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A one-year study of bacterial isolates causing Healthcare Acquired Infection (HAI) and their antibiotic resistance pattern at a private hospital in Egypt

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Abstract

The present study included 210 inpatients, out of them 180 were diagnosed as suspected cases of sepsis in the period over 12 months from December 2018 to December 2019, at a private hospital in Cairo, Egypt. Out of 210 investigated cases 180 (85.7%) were infected. Single bacterial isolates were recovered from 120 cases (66.7%), while 60 (33.3%) cases were found to be carrying more than one bacterial isolate. The results of the current study revealed that 34.4% (62/180) of isolates were Gram-positive and 65.6% (118/180) were Gram-negative.

In the current study, *Staphylococcus aureus* was mostly isolated from burn unit (55.5%), meanwhile, *Staphylococcus epidermidis* was the greatest species isolated from otolaryngology department (30.0%). However, *Pseudomonas aeruginosa* was mostly isolated from surgery department (62.5%), *Acinetobacter baumannii* was mainly isolated from pediatrics department (50%), *Klebseilla pneumonia* and *Escherichia coli* isolates were the lowest number of bacterial pathogens recovered from the examined clinical specimens. *Klebseilla pneumonia* was mostly isolated from urology department (31.6%) and, *Escherichia coli* isolates were recovered only from urology department (13.2%). All of the percentages were correlated to total number of bacterial isolates isolated from each department.

The order of antibiotic efficacy among the different bacterial isolates in this study was quite different. Most bacterial isolates recovered from different hospital departments were multi drug resistant. The highest resistance percentages were observed with ampicillin. The greatest resistance to ampicillin was *Pseudomonas aeruginosae* (96.6%), similarly highest resistance to Amoxicillin/clavulanate were recorded with *Pseudomonas aeruginosae* (86.2%). Cefotaxime resistance more or less resembled Amoxicillin/clavulanate with *Pseudomonas aeruginosae* (86.2%).

Concerning ciprofloxacin resistance; *Acinetobacter baumannii* showed greatest resistance (80%), while all *Escherichia coli* isolates were sensitive to ciprofloxacin. Vancomycin was the most effective antibiotic tested against the isolated bacteria. All *Staphylococcus aureus*, *Klebseilla pneumonia* and *Escherichia coli* isolates were (100%) sensitive to vancomycin. Meanwhile, 48.3% of Pseudomonas aeruginosae isolates were resistant to vancomycin. Resistance to azithromycin was highest among *Pseudomonas aeruginosae* isolates (34.5%), but all *Escherichia coli* isolates were sensitive to azithromycin.

Rifampicin resistance of the tested bacterial isolates showed highest percentage with *Pseudomonas aeruginosae* isolates with 51.7%, all *Escherichia coli* isolates were sensitive to rifampicin. The frequencies of the aminoglycoside gentamycin resistance were similar to previous results as *Pseudomonas aeruginosae* showed greatest percentage (82.8%). Resistance to chloramphincol were mostly found in *Acinetobacter baumannii* isolates (75%). Finally, the resistance of all bacterial isolates to tetracycline was also similar to previous results as *Acinetobacter baumannii* had the greatest resistance (80%). High antimicrobial consumption, illustrates the urgent need for antimicrobial stewardship programs and also infection control programs. The ratios above were used to calculate the corresponding susceptibility percentage for every species to every used antibiotic to make an antibiogram to facilitate the empiric antibiotic prescription by physicians and as the first step towards antibiotic stewardship.

Introduction

World Health Organization defines healthcare associated infection as 'an infection occurring in a patient during the process of care in a healthcare facility which was not present at the time of admission. Whilst this is similar to the definition of hospital-acquired infection, it importantly also includes patients in long-stay residential or nursing homes' [1].

Healthcare associated infections are infections that first appear three days or more after hospitalization or within 30 days after having received health care. Multiple studies indicate that the most common types of adverse events affecting hospitalized patients are adverse drug events, healthcare associated infection, and surgical complications [2][3].

