

Existence of Multi Drug Resistance Pathogen in Soil of Hospital Disposal Site

Kavya Unni, Anirudh Gupta², and Indu Sharma²

¹Department of Microbiology, Nims Institute of Allied Medical Science and Technology, NIMS University, Rajasthan, Jaipur 303121, Rajasthan (India)

²Department of Biotechnology, Nims Institute of Allied Medical Science and Technology, NIMS University, Rajasthan, Jaipur 303121, Rajasthan (India)

ARTICLE INFO

Keywords:

Correspondence:

E-mail: endusharma@gmail.com

ABSTRACT

Hospital waste is a potential health hazards to the health care workers and public acquired infection, transfusion transmitted diseases. Increasing land and water pollution lead to increasing possibility of contracting many diseases. Study has compelled the authorities to think seriously about hospital wastes and the disease transmitted through their improper disposal. Soil samples were collected from hospital in Jaipur city. A total of six isolated were identified and purified from the samples, further screened for individual antibiotics at their respective concentrations and all the six isolates were found to be strong resistant against antibiotics selected in the study. In this present study, *Pseudomonas* sp only possessed a positive virulence characteristic such as hemolysis, protease, coagulase, lecithinase, pyocyanin and lipase when compare to other species like *E. coli*, *Proteus*, *Bacillus*, *Klebsiella*, *Staphylococcus* sp. These are all very low in prevalence of enzymes associated with bacterial pathogens. The total of 11 antibiotics were used included; Tetracycline, Ampicillin, Sparfloxacin, Co-timoxazole, Gatifloxacin, Ceftrizoxime, Amoxicillin, Ciprofloxacin, Nitrofurantoin, Ofloxacin and Streptomycin. All strains were found resistant to Tetracycline, Ampicillin Sparfloxacin, Co-timoxazole and Gatifloxacin. 8 antibiotics showed an intermediate resistance. All isolates were found sensitive to Nitrofurantoin, Ofloxacin and Streptomycin. The aim of the study is isolation, identification and antibiotic profiling of microbes from soil contaminated with hospital waste dumping..

1. INTRODUCTION

Hospital is the place, which is frequently accessed by the people irrespective of age, sex, race, religion, region and even nationality. The waste generated during entire healthcare activities has higher potential to produce health and environmental hazards than the wastes of other places (Boyce et al., 1997). Hospital wastes are one of the most dangerous causes of pollution. Hospital wastes are generated the diagnosis and treatment or immunization of human beings or animals. It is a universal set having subsets like infectious and hazardous wastes. Wrongly managed hospitals wastes can result in severe health hazards. Hospital wastes refer to all biological or non-biological wastes from hospitals, which are discarded directly to soil. This makes land pollution and environmental pollution in Hospital premises. Some of the pathogenic organisms are dangerous, because they may be resistant to treatment and possess high pathogenicity .

According to the World Health Organization (WHO), medical waste has been defined as an end product of medical services and includes items such as medical devices, sharps, blood,

body parts, chemicals, pharmaceuticals, and radioactive materials. Health care facilities are the main source of medical waste, but other sources include household and communal settings where health care activities conducted in these settings also generate medical waste. Globally, it has been estimated that 5.2 million people, including 4 million children, die annually from waste-related diseases and the situation is likely to get worse if appropriate measures are not developed to curb it [WHO]. There is a growing awareness about solid medical waste (SMW) and its associated hazards, but its management has not received prioritized attention in developing countries.

Although hazardous waste it represents an immediate threat to humans through the transmission of pathogens from biological fluids. Examples of hazardous waste include soiled and/or blood-stained gauze bandages, used lancets and syringes, used and discarded specimen containers, cultures, stocks of infectious agents and human tissue. If medical waste is not treated in a way that destroys the pathogenic organisms (viruses, bacteria, parasites or fungi), these pathogens will be present in harmful quantities. They enter the body through punctures and other breaks in the skin, mucous membranes in the mouth, by inhalation, ingestion, or transmission by a vector organism. People who come in direct contact with the waste are at greatest risk. Sharps waste pose a specific hazard through cuts and punctures. The healthcare workers or waste handlers who handle sharps waste can become infected with HIV/AIDS and hepatitis B and C viruses through pricks or reuse of syringes/needles. Commonly identified bacterial pathogens such as *Pseudomonas spp.*, *Corynebacterium diphtheriae*, *Escherichia coli*, *Staphylococcus spp.* which are known to cause respiratory tract infections and other diseases have been reported in SMW and should be carefully controlled to prevent associated nosocomial infection. Some of these bacteria exhibit resistance to antibiotics. Drugs that were used to treat associated diseases are now losing their impact due to emerging drug resistant microorganisms including *Escherichia coli* and *Klebsiella pneumonia*. This resistance threatens the effective control against these bacteria that cause UTI, pneumonia and bloodstream infections, resulting in longer hospital stay and higher costs of care. Pathogens present in untreated waste can also leach out and contaminate the soil and surface water. Microorganisms that are resistant to one or more therapeutic classes of antimicrobial agents are called Multidrug resistant organisms (MDROs). World Journal of Pharmaceutical Research condition enabling a disease-causing organism to resist distinct drugs or chemicals of a wide variety of structure and function targeted at eradicating the organisms.

