The prevalence of multidrug-resistant organisms in the burn units of three hospitals in the Free State and Northern Cape before, during and after the COVID-19 pandemic (1 March 2018–31 July 2024)

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DECLARATION OF ORIGINAL WORK

I, Chene' Beetge (Student number: ________) hereby declare that all the work submitted in this project report titled "The prevalence of multidrug-resistant organisms in the burn units of three hospitals in the Free State and Northern Cape before, during and after the COVID-19 pandemic (1 March 2018–31 July 2024)" is my original and independent academic effort that was carried out under the able supervision of scientists: Prof P.M. Makhoahle and Dr B. Malope-Kgokong and it is hereby submitted to the CUT for the fulfilment of the degree: Master of Health Sciences – Biomedical Technology. I confirm that I know the meaning of plagiarism, and this work has never been presented or submitted to another institution for any other degree or part of a qualification, by me or by any other person.

25/09/2025

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Signed Date

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This research is dedicated to my husband, JD, and my Pieterse family for their constant love, encouragement and support throughout this journey.

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LIST OF ABBREVIATIONS

AARMS: Academic Affairs and Research Management System

AMR: Antimicrobial resistance

ASP: Antimicrobial Stewardship Program

CDC: Center for Disease Control and Prevention

CDW: Central Data Warehouse

COVID-19: Coronavirus disease of 2019

HAI: Hospital Acquired Infections

HIV: Human Immunodeficiency Virus

MDR: Multidrug-Resistant Bacteria

NHLS: National Health Laboratory Services

US: United States

WHO: World Health Organization

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ABSTRACT

INTRODUCTION

Antimicrobial resistance (AMR) is a global health threat; it has no limits and knows no borders and can be found on every continent across the globe. The impact is evident with the sixfold increase of AMR that has been seen in the world since 2017. It has been estimated by researchers that by the year 2050 there will be 10 million deaths across the world because of untreatable strains, due to the misuse and overuse of antimicrobials that led to the extensive development of AMR. This study was done to determine the impact that the COVID-19 pandemic had on the further development of AMR by comparing the bacterial resistance pattern before, during and after the pandemic.

METHODOLOGY

This was a retrospective study, and retrospective data were used to conduct this research. The data that were requested were pre-pandemic (1 March 2018 until 30 February 2020), during the pandemic (1 March 2020 until 30 June 2022), and post-pandemic (1 July 2022 until 31 July 2024). The burn units of the public hospitals of the Free State and Northern Cape were chosen as the study site for the research. The hospitals in the Free State include Pelonomi, National, Universitas, Bongani Regional Hospital, Boitumelo and Mofumahadi Manapo Mopeli Hospital. The hospital of the Northern Cape is Robert Mangaliso Sobukwe. In this study we wanted to determine the resistance profile of the bacteria in the burn units and compare the resistance before, during and after the pandemic.

RESULTS AND DISCUSSIONS

This study compared the resistance in the burn units of the Northern Cape and Free State before, during and post-COVID-19. It was noted that in most cases, resistance increased during the pandemic, and it did not decrease to the levels it had been before the pandemic.

A total of 1 606 samples were collected for this study, of which 1 549 were from the Free State and 57 from the Northern Cape. Due to the small sample size of the North Cape, the results largely reflect those of the Free State. In both provinces

Pseudomonas aeruginosa and *Staphylococcus aures* were the most frequently isolated organisms. In both provinces *Acinetobacter baumannii* showed a concerning and persistently high resistance (above 80%). The overall resistance increased in the Free State during COVID-19 (35.01% to 38.64%) and increased further post-COVID-19 (39.59%). The resistance rose sharply during COVID-19 in the Northern Cape (28.77% to 52.78%), but the levels decreased again post-COVID-19 (21.05%). The decrease could be attributed to the small sample size for this timeframe.

At the organism level, the resistance of *Staphylococcus aureus* decreased in the Free State from 32.19% pre-COVID-19 to 21.9% post-COVID-19, while the resistance increased drastically to 50% in the Northern Cape during COVID-19. The resistance of *Pseudomonas aeruginosa* was quite high. It spiked during COVID-19 in the Free State (37.39% to 57.49%) and then decreased slightly (49.49%). The resistance was 34.72% pre-COVID-19 in the Northern Cape with no further comparable data. The resistance of *Klebsiella pneumoniae* rose markedly during COVID-19 (36.65% to 53.33%) in the Free State and decreased slightly post-COVID-19 (45.03%). *Streptococcus pyogenes,* while *Proteus mirabilis* showed a slight decrease in resistance in the Free State over the three timeframes. *Escherichia coli* showed a sharp decrease in resistance during the pandemic in the Free State (21.5% to 3.92%), and this then increased slightly post-COVID-19 (9.62%).

The patterns of increased in resistance post-COVID-19 levels confirm the need for the reinstatement of antimicrobial stewardship programmes in South Africa along with infection-prevention measures in the burn units in South Africa.

CONCLUSION

Overall, this study has shown that during COVID-19, the AMR increased in the burn units of the Northern Cape and the Free State. Post-pandemic it is shown that the resistance levels remained higher than the levels pre-pandemic. The organisms that are the most concerning was *Pseudomonas aeruginosa*, *Acinetobacter baumannii*, and *Klebsiella pneumoniae* due to the persistence in elevated resistance levels. The resistance patterns vary across the different timeframes for the different antimicrobial classes, with the most worrisome trends seen among the last-resort antimicrobials. However, a common trend is seen, and this is that the resistance levels did not go back to pre-COVID-19 levels. This highlights the urgency of the matter to firstly

strengthen and then re-establish the antimicrobial stewardship programmes in South Africa, and then also to prioritize the infection prevention measure in the burn units. If there is no intervention, the resistant infections that are currently in these burn units will continue to threaten and cause further harm to these burn unit patients that are already vulnerable and further strain the South African healthcare systems.

Keywords: antimicrobial resistance, COVID-19 pandemic, multidrug resistance.

CHAPTER 1: BACKGROUND

1.1 INTRODUCTION

The impact of the coronavirus disease of 2019 (COVID-19) on South Africa was undeniably significant, affecting not only the country's financial landscape, but also leaving its mark on the medical sector. Hospitals were drastically understaffed during this period and without the necessary equipment, drugs and infrastructure to fight this pandemic led to a poor health outcome, which was unavoidable in certain situations. There was a wide range of adverse impacts on the health services and one of the impacts that were not noticed was the further development of antimicrobial resistance (AMR) (Shomuyiwa et al., 2022). The antimicrobial stewardship programmes were shifted aside during the pandemic, and more focus was put on the lockdown, rapid testing and vaccinations for COVID-19. Virtual patient consultations and intensive care units with the focus on treating COVID-19 patients unintentionally contributed to the further development of multidrug-resistant bacteria, and while the laboratories were flooded with COVID-19 samples, there was a decrease in the testing of bacteria (Khaznadar et al., 2023).

Factors that contributed to the increased consumption of antibiotics included misconceptions, lack of awareness about the virus outbreak, cultural stigma and misinformation, and the overall fear of the population to get infected with the virus led to patients asking for antibiotics without a bacterial infection being present. This occurred in low-and-middle-income countries in particular, for example, South Africa (Djuikoue et al., 2023). In 2017, a list was released by the World Health Organization (WHO) to researchers and doctors that included 12 of the families of bacteria that were the most resistant to antimicrobials to promote the development of new antimicrobials by researchers. The organisms that were on the list included multidrug-resistant (MDR) bacteria that can be found in hospitals or that pose a threat to patients with indwelling devices (Med, 2019). Bacterial infections could be fatal if left untreated due to the unavailability of antimicrobials to which they are sensitive. These infections include pneumoniae and blood infections. This was done in an attempt to decrease the further development of AMR, but before any plans or strategies could be put in place, the COVID-19 pandemic occurred and since then attempts have been placed on hold (Djuikoue et al., 2023).

When looking at low-and-middle-income countries, it is noted that 80% of all of the prevalent illnesses show resistance to at least one of the antibiotics that are commonly utilized for that specific bacterium (Gulumbe et al., 2023). This clearly highlights the severity of the new pandemic that could be seen as AMR in these countries. Further research confirmed that there is a direct correlation between the high levels of resistance and the overuse and misuse of antibiotics (Gulumbe et al., 2023). South Africa lacks the presence of a proper and working antimicrobial stewardship programme (ASP) (Gulumbe et al., 2023). Without this programme in place the presence of AMR will increase further due to the lack of education regarding the proper use of antimicrobials. A WHO report states that in South-East Asia and in Africa, 45% of deaths are due to MDR bacteria (Gulumbe et al., 2023). These troubling numbers are said to increase even more, since this data had been collected before the COVID-19 pandemic, and current research confirms that the pandemic might have increased the prevalence of MDR bacteria across the world (Gulumbe et al., 2023).

Antimicrobial resistance led to prolonged hospital stays for patients, with an increased risk for the further development of a hospital-acquired infection. The effects of AMR do not only stop at the patient but also has a tremendous impact on the strain of the economy. An increased amount of money is spent on the treatment of patients in public hospitals, which could be avoided. This causes a decrease in the finances made available to other clinics or other areas in the health sector that are in dire need of funding. Examples of this financial implication in other countries include 9 billion euros that are spent in Europe each year due to AMR (Dadgoster, 2019). AMR also adds 20 billion dollars in direct healthcare costs according to the Centers for Disease Control and Prevention (CDC) in the United States (Dadgoster, 2019). Another 35 billion dollars is lost annually due to a decrease in productivity (Dadgoster, 2019). These numbers are drastically high, especially since these two countries are high-income populations, and this should be taken into consideration when looking at the impact that AMR had on South Africa. There is no definite number to say how much money South Africa has lost in productivity or how much is spent in healthcare costs, but since it is a middle-income country, and just by looking at the high-income countries, it can clearly be estimated to be billions of rands (Dadgoster, 2019).

1.2 PROBLEM STATEMENT

Antimicrobial resistance is a global health threat that has no limits. It knows no borders and can be found in every continent across the globe. The impact is evident, with the sixfold increase of AMR seen in the world since 2017. It has been estimated by researchers that by the year 2050, there will be 10 million deaths across the world due to untreatable strains because of the misuses and overuse of antimicrobials that lead to the extensive development of AMR (Shomuyiwa et al., 2022). This study was done to determine the impact the COVID-19 pandemic had on the further development of AMR by comparing the bacterial resistance pattern before, during and after the pandemic.

1.3 JUSTIFICATION OF THE STUDY

It is commonly said that we have to fight the war against bacteria, but this is a common misconception. We have to fight the war against AMR bacteria to reduce their effects on humans. As global research has stated, by the year 2050, ten million people will die each year directly due to antibiotic-resistant infections. When comparing this to the COVID-19 pandemic, four times more people died due to COVID-19 across the world in the first year of the pandemic. Rather than looking at the pandemic of AMR as a problem that can be solved in the future, it should be handled like the COVID-19 pandemic as a threat that is currently present. South Africa has to make drastic plans to counteract this effect of AMR, since 88% of the ten million people who are said to die by 2050 would be from Africa and Asia. To counteract the effect of AMR, it is said that five main points need to be considered: the development of new antibiotics, promoting antibiotic and bacterial education and awareness, good surveillance, infection prevention, and optimization of antibiotic use. The antibiotics that are currently available should be used sparingly and with much reserve to prevent the further development of AMR. This has been seen as a faceless pandemic due to the fact that there is not only one single pathogen, but rather a whole list (Davids, 2021).

Spending the necessary time and funding to understand AMR better in a post-COVID-19 South Africa could prevent thousands of deaths. However, not a lot is known about the AMR post-COVID-19 in South Africa. Therefore, research needs to be conducted to gain facts to later develop policies and strategies to ensure recovery not just from

COVID-19, but also from AMR. This has to be done to ensure the safety of our population (Shomuyiwa et al., 2022).

1.4 RESEARCH QUESTIONS

- What was the bacterial resistance profile of the organisms from the swabs that were sent to the laboratories from the burn units of the public hospitals in the Free State and Northern Cape before, during and after the COVID-19 pandemic?
- How did the years before, during and after the COVID-19 pandemic compare with one another in terms of the bacterial resistance profile?

1.5 AIM

This research aimed to determine what the bacterial resistance profile of the identified microorganisms detected in swabs taken from the patients in the burn units of the public hospitals in the Free State and Northern Cape was between 1 March 2018 and 31 July 2024.

1.6 OBJECTIVES

1. To determine the bacterial resistance profile of the organisms from the swabs that were sent to the laboratories from the burn units of the public hospitals in the Free State and Northern Cape pre-pandemic (1 March 2018 until 30 February 2020), during the pandemic (1 March 2020 until 30 June 2022), and post-pandemic (1 July 2022 until 31 July 2024).
2. To determine how the years before, during and after the pandemic compared to one another in terms of the bacterial resistance profile from the swabs that were sent from the burn units to the laboratories.

CHAPTER 2: LITERATURE REVIEW

2.1 INTRODUCTION

The world is faced with an increase in bacterial resistance and burn units are not excluded, as Africa still relies on non-electrical energy and other burn-associated incidents. A normal human carries around two kilograms of bacteria, meaning there are more bacteria than human cells in the body (Davids, 2021). These bacteria are good for the body and aid in the proper functioning of the body. The problem occurs when these bacteria become pathogenic, or when a bacterial infection is acquired and the bacterium is resistant. It was noted that in the past few decades there has been a drastic increase in the amount of unnecessary antibiotic consumption in South Africa (Davids, 2021). Because of these unnecessary prescriptions that were given to patients, infections are now seen in hospitals that are resistant to first- and second-line antibiotics (Davids, 2021). Another problem is the pan-drug-resistant infections that are seen in patients. This is seen when the bacteria are resistant to nearly all antimicrobials that could be used against it (Davids, 2021).

