A retrospective review of colistin utilization and patient outcomes across four private sector hospitals in South Africa to identify opportunities to optimise colistin stewardship in hospitalised patients with multi-drug resistant Gram-negative infections.

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A dissertation submitted to the Faculty of Health Sciences, University of the Witwatersrand, in fulfilment of the requirements for the degree of Master of Pharmacy

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DECLARATION

I, Angeliki Phroso Messina, declare that this dissertation is my own original work, with all other sources of information acknowledged by means of a complete reference list. This dissertation is being submitted in fulfillment for the degree of Master of Pharmacy at The University of the Witwatersrand, Johannesburg. This work has not been submitted before for any degree or examination at this or any other University.

18 May 2018

Angeliki Phroso Messina **Date**

ABSTRACT

The increased prevalence of multi-drug resistant (MDR) Gram-negative infections in critically ill patients has resulted in the re-introduction of colistin as rescue therapy. Various guidelines for colistin administration have led to confusion in establishing the appropriate dose which has potential for adverse consequences including treatment failure or toxicity. Colistin, also known as Polymixin E, is a concentration-dependent bactericidal antibiotic considered to be highly nephrotoxic and neurotoxic. Colistin is used either intravenously to treat life threatening systemic infections or by nebulisation for the treatment of respiratory tract infections. Although colistin resistance has been documented in South Africa, there is no local evidence as to why and how colistin is used in hospitals and similarly compliance with current dosing guidelines is unknown. This study aimed to evaluate the utilization of colistin in order to identify stewardship opportunities regarding its' appropriate use in the future.

A retrospective electronic record review of adult patients treated with intravenous (IV) and aerosolised colistin therapy in four Gauteng private hospitals was conducted between 1 September 2015 - 30 June 2016. The following data were collected on a standardized template; patient demographics including: age, gender, weight and hospital location; laboratory indicators including: renal function markers of creatinine and estimated Glomerular Filtration Rate (eGFR), as well as, culture specimens taken and their corresponding results. With regards to the colistin therapy: the indication for use, admitting diagnosis, the prescribed dose, frequency and route of administration, duration of treatment and if prescribed in combination with another Gram-negative antibiotic was considered. The following stewardship principles were monitored in addition to appropriate dose and duration; if a culture was taken prior to the initiation of treatment, if therapy was de-escalated and if a loading dose was prescribed. Outcome measures included overall in-hospital mortality, intensive care unit length of stay and overall hospital length of stay. Furthermore, compliance to two local colistin dosing guidelines was measured and a colistin stewardship bundle was developed, including nine process measures, to enhance the appropriate use of IV colistin.

A total of 237 patients were included in the study of which 212 received colistin IV and, 25 via nebulisation. The results of patients who received IV colistin therapy demonstrated an 81.2% overall compliance to the proposed colistin stewardship bundle developed from this study. Non-compliance was mainly due to incorrect maintenance doses prescribed (50%), 'hang time' (66%) and poor de-escalation practices (69%). Significantly shorter durations of treatment were found in patients who received higher loading doses (p=0.040) and in those that received maintenance doses of 4.5 Million Units (MU) twice daily vs 3 MU three times daily (p=0.0027). In addition, more of the patients that demised received the 3 MU three times daily maintenance doses, compared to those who survived (p=0.0037).

Aerosolised colistin was only prescribed in one of the four hospitals studied. Of those patients who received aerosolised colistin, 13 were for cystic fibrosis and 12 for other nosocomial lower respiratory tract infections (LRTI's). Compliance to appropriate dose for the cystic fibrosis patients was good at 92.3%, however, for other LRTI's was poor at only 41.7%.

This study demonstrated that there is noteworthy prevalence of MDR Gram-negative infections in South African hospitals which requires the use of colistin. In addition, the study identified many stewardship related opportunities to improve appropriate colistin utilization in particular relating to dose for both routes of administration. The implementation of a colistin stewardship bundle is necessary, as a matter of urgency, to preserve the efficacy of this last resort antibiotic.

DEDICATION

I dedicate this dissertation to my large Mediterranean family.

In particular my husband, Lorenzo Messina, mother, Emilia Stephanou, father, Stephanos Stephanou, brother, Andreas Stephanou, cousins Elena Philippou and Andri Apostolellis, grandmother, Phroso Stephanou, Godmother, Kitsa Phillippou and my mentor and friend Dena van den Bergh.

Thank you for making this possible, for your belief in me, and for all the support, encouragement, reassurance and unconditional love, without which this work would not have been imaginable.

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LIST OF PRESENTATIONS AND PUBLICATIONS ARISING FROM THIS STUDY

Presentations:

Refer to Appendix A

1. Messina AP., Brink AJ., van Vuuren SF. Evaluation of colistin utilization in patients with multidrug- and extensive drug-resistant Gram-negative infections in four private hospitals in South Africa. All Africa congress on pharmacology and pharmacy, Gauteng, South Africa. October 5-8 2016.

   Winner of the Academy of Pharmaceutical Sciences Young Scientist Award (Pharmacy Practice), 2016.

2. Messina AP., Brink AJ., van Vuuren SF. Evaluation of colistin utilization across multiple South African private hospitals: Indeed it's time for colistin stewardship. Federation of Infectious Disease Society of South Africa (FIDSSA) conference, Cape Town, South Africa. November 9-11 2017.

   Winner of the Institut Merieux and Federation of Infectious Diseases Societies of Southern Africa Young Investigator Award, 2017.

Publications:

1. Messina AP., van Vuuren SF., Brink AJ. Our collective contribution matters: Pharmacists unite in tackling antibiotic resistance for South Africa. South African Pharmacy Journal 2017; 84(2): 53-55. (Invited article)

2. Messina AP., Brink AJ., Richards GA., van Vuuren S. Opportunities to optimize colistin stewardship in hospitalized patients in South Africa: Results of a multi-site utilization audit. South African Medical Journal 2018; 108(1):28-32. DOI:10.7196/SAMJ.2018.v108i1.12561

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LIST OF ABBREVIATIONS

%	Percent
APACHEII	Acute Physiology and Chronic Health Evaluation Score
ASP	Antimicrobial Stewardship Program
AUIC	Area Under the Inhibitory Curve
BRICS	Brazil, Russia, India, China, South Africa
CBA	Colistin Base Activity
CDC	Centre for Disease Control
CF	Cystic Fibrosis
CMS	Colistimethate Sodium
CrCl	Creatinine Clearance
CPE	Carbapenemase Producing *Enterobacteriaceae*
DMSA	Data Management and Statistical Analysis
eGFR	Estimated Glomerular Filtration Rate
EMA	European Medicines Agency
ESBL	Extended Spectrum Beta Lactamases
ESCMID	European Society of Clinical Microbiology and Infectious Diseases
FDA	Food and Drug Administration
FEV	Forced Expiratory Volume
FIDSSA	Federation of Infectious Diseases Society of South Africa
GES	Guiana extended-spectrum β-lactamases
ICU	Intensive Care Unit
IQR	Interquartile Range
IU	International Units
IV	Intravenous
JHB	Johannesburg
LOS	Length of Stay
LRTI	Lower Respiratory Tract Infection

MCC	Medicines Control Council
MDR	Multi Drug Resistant
mg	Milligrams
MIC	Minimum Inhibitory Concentration
MRSCA	Medicines and Related Substances Control Act
MU	Million Units
NDM	New Delhi metallo-β -lactamase 1
NDoH	National Department of Health
OXA-48	Oxacillinase-type carbapenemases
PD	Pharmacodymics
PK	Pharmacokinetics
PTA	Pretoria
SAASP	South African Antibiotic Stewardship Program
SAPJ	South African Pharmacy Journal
SAMJ	South African Medical Journal
SASOCP	South African Society of Clinical Pharmacy
SCr	Serum Creatinine
SSI	Surgical Site Infection
STG	Standard Treatment Guidelines
TDM	Therapeutic Drug Monitoring
VAP	Ventilator Associated Pneumonia
VIM	Verona integron-encoded metallo β-lactamases
XDR	Extensive Drug Resistance
β	Beta
>	Greater than
≤	Less than or equal to

CHAPTER ONE

GENERAL INTRODUCTION AND LITERATURE REVIEW

1.1. A global view of antimicrobial resistance

The discovery of antibiotics has been a critical resource to the advancement of modern medicine. Since the initial unearthing of antibiotics in the 1940's, their essential role has contributed to the treatment of serious bacterial infections. Antibiotics have helped to prevent infections in surgeries and the immune compromised patient population; ultimately prolonging the life span of humans (CDDEP, 2015). Today, however, the efficacy of antibiotics is dwindling worldwide with the resultant emergence of life-threatening multidrug-resistant (MDR) bacteria. The persistent misuse (including under dosing, inappropriate duration and incorrect indications) of antibiotics globally has over time hastened the natural process of antibiotic resistance which Sir Alexander Fleming cautioned the public of in 1945. Antibiotic resistance is also known to be compounded as a consequence of antibiotic use which has been caused by two global factors: a) increased access to antibiotics due to increased earnings and availability and, b) a greater global demand for protein forcing the use of antibiotics as growth promoters in livestock (CDDEP, 2015). In line with natures mantra of 'survival of the fittest;' the larger the consumption of antibiotics and antibiotic pressure on a system, the higher the risk of MDR bacterial population selection (CDDEP, 2015).

The 'dawn of the post antibiotic era' has been widely documented as a global threat to society which was echoed by the World health Organization (WHO, 2014). It is estimated that by the year 2050, infections by antimicrobial resistant organisms will be the leading cause of death worldwide (one person dying every three seconds) (O'Neill, 2014). According to the Centre for disease Control and Prevention (2013) each year in the United States, two million patients are infected by MDR organisms, defined as resistance to more than three different classes of antimicrobials, of which 23000 patients succumb to these infections annually. The devastating consequences of this crisis includes increased hospital and antibiotic costs, prolonged hospital stays, poor patient outcomes and increased infection risks in hospitals and communities (CDDEP, 2015).

In addition, global health security has been compromised as a result of the advancements in aviation and population migration, which has provided an opportunistic platform for MDR organisms and other diseases to spread. A recent review of global antimicrobial resistance highlighted that antibiotic resistance is no longer a forecast for the future but rather a phenomena that is currently occurring extensively worldwide and which requires imperative collective attention and action (WHO, 2014). Furthermore, a report entitled: "The state of the world's antibiotics" exposed the alarming prevalence of MDR Gram-negative organisms especially amongst the *Enterobacteriaceae* (CDDEP, 2015). Low and middle income countries are particularly affected by antibiotic resistance due to the enhanced infection rates experienced, lack of infection surveillance programs in place, and the exorbitant costs associated with treating these in already weak and strained health systems (CDDEP, 2015).

A global review of antibiotic consumption over a ten year period showed that utilization of antibiotics increased overall by 36%. The most notable increase in consumption was reflected in the carbapenem and polymixin classes of antibiotics (van Boeckel et al., 2014). This implies a greater global need for the use of broad-spectrum Gram-negative antibiotics in healthcare settings. Because these so-called 'super-bugs' have gained resistance mechanisms to almost all antibiotics currently available, the use of older, more toxic drugs such as colistin is necessary, as a last resort therapy to help treat severe infections particularly by MDR Gram-negative pathogens (Goff et al., 2014).

Experts predict that in the foreseeable future there will be no new antibiotics with novel mechanisms of action available for the treatment of Gram-positive and Gram-negative bacterial infections (Nation and Li, 2009; Yamamoto and Pop-Vicas, 2014). Unfortunately, there is limited support available within the pharmaceutical industry to promote research and development for antimicrobials because discovery and development of these agents is difficult and return on investment is poor (Rex et al., 2014).

1.2. Gram-negative organisms and resistance

Gram-negative bacilli are found within the natural environment including soil and water and are often characterized by resistance to multiple antibiotics (Vincenti et al., 2014). These

organisms pose a particular threat to hospital environments as they are the most widespread which cause nosocomial infections (Vincenti et al., 2014). The rates of MDR Gram-negative organisms are escalating at an alarming pace. A recent report by the Centre for Disease Dynamics Economics and Policy (2015) showed that resistance to all first line and last resort antibiotics is on the rise universally. The ESKAPE organisms including: *Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumanni*, *Pseudomonas aeruginosa*, and *Enterobacter* species, have been recognized globally as the pathogens shown to be rapidly developing resistance for which therapeutic options are diminishing, especially in the critically ill hospital care setting (Rice, 2008; Boucher et al., 2009).

In non-fermenting Gram-negative bacilli such as *P. aeruginosa*, MDR and extensive-drug resistance (XDR) may emerge following sequential chromosomal mutations, which may lead to the overproduction of intrinsic β-lactamases, such as AmpC and hyper-expression of efflux pumps, target modifications and cell permeability alterations (Ruppe et al., 2015). *Pseudomonas aeruginos*a also has the ability to acquire mobile genetic elements encoding for resistance, including carbapenemases (Ruppe et al., 2015). The spontaneous mutation rate for expression of resistance may occur as frequently as 1 in 10^{6-7} wild type strains. This process may be accelerated by overuse of antibiotics with anti-pseudomonal activity particularly if therapy is prolonged (Ruppe et al., 2015).

Historically resistance amongst *Enterobacteriaceae* began with the emergence of the β-lactamase enzymes around thirty years ago which rendered certain β-lactam antibiotics including cephalosporins ineffective (Bradford et al., 2004). As a result of the global spread of extended spectrum β-lactamases (ESBL) amongst *Enterobacteriaceae,* carbapenems -which are a broader spectrum class of antibiotics that cover most resistant Gram-negative species, were increasingly used. In the simplistic explanation of this phenomenon, the selective pressure caused by antimicrobial overuse, in hospital and community settings, has driven these organisms to acquire such resistance mechanisms (Vincenti et al., 2014).

Evidence of the first carbapenem resistant *Enterobacteriaceae* species was documented in 1993 (Ah et al., 2014). Carbapenem resistance amongst the *Enterobacteriaceae* can occur through various mechanisms; however, the most common is through the production of

carbapenemases, a bacterial enzyme which hydrolyses carbapenems and all other β-lactam antibiotics including penicillins and cephalosporins. These carbapenemase-producing *Enterobacteriaceae* (CPE) often contain additional mechanisms of resistance to the aminoglycosides and the flouroquinolone class of antibiotics, rendering all standard antibiotic therapies ineffective (Brink et al., 2012). The CPE pathogens produce various epidemiological classes of carbapenemases including: Guiana extended-spectrum β-lactamases (GES), Verona integron-encoded metallo β-lactamases (VIM), oxacillinase-type carbapenemases (OXA-48 and its derivatives) and New Delhi metallo-β -lactamase 1 (NDM-1), all of whom are a major concern and risk to modern medicine and healthcare systems today (Brink et al., 2012).

The international spread of CPE is showcased through the discovery of the NDM-1 enzyme in particular. This enzyme was first revealed in 2008 from a Swedish patient following travel to New Delhi, India (Johnson and Woodford, 2013). To date *Enterobacteriaceae* containing NDM-1 genes have been reported in over 70 countries across the world which indicates how rapidly these organisms can spread globally (Johnson and Woodford, 2013). It has also been detected in environmental samples including water reservoirs in India and Vietnam, indicating emergence in both hospital and community locations (Johnson and Woodford, 2013). Risk factors for CPE organisms include: previous antibiotic exposure, prolonged hospitalization, severe illness and surgery to name a few (Brink et al., 2012). Often, patients infected with CPE's are treated with salvage agents such as tigecycline, colistin and or fosfomycin as last resort therapy for these life threatening infections (Brink et al., 2012).

MDR Gram-negative organisms such as *K. pneumoniae* and *P. aeruginosa* are recognised as particularly life-threatening pathogens. Although patterns of resistance vary worldwide there is a definite increase in these organisms including the CPE's which has resulted in the Centres for Disease Control naming this group of organisms one of the topmost critical antibiotic resistant challenges today (Karam et al., 2016).

Mechanisms of bacterial resistance are not a novel phenomenon but rather something that has always intrinsically existed even prior to the discovery of antibiotics (Karam et al., 2016). Therefore, it is thought that the emergence and spread of resistant bacteria following antibiotic therapy is a result of the selection of naturally occurring resistant bacteria. This is

due to the destruction of sensitive bacteria following antibiotic therapy and the environment created for those resistant kinds to proliferate (Karam et al., 2016). The risk of carbapenem resistance occurring in Gram-negative pathogens has been shown to be 5.9 times higher in patients exposed to only one to three days of a carbapenem and 7.8 times higher in patients exposed to longer therapy (Armand-Lefevre et al., 2013). The Gram-negative pathogens are most likely to display resistance to multiple classes of antibiotics and there are three mechanisms in which resistance can occur, particularly to β-lactam antibiotics (Poole, 2001; Karam et al., 2016):

a) The production of a β-lactamase enzyme which hydrolyses multiple antibiotics and can result from a single amino acid change (plasmid-mediated) of the conventionally produced enzyme. In addition, chromosomally-mediated enzymes known as AmpC β-lactamases can also develop and have been detected in *K. pneumoniae* and *P. aeruginosa*.
b) Closure of porin channels within the bacterial cell wall which results in the inability of antibiotics to penetrate the pathogen. This mechanism is most commonly seen in organisms resistant to the carbapenem class of antibiotics.
c) Development of efflux pumps that can be intrinsic or acquired by the pathogen and which expel antibiotics out of the bacteria cell walls.

The factors that result in the emergence of carbapenemase production have not been linked to the single exposure of a particular antibiotic but rather to the repetitive broad-spectrum exposure and prolonged duration of all antibiotic therapies (Karam et al., 2016).

The escalating global prevalence of antibiotic resistant organisms requires antibiotic therapy to be optimized in an attempt to effectively manage these infections (Cassir et al., 2014). Optimising antibiotic therapy in the critically ill patient, requires a deep understanding of the PK and PD parameters of the antibiotic as well as the MIC of the organism (Goff and Nicolau, 2013; Abdul-Aziz et al., 2015). When considering the PD of antibiotics, the antibiotic concentration achieved is directly related to the ability to exert bactericidal or bacteriostatic effects (Abdul-Aziz et al., 2015). The activity of an antibiotic is a result of the amount of free drug concentration available and this is influenced by drug, patient and/or severity of illness factors including: the hydrophilicity or lipophilicity of the drug affecting its' volume of

distribution; degree of protein binding of the drug and the serum albumin levels of the patients; augmented renal clearance and degree of capillary leakage; amongst other factors which could influence the drugs absorption, distribution, metabolism and elimination (Abdul-Aziz et al., 2015; Richards et al., 2015).

Multidrug resistant organisms often have much higher MIC's then their sensitive counterparts (Richards et al., 2015). Therefore, when attempting to treat MDR organisms, optimal drug concentrations required are considerably greater than those usually accepted to be sufficient (Richards et al., 2015). Recommendations of drug concentrations more than four times the MIC exist to ensure adequate therapeutic efficacy and prevent the selection of resistant pathogens in critically ill patients (Richards et al., 2015).

In January 2017, the first published case report of a carbapenem resistant *K. pneumoniae* wound isolate revealed plasmid-mediated resistance to 26 antibiotics - all antibiotics currently commercially available (Chen et al., 2017). This pathogen was identified from an elderly female patient with a history of multiple hospital admissions (in both India and United States of America) and frequent antibiotic exposure. The patient succumbed to the infection following bacteraemia and multiple organ failure due to the absence of antibiotic agents as effective therapy (Chen et al., 2017).

This case generated much concern from the healthcare fraternity and public alike as it highlighted the current reality of antibiotic resistance and the imminent inability to treat severe bacterial infections (Chen et al., 2017).

1.3. Background to colistin

Colistin, (trade name Colimycine® and also known as Polymixin E) is considered to be a highly nephrotoxic and neurotoxic concentration-dependent bactericidal antibiotic. It became accessible for use in the late 1950's, however, its' utilization diminished over time as newer 'less toxic' antibiotics with more favorable safety profiles, such as the aminoglycosides, became available (Biswas et al., 2012 ; Pike and Saltiel, 2014). Colistin can be used either intravenously (IV) to treat life threatening systemic infections or by nebulisation for the treatment of respiratory tract infections including ventilator associated pneumonia (Nation

et al., 2014; Gu et al., 2014). There are two kinds of colistin preparations available; the first is in the form of a sodium salt known as colistimethate sodium (CMS) which is an inactive pro-drug that requires *in vivo* conversion to its' active form, colistin; and the second is in the form of colistin Base Activity (CBA), based upon microbiological standardization (Nation et al., 2014).

Unfortunately, the current dosing guidelines for colistin administration are outdated and confusing as package insert information has not been revised with new information since its' initial launch, therefore, healthcare practitioners are using decades old information to make clinical decisions if and when referring to the package insert (Nation et al., 2014). This information is still based on pharmacokinetic (PK) properties concluded from improper microbiological assay studies (Nation et al., 2014). An incongruity arises as a result of this methodology as continuous conversion from the inactive prodrug, CMS, to its active form, colistin, occurs during the incubation period of these assays. Therefore, it is difficult to establish what the exact blood concentration of active colistin was at the time of blood sample collection and thus, product information is based on inflated colistin concentration values (Li and Nation, 2006; Nation et al., 2014). A study by Ortwine et al. (2014) concludes that because of these discrepancies the dosing guideline of colistin in the package insert is inaccurate and thus, the appropriate use of colistin cannot completely be determined due to the inadequate PK and pharmacodynamic (PD) data available.

Furthermore, colistin guidelines are complicated because the literature provides recommendations in international units (IU) and milligrams (mg) of CMS, as well as the mg of CBA. The recommendations also differ between European and American literature depending on which metric convention the country adopts (Biswas et al., 2012). As a result, there is a misunderstanding in establishing the appropriate therapeutic dose of colistin which often results in the incorrect dose being administered to the patient (Nation et al., 2014). These pharmacological concerns further compromise the management of patients with life-threatening Gram-negative infections who are on colistin treatment (Kassamali et al., 2013). If colistin is not dosed appropriately it can take 2-3 days to reach steady state concentration and thus the administration of a loading dose is essential to improved patient outcomes. Sub-

therapeutic doses of colistin prohibit the drug from achieving optimal tissue concentrations for bacterial killing and are therefore ineffective (Kassamali et al., 2015).

In South Africa, colistin is available in the IU of CMS as a Section 21 medicine since it is not registered for use in the country (Visser-Kift et al., 2014). It can be procured according to the Medicines and Related Substances Control Act (MRSCA) through special application and approval granted by the Medicines Control Council (MCC). Often, this approval process can lead to a delay in therapy which ultimately can have detrimental effects for the patient (Tigen et al., 2013; Wertheim et al., 2013). This onerous process makes colistin unique as it is the only antibiotic 'restricted' as a result of it not being registered for use in the country (Nation et al., 2014). Therefore, colistin in South Africa is only authorized in critical and crucial circumstances as stipulated by the MRSCA (Act 101 of 1965).

As a result of the mounting prevalence of MDR and XDR *Enterobacteriaceae* (such as *K.pneumoniae*) and non-fermentative Gram-negative pathogens particularly *P. aeruginosa,* and *A. baumanii*, globally and in South Africa, polymixins are often the only remaining class of antibiotic that can be used as a final treatment option in critically ill patients (Li et al., 2006). New data suggests that the adverse effects of nephrotoxicity previously reported with the initial use of colistin was indeed a consequence of a misunderstanding of the antibiotics' PK and PD properties and inappropriate dosing due to confusion and variations in the dosing metrics adopted by different countries (Dalfino et al., 2012; Nation et al., 2014; Nation et al., 2017). As such, limited recent data supports the neurotoxic nature of colistin and some studies have suggested similar or safer toxicity profiles of colistin compared to the aminoglycosides when considering nephrotoxicity (Li et al., 2006). In most instances, the reported side effects of colistin are not long lasting and have been described to dissipate following discontinuation of therapy (Li et al., 2006; Dalfino et al., 2012). The risk-benefit ratio of prescribing colistin requires consideration in the fight against severe MDR and XDR infections.

In early 2017, Nation and colleagues described a novel algorithmic approach for IV colistin dosing following interpretation of results from a large multi-centre PK study including 215

patients. This study established for the first time colistin dosing recommendations which were based on accurate PK data.

Utilization reviews on colistin are scarce since the antibiotics' use diminished from the market following the introduction of the aminoglycoside antibiotics in the early 1960's (Biswas et al., 2012 ; Pike and Saltiel, 2014). A search conducted on global databases including Pubmed and Science-Direct resulted in five studies relating to colistin utilization reviews (Table 1.1). The subsequent gap in the availability of literature over numerous years has left many questions regarding the appropriate use of colistin unanswered. In recent years, literature on colistin has become more abundant as a direct consequence of the worldwide emergence of MDR Gram-negative bacteria, ultimately resurrecting the use of colistin (Landersdorfer and Nation, 2015).

