)	25.0	50.0	25.0	95.0	60.0
4	***Pseudomonas* sp.**	16 (9.14)	13 (81.25)	23.07	46.14	30.76	92.31	66.66
5	***Enterobacter* sp.**	8 (4.57)	6 (75.0)	16.66	66.64	16.66	83.34	50.0
6	***Proteus mirabilis***	3 (1.71)	3 (100)	33.33	66.66	0.0	100	0.0
7	***Citrobacter freundii***	2 (1.14)	1 (50.0)	0.0	100	0.0	100	0.0
8	***Serratia marcescens***	1 (0.57)	0.0	0.0	0.0	0.0	0.0	0.0
9	***Cedecea davisae.***	1 (0.57)	0.0	0.0	0.0	0.0	0.0	0.0
10	***Burkholderia cepacia***	1 (0.57)	0.0	0.0	0.0	0.0	0.0	0.0
11	***Pantoea agglomerans***	1 (0.57)	0.0	0.0	0.0	0.0	0.0	0.0
12	**Total**	175 (100%)	132 (75.4)	0.0	0.0	0.0	0.0	0.0

Table (4)
Production of siderophore by bacterial isolates.

No.	Bacteria species	Number of bacterial isolates (%)	Number of bacterial isolates produce siderophore (%)
1	***Escherichia coli***	63 (36)	52 (82.16)
2	***Klebsiella pneumonia***	54 (30.85)	49 (90.65)
3	***Acinetobacter baummannii***	25 (14.28)	18 (72)
4	***Pseudomonas* sp.**	16 (9.14)	16 (100)
5	***Enterobacter* sp.**	8 (4.57)	8 (100)
6	***Proteus mirabilis***	3 (1.71)	3 (100)
7	***Citrobacter freundii***	2 (1.14)	1 (50)
8	***Serratia marcescens***	1 (0.57)	0.0
9	***Cedecea davisae.***	1 (0.57)	0.0
10	***Burkholderia cepacia***	1(0.57)	0.0
11	***Pantoea agglomerans***	1 (0.57)	0.0
12	**Total**	175 (100%)	147 (83.93)

sp. (9.14%) *Enterobacter* sp. (4.57%) and *Proteus mirabilis* (1.71%). A previous study by Eldomany and Abdelaziz reported that *E. coli* was the main isolated Gram-negative bacteria from all clinical specimens with a frequency rate of (30%) followed by *P. aeruginosa* (24.5%) and *A. baumannii* (18.7%).[20] According to a study by Ashour and El-Sharif, the most frequent isolates among patients with leukemia and solid tumors were *K. pneumoniae* (31.2%) followed by *E. coli* (22.2%).[19] Gram-negative bacteria are the most common isolates from febrile neutropenic cancer patients with a frequency rate of 84.9% (180/212), *Escherichia coli* was the most frequently isolated bacterial species (38.68 %), followed by *Klebsiella* sp. (14.15%) and *Acinetobacter* sp. (11.32%).[21] Moreover, Trecarichi and Tumbarello reported that the rate of Gram-negative bacteria recovery from cancer patients ranged from 24.7% to 75.8%, and *E. coli* represented the most common species (mean frequency of isolation 32.1%).[22]

Our study showed that the susceptibility of Gram-negative bacilli to commonly used antibiotics was found to be highly resistant to ampicillin, cefazolin, ceftriaxone, ampicillin-sulbactam, and trimethoprim/sulfamethoxazole with a different ratio, but sensitive to amikacin and gentamicin with ratios of (57.67%) and (38.25%) respectively. Eldomany and Abdelaziz[20] reported that Gram-negative bacteria isolated from cancer patients were highly resistant to cefotaxime and ceftazidime. *Acinetobacter* sp. exhibited resistance percentages of 84.1% and 81.2% to cefotaxime and ceftazidime, respectively. The percentage of resistance to ceftazidime and cefotaxime was also high in *Klebsiella, Escherichia, Pseudomonas,* and *Enterobacter* species. The decreased susceptibility to most antibiotics tested including non-β-lactam antibiotics such as aminoglycosides (gentamicin) and quinolones (ciprofloxacin, levofloxacin) was observed in isolates of *Escherichia coli, Enterobacter, Klebsiella, Pseudomonas,* and *Acinetobacter* species.[20] Jacobson et al. showed that resistance among Gram-negative bacilli at their cancer canter increased to third-generation cephalosporins, quinolones, β-lactams, and aminoglycosides. They suggested that imipenem, meropenem, cefepime, and piperacillin/tazobactam were suitable choices for treating the infection of cancer patients.[23] On the basis that bacterial isolates were regarded as MDR when they were resistant to one or more antibiotics in three or more classes of antimicrobials, a significant proportion of bacterial isolates recorded in the present study are considered to be MDR bacteria. Previous studies reported MDR bacterial isolates with a ratio of 95%.[24] According to the data analysis of the (ESβLs) in our study, *E. coli* produced (ESβLs) with a percentage of 25.28% (n = 16), *Klebsiella pneumonia* 11.10% (n- = 6), and *Pseudomonas* sp. 25.0% (n = 4). A similar finding was reported by Garba et al., who reported that *Escherichia coli* is the highest producer of ESβLs enzymes from clinical samples followed by *Klebsiella pneumonia* and *Pseudomonas aeruginosa*.[25] Liu et al. reported that *Klebsiella pneumoniae* and *Escherichia coli* isolated from hospitals were the top two detected isolates that produced ESβLs with a rate of 14.1% and 68.2%, respectively.[26]