When patients are given antibiotics, they expect to take the medication and become better afterwards. However, this road from sickness to health is becoming even more and more of a rocky road. This is because the antibiotics that are designed to work for certain bacterial infections, are no longer working due to the resistance of the bacteria. This causes practitioners to try a range of different antibiotics before one works for a patient. However, in certain cases, there is no antibiotic that can help patients and, in the end, they die due to bacteria being resistant (Barron, 2024). When patients are prescribed antibiotics, the patients are told to complete the entire course of antibiotics, but patients do not completely understand how antibiotics work. Thus, when they feel better, they tend to stop using the antibiotics. This will then cause a form of resistance in the bacterium that the patient has, since the dosage of the antibiotic is not constant or enough to kill the bacterium, causing the bacterium to adapt and be able to grow in the presence of the antimicrobial (Mendelson, 2017).

2.2 THE OVERALL USE OF ANTIBIOTICS IN SOUTH AFRICA ACCORDING TO STATISTICS AND HOW SOUTH AFRICANS VIEW ANTIBIOTICS

When looking at the percentage of antibiotics that were used in South Africa, the public sector used 28% of the total amount of antibiotics that were prescribed and these antibiotics were extended-spectrum penicillin. Oral trimethoprim-sulfamethoxazole and metronidazole were used 13% and 12% overall. In contrast, within the private sector, extended-spectrum penicillin, carbapenems, and third-generation cephalosporins constituted 41%, 20% and 13%, respectively. The utilization of macrolides has more than doubled from 5% to 11% between 2018 and 2020, which might indicate potential increases in usage during the COVID-19 pandemic in South Africa, starting from March 2020 and continuing at the time of drafting this report. The private sector exhibited a higher overall proportion of carbapenem usage compared to the public sector, although both sectors displayed an elevated usage of broad-spectrum penicillin. It is worth noting that antibiotic usage has the potential to drive resistance (Health, 2018).

Based on a study that was done in 2018 that focused on the behaviour and understanding amongst patients and practitioners regarding antibiotics, it was found that 97.1% of these prescribers believed that antibiotics were overused, while 95.8% of these prescribers believed that AMR is a drastic problem. Patients were also believed to play a role in AMR according to these prescribers, saying that 87.4% believed it was due to patient non-adherence, while 66.5% of these prescribers felt that they were pressured by patients to prescribe antibiotics to them. This study also showed that patients who were more aware of AMR had lower expectations to receive antibiotics than those who did not have any knowledge about AMR. Practitioners mentioned that antibiotics were prescribed to these patients without hesitation to maintain a good relationship with their patients. It was found that only 33% of the final-year medical students in South Africa felt that they were confident enough to prescribe antibiotics, and 95% agreed that they would appreciate an improvement in undergraduate training regarding AMR to understand how to prescribe and use antibiotics appropriately (Farley et al., 2018).

2.3 THE IMPACT OF THE COVID-19 PANDEMIC ON THE PREVALENCE OF ANTIMICROBIAL RESISTANCE.

Social distancing, masks and travel restrictions were implemented to prevent the spread of COVID-19 and isolation was requested for patients who tested positively for the virus. The travel restrictions that were implemented could increase the presence of AMR, since people who travelled to countries that have a higher prevalence of AMR could become infected and move these pathogens from one country to another. This then caused the further spreading of AMR pathogens. These restrictions were only short term, so their effect on AMR could not exactly be determined. The scientific community agrees that the face masks greatly reduced germ transmission. This fact is supported by the decrease in the influenza and pneumonia cases that were reported. On the other hand, these face masks were a great hiding spot for antimicrobial-resistance genes, especially in the marine ecosystem, and it could contribute to the increased AMR (Khaznadar et al., 2023).

While most could say that COVID-19 could only have had a positive effect on the development of AMR due to isolation and personal protective equipment, some research suggests otherwise. Due to the attention being mostly on COVID-19 patients, samples were mostly not tested for any bacterial organism and only tested for the virus. Bacterial co-infections could also occur in these hospitalized patients, and this requires antibiotics, but data suggest that the prescription of antibiotics to these patients is much higher than the actual prevalence of antimicrobial co-infections. This also contributes to the overuse of antibiotics and further development of AMR. While most healthcare workers were obligated to attend to the COVID-19 patients, not a lot of attention was placed on AMR. This allowed self-medication of antibiotics to increase and the testing of samples for antimicrobial susceptibility to decrease (Khoshbakht et al., 2022).

According to an article written in the United States, over 90% of their hospital beds were occupied in their hospitals from May 2020 until April 2021. These patients were in hospital for an average of 14 days and many of them were ventilated. Due to them being in hospital for so long, and also being on a ventilator, this increased their risk of a hospital-acquired infection. Because the clinicians were concerned about co-infections in their patients they prescribed antibiotics more easily to patients. This meant that around 80% of all patients that were hospitalized in the US during the pandemic were prescribed antibiotics. Data that were obtained showed that multidrug-resistant infections were five times more likely to be seen in the COVID-19 wards than in wards that were not for COVID-19 patients (Dall, 2025).

Around 50% of the overall antibiotics that are prescribed to patients are done so unnecessarily. In general, prescriptions are given to patients that present with cold or flu-like symptoms by doctors, but these illnesses are caused by viruses and antibiotics have no effect on a virus. A study that was done in 2015 by the WHO indicates that South Africans held the belief that flu, colds, HIV, measles, headaches and body aches could be cured by antibiotics. Since COVID-19 had cold or flu-like symptoms, patients requested antibiotics, since they believed it would help them (Mendelson, 2017).

Due to the high pressure that was placed on the healthcare system during the pandemic, antibiotics were prescribed to patients due to the initial uncertainty in patients that were presenting with a respiratory illness. This was made even more difficult due to the possibility of a co-infection with COVID-19 or a secondary infection in COVID-19-positive patients. Virtual consultations also contributed to the increased prescriptions of antibiotics due to the difficulty in diagnosis over the phone (Langford et al., 2023).

2.4 HOW THE USE OF THE ANTIMICROBIAL STEWARDSHIP PROGRAMS COULD REDUCE THE FURTHER DEVELOPMENT OF AMR AND WHY IT IS NECESSARY

Antimicrobial stewardship is a structured initiative aimed at promoting the prudent utilization of antimicrobial agents, encompassing antibiotics. Its key objectives include enhancing patient outcomes, curbing microbial resistance, and stemming the spread of infections triggered by multidrug-resistant organisms. Collaborating with healthcare practitioners to adhere to the principles of the 5 "Ds" are: selecting the appropriate drug, ensuring accurate dosage, choosing the suitable route of administration, prescribing the necessary duration promptly, and considering the de-escalation of antimicrobial therapy. This aims to prevent overuse or misuse of antimicrobials by patients, thereby reducing adverse effects associated with antibiotics and minimizing the emergence of resistance while also cutting down on healthcare-related expenses (Khadse, Ugemuge, & Singh, 2023).

According to the Centre for Disease Control and Prevention (CDC), AMS involves assessing and enhancing how antibiotics are prescribed by healthcare providers and utilized by patients. Enhancing antibiotic prescription practices is crucial for treating infections effectively, safeguarding patients from potential harm associated with unnecessary antibiotic use, and addressing antibiotic resistance. The CDC's Core Elements of Antibiotic Stewardship provide healthcare providers and institutions with fundamental principles to guide efforts aimed at enhancing antibiotic usage, thereby promoting patient safety and better outcomes. These guidelines complement existing recommendations and standards from prominent healthcare organizations (Centre for Disease Control and Prevention, 2023).

AMS plans should be implemented due to the fact that the amount of AMR is only increasing as acknowledged by the WHO, and only a few new antibiotics in the process of being developed. With the amount of antibiotics that are able to work against microbials decreasing, and the fact that South Africa has a higher-than-normal burden of infectious diseases, the implementation of AMS programmes could only benefit South Africa. Based on statistics that were collected in 2019, South Africa has the highest number of people with the human immunodeficiency virus (HIV), namely 7.97 million people. South Africa also has the fourth-highest number of people living with tuberculosis globally. Other factors that could contribute to the amount of AMR and increased infections in South Africa include malnutrition, poverty, shortage of trained medical professionals and a high number of non-communicable diseases. All of these factors are great examples of why South Africa needs an AMS programme (National Library of Medicine, 2021).

2.5 HOW THE ANTIMICROBIAL STEWARDSHIP PROGRAMMES MITIGATED THE IMPACT OF THE PANDEMIC ON AMR.

Quite an in-depth study was done in the United Kingdom regarding AMS programmes and how they were done before and during the pandemic. The researcher interviewed healthcare professionals who were part of the AMS programme. They noted that the most effective way to implement the AMS strategies was by doing ward rounds, but due to the pandemic that was not possible, and the programme was neglected. Before the pandemic there were regular workshops and educational seminars held to ensure that practitioners were kept up to date and engaged, but due to the pandemic this has been stopped and has largely been forgotten. Certain healthcare professionals mentioned that it has been as if practitioners believe that AMR does not exist anymore and that they have forgotten about how big a danger it truly is (Khan et al., 2022).

It is quite clear that the AMS programmes were not as effective during the pandemic as they should have been, not due to the programmes being faulty, but due to the focus shifted to the pandemics and AMR being placed at the back of the row as the least important. In the UK alone, 60% of the participants in the AMS programmes agreed that the pandemic had a negative effect on the programme. Only in a small percentage of hospitals and wards was the programme still followed, and they reported a decrease in the consumption of antibiotics during the pandemic, but this cannot be said for most of the larger hospitals that had a lot more patients (Comelli et al., 2022).

CHAPTER 3: METHODOLOGY

3.1 STUDY LOCATION

The burn units of the public hospitals of the Free State and Northern Cape were chosen as the study site for the research. The hospitals in the Free State include Pelonomi, National, Universitas, Bongani Regional Hospital, Boitumelo and Mofumahadi Manapo Mopeli Hospital. The hospital of the Northern Cape was Robert Mangaliso Sobukwe. The burn units specialize in patients who have burn wounds. The two provinces that were chosen gave a more accurate representation of what the bacterial resistance profile was, since the general population of these two provinces together are much larger.

3.2 STUDY DESIGN

This was a retrospective study. Therefore, retrospective results were collected for samples that had already been tested. The data were only collected, and no changes were made to these data. The data that were requested were regarding pre-pandemic (1 March 2018 until 30 February 2020), during the pandemic (1 March 2020 until 30 June 2022), and post-pandemic (1 July 2022 until 31 July 2024).

3.3 EXPECTED RESEARCH OUTCOMES

This study contributes to the body of knowledge by being published in scientific journals and conferences. It gives a better understanding of our antimicrobial resistance and how the pandemic influenced this. The research will also influence policy change if the current treatment regime is found to have changed due to the COVID-19 pandemic.

3.4 STUDY POPULATION

3.4.1 The number of participants

The study consisted of at least 40 samples per each period, meaning 120 per each identified laboratory, making a total of 720 samples. The samples from the burn units were collected from the public hospitals of the Free State and Northern Cape between

1 March 2018 until 30 February 2020 before the COVID-19 pandemic, between 1 March 2020 until 30 June 2022 during the pandemic, and lastly between 1 July 2022 until 31 July 2024, which was after the pandemic.

3.4.2 Participant identification

An application was submitted on the Academic Affairs and Research Management System (AARMS) to request access to the data from the NHLS central data warehouse (CDW). Only patients from the burn units of Pelonomi, National, Universitas, Bongani Regional Hospital, Boitumelo, Mofumahadi Manapo Mopeli and Robert Mangaliso Sobukwe Hospitals were included in the study. The timeframe from 1 March 2018 until 30 February 2020 before the COVID-19 pandemic, between 1 March 2020 until 30 June 2022 during the pandemic, and lastly between 1 July 2022 until 31 July 2024, which was after the pandemic.

3.4.3 Inclusion and exclusion criteria

3.4.3.1 Inclusion criteria

1. All participants were admitted patients from the burn units of the following hospitals: Pelonomi, National, Universitas, Bongani Regional Hospital, Boitumelo, Mofumahadi Manapo Mopeli and Robert Mangaliso Sobukwe.
2. The sample should be a swab that was sent to the laboratory for antimicrobial susceptibility testing.
3. The sample should have been collected from 1 March 2018 until 29 February 2020, or from 1 March 2020 until 30 June 2022, or lastly from 1 July 2022 until 31 July 2024.
4. The patient should be 18 years or older.

3.4.3.2 Exclusion criteria

1. The patient was not admitted to the burn units of the following hospitals: Pelonomi, National, Universitas, Bongani Regional Hospital, Boitumelo, Mofumahadi Manapo Mopeli and Robert Mangaliso Sobukwe.
2. The sample was not a swab, nor it was not sent for antimicrobial susceptibility.

3. The sample was not collected from 1 March 2018 until 29 February 2020, or from 1 March 2020 until 30 June 2022, or lastly from 1 July 2022 until 31 July 2024.
4. The patient was younger than 18 years.

3.5 METHODS AND MATERIAL

3.5.1 Preparation

A data sheet was compiled by the data analyst for the bacterial resistance profile before, during and after the pandemic. All seven of the hospitals of both provinces were combined into this spreadsheet into their respective time periods. De-identified data were received from the NHLS CDW so there was no need to remove patient personal information. The type of organism and the bacterial resistance profile for each organism were entered in the spreadsheet.

3.5.2 Data collection

This was a retrospective study, and the data had already been available after the end of July 2024, but arrangements for the timeframe of the data were made prior to the date. The samples were collected and processed over different timeframes at each hospital. After the necessary approval from the NHLS academic affairs and research management system (AARMS) for the data use and the HSREC, data were obtained from the CDW. The inclusion and exclusion criteria were also given to the NHLS to ensure that the data meet the necessary requirements. The organism and the antimicrobial resistance pattern for each patient was then entered into the respective data sheets.

3.5.3 Documentation and metadata

The Research Electronic Data Capture (REDCap) platform served as the primary tool for data capturing and analysis of the results obtained from NHLS CDW due to its robust security features. The platform was accessed through a secure web application, and all data were safeguarded through password protection, with access restricted solely to the private investigator.

Data were requested and stored in CSV file format to ensure data integrity and stored securely in a double password-protected file or computer on the Figshare storage system in its original form. Analysis of the data was conducted using Excel files, also password protected, with complete de-identification of the data. These files were stored separately from any de-identified or pseudo-anonymous information, ensuring anonymity. Furthermore, any numerical identifiers were stored in a separate file devoid of metadata from the original study data. The anonymous data were then analysed in Excel to generate documents bearing the initials of the principal investigator, along with the date of creation, all of which were protected with passwords.