The US Center for Disease Control and Prevention identifies that nearly 1.7 million hospitalized patients annually acquire healthcare associated infections while being treated for other health issues and that more than 98,000 of these patients (one in 17) die due to healthcare associated infections. The Agency for Health care Research and Quality reported that healthcare associated infections are the most common complications of hospital care and one of the top 10 leading causes of death in the USA. Out of every 100 hospitalized patients, seven patients in advanced countries and ten patients in emerging countries acquire healthcare associated infections [4][5].

Other studies conducted in high-income countries found that 5%–15% of the hospitalized patients acquire healthcare associated infections which can affect from 9% to 37% of those admitted to intensive care units [6].

Multiple research studies report that in Europe hospital-wide prevalence rates of healthcare associated infections range from 4.6% to 9.3%. The WHO reports however that healthcare associated infections usually receive public attention only when there are epidemics [7][8]. Health care associated infections also have impact on critically ill patients with around 0.5 million episodes of healthcare associated infections being diagnosed every year in intensive care units alone. Intensive care units' patients are often in a very critical, immuno-compromised status, which increases their susceptibility to healthcare associated infections [9].

Residual antibiotics in the freshwater are possibly from different sources such as agriculture, wastewater discharge and animal farming. The residual antibiotics serve as the selective pressure and main concern for the development of antibiotic resistant bacteria. A study was done in Malaysia to identify the antibiotic resistant bacteria present in selected rivers, found that fresh river water contained antibiotic resistant bacteria [10].

Aim of the work

The present study has been undertaken to investigate the following: Study of prevalence of bacteria associated with hospital acquired infections among hospital staff, patients and medical instruments at a private hospital during the period from December 2018 to December 2019; Study of antibiotic resistance pattern of the isolated bacteria to the commonly prescribed antibiotic; Analysis of the results in order to know the main sources of infection.

Materials and methods

Clinical specimens

Over a period of 12 months between December 2018 to December 2019 a total of 210 samples as shown in tables (1 and 2) were collected from inpatients, 10 samples from nurses, 10 samples from physicians and 20 samples from medical instruments with clinical suspicion of sepsis at different clinical departments. Hospital management approval was obtained prior to conducting of this study. No disclosure of patients' information was done; hence no personal consents were needed. Approval of hospitals ethical committee for the present study was obtained.

Specimen collection, transport and processing

Using sterile cotton swabs, a deep surface swab was taken from septic wounds. Swabs were transported in sterile tubes containing 2 ml of trypticase soy broth and incubated aerobically for 18 hours at 37 ºC. The broth was sub cultured onto MacConkey's agar, Mannitol Salt Agar, Blood Agar, and Cetrimide Agar plates, and incubated aerobically at 37°C for 24-48 hours. After incubation, plates were examined for growth of colonies. Each pure colony was subjected to Gram stain and biochemical reactions, for isolation and identification of isolates, using API 20E Multi Test (Analytic Profile Index) System.

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Table 1: Distribution of clinical samples among different departments of a private hospital

Department	Number of samples			
	Patients	Physician	Nurses	Instruments and hospital environment
Burn Unit	18	1	1	3
Intensive care unit	30	2	2	3
Urology department	24	1	1	2
Surgery department	48	2	2	5
Otolaryngology	20	1	1	2
Pediatrics	26	1	1	2
Internal medicine	44	2	2	3
Total	210	10	10	20
Total number samples	250			

Table 2: Source and number of instruments and hospital environment samples collected

Source	Site	Number of samples
Burn Unit	Patient's beds	3
ICU	Ventilators	3
Urology department	Casters	2
Surgery department	Surgical instruments	5
Otolaryngology	Tongue depressor	2
Pediatrics	Patient's beds	2
Internal medicine	Patient's beds	3
Total	20	

Antimicrobial sensitivity tests

The antimicrobial susceptibility tests of the isolated strains were performed by the Disk Diffusion Method Mueller-Hinton agar plates according to: National Committee For Clinical Laboratory Standards (NCCLS) [11].