Table 1.1. A summary of publications pertaining to previously conducted colistin utilization reviews worldwide

Country	Number of hospitals included in the study	Number of patients meeting the study inclusion criteria	Reference
Greece	One	24	(Markou et al., 2003)
Greece	One	43	(Michalopoulos et al., 2005)
Canada	Twelve	22	(Sabuda et al., 2008)
Greece	One	258	(Falagas et al., 2009)
Brazil	One	109	(Tanita et al., 2013)

It is not surprising that the early utilization studies on colistin were derived mostly from healthcare settings in Greece, as Miyakis et al. (2011) demonstrated, the country had one of the highest rates of MDR organisms in Europe according to European surveillance data reports from 2009, thus necessitating the need for colistin as salvage therapy as a direct consequence of the increased antibiotic resistance rates experienced.

The studies (Table 1.1) collected data similar to the scope of this study including; patient demographics, indication for use, organism resistance patterns, colistin dose, duration of therapy, effects on renal function, and overall outcome. The aims of these studies varied slightly but each contributed to the body of knowledge as they described their experiences with colistin in hospitals following the global rise of antibiotic resistance.

1.4. Intravenous colistin dosing in the critically ill patient population

It has been shown that patient outcomes improve and mortality is reduced with the timeous and appropriate administration of antibiotic therapy (Kumar et al., 2006). The benefits of administering the correct drug, however, are often reversed by suboptimal drug concentrations *in vivo* as a direct consequence of inappropriate dosing (Dalfino et al., 2012). Therefore, the prescription of the right drug at the right dose (cornerstones of antibiotic stewardship) is imperative to safeguard favourable patient outcomes and aid the resolution of infectious diseases. Dosing strategies of antibiotics are often devised from studies undertaken in patients who are not critically ill and therefore achieving the appropriate antibiotic dose in these at risk patients is a challenge (Roberts et al., 2014). As a result, the mortality rates are high and outcomes often poor for the critically ill (Roberts et al., 2014) and thus, it is imperative that dosing is accurate for these patients in order for them to have the best possible chance of survival.

Optimising antibiotic dosing in the critically ill is complex due to the altered physiological state of the host such as: increased volumes of distribution and prevalence of organ dysfunction both affecting the PD of the drug (Roberts et al., 2014). This is further compounded by higher organism resistance rates (Richards et al., 2015). These factors make the attainment of the Minimum Inhibitory Concentration (MIC) required for the appropriate bactericidal or bacteriostatic antibiotic effect extremely problematic in these patients as often MIC's can be doubled or tripled as a result of MDR.

The optimization of antibiotic dosing is based on the PK and PD properties of the drug. The aim of these principles is to ensure that the suitable concentration of antibiotic is at the tissue site target in order to destroy or inhibit the growth of the infecting bacteria. The PK of antibiotics can be classified as either: a) time dependant agents, where a specific time is

required above the MIC (Time > MIC) to execute efficacy or, b) concentration dependant agents, where the area under the inhibitory curve (AUIC) to the MIC or concentration peak > MIC is important for effect (Figure 1.1) (Richards et al., 2015). The significance of optimal dosing cannot be stressed enough, as sub-therapeutic doses can lead to treatment failure and antibiotic resistance, whilst over dosing can lead to toxicities and side effects (Landersdorfer and Nation, 2015). The range of dosing of a drug which allows the exertion of its therapeutic effect safely is known as the 'drug therapeutic window' and drug prescribing, dosing and frequency of administration should aim to fall within this window (Landersdorfer and Nation, 2015). This is difficult for drugs that have narrow therapeutic windows, such as colistin, as the difference in serum concentrations between efficacies (antibiotic effect), safety and toxicity (nephrotoxicity) is slight (Landersdorfer and Nation, 2015).

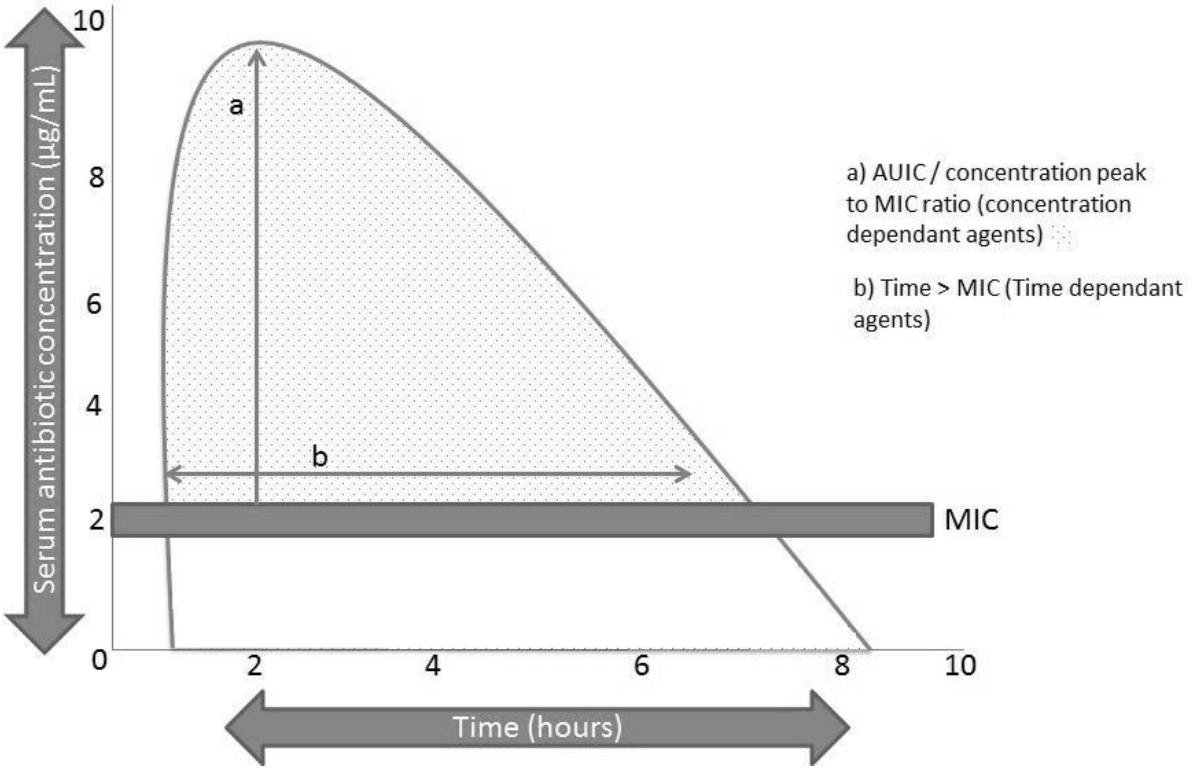

Figure 1.1. The pharmacokinetics of antibiotics adapted from Richards et al., 2015. (MIC = Minimum inhibitory concentration; Time > MIC = time above the MIC; AUIC = area under the inhibitory curve)

As mentioned, colistin was brought to market in the early 1960's and was not subjected to the stringent drug development safety and efficacy studies as would be required today by

organisations such as the Food and Drug Administration (FDA) and locally the MCC (Nation and Li, 2009). Furthermore, many dosing guidelines of colistin, including those of the package insert, have been based on decades old information from inaccurate kinetic studies and therefore much of the information to date required to understand the appropriate use of colistin has been invalid or unknown (Garonzik et al., 2011; Nation et al., 2014; Landersdorfer and Nation, 2015).

Since colistin is administered as a prodrug (CMS), it was difficult to establish the exact concentration of colistin in blood samples drawn during PK studies (Nation et al., 2014). To overcome this, in recent years, researchers placed extracted samples on ice and immediately centrifuged these at low temperatures. The resultant plasma was then stored at between -70°C and -80°C to prevent *in vitro* conversion of CMS to colistin (Plachouras et al., 2009; Garonzik et al., 2011). Additional studies also showed that an estimated 60% - 80% of inactive CMS is excreted unchanged in the urine suggesting that the conversion rate of CMS to active colistin *in vivo* is very slow, with maximum concentrations achieved approximately seven hours after each dose. This highlights the inefficiencies of CMS as a prodrug (Michalopoulos and Falagas, 2011; Ortwine et al., 2014; Landersdorfer and Nation, 2015).

Plachouras and colleagues (2009), published the first report of population pharmacokinetics following IV colistin administration in critically ill patients. Although the study only included a sample of 18 patients, a better understanding of colistin dosing was brought to light. Many important findings were revealed including;

- The half-life of the inactive prodrug, CMS, was 2.3 hours.
- The half-life of the active drug colistin was 14.4 hours. This was later found to vary between patients (Nation et al., 2017).
- Based on previously recommended dosing guidance of 3 MU eight hourly, it would take three days for colistin to reach a plasma concentration of 2 mg/L (the recommended MIC break point).
- The administration of a colistin loading dose (9 MU or 12 MU) allowed for the attainment of the recommended MIC breakpoint much faster (Figure 1.2).

The implication of these findings were ground-breaking in obtaining a better understanding of how colistin should be dosed and provided the necessary evidence for a colistin loading dose, as it was demonstrated that the administration of a colistin loading dose ensures the quickest attainment of the MIC breakpoint (therapeutic colistin concentrations) resulting in optimised patient care (Plachouras et al., 2009; Karaiskos et al., 2015). The impact of these findings was verified by Mohamed et al. (2012), who showed that the administration of a 9 MU colistin loading dose was associated with a rapid increase in the bactericidal effect of the drug within the first six and a half hours of treatment.

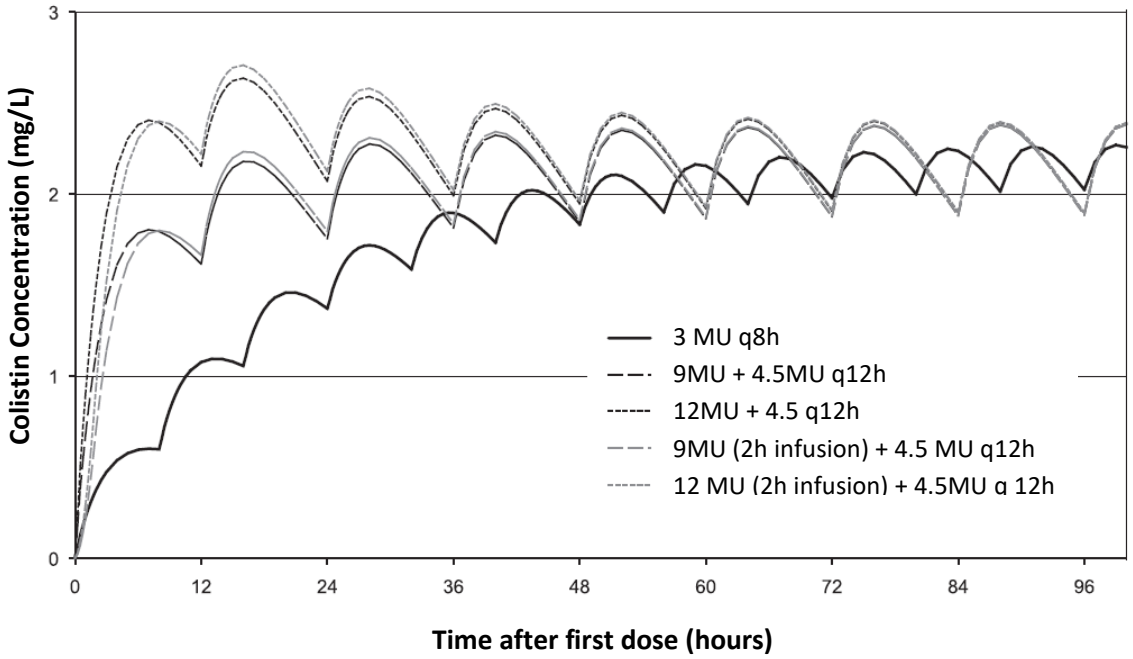

Figure 1.2. The Pharmacokinetic predicted model of colistin concentrations in a typical patient following the administration of various colistin dosing strategies as described by Plachouras et al., 2009.

Garonzik and colleagues (2011), expanded on the study by Plachouras et al. (2009) and published PK data following colistin IV administration in 105 patients including 12 patients on haemodialysis and four on continuous renal replacement therapy. The study made detailed colistin loading and maintenance dose suggestions for various categories of renal function and also based on body weight (Garonzik et al., 2011; Roberts and Lipman, 2012). Both studies

by Plachouras et al. (2009) and Garonzik et al. (2011), recommended higher daily doses of colistin that had previously ever been recorded or used.

The outcomes of these studies were confirmed by Dalfino et al. (2012), who showed that loading patients with 9 MU of colistin and administering a 4.5 MU dose every 12 hours totalling an average daily dose of 9 MU (rather than 3 MU every 8 hours) resulted in a clinical cure rate of 82% in the 25 study patients. The study thus advocated for the administration of a colistin loading dose followed by high individual doses administered at longer intervals. Furthermore, acute kidney injury was observed in 18% of colistin courses administered which was found not to be severe and the effects of which were reversed following course completion (Dalfino et al., 2012). Another study also found that patients who survived received higher colistin doses (9 MU per day in divided doses) with limited toxic side effects and concluded that the average daily dose of colistin was an independent factor for mortality (Falagas et al., 2009). In addition, studies by both Vicari et al. (2013) and Gibson et al. (2016) demonstrated better seven day clinical cure rates for patients who received high dose colistin. The aforementioned evidence reiterates the need to optimise colistin dosing to improve patient outcomes.

In 2015, the European Medicines Agency (EMA) and the United States FDA also updated their colistin dosing recommendations, however; the recommendations made between the two agencies were conflicting (Nation et al., 2016). The FDA recommendations did not include that of a loading dose for colistin and often, in a large proportion of patients with varying categories of renal dysfunction, the necessary plasma concentrations were unattainable based on the dosing suggestions made by either organisation (Nation et al., 2016). Thus, global consensus on the appropriate dosing of colistin following the release of these updated recommendations could still not be reached and further added to the confusion in optimal colistin dosing; since all the evidence published until then were based on relatively small patient samples.

The largest population IV colistin PK study conducted including 215 critically ill patients from three different countries with varying degrees of renal function was published in March 2017 by Nation and colleagues. This study clarified the appropriate recommended daily dosing of colistin according to various categories of renal function for the first time (Nation et al., 2017).

The findings from this study demonstrated the influence of renal function on the elimination of colistin and the drastic inter patient variability observed even at similar renal function categories. This effect is most likely due to the patient dependant conversion of the inactive pro drug to its active form, colistin, *in vivo* (Nation et al., 2017). The inter patient variability of *in vivo* colistin conversion further contributes to the complexity of the appropriate dosing of colistin, however, the recommendations from this study provide a verified dosing guideline and best available data to date. In addition, the findings push the colistin dosing boundaries and indicate that patients should possibly receive higher doses than what is currently being utilised.

1.5. Colistin dosing guidelines

At the time this study was conducted, the international dosing guidelines from Nation and colleagues (2017) were not yet published. Therefore, the best available colistin dosing guidelines available for South Africa were used to evaluate and determine the accuracy and appropriateness of colistin doses prescribed for the study patients. These guidelines were based on the best available international and national evidence at the time of their compilation. As discussed previously, consensus on appropriate colistin dosing globally is lacking. The advantages and disadvantages of these guidelines are summarised in Table 1.2.

Table 1.2: Advantages and disadvantages of available South African colistin dosing guidelines.

South African Society of Clinical Pharmacy Colistin dosing guideline (Labuschagne et al., 2016)	
Advantages	**Disadvantages**
• Most recent South African colistin dosing guideline. • Comprehensive and specific dosing guideline for intravenous and inhaled colistin therapy in adults and peadiatrics. • Authored by a large panel of experts in pharmacokinetics and clinical pharmacy.	• Dosing recommendations are made according to creatinine clearance which requires additional metrics to calculate including patient weight. • Recommendations not based according to randomised controlled trials.

South African Society of Clinical Pharmacy Colistin dosing guideline (Labuschagne et al., 2016)	
Advantages	**Disadvantages**
• Dosing recommendations are provided for various categories of renal function including renal replacement therapy. • Additional information is provided including reconstitution of colistin and administration guidance. • Definitive loading dose recommendations are made.	• Dosing recommendations not tiered according to strength of available evidence. • Different frequencies of administration are provided for various categories of renal function which may cause confusion in clinical practice.

Colistin Dosing Guideline (Visser-Kift et al., 2015)	
Advantages	**Disadvantages**
• Dosing recommendations are made according to GFR categories which is easier to retrieve from laboratory reports. • Dosing recommedations are provided as part of a systematic review of available literature at the time. • Dosing recommendations are provided for various categories of renal function including renal replacement therapy. • Additional information including: dose adjustment in renal failure, combination therapy, loading doses and a comprehensive discussion is provided. • Only 12 hourly frequency of dose administration recommendations are made in an attempt to standardise administration of colistin.	• Recommendations are not based according to randomised controlled trials. • Dosing recommendations are not tiered according to strength of available evidence. • Loading dose recommedations are somewhat vague recommending 9-12 MU. • Does not contain dosing recommendations for inhaled colistin therapy.

1.6. Colistin effects on renal function

Colistimethate Sodium (prodrug) is excreted primarily through the kidneys via glomerular filtration and a small portion is thought to be removed via tubular secretion. In contrast, the bulk of colistin following glomerular filtration is absorbed via tubular reabsorption and therefore the exact mechanisms of colistin clearance remains ambiguous (Landersdorfer and

Nation, 2015). Figure 1.3 depicts the pharmacokinetics of the renal excretion mechanisms of CMS and colistin. The process of reabsorption of colistin through renal tubular cells is considered to be the most likely cause of resultant nephrotoxicity (Roberts and Lipman, 2012; Landersdorfer and Nation, 2015).

As a consequence of this mechanism of clearance, those patients with decreased renal activity have a larger proportion of CMS converted to Colistin; as the elimination of CMS would be delayed as a result of the decrease in renal function, allowing more time and opportunity for CMS to be hydrolysed. For this reason, dose adjustments in patients with renal insufficiencies are necessary in order to avoid toxic plasma concentrations of colistin *in vivo* (Ortwine et al., 2014).

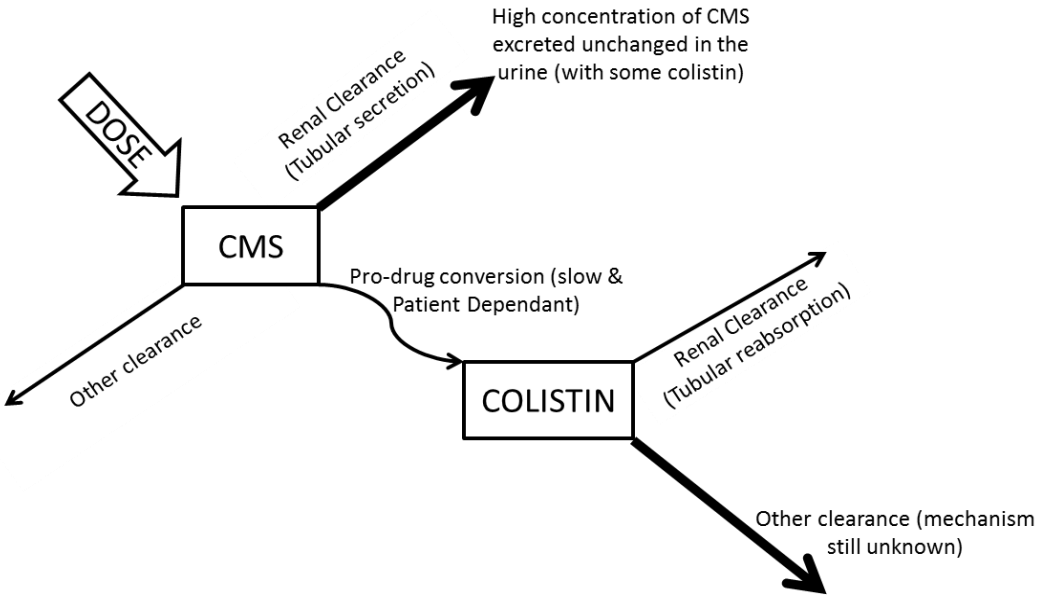

Figure 1.3. A schematic diagram of the mechanism of renal excretion of Colistimethate Sodium (CMS) and colistin. During normal kidney function, the thickness of the arrows represent the degree of clearance for each component (Adapated from Landersdorfer and Nation, 2015).

Colistin related nephrotoxicity has been linked to drug concentrations (dose-dependent phenomenon) and duration of therapy (longer treatment periods) (Nation and Li, 2009; Pogue et al., 2011; Dalfino et al., 2012). Toxicity rates between 30-45% have been reported recently (Akajagbor et al., 2013). A study conducted in a young cohort of 66 patients with few

comorbidity covariates that could attribute to renal dysfunction found that 45% obtained a degree of renal dysfunction following exposure to colistin (Hartzell et al., 2009). The study also found that the likelihood of toxicity occurring was 3.7 times greater if therapy continued for more than two weeks (Hartzell et al., 2009). DeRyke and colleagues (2010) demonstrated that 33% of patients studied developed nephrotoxicity during the first five days of colistin therapy. In this study, risk factors for the development of toxicity included advanced age, elevated illness severity scores, previous hospital admission, treatment in an intensive care environment and, renal dysfunction prior to the commencement of colistin therapy. Results from a study conducted by Pogue et al. (2011) revealed that nephrotoxicity occurred in 43% of patients following treatment with a higher dose colistin, however, kidney injury was short lived and mortality rates were not different when comparing those patients who developed nephrotoxicity and those who did not.

A systematic review of the literature relating to colistin induced nephrotoxicity conducted by Pike and Saltiel (2014) concluded that although the administration of colistin is associated with exceptionally high rates of nephrotoxicity, the toxicity is often reversible and does not warrant the discontinuation of therapy. This suggests that in patients where colistin may be the only viable treatment option, the risk benefit ratio should be considered and perhaps the associated toxicity can be managed once the infection has been resolved.

1.7. Prescription of colistin in combination with other Gram-negative spectrum antibiotics

When attempting to treat MDR organisms, prescribers are often faced with the following options:

a) Increase the doses of otherwise standard antibiotic regimens or,
b) Use a combination of antibiotic therapies to exert a potentially novel effect on the bacteria (Roberts and Lipman, 2012).

The rationale for the prescription of colistin in combination with other Gram-negative antibiotic agents is to create synergy; a phenomenon whereby the combination of the two antibiotics creates an effect or exerts an efficacy that is greater than their individual contributions.

Patients with adequate renal function clear CMS, faster and therefore lower concentrations of the prodrug is available in the plasma for conversion to active colistin *in vivo* – a process further compounded by the slow conversion rate previously described. As such combination therapy is strongly advocated in patients with good renal function (CrCl >80 mL/min) for a synergistic antibiotic effect and to attain the required plasma MIC for bacterial killing (Landersdorfer and Nation, 2015; Nation et al., 2017). Furthermore, the recommended daily dose of patients with adequate renal function according to Nation et al. (2017) is 10 MU. Since the risk of colistin-associated nephrotoxicity is increased at this dose, it is recommended that colistin is administered at a maximum daily dose of 9 MU and combined with an additional Gram-negative spectrum antibiotic for synergy to achieve therapeutic efficacy (Nation et al., 2017). In addition, since reports of colistin resistance have surfaced globally, administering colistin in combination is a method that could curtail the development of resistance to colistin, compared to the continuous use of colistin in monotherapy (Nation and Li, 2009; Visser-Kift et al., 2014; Richards et al., 2015; Coetzee et al., 2016).

Colistin combination therapy has also been studied for the treatment of CPE infections in the critically ill since routine antibiotic regimens were rendered ineffective as a result of the emergence of these pathogens. Numerous observational studies have shown some success in using this strategy for the treatment of carbapenem resistant Gram-negative infections and as such, this has become the standard of care (Paul et al., 2014). However, there is very limited evidence to show that combination therapy improves clinical outcomes (Paul et al., 2014). Dalfino and colleagues (2012) initiated colistin monotherapy in 50% of study patients and found no difference in clinical cure compared to those patients who received combination therapy. Durante-Mangoni et al. (2013) studied the effects of colistin-rifampicin combination compared to colistin alone for XDR *A. baumannii* infections and found no change in 30-day mortality, infection related death or length of hospital stay in the two patient groups. Parchem et al. (2016) showed significant results with the administration of colistin combination therapy for microbiological cure but this was not found for clinical cure when compared to patients on monotherapy. In contrast, however, Daikos and colleagues (2014) demonstrated that significantly higher mortality rates were found in patients with carbapenemase producing blood stream infections treated with monotherapy compared to those treated with combination therapy.