3.5.4 Data analysis

Statistical methods were employed to assess the significance of results, focusing on organisms and their antibiotic profiles. Various graphical representations and tables were utilized to illustrate differences in resistance patterns amongst the three timeframes that were included in this research.

3.5.5 Data storage

This research was categorized as low risk. Data storage was facilitated through Figshare, a POPIA-approved platform known for securely storing research data on the cloud. This enabled private sharing of data among the research team. To mitigate any potential security risks, the device underwent thorough malware scans using reputable antivirus software, ensuring compatibility with the Figshare system. Additionally, regular backups were conducted to guarantee the safe storage of data. Before data were uploaded onto Figshare, they were firstly processed through RedCap. RedCap was configured to only record necessary de-identified data, and encryption was enabled. Training sessions were conducted to ensure proficiency in RedCap programming.

3.6 DATA MANAGEMENT AND STATISTICAL ANALYSIS

Primary data were extracted from the NHLS CDW from the seven hospitals for the patients that were tested for antimicrobial susceptibility in the burns units that met the criteria. This data was entered into the spreadsheet based on the date of collection. Statistical analysis was used to determine the different organisms and the resistance patterns. The change over the periods selected was used to calculate the *p*-value, any sample error margin (SEM) was identified and analysed to determine the trueness of the results obtained.

3.7 ETHICAL ASPECTS AND GOOD CLINICAL PRACTICE

3.7.1 Ethical clearance and confidentiality

Ethical approval was requested from the Health Sciences Research Ethics Committee (HSREC) and permission for the use of the data was obtained from the regional manager of the National Health Laboratory Services (NHLS). De-identified patient results were obtained from the NHLS CDW. As per the guidelines outlined in the Protection of Personal Information Act (POPIA) (RSA, 2013) for researchers, any study involving human subjects must receive ethical clearance from a recognized research ethics committee. Moreover, it is imperative to adhere to the regulations specified in sections 14, 15 and 17 of the National Health Act 61 of 2003 (RSA, 2003) concerning confidentiality, access to health records, and safeguarding health information.

Data provided by the NHLS were utilized in accordance with ethical standards and solely for the intended research purposes. Strict confidentiality protocols were upheld at both participant and institutional levels, ensuring no divulgence of personal or sensitive data, as stipulated by NHLS policy and POPIA. Only pertinent information necessary for the study was accessed and utilized. The data utilized in this investigation originated from routine clinical examinations, obviating the necessity for additional inquiries. Commencement of this study was contingent upon obtaining ethical clearance from the HSREC and acquiring data from the NHLS.

3.7.2 Safety variables

3.7.2.1 Project safety

No interviews were carried out in this study, and no experiments were performed on patient samples. All information that was extracted from the NHLS CDW complied with the POPIA and Excel sheet be kept confidential and password protected for access by the study principal investigator and the supervisors.

3.7.2.2 Patients' safety

This research posed no threat to any patients, since only patient results were collected in a way to ensure patient confidentiality.

3.7.2.3 Financial implication to the participants

There were no financial implications to the participants in this study.

3.7.2.4 Withdrawal criteria

Not applicable to this particular study.

3.7.2.5 Subject information and informed consent

Not applicable to this particular study.

3.7.2.6 Data sharing and accountability

The research data remained strictly confined within my research team; no external parties had access to it. As the designated Private Investigator, I bore the responsibility for acquiring, securely storing, and safeguarding the research data in accordance with the principles outlined in the POPIA code of conduct. Any breaches of compliance were my sole responsibility. The data was retained for a period of five years, after which it was securely disposed of. In the event of my departure from NHLS/CUT, all data under my custody was promptly deleted or securely erased. Final oversight of data management will rest with my supervisor at CUT, or an appointed staff member should the supervisor be unavailable.

CHAPTER 4: RESULTS

4.1 RESULTS

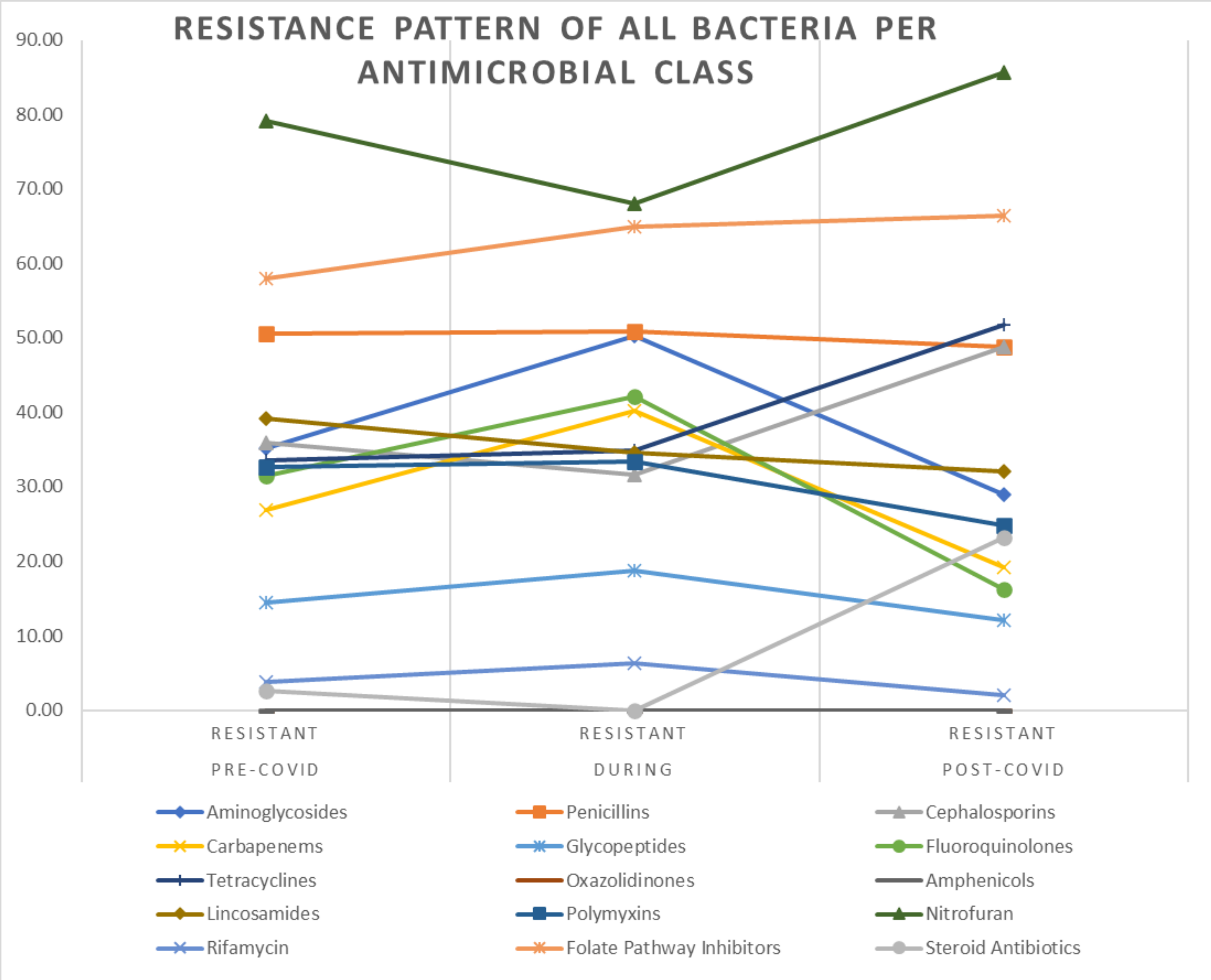

Figure 1: Overall resistance pattern of all bacteria to each antibiotic class

Figure 1 shows the overall resistance pattern of all the bacteria to each antimicrobial class in both provinces. Penicillins and Lincosamides showed a gradual decrease in resistance percentage across all three timeframes. In contrast, Folate path inhibitors showed a progressive increase across all three timeframes. Nitrofuran, Tetracyclines, Cephalosporins and the Steroid antibiotics decreased during COVID-19 and subsequently increased again post-COVID-19. Conversely, Aminoglycosides, Fluoroquinolones, Carbapenems, Rifamycin, Glycopeptides, Polymyxins and

Fluoroquinolones increased during the COVID-19 timeframe and declined again in the post-COVID-19 timeframe.

Table 1: Bacterial isolate samples per province

	Free State				Northern Cape				
	Pre-COVID-19	**During COVID-19**	**Post-COVID-19**	**Total**	**Pre-COVID-19**	**During COVID-19**	**Post-COVID-19**	**Total**	**Grand Total**
PSEUDOMONAS AERUGINOSA	210	139	186	535	12	0	0	12	547
STAPHYLOCOCCUS AUREUS	169	136	118	423	14	1	0	15	438
PROTEUS MIRABILIS	30	49	100	179	9	0	1	10	189
ACINETOBACTER BAUMANNII	32	27	64	123	8	1	0	9	132
KLEBSIELLA PNEUMONIAE SUBSP PNEUMONIAE	40	13	27	80	2	0	0	2	82
STREPTOCOCCUS PYOGENES	23	22	6	51	2	0	0	2	53
ESCHERICHIA COLI	6	11	20	37	1	0	0	1	38
PROVIDENCIA STUARTII	22	1	3	26	0	0	0	0	26
ENTEROBACTER CLOACAE SUBSP CLOACAE	6	12	1	19	0	0	0	0	19
SERRATIA MARCESCENS	12	1	3	16	2	0	0	2	18
ENTEROCOCCUS FAECALIS	11	5	2	18	0	0	0	0	18
PSEUDOMONAS PUTIDA	0	0	11	11	1	1	0	2	13

CITROBACTER FREUNDII	12	0	0	12	0	0	0	0	12
ENTEROBACTER CLOACAE COMPLEX	6	0	4	10	1	0	0	1	11
ENTEROBACTER AEROGENES	6	0	3	9	1	0	0	1	10
Grand Total	**585**	**416**	**548**	**1549**	**53**	**3**	**1**	**57**	**1606**

Table 1 represents the different bacteria from the swabs of the burn unit patients from the Free State province for pre-, during and post- COVID-19. In the Free State a total of 1 549 samples were collected. In the Northern Cape only 57 samples were collected, which make this a total of 1 606 samples in this study. The study sample size shows that the majority of the samples are from the Free State and therefore the findings will largely highlight the patterns in this province. Even though the Northern Cape is included in this study the results should be interpreted with caution due to the small sample size.

During COVID-19 the sample size for the Free State declined from 585 to 416 and then increased again post-COVID-19 (548). In the Northern Cape the sample size decreased from 53 to 3 during COVID-19 and decreased even further post-COVID-19 (1). *Pseudomonas aeruginosa* is found to be the most isolated in the Free State (535) and has the second-most isolates in the Northern Cape (12). *Staphylococcus aureus* is the most commonly isolated bacterium in the Northern Cape (15).

In this study, *Pseudomonas aeruginosa* was found to be the most frequently isolated, along with *Staphylococcus aureus* as the second-most common isolate with 423 samples in the Free State and 15 in the Northern Cape. This is also the leading bacterium in the Northern Cape. This clearly shows that the Free State carried the overwhelming burden of the isolates. The sample size decreased slightly in the Free State for the bacterium, while in the Northern Cape the sample size also decreased to 0 samples post-COVID-19.

Proteus mirabilis showed a steady increase in sample size in the Free State over the three timeframes (30, 49 and 100 samples) and the Northern Cape showed the opposite trajectory in sample size since the sample size decreased (9, 0 and 1 samples).

Acinetobacter baumannii had a decrease in sample size in the Free State during COVID-19 (32 to 27) and the sample size then increased post-COVID-19 (64). The Northern Cape had a much smaller sample size compared to the Free State, with 8 samples pre-COVID-19 and only one sample during COVID-19.

Overall, *Klebsiella pneumoniae subsp. pneumoniae* was isolated 80 times in the Free State, which is a lot more than the 2 samples that were isolated in the Northern Cape. The sample size decreased from 40 to 13 during COVID-19 in the Free State and then increased slightly post-COVID-19 to 27 samples. In the Northern Cape there were only two samples pre-COVID-19. *Streptococcus pyogenes* showed a similar trajectory to *Klebsiella pneumoniae*. The resistance decreased in the Free State over the three timeframes (23, 22 and 6 samples) while in the Northern Cape there were only 2 samples pre-COVID-19.

The sample size for *Escherichia coli* increased gradually over the three timeframes in the Free State (6, 11 and 20 samples) while there was only one sample pre-COVID-19 in the Northern Cape. *Providencia stuartii* and *Enterobacter cloacae subsp. cloacae* had no samples in the Northern Cape. The sample size for *Providencia stuartii* decreased during COVID-19 (from 22 to 1 sample) and then increased slightly post-COVID-19 (3 samples). The sample size for *Enterobacter cloacae subsp. cloacae* increased during COVID-19 in the Free State (from 6 to 12) and then decreased again post-COVID-19 to only one sample.

The sample size for *Serratia marcescens* in the Free State decreased during COVID-19 (from 12 to 1) and then increased again slightly post-COVID-19 (3). There were only two samples in the Northern Cape pre-COVID-19. There were no samples for *Enterococcus faecalis* in the Northern Cape, while there were 18 samples in total for the Free State. The sample size decreased gradually over the three timeframes (11, 5 and 2 samples).

There were only 11 samples for *Pseudomonas putida* post-COVID-19 in the Free State. This is still much higher than the total sample size of two for the Northern Cape. There was one sample pre- and during COVID-19 in the Northern Cape. There were only 12 samples for *Citrobacter freundii* pre-COVID-19 in the Free State and no samples in the Northern Cape. The sample size decreased for *Enterobacter cloacae complex* from 6 samples pre-COVID-19 to 4 samples post-COVID-19 in the Free State. However, in the Northern Cape there was only one sample pre-COVID-19. The results for the sample size of *Enterobacter aerogenes* are similar to those of *Enterobacter cloacae complex*. The sample size decreased from 6 pre-COVID-19 to 3 post-COVID-19 in the Free State, while there was only one sample pre-COVID-19 in the Northern Cape.