The present study is an attempt to isolate and characterize the multi-drug resistant (MDR) bacteria from hospital waste which was obtained from various site of hospitals. The isolates were investigated for their sensitivity against eleven antibiotics and to check the activity of culture in the presence of various antibiotics.

The Soil is the medium where sharps and other wastes are disposed by the land burials etc rich in tetanus spores or blood borne pathogens have gained the significant attack of HIV, HBV, HCV which leads to AIDS and Hepatitis B, C and other viral and bacterial infections. Multidrug resistance is a condition enabling a disease-causing organism to resist distinct drugs or chemicals of a wide variety of structure and function targeted at eradicating the organisms. Pathologic cells, including bacterial and neoplastic cells can display multidrug resistance or multiple drug resistance. Antibiotic resistance occurs when bacteria change in some way that reduces or eliminates the effectiveness of drugs, chemicals, or other agents designed to cure or prevent infectious. The bacteria survive and continue to multiply causing more harm. Bacteria can do this through several mechanisms. Some bacteria develop the ability to neutralize the antibiotic before it can do harm, others can rapidly pump the antibiotic out, and still others can change the antibiotic attack site so it cannot affect the function of the bacteria. The number of MDROs will increase if the selective pressure of antibiotic use continues and the resistant organisms are able to spread from one person to another. Now, multiple-drug resistant (MDR) bacteria are a big public health problem in our world. It is really urgent to improve the current strategies to control this global public health threat.

The presence of bacterial and fungal agents in the hospitals' waste was determined using the conventional bacteria, and fungi identification methods. Medical waste has a high potential of carrying microorganisms that can infect people who are exposed to it, as well as the

community at large if it is not properly disposed. Mainly infection causing hospital dumping soil pathogens are Multi Drug Resistant pathogens. These drug resistant strains initially appeared in hospitals. There are many adverse and harmful effects to the environment and human being which are caused by the hospital waste generated during the patient care. These resistance mechanisms have led to the appearance of MDR bacteria. These MDR bacteria, via multiple mutations, coupled with the acquisition of antibiotic resistance genes by HGT, have earned the name of 'superbugs' and have been responsible for many human infections. Hospitals' solid waste may contain varieties of bacteria. The types and quantity of bacteria depend on the hospitals' solid waste compositions and its generation source. Commonly identified bacterial pathogens such as *Pseudomonas spp.*, *Corynebacterium diphtheriae*, *Candida spp.*, *Klebsiella spp.*, *E. coli*, and *Staphylococcus spp.*, have been reported to be part of the hospitals' wastes. Hospital's waste should be carefully controlled. The present study was undertaken to determine the bacterial and fungal agents present in different various of the hospitals' solid waste.

Types of Wastes

- **Infectious waste:** waste contaminated with blood and other bodily fluids (e.g., from discarded diagnostic samples), cultures and stocks of infectious agents from laboratory work (e.g., waste from autopsies and infected animals from laboratories), or waste from patients in isolation wards and equipment (e.g., swabs, bandages, and disposable medical devices);
- **Pathological waste:** human tissues, organs or fluids, body parts, and contaminated animal carcasses;
- **Sharps:** syringes, needles, disposable scalpels and blades, etc.;
- **Chemicals:** for example, solvents used for laboratory preparations, disinfectants, and heavy metals contained in medical devices (e.g., mercury in broken thermometers) and batteries;
- **Pharmaceuticals:** expired, unused, and contaminated drugs and vaccines;
- **Genotoxic waste:** highly hazardous, mutagenic, teratogenic, or carcinogenic, such as cytotoxic drugs used in cancer treatment and their metabolites;
- **Radioactive waste:** such as products contaminated by radionuclides including radioactive diagnostic material or radiotherapeutic materials; and
- **Nonhazardous or general waste:** waste that does not pose any particular biological, chemical, radioactive, or physical hazard.