An *in vitro* analysis of the activity of colistin alone versus in combination with carbapenems indicated that doripenem most consistently achieved synergistic effects and in general the co-administration of colistin and a carbapenem showed enhanced bactericidal efficacy and no evidence of emergence of resistance (Zusman et al., 2013). A systematic review of the available evidence including 20 articles for the treatment of CPE infections revealed that numerous combinations of adjunctive therapy to colistin have been tested with varying degrees of success, albeit in small patient populations and, significant variations regarding site and severity of infection. The review concluded that combinations of tigecycline-colistin and colistin-carbapenem may result in lower mortalities for infections caused by *Klebsiella* spp. (Falagas et al., 2014). One study also suggested triple therapy of colistin-tigecycline-carbapenem as a strategy to combat these infections (Falagas et al., 2014).

The disadvantages of combination therapy includes increased treatment costs, and risks of drug related toxicity with the patients' broad exposure to antibiotics (Bergen et al., 2015). In addition, it is unclear if higher dosing of colistin has better outcomes than the administration of combination therapy (Delfino et al., 2012). A study including 12 countries sought to establish the effect of combination therapy versus monotherapy and mortality in patients with MDR blood stream infections for the first time. The most common combinations described in the study included: colistin-tigecycline (31%), aminoglycoside-tigecycline (35%) and colistin-carbapenem (44%) (Gutierrez-Gutierrez et al., 2017). Combinations including tigecycline, the aminoglycosides and colistin were associated with better outcomes compared to colistin monotherapy. However, due to the small sample of patients, the advantageous effects of the addition of a carbapenem to colistin could not be established. The study concluded that combination therapy was associated with improved survival rates only in patients that had high illness severity scores and that monotherapy should be used in patients with low scores (Gutierrez-Gutierrez et al., 2017).

There are many unanswered questions regarding the outcome of combination antibiotic therapy and verification of the effectiveness of this strategy versus monotherapy is currently underway internationally by other study groups in a randomised control trial (Gutierrez-Gutierrez et al., 2017). To date, the evidence is based on small, retrospective cohort type analysis (Falagas et al., 2014). Until additional evidence is available, the prescription of colistin

combination therapy is still recommended as best practice (Richards et al., 2015; Coetzee et al., 2016).

1.8. Inhaled colistin therapy

The delivery of active therapeutic compounds directly to the respiratory tract dates back to the times of ancient Greek mythology where tales are told of the oracle of Delphi inhaling fumes from the temple of Apollo (Wenzler et al., 2016). In 1932, Whitlaw and Patterson termed the word 'aerosol' directly meaning air (aer) solution (sol) (Wenzler et al., 2016). Over the years many drug delivery advancements to the respiratory tract have been made culminating in the use of aerosolised antibiotics for the treatment of bacterial infections of the airways; since these infections are thought to be the most common cause of human illness in both in- and out- patient settings (Wenzler et al., 2016).

It is shared knowledge that the effectiveness of antibiotic therapy relies on the achievement of sufficient concentrations of drug at the infection target site. Unfortunately, infections of the lower respiratory tract, such as pneumonia, are often difficult to treat as suboptimal concentrations of the drug penetrate the lung parenchyma to reach the deep alveolar level of the airways following systemic administration (Wenzler et al., 2016). To circumvent this, the delivery of the drug directly into the lungs, via inhalation, ensures optimal drug concentrations are achieved for microbial killing at the infection site whilst limiting the unintended consequences of commonly accompanying adverse effects, toxicities and the possible development of MDR intestinal flora that is associated with systemic delivery and exposure (Kofteridis et al., 2010; Tumbarello et al., 2013; Wenzler et al., 2016; Wunderink, 2016).

It is important to consider the practicality of inhaled drug delivery. Some of this includes consideration of the actual device used for aerosolisation (nebulizers versus dry powder inhalers), the size of drug droplets formed and the distribution of these in the lungs - small particles gravitate towards the lower airways including the bronchioles and alveoli compared to large particles which stay in the upper airways (Michalopoulos and Papadakis, 2010). The PK and PD of inhaled antibiotics are challenging, thus determining the link of drug delivery and clinical outcome is complex (Wenzler et al., 2016). Due to the lack of robust clinical data

and low numbers of large randomised controlled trials to establish accurate efficacy, the widespread use of aerosolised antibiotic administration is limited.

Colistimethate Sodium can be administered through nebulisation, the effects of which are complicated by it requiring conversion to its active form. Frequently occurring reported side effects, albeit reversible, of inhaled colistin include: cough, tightness of the chest, bronchoconstriction as a result of histamine release and, apnea due to neuromuscular blockade (Cunningham et al., 2001; Westerman et al., 2004; Michalopoulos and Papadakis, 2010;). As such, the concomitant prescription of a β-2 agonist (bronchodilator) is recommended (Beringer, 2001).

In South Africa, the IV colistin formulation is reconstituted and nebulised. However, this IV formulation foams profusely when used for nebulisation which enhances the complexity of optimising drug delivery (Beringer, 2001). This is in contrast to the aerosolised colistin preparations available in Europe, including dry powder inhalers and specially prepared colistin solutions for nebulisation.

Of concern, is the poor patient outcomes related to nosocomial pneumonia, including ventilator-associated pneumonia (VAP), affecting 10-20% of critically ill patients on mechanical ventilators in hospitals (Tumbarello et al., 2013). In these circumstances, mortality is affected by co-morbid diseases and the virulence of the infecting organism which is further compounded by MDR. Recently it has been recommended that aerosolised antibiotics should be used routinely, given the high failure rates of IV therapy and the current context of MDR infections for patients with VAP (Wunderink, 2016). Although, experience is limited with the use of aerosolised colistin for the treatment of critically ill patients with MDR Gram-negative lower respiratory tract infections (LRTI's), a plethora of evidence exists for its use in patients with cystic fibrosis (CF) and the prevention and treatment of *P. aeruginosa* (Falagas et al., 2006; Wenzler et al., 2016).

1.9. Review of cystic fibrosis

In the western world CF is one of the most common congenital hereditary diseases characterised by recurrent LRTI's (Li et al., 2001). It affects 1 in 2500 births per year and over

70 000 people in the world today are currently living with the condition (Doring et al., 2000; Ciofu et al., 2015). Infection of the airways in CF is recognized as the biggest contributor to morbidity and mortality causing over 90% of patients to succumb to the condition (Doring et al., 2000; Langan et al., 2015). The disease is a ramification of a single genetic mutation of chromosome seven which causes reduced production of chloride and water secretions in the airways and results in the development of thick, viscous secretions and diminished mucociliary clearance (Doring et al., 2000; Hoiby, 2011). Therefore, elimination of inhaled bacteria from the lungs is inhibited allowing pathogenic organisms to harbour and cause colonisation and infection within the respiratory tract. Consequently, in the fight against infection, the patient's non-inflammatory defence mechanisms breakdown, triggering a premature response of the inflammatory defence mechanisms including: polymorphonuclear leukocytes, cytokines and antibodies in the airways. The intense inflammatory response ultimately results in severe lung tissue damage (Hoiby, 2011).

Patients with CF are prone to repeated and persistent respiratory tract infections from early childhood which can lead to respiratory failure, lung transplantation or death (Hoiby, 2011; Koerner-Rettberg and Ballmann, 2014). The timeous and aggressive treatment with antibiotic therapy can extend the CF patient's life expectancy to 35-50 years, however, if left unattended to this can be drastically reduced (Hoiby, 2011). The primary aim of antibiotic therapy in CF is to steady lung function, prevent further lung tissue damage and if possible, reinstate previously diminished lung function (Hodson et al., 2002; Dalhoff, 2014). Although it is currently unclear if CF patients benefit more from aerosolised therapy compared to IV or oral, this method of drug delivery has certainly minimized the treatment burden and increased treatment compliance for these patients - as medication can be administered at home – and, it ensures sufficient therapeutic drug concentrations within the lungs whilst limiting associated systemic adverse effects (Dalhoff, 2014; Koerner-Rettberg and Ballmann, 2014).

In CF patients of all ages, *P. aeruginosa* is the most commonly identified opportunistic bacteria in sputum and bronchial washing samples (Doring et al., 2000; Li et al., 2001; Hodson et al., 2002; Hoiby, 2011). This organism is known to be responsible for the on-going lung damage and the consequent respiratory failure that transpires from the disease as an approximate 2% of lung function is thought to diminish each year once chronic infection has been established

(Li et al., 2001; Hodson et al., 2002). It is estimated that 81.3% of CF patients between the ages of 26-30 are infected with *P. aeruginosa* and spread is thought to occur via direct transmission from one patient to another or via contaminants in the environment (Doring et al., 2000; Ciofu et al., 2015).

There are two types of *P. aeruginosa* identified through the stages of CF infection illustrating the adaptive mechanism of the organism (Hoiby, 2011; Ciofu et al., 2015). Non-mucoid *P. aeruginosa* is typically recognized in initial infection episodes and is more responsive to antibiotic therapy compared to mucoid *P. aeruginosa* (Langan et al., 2015). The mucoid type often indicates chronic infection and contains a biofilm layer making the infiltration of antibiotics extremely difficult (Doring et al., 2000; Ciofu et al., 2015; Stefani et al., 2017). Biofilms are ever-present in nature, the components of which are often produced by bacteria themselves, and are formed through interconnecting extracellular substances that create a defence like matrix shell (Ciofu et al., 2015). The biofilm forms as a mechanism to protect the pathogen and this type of infection often follows repeated and on-going antibiotic exposure (Hoiby, 2011). Antibiotic concentrations of 100-1000 times more than that required for efficacy against the non-mucoid type are necessary to penetrate the biofilm during treatment (Doring et al., 2000; Ciofu et al., 2015). However, evidence has shown that the local concentrations of colistin achieved in the airways when nebulised are often optimal for efficacy against biofilm infections (Ciofu et al., 2015).

The mucoid category of infection provides an explanation as to how the organism is able to outlast and endure in the airways of CF patients for numerous years despite the patient's immune response and exposure to antibiotic therapy (Hoiby, 2011; Dalhoff, 2014). Chronic mucoid *P. aeruginosa* infection of the lungs in CF is similarly categorised to that of a type III hypersensitivity reaction whereby the large inflammatory response generated leads to profuse amounts of neutrophil production and the resultant decay causes the formation of large pus zones around the incessant bacteria which can end in total obstruction of the bronchi, bronchioles and alveoli ensuing irreversible lung tissue damage (Doring et al., 2000; Ciofu et al., 2015).

A study including 146 CF patients demonstrated a 40- month mean duration from diagnosis of CF to first *P. aeruginosa* isolation in the lungs with initial onset at increased age (> 2 years)

recognised as an independent factor for risk of development of chronic colonization of *P. aeruginosa* (Emiralioglu et al., 2016). Various antibiotic regimens can be used to eliminate the initial *P. aeruginosa* infection with median relapse periods ranging from 8-18 months (Emiralioglu et al., 2016). Although the ideal therapeutic options and duration of treatment have not yet been concluded (Emiralioglu et al., 2016), nebulised colistin is a well-established therapeutic agent for the early eradication of initial *P. aeruginosa* colonization, treatment of acute *P. aeruginosa* exacerbations and maintenance therapy for chronic infections in CF (Doring et al., 2000; Beringer, 2001; Michalopoulos et al., 2005). Early therapy has also been shown to delay the onset of chronic infection and generally improve the patient's health status (Beringer, 2001; Emiralioglu et al., 2016). Mayer-Hamblett et al. (2015) showed that in those patients in which early eradication of *P. aeruginosa* is sustained, time to infection relapse and chronic infection is prolonged compared to those patients who are unable to achieve early eradication of the organism; although the study could not establish a difference in the improvement of lung function between both patient sets.

1.9.1. Aerosolised colistin dosing and duration of therapy for cystic fibrosis patients

The PK of aerosolised colistin has not been vigorously established since the large drug particles get trapped within respiratory secretions. Absorption rates are reliant on numerous factors such as respiratory secretion volumes and mechanical factors which also postpone drug elimination (Ratjen et al., 2006; Bos et al., 2017). Delayed drug elimination may be advantageous since the antibiotic exposure time at the target sight may be favourably extended allowing for the additional conversion to active colistin (Ratjen et al., 2006; Yapa et al., 2014). Although colistin has been shown to be superior to other antibiotics in anaerobic conditions, data is conflicting with regards to the PK of aerosolised colistin due to different study methods used and as such, results should be interpreted with caution (Dalhoff, 2014; Bos et al., 2017). Also important to consider, mainstream clinical studies often determine efficacy by defining clinical cure and organism suppression which is difficult to attain in CF patients since once chronic infection is established, eradication of *P. aeruginosa* is impossible (Emiralioglu et al., 2016). Alternative outcome measures for these patients should include enhanced lung function and decreased bacterial density (Dalhoff, 2014).

Ratjen and colleagues (2006), aimed to establish the PK of colistin following an inhaled dose of 2 MU in 30 CF patients. The study reiterated the minimal systemic concentrations of colistin achieved following inhaled therapy with only 1.3% of the dose detected in the urine of study patients. Peak sputum concentrations were achieved one hour after the inhaled dose. Extremely high drug concentrations with MIC values ten times greater than international colistin breakpoints were reflected and maintained above the MIC value for at least eight hours post dose administration. As a consequence of these findings, a twice a day dosing regimen was recommended (Ratjen et al., 2006). Yapa et al. (2014), aimed to establish the PK of inhaled colistin in six patients with CF. Results of this study were similar to those of Ratjen et al. (2006) and demonstrated the advantages of inhaled drug delivery in achieving concentrations efficacious against *P. aeruginosa* strains with high MIC's (Yapa et al., 2014). A randomised clinical trial conducted by Hodson et al. (2002) compared the outcomes of inhaled colistin versus tobramycin in CF patients with chronic *P. aeruginosa* infection. This study found that nebulised colistin significantly reduced the prevalence of *P. aeruginosa* in sputum, however, it did not improve or restore lung function (measured as a change in the study populations forced expiratory volume (FEV) which tobramycin was able to achieve (Hodson et al., 2002). In contrast, a multi-centre study comparing inhaled dry powder colistin to tobramycin nebulized solution showed colistin to be non-inferior to tobramycin in terms of improvement in lung function (Schuster et al., 2013).

A 2011 Cochrane review on inhaled antibiotic therapy in CF concluded that a true meta-analysis to determine the superiority between colistin and tobramycin is impossible due to the large variation in study designs but recommended inhaled therapy for CF patients to improve lung function and prevent infection exacerbations (Ryan et al., 2011). However, a network meta-analysis on this matter concluded that all available inhaled antibiotic agents for CF have comparable efficacy with a slight advantage of tobramycin over colistin and aztreonam (Littlewood et al., 2012). Members of the pulmonary clinical practice guidelines committee established by the United States CF foundation, believe that evidence relating to the efficacy of inhaled antibiotics other than tobramycin in the treatment of CF patients was insufficient to assess appropriate outcomes due to the limited number of studies available (Flume et al., 2007; Mogayzel et al., 2013). Due to the conflicting literature, there is inadequate evidence to conclusively support the recommendation of one antibiotic regimen

for the treatment of *P. aeruginosa* (Langan et al., 2015). This was reiterated by another Cochrane review conducted recently which was still inconclusive as to the appropriate antibiotic regimen to be used in CF to eradicate *P. aeruginosa* infections and improve associated morbidity and mortality (Langton and Smyth., 2017).

Although general consensus is yet to be established, recent evidence in the literature supports inhaled and adjunctive IV therapy is recommended for the treatment of acute MDR *P. aeruginosa* exacerbations in CF. Suggested combinations include cephalosporins or carbapenems (IV) along with inhaled tobramycin or colistin for a duration of 14 days in order to optimise survival and prevent lung function decline (Antoniou and Elston, 2016; Elborn, 2016).

According to the limited PK studies conducted on inhaled colistin administration in CF patients, the recommended appropriate dosing strategy is 2 MU given in 12 hourly intervals (Ratjen et al, 2006; Yapa et al., 2014). In addition, the South African CF Association published a CF consensus guideline in 2007 which also supports the 2 MU 12 hourly colistin dosing strategy for these patients (SACFA, 2007).

1.10. Colistin use in lower respiratory tract infections and ventilator associated pneumonia

Pneumonia, an infection of the lung tissue, is a serious illness that causes millions of admissions into hospitals each year and is associated with high morbidity and poor patient outcomes. Many patients develop pneumonia due to common community acquired pathogens, however, nosocomial pneumonias are problematic to treat and those acquired as a result of mechanical ventilation (VAP) can be even more detrimental to patient prognosis (Tumbarello et al., 2013). For critically ill patients in high level care units with such LRTI's, Gram-negative organisms account for approximately 65% of cases (Wenzler et al., 2016). Due to the steady increased prevalence of MDR and XDR Gram-negative organisms in the hospital setting colistin has become an appropriate treatment option for these infections as salvage therapy, however consensus on the appropriate use of inhaled colistin for this patient population is still yet to be established (Kofteridis et al., 2010).

1.10.1. Adjunctive (dual) versus mono-therapy of aerosolised colistin in lower respiratory tract infections.

In general, it is thought that aerosolised colistin should be used as adjunctive therapy to IV colistin for the treatment of serious MDR Gram-negative LRTI's (Kofteridis et al., 2010; Wenzler et al., 2016) This recommendation follows the consideration that lung tissue concentrations of colistin after IV administration are low (Li et al., 2006) and studies have shown that high concentrations of colistin are achieved in sputum and bronchial secretions following aerosolised colistin administration (Michalopoulos and Papadakis, 2010). These concentrations have been shown to sustain for 8-12 hours in the lung tissue of most patients following inhalation (Michalopoulos and Papadakis, 2010). As such, the practice of concurrent IV and inhaled colistin is considered most appropriate for this subset of patients in order to achieve best possible outcomes; however, evidence to support this approach has been limited.

Michalopoulos et al. (2008), evaluated 60 VAP patients who received inhaled colistin, 57 of which were in combination with IV colistin or other antibiotic agents. The findings revealed that 83.3% of patients achieved clinical or microbiological resolution and concluded that inhaled colistin may be considered as adjunctive therapy for VAP patients. However, it did not directly assess the impact of inhaled colistin with IV colistin compared to other IV antibiotics (Michalopoulos et al., 2008). A randomised control trial conducted by Rattanaumpawan et al. (2010), aimed to establish if aerosolised colistin as adjunctive therapy for the treatment of MDR Gram-negative VAP was safe and advantageous. Results of 51 patients who received inhaled colistin with other systemic antibiotics were compared to 49 patients who received inhaled sterile normal saline solution with other systemic antibiotics. Although the study could not establish an advantageous outcome of inhaled colistin over the placebo, it did reveal that the duration of systemic antibiotics of those patients who received inhaled colistin was reduced by two days (Rattanaumpawan et al., 2010).

Kofteridis and colleagues (2010), undertook a matched case control study comparing the outcomes of nebulised and IV colistin to IV colistin alone (43 patients in each treatment arm) for the treatment of VAP. The study could not establish any clinical, microbiological or mortality benefit with the addition of aerosolised colistin to IV colistin therapy. Similar findings were also demonstrated by Demirdal et al. (2016) and Gu et al. (2014). Tumbarello

et al. (2013), found no improvement in ICU length of stay or mortality comparing aerosolised colistin in combination with IV colistin to colistin monotherapy. However, the study did find improved clinical cure in patients treated with colistin dual therapy regimens. In addition, the length of mechanical ventilation in patients who received adjunctive inhaled colistin was reduced by four days (Tumbarello et al., 2013). Korbila et al. (2010), found that the use of nebulised colistin combined with IV colistin to be an independent factor of VAP cure compared to IV only treatment. These studies all involved relatively small patient cohorts and none demonstrated improvement in overall mortality between the two patient groups (Kofteridis et al., 2010; Korbila et al., 2010; Tumbarello et al., 2013; Gu et al., 2014; Demirdal et al., 2016).

However, a systematic review and meta-analysis conducted by Liu et al. (2015), aimed to clarify the incongruent findings of the preceding studies and establish the efficacy and safety of combined aerosolised and IV colistin versus IV colistin alone for the treatment of MDR Gram-negative nosocomial pneumonia. With a pooled sample of 672 patients, significance and improvement was established in patients who received both IV and inhaled colistin in terms of clinical cure, microbiological cure and all-cause mortality with no evidence of additional side effects (Liu et al., 2015). This study established for the first time the clinical benefits and improved outcomes of the dual therapy colistin strategy for nosocomial LRTI's (Liu et al., 2015). Valachis et al. (2015), also conducted a systematic review and meta-analysis, the findings of which were similar to those of Liu et al. (2015). However, in the analysis, improvement in infection related mortality could be established but not in overall mortality between patients receiving inhaled and IV colistin compared to IV colistin monotherapy (Valachis et al., 2015).

Tulli and colleagues (2017), found that therapeutic regimes including colistin either inhaled or systemic for the management of VAP did not demonstrate inferior outcomes when compared to standard treatment regimens. Vardakas et al. (2017), conducted a systematic review and meta-analysis of 373 patients to establish the efficacy and safety of aerosolised colistin alone (without concomitant IV therapy) for the treatment of MDR nosocomial pneumonia since the combined therapy may lead to increased healthcare costs and systemic related toxicities. The review revealed that no difference in mortality could be established and microbiological and clinical cure was as effective using inhaled colistin monotherapy. Jang

et al. (2017), and other previous studies (Kwa et al., 2005; Lu et al., 2012), demonstrated similar findings concluding that aerosolised colistin on its own represents a valid alternative for the treatment and management of MDR VAP. However, Gutierrez-Pizarraya et al. (2017), cautioned against this strategy since the risk of bacterial systemic dissemination to cause bacteraemia in critically ill patients with nosocomial pneumonia is high and recommended that aerosolised colistin be administered in combination with IV antibiotics to ensure clinical and microbiological cure for these fatal infections.

In contrast, however, Rello et al. (2017) issued an ESCMID position paper on the use of aerosolised antibiotics for LRTI's in mechanically ventilated patients. The panel concluded against recommending the utilization of inhaled antibiotics for the treatment of VAP due to the lack of robust supporting evidence and the rates of related respiratory complications when antibiotics are administered through this route (Rello et al., 2017). Although recognised as common practice, the utilization of aerosolised colistin alone without concomitant IV therapy was also not recommended whilst the use of dual route colistin therapy was cautioned due to patient safety concerns (Rello et al., 2017). As such, consensus on adjunctive IV colistin with inhaled colistin versus monotherapy of inhaled colistin with or without IV therapy of other antibiotics for the treatment of nosocomial pneumonia in general is lacking.

1.11. Antibiotic resistance in South Africa

In an editorial entitled "Wake up, South Africa! The antibiotic horse has bolted," it was stated that South Africa has become reliant on colistin as a final option for the treatment of MDR Gram-negative infections including the CPE labeling it as a "home grown" multifaceted problem (Mendelson et al., 2012). Furthermore, carbapenem susceptibility was shown to decrease by 18% over a four year period in South African public sector hospitals and in 2011, 13.6% of blood stream infections caused by *A. baumanii* were resistant to colistin in a public Cape Town hospital (Visser-Kift et al., 2014). A point prevalence study conducted locally by Paruk et al. (2012), evaluated antibiotic prescription practices in the intensive care units (ICU) of both public and private sector hospitals in five provinces. This study found that unsuitable antibiotics were initiated in over 50% of patients reviewed and 72% of these patients received antibiotic therapy for an inappropriate duration. Alarmingly, van Boeckel et al. (2014) noted

that antibiotic consumption increased dis-proportionately to population growth during 2000-2010 in the BRICS countries, of which South Africa is one. The problems experienced currently with drug-resistant tuberculosis and non-*albicans Candida* infections, which are resistant to first line antifungal therapy, further enhances the crisis South Africa is facing with MDR organisms (Mendelson and Matsoso, 2014). In addition, the CPE organisms have been detected across the country in most cities and towns such as Johannesburg, Pretoria, Cape Town, Bloemfontein, Port Elizabeth and Witbank (Brink et al., 2012).

The current state of antibiotic resistance in South Africa and its impact on public health was summarized by Sekyere (2016) whereby evaluation of carbapenem resistance amongst *Enterobacteriaceae* over a six year period revealed detection of over 2300 isolates with increased prevalence in Gauteng province followed by KwaZulu-Natal. The NDM-1 and OXA-48 carbapenemases were most abundantly identified. Investigations of these cases revealed that the majority of patients had no travel history outside of the country which may indicate that these enzymes emerged as a direct result from increased carbapenem use and exposure locally. In South Africa, an exponential increase in carbapenem utilization occurred between 2009 and 2011 as a consequence of rising rates of ESBL producing *Enterobacteriaceae* infections. This in turn may have contributed to the risk of selective pressure for the emergence of CPE in the country (Sekyere, 2016).

1.12. The emergence of colistin resistance

In recent years an alarming increase in documented reports have emerged worldwide indicating instances of colistin-resistance in Gram-negative pathogens (Jayol et al., 2014; Coetzee et al., 2016). The first reports of colistin resistant organisms were described in 1999 from the Czech Republic and these remained isolated and sporadic until very recently (Coetzee et al., 2016). Locally, Brink et al. (2013) reported a case of pan-resistant OXA-181 producing *K. pnuemoniae*. In such instances the consequences of MDR and pan-resistant organisms are dire and are associated with an increased risk of patient mortality. This is because there are no antibiotics available to treat these fatal infections.