Table 2: Resistance pattern of bacteria in the Free State

FREE STATE BACTERIAL SENSITIVITY RESULTS									
	PRE-COVID-19			DURING COVID-19			POST-COVID-19		
	SENSITIVE	RESISTANT	TOTAL	SENSITIVE	RESISTANT	TOTAL	SENSITIVE	RESISTANT	TOTAL
PSEUDOMONAS AERUGINOSA	1204 (62.61)	719 (37.39)	1923	556 (42.51)	752 (57.49)	1308	1230 (50.51)	1205 (49.49)	2435
STAPHYLOCOCCUS AUREUS	1584 (67.81)	752 (32.19)	2336	1216 (67.97)	573 (32.03)	1789	1202 (78.10)	337 (21.90)	1539
PROTEUS MIRABILIS	398 (74.53)	136 (25.47)	534	678 (76.27)	211 (23.73)	889	1184 (79.95)	297 (20.05)	1481
ACINETOBACTER BAUMANNII	77 (21.21)	286 (78.79)	363	53 (15.59)	287 (84.41)	340	84 (11.11)	672 (88.89)	756
KLEBSIELLA PNEUMONIAE	408 (63.35)	236 (36.65)	644	112 (46.67)	128 (53.33)	240	260 (54.97)	213 (45.03)	473
ESCHERICHIA COLI	84 (78.50)	23 (21.50)	107	196 (96.08)	8 (3.92)	204	282 (90.38)	30 (9.62)	312
STREPTOCOCCUS PYOGENES	232 (91.34)	22 (8.66)	254	189 (94.97)	10 (5.03)	199	36 (100.00)	0 (0.00)	36
PROVIDENCIA STUARTII	223 (57.47)	165 (42.53)	388	9 (50.00)	9 (50.00)	18	30 (58.82)	21 (41.18)	51
ENTEROBACTER CLOACAE SUBSP CLOACAE	66 (61.68)	41 (38.32)	107	136 (62.10)	83 (37.90)	219	12 (80.00)	3 (20.00)	15
SERRATIA MARCESCENS	126 (63.32)	73 (36.68)	199	11 (64.71)	6 (35.29)	17	30 (66.67)	15 (33.33)	45
CITROBACTER FREUNDII	180	48	228	0	0	0	0	0	0

	(78.95)	(21.05)		(0.00)	(0.00)		(0.00)	(0.00)	
ENTEROBACTER CLOACAE COMPLEX	81 (72.97)	30 (27.03)	111	0 (0.00)	0 (0.00)	0	50 (76.92)	15 (23.08)	65
ENTEROBACTER AEROGENES	66 (57.89)	48 (42.11)	114	0 (0.00)	0 (0.00)	0	33 (68.75)	15 (31.25)	48
ENTEROCOCCUS FAECALIS	95 (82.61)	20 (17.39)	115	33 (78.57)	9 (21.43)	42	6 (100.00)	0 (0.00)	6
KLEBSIELLA OXYTOCA	0 (0.00)	0 (0.00)	0	119 (94.44)	7 (5.56)	126	0 (0.00)	0 (0.00)	0
ACINETOBACTER BAUMANNII COMPLEX	0 (0.00)	0 (0.00)	0	0 (0.00)	0 (0.00)	0	22 (18.03)	100 (81.97)	122
OTHER BACTERIA	4824 (64.99)	2599 (35.01)	7423	3308 (61.36)	2083 (38.64)	5391	4461 (60.41)	2923 (39.59)	7384
TOTAL	**9648 (64.99)**	**5198 (35.01)**	**14846**	**6616 (61.36)**	**4166 (38.64)**	**10782**	**8922 (60.41)**	**5846 (39.59)**	**14768**

The bacterial isolates were tested for antimicrobial sensitivity, and the results are represented in Table 2. Pre-COVID-19, a total of 14 846 results were obtained, of which 35.01% were resistant and 64.99% were sensitive. During COVID-19, a total of 10 782 results were obtained, of which 61.36% were sensitive and 38.64% were resistant. Post-COVID-19, a total of 14 768 results were obtained, of which 60.41% were sensitive and 39.59% were resistant. Overall, it is clear that there is resistance across all three time periods that are below 40%.

Pseudomonas aeruginosa is the organism with the highest number of samples in the Free State. Pre-COVID-19, the resistance was 37.4% and this increased drastically to 57.5% during the COVID-19 period. The resistance decreased slightly post-COVID-19 to 49.5%, but it did not decrease back to the percentage it was before COVID-19.

Staphylococcus aureus had an interesting trajectory. The resistance pre-COVID-19 was 32.2% and remained stable during COVID-19 at 32%, but there was a drastic drop in resistance post-COVID-19 to 21.9%. The decrease in resistance could be due to better antimicrobial stewardship programmes or due to the increased hygiene measures during the pandemic.

Proteus mirabilis, however, showed a steady decrease in resistance over the three timeframes from 25.5% pre-COVID-19, to 23.7% during COVID-19 and 20.1% post-COVID-19. These infections could possibly be due to cross-contamination from urinary tract infections and the decrease in resistance could also be due to improved hygiene during the pandemic.

The results for *Acinetobacter baumannii* showed an increasing trend over these three timeframes. Pre-COVID-19 the resistance was 78.8%, which is alarmingly high. During COVID-19, this rose to 84.4% and even further post-COVID-19 to 88.9%. This bacterium is seen as a critical threat due to the consistent high levels of multidrug resistance.

Klebsiella pneumoniae also had an increase in resistance during the pandemic and increased from 36.7% to 53.3% but dropped slightly to 45% post-COVID-19. The resistance remained high across all three timeframes. However, even though it decreased post-COVID-19, it still did not go back to the levels it had been before the pandemic.

The resistance of *Escherichia coli* increased during COVID-19 (78.5% to 96.08%) and decreased slightly post-COVID-19 (90.38%). The resistance remained alarmingly high across all three the timeframes and even though the resistance decreased, it is still above the level it had been before the pandemic.

Streptococcus pyogenes had quite a low resistance level during all three timeframes. The level decreased during COVID-19 (8.66% to 5.03%) and the level decreased even further to 0% post-COVID-19. The decrease in resistance could also be due to the decrease in the sample size over the three timeframes.

The resistance of *Providencia stuartii* increased during COVID-19 (42.53% to 50%), but post-COVID-19 the level decreased even below the resistance level pre-COVID-19 (41.18%). Even though the resistance level decreased, the high level of resistance is still quite high and still concerning.

The *Enterobacter species (E.cloacae, E cloacae complex* and *E.aerogenes*) had fluctuating levels of resistance with moderate to high levels of resistance*. E.cloacae subsp. Cloacae* had a relatively stable resistance during COVID-19 (62%), while *E.aerogenes* had quite a decrease in resistance (31.25%) post-COVID-19.

Serratia marcescens had a relatively stable level of resistance across all three timeframes. The resistance decreased slightly during COVID-19 (35.29 to 36.68%), but the resistance then decreased slightly post-COVID-19 to 33.33%. Before COVID-19, *Citrobacter freundii* had a 21.05% resistance, which then decreased during and post-COVID-19 to 0%. The decrease in resistance, however, is due to no samples testing positively for the organism in these two timeframes.

Enterococcus faecalis had a relatively low level of resistance during all three timeframes. The resistance level increased slightly during COVID-19 (17.39% to 21.43%), but the resistance decreased again post-COVID-19 to 0%. The 0% in resistance, however, is due to no samples that tested positively for the organism during this timeframe post-COVID-19.

Klebsiella oxytoca had no samples that tested positively for the organism before and after COVID-19. The resistance level, however, was quite low during COVID-19, since it was 5.56%. *Acinetobacter baumannii* had no samples that tested positively before and during COVID-19, but post-COVID-19, the resistance level was extremely high at 81.97%.

All of the other bacteria with low sample sizes (less than 10) were grouped together. The resistance level decreased gradually over all three timeframes: 64.99% pre-COVID, 61.36% during, and 60.41% post-COVID. There was an overall rise in the antimicrobial resistance overall from pre- to post-COVID-19.

Table 3: Resistance pattern of bacteria in the Northern Cape

NORTHERN-CAPE BACTERIAL SENSITIVITY RESULTS									
	PRE-COVID-19			**DURING COVID-19**			**POST-COVID-19**		
	SENSITIVE	**RESISTANT**	**TOTAL**	**SENSITIVE**	**RESISTANT**	**TOTAL**	**SENSITIVE**	**RESISTANT**	**TOTAL**
PSEUDOMONAS AERUGINOSA	94 (65.28)	50 (34.72)	**144**	0 (0.00)	0 (0.00)	**0**	0 (0.00)	0 (0.00)	**0**
STAPHYLOCOCCUS AUREUS	196 (94.23)	12 (5.77)	**208**	7 (50)	7 (50)	**14**	0 (0.00)	0 (0.00)	**0**
PROTEUS MIRABILIS	108 (68.35)	50 (31.65)	**158**	0 (0.00)	0 (0.00)	**0**	15 (78.95)	4 (21.05)	**19**
ACINETOBACTER BAUMANNII	18 (17.65)	84 (82.35)	**102**	2 (18.18)	9 (81.82)	**11**	0 (0.00)	0 (0.00)	**0**
KLEBSIELLA PNEUMONIAE	32 (88.89)	4 (11.11)	**36**	0 (0.00)	0 (0.00)	**0**	0 (0.00)	0 (0.00)	**0**
ESCHERICHIA COLI	16 (88.89)	2 (11.11)	**18**	0 (0.00)	0 (0.00)	**0**	0 (0.00)	0 (0.00)	**0**
STREPTOCOCCUS PYOGENES	4 (100)	0	**4**	0 (0.00)	0 (0.00)	**0**	0 (0.00)	0 (0.00)	**0**
SERRATIA MARCESCENS	16 (72.73)	6 (27.27)	**22**	0 (0.00)	0 (0.00)	**0**	0 (0.00)	0 (0.00)	**0**

ENTEROBACTER CLOACAE COMPLEX	13 (76.47)	4 (23.53)	**17**	0 (0.00)	0 (0.00)	**0**	0 (0.00)	0 (0.00)	**0**
ENTEROBACTER AEROGENES	14 (82.35)	3 (17.65)	**17**	0 (0.00)	0 (0.00)	**0**	0 (0.00)	0 (0.00)	**0**
OTHER BACTERIA	56 (80)	14 (20)	**70**	8 (72.73)	3 (27.27)	**11**	0 (0.00)	0 (0.00)	**0**
TOTAL	**567 (71.23)**	**229 (28.77)**	**796**	**17 (47.22)**	**19 (52.78)**	**36**	**15 (78.95)**	**4 (21.05)**	**19**

Table 3 shows the Northern Cape resistance profile of the different bacterial isolates for the three different timeframes. The Free State showed a much higher number of samples than the Northern Cape, since the Northern Cape only had 57 samples compared to the 1 549 samples of the Free State. Even though the sample size is much smaller, it still gives a picture of AMR in the Northern Cape.

Pseudomonas aeruginosa showed a 34.72% resistance pre-COVID-19 in the Northern Cape. This value could not be compared to, during, or post-COVID-19, since no samples tested positively for this bacterium in these two timeframes. *Staphylococcus aureus* showed a 5.77% resistance pre-COVID-19, which is considerably low. During COVID-19, the bacterium had an increase in resistance (50%), but the resistance post-COVID-19 was 0%, since there were no isolates post-COVID-19.

The resistance for *Proteus mirabilis* decreased slightly during COVID-19 (from 31.65% to 21.05%). However, there were no isolates for the timeframe post-COVID-19 to compare these results with. *Acinetobacter baumannii* had the highest level of resistance pre-COVID-19 (82.4%). During COVID-19, the resistance decreased slightly (81.82%) in the Northern Cape, which is still extremely high. Post-COVID-19, however, no samples tested positively for *Acinetobacter baumannii*.

Klebsiella pneumoniae and *Escherichia coli* both had a relatively low level of resistance (11.11%) pre-COVID-19, but during and post-COVID-19, no samples tested positively for the bacterium and therefore the results could not be compared. *Streptococcus pyogenes* had no samples that tested positively for this bacterium in any of the three timeframes and the resistance level could not be evaluated.

Serrata marcescens, Enterobacter cloacae complex and Enterobacter aerogenes all three had a relatively low level of resistance during COVID-19 (27.27%, 23.53% and 17.65%, respectively). During and post-COVID-19, no samples tested positively for these bacteria, so the results could not be compared. For all of the other bacteria there was a relatively low level of resistance pre-COVID-19 at 20%, which then increased during COVID-19 to 27.27%. Post-COVID-19, however, no samples tested positively for these isolates and therefore the results could not be compared.

Out of the 796 isolates pre-COVID-19 in the Northern Cape there was a 28.77% resistance. During COVID-19, the sample size declined to only 36 isolates and the resistance level rose to 52.78%. Post-COVID-19, the sample size was extremely low at only 19 samples and the resistance decreased to 21.05%.