Health Impacts of Health-Care Waste

1. Types of Hazards

Exposure to hazardous health-care waste can result in disease or injury. The hazardous nature of health-care waste may be due to one or more of the following characteristics:

- It contains infectious agents;
- It is genotoxic;
- It contains toxic or hazardous chemicals or pharmaceuticals;
- It is radioactive;
- It contains sharps.

2. Persons at Risk

All individuals exposed to hazardous health-care waste are potentially at risk, including those within health-care establishments that generate hazardous waste, and those outside these sources who either handle such waste or are exposed to it as a consequence of careless management. The main groups at risk are the following: medical doctors, nurses, health-care auxiliaries, and hospital maintenance personnel; patients in health-care establishments or receiving home care; visitors to health-care establishments; workers in support services allied to health-care establishments, such as laundries, waste handling, and transportation; workers in waste disposal facilities (such as incinerators), including scavengers.

The hazards associated with scattered, small sources of health-care waste should not be overlooked; waste from these sources includes that generated by home-based health-care, such as dialysis, and that generated by illicit drug use (usually intravenous).

3. Sharps-Related

Worldwide, an estimated 16 billion injections are administered every year. Not all needles and syringes are disposed of safely, creating a risk of injury and infection and opportunities for reuse.

Injections with contaminated needles and syringes in low- and middle-income countries have reduced substantially in recent years, partly due to efforts to reduce reuse of injection devices. Despite this progress, in 2010, unsafe injections were still responsible for as many as 33,800 new HIV infections, 1.7 million hepatitis B infections, and 315,000 hepatitis C infections.

A person who experiences one needle-stick injury from a needle used on an infected source patient has risks of 30%, 1.8%, and 0.3%, respectively, of becoming infected with HBV, HCV, and HIV.

Additional hazards occur from scavenging at waste disposal sites and during the manual sorting of hazardous waste from health-care facilities. These practices are common in many regions of the world, especially in low- and middle-income countries. The waste handlers are at immediate risk of needle-stick injuries and exposure to toxic or infectious materials.

In 2015, a joint WHO/UNICEF assessment found that just over half (58%) of sampled facilities from 24 countries had adequate systems in place for the safe disposal of health-care waste.

Disease which can be caused due to improper waste disposal:

Parasitic Infections

It's not surprising that labs who regularly test for parasitic infections would have specimens that are positive for parasites. Typically, labs will incubate body fluids to test for these parasites, then dispose of them as medical waste. Some of the parasites can still live and thrive in the waste and potentially cause infection

Lung Infections

It is common for medical waste to release airborne aerosols that could potentially contain pathogens, such as a respiratory syncytial virus (RSV), or other viruses that can cause lung infections like influenza, tuberculosis and pneumonia.

Skin infections

It's very possible to contract a skin infection from medical waste, even something as dangerous as anthrax. Yes, anthrax cases are rare, but anthrax vaccine-producing labs may generate anthrax-contaminated medical waste and, if handled improperly, this waste poses a serious threat to the community.

HIV and Hepatitis B and C Viruses

There is always the chance that medical waste contains sharps that may contain blood contaminated with viruses that cause AIDS, Hepatitis B and C. A needle stick or puncture from a scalpel blade can result in deadly infections. This is why sharps are required to be disposed of in rigid, sealed, and clearly marked containers.

Candida

Candida is a disease caused by the yeast *Candida albicans*. *Candida albicans* associated with prolonged hospital stays, so it goes without saying that untreated waste from hospital facilities may contain significant amounts of this pathogen. Candida poses significant and even life-threatening risk to the elderly, pregnant mothers, small children, and people with weakened immune systems.

Meningitis

Medical waste may contain pathogens that can transmit meningitis. Meningitis, which is transmitted via body fluids, causes inflammation of the membranes surrounding the brain and spinal cord.

Bacteremia

Bacteremia is a life-threatening infection. Bacteremia infection occurs when bacteria are present in the bloodstream, where it can easily infect other organs and cause inflammation. A particular type of medical waste, sharps, can introduce pathogens into the bloodstream that may result in bacteremia.

