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Original Research Article

Statistical Study of Cholera Outbreak According to Gender, Age and Antibiotic Sensitivity in Baghdad Province, Iraq for a Period During 2022

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Abstract: ***Objective***: The gram-negative bacterium *Vibrio cholera* is the cause of the acute infectious bacterial illness cholera. It often spreads via ingesting contaminated food or water. Diarrhea and severe dehydration are symptoms of cholera. It can be lethal within hours if untreated. A fresh cholera outbreak struck many Iraqi cities in 2022 based on the World Health Organization's verified data. ***Methodology***: A total 32 instances in all were recorded in the Medical City in Baghdad between June and November 2022, with the causal agent being found in stool samples from individuals who had been diagnosed as being infected. Distribution of the collected data is based on age and gender. Additionally, the isolated bacteria were tested for susceptibility to a variety of antibiotics, including Ciprofloxacin, Tetracycline, Ampicillin, Trimethoprim, and Supremethoprim. Statistics were applied to the obtained data. ***Results***: Male were shown to have 55% more infections than female, who had 45%. Regarding demographics, it mostly targets the 22–42 age bracket. The greatest instances of cholera occurred in July and August, with the infection rising in the summer more than in the autumn. Statistics revealed that each antibiotic's sensitivity and resistance regardless of the patient's age and gender. The bacterial isolates were sensitive to Ciprofloxacin and Tetracycline. And resistant to other tested antibiotics. ***Conclusion***: The antibiotic sensitivity of cholera was regardless to age and gender. Further studies needed to study the disease distribution according to geographical area in Baghdad as well as study the medical profile of infected people. Study sensitivity of the infected bacteria to other recent antibiotics is also recommended.

Keywords: Baghdad, disease, medical profile, population, sensitivity, Cholera.

INTRODUCTION

Essentially, cholera is an enteric bacterial infection caused by vibrio cholerae, a flagellated bacterium. Consuming vegetables that are contaminated with the pathogen or drinking water that contaminated with the pathogen causes infection. Poor sanitation, lack of awareness, and/or lack of access to clean drinking water make the disease particularly prevalent in developing countries (Ali *et al*., 2015).

The major symptoms of the disease include severe watery diarrhea, or "rice-watery" stool, odorless or with a mild fishy smell (Endris *et al*., 2022). Since 2020, 26 countries have reported cases of cholera with 857 deaths (WHO 2021). In Iraq, the first recorded cholera epidemic occurred in Basra in 1820, causing a large number of deaths (Al-Abbassi *et al*., 2005). Another study reported that 4,667 cases of cholera were reported in Iraq in 2007; 136 (2.9%) were recorded in Baghdad's capital city, and 33% of the confirmed cases were aged 0–4 years and 15–45 years (Khwaif *et al*., 2010). Different cities in Iraq were affected by a new cholera outbreak in 2022. A total of 449 confirmed cases of cholera were reported in Iraq in 2018, according to WHO data (Sabir *et al*., 2023).

As part of its pathogenesis, V. cholera infects and colonizes the epithelium of the small intestine. Virulence factors expressed by V. cholera in the intestine, such as toxin coregulated pilus (TCP) and cholera toxin (CT), are essential to colonize the host and cause enterotoxicity (Jameel *et al*., 2016).When patients experience severe dehydration due to

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diarrhea caused by V. cholera, they get intravenous fluid replacement or oral rehydration fluid therapy. Even though using antibiotics to treat cholera can result in the evolution of resistance, they are not a necessary element of the treatment (Jameel *et al*., 2016).

Researchers also confirmed the use of antibiotics like trimethoprim-sulfamethoxazole, tetracycline, erythromycin, azithromycin, and ampicillin, which were very helpful to decrease cholera diarrhea duration (Saleh *et al*., 2011). A variety of diagnostic laboratory tests are applied to confirm V. cholera infection (Buss *et al*., 2015; Keddy *et al*., 2013). Including stool culture, biochemical test, serological test, specialized microscopy which include polyvalent, Inaba antisera testing, Ogawa, and single plex PCR, conventional coupled PCR or real-time PCR with fluorescence; to identify the toxin-encoding genes (Koskela *et al*., 2009).

We report the analysis of cholera outbreak data and antibiotic sensitivity of the pathogen in a group of hospitals in the Medical City, including the Baghdad Teaching Hospital, Al-Talmia Laboratories, Al-Ghazi Al-Hariri Hospital, and the Digestive System Hospital.from20thJune to 9th November 2022.

MATERIALS AND METHODS

Epidemiological Data Collection and Statistical Analysis

Epidemiological data obtained from hospitals in the Medical City, including the Baghdad Teaching Hospital, Al-Talmia Laboratories, Al-Ghazi Al-Hariri Hospital, and the Digestive System Hospital. A 32 suspected cholera stool specimens were collected from both male and female patients in different ages between June and November 2022. For bacterial growth enrichment, the samples were examined microscopically and immediately placed in alkaline peptone water (APW; BiomarkTM, India) at 8.5 pH and incubated at 37 C for 6 hrs. The cells were streaked onto TCBS, MacConkey agar, and blood agar (BiomarkTM, India) using an inoculating loop and cultured at 37°C for 18 to 24 hours. A biochemical diagnosis was made after bacterial isolation (Jameel *et al*., 2016). A Kirby-Bauer susceptibility test was conducted on all bacterial isolates, using the disk diffusion technique. It was conducted on Hinton-Muller agar (Samanta *et al*., 2015). Antibiotics used include Ciprofloxacin, Tetracycline, Ampicillin, Trimethoprim, and Supremethoprim.

In order to analyze data statistically, the collected data including sensitivity to the selected antibiotics distributed based on age and gender. According to age, they were divided into adults and children.

RESULTS AND DISCUSSION

Statistical data showed that it affects males more than females Males 55% and females 45%. As for age, statistical data showed that it is more targeted at the age group (22_42). Cholera infection increases in summer more than in autumn, and the most cases were in July and August.

Figure 1A shows the percentage of adults to children within the study symbols. It can be seen that the adults are about 75% of the overall symbols while the children are about 25%. Figure 1, b shows the percentage of each age quarter, it is obvious that the study symbols have 48% of people in the ages of 0-25 years old about half the symbols while the people of midges (26-50 years old) are 39% finally the people over51 years old are 13% of the overall symbols. The dataset consisted of 31 symbols to study the sensitivity and resistivity of both males and females within a range of ages between 2 years to 80 years. Additionally, we found that males were slightly more likely to contract cholera than females; those aged 20–44 were more likely to contract it

A major cause of cholera disease outbreaks probably a lack of clean water and sanitation during the summer months. Because of the hot weather, Individuals are consuming more water, which can be contaminated, in summer days to gain the lost fluids, increasing the microorganisms growing chance on food and causing food poisoning (Anderson *et al*., 2020). Further, hot air and low water levels, which create the perfect environment for bacterial development, are probably the causes of summertime cholera epidemics (Asadgol *et al*., 2019).

They are known as a food-borne and waterborne infection , and are more prevalent in summer when there is a shortage of water, higher water consumption, outdoor activities and recreational water use such as camping or swimming, subsequently transfer diseases from person to person and might intensify enteric disease outbreaks (Stuart *et al*., 2003). Many parts of Iraq have suffered from these conditions after 2003,including inadequate environmental hygiene, combined with unique occurrences like hunger, flooding, and refugee migrations, who live in congested camps that may contribute to the rise of enteric diarrheal illnesses like cholera (Calzada *et al*., 2005). In 2009 and 2014, studies found that cholera usually affects children between 1 and 5 years of age and sometimes neonates between 2 and 30 days of age (Jameel *et al*., 2016). It's important to note that children are less likely to have cholera, perhaps because secretory antibodies in mother's milk provide protection (Qureshi *et al*., 2006). However, there have been theories that suggest inadequate sanitation and education for children under five may be to responsible for cholera cases (Faulkner *et al*., 2003).

On the other hand, because they have acquired immunity via repeated exposure to infections, older individuals appear to have lower incidence of cholera infection, whether they are symptomatic or asymptomatic. Additionally, since the gut mucosa plays a role in local immune protection, cholera infections are uncommon to reoccur. Conversely, the damage of infrastructure carried by the various crises Iraq has experienced, as well as natural disasters, hunger, and war, as well as the forced relocation of people into overcrowded and subpar camps for internally displaced people or refugees , these factors have contributed to the diffusion of cholera infection in Iraq, a disease that represents a threat to other nations as well since bacterial infections can spread through birds, the air, food, and waterways (Slanthia and Bansal, 1999; Khwaif *et al*., 2010).

In terms to gender, it might be due to the fact that individuals at this age, particularly males, eat more often outside, increasing the risk that they will interact with infections like V. cholerae. According to a study on a cholera outbreak in Uganda in 2011, male cholera cases were 1.6 times higher than female cases (Bwire *et al*., 2017). Ten cholera outbreaks were studied in Ugandan fishing communities from 2011 to 2015, and the male to female case-fatality ratio was much greater (Bwire *et al*., 2017; SABIR *et al*., 2023).

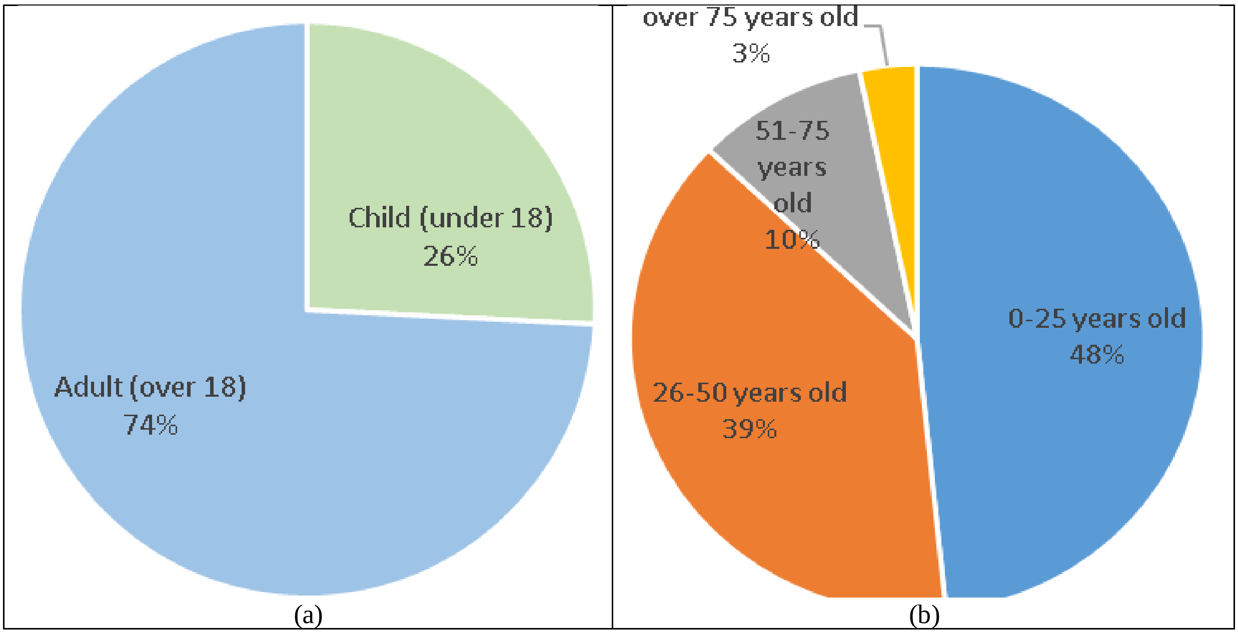

Figure 1: a) The testing data distributed according to the childhood age, b) The testing data distributed according to the ages quarters

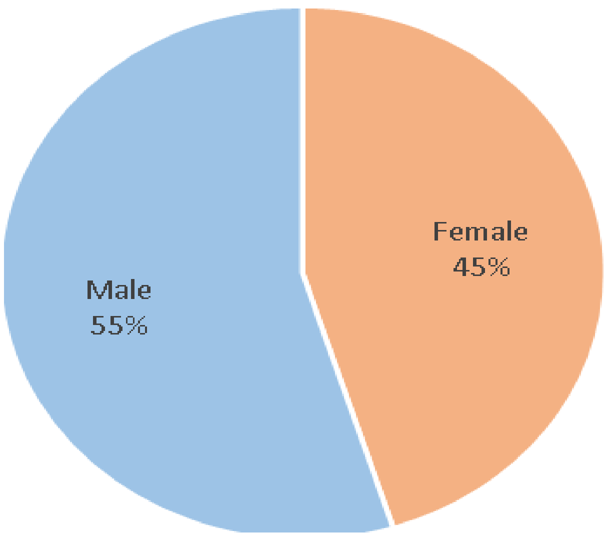

Figure 2: The testing data distributed according to the genders

Figure 3 shows the histogram of the range of ages it can be seen that about half the symbols are in the youth age range (22-42) years old.

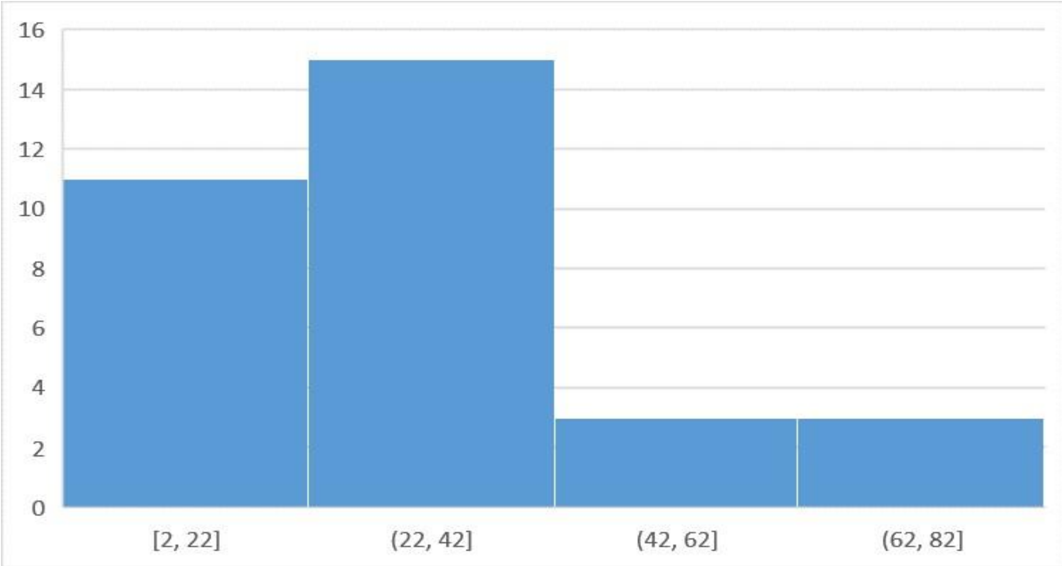

Figure 3: The testing data histogram according to the ages of the subject

Statistical data showed that the sensitivity and resistance to each antibiotic regardless of the age and gender of the infected person is that Vibrio cholerae isolated from feces are sensitive to antibiotics Ciprofloxacin and Tetracycline. Data displayed in Table 1.

Table 1: Data distribution according to age, gender and antibiotic sensitivity of *Vibrio cholera* isolated from stool specimens of infected patients

Specimen no.	Antibiotic	Age	Gender	Specimen no.	Antibiotic	Age	gender	Specimen	Antibiotic	age	gender
1	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	16	female	11	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	27	female	21	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	80	Female
2	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	14	male	12	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	35	male	22	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	60	female
3	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	29	female	13	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	70	female	23	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	37	male

male	14	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	29	male	21	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) 10Suplphamethoprim(R)	19	male	3	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	9
male	33	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	28	female	22	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	18	female	2	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	8
female	30	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	27	male	38	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Triimethoprim(R) Suplphamethoprim(R)	17	male	10	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	7
male	25	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	26	female	65	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	16	male	17	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	6
male	8	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	25	female	36	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	15	male	30	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	5
male	44	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	24	female	47	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	14	male	25	Ciprooxacin(s) Ampicillin(R) 5Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	4

10	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	31	female	20	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	40	male	30	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	23	female
31	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	18	male	32	Ciprooxacin(s) Ampicillin(R) Tetracycline(s) Trimethoprim(R) Suplphamethoprim(R)	25	male				

There were irregular intervals between cholera epidemics, making the prediction of the next outbreak difficult. In addition, there was a significant decrease in the number of cholera cases throughout the outbreaks, with about 30 people dying as a result. These could be the results of community health, naturally developed immunity to cholera, and successful preventative measures (Sidiq, 2023). The WHO recommends antibiotics for severe cholera patients irrespective of their age and for those who require hospitalization (Nelson *et al*., 2011). Infection diarrhea can be shortened by antibiotics (from 5 days to 1 to 2 days) and stool output can be reduced by up to 50% (Rashed *et al*., 2017). Tetracycline, doxycycline, co-trimoxazole, fluoroquinolones, ciprofloxacin, trimethoprim-sulfamethoxazole, azithromycin, and erythromycin are commonly prescribed antibiotics for cholera patients, but resistance is becoming increasingly common (Davies *et al*., 2017). Local antibiotic susceptibility patterns determine the choice of antibiotics. For adults (including pregnant women), doxycycline is advised as a first-line therapy in most countries. For children, the suggested dosage is two to four milligrams per kilogram. Azithromycin and ciprofloxacin are substitute medications in the case that doxycycline resistance is confirmed (Chowdhury *et al*., 2022).

Between 1990 and 2010, there was an increase of antimicrobial resistance (AMR) to trimethoprim-sulfamethoxazole, furazolidone, nalidixic acid, ciprofloxacin, and tetracycline. Since multidrug resistance (MDR) has emerged, antimicrobial testing of V. cholerae clinical isolates is thus required for treating cholera patients in areas where cholera is endemic because of frequent chromosomal mutation. As a result of whole-genome sequencing of V. cholerae isolates over the past decade, plasmids, conjugative elements, superintegrons, and supplemental sequences have been shown to contribute to AMR (Das *et al*., 2020).

CONCLUSIONS

The outbreak of cholera in Iraq occurs several times a year, often during the summer season. Climate change, population growth, and a decline in human immunity might be the main reasons. Besides, it was more common in ale than female, as well as higher among adults than among other ages. A person infected with the commonly used antibiotic will be susceptible and resistant to it regardless of their age and gender. The distribution of diseases according to geographical area in Baghdad and the medical profile of infected individuals need further study. Also, it is recommended that the infected bacteria be tested for sensitivity to other recent antibiotics.

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