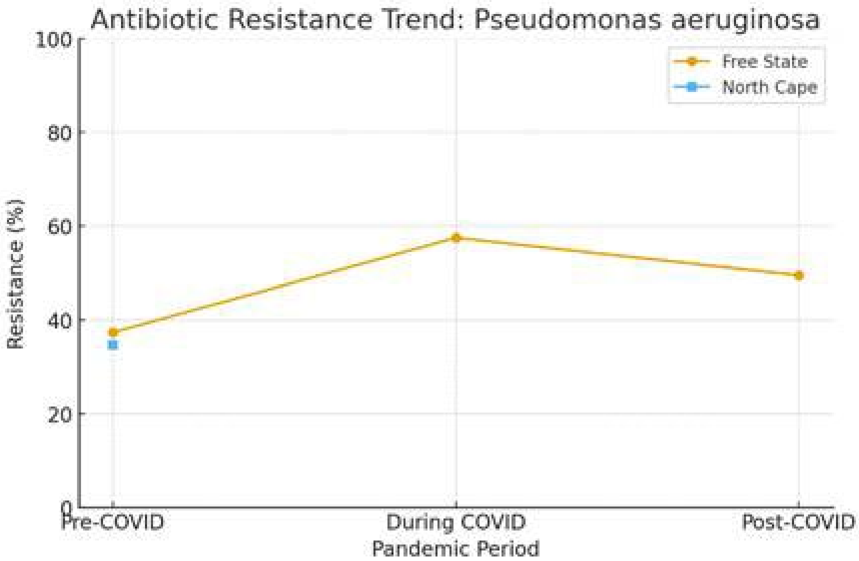

Figure 2: Antibiotic resistance trend of Pseudomonas aeruginosa

Pseudomonas aeruginosa had a much larger sample size in the Free State compared to the Northern Cape. The resistance in Free State pre-COVID-19 was 37.39%, which was slightly higher than the Northern Cape at 34.72%. During and post-COVID-19 no samples that tested positively for the bacterium in the Northern Cape; therefore, there are no results to compare with the results of the Free State. While the Free State shows a clear problem in resistance for this organism, the Northern Cape lacks enough data to make the long-term trends clear.

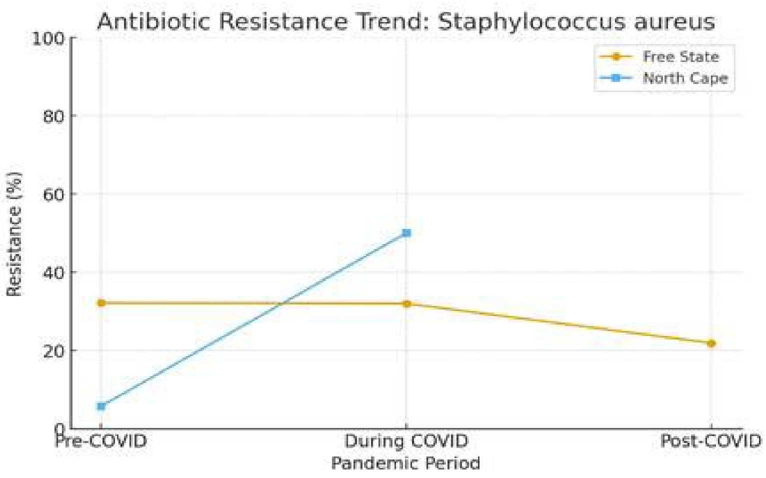

Figure 3: Antibiotics resistance trend of Staphylococcus aureus

Staphylococcus aureus had two completely different resistance trends for the two provinces. In the Free State the resistance pre-COVID-19 was 32.19%, which is higher than that of the Northern Cape at 5.77%. The increase rose sharply during COVID-19 for the Northern Cape (50%) but then decreased slightly for the Free State (32.03%). The resistance of the Free State post-COVID-19 decreased even further (21.90%), but this could not be compared to the resistance in the Northern Cape due to the absence of samples that tested positively for the isolate.

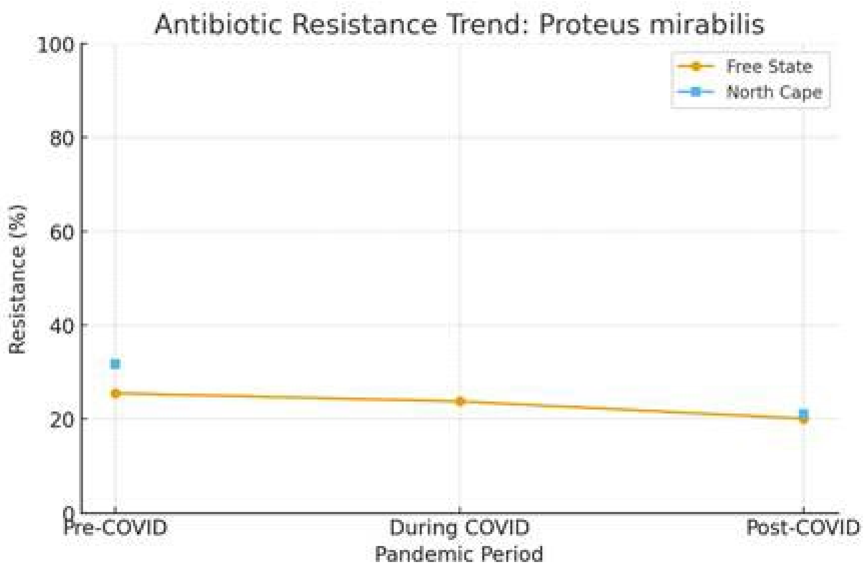

Figure 4: Antibiotic resistance trend of Proteus mirabilis

The resistance for *Proteus mirabilis* in the Free State was slightly lower than that of the Northern Cape when looking at the overall graph. The resistance pre-COVID-19 was lower for the Free State (25.47%) compared to the Northern Cape (31.65%). The resistance of the Free State during COVID-19 (23.73%) could not be compared to that of the Northern Cape, since no samples tested positively for the isolate. The resistance for the Free State decreased even further post-COVID-19 (20.05%), which remains lower than that of the Northern Cape (21.05%).

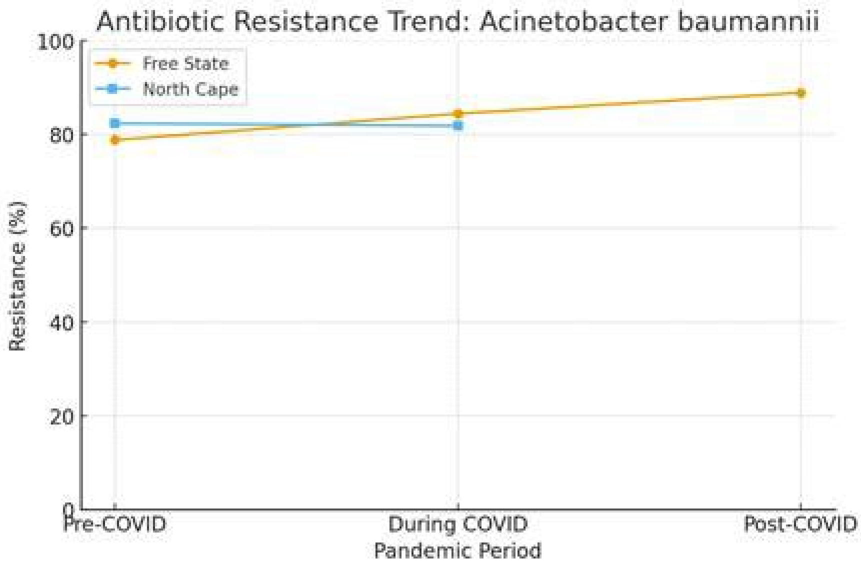

Figure 5: Antibiotic resistance trend of Acinetobacter baumannii

The resistance of *Acinetobacter baumannii* remained alarmingly high across all three timeframes for both provinces. Pre-COVID-19, the resistance for the Free State (78.79%) was slightly lower than the resistance of the Northern Cape (82.35%). The resistance for the Free State increased (84.41%) during COVID-19, while the resistance for the Northern Cape decreased slightly to 81.82%. The resistance of the Free State post-COVID-19 increased even further (88.89%), but this could not be compared to the Northern Cape, since there were no samples that tested positively for the bacterium.

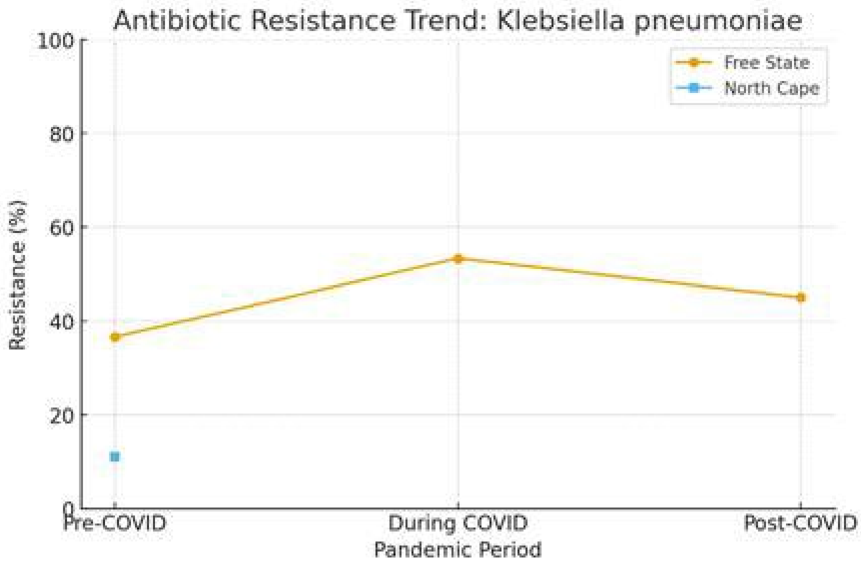

Figure 6: Antibiotic resistance trend of Klebsiella pneumoniae

The resistance of *Klebsiella pneumoniae* was quite high for the Free State pre-COVID-19 (36.65%) compared to the Northern Cape at 11.11%. The lack of samples during and post-COVID-19 for the Northern Cape makes it impossible to compare the resistance of the two provinces. However, the resistance increased in the Free State during COVID-19 (53.33%) and then decreased slightly post-COVID-19 (45.03%).

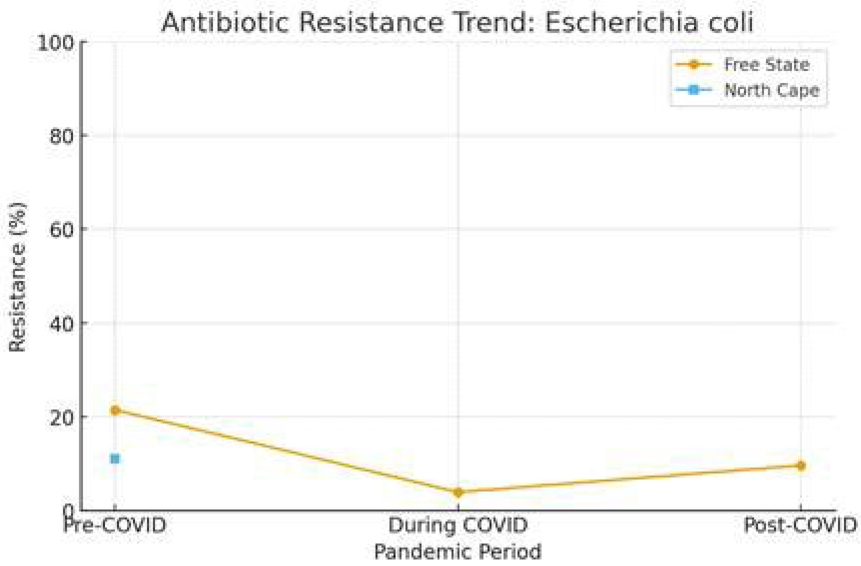

Figure 7: Antibiotic resistance trend of Escherichia coli

The resistance for *Escherichia coli* was slightly higher pre-COVID-19 in the Free State (21.50%) compared to the Northern Cape (11.11%). The resistance decreased tremendously during COVID-19 in the Free State (3.92%), but this could not be compared to the results of the Northern Cape, since there were no samples that tested positively for this bacterium during and post-COVID-19. The resistance increased slightly post-COVID-19 for the Free State (9.62%).

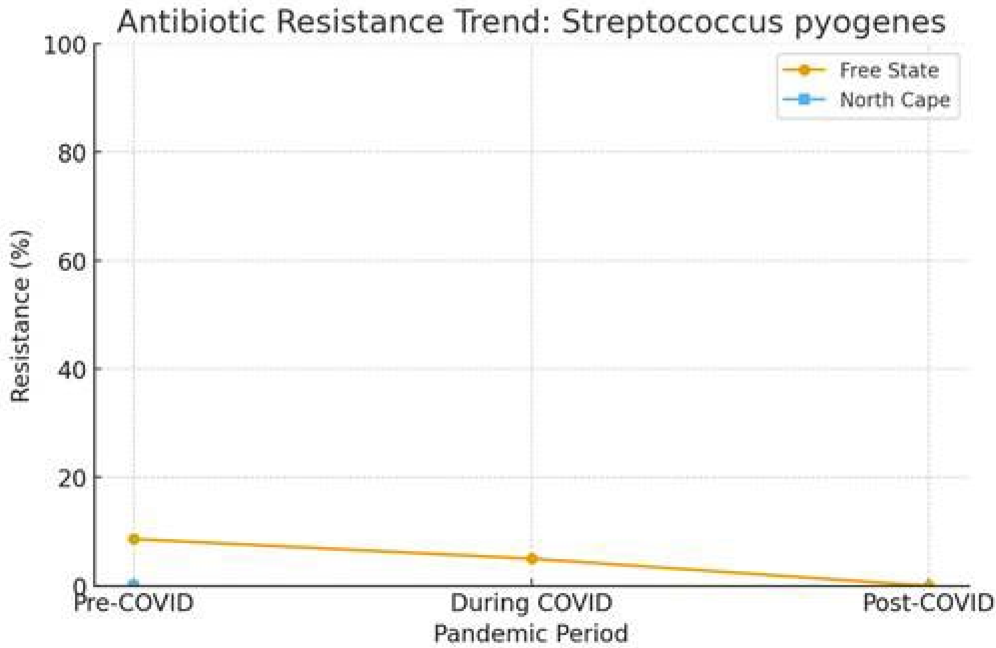

Figure 8: Antibiotic resistance trend of Streptococcus pyogenes

The resistance for *Streptococcus pyogenes* in the Free State showed a steady decline over the three timeframes (8.66% to 5.03%). There were, however, no samples that tested positively for the organism post-COVID-19 and that is why the resistance is 0%. The results of the Free State cannot be compared to those of the Northern Cape, since no samples tested positively for the bacterium in the Northern Cape.