Strains of Pathogenic Bacteria:

1. *Klebsiella pneumonia*

Klebsiella pneumonia is a gram-negative bacterium that causes significant blood stream infections that result to morbidity and mortality if not treated on time. It is one of the most common causes of bacteremia associated with gram negative bacteria. It is also a causative agent of urinary tract infections, nosocomial pneumonia and intra-abdominal infections (Lin et al, 2010). The bacterial pathogen can be acquired in the hospital during hospital admissions and visits or within the community. *K. pneumonia* is mainly found in the environment; it is found in water, sewage, soil and plants and on the mucosal surface of mammals (Bagley, 1985). The bacterium inhabits the nasopharynx and the intestinal tract of humans with higher detection rates being in the latter via the stool cultures (Pods Chun and Ullmann, 1998). Underlying conditions commonly associated with *K. pneumonia* bacteremia include diabetes mellitus, neoplastic disease, chronic lung disease, hepatobiliary diseases and alcoholism (Ko et al., 2002; Tsai et al., 2002; Lin et al, 2010). It is worth noting that the rates of *K. pneumoniae* blood stream infections differ significantly in different temperature and humid conditions. High rates of *K. pneumoniae* infections occur during the warm months of the year (Deerlick et al., 2008). Most clinical isolates of *K. pneumoniae* have developed resistance to most antibiotics used for empirical treatment thereby reducing the treatment options to third generation cephalosporins and quinolones to which resistance is also slowly emerging (Ko et al., 2002). Resistance has been associated with the increasing prevalence of extended-spectrum β -lactamase producing strains (Pods Chun and Ullmann, 1998). Prevention strategies put in place by the public health ministries in Kenya and around the world include proper hand washing and proper hygiene conditions and access to sanitation facilities

2. *Staphylococcus aureus*

Staphylococcus aureus is a gram-positive bacterium frequently associated with community acquired-invasive bacterial infections. These include Ritter disease, folliculitis, furuncle and carbuncle, septic arthritis, endocarditis, toxic shock syndrome, pneumonia and thrombophlebitis. The bacterium is also responsible for Staphylococcal food poisoning that arises from ingestion of food contaminated with the organism. Contamination occurs when food is prepared and stored in areas of poor hygiene where the bacterium is present. Most resource-poor settings lack clean water services and sanitation facilities therefore with the high numbers of people in these congested areas, contamination of water and food with pathogens including *S. aureus* normally occurs and is responsible for significant morbidity and mortality. Strains of *S. aureus* which are resistant to methicillin are commonly referred to as methicillin-resistant *S. aureus* (MRSA) and continue to cause diseases among populations. In addition, multidrug resistant *S. aureus* strains have emerged, a trend that continually exhausts the available treatment options available (Appelbaum, 2006). *Staphylococcus aureus* infections cause fatal disease in patients whose immune system are impaired or weakened as the bacterium is highly aggressive and is able to invade and destroy tissues (Owais et al., 2010).

3. *Escherichia coli*

Escherichia coli is a gram-negative bacterium that inhabits many parts of the body. It is the most prevalent normal flora bacteria in the body and is usually non-pathogenic in the areas it colonizes (Nataro and Kaper, 1998). However, in immuno-compromised hosts or when the gastrointestinal barriers are violated, the non-pathogenic strains of *E. coli* can cause infection. There have emerged *E. coli* strains that produce enterotoxins that are responsible for traveler's diarrhea and infant diarrhea (Nataro and Kaper, 1998). These strains of *E. coli* mostly invade the mucosal surfaces but could at times spread to other parts of the body.

Diseases that are as a result of *E. coli* bacteremia include urinary tract infection, meningitis and diarrheal disease (Nataro and Kaper 1998; Pathak et al., 2012).

The infection rates of *E. coli* are varied with considerably high incidences being recorded in the warmest months of the year (Al-Hasan et al., 2009). Studies have reported that *E. coli* strains have shown high resistance to beta-lactam antibiotics than other classes of antimicrobials (Alam et al, 2011). The β -lactam antibiotics including penicillin, cephalosporins and related compounds, are commonly given to patients presenting with bacterial infections.