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Table 3: Antimicrobial discs; Their abbreviations and concentrations (Oxoid, UK)

Antimicrobial	Code	Disc concentration (µg)
Ampicillin	AMP	10
Amoxicillin/clavulanate	AMC	30
Cefotaxime	CTX	30
Ciprofloxacin	CIP	5
Vancomycin	VA	30
Azithromycin	AZM	15
Rifampicin	RD	5
Chloramphincol	CHA	30
Gentamicin	GN	10
Tetracycline	TET	10

Results

The present study included 210 inpatients, out of them 180 were diagnosed as suspected cases of sepsis in the period over 12 months from December 2018 to December 2019, at a private hospital in Cairo, Egypt. In the current study there was no pathogenic bacterial species isolated from the examined physician or nurses, but 50% of the examined instruments were contaminated, Table 4.

Table 4: The incidence of bacterial isolation from physician, nurses and medical instruments

	Total number	No. of suspected sepsis	%
Physician	10	0.0	0.0
Nurses	10	0.0	0.0
Instruments	20	10	50.0

N.B. % was correlated to the total number of each group.

Out of 210 investigated cases 180 (85.7 %) were infected. Single bacterial isolates were recovered from 120 cases (66.7%), while 60 (33.3%) cases were found to be carrying more than one bacterial isolate, table 5.

Table 5: Frequencies of bacterial isolates among different clinical departments

Sex	No. of cases carrying single bacterial isolate	%*	No. of cases carrying more than bacterial isolate	%*
Male	90	81.8	20	18.2
Female	50	71.4	20	28.6

*% was correlated to the total number of male or female patients.

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Concerning the frequencies of Gram-positive and Gram-negative isolates; the results of the current study revealed that 34.4% (62/180) of isolates were Gram-positive and 65.6% (118/180) were Gram-negative as described in table 6.

Table 6: Frequencies of Gram-positive and Gram-negative isolates

Microorganism	Number	%*
Staphylococcus aureus	42	23.3
Staphylococcus epidermids	20	11.1
Pseudomonas aeruginosa	58	32.2
Acinetobacter baumannii	40	22.2
Klebseilla pneumonia	15	8.3
Escherichia coli	5	2.8

*Percentage was correlated to total number of isolates (180).

Frequencies of each isolate among different hospital department acquired infections

Investigation of the cases with established burn infections revealed that *Staphylococcus aureus* 55.5% was the most recovered pathogen from burn department followed by *Pseudomonas aeruginosa* 27.8% and *Staphylococcus epidermidis* 16.7%. Regarding to surgery department infected surgical wounds were mainly caused by *Pseudomonas aeruginosa* 62.5% were the most prevalent pathogen followed by *Staphylococcus aureus* 25.0% (12/157) and *Acinetobacter baumannii* 12.5%. On the other hand, *Acinetobacter baumannii* were the prevalent pathogen causing infection among patients at intensive care unit 37.5% followed by *Pseudomonas aeruginosa* 33.3%, *Staphylococcus aureus* 20.8% and *Staphylococcus epidermidis* 8.3%.

Patients suffering from urinary tract infection the prevalent pathogen causing infection were *Klebseilla pneumonia* 31.6%, *Staphylococcus aureus* 26.3%, *Pseudomonas aeruginosa* 21.1%, *Escherichia coli* 13.2% and *Acinetobacterbaumannii* 7.9%. Concerning infection among patients in otolaryngology department the most common isolated bacterial pathogen was *Acinetobacterbaumannii* 50.0%, followed by *Staphylococcus epidermidis* 30.0% and *Pseudomonas aeruginosa* 20%. Also, *Acinetobacter baumannii* was the most isolated pathogen causing infection in pediatric patients 53.8%, followed by *Staphylococcus epidermidis* 26.9% and *Staphylococcus aureus* 19.2%.

Finally, the common bacterial isolates recovered from internal medicine department patients were in descending order, *Pseudomonas aeruginosa* 41.6%, *Staphylococcus epidermidis* 20.8%, *Acinetobacter baumannii* 16.7%, *Klebseilla pneumonia* 12.5% and *Staphylococcus aureus* 8.3%.

(N.B.- Percentage was correlated to total number of bacterial isolates isolated from each department).