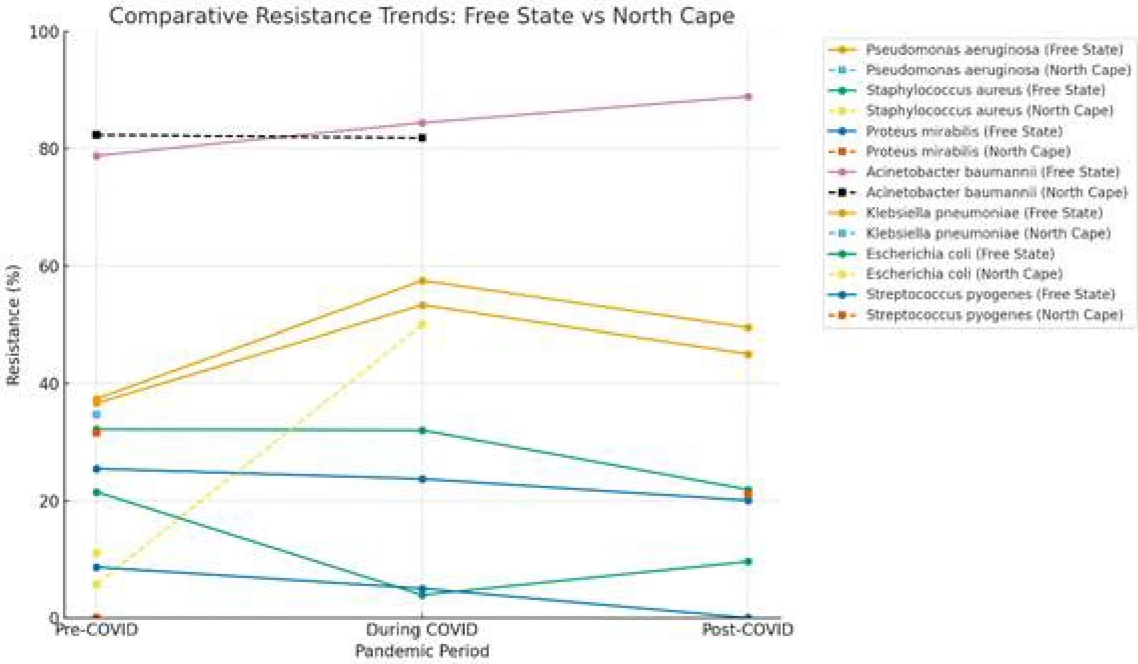

Figure 9: Antibiotic resistance trend of the Northern Cape and Free State

In this graph it is clear that *Acinetobacter baumannii* maintained the highest resistance trend in both provinces (above 80%). The resistance of *Pseudomonas aeruginosa* and *Klebsiella pneumoniae* were much higher in the Free State than in the Northern Cape. The resistance of *Staphylococcus aureus* remained moderate in the Free State while the resistance spiked to 50% during COVID-19 in the Northern Cape.

CHAPTER 5: DISCUSSION AND CONCLUSION

5.1 DISCUSSION

This study was conducted to determine the resistance profile of the bacteria that were isolated from the burn units of the Northern Cape and Free State and then to compare how the resistance pattern changed pre-pandemic (1 March 2018 until 30 February 2020), during the pandemic (1 March 2020 until 30 June 2022), and post-pandemic (1 July 2022 until 31 July 2024). Two objectives were set out for this study; firstly, to determine the bacterial resistance profile of the organisms from the swabs that were sent to the laboratories from the burn units of the public hospitals in the Free State and Northern Cape before, during and after the COVID-19 pandemic; secondly, to determine how the years before, during and after the pandemic compared to one another in terms of the bacterial resistance profile from the swabs that were sent from the burn units to the laboratories. Overall, the findings confirmed that AMR is still an escalating problem in South Africa. This was accelerated by the pandemic while the level post-pandemic has not decreased back to the level's pre-pandemic.

The overall findings shows that COVID-19 had an indirect impact on the microbial ecology of the burn units in these hospitals due to the fact that the resistance levels of the bacteria did not return to the levels it was pre-COVID-19. Due to the loss of the skin barrier on these burn patients and the necrotic tissue, an ideal environment is created for opportunistic infections to occur along with the invasive devices used on these patients (Gulumbe et al., 2023). The results of this study, along with the already existing literature, suggest that COVID-19 amplified the AMR crisis by stopping the antimicrobial stewardship in South Africa and increasing the use of broad-spectrum antibiotics (Khan et al., 2022). This is shown by the increase in resistance that was seen during COVID-19 in the Free State (35.01% to 38.64%) and then the further increase post-COVID-19 (39.59%). The resistance also increased during COVID-19 in the Northern Cape (28.77% to 52.78%), but the levels decreased again post-COVID-19 (21.05%). The decrease that was seen post-COVID-19 in the Northern Cape could be caused by the small sample size of only 19 samples for this timeframe.

As previously stated by literature, during the COVID-19 pandemic there was an increase in antibiotic prescriptions without the necessary testing to confirm the type of infection. Due to the threat of the pandemic and the prioritization of COVID-19 sample testing, the antimicrobial surveillance programs were sidelined, causing a wide range of resistant bacterial strains to dominate in the hospital environments. The results that were stated in this research reinforced the world-wide concern that the pandemic was a catalyst that caused the current AMR crisis to accelerate even more (Langford et al., 2023). This is supported by the results that were seen especially in the Free State. An increase in resistance was seen during COVID-19 in the Free State (35.01% to 38.64%) and then a further increase post-COVID-19 (39.59%). The continual high levels of resistance that was seen post-COVID-19 suggest that the results of the ecological changes during the pandemic were not temporary, but this may have altered the baseline of the resistance in the South African burn units permanently. The resistance level in the Free State continued to increase post-COVID-19 (39.59%) and remained high. The resistance decreased post-COVID-19 in the Northern Cape (21.05%), but the decrease could be caused by the low sample size for this timeframe.

When comparing the sample size of the two provinces it is quite clear that the Free State had a much larger sample size (1 549 samples) compared to the Northern Cape (57 samples). However, it was still clear that the resistance in the Free State increased during the pandemic (from 35% to 38.6%) and continued to increase post-COVID-19 (39.6%). The resistance in the Northern Cape showed a different pattern, since the resistance rose drastically during COVID-19 (from 28.8% to 52.8%) and then decreased again sharply post-COVID-19 (21.1%). The decrease in resistance for the Northern Cape could be misleading, since there was only one sample post-COVID-19. The increase in resistance during the pandemic is consistent with the literature, which states that there was an increase in the samples sent to laboratories, increased broad-spectrum prescriptions and reduced ASP (Khaznadar et al., 2023).

It is important to take note of the difference in sample size between the Northern Cape and the Free State. The larger sample size of the Free State provides a more stable and reliable resistance pattern and organism prevalence, while the smaller size in the Northern Cape could exaggerate certain fluctuations in results (Farley et al., 2018). The hospitals in the Free State received higher patient loads due to the large population, which increased the risk of the spread of AMR (Davids, 2021). The Northern Cape is more rural with a much smaller population compared to the Free State.

The difference in the sample size for the two provinces could also be caused by a difference in staffing levels, healthcare infrastructure and laboratory testing capacity. Since the hospitals in the Free State serve a larger population size it could also mean that they have a more constant and larger flow of samples, which could also explain the larger sample size. The Northern Cape has smaller hospitals and a smaller population size and during the pandemic, these hospitals could have been overwhelmed, focusing solely on COVID-19 cases, which could contribute to the small sample size. When comparing the results of the Northern Cape it should be done with caution, since it does not necessarily reflect the decline in bacterial prevalence, but more likely the surveillance gaps in this province. However, the increase in resistance that is seen in the Northern Cape during COVID-19 still demonstrates how even a slight misuse in antibiotic use could still rapidly elevate the resistance levels of different bacteria. The large difference in sample size highlights the unequal burden of AMR across South Africa, especially that the more rural provinces might be more vulnerable to the shifts in resistance (Health, 2018).

Pseudomonas aeruginosa and *Staphylococcus aureus* were the most commonly found isolates from the swabs from burn wounds due to their ability to colonize the skin. These are well-documented organisms in burn units (Gulumbe et al., 2023). *Proteus mirabilis*, *Acinetobacter baumannii* and *Klebsiella pneumoniae* were also prevalent in the burn wound swabs, due to these bacteria causing hospital-acquired infections (Khaznadar et al., 2023).

Pseudomonas aeruginosa resistance rose sharply during COVID-19 (from 37.39% to 57.49%) in the Free State. These results confirm what has been said in the literature that during COVID-19 there was an increase in the prescriptions of broad-spectrum antibiotics without the confirmation of bacterial co-infections, which then caused an increase in resistance (Comelli et al., 2022). Post-COVID-19, the results only decreased slightly (49.49%), but it did not reach pre-COVID-19 levels, which shows the persistence of these bacteria in the hospital environments. In the Northern Cape the resistance was similar to the Free State pre-COVID-19 (34.72%), but there were no isolates during and post-COVID-19 to compare this result with.

It is quite concerning that the resistance continued to be high post-COVID-19, since this could suggest that the resistant strains of *Pseudomonas aeruginosa* have become embedded in the hospital environment and are difficult to eradicate. Since the levels post-COVID-19 has not decreased to the baseline level of pre-COVID-19, this raises concerns about whether the current infection control measures are effective. Moreover, it highlights the need for antimicrobial stewardship programmes to be re-instated.

The increase in resistance that is seen for *Pseudomonas aeruginosa* is concerning due to the high resistance to many different antibiotics (Med, 2019). This organism commonly causes septicaemia in burn-wound patients and, if not treated promptly, it could lead to a life-threatening infection (Dadgoster, 2019). The remaining high levels of resistance seen post-COVID-19 indicate that not just an ecological shift had occurred, but that this shift has now been entrenched in these environments, which could be due to the resistant colonies that established dominance in the hospital environments (Khaznadar et al., 2023). The findings for this organism confirm global concerns, since the organism is known to thrive in a hospital environment and evade antibiotic therapy (Comelli et al., 2022).

The trajectory of *Staphylococcus aureus* is slightly different. The resistance remained constant pre- and during COVID-19 (32.2% and 32%) in the Free State but then declined drastically post-COVID-19 (21.9%). This decrease in resistance post-COVID-19 could be caused by the increase in infection control measurements that were put in place with hygiene control during the pandemic (Shomuyiwa et al., 2022). However, the resistance increased from 5.77% to 50% during the pandemic in the Northern Cape. Post-COVID-19 no samples tested positively for *Staphylococcus aureus* and therefore the resistance was 0%.

The decline in resistance that is seen in the Free State post-COVID-19 could reflect the effect of the use of personal protective equipment such as masks, increased hand hygiene and environmental decontamination, especially in hospitals during this pandemic (Khan et al., 2022). It is known that *Methicillin-resistant Staphylococcus aureus* is one of the most commonly found hospital-acquired pathogens, especially in South Africa, but strict hygiene protocols could decrease the transmission (Farley et al., 2018). The increase in resistance that was seen during the pandemic in the Northern Cape is concerning, but this could be due to a single outbreak in a facility and not necessarily a provincial trend when looking at the small sample size. The disappearance in isolates in the Northern Cape could just reflect the absence in tested samples and not necessarily the eradication of the organism (National Library of Medicine, 2021).

The stricter patient isolation measures and hospital hygiene that were put in place during the pandemic could be the reason for the decline in the resistance seen in the Free State. These measures could have prevented the transmission of *methicillin-resistant Staphylococcus aureus,* which could cause a permanent decrease in the prevalence of this resistant organism. However, the sudden spike in the Northern Cape to 50% during the pandemic could indicate a sudden outbreak in one hospital or the burn unit of one of these hospitals, amplified by the small sample size of the Northern Cape. Due to no samples post-COVID-19 in the Northern Cape, it is not possible to determine if this sudden increase is an anomaly or a larger regional increase.

Acinetobacter baumannii had the highest resistance in the Free State (pre-COVID-19 78.79%, during 84.41%, post 88.89%) and Northern Cape (pre-COVID-19 82.35%, during 81.82%) when compared to other bacteria. This organism's resistance reflects its persistent presence in the hospital environment along with its ability to colonize medical equipment and its resistance to a wide variety of antimicrobials. Therefore, the resistance was expected, and these results reflect other reports (Khoshbakht et al., 2022). This organism has already been mentioned as a high-priority pathogen by the WHO (Med, 2019).

The constantly high resistance of *Acinetobacter baumannii* as stated is quite concerning, since it appears that the bacterium is entrenched in the hospital environment, particularly the burn units (Dadgoster, 2019). The bacterium forms biofilms on surfaces and it is therefore quite difficult to eradicate (Khaznadar et al., 2023). The nearly 90% resistance that is seen in the Free State post-COVID-19 shows that this is a threat due to the limitations in therapeutic options (Med, 2019). *Carbapenem*-resistant *Acinetobacter baumannii* outbreaks in South Africa have been linked to shortages of single-use equipment, also due to poor infection control and prevention (National Library of Medicine, 2021). The presence of the pandemic likely aggravated this, since most of the resources and staff were used for COVID-19 cases and therefore the burn units could have been under-monitored. The persistent increase in resistance post-COVID-19 suggests that the decrease in antimicrobial stewardship during the pandemic allowed resistant strains to dominate in the hospital environment (National Library of Medicine, 2021).

The same trend could be seen with *Klebsiella Pneumoniae* as was seen with *Acinetobacter baumannii* in the Free State, where the resistance increased drastically during COVID-19 (from 36.65% to 53.33%) and then decreased slightly post-COVID-19 (45.03%). As stated by the literature, there was an increase in the prescription of broad-spectrum antibiotics during the pandemic to patients, although bacterial infections were not confirmed, causing an increase in resistance during COVID-19. Post-COVID-19 the resistance decreased slightly as other organisms re-emerged and the excessive use of antibiotics decreased (Comelli et al., 2022). Due to the heavy reliance on carbapenems and cephalosporins during COVID-19, the rise in resistance is not surprising (Langford et al., 2023). This bacterium has already been classified by the WHO as a critical priority due to the bacterium's ability to disseminate the genes of carbapenems, which then cause the bacterium to gain resistance to last-line agents (Med, 2019). The slight decrease in resistance seen post-COVID-19 could be attributed to the normalization in prescription patterns in the hospitals, but the fact that the level is still above pre-COVID-19 levels confirms that this organism is resilient and difficult to eradicate (Djuikoue et al., 2023).