The colistin resistant plasmid mediated *mcr*-1 gene was recently identified in human and animal samples in China (Liu et al., 2016). Reports of this gene have since been identified in over 17 countries. In South Africa, detection of this gene has occurred in multiple cities including Johannesburg, Pretoria and Cape Town in clinical isolates from hospitalized (n=3) and community (n=6) patients, as well as in poultry samples (Coetzee et al., 2016). Such evidence further highlights the antibiotic resistant problems the country is facing (Coetzee et al., 2016; Al-Tawfiq et al., 2017). Since then, *mcr*-2 (Xavier et al., 2016) and *mcr*-3 (Litrup et al., 2017; Yin et al., 2017) colistin resistant genes have been identified suggesting the prompt adaptability of the resistance mechanism of this gene. The plasmid mediated mechanism of resistance displayed by the gene allows for the swift capability of horizontal transmission between and within bacterial organisms and as such many more pathogens could become affected in the future (Litrup et al., 2017).

Brink et al. (2012) believe that "suboptimal dosing may also be a contributing factor for the development of resistance" and that antibiotics reserved as final options, such as colistin, should be dose optimized and avoided as mono-therapy administration in an attempt to curtail the current resistance crisis.

1.13. Antibiotic stewardship

There are multiple factors that contribute to the crisis of antibiotic resistance thus it is naïve to believe that one single solution can solve the problem. However, numerous initiatives globally and nationally are orchestrating mechanisms in which to minimize the threat of MDR organisms primarily through the promotion of appropriate infection control programs and advocating the judicious use of antimicrobial agents through antibiotic stewardship programs (ASP). These initiatives are to ensure the sensible use of antibiotics and the most positive outcomes for patients in an attempt to decrease antimicrobial resistance; thus the primary goal of any antibiotic stewardship program is to improve patient care and healthcare outcomes (Dodds Ashley et al., 2014). Antibiotic stewardship is a colloquial term used to describe initiatives and interventions that can improve antibiotic prescribing practices. It includes the evaluation and monitoring of the appropriate drug, dose, duration and route of antibiotics to optimize patient safety and outcomes (File et al., 2014).

International organizations including the Centre for Disease Control (CDC), and the European Society of Clinical Microbiology and Infectious Disease (ESCMID), as well as, locally the Federation of Infectious Diseases Society of South Africa (FIDSSA) and the South African Antibiotic Stewardship Program (SAASP) recommend antibiotic stewardship programs to manage organism resistance problems in all hospitals. Mendelson et al. (2012) advocated the "return to rational antibiotic prescribing through strong antibiotic stewardship" guided by specific programs for South Africa. This message is further enhanced by the 'Best Care Always' South African organization which provides guidelines relating to the implementation of antibiotic stewardship practices.

The favorable impact of ASP initiatives internationally has demonstrated reductions in antibiotic costs, antibiotic resistance, hospital length of stay and, unintended consequences of antibiotic therapy such as *Clostridium difficile* (Goff et al., 2012). Such stewardship interventions include: formulary restriction, IV to oral therapy conversion, prospective audit and feedback methodologies pertaining to, amongst others, dose, duration, compliance to obtaining a culture prior to antibiotic administration and, streamlining of antibiotic therapy following such results (Goff et al., 2012; File et al., 2014). Additional stewardship processes include: therapeutic drug monitoring of numerous antibiotics, vaccination campaigns, automatic stop orders and antibiotic batching. Many of the initiatives described are led by infectious disease specialist physicians and pharmacists, however, such models are difficult to replicate in the South African setting as such expertise are limited (Brink et al., 2016).

In order to adapt these initiatives to the South African context, existing resources such as pharmacists and nurses are ideally placed to develop, and execute antibiotic stewardship initiatives in healthcare settings (Schellack et al., 2016). The collective impact of hospital pharmacists and their critical role as pivotal members of multi-disciplinary teams in various antibiotic stewardship initiatives has been demonstrated recently across a private hospital network in South Africa (Table 1.3). As is evident from Table 1.3, the general pharmacist can lead and make a difference in antibiotic stewardship initiatives which have a direct and positive impact on overall patient care. Although much of this work occurred in the South African private hospital sector, it is applicable and can be adapted for implementation in public hospitals too, with appropriate institutional support and allocated 'protected

stewardship time,' as these principles are universal and applicable to all settings where antibiotics are prescribed. Boyles et al. (2013) showed that the implementation of a dedicated antibiotic prescription chart and weekly antibiotic stewardship ward rounds reduced antibiotic consumption and cost without impacting readmission rates and patient mortality in a Western Cape public hospital.

Table 1.3. The hospital pharmacists impact in various antibiotic stewardship initiatives in South African private hospitals

Antibiotic Stewardship Initiative	Pharmacists Impact
Implementation of a "hang time" intervention to improve the time from antibiotic prescription to administration in hospital settings (Messina et al., 2015)	With every hour in delay of antibiotic administration mortality can increase by 7.6% in patients with sepsis and septic shock (Kumar et al., 2006). Therefore, ensuring the timely administration of antimicrobials is critical in the management of patients with infections. Implementation of a pharmacist driven initiative to ensure the prompt administration of antibiotics within one hour following prescription (commonly known as antibiotic 'hang time') significantly increased compliance to a 'hang time' by 47%.
Implementation of "low – hanging fruit" stewardship interventions to decrease antibiotic consumption (Brink et al., 2016)	Pharmacists undertook a prospective audit and feedback method to implement and monitor five foundational stewardship interventions including: Duration of antibiotics greater than seven and 14 days; ensuring a culture is taken prior to the commencement of antibiotic therapy; inappropriate duplicate antibiotic cover and the concurrent co-administration of more than four antibiotics. An intervention was required for one in every 15 prescriptions and overall antibiotic consumption significantly decreased over the study period by 18%.

Antibiotic Stewardship Initiative	Pharmacists Impact
Improving compliance to surgical prophylaxis guidelines to decrease surgical site infections (Brink et al., 2016)	Appropriate peri-operative antibiotic prophylaxis is critical in minimizing the risk of surgical site infections (SSI) post-operatively. Pharmacists undertook to improve compliance to a bundle of four antibiotic prophylactic measures including: appropriate agent, appropriate dose, appropriate time of administration and appropriate duration of prophylaxis based on a recommended peer reviewed guideline mainly for caesarean sections and orthopedic surgeries. There was a significant improvement in compliance with all process measures and overall bundle compliance significantly increased by 24.7%. This had a direct impact on the SSI rate which decreased by 19.7%.

These proven strategies are important processes that can contribute to the appropriate use of antibiotics and the limitation of antibiotic resistance in low to middle income countries such as South Africa.

1.13.1. Obtaining microbiological cultures as a fundamental stewardship tenet

One of the foundational principles of antibiotic prescribing includes obtaining an accurate infectious disease diagnosis. This is done through: a) establishing the site of infection, b) understanding the co-morbidities of the patient and, c) establishing a microbiological diagnosis (Leekha et al., 2011). The effective management of resolving infectious diseases relies heavily on isolating the specific organism or pathogen that may be causing the illness. In order to optimize microbiological diagnoses, specimens should be collected timeously, appropriately minimising contamination risks and, prior to the initiation of antibiotic therapy, to obtain an accurate result of the infecting organism (Leekha et al., 2011). The practice of performing microbiological cultures prior to the administration of antibiotic therapy and their corresponding results also form the foundation of antibiotic stewardship since antibiotic

therapy can then be tailored to the most suitable, narrowest spectrum agent according to the identified pathogen. This then attempts to minimise the patients' exposure to broad-spectrum antibiotic therapy in an effort to reduce the selection of antibiotic resistant organisms (Dellit et al., 2007).

In South Africa, it remains the responsibility of the prescriber to request and order a culture to be taken for the patient.

1.13.2. Antibiotic de-escalation as a fundamental stewardship tenet

De-escalation is defined as the "reduction on the spectrum of administered antibiotics through the discontinuation of antibiotics providing activity against non-pathogenic organisms, discontinuation of antibiotics with similar activity or switching to an agent with narrower spectrum" (Garnacho-Montero et al., 2015). De-escalation following knowledge of the causative organism and its sensitivity profile is a fundamental component of an ASP as it limits exposure to broad-spectrum therapy and helps to tailor empiric treatment thus aiming to minimise antibiotic resistance risks. The practice of de-escalation has been shown not to influence patient outcomes negatively and also shorten durations of therapy (Lew et al., 2015; Garancho-Montero et al., 2015) dispelling the myth that long durations of broad-spectrum therapy render more favourable outcomes.

The uniqueness of this colistin utilization study is that it is the first of its kind from South Africa, conducted across multiple hospitals, and including a reasonably large sample of patients. It also evaluates the compliance to locally available colistin dosing guidelines and reviews the utilization of colistin in relation to antibiotic stewardship principles and parameters.

1.14. Rationale of study

Many unanswered questions regarding colistin use including the appropriate dosing schedule, duration and combination of treatment exist in the literature. This highlights the importance of establishing a baseline of how this drug is prescribed in clinical practice and as such the need for a local utilization review is evident. To the best of my knowledge, scientific peer

reviewed reports regarding how and why colistin is used in South Africa are unavailable and compliance to current dosing guidelines is unknown. Establishing this is therefore an essential step forward in elevating the stewardship processes in South African hospitals for this last resort antibiotic agent.

1.15. Research aims and objectives

The aim of this study was to retrospectively evaluate the use of colistin and consider the clinical outcomes of patients while on colistin in four private sector South African hospitals; in order to establish a baseline of how the drug is used and provide insight to enhance the appropriate use of this antibiotic in the future.

The objectives of this study were therefore;

2. To ascertain colistin utilization including: dose, dose frequency, route of administration and duration of treatment.
3. To ascertain which were the most prevalent infecting micro-organisms and source of infections that necessitated the use of colistin.
4. To establish if appropriate antimicrobial stewardship principles are practiced during colistin therapy.
5. To establish patient outcomes while on colistin therapy including effects on renal function, hospital length of stay and overall in-hospital mortality.

CHAPTER TWO

STUDY METHODOLOGY

2.1. Study design

This study was a multi-center retrospective electronic record review conducted to investigate the appropriateness of colistin utilization in adult patients across four private sector hospitals in South Africa (including two hospitals each in Johannesburg and Pretoria). The study was conducted over a ten month period from 1 September 2015- 30 June 2016.

2.2. Hospital selection

The four participating hospital sites were purposefully selected for this study as they are large and highly specialized referral centers of excellence for complicated medical conditions. In addition, the four hospitals were identified as high colistin usage hospitals by the hospital group as together they accounted for over 70% of the groups' overall consumption of colistin. Furthermore, information on colistin use was readily available from these hospitals as they had already transitioned onto the electronic antibiotic and infection surveillance system namely Bluebird® in early 2015.

For the purposes of this dissertation and any related publications, the hospital names will remain anonymous in order to comply with the study approval requirements set out by the study ethics approval and hospital group research committee approval. This is also to protect the hospitals from any positive or negative feedback that may result from the study findings. Each hospital was able to request their results and the information provided does not reflect on the results of the other hospitals that participated in this study. When publishing the results of this study, the hospitals are referred to as Hospital 1, 2, 3 and 4.

2.2.1. Preliminary analysis of colistin utilization

Preliminary baseline data analysis indicated that 325 patients received colistin therapy in the four selected hospitals over a twelve month period in 2014 (Hospital group antibiotic utilization report, January 2015) thus indicating that a substantial number of patients could potentially be reviewed and incorporated in the study through the inclusion of these hospitals. Hospital one is a 346-bed, level one trauma hospital situated in Johannesburg which offers expertise in all medical disciplines except for maternity and pediatrics. Hospital two is a 222-bed Johannesburg based hospital with expertise in trauma and cardiology. Hospital three is a 358-bed, specialized healthcare center of excellence in Pretoria for trauma, general medicine, surgery, critical care and hematologic oncology. Hospital four, also in Pretoria, is a 470-bed institution with a wide variety of medical specialties including general surgery, cardiology and oncology. Table 2.1 shows the number of specialized care beds per site and the total number of patients on colistin at the time of preliminary baseline data analysis (1 January 2014 - 31 December 2014).

Table 2.1. **Number of specialized care beds and number of patients on colistin therapy at the four selected private hospitals (data collected 2014)**

Hospital	Number of adult ICU beds	Number of adult high care beds	Total number of patients on colistin*
Hospital 1	66	29	114
Hospital 2	32	8	16
Hospital 3	35	29	170
Hospital 4	65	33	25
Total	**198**	**99**	**325**

*This data comprises of all patients, adult and pediatric, for which colistin was dispensed including all possible routes of administration (intravenous, aerosolised and irrigation).

2.2.2. Antibiotic prescribing in the private healthcare sector of South Africa

It is important to note that in South Africa, private healthcare clinicians consult their services to this sector and are not employees of the hospital. Due to the autonomous nature of the private sector and the consequential inability by private hospital groups to be prescriptive in

prescribing practices required from the private practitioners; no restrictions or guidelines have been imposed for antimicrobial prescribing in private hospitals to date. The hospital group in which this study was conducted has a medication formulary which in itself is all inclusive and contains all antibiotics available on the South African market. In contrast to the South African public sector hospital system, private practitioners are also not bound to prescribe only according to the National Department of Health (NDoH) Standard Treatment Guidelines (STG's). As a result of the aforementioned factors, a true reflection of the prescribing of colistin could be determined for this study as it would not be influenced or biased by hospital or group imposed protocols or formularies.

2.3. Sample selection

2.3.1. Inclusion criteria

Adult patients (over the age of 18 years) at the participating hospitals who were deemed to have received colistin therapy as a result of the product being dispensed, and thus billed to the patients profile, via the IV and aerosolised routes of administration, were included in the study.

2.3.2. Exclusion criteria

Pediatric and neonatal patients and those receiving colistin via an alternative route of administration were excluded from the study. These exclusions were made because limited evidence exists regarding the appropriate use of colistin in the pediatric and neonatal patient populations, since safety and efficacy studies have not been conducted in these patient categories. Nor do approved recommendations for other routes of administrations, such as irrigations, exist. These practices are mostly off label and therefore were not included in the study. Patients who were dispensed and billed colistin but whose profiles were not appropriately updated on the Bluebird® electronic system and those with a large amount of missing electronic data were also excluded.

2.4. Ethical considerations

The necessary approvals were obtained from the individual participating hospitals and the hospital groups' research committee (Appendix B) and ethical clearance (M150404) was granted by the University of the Witwatersrand Human Research Ethics Committee (Appendix C) prior to the commencement of data collection.

2.5. The Bluebird® system and identification of patients

The retrospective electronic record review of adult patients on colistin treatment was conducted using the Bluebird® system. This web based electronic system integrates laboratory data from the main private laboratories, as well as, hospital medication dispensing data and the patient's admission master file (Figure 2.1). The electronic patient record derived from the system allowed for the identification of patient's in each hospital to whom colistin had been dispensed, the monitoring of laboratory culture results, their drug prescription data, hospital movements and overall in-hospital outcome. All aspects of a patients' antibiotic therapy were captured onto the Bluebird® system by local hospital ward pharmacists. Clinical biochemistry, hematology and serology results, organism culture results and corresponding sensitivity profiles were also available electronically on the system for review. The researcher was granted access to the Bluebird® system of the four participating hospitals for the duration of the study period by the hospital group.

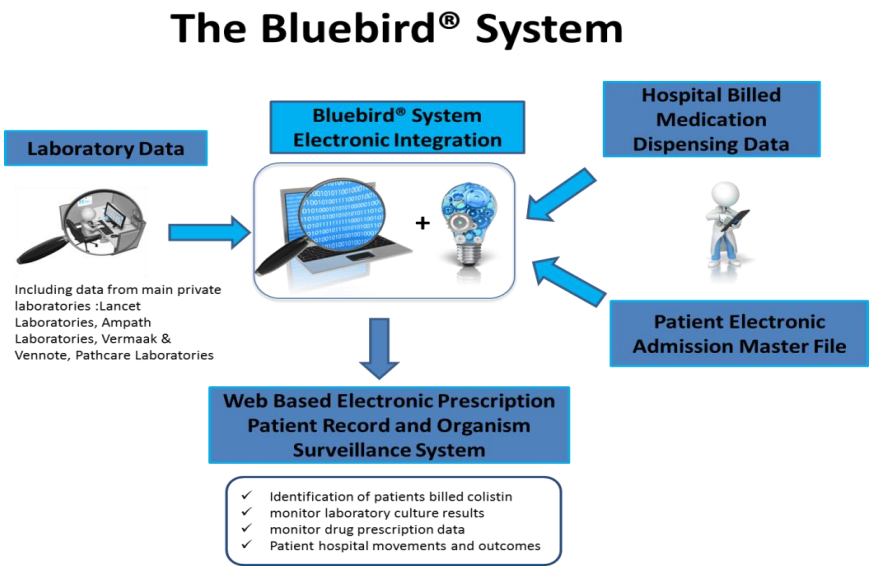

Figure 2.1. The integration processes of the Bluebird® electronic surveillance system used to retrospectively identify and evaluate records of patients on colistin therapy.

2.6. Data collected of patients prescribed colistin

The data described in Table 2.2 were collected per study patient on a standardized data collection template (Appendix D). Data was manually collected to include all the required demographic, clinical and therapeutic data for each patient on colistin (according to the study inclusion criteria) following review of patient records on the Bluebird® system. Findings were entered onto a spreadsheet using Microsoft® Excel for statistical analysis and qualitative interpretation. Data collection was coded to ensure patient and hospital confidentiality and any traceable information was kept electronically in a password protected folder.

Table 2.2. Data collected for patients prescribed colistin in four private sector hospitals in South Africa

Data indicator	Reason required for the study
Hospital identification code	To categorize patients according to hospitals for comparison of colistin prescription between hospitals.
Unique patient study number	To link the manually completed patient data collection forms (Appendix D) to the Bluebird® system for follow up of patient information throughout the hospital length of stay.
Date	To document the date of record review commencement of the study patient by the researcher.
Ward	To document the ward in which the study patient commenced colistin therapy for comparison of prescribing between high level care units and general wards (if any).
Patient gender	To document the gender of patients included in the study for patient demographic information.
Patient Age	To document the age of study patients to determine the age demographic range of adult patients requiring colistin therapy.
Patient weight	Weight is a variable required for the calculation of creatinine clearance to determine the drugs' effect on

Data indicator	Reason required for the study
	patient renal function pre and post colistin exposure and was recorded for this purpose.
Patient admitting diagnosis	To establish the primary reason for study patients' hospital admission.
Acute Physiology and Chronic Health Evaluation (APACHE II) score	To measure the severity of disease of study patients admitted to intensive care units.
The indication for commencement of colistin therapy	To determine the reason colistin therapy was required and prescribed. This was categorized according to the following: a) empiric therapy (if no evidence of an MDR or XDR Gram-negative organism was found prior to or during the course of treatment), b) directed therapy (infection with an MDR or XDR organism of known sensitivity), c) salvage therapy (failure of an alternative treatment where colistin was used as escalation therapy), d) No clinical reason (if there was no evidence to indicate a reason for colistin therapy at any point during the patients hospitalization including the review of sepsis markers deemed to be normal), e) Other (if the reason for colistin therapy did not fit any of the above mentioned categories).
Start date of colistin therapy	To record the date in which colistin therapy first commenced in order to assist in establishing total treatment days of colistin per study patient.
End date of colistin therapy	To record the date in which colistin therapy terminated (date of last colistin dose administered) in order to

Data indicator	Reason required for the study
	assist in establishing total treatment days of colistin per study patient.
Total days of colistin therapy	To determine the number of treatment days of colistin therapy per patient. The first day of colistin administration was counted as day one of therapy.
Route of colistin administration	To establish if colistin was prescribed either IV or via nebulisation in order to categorize patients according to route of administration.
The prescription of a colistin loading dose	To establish if a colistin loading dose was prescribed as is recommended best practice by local colistin dosing guidelines (Labuschagne et al., 2016 ; Visser-Kift et al., 2014)
Actual colistin loading dose prescribed	To record the actual colistin loading dose in million international units (MU) prescribed per patient.
Colistin maintenance dose and frequency prescribed	To record the maintenance dose prescribed per patient including the dose (MU) and the frequency of administration prescribed (hourly intervals of colistin administration).
First or repeat course of colistin	To establish if this was the first exposure to colistin for the patient or not.
Compliance to antibiotic 'hang time'	This data was recorded to establish if colistin was administered within one hour following prescription as is recommended for patients with sepsis (Kumar et al., 2006)
Laboratory cultures taken	To determine if an appropriate culture was taken prior to the commencement of colistin therapy in order to identify the possible causative organism of the infection.

Data indicator	Reason required for the study
Culture specimen type	To establish which clinical specimens were tested for possible organism growth. These were categorized as follows: - Urine - Blood - Trachael aspirate - Sputum - Other
Infecting organism cultured	To record and review the results of the organism identified following laboratory and microbiological review necessitating the use of colistin. This data was established from the laboratory report.
Organism sensitivity profile	To determine the resistance patterns of the organisms cultured and to establish appropriate drug-bug match (e.g.: if alternative therapeutic options were available to treat the organism or if colistin was the only option).
Minimum inhibitory concentration (MIC)	To document the MIC of antibiotics for the infecting organism identified in the study (when available) to provide further insight into the severity of resistance of the organism.
Other co-prescribed antibiotics	To record other Gram-negative antibiotics prescribed along with colistin since colistin monotherapy is not considered best practice (Richards et al., 2015)
Serum Creatinine (SCr) prior to and post colistin treatment	Used to establish the patient's creatinine clearance on the first and last days of colistin therapy in order to determine the antibiotics effect on renal function per patient (if any). This data was recorded from the serology test results from laboratory reports. The Cockcroft-Gault Equation was used to establish this:

Data indicator	Reason required for the study
	(((140-age in years) x (wt in kg)) x 1.23) / (serum creatinine in micromole/L) - A published South African colistin dosing guideline made dosing recommendations according to renal function based on creatinine clearance criteria. Therefore, this variable was required to establish colistin dosing compliance to this guideline (Labuschagne et al., 2016).
Estimated glomerular filtration rate (eGFR) prior to and poste colistin treatment	The eGFR was also recorded to establish the patients renal function on the first and last days of colistin treatment to determine the antibiotics effect on renal function per patient (if any). This data was recorded from the serology test results from laboratory reports. - Another South African colistin dosing guideline made dosing recommendations according to renal function based on eGFR criteria. Therefore, this variable was required to establish colistin dosing compliance to this guideline (Visser-Kift et al., 2014)
De-escalation of colistin therapy	De-escalation or streamlining of antibiotic therapy refers to the practice of tailoring therapy from broad-spectrum to narrow-spectrum following culture sensitivity results (Garnacho-Montero et al., 2015). De-escalation practices were reviewed to determine compliance to this antibiotic stewardship principle.
Choice of de-escalated antibiotic	To establish which antibiotic was the agent of choice on occasions when therapy was de-escalated.
Total length of stay (LOS) in the Intensive Care unit (ICU).	To determine the number of days spent by each study patient in the ICU as a study outcome measure.
Total LOS in hospital	To determine the overall duration of hospital admission (including ICU and general ward stay) spent by each

Data indicator	Reason required for the study
	study patient (measured in days) as an additional study outcome measure.
Overall in-hospital patient outcome	To establish if the study patients were discharged from the hospital or demised in hospital as a third study outcome measure.

2.7. Patients

Due to the retrospective record review nature of this study, there was no direct contact with patients. As such, no informed patient consent from the patient was needed and permission to use aggregated anonymised data for research purposes is granted as part of the hospital admission process. The patients' antibiotic dosing regimens were not affected in any way by this study. Confidentiality of patient and hospital data has been maintained throughout and only cumulative data is presented, therefore, data cannot be traced back to an individual patient.

2.8. Statistical analysis

Statistical analysis was conducted with the assistance of a statistician from the Data Management and Statistical Analysis (DMSA) consortium (http://www.dmsa.co.za/).

2.8.1. Sample size

Sample size was determined by the key research question to be answered. For the determination of the prevalence of patients with a particular characteristic (e.g. the percentage of females in the study group), a sample size estimation was based on a 50% prevalence (worst-case in terms of sample size), 5% precision and a 95% confidence interval. Based on this methodology and taking this study into consideration, a sample size of 385 patients was required. The actual sample size of 237 patients in this study corresponds to a precision of 6.4% (rather than 5.0%), which is acceptable.

Sample size for prevalence was determined using the formula (Daniel, 1999):

$$n=(Z^2 P(1-P))/d^2$$

where; n=sample size, Z=Z-statistic for the chosen level of confidence, P=expected prevalence or proportion and, d=precision.