Resistance patterns of all bacterial isolates

Most bacterial isolates recovered from different hospital departments were multi drug resistant. The highest resistance percentages were observed with ampicillin. The rates of resistance to ampicillin were as follow; *Pseudomonas aeruginosae* (96.6%), *Acinetobacter baumannii* (90%), *Klebseilla pneumonia* (86.7%), *Staphylococcus aureus* (83.3%), *Staphylococcus epidermidis* (70%), and *Escherichia coli* (40%). Also resistance to Amoxicillin/clavulanate were recorded in descending order starting with *Pseudomonas aeruginosae* (86.2%), *Acinetobacter baumannii*(80%), *Klebseilla pneumonia* (73.3%), *Staphylococcus epidermidis* (60%), *Staphylococcus aureus* (59.5%) and *Escherichia coli* (20%).

Cefotaxime resistance more or less resemble Amoxicillin/clavulanate; the resistance pattern was as follow; *Pseudomonas aeruginosae* (86.2%), *Acinetobacter baumannii* (80%), *Staphylococcus aureus* (59.5%) *Staphylococcus epidermidis* (55%), *Klebseilla pneumonia* (53.3%) and *Escherichia coli* (20%). Concerning ciprofloxacin resistance; *Acinetobacter baumannii* (80%), *Pseudomonas aeruginosae* (69%), *Staphylococcus epidermidis* (40%), *Klebseilla pneumonia* (13.3%) and *Staphylococcus aureus* (9.5%). On the other hand, all *Escherichia coli* isolates were sensitive to ciprofloxacin. Vancomycin was the most effective antibiotic tested against the isolated bacteria. All *Staphylococcus aureus, Klebseilla pneumonia* and *Escherichia coli* isolates were (100%) sensitive to vancomycin. Meanwhile, 48.3% of *Pseudomonas aeruginosae* isolates, 25% of *Acinetobacterbaumannii* and 5% of *Staphylococcus epidermidis* were resistant to vancomycin.

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Resistance to azithromycin was in the following order; *Pseudomonas aeruginosae* (34.5%), *Staphylococcus aureus* (28.6%), *Acinetobacter baumannii* (25%), *Staphylococcus epidermidis* (20%) and *Klebseilla pneumonia* (6.7%). However, all *Escherichia coli* isolates were sensitive toazithromycin.

Resistance to rifampicin was illustrated in. *Pseudomonas aeruginosae*, *Acinetobacter baumannii*, *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Klebseilla pneumonia* isolates were 51.7%, 37.5%, 31%, 25% and 13.3% respectively. However, all *Escherichia coli* isolates were sensitive to rifampicin.

The results of the current study concluded that bacterial isolates resistant to chloramphincol were in the following descending order; *Acinetobacter baumannii* (75%), *Pseudomonas aeruginosae* (55.2%), *Staphylococcus aureus* (50%), *Staphylococcus epidermidis* (30%), *Escherichia coli* (20%) and *Klebseilla pneumonia* (13.3%).

Resistance to the aminoglycoside, gentamycin wasas following; *Pseudomonas aeruginosae* (82.8%), *Acinetobacter baumannii*(55%), *Staphylococcus aureus* (52.4%), *Staphylococcus epidermidis* (40%), *Escherichia coli* (40%) and *Klebseilla pneumonia* (20%).

Finally, the resistance of all bacterial isolates to antibiotic tetracycline was as follow; *Acinetobacter baumannii* (80%), *Pseudomonas aeruginosae* (65.5%), *Staphylococcus epidermidis* (45%), *Staphylococcus aureus* (36%), *Klebseilla pneumonia*(26.7%) and *Escherichia coli* (20%).

Discussion

Healthcare associated infections are a common cause of morbidity and mortality all over the world and are among the most common adverse events in healthcare. Infectious agents causing healthcare associated infections may come from endogenous or exogenous sources [11]. Recently, new emphasis on healthcare associated infections as a patient safety and public health problem has underscored the need for systematic healthcare associated infections surveillance as part of a broad-based prevention and control strategy [12].

Healthcare associated infections are additional burdens on individual hospitals and health care systems. They can increase the costs of patient care from several economic perspectives, (e.g. health insurance companies). Excess costs of healthcare associated infections are related to additional diagnostic tests and treatment, additional hospital days, and post discharge complications, among others. Quantifying the exact economic burden attributable to healthcare associated infections still remains a challenging issue [13].