Biofilm is one of the most important virulence factors that helps bacterial species adhere to the living surface of host cells. Most of the biofilm-forming bacteria are resistant to antibiotics.[27] The development of microbial biofilms results in persistent infections and is a key contributor to treatment failure.[28] Our result elucidated that 75.4% of bacterial isolates have the ability to produce biofilm with different degrees, similar findings were reported by other studies.[29] Biofilm formation is an essential part of pathogenicity (or a combination of virulence and antibiotic resistance); thus, the formation is highly regulated by a complex network of transcriptional regulations.[30,31] Biofilm-related resistance to antimicrobial drugs is conferred by the physiological characteristics of biofilm organisms and the structure of biofilms.[32] Some of the possible reasons for the resistance of biofilms to antimicrobials are altered gene expression in biofilm-specific resistance genes (as efflux pumps), decreased sensitivity of most antibiotics to slower cell growth and decreased metabolic activity, degradation of antimicrobials, impaired antibiotic penetration into the biofilm matrix, the stress response to a hostile environment, and destruction of antibiotics by enzymes in the biofilm matrix.[33,34] Greater horizontal gene transmission occurs in biofilm-grown bacteria than in planktonic bacteria.[35] Biofilms experience a substantially higher rate of mutation than planktonic cells do.[36] Through the use of virulence factors and antibiotic-resistant genes from resistant to susceptible bacterial species, biofilms increase the likelihood of gene transfer and the development of antibiotic resistance.[37,38] Bacteria forming biofilms show resistance

against both the human immune system and antibiotics.[39] Dumaru et al. found a significant association between ESβL production and biofilm formation to antibiotic resistance.[40] In our study, the bacterial isolates' biofilm formation is highly resistant to antibiotics compared with non-biofilm formation. The biofilm-forming bacteria were more multidrug-resistant and produced more virulence genes compared with the non-biofilm-forming bacteria.[41] Infections related to biofilms can be roughly categorized into two categories. Infections linked to indwelling medical devices[42,43] and native biofilm infections of host tissue.[44] The biofilm that first developed on medical implants can result in infections of the urinary tract and bloodstream.[45] Chronic infections of the host tissue that are caused by biofilms include cystic fibrosis patients' chronic lung infections, chronic wounds, chronic otitis media, recurrent UTIs, chronic osteomyelitis, endocarditis, and others.[44]

In this study, the majority of bacterial species produce siderophores. A previous study in India examined 200 uropathogenic *Escherichia coli* isolates that were separated from UTI patients. The isolates were screened using the Chrome azurol assay (CAZ) for siderophore production, and the results showed that 95% of the isolates produced siderophores.[46] *Acinetobacter baumannii* and other bacteria require siderophores to adhere to surfaces and synthesize extracellular polysaccharides, which are necessary to produce biofilms and microbial communities that exhibit enough iron mutualism.[47] Pyoverdine and pyochelin are the two siderophores that *Pseudomonas aeruginosa* makes; the production of pyochelin is linked to biofilm development, a characteristic of *P. aeruginosa* that causes persistent lung infections in cystic fibrosis patients.[48] The virulence factors comprise a broad range of elements such as protective capsules, bacterial toxins, adhesion factors, and siderophores that participate in the colonization of pathogenic microbes in the host and increase the severity of the disease. The ability of bacteria species to develop strategies to acquire Fe from their host, which includes the expression of siderophores taking Fe from proteins bound to the host Fe. The absence of the genes regulating siderophore expression or other Fe harvesting systems is usually linked with decreased virulence of pathogens involving infectious bacteria.[49] In actuality, the bacterial strains that can produce additional levels of siderophores are highly virulent, and those incapable of secreting and synthesizing siderophores are less capable of virulence and colonization through infection.[50]

5. Conclusion

We can conclude that cancer patients have a significant distribution of multidrug-resistant pathogenic bacteria. Bacterial strains isolated from cancer patients are highly resistant to most available antibiotics, forming biofilm, producing siderophores, and producing ESβLs enzymes. These enzymes and other resistance traits give strong testimony to the resilience of bacteria and their ability to adapt. Treatment of highly resistant bacterial strains demands a multifactorial approach combining continued research, development of novel antibiotics, more prudent use of existing agents, and a continuous emphasis on more effective infection control measures.

Authors contribution statement

Gamal M. El-Sherbiny: conceptualization, formal analysis supervision, writing - review & editing. **Eman E. Farghal**: conceptualization, formal analysis, methodology, writing – review, and editing. **Mohamed K. Lila**: methodology, investigation. **Yousseria M. Shetaia**: methodology, data curation writing - review & editing. **S.S. Mohamed:** methodology, resources, writing and review **Marwa MF Elswify**: methodology, resources, visualization, data curation.

Ethics approval and consent to participate

All samples collected were approved by the Ethics Committee of the National Cancer Institute, No. 201617025.3 Cairo University, Cairo, Egypt.

Human participants and/or animals

N/A.

Informed consent

N/A.

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Declaration of competing interest

We declare that we have no conflict of interest.

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