2.8.2. Statistical methodology

Descriptive analysis of the data was carried out as follows: categorical variables were summarized by frequency and percentage tabulation, and illustrated by means of bar charts. Continuous variables were summarized by the mean, standard deviation, median and interquartile range (IQR), and their distribution illustrated by means of histograms. The X2 test was used to assess the relationships between categorical variables. Fisher's exact test was used for 2 x 2 tables or where the requirements for the X2 test could not be met. The strength of the associations was measured by Cramer's V and the phi coefficient respectively. The scale of interpretation used is summarized in Table 2.3.

Table 2.3. Interpretation of statistical associations between categorical variables

Statistical association strength	Interpretation of association
0.50 and above	high/strong association
0.30 to 0.49	moderate association
0.10 to 0.29	weak association
below 0.10	little if any association

The relationship between continuous and categorical variables was assessed by the t-test (or ANOVA for more than two categories). Where the data did not meet the assumptions of these tests, a non-parametric alternative, the Wilcoxon rank sum test (or the Kruskal-Wallis test for more than two categories) was used. The strength of the associations was measured by the Cohen's d-value for parametric tests and the r-value for the non-parametric tests. The scale of interpretation used is reflected in Table 2.4.

Table 2.4. Interpretation of statistical associations between continuous and categorical variables

Statistical association strength	Interpretation of association
0.80 and above	large effect
0.50 to 0.79	moderate effect
0.20 to 0.49	small effect
below 0.20	near zero effect

The relationship between the two continuous variables was assessed by Pearson's correlation coefficient. Where the data did not meet the assumptions of these tests, a non-parametric alternative, Spearman's rank correlation coefficient was used. The strength of the associations was measured by interpreting the absolute value of the correlation coefficient. The scale of interpretation used is described in Table 2.5.

Table 2.5. Interpretation of statistical associations between two continuous variables

Statistical association strength	Interpretation of association
0.50 and above	large effect
0.3 to 0.49	moderate effect
Below 0.3	small effect

Data analysis was carried out using SAS® version 9.4 for Windows. The 5% significance level was used. In other words, p-values <0.05 indicate significant results. Determination of significance was only conducted on the results of patients who received IV colistin therapy as the sample size of those who received nebulized colistin was too small to render valid appropriate interpretations.

2.9. Summary of study process

A summary of the complete methodical process undertaken for this study is depicted in Figure 2.2.

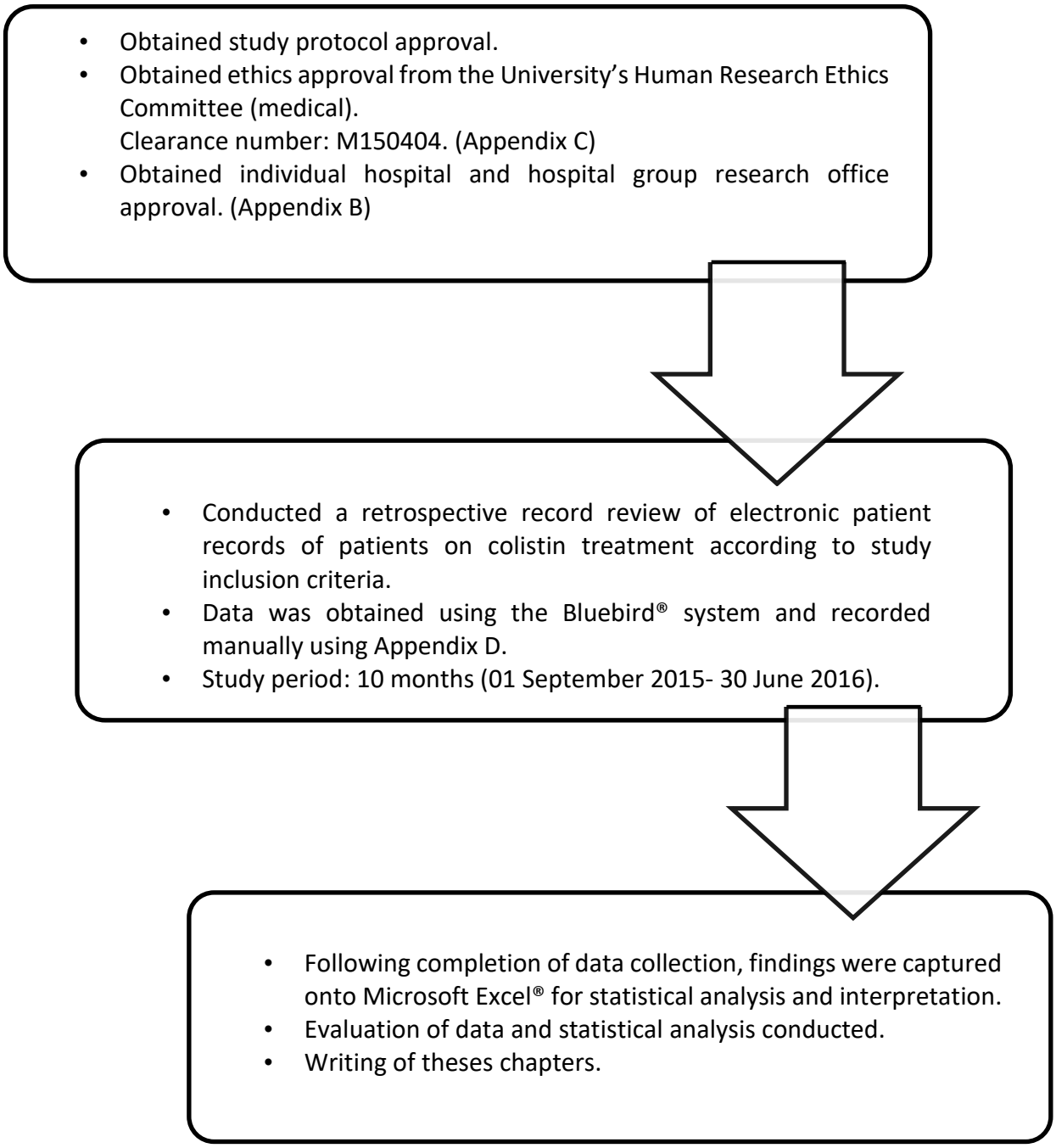

Figure 2.2. Summary of the study process and methodology

CHAPTER THREE

REVIEW OF INTRAVENOUS (IV) COLISTIN UTILIZATION ACROSS FOUR PRIVATE SECTOR HOSPITALS IN SOUTH AFRICA

3.1 Introduction

A total of 237 patients on colistin therapy from the four participating hospitals during the ten month study period were included in this study. Of these, 89.5% (n=212) of patients received colistin via the IV route of administration. For the purposes of this chapter, the results of the colistin utilization review conducted in these patients will be discussed. The remaining 10.5% (n=25) of patients received aerosolised colistin, the findings of which will be discussed in the chapter to follow.

3.2. Results

3.2.1. General patient demographics

The number of patients on IV colistin contributing to the study per hospital is depicted in Figure 3.1. Hospital four contributed the most number of patients 55.2% (n=117) followed by hospital one 26.4% (n=56), hospital two 10.4% (n=22) and hospital three 8.0% (n=17). The majority of patients were male 56.1% (n= 119) (Figure 3.2) and 80.7% (n= 171) received colistin whilst in the ICU (Figure 3.3). A smaller population of patients received colistin in general wards 19.3% (n=41). For 82.1% (n=174) of patients, this was their first exposure to colistin whilst 17.9% (n=38) had previously received a course of colistin therapy during their hospital admission (Figure 3.4). The mean age of patients included in the study was 50.9 years (SD 16.6; range 18-93 years). This mean age is similar to that described in other colistin utilization reviews conducted (Table 1.1).

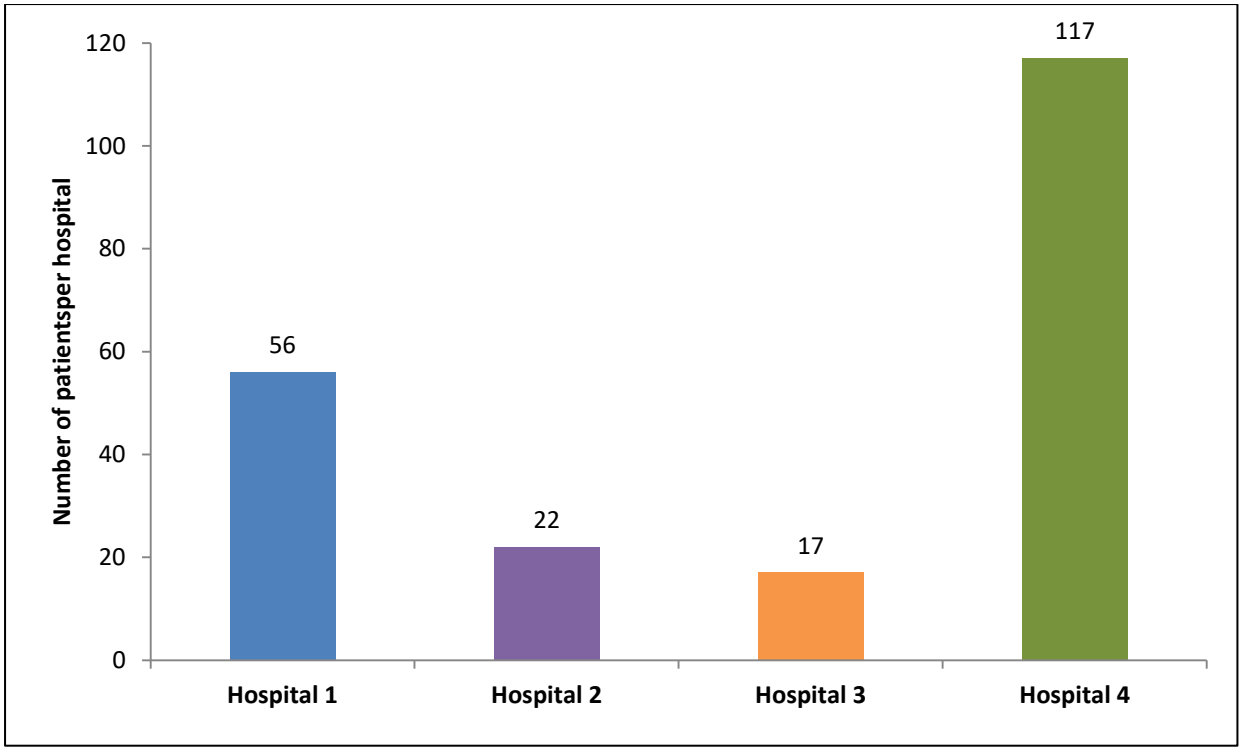

Figure 3.1. The number of patients per hospital on intravenous colistin therapy included in the study (n= 212)

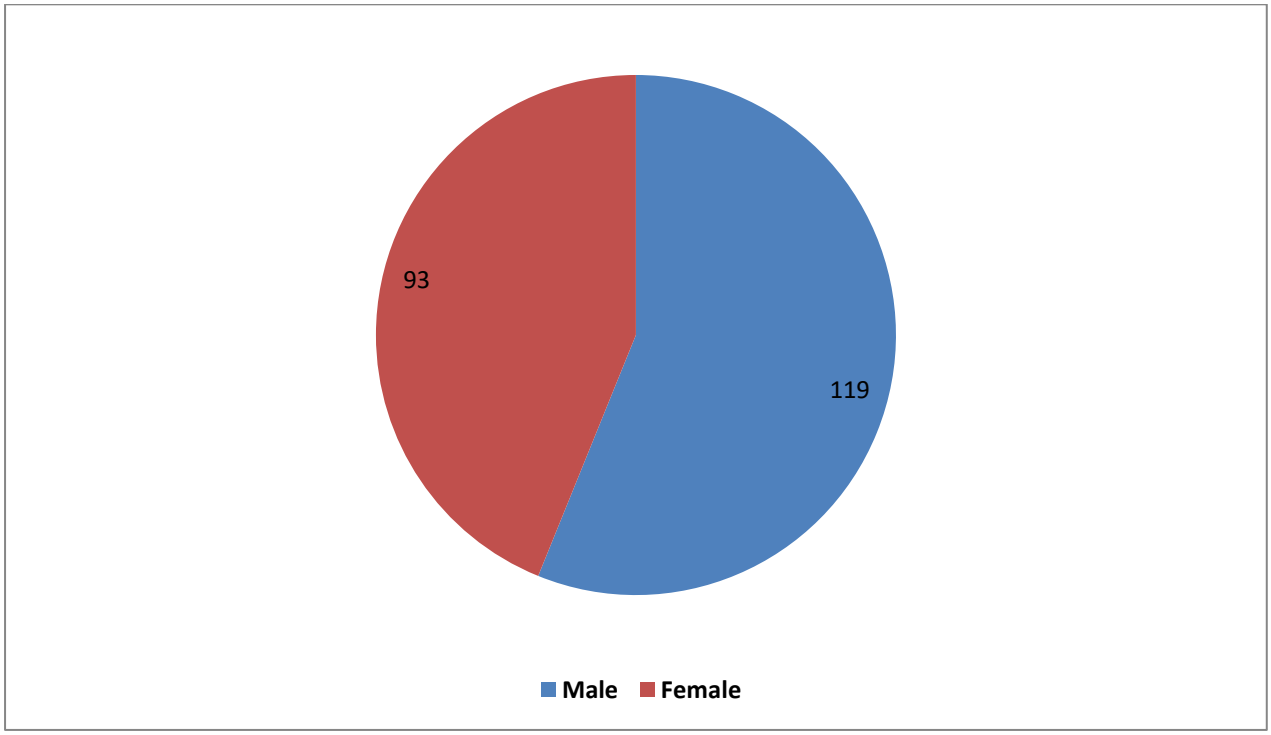

Figure 3.2. A breakdown of the number of patients on intravenous colistin therapy included in the study according to gender (n=212)

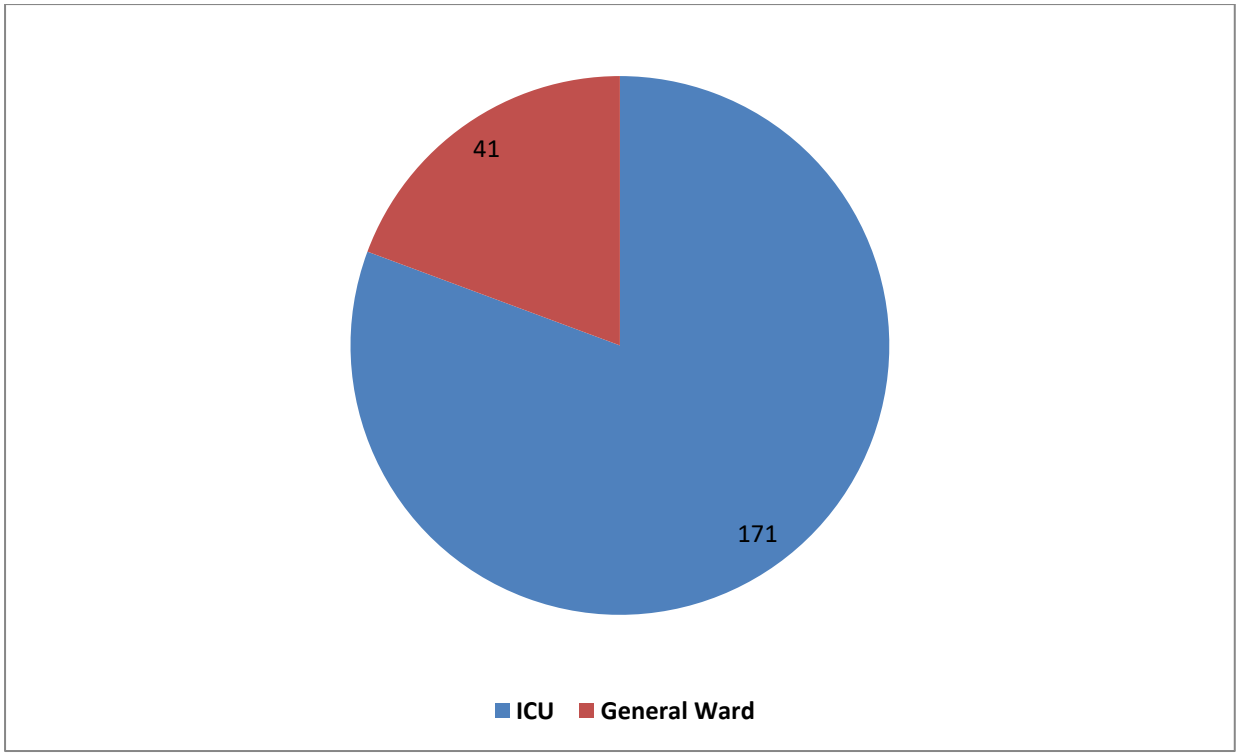

Figure 3.3. A breakdown of the number of patients on intravenous colistin therapy included in the study according to hospital ward location (n=212)

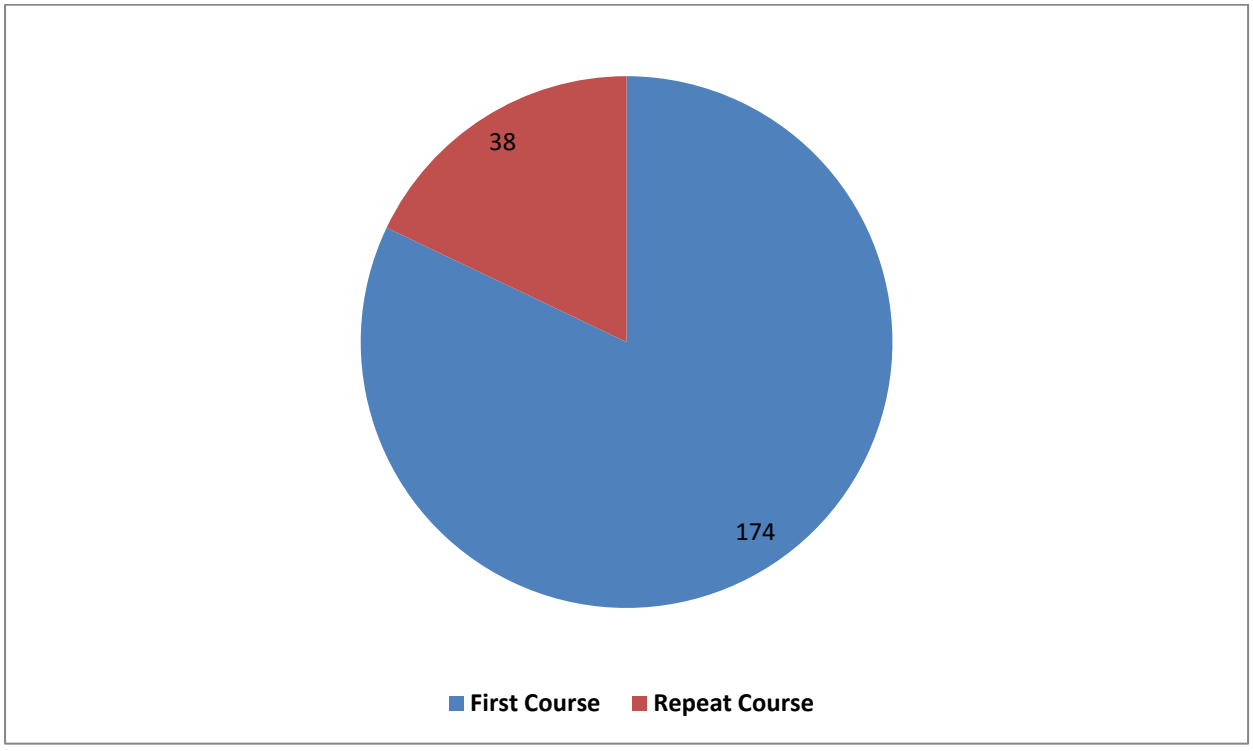

Figure 3.4. A breakdown of the number of patients for which an initial colistin course versus repeated colistin course was prescribed during their hospital admission (n=212)

A breakdown of the broad categories of admission diagnosis of the patients is depicted in Figure 3.5. Classification of admitting diagnosis according to exact ICD 10 codes could not be established from the electronic record as the exact admitting diagnosis was often not completed, nor was there a field on the system to record ICD 10 codes and, as such, broad categories of disease state admitting diagnosis were used to classify patients. All patients could be classified as critically ill receiving treatment in highly specialised tertiary level care hospitals. The most prominent of which was immune compromised, neutropenic, bone marrow oncology 40.1% (n=85) followed by trauma 11.3% (n=24) and blood stream infections 7.1% (n=15). Although an initial metric of this study, the illness severity score (APACHE II) score could not be recorded per patient as this was not documented on the electronic patient record. Upon further investigation, it was determined that this score is mostly not recorded on paper prescription charts in hospitals but rather kept by prescribers in their personal notes and records. Therefore patient risk in relation to mortality or outcome could not be corrected for or determined.

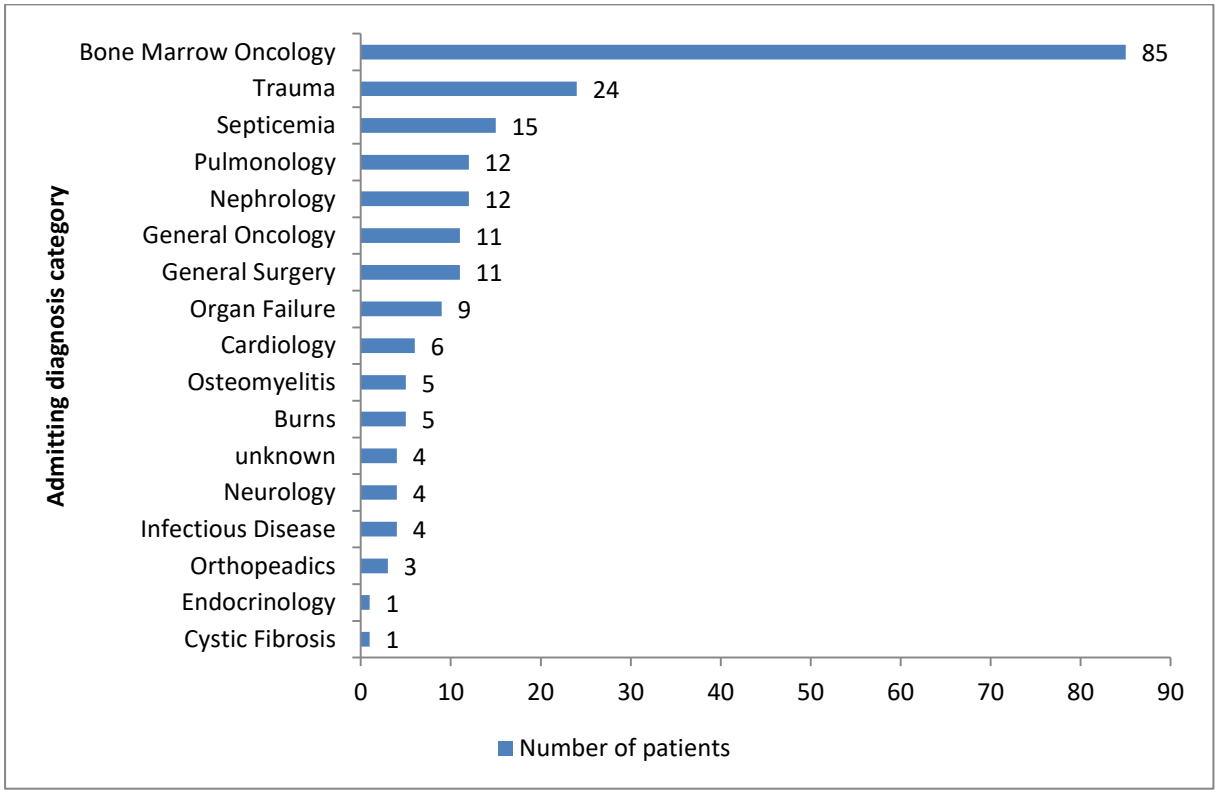

Figure 3.5. A breakdown of the number of patients per category of admitting diagnoses for patients included in the study on intravenous colistin (n=212)

The indication for colistin treatment (Figure 3.6) was mostly as directed therapy as a result of evidence of an infection with an MDR organism 57.1% (n=121), followed by empiric therapy 33.5% (n=71) and salvage therapy 9.4% (n=20), the study definitions of which are described in Table 2.2 (Chapter 2).