Klebsiella pneumoniae forms biofilms in the hospital environment as well as on devices that are used in the hospital setting. This is particularly concerning, since this bacterium is clearly embedded in the burn units of these hospitals and is particularly dangerous to these susceptible burn-wound patients (Djuikoue et al., 2023). The continual high levels of resistance seen post-COVID-19, even though the overprescription of antibiotics have decreased, could be due to environmental contamination in these hospitals. This shows that there is a need for active environmental surveillance along with the reinstatement of the antimicrobial stewardship programmes (Khan et al., 2022).

A completely opposite resistance could be seen with *Proteus mirabilis* where the resistance decreased slightly across the three timeframes. This could be caused by the fact that *Proteus mirabilis* is not a pathogen commonly seen in burn units when compared with *Pseudomonas aeruginosa* and *Staphylococcus aureus* (Khaznadar et al., 2023). When comparing the resistance of the other organisms, the results fluctuated across the three timeframes and also across all of the bacteria. The reason for this could, however, be due to the small sample sizes of these organisms.

Proteus mirabilis is more commonly seen with urinary tract infections, but it can become an opportunistic pathogen in wounds, especially where there is an increase in contamination risk or with prolonged catheterisation (Shomuyiwa et al., 2022). Most of the broad-spectrum antibiotics that were used during COVID-19 were for Gram-negative bacteria and therefore *Proteus mirabilis* could have experienced less selective pressure, causing the decrease in resistance (Langford et al., 2023). The lower prevalence of the bacterium in the burn units decreases the selective pressure for multidrug resistance. During the pandemic improved infection prevention measures were put in place. This could have improved catheter care, which decreased the cross-transmission to the burn wounds of the patients (Khadse, Ugemuge, & Singh, 2023).

Enterobacter cloacae complex and *Enterobacter aerogenes* are commonly seen environmental organisms that colonize medical devices, disinfectant solutions and even hospital plumbing. Once these organisms are found in an environment such as burn units, they can persist for long periods of time and cause outbreaks (Gulumbe et al., 2023). This pathogen is an opportunistic pathogen that can commonly be seen in immunocompromised patients, and that is why it is seen in this study with burn-wound patients (Med, 2019). These burn patients are repeatedly exposed to antibiotic use and prolonged hospitalization, which create the perfect environment for COVID-19 to thrive in (Langford et al., 2023).

Enterobacter cloacae subsp. cloacae showed a gradual decrease in resistance over the three timeframes in the Free State. The resistance decreased slightly during COVID-19 from 38.32% to 37.9% in the Free State and then decreased even further post-COVID-19 (20%). In the Northern Cape the resistance was 23.53% pre-COVID-19 and there were no further samples after this timeframe with which to compare this result. It is, however, close to the resistance level in the Free State. The resistance for *Enterobacter aerogenes* was quite similar. The resistance was high pre-COVID-19 in the Free State (42.11%), which then decreased post-COVID-19 (31.25%), but there were no samples during COVID-19 to show a clear trend. There were also only samples for pre-COVID-19 in the Northern Cape, and the resistance was slightly lower than that of the Free State (17.65%). The difference in resistance over the three timeframes and also between the two provinces could be due to the variations between the different wards and hospitals in infection control efficacy.

The resistance of *Escherichia coli* rose slightly during COVID-19, likely due to the overuse of antibiotics such as cephalosporins. Post-COVID-19, the levels decreased, but it did not go back to pre-COVID-19 levels, which could have been caused by a shift in resistant ecology (Farley et al., 2018). The presence of this bacterium is not commonly seen in burn wound swabs and could be associated with a secondary infection from urinary or gastrointestinal sources from devices such as catheters (Djuikoue et al., 2023). The trend in resistance that is seen corresponds with the literature that states that there has been a surge in extended-spectrum-beta-lactamase producing *Escherichia coli* during COVID-19, due to the increase in the use of antibiotics such as fluoroquinolone and cephalosporin (Khoshbakht et al., 2022). This is seen as a great challenge, since this bacterium is able to transfer its plasmids carrying resistance genes to other bacteria that are also Gram-negative such as *Proteus mirabilis* or *Klebsiella pneumoniae* (Khoshbakht et al., 2022).

Serratia spp. was less commonly isolated in this study, but the trend in resistance correlated with those of other Gram-negative pathogens, since there was an increase in resistance during COVID-19. This pathogen causes opportunistic infections and are commonly found in moist hospital environments. Although the small sample size limits the proper interpretation, the presence of this pathogen in the burn units shows that proper environmental hygiene and infection prevention have not been followed. This bacterium has been noted to contaminate soap dispensers and decontamination devices in hospital environments and since these devices are commonly used in burn units, this could be the source of the spread of the bacterium (Khaznadar et al., 2023).

When comparing the overall resistance across both provinces, the same trajectory can be seen as the resistance increased during the pandemic. In the Free State it rose slightly (35.01% to 38.64%) and continued to rise post-COVID-19 (39.59%). The Northern Cape, however, showed a slightly different trajectory post-COVID-19, as the resistance decreased (21.05%) even below pre-COVID-19 levels (28.77%). The results should be considered with caution due to the extremely low sample size during and especially post-COVID-19.

The increase in resistance during COVID-19 could be caused by the overuse of antibiotics and the pause that was placed on the ASP during this time (Langford et al., 2023). The fact that the overall resistance did not return to pre-COVID-19 levels could be caused by the consistent presence of these bacteria in hospitals, as they cause hospital-acquired infections that are not easy to eradicate (Comelli et al., 2022). These findings correspond with literature that confirms that COVID-19 produced ideal conditions for the prolonged increase in AMR (Shomuyiwa et al., 2022).

The findings of this study support the fact that *Pseudomonas aeruginosa* and *Acinetobacter baumannii* are MDR organisms that are entrenched in the burn units in hospitals. The burn patients are extremely vulnerable to these due to their extensive wound exposure, extended hospital stays and the use of invasive devices. The increase in resistance that was seen during COVID-19 is due to the overprescription of antibiotics during the pandemic and the halt that was put on the antimicrobial stewardship programmes (Khan et al., 2022).

The continual increase in resistance of the organism post-COVID-19 suggests that a long-term ecological shift has occurred in the hospital microbial population. It is known that the resistant strains found in hospitals are particularly difficult to eradicate, especially in high-risk environments such as burn units (Dadgoster, 2019). This highlights the urgency of the matter to firstly strengthen and then re-establish the antimicrobial stewardship programmes in South Africa (National Library of Medicine, 2021).

Overall, these findings across the multiple organisms showed that the COVID-19 pandemic placed stress on the antibiotic stewardship and infection prevention in South Africa, particularly in the burn units. Certain species such as *Acinetobacter* and *Klebsiella* showed an increase in resistance under the high levels of antibiotic use, while for other bacteria such as *Enterococcus* and *Proteus,* the resistance remained lower, but they still acted as reservoirs for the transfer of resistant genes. These results show that AMR is not one single problem, but a complex system that is influenced by environmental factors, antibiotic use and patient vulnerability (National Library of Medicine, 2021).

Since there are quite a few pathogens that have the ability to form biofilms in the hospital environment, patient wounds and even invasive devices, it is important to note that even if the number of unnecessary prescriptions are decreased to how it was before the pandemic these resistant bacteria can still remain in the hospital environment and cause infections. Therefore, while it is necessary to reinstate the antibiotic stewardship programmes in the hospitals, it is also important to add additional intervention strategies such as the use of antimicrobial dressing in burn wound patients and the regular cleaning of equipment that are used on these patients to prevent the further spread of the bacteria (Djuikoue et al., 2023).

5.2 CONCLUSIONS

Overall, this study has shown that during COVID-19, the AMR increased in the burn units of the Northern Cape and the Free State. Post-pandemic it is shown that the resistance levels remained higher than the levels pre-pandemic. The organisms that were the most concerning was *Pseudomonas aeruginosa*, *Acinetobacter baumannii*, and *Klebsiella pneumoniae* due to the persistence in elevated resistance levels. The resistance patterns varied across the different antimicrobial classes, with the most worrisome trends seen among the last-resort antimicrobials. This highlights the urgency of the matter to firstly strengthen and then re-establish the antimicrobial stewardship programmes in South Africa, and then also to prioritize the infection prevention measure in the burn units. If no intervention takes place, the resistant infections that are currently in these burn units will continue to threaten and cause further harm to these burn unit patients that are already vulnerable, as well as further strain the South African healthcare systems. Future research should be done to determine exactly what antibiotic classes are no longer effective against what bacteria and to determine what options of antibiotics are still available for different organisms. How antibiotics are used and to what extend should also be studied in South Africa to determine if and how these antibiotics are being misused.

5.3 LIMITATIONS

Two limitations for this study should be acknowledged. Firstly, since this study was retrospective, the data therefore relied on the laboratory submissions that were reduced due to the prioritization of the viral samples during COVID-19. Secondly, the small sample size of the Northern Cape during and after COVID-19 limited the comparison of the provinces.

REFERENCES

Barron, M. 2024, October 08. *American Society for Microbiology*. Retrieved from The Antimicrobial Resistance Pandemic: Breaking the Silence: https://asm.org/articles/2024/october/antimicrobial-resistance-pandemic-breaking-silence?utm_source=chatgpt.com

Centre for Disease Control and Prevention. 2023. *Laboratory Quality Assurance and Standardization Programmes*. Retrieved April 7, 2023, from: https://www.cdc.gov/labstandards/overview.html

Comelli, A., Genovese, C., Lombardi, A., Bobbio, C., Scudeller, L., Restelli, U., & Muscatello, A. 2022, August 29. *BMC*. Retrieved from Antimicrobial Resistance & Infection Control: https://aricjournal.biomedcentral.com/articles/10.1186/s13756-022-01152-5?utm_source=chatgpt.com

Dadgoster, P. 2019, December 20. *PubMed Central*. Retrieved from National Library of Medicine: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6929930/

Dall, C. 2025, January 15. *University of Minnesota*. Retrieved from Antimicrobial Stewardship, COVID-19: https://www.cidrap.umn.edu/antimicrobial-stewardship/report-highlights-how-covid-hindered-fight-against-antimicrobial?utm_source=chatgpt.com

Davids, N. 2021. *University Of Cape Town News*. Retrieved October 19, 2022, from: https://www.news.uct.ac.za/article/-2021-04-15-antibiotic-resistance-a-faceless-pandemic

Djuikoue, C. I., Djonkouh, W. Y., Bekolo, C. E., Wouambo, R. K., Founou, R. C., Djoulako, P. D., . . . Apalata, T. R. 2023, May 18. *MDPI*. Retrieved from Antibiotics: https://www.mdpi.com/2079-6382/12/5/929

Farley, E., Stewart, A., Davies, M.-A., Govind, M., van den Bergh, D., & Boyles, T. 2018. Antibiotic use and resistance: Knowledge, attitudes and perceptions among primary care prescribers in South Africa. *South African Medical Journal,12*.

Gulumbe, B. H., Sahal, M. R., Abdulraim, A., Faggo, A. A., Yusuf, Z. M., Sambo, K. H., . . . Lawan, K. A. 2023, September 13. *PubMed Central*. Retrieved from National Library of Medicine: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC10498429/

Health, N. D. 2018, November. *Surveillance for antimicrobial resistance and consumption of antibiotics in South Africa.* Retrieved October 05, 2022, from: https://www.knowledgehub.org.za/system/files/elibdownloads/2020-03/AMR%20Surveillance%20report%20South%20Africa%20-%20Nov2018.pdf

Khadse, S. N., Ugemuge, S. & Singh, C. 2023, December 04. *PubMed Central*. Retrieved from Impact of Antimicrobial Stewardship on Reducing Antimicrobial Resistance: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC10765068/

Khan, S., Bond, S. E., Bakhit, M., Hasan, S. S., Sadeq, A. A., Conway, B. R. & Aldeyab, M. A. 2022, November 11. *National Library of Medicine*. Retrieved from PubMed Central: https://pmc.ncbi.nlm.nih.gov/articles/PMC9686587/?utm_source=chatgpt.com#sec3-antibiotics-11-01600

Khaznadar, O., Khaznadar, F., Petrovic, A., Lucija, K., Loncar, A., Kolaric, T. O. . . . Smolic, M. 2023, May 31. *MDPI*. Retrieved from https://www.mdpi.com/2036-7481/14/2/52

Khoshbakht, R., Kabiri, M., Neshani, A., Khaksari, M. N., Sadrzadeh, S. M., Mousavi, S. M. . . . Ghaidel, M. 2022, October 01. *Assessment of antibiotic resistance changes during the Covid-19 pandemic in northeast Iran during 2020–2022: an epidemiological study*. Retrieved from Biomedical Central: https://aricjournal.biomedcentral.com/articles/10.1186/s13756-022-01159-y

Langford, B. J., Soucy, J.-P. R., Leung, V., So, M., Kwan, A. T., Portnoff, J. S., . . . Daneman, N. 2023, March 29. *National Library of Medicine*. Retrieved from Antibiotic Resistance associated with the COVID-19 pandemic: a systematic review and meta-analysis: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC9733301/

Med, O. J. 2019, May. *National Library of Medicine*. Retrieved from PubMed Central: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6505350/

Mendelson, M. 2017, August 08. *Keep taking the tablets: Three reasons to stay the full antibiotic course*. Retrieved from University of Cape Town News: https://www.news.uct.ac.za/article/-2017-08-08-keep-taking-the-tablets-three-reasons-to-stay-the-full-antibiotics-course

National Library of Medicine. 2021, September 28. Retrieved from South Africa's capacity to conduct antimicrobial stewardship: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8517762/

Republic of South Africa [RSA]. 2003. National Health Act 61 of 2003. Pretoria: Government Printers.

Republic of South Africa [RSA]. 2013. Personal Information Act no. 4 of 2013. Pretoria: Government Printers.

National Health Act 61 of 2003. Pretoria: Government Printers.