There are several risk factors which are associated with healthcare associated infections such as poor hygiene, interacting with people especially with patients without washing hands, prolonged stay in the hospital wards, and lack of awareness regarding risk factors, prevention, and treatments [14]. The variation in incidence of healthcare associated infections between developing countries could be due to variation in the technical advancement of health facilities, study designs and diagnostic criteria of sepsis, while the low incidences in developed countries reflect the high standard measure of health care, hospital services and the quality of life.

The most common pathogens implicated in intensive care unit were Gram-negative infections, Gram-positive infections were less compared to Gram-negative infections, conversely, other studies reported that Gram-positive cocci are the most common pathogenic isolate in intensive care unit especially in developed countries. This can be explained by the fact that causative agents vary with the geographical area and time [15].

Data presented in this study indicate that the *Pseudomonas aeruginosa* was the most common organism observed among the patients of surgical wards followed by *Staphylococcus aureus,* similar finding have been seen in Philippines [16] and Turkey [17].

Infection can lead to deterioration of the wound healing process and severe systemic complications and is the leading cause of morbidity and mortality in patients with burns. Therefore, knowledge of the bacteriology of burns is of prime importance to fast and clinically sound therapeutic decisions in critically ill burn patients. In the current study, investigation of the cases with established burn infectionsrevealed that, the three most frequent nosocomial pathogens in our Burns Clinic for the study period were *Staphylococcus aureus* (55.5%), *Pseudomonas aeruginosa* (27.8 %).

Many of the published data indicate the leadership of *Staphylococcus aureus* as a nosocomial pathogen but in most Asian and Arab countries *Acinetobacter baumannii* still prevails. These differences may be due to different local conditions, such as climate, topical and systemic treatment regimens, sampling procedures, infection prevention protocols as well as the study period [18]. However, in many cases the isolation of *Acinetobacter baumannii* was due to room contamination rather than infection [19].

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In the present study, patients suffering from urinary tract infection, the most prevalent pathogens causing infection were *Klebseilla pneumonia* (31.6%). Our findings, concerning the common pathogens isolated from urinary tract infected patients, were more or less different than those isolated from other researches were, *Escherichia coli* has been found as the most common uropathogenic bacteria in different seasons during the two-year surveillance. The second most common uropathogenic agent has been shown to be *Klebsiella pneumonia* [20]. We suggest that these categories of studies should be done at regular intervals to follow any changes in the pathogenic agents' patterns.

Ear, nose and throat infection is a more frequent treatable health care problem worldwide, yet if left untreated; it can cause a serious complication such as a speech disorder, pain in patients and their family quality of life, and economic burden on the health care system. The burden and prevalence of ear, nose and throat infection are more intense in developing countries due to the poor living standard and hygienic conditions along with lack of proper nutrition [21].

In the current study we found that infection among patients in otolaryngology department, the most common isolated bacterial pathogen was *Acinetobacter baumannii* 50.0%, Kumar reported that from Gram-negative bacteria spp., Proteus spp. where responsible for most cases of ear, nose and throat infections, whereas Edwin find out that Pseudomonas spp., Proteus spp. and Klebsiella spp., are the common bacteria that cause ear, nose and throat infection in Japura India, similarly Argaw-Denboba find out that *Pseudomonas aeruginosa*, Klebsiella spp. *Escherichia coli* are the bacteria from Ethiopia, Nigeria, and Egypt [22][23].

Infection in children remains a significant cause of morbidity and mortality worldwide. Sepsis can be defined as the body's response to an infection. It is a potentially fatal, whole-body inflammation. Severe sepsis is complicated by organ dysfunction. Bacteria are the main cause of sepsis; however, even fungi, viruses, and parasites in the blood, urinary tract, lungs, skin, and other tissues can cause sepsis. The frequency of pediatric sepsis was found to be greater than 42,000 cases annually, with an associated mortality rate of 10% in the United States [24].

The common sources of infection for sepsis in the study were pneumonia, meningitis, and respiratory tract infections in pediatrics as well as neonates. Inwald conducted studies in tertiary care hospitals in the United States, finding that pneumonia is the most common type of community and hospital acquired infection in pediatrics. It has a significantly high rate compared to other sources of infection, such as meningitis, urinary tract infection, etc., because almost all pediatric admissions undergo ventilation and catheterization (like tracheostomy), which creates an access point for microorganisms to infect the lungs [25].