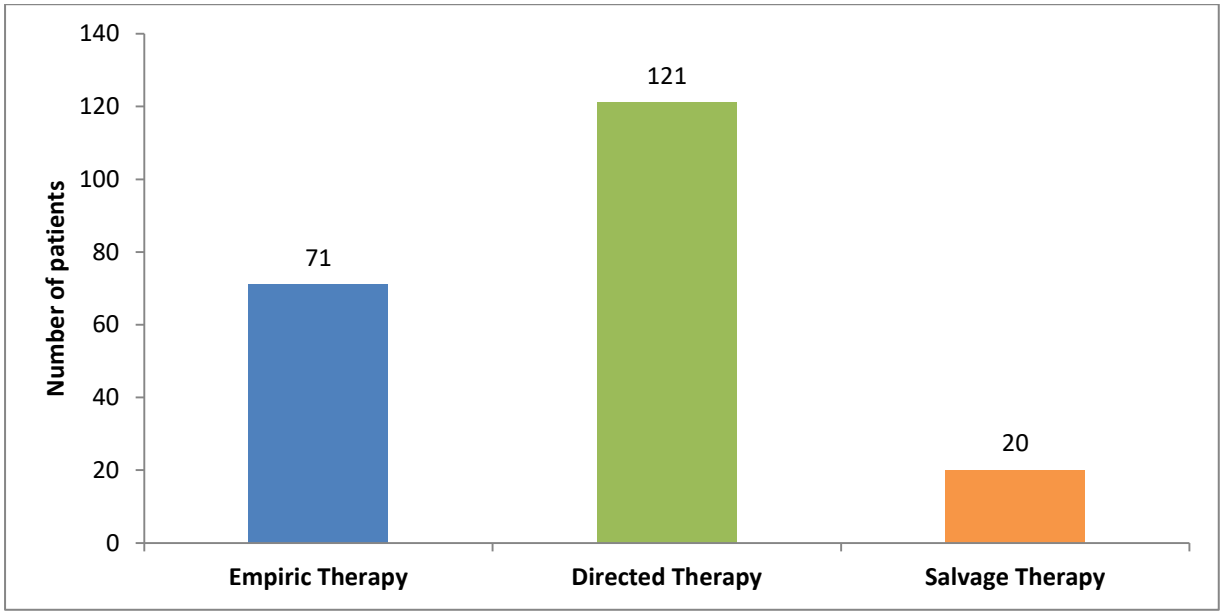

Figure 3.6. A depiction of the number of patients per indication for intravenous colistin therapy (n=212)

3.2.2. Microbiological assessment

Cultures were performed prior to the initiation of colistin therapy in all but two patients (Figure 3.7) thus, reflecting good compliance to this stewardship principle.

For the remaining 99.1% of patients (n=210), a breakdown of the specimen types tested for possible organism identification are described in Figure 3.8. Blood cultures were the most frequently tested specimens 57.4% (n=120) followed by urine 9.9% (n=21), sputum 8.1% (n=17) and tracheal aspirates 7.1% (n=15). The assumed inappropriateness of the 1.9% (n=4) of patients who were prescribed colistin following a result from a CPE rectal screening swab should be noted, as this is not considered a clinical specimen and positive results on such a screen indicate colonisation and not necessarily infection (Ruppe and Andremont, 2013). The entire clinical overview of these cases was not known, nor could have been established form the limited data available of the records available, and therefore it should be considered that

there may have been valid reasons for the commencement of colistin albeit deemed empirically for these patients.

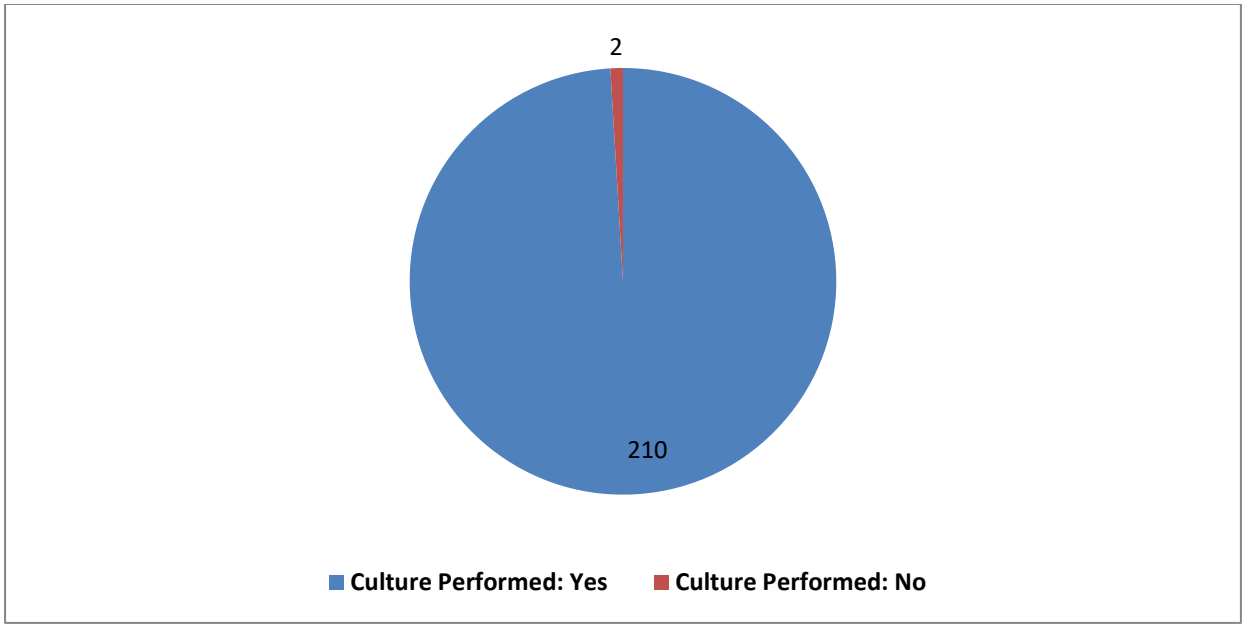

Figure 3.7. The number of patients who had a culture taken prior to the initiation of colistin therapy (n=212)

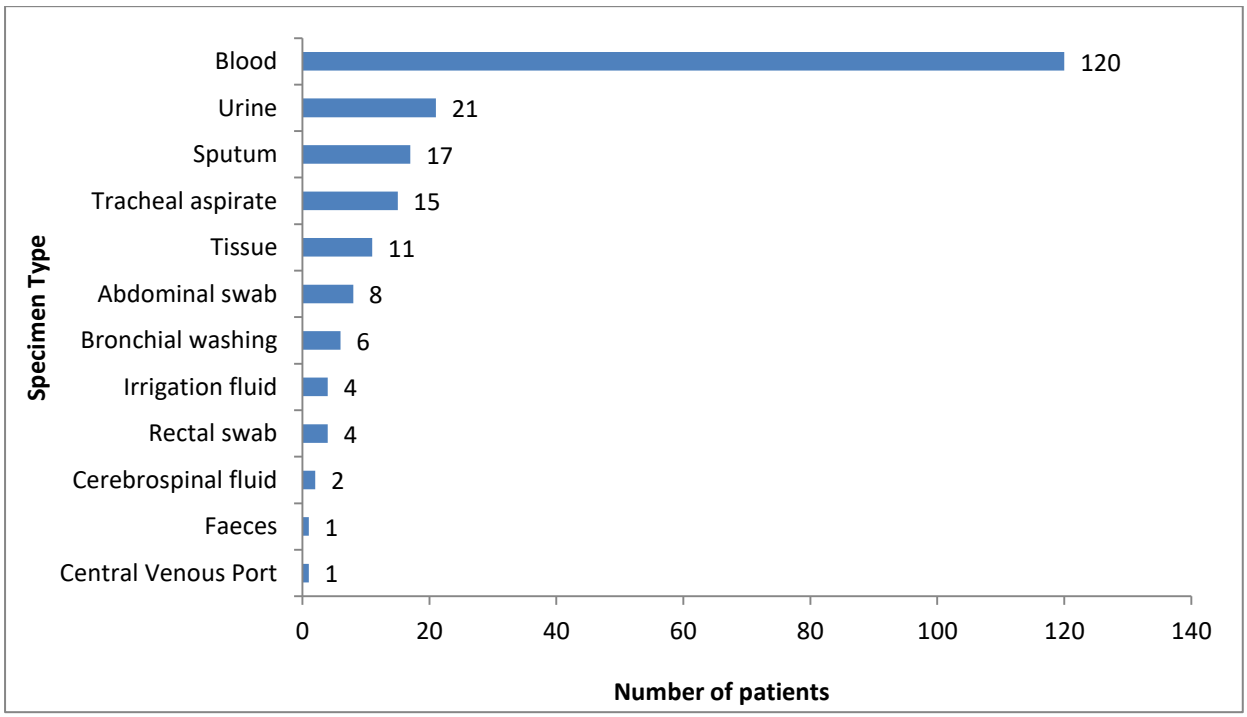

Figure 3.8. A breakdown of the number of patients and the various specimen types tested in patients who had cultures taken (n=210)

The prevalence of the various organisms identified is shown in Figure 3.9. *Klebsiella pneumoniae* 34.2% (n=81), *P. aeruginosa* 24.9% (n=59) and *A. baumanii* 9.3% (n=22) were the most predominant pathogens identified necessitating the use of colistin. These organisms also form part of the Gram-negative cluster of the "ESKAPE" pathogens, globally recognised as a group of concerning pathogens that have shown to be increasing in prevalence and highly resistant, drastically limiting treatment options for hospitalised patients and, impacting negatively on patient outcomes (Boucher et al., 2009).

For a substantial amount of patients in this study, however, no organism was identified 25.7% (n=54) although IV colistin therapy was still prescribed (Figure 3.9). For the remaining patients who were deemed to have received colistin empirically (Figure 3.6), 0.9% (n=2) did not have microbiological cultures performed and 7.0% (n=15) cultured an organism which was shown to be sensitive to antibiotic agents other than colistin. Due to the limitation of data available for review on the Bluebird system, in the absence of clinical notes and other clinical parameters including: fever, blood pressure, prescriber notes, severity of illness scores, and justification of prescriptions; determination of the exact reasons for empiric therapy could not be established.

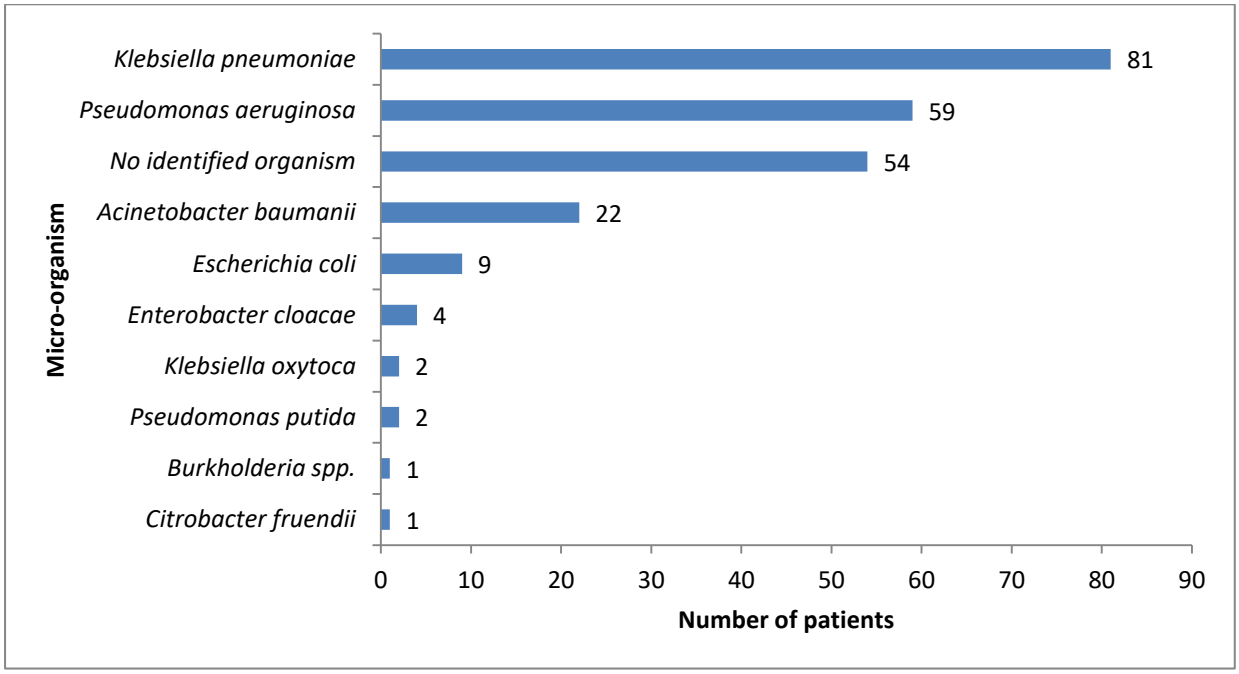

Figure 3.9. A breakdown of the number of organisms identified following laboratory culture results. Note that numbers do not sum to n=210 since some patients had more than one organism identified.

3.2.3 Evaluation of loading doses, maintenance doses and frequency of colistin doses prescribed in IV study patients

3.2.3.1 Evaluation of colistin loading doses prescribed

Compliance to the prescription of a loading dose was high with 93.9% (n=199) of patients prescribed a colistin loading dose. However, as depicted in Table 3.1, huge variation in the actual loading dose prescribed was noted. Therefore, of the patients who were prescribed a loading dose, 90.4% (n=180) received an appropriate loading dose (9-12 MU) indicating that 9.6% (n=19) received sub-optimal loading doses. For 15.1% (n=32) of study patients, loading doses were either not prescribed (n=13) or were too low (n=19) indicating inappropriate management of this process measure for this cohort of patients.

Table 3.1. Prescribed colistin loading doses for intravenous study patients (n=199)

Loading Dose (MU)	*%* of patients *(n)*
4	1.5 (3)
6	5.5 (11)
8	2.5 (5)
9	22.6 (45)
11	0.5 (1)
12	67.3 (134)

3.2.3.2 Evaluation of colistin maintenance doses and frequency of administration prescribed

Table 3.2 describes the colistin maintenance doses and frequency of administration prescribed for the study patients. The large variation of colistin maintenance doses prescribed is evident with dose ranges from 1-4.5 MU prescribed and frequencies including six, eight and 12 hourly intervals (Table 3.2). The majority, 99.5% (n=211), of patients studied were prescribed colistin at the appropriate frequency of administration. However, one patient in this study was prescribed colistin six- hourly which should be deemed as inappropriate as it could contribute to elevated risks of drug induced toxicity.

Table 3.2. Prescribed colistin maintenance doses and frequency of administration for intravenous study patients (n=212)

Maintenance Dose (MU)	% of patients *(n)*	Frequency of administration (hourly)	% of patients *(n)*
1	1.3 (3)	6	0.5 (1)
1.5	6.1 (13)	8	35.9 (76)
2	8.0 (17)	12	63.7 (135)
2.5	0.9 (2)		
3	30.7 (65)		
4.5	52.8 (112)		

3.2.4 Compliance of study patients to two South African colistin dosing guidelines.

3.2.4.1 Evaluation of the study patients compliance to the South African Society of Clinical pharmacy (SASOCP) colistin dosing guidelines

Compliance of the study patients to the SASOCP colistin dosing guidelines is described in Table 3.3. Dosing recommendations in this guideline are made according to Creatinine Clearance (CrCl). The Cockcroft-Gault equation was used to establish this based on the patients Serum creatinine (SCr), gender and weight (described in Chapter 2). Patient weight was difficult to obtain from the electronic record review as often this data was not recorded on the electronic patient profile. As such, CrCl was impossible to establish for these patients and, therefore, was the primary factor which contributed to the large number of patients (n=64) where guideline compliance could not be determined. For the remaining patients, compliance to prescribed colistin doses was very poor, 34.9% (74/212). Considering only those patients for which data was available, compliance to these guidelines was still poor at 50% (74/148). As such, this evaluation emphasized that not recording weight in this patient cohort is a process that should be targeted for improvement.

Table 3.3. Study patients' compliance to the South African Society of Clinical Pharmacy colistin dosing guideline (Labuschagne et al., 2016)

Guideline Recommendation	
Normal renal function	Loading dose: 12 MU, then maintenance dose: 3 MU 8 hourly or 4.5 MU 12 hourly
CrCl* 40-60ml/min	2 MU 12 hourly
CrCl* 10-40 ml/min	2 MU 24 hourly
CrCl* < 10 ml/min	1.5 MU 36 hourly
Study patients compliance to recommended dosing guidelines	
All categories of renal function	Unknown*: 30.2% (n=64) Compliant: 34.9% (n=74) Non-compliant: 34.9% (n=74)

*Contributions to unknown compliance are as a result of data not documented on records for variables including weight.

A detailed evaluation of patient renal function according to CrCl categories, colistin doses prescribed and guideline compliance is presented in Table 3.4. This table highlights the huge variation in colistin doses prescribed according to the various categories of renal function for the patients studied and informs the poor dosing compliance demonstrated by Table 3.3.

Table 3.4. Detailed evaluation of study patients renal function according to CrCl categories, colistin doses prescribed and guideline compliance

Cr clearance Prior	**Loading Dose (MU)**	**Dose (MU)**	**Frequency (hrs)**	***(n)***	**Percentage (%)**	**Compliance to guideline (Labuschagne et al., 2016)**
.	.	2	8	2	0.94	unknown
.	.	3	8	1	0.47	unknown
.	4	2	12	1	0.47	unknown
.	6	1.5	12	1	0.47	unknown
.	6	3	8	2	0.94	unknown
.	8	3	8	4	1.89	unknown

Cr clearance Prior	Loading Dose (MU)	Dose (MU)	Frequency (hrs)	*(n)*	Percentage (%)	Compliance to guideline (Labuschagne et al., 2016)
.	9	1.5	12	2	0.94	unknown
.	9	3	8	7	3.3	unknown
.	9	3	12	1	0.47	unknown
.	9	4.5	12	2	0.94	unknown
.	11	4.5	12	1	0.47	unknown
.	12	1	8	1	0.47	unknown
.	12	1.5	8	1	0.47	unknown
.	12	3	8	3	1.42	unknown
.	12	4.5	12	35	16.51	unknown
10-40	.	2	8	1	0.47	no
10-40	.	3	8	2	0.94	no
10-40	4	2	8	1	0.47	no
10-40	9	2	8	2	0.94	no
10-40	9	3	8	1	0.47	no
10-40	9	3	12	1	0.47	no
10-40	12	1	8	1	0.47	no
10-40	12	1.5	12	5	2.36	no
10-40	12	2	12	2	0.94	no
10-40	12	2.5	12	1	0.47	no
10-40	12	3	12	1	0.47	no
10-40	12	4.5	12	3	1.42	no
40-60	4	3	8	1	0.47	no
40-60	6	1	8	1	0.47	no
40-60	6	2	8	1	0.47	no
40-60	6	2.5	6	1	0.47	no
40-60	9	3	8	4	1.89	no
40-60	9	4.5	12	2	0.94	no
40-60	12	1.5	8	1	0.47	no
40-60	12	2	12	3	1.42	yes
40-60	12	4.5	12	2	0.94	no
>60	.	2	8	1	0.47	no
>60	.	3	8	5	2.36	no
>60	.	3	12	1	0.47	no
>60	6	3	8	5	2.36	no
>60	8	3	8	1	0.47	no
>60	9	1.5	8	1	0.47	no
>60	9	2	8	1	0.47	no

Cr clearance Prior	Loading Dose (MU)	Dose (MU)	Frequency (hrs)	*(n)*	Percentage (%)	Compliance to guideline (Labuschagne et al., 2016)
>60	9	3	8	12	5.66	no
>60	9	3	12	2	0.94	no
>60	9	4.5	12	7	3.3	no
>60	12	1.5	8	1	0.47	no
>60	12	1.5	12	1	0.47	no
>60	12	2	8	1	0.47	no
>60	12	2	12	1	0.47	no
>60	12	3	8	11	5.19	yes
>60	12	4.5	12	60	28.3	yes
				212	99.94	

-Unknown variable

3.2.4.2 Evaluation of the study patients compliance to a colistin dosing guideline published by Visser-Kift et al., 2014

Compliance of the study patients to the colistin dosing guideline by Visser-Kift et al. (2014) is described in Table 3.5. This guideline made dosing recommendations based on eGFR which is often easier to determine compared to CrCl as the estimation is calculated from biochemistry results. As such, less patient data was missing, as only four patients did not have an eGFR performed pre and post colistin therapy. Evaluation of colistin dosing for the remaining patients could be determined; however, compliance to appropriate dosing based on renal function was also low, 41.5% (88/212). When considering only those patients for which data was available, compliance to these guidelines was still poor at 42.3% (88/208).

A detailed evaluation of patient renal function according to eGFR categories, colistin doses prescribed and guideline compliance is presented in Table 3.6. Variation in dosing compliance is again demonstrated which reiterates the poor compliance to the recommended dosing guideline as per Table 3.5.

Table 3.5. Study patient compliance to a colistin dosing guideline (Visser-Kift et al., 2014)

Guideline recommendations	
Critically ill or severe sepsis	Loading dose: 9-12 MU
eGFR > 60 ml/min	4.5MU 12 hourly
eGFR 30-60 ml/min	3 MU 12 hourly
eGFR 10-30 ml/min	2 MU 12 hourly
eGFR <10 ml/min	1 MU 12 hourly
Study patients compliance to recommended dosing guidelines	
All categories of renal function	Unknown*: 1.9% (n=4)
	Compliant: 41.5% (n=88)
	Non-compliant: 56.5% (n=120)

*Contributions to unknown compliance are as a result of data not documented on records for variables including eGFR.

Table 3.6. Detailed evaluation of study patients renal function according to eGFR categories, colistin doses prescribed and guideline compliance.

GFR_Prior	**Loading Dose (MU)**	**Dose (MU)**	**Frequency (hrs)**	***(n)***	**Percentage (%)**	**Compliance to guideline (Visser-Kift et al., 2014)**
.	9	1.5	12	1	0.47	unknown
.	12	4.5	12	3	1.42	unknown
<10	9	3	12	1	0.47	no
<10	12	1.5	12	3	1.42	no
10-30	.	3	8	2	0.94	no
10-30	4	2	8	1	0.47	no
10-30	6	1.5	12	1	0.47	no
10-30	6	2.5	6	1	0.47	no
10-30	9	2	8	1	0.47	no
10-30	9	3	8	4	1.89	no
10-30	9	3	12	1	0.47	no
10-30	9	4.5	12	2	0.94	no
10-30	12	1	8	1	0.47	no
10-30	12	1.5	8	1	0.47	no

GFR_Prior	Loading Dose (MU)	Dose (MU)	Frequency (hrs)	*(n)*	Percentage (%)	Compliance to guideline (Visser-Kift et al., 2014)
10-30	12	1.5	12	2	0.94	no
10-30	12	2	12	2	0.94	yes
10-30	12	2.5	12	1	0.47	no
10-30	12	3	12	1	0.47	no
10-30	12	4.5	12	2	0.94	no
30-60	.	2	8	2	0.94	no
30-60	4	2	12	1	0.47	no
30-60	6	1	8	1	0.47	no
30-60	6	2	8	1	0.47	no
30-60	9	1.5	8	1	0.47	no
30-60	9	1.5	12	1	0.47	no
30-60	9	2	8	1	0.47	no
30-60	9	3	8	6	2.83	no
30-60	9	4.5	12	5	2.36	no
30-60	12	1	8	1	0.47	no
30-60	12	1.5	8	2	0.94	no
30-60	12	1.5	12	1	0.47	no
30-60	12	2	12	3	1.42	no
30-60	12	3	8	2	0.94	no
30-60	12	4.5	12	14	6.6	no
>60	.	2	8	2	0.94	no
>60	.	3	8	6	2.83	no
>60	.	3	12	1	0.47	no
>60	4	3	8	1	0.47	no
>60	6	3	8	7	3.3	no
>60	8	3	8	5	2.36	no
>60	9	2	8	1	0.47	no
>60	9	3	8	14	6.6	no
>60	9	3	12	2	0.94	no
>60	9	4.5	12	4	1.89	yes
>60	11	4.5	12	1	0.47	yes
>60	12	2	8	1	0.47	no
>60	12	2	12	1	0.47	no
>60	12	3	8	12	5.66	no
>60	12	4.5	12	81	38.21	yes
				212	99.94	

-Unknown variable

3.2.5 Compliance to the timely administration of colistin

Compliance to colistin 'hang time' in this study was poor with 54.7% (n=116) of patients deemed compliant and 27.8% (n=59) non-compliant. For 17.5% (n=37), compliance was unknown due to missing documentation of the prescription time on the electronic record. Only taking into account those patients for which 'hang time' could be determined, overall compliance was 66.3% (n=116/175).

The compliance to hang time was measured as per the data collection tool (Appendix D) as a simple "yes or no" and the reasons for hang time delay per patient were not available on the Bluebrid system nor were they recorded as part of the data collection process. As such, the exact reasons for the delay could not be established from this analysis.

3.2.6 Combination therapy

Combination therapy was prescribed in 98.6% (n=209) patients. For 63.7% (n=131) of the patients a single other antibiotic was prescribed, while 33.5% (n=70) and 3.8% (n=8) had two and three other antibiotics prescribed, respectively. A breakdown of the concurrently administered antibiotics is depicted in Figure 3.10. Meropenem was the most common co-administered antibiotic 62.7% (n=131) followed by tigecycline 28.7% (n=60). Furthermore, Table 3.7 describes the number of patients and the various antibiotic combinations prescribed in addition to colistin therapy. In total, 31 unique combinations were prescribed for the 209 patients who received combination therapy. All co-administered antibiotic agents, with the exception of rifampicin, possess Gram-negative spectrum of activity. The addition of meropenem is currently mostly recommended for the treatment of the CPE's and tigecyline in combination for *Acinetobacter* spp. infections (Richards et al., 2015). The reasons for combination therapy with other antibiotics were not determined. It is assumed that the addition of these alternative agents would be an attempt by clinicians to add supplementary mechanisms to inhibit the growth of the MDR organisms cultured in the study patients. Rifampicin, a Gram- positive and tuberculosis antibiotic, has been used as a combination agent with colistin for its anti-biofilm activity, however, the effectiveness of this strategy has not been well established and is not routinely recommended (Durante-Mangoni et al., 2013).

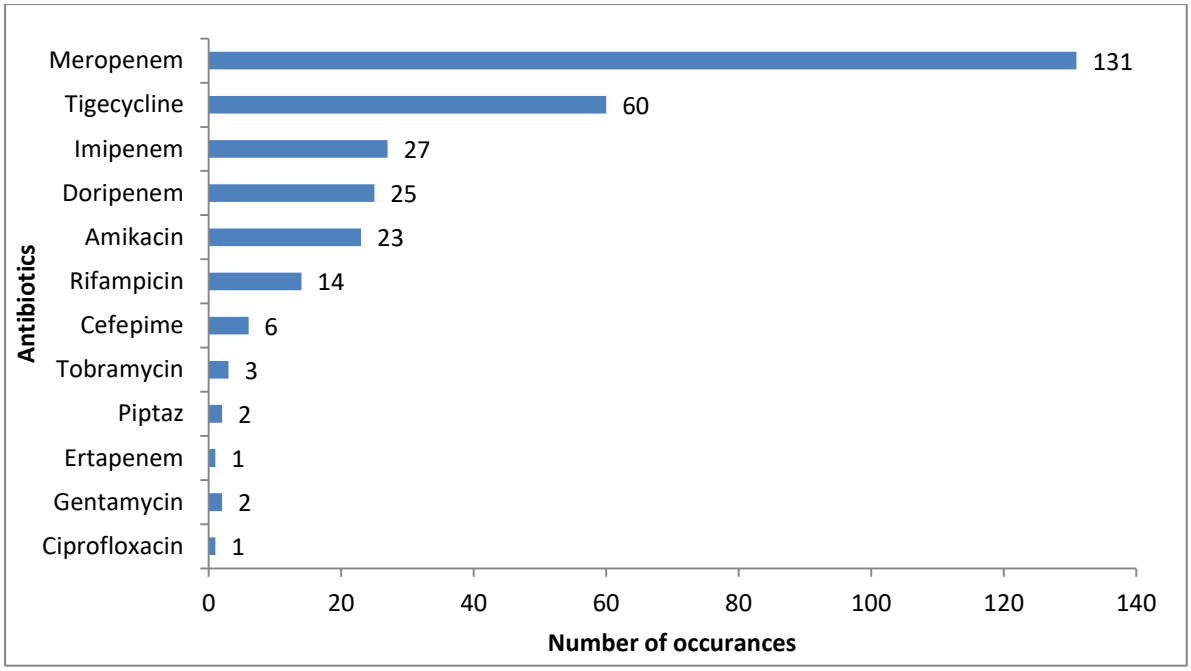

Figure 3.10. A breakdown of combination agents of choice prescribed with intravenous colistin therapy. Note values do not add up to 100% as some patients received more than one agent in combination.

Table 3.7. The number of patients and various antibiotic combinations prescribed in addition to colistin therapy (n=209).

Antibiotic	Number of Patients
One Additional Agent	
Cefepime	1
Doripenem	12
Gentamicin	1
Imipenem	17
Meropenem	84
Pipperacillin Tazobactam	1
Rifampicin	3
Tigecycline	12
Total	**131**
Two Additional Agents	
Amikacin/Doripenem	2
Cefepime/Ertapenem	1
Cefepime/Tigecycline	1
Imipenem/Amikacin	1
Imipenem/Rifampicin	1
Imipenem/Tigecycline	8
Meropenem/Amikacin	11

Antibiotic	Number of Patients
Meropenem/Cefepime	2
Meropenem/Doripenem	1
Meropenem/Rifampicin	1
Meropenem/Tigecycline	26
Rifampicin/Amikacin	3
Rifampicin/Doripenem	1
Rifampicin/Tigecycline	1
Rifampicin/Tobramycin	2
Tigecycline/Doripenem	7
Tobramycin/Doripenem	1
Total	**70**
Three Additional Agents	
Cefepime/Rifampicin/Amikacin	1
Meropenem/Amikacin/Ciprofloxacin	1
Meropenem/Rifampicin/Amikacin	1
Meropenem/Tigecycline/Amikacin	3
Meropenem/Tigecycline/Gentamicin	1
Tigecycline/Doripenem/Pipperacillin Tazobactam	1
Total	**8**

3.2.7 De-escalation practices

Of the patients who had cultures taken (n=210), in 74.3% (n=156) an organism was identified for which corresponding antibiotic sensitivity results were available to evaluate compliance to de-escalation practices. In 46.8% (73/156) of these patients, colistin was the only susceptible agent and de-escalation was therefore not possible. This demonstrates the extent of XDR infections in these settings and the reliance on colistin as the only viable treatment option for a substantial subset of patients. However, in 53.2% (n=83/156) of patients, where sensitivity to at least one other feasible antibiotic agent was demonstrated, de-escalation or tailoring of directed therapy to an appropriate alternative antibiotic only occurred in 69.9% (58/83) of cases. No particular pattern in de-escalation could be established as this is due to prescriber preference. The de-escalated antibiotic selected was either a continuation of one of the combined antibiotics as monotherapy in 31% (n=18) of cases or, a completely new antibiotic agent in 69% (n=40). Detailed clinical information of these patients is unknown and whilst there may have been valid reasons to continue colistin therapy in these cases, the results suggest another colistin process measure that could be targeted for improvement.

For those patients for which therapy was de-escalated, the predominant antibiotic following de-escalation was meropenem 22.7% (n=15), levofloxacin 18.2% (n=12) and tigecycline 15.2% (n=10) (Figure 3.11).

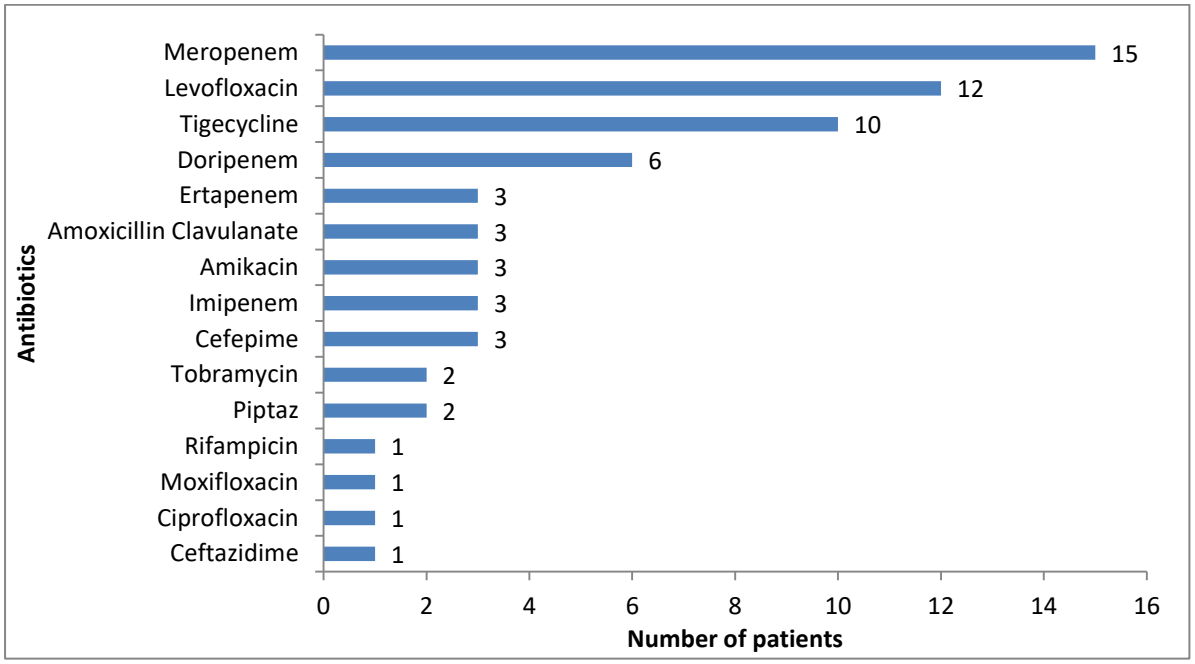

Figure 3.11. A breakdown of de-escalated antibiotic agents for the study patients

3.2.8 Duration of therapy

Duration of therapy was calculated as the sum of treatment days. A total of 6.1% (n=13) of study patients received a duration of colistin therapy for less than 72 hours. In order to avoid bias in the results, these patients will be excluded and thus the results pertaining to duration of therapy, effects on renal function and overall outcome of the remaining 199 patients will be discussed.

The median duration of colistin therapy was nine days (interquartile range (IQR) 6-16 days; range 3-63 days). Most patients, 57.8% (n=115), received a course of therapy ≤10 days, 13.6% (n=27) between 11-14 days and 28.6% (n=57) ≥ 15 days.

For those patients who received colistin for 15 days or more, the mean age was 49.7 years (range 19-83 years). The majority were male patients 64.9% (n=37) and were treated within the ICU 82.5% (n=47). At least one of the three major Gram-negative organisms were cultured

in 98.2% (n=56) of the patients. The average length of stay of these patients was 73.45 days (range: 16-227 days) and 36.8% (n=21) demised in hospital.

3.2.9 Analysis of renal function

Prior to the commencement of colistin, 2.0% (n=4) of patients were deemed to have kidney failure, 10.7% (n=21) severe kidney injury, 20.4% (n=40) moderate kidney injury and, 66.8% (n=131) normal kidney function, according to the kidney disease improving global outcomes (KDIGO) classification (KDIGO, 2013).

The effects on renal function for patients who received IV colistin therapy in this study were found to be insignificant and no changes in renal function measured through SCr or eGFR were noted. The change in SCr level, as well as, the change in eGFR of study patient's pre and post exposure to colistin is described in Figures 3.12 and 3.13. These study findings are similar to those described by Falagas et al. (2005) and Gibson et al. (2016) and could be as a result of the lower doses prescribed and shorter treatment durations found in this study.

Furthermore, although the range in SCr of the patients studied was wide (Figure 3.12), most often, a medication is considered to have an adverse effect on the kidney if SCr increases by 100% post exposure to the agent. In this study, no patient's SCr post colistin exposure increased by 100% and only in a very small cohort of 5.6% (n=12) of patient's, did SCr increase by more than 50% (range 54.3%-89.5%) following exposure to colistin. The wide range in SCr demonstrated by the 212 patients in the study could be attributed to the critically ill nature of the patient population for which variable kindey function is to be expected. The various categories of renal function according to the KDIGO classification of the patients studied prior commencement of colistin has already been described. Unfortunately, no additional variables such as the presence or absence of renal replacement therapy, other medications prescribed and clinical notes were available to elucidate the exact reasons for the wide range in SCr of patients studied.

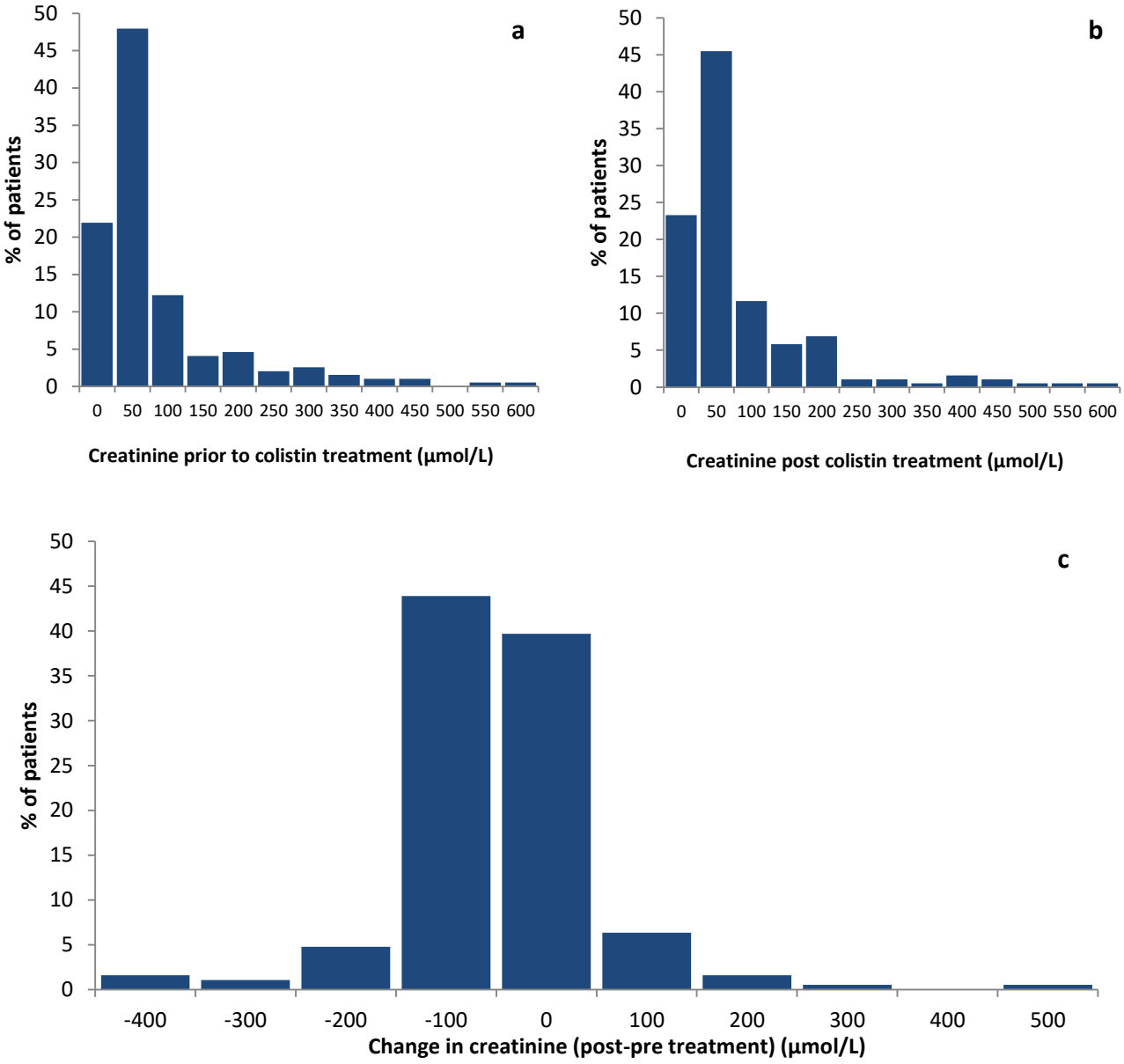

Figure 3.12. Evaluation of the effects of serum creatinine (SCr) pre and post study patient's exposure to colistin therapy. a) The median creatinine level before colistin therapy was 73 µmol/L (IQR 53-110 µmol/L; range 21-601 µmol/L). b) The median creatinine level after colistin therapy was 73 µmol/L (IQR 51-128 µmol/L; range 20-645 µmol/L). c) The mean change in creatinine level (post-pre treatment) was 0 µmol/L (sd 95; range -384 to 577 µmol/L). This mean change was not significantly different to zero (95% confidence interval for mean change: -14 to 13 µmol/L).

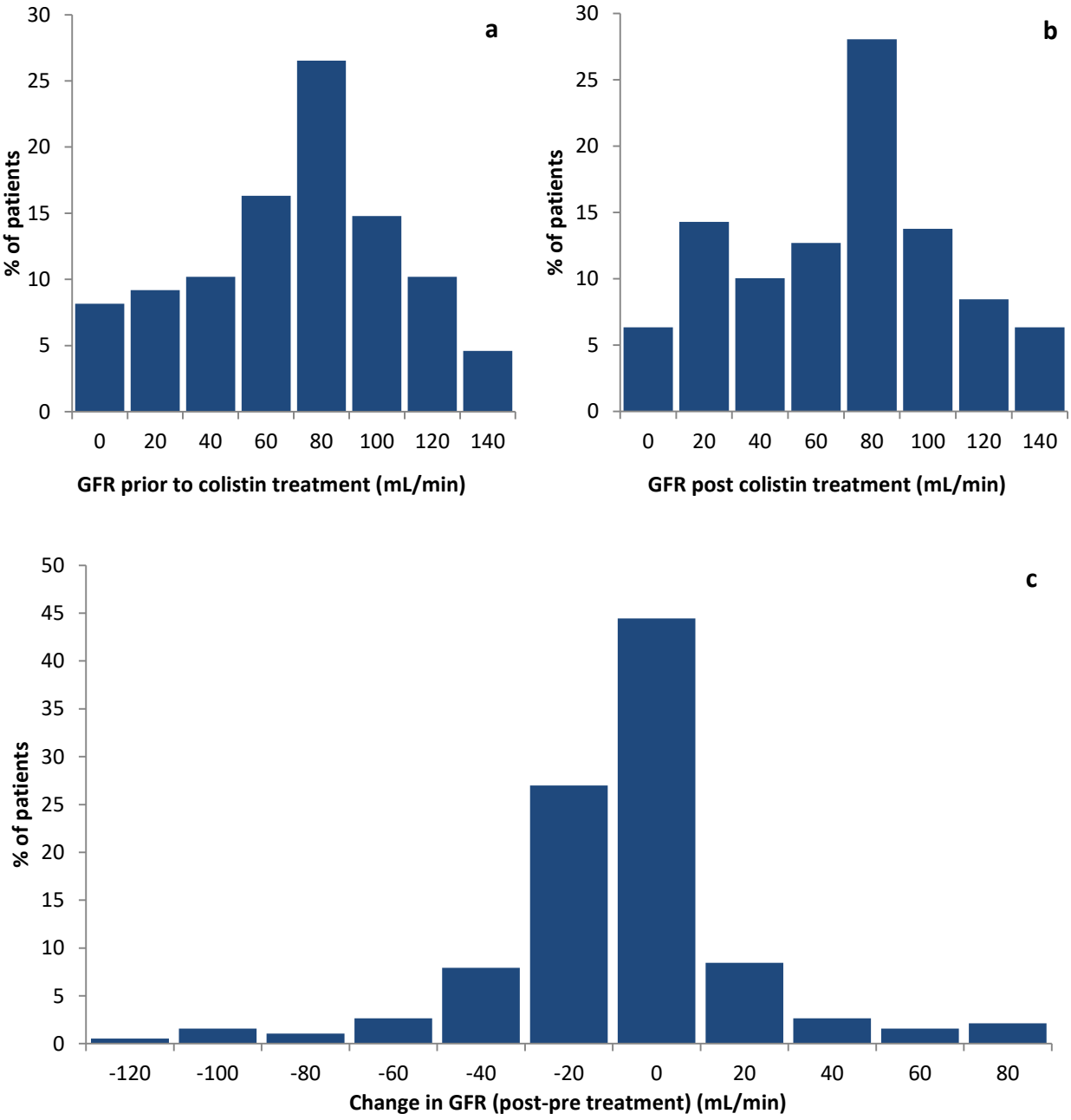

Figure 3.13. Evaluation of the effects of estimated Glomerular Filtration Rate (eGFR) pre and post study patients exposure to colistin therapy. a) The mean eGFR level before treatment was 79 mL/min (sd 37 mL/min; range 8-150 mL/min). b) The mean eGFR level after treatment was 79 mL/min (sd 38 mL/min; range 7-150 mL/min). c) The mean change in eGFR level (post-pre treatment) was 0 mL/min (sd 28 mL/min; range -112 to 89 mL/min). This mean change was not significantly different to zero (95% confidence interval for mean change: -4 to 4 mL/min).

3.2.10 Overall compliance to antibiotic stewardship process measures for study patients on intravenous colistin therapy

In order to establish appropriate stewardship practices for colistin in this study, a stewardship bundle comprising of eight process measures was designed to determine the compliance to essential stewardship related principles for colistin utilization. The compliance of the antibiotic stewardship related process measures of the patients on IV colistin in this study are summarised in Table 3.8. The lowest compliance rate from this evaluation pertains to maintenance doses (50.0%), 'hang time' (66.3%) and de-escalation practices (69.9%). Appropriate compliance to duration of colistin therapy could not be audited since evidence alluding to what the appropriate duration should be is lacking and as a result, in real world practice, this is based on the patients' clinical response to treatment. As such, the study patient's composite compliance to the proposed colistin stewardship bundle is 81.2% at best (using only seven process measures due to duration not able to be assessed and dose compliance according to the SASOCP guideline). As a result of this evaluation, important stewardship related targets were identified to enable future recommendations. In addition, ensuring compliance with recording weight and incorporation of South African guidelines for colistin dosing in hospital policies, may further improve utilization.

Table 3.8. A summary of the compliance rate of antibiotic stewardship process measures for patients prescribed intravenous colistin therapy

Process measures	Compliance rate % (*n*)
1. Obtaining an appropriate culture prior to the commencement of colistin therapy	99.1 (210)
2. Prescription of a loading dose	93.9 (199)
3. Prescription of an appropriate loading dose	90.4 (180)
4. Prescription of appropriate maintenance dosing including adjustment according to renal insufficiency	50.0 (74/148)[*] 42.3 (88/208)[†]
5. Compliance to antibiotic 'Hang time'	66.3 (116)
6. Prescription of colistin in combination with another Gram-negative susceptible antibiotic	98.6 (209)
7. De-escalation of colistin therapy	69.9 (58)

Process measures	Compliance rate % (*n*)
8. Median duration of therapy	9 days

*SASOCP guidelines † Visser Kift et al., 2014

3.2.11 Patient outcome measures

Infection with MDR pathogens and critical illness are factors that impact hospital LOS and overall outcome. For the patients studied (n=199), median ICU LOS was 31 days (IQR 15-52 days; range 0-152 days). The overall hospital admission median LOS for these patients was 46 days (IQR 25-83 days; range 3-227 days). The majority of patients, 70.4% (n=140), were discharged indicating a 29.6% (n=59) in-hospital mortality rate (Figure 3.14).

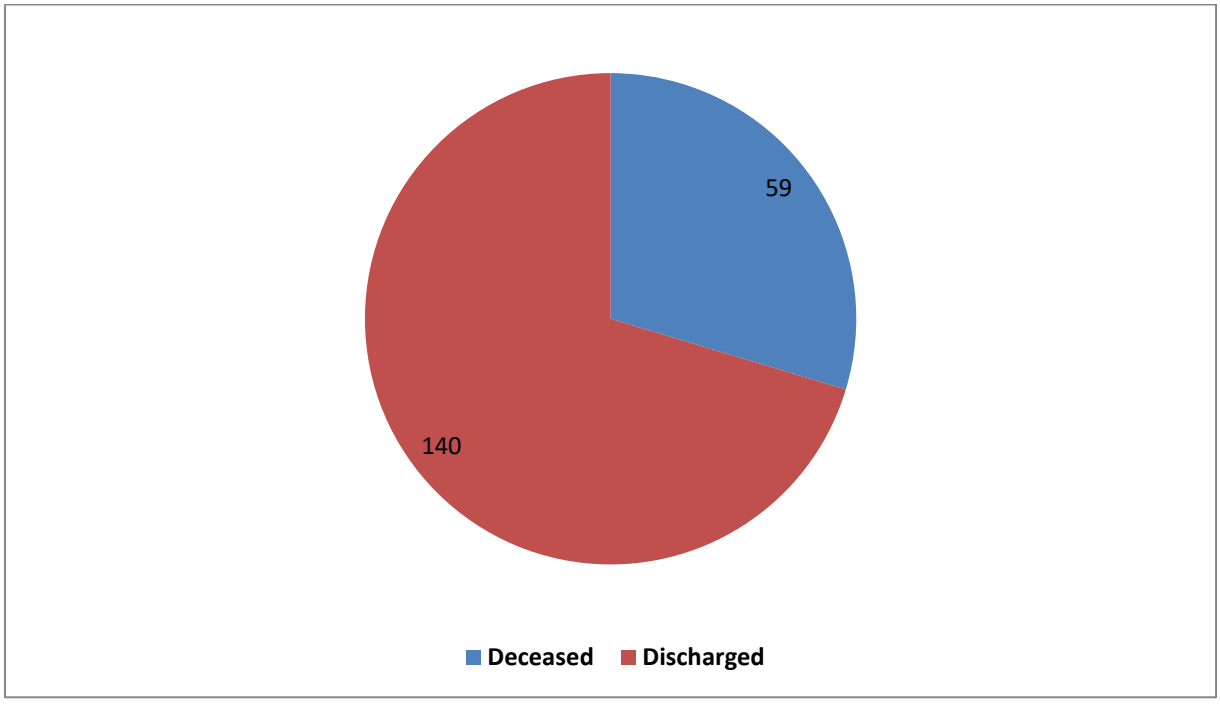

Figure 3.14. Overall in hospital patient outcome for patients on intravenous colistin (n=199)

3.3 Evaluation of the study associations of patients who received intravenous colistin

Further statistical analysis was conducted on the patients who received IV colistin therapy in order to establish any possible associations between colistin utilization and other variables such as renal function, dose, treatment duration, hospital location, specimen type and organisms. Any associations established could further inform suitable stewardship recommendations for the appropriate use of colistin in the future.

3.3.1. The association between colistin dose (loading and maintenance) and renal function (eGFR)

No significant associations were found between various categories of renal function and loading doses prescribed (Fisher's exact test; p= 0.13). However, although obvious and expected, a strong significant association was noted between different categories of renal function and colistin maintenance dose prescribed; those patients with eGFR categories of more than 60 mL/min received higher colistin maintenance doses (chi-square test; $p < 0.0001$; phi coefficient= 0.52) which is in line with the renal function based dosing strategy of colistin. Figure 3.15 graphically indicates the percentage of patients per various categories of colistin dose received according renal function classifications. It is evident that those patients in the severe kidney injury classification (GFR <30mmol/L) received lower colistin doses; 48% of patients in this category received a range of colistin dose between 1-2.5 MU. This is contrasted by the patients classified with normal renal function (GFR >60mmol/L) where 96.1% of patients in this category received a colistin dose of 3MU or higher.

3.3.2. The association of the presence of a blood stream infection on duration of treatment and overall outcome

The analysis could not establish significance in the median duration of therapy (Wilcoxon rank sum test; p= 0.39) nor overall outcome (Fisher's exact test; p= 0.86) between patients who did or did not have a blood stream infection.

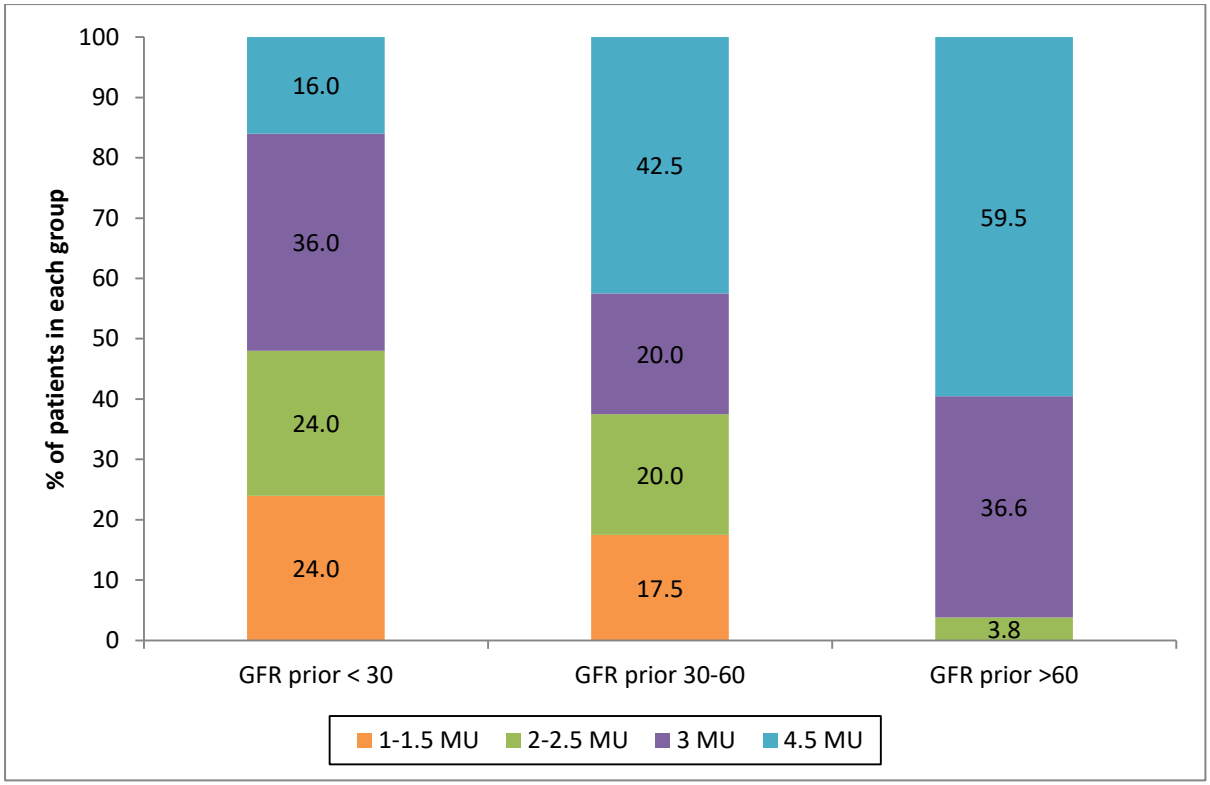

Figure 3.15. The various colistin doses prescribed according to renal function classifications for intravenous (IV) study patients.

3.3.3. The associations between the presence or absence of the three prevalent Gram-negative organisms and duration of therapy

The median duration of colistin treatment for patients with *P. aeruginosa* (12 days; IQR 8-23 days) was significantly longer than for those patients who did not isolate this organism (9 days; IQR 6-15 days)(Wilcoxon rank sum test; p=0.044; r=0.14; small effect size). The reasons for this are unknown but in general perceptions are such that *P. aeruginosa* infections require longer durations of treatment. No significant differences in the median duration of colistin treatment were found between those patients who did or did not have *K. pneumoniae* (Wilcoxon rank sum test; p= 0.68) nor those patients who did or did not have *A. baumannii* (Wilcoxon rank sum test; p= 0.54).

3.3.4. Comparison between hospitals one and two (Johannesburg) to hospitals three and four (Pretoria)

Colistin loading doses (chi-square test; p<0.0001; Cramer's V=0.46) and maintenance doses (p< 0.0001) were found to be moderately significantly higher in the Pretoria set of hospitals (Figure 3.16 and 3.17) which indicates a prescriber preference of higher dose utilization in hospitals three and four. In patients who survived, the median duration of colistin treatment in the Johannesburg hospitals (11 days; IQR 7-16 days) was significantly longer (Wilcoxon rank sum test; p= 0.44; r=0.17; small effect size) than for those in the Pretoria hospitals (8 days; IQR 6-13 days). In addition, the proportion of patients with *A. baumannii* infections was significantly (moderate) higher (Fischer's exact test; p<0.0001; phi coefficient= 0.32) in the Johannesburg hospitals (20.8%) compared to the Pretoria hospitals (1.7%) and similarly for *P. aeruginosa,* although a weak significance (Fisher's exact test; P=0.0010; phi coefficient=0.24), the proportion of patients was higher in the Johannesburg hospitals (32.5%) compared to the Pretoria hospitals (12.5%). No significant associations between cities and *K. pneumoniae* prevalence could be established (Fisher's exact test; p=0.46).

3.3.5. Comparison of patients in general wards and Intensive Care Units (ICU) with regards to colistin loading and maintenance doses and duration of therapy

Colistin loading (chi-square test; p= 0.048; Cramer's V=0.18) and maintenance doses (Fisher's exact test; p= 0.028; phi coefficient=0.21) were significantly (weak association) higher in patients in ICU's versus general wards (Figure 3.18 and 3.19). This may be due to the critically ill nature of patients in ICU's and the need to optimise dosing for these patients to enhance treatment success. No significance could be established in the median duration of treatment for patients in ICU's versus general wards (Wilcoxon rank sum test; p=0.41).

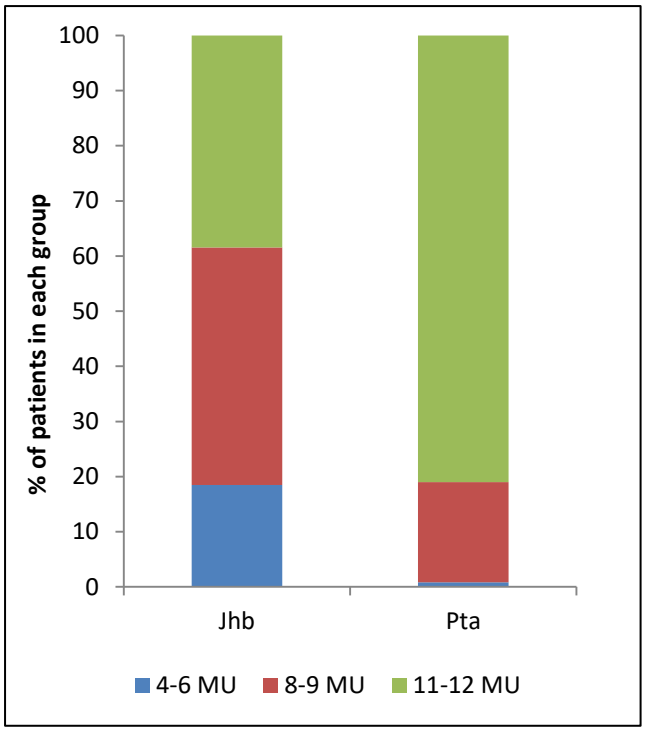

Figure 3.16. Comparison of colistin loading doses prescribed between the hospitals located in Johannesburg (Jhb) and Pretoria (Pta)

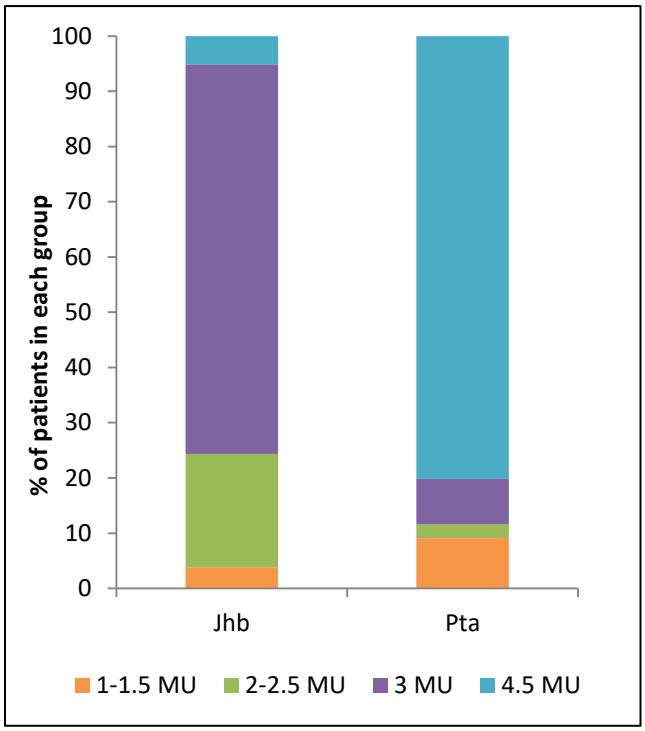

Figure 3.17. Comparison of colistin maintenance doses prescribed between the hospitals located in Johannesburg (Jhb) and Pretoria (Pta)

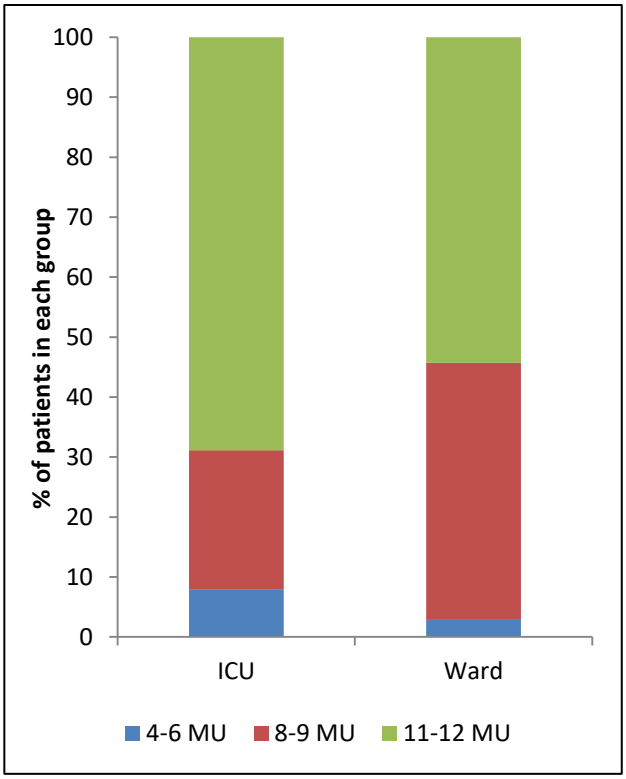

Figure 3.18. Comparison of colistin loading doses prescribed between general wards and Intensive Care Units

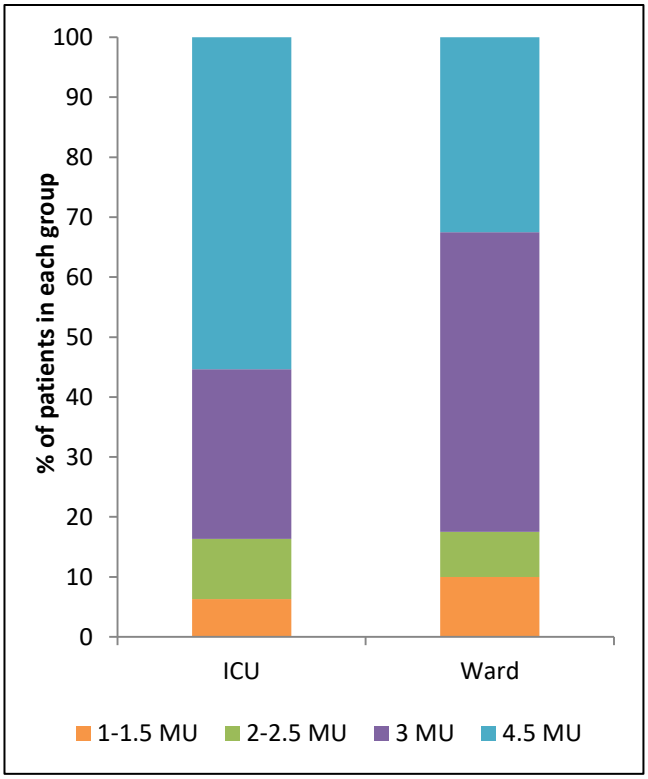

Figure 3.19. Comparison of colistin maintenance doses prescribed between general wards and Intensive Care Units

3.3.6 The association between duration of therapy and loading and maintenance dose

Importantly, the median duration of colistin treatment for patients who received lower loading doses of 4-6 MU (20 days) was significantly longer than those patients who received higher loading doses of 11-12 MU (8 days) (Kruskal-Wallis test; p=0.040) (Figure 3.20). This concept of higher doses resulting in shorter durations of treatment was also found with higher colistin maintenance doses prescribed, as the median duration of treatment of those patients who received a 3 MU eight hourly maintenance dose (12 days) was significantly longer than that of patients who received a 4.5 MU twelve hourly maintenance dose (8 days) (Kruskal-Wallis test; p=0.027) (Figure 3.21). It should be noted that only 17 patients received a dose of 1-1.5 MU compared to 112 patients who received a 4.5 MU dose and, as such, the implied similar duration of therapy for these two categories of patients should be interpreted with caution as this does not reprersent a valid sample to suggest that lower doses of colistin render shorter durations of treatment (Figure 3.21). Since duration in this study was shorter for those patients who received higher doses this may infer that clinical stability and therapeutic efficacy might have been achieved sooner for these patients (which is in line with supporting evidence that attributes this to the faster attainment of steady state and optimal colistin drug concentrations) and thus allowing the ability to stop therapy sooner.

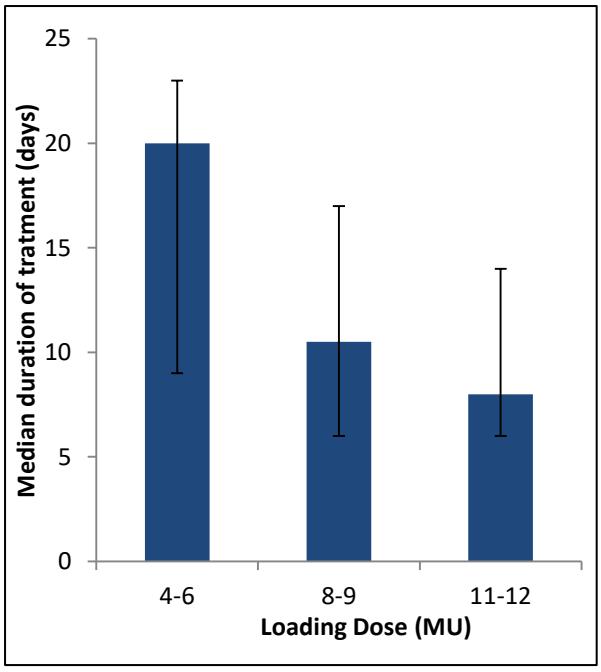

Figure 3.20. Associations between colistin loading dose and median duration of treatment. The error bars denote the interquartile range.

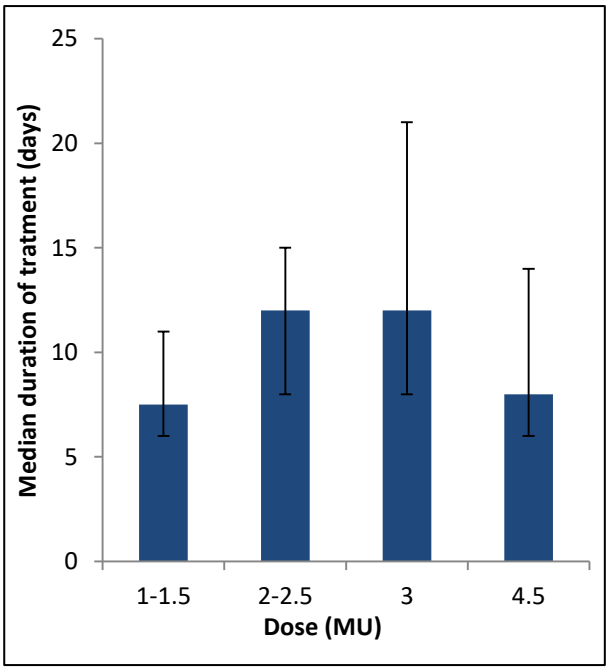

Figure 3.21. Associations between colistin maintenance dose and median duration of treatment. The error bars denote the interquartile range.

3.3.7. The association between overall patient outcomes and the presence or absence of each of the three qualifying organisms, colistin dose or duration of therapy.

No significant associations could be found between patient outcome and a particular organism (Fisher's exact test; *K. pneumoniae* p=0. 87; *P. aeruginosa* p= 0.44; *A. baumannii* > 0.99). Furthermore, no significance could be established between patient outcome and colistin loading dose received (chi-square test; p= 0.83) and duration of treatment (Wilcoxon rank sum test; p= 0.20). However, probably the most crucial finding of this study, despite lack of severity of illness data, is that a significant moderate association between overall patient outcome and IV colistin maintenance dose prescribed was found. Deceased patients were associated with lower maintenance doses per interval compared to patients that survived (Fisher's exact test; p=0.0037; phi coefficient= 0.26). This is evidenced by the proportion of patients per category described in Figure 3.22 where 31.6% of patients who demised received a maintenance dose of 1-2.5 MU versus 11.6% in the group that were discharged.

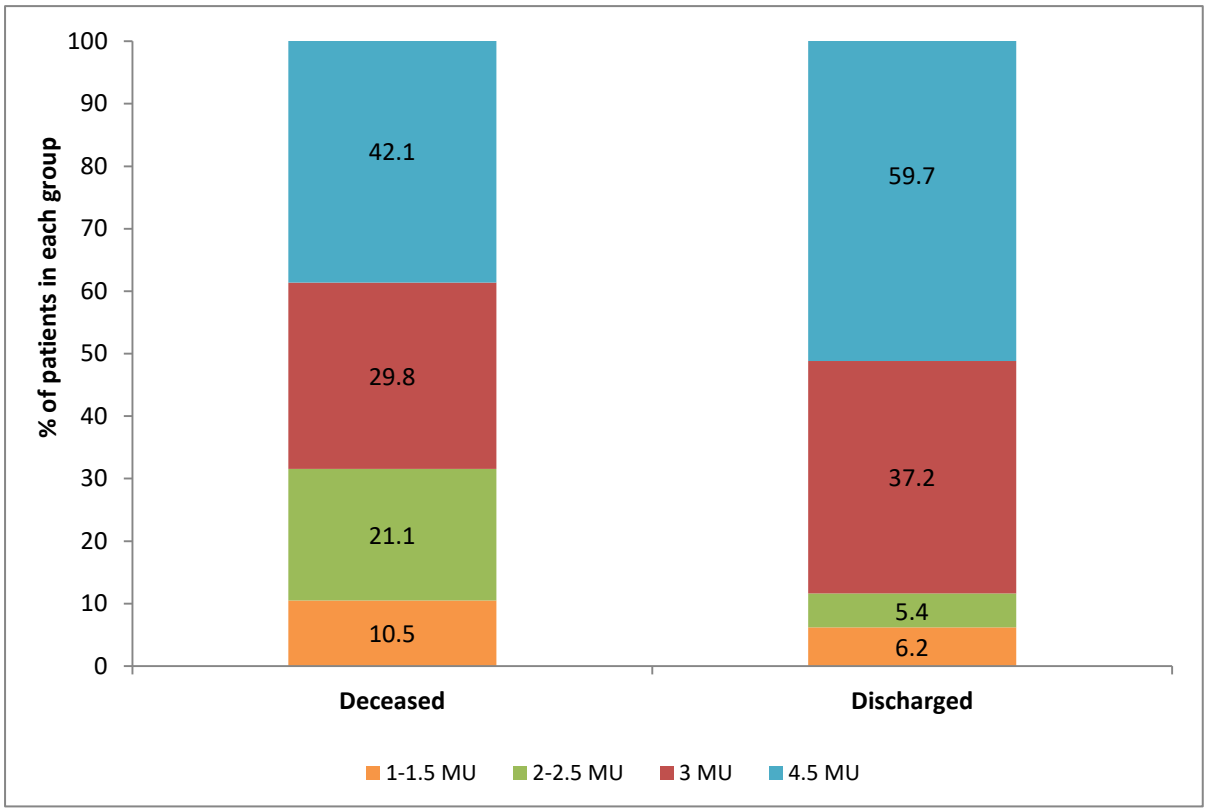

Figure 3.22. Associations between colistin maintenance doses prescribed and patient outcome

3.4. Discussion

This results of this study evaluated the current utilisation of IV colistin across multiple South African hospitals involving a large sample of patients. Through this process numerous opportunities for improved stewardship were identified.

Recent recommendations in response to the emergence and spread of plasmid-mediated colistin resistance include: preserving colistin use for definitive treatment based on susceptibility testing, use of PK/PD indicators to ensure appropriate dosing, and use of empirical therapy in selected cases only (Al-Tawfiq et al., 2017). As is evidenced through the results presented, MDR of the three major Gram-negative organisms which compelled the use of colistin in this study was widespread. Compliance to obtaining a culture in this study was good (99.1%), however, the use of colistin in the 33.5% of patients studied that was deemed to be empiric is a concern and could be as a result of the severely immune compromised, neutropenic state of the majority of patients included in the study, where colistin may have been prescribed following poor clinical response to other broad-spectrum antibiotic agents including the carbapenem class. Other factors could comprise: continuous spikes in temperatures; numerous previous admissions with broad spectrum antibiotic exposure and; the relatively high suspicion and risk of MDR infections due to the prolonged hospital length of stay in this patient population.

There is wide-spread global consensus that the empiric use of colistin outside of “clearly defined circumstances or for certain at risk categories of patients” is strongly discouraged in order to preserve the efficacy of the antibiotic for the future generations (Al-Tawfiq et al., 2017). The “clearly defined circumstances” and “at risk categories of patients” referred to should be outlined and defined, as it is difficult to say currently that the use of colistin in the 71 patients deemed to have received empiric therapy in this study is justifiable or not, as a consequence of the critically ill nature of this study patient population.

Use of colistin in general wards is distressing due to the high risk and toxic nature (nephrotoxic and neurotoxic) of the antibiotic, which requires appropriate monitoring and supervision when administered for signs and symptoms including: changes in renal function, muscle weakness, peripheral neuropathy and visual disturbances to name a few.

Repeat courses