Shomuyiwa, O. D., Lucero-Prisno, D. E., Manirambona, E., Suleman, M. H., Rayan R. A., Huang, J. & Musa, S. S. 2022, August 8. *PubMed Central*. Retrieved from National Laboratory of Medicine: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC9358668/

Central University of Technology, Free State

APPENDIX A: LETTER TO ETHICS COMMITTEE

UNIVERSITY OF THE FREE STATE
UNIVERSITEIT VAN DIE VRYSTAAT
YUNIVESITHI YA FREISTATA

UFS HEALTH SCIENCES

Health Sciences Research Ethics Committee

28-May-2025

Dear **Mrs Chene Beetge**

Ethics Clearance: **The prevalence of multidrug-resistant antimicrobials in the burn units of three hospitals in the Free State and Northern Cape before, during and after the COVID-19 pandemic (1 March 2018 - 31 July 2024).**

Principal Investigator: **Mrs Chene Beetge**

Department: **CUT - Central University of Technology**

Submission Page

APPLICATION APPROVED

Please ensure that you read the whole document.

With reference to your application for ethical clearance with the Faculty of Health Sciences, I am pleased to inform you on behalf of the Health Sciences Research Ethics Committee that you have been granted ethical clearance for your project.

Your ethical clearance number, to be used in all correspondence is:**UFS-HSD2024/1262/2705**

The ethical clearance number is valid for research conducted for one year from issuance. Should you require more time to complete this research, please apply for an extension.

We request that any changes that may take place during the course of your research project be submitted to the HSREC for approval to ensure we are kept up to date with your progress and any ethical implications that may arise. This includes any serious adverse events and/or termination of the study.

A progress report should be submitted within one year of approval, and annually for long term studies. A final report should be submitted at the completion of the study.

Research conducted in any Department of Health facility: Researchers are required to sign and return the HSREC approval letters to the provincial Department of Health where they applied. It is also a requirement for researchers to submit electronic copies of their final research findings, and/or make a presentation of their findings and recommendations at departmental research days when and where indicated.

The HSREC functions in compliance with, but not limited to, the following documents and guidelines: The SA National Health Act. No. 61 of 2003; Ethics in Health Research: Principles, Structures and Processes (2015); SA GCP(2020); Declaration of Helsinki; The Belmont Report; The US Office of Human Research Protections 45 CFR 461 (for non-exempt research with human participants conducted or supported by the US Department of Health and Human Services- (HHS), 21 CFR 50, 21 CFR 56; CIOMS; ICH-GCP-E6 Sections 1-4; International Council for Harmonisation (ICH) Harmonised Guideline, Integrated Addendum to ICH E6(R1), Guideline for Good Clinical Practice (GCP) E6(R2), 2016, SAHPRA Guidelines as well as Laws and Regulations with regard to the Control of Medicines, Constitution of the HSREC of the Faculty of Health Sciences.

The Principal Investigator (PI) bears final responsibility for the RIMS application. In the event of any misconduct or improper activities perpetuated by a third party, the PI will be held vicariously liable. The HSREC will bear no responsibility or liability for any actions of a PI and/or third party or breach of confidentiality caused by the PI and/or third party.

For any questions or concerns, please feel free to contact HSREC Administration: 051-4012650/9860 or email EthicsFHS@ufs.ac.za.

Thank you for submitting this proposal for ethical clearance and we wish you every success with your research.

Yours Sincerely

Dr. C. Armour (Barrett)
Chairperson: Health Sciences Research Ethics Committee

Health Sciences Research Ethics Committee
T: +27 (0)51 401 2650/9860 | E: ethicsfhs@ufs.ac.za
IRB 00011992; REC 230408-011; IORG 0010096; FWA 00027947

APPENDIX B: REQUEST TO ACCESS NHLS RESOURCES

NATIONAL HEALTH LABORATORY SERVICE

Academic Affairs and Research
1 Modderfontein Road, Sandringham, 2031
Tel: +27 (0)11 555 0367/0406
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11 November 2024

Applicant: Chene' Beetge
Institution: Central University of Technology
E-mail Address: chenepieterse10102001@gmail.com
Cell: 071 474 9290

Project Title: The prevalence of multi drug resistant antimicrobials in the burn units of the public hospitals of the Free State and North Cape before, during and after the COVID-19 pandemic (1 March 2018-31 July 2024)

Reference Number: PR2455119

Research Application Type(s):
1. Request for Data

RE: APPROVAL LETTER: REQUEST TO ACCESS NHLS RESOURCES FOR RESEARCH PURPOSES

This letter serves to advise that the application requesting permission to conduct the above-mentioned research using the listed NHLS resources has been reviewed and **"Approved"**. Please note that the approval is granted **without undergoing the full internal peer review process** on the condition of the **urgency of the request and its time-sensitive nature, therefore further clarity may be required by the processing unit.** You are required to comply with the NHLS Research Material and Data Access Policy and requirements stated below.

1. All material and data requested shall be used as per the research protocol submitted to the NHLS and as approved by the relevant Health Research Ethics Committee (HREC) in South Africa.
2. Access to the NHLS material and/or data shall be limited to the minimum required for successful completion of the approved study and shall be made available ***without patient names and other patient identifiers (including, but not limited to, national identity numbers, hospital/clinic file numbers, addresses and telephone numbers)***.
3. Confidentiality shall be maintained at the participant and institutional level and there shall be no disclosure of personal information or confidential information.
4. Data and/or material shall not be shared with other parties unless approved by the NHLS
5. The material and/or data obtained from the NHLS shall be anonymised and not, for any reason, be used to track or recruit patients as no pre-approval/consent is obtained from patients.
6. Processes shall be discussed with the relevant NHLS departments (i.e. Corporate Data Warehouse (CDW), NHLS Laboratory Management, Operations Office, etc.) and agreed upon.
7. Any amendments to the study requirements, including the use of the material and/or data for purposes not initially disclosed to the NHLS) shall be cleared by an approved HREC and submitted to the NHLS for approval via the AARMS system – https://aarms.nhls.ac.za.
8. The NHLS shall be acknowledged as a source of material and/or data in any output, such as abstracts and journal articles, emanating from the project.
9. A final report of the research study and any published output resulting from this study shall be submitted to the NHLS via AARMS

Please note that this letter constitutes approval by the NHLS Academic Affairs and Research Office. The NHLS entities tasked with providing the material and/data may have additional requirements for access. Data related queries may be directed to NHLS CDW, email: zarina.sabat@nhls.ac.za; contact number: 011 386 6074 and sample related queries (if applicable) shall be directed to the relevant business manager.

Dr Babatyi Malope-Kgokong
National Manager: Academic Affairs and Research

APPENDIX C: UFS HSREC ETHICS COMMITTEE

FACULTY OF HEALTH AND ENVIRONMENTAL SCIENCES

30 May 2024

ATTN: UFS HSREC Ethics Committee

Re: Scientific Review; CHENE' BEETGE
Topic: The prevalence of multi drug resistant antimicrobials in the burn units of the public hospitals of the Free State and North Cape before, during and after the COVID-19 pandemic (1 March 2018-31 July 2024)

To Whom it may concern

This letter serves to confirm that the research protocol, titled, "***The prevalence of multi drug resistant antimicrobials in the burn units of the public hospitals of the Free State and North Cape before, during and after the COVID-19 pandemic (1 March 2018-31 July 2024,"*** has been reviewed the Faculty Research and Innovative Committee (FRIC) of the Faculty of Health and Environmental Sciences, Central University of Technology on the 23rd May 2024 and has been judged to be relevant, designed in accordance with accepted scientific practices and norms.

Student: CHENE' BEETGE
Student number: 220022618

Should you require additional information, please contact Prof TJ Makhafola at jmakhafola@cut.ac.za

Sincerely;

Tel: +27 51 507 3369
Prof TJ Makhafola
Assistant Dean; Research, Innovation and Engagement
Faculty if Health and Environmental Sciences

• Private Bag X20539 • Bloemfontein • SOUTH AFRICA • 9300 •
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APPENDIX D: FIRST ARTICLE PUBLISHED


International Journal of Environmental Sciences,
ISSN: 2229-7359

CERTIFICATE

OF PUBLICATION

This certificate is proudly presented to

Beetge C, Makhoahle P

In recognition of the publication of the paper entitled

The Prevalence Of Multi-Drug Resistant Pseudomonas
Aeruginosa In The Burn's Unit Of Pelonomi Hospital.

has been published in International Journal of Environmental Sciences
(2229-7359)

Volume 11 Issue 20s (2025) , Date of Publication : August 2025

Mihai V. Putz

MIHAI V. PUTZ

Editor in Cheif

ELSEVIER

APPENDIX E: SECOND ARTICLE PUBLISHED

International Journal of Environmental Sciences
ISSN: 2229-7359
Vol. 11 No. 15s, 2025
https://www.theaspd.com/ijes.php

Antimicrobial Resistance In The COVID-19 Era: A Global Literature Review Highlighting The Need For Focused Surveillance In South Africa's Free State And Northern Cape Provinces

Beetge C[1], Makhoahle PM[1*] and Malope-Kgokong B[2]

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Received- 23/06/2025 , Acceptance - 25/06/2025

Abstract
Antimicrobial resistance (AMR) represents a mounting global health crisis that transcends geographic, political, and economic boundaries, with its presence documented across every continent. Since 2017, the burden of AMR has surged sixfold globally, signalling an urgent call for coordinated intervention. This literature review synthesizes current findings from peer-reviewed academic databases to examine the role of the COVID-19 pandemic in accelerating the trajectory of AMR and highlights persistent gaps in the global research agenda. Notably, the United States reported a 20% increase in six major bacterial antimicrobial-resistant hospital-onset infections during the pandemic compared to pre-pandemic periods. This surge peaked in 2021 and, alarmingly, remained elevated throughout 2022, underscoring the pandemic's compounding effect on existing resistance trends and the fragility of global healthcare infrastructures.

Keywords: *COVID-19 pandemic, antimicrobial resistance, multi-drug resistance.*

INTRODUCTION

The impact of the coronavirus disease of 2019 (COVID-19) on South Africa was undeniably significant, affecting not only the country's financial landscape but also leaving its mark on the medical sector. Hospitals were drastically understaffed during this period and without the necessary equipment, drugs, and infrastructure to fight this pandemic lead to a poor health outcome which was unavoidable in certain situations. There was a wide range of adverse impacts on the health services and one of the impacts that were not noticed was the further development of antimicrobial resistance (AMR) (Shomuyiwa , et al., 2022). The antimicrobial stewardship programs were shifted aside during the pandemic and more focus was put on the lockdown, rapid testing and vaccinations for COVID-19. Virtual patient consultations and intensive care units with the focus on treating COVID-19 patients unintentionally contributed to the further development of multi-drug resistant bacteria, while the laboratories were flooded with COVID samples there was a decrease in the testing of bacteria (Khaznadar, et al., 2023).

Factors that contributed to the increased consumption of antibiotics included misconceptions, lack of awareness about the virus outbreak, cultural stigma, misinformation and the overall fear of the population to get infected with the virus lead to patients asking for antibiotics without a bacterial infection being present. This especially occurred in low-and-middle-income countries for example South Africa (Djuikoue, et al., 2023). In 2017 a list was released by the World Health Organization (WHO) to researchers and doctors that included 12 of the families of bacteria that was the most resistant to antimicrobials, to promote the development of new antimicrobials by researchers. The organisms that were on the list included multi-drug resistant (MDR) bacteria that can be found in hospitals or that poses a threat to patients with indwelling devices (Med, 2019). Bacterial infections could be fatal if left untreated due to the unavailability of antimicrobials to which it is sensitive to, these infections included pneumoniae and blood infections. This was done in an attempt to decrease the further development of AMR, but before any plans or strategies could be put in place the COVID-19 pandemic occurred and since then the attempts have been placed on hold (Djuikoue, et al., 2023).

APPENDIX F: PROOF OF LINGUISTIC EDITING

CORNELIA GELDENHUYS

083 2877088
corrieg@mweb.co.za

25 September 2025

TO WHOM IT MAY CONCERN

Herewith I, **Cornelia Geldenhuys (ID 521114 0083 088)** declare that I am a qualified, accredited language practitioner and that I have edited the following dissertation:

THE PREVALENCE OF MULTIDRUG-RESISTANT ANTIMICROBIALS IN THE BURN UNITS OF THREE HOSPITALS IN THE FREE STATE AND NORTHERN CAPE BEFORE, DURING AND AFTER THE COVID-19 PANDEMIC (1 MARCH 2018–31 JULY 2024)

by

Chene' Beetge

All changes were indicated by track changes and comments **for the author to verify, clarify aspects that are unclear, make the necessary adjustments, and finalise.** The editor takes no responsibility in the instance of this not being done. The document remains the final responsibility of the author.

………………………………………………………

C GELDENHUYS
MA (Lin) cum laude, MA (Mus), BA Hons (French), HED, HDL, UELM

Accredited member/Geakkrediteerde lid, SATI, Membership/Lidmaatskap: 1001474 (A/E-E/A)
Full member/Volle lid, Professional Editors Guild (PEG, Membership GEL001)
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European Association of Scientific Editors (EASE: Membership 5523)

Central University of Technology, Free State

APPENDIX G: TURNIT RECEIPT

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This receipt acknowledges that Turnitin received your paper. Below you will find the receipt information regarding your submission.

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The prevalence of multidrug-resistant antimicrobials in the burn units of three hospitals in the Free State and Northern Cape before, during and after the COVID-19 pandemic (1 March 2018–31 July 2024)

Chené Beetge

Student number – [illegible]

Central University of Technology Free State Campus

Faculty of Health and Environmental Sciences

Department of Health Sciences

Program: Masters in Biomedical Technology

Bloemfontein

South Africa

Supervisor: Prof Pierre Makhoahle (Biomedical Technology)

Co-supervisor: Dr Babatyi Malope-Kgokong

APPENDIX G: TURN IT IN SUMMARY

Beetge C Masters

ORIGINALITY REPORT

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SIMILARITY INDEX	INTERNET SOURCES	PUBLICATIONS	STUDENT PAPERS

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