The results of the present study raveled that *Acinetobacter baumannii* was the most isolated pathogen causing infection in pediatric patients 53.8%, in a study performed in India, the most commonly isolated pathogens *Klebsiella pneumoniae*, *Escherichia coli*, and *Staphylococcus aureus* were more isolated in number [26].

These results were in agreement with other previous studies in other countries, in India, Tiwari et al., mentioned that Gram-negative bacteria represented about (71.87%) of total isolates and Gram-positive bacteria accounted for (28.13%) in their study [27].

One of the objectives of the current study is to gain knowledge about antimicrobial resistance patterns of the isolated bacterial isolates which help us to optimize treatment and decrease mortality rates. Many of these microorganisms that are resistant to antibiotics and can easily spread by hospital personnel. Guidelines for antibiotic therapy can be helpful for clinicians to select more appropriate antibiotics for effective treatment and prevent the development of drug resistance.

This study showed the distribution of antibiotic resistance of bacterial species associated with nosocomial infections at a private Hospital in Cairo, Egypt, and the results reveled that they have become multi-resistant to these therapeutic agents, thus rendering these drugs ineffective as treatments of choice for infections caused by these pathogens. The results of the study indicate the importance of developing policies and regulations for antibiotic use at the country level, implementing antibiotic stewardship programs to promote appropriate use of antibiotics, and increasing the awareness of clinicians and the public on rational use of antibiotics.

Extensive use and misuse of antibiotics in medication, veterinary, agriculture and aquaculture have caused antibiotic-resistant bacteria to be widespread. Most bacterial isolates recovered from different hospital departments were multi drug resistant. The highest resistance percentages were observed with ampicillin. The rates of resistance to ampicillin agree with other results recorded in other countries [28][29].Also, resistance to Amoxicillin/clavulanate were similar to results obtained by Leflon-Guibout et al. [30].

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Ciprofloxacin is a synthetic broad-spectrum fluoroquinolone antibiotic. Ciprofloxacin binds to and inhibits bacterial DNA gyrase, an enzyme essential for DNA replication. The results of other studies showed resistance to ciprofloxacin but to less extent than that detected in the current study this may be attributed to a policy of the rational use of antimicrobials [31][32].

The results of this study proved that vancomycin was the most effective antibiotic tested against the isolated bacteria. Same conclusion was reported by Manav Khera et al. [33].

The susceptibility pattern of the isolated microorganisms in this study proved that the resistance to azithromycin All *Escherichia coli* isolates were sensitive to azithromycin, these results are quite different with results obtained in other research especially with *Escherichia coli* isolates [34].

The results of the current study concluded that bacterial isolates resistant to chloramphincol agree with that obtained by Hongyue Dang, et al. [35].

The results of the current study concerning gentamycin resistance, agree with a community setting, a study was performed earlier in Spain. The study showed that despite 4 years of official banning of antibiotic growth promoters in animals, enterococci isolated from food handlers were more resistant than those from healthy volunteers [36]. Another noteworthy study performed by Don-abedian et al. showed a commonality of gentamicin-resistant determinants and gentamicin resistant enterococcal isolates among humans, food, and food-producing animals over a broad geographical area [37].

Tetracyclines are generally used in the treatment of infections of the urinary tract, respiratory tract, and the intestines and are also used in the treatment of chlamydia, especially in patients allergic to β-lactams and macrolides; however, their use for these indications is less popular than it once was due to widespread development of tetracycline-resistant isolates of clinically important bacteria [38].

Finally, in the present study, all bacterial isolates isolated from different clinical departments were tested for their susceptibility to tetracycline and the results revealed their resistance to tetracycline. This finding more or less similar to the results reported in many other researches [39] [40].

Conclusion

This unnecessary exposure to antibiotics by large numbers of people may lead to increased prevalence of antibiotic-resistant bacteria in the community. Thus, education of the public, health authorities, and clinicians is needed now more than ever to eliminate home stockpiling of antibiotics, to ensure the correct use for all bacterial infections, and to prevent use when antibiotics are not needed, such as use for treatment of viral infections.

Conflict of interest

The author declares no conflict of interest.

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