CONCLUSION

Finally concluded that, in the present study most of the predominant organisms present in the hospital waste dumping site area. In the study area were virulent possess up antibiotic sensitivity potential source and reservoir of multiple drug resistance organisms. A major factor in the emergence of Multi Drug Resistant Organisms (MDRO's) is overuse of antibiotics in any setting, the hospital or the community. Many human pathogenic bacteria are examples of this emerging crisis, and they are extremely difficult to treat with present existing extended-spectrum antibiotics, leading to increased morbidity and mortality in human. The population and pollution of the whole world is increasing day by day. The main source of the Multi Drug Resistant Organisms is the hospital wastes. Thus Multi Drug Resistant Organisms are increasing in such cities and is becoming a serious threat in the whole world. It is really urgent to improve the current strategies to control this global public health threat. The presence of high number of pathogenic organisms resident in the hospital waste dumping site area is a treat to such environment and has a serious public health implication. It is imperative that all hospital wastes should be incinerated and treated properly before discharge into the environment and there should be an efficient waste management practices such as landfill where wastes could be channelled in both public and private hospitals.

In our present study we have tried to isolate and identify using both biochemical and molecular methods to ascertain the MDR microbes upto species level. We have also done the detailed analysis of MDR to understand and decide the possible control methods for the above mentioned strains/microbes.

REFERENCES

1. WHO. Management of Solid Health-Care Waste at Primary Health-Care Centers: A Decision-Making Guide. Geneva, Switzerland: WHO Press, World Health Organization; 2005.
2. Lubasi-Kapijimpanga BI. Medical Waste Report. Lusaka, Zambia: Office of the Auditor General: 24 p.
3. Udofia EA, Fobil J, Gulis G. Stakeholders' Practices and Perspectives on Solid Medical Waste Management: A Community Based Study in Accra, Ghana. AJEST. 2018; 534–54. doi: 10.4236/jep.2018.913081
4. WHO. Definition and characterization of health-care waste. In: Chartier Y. et al. (eds.), Safe management of wastes from health-care activities (2nd ed.). Geneva, Switzerland: World Health Organization Press, 2014; 1:3–23.
5. Johansson E., Steffens J. J., Emptage M., Lindqvist Y., Schneider G., (2010). Cloning, expression, purification and crystallization of saccharopinereductase from *Magnaporthe grisea*. *Acta Crystallogr D Biol Crystallogr*;56(Pt 5):662–664. doi: 10.1107/s0907444900003735.
6. Nascimento TC, Januzzi WD, Leonel M, Silva VL, Diniz CG. Occurrence of clinically relevant bacteria in health service waste in a Brazilian sanitary landfill and antimicrobial

susceptibility profile. *Revista da Sociedade Brasileira de Medicina Tropical*. 2009. Aug;42(4):415-9. doi: 10.1590/s0037-86822009000400011.

7. Manzoor J, Sharma M. Impact of biomedical waste on environment and human health. *Environmental Claims Journal*. 2019. Oct 2;31(4):311-34. doi: 10.1080/10406026.2019.1619265.

8. Anitha J et al. Isolation and Identification of Bacteria from Biomedical Waste (BMW). *International Journal of Pharmacy and Pharmaceutical Sciences* 2012;

9. Hemalatha et al., *J Pharm Biol Sci* 2017; 5(3): 126-133 129.

10. Shouviksaha et al. Actinomycetes from hospital dump soil produce highly active antibiotic. *International Journal of Microbiology research* 2012; 4(6):258-267

11. Edberg SC et al. Analysis of the virulence characteristics of bacteria isolated from hospital dumping soil. *Journal of microbial ecology in health and disease* 2000; 64-69.

12. Achudume AC, Olawale JT. Enumeration and identification of gram negative bacteria present in soil underlying urban waste-sites in southwestern Nigeria. *Journal of Environmental Biology* 2010; 31(5):643-648.

13. Prasad Chandan et al. Isolation and Characterization of Multi drug Resistant Super Pathogens from soil Samples Collected from Hospitals. *Research Journal of Recent Sciences* 2013; 2: 124-129

14. Moni VT et al. Prevalence of Multidrug Resistant bacteria Isolated from Biomedical waste generated in hospital. *British Microbiology Research Journal* 2015; 10(3): 1-10.

15. Senthil Velsivasakthivel, Natarajan Nandini. Airborne Multiple Drug Resistant Bacteria Isolated from Concentrated Municipal Solid Waste Dumping Site of Bangalore, Karnataka, India. *International Research Journal of Environment Sciences* 2014; 3(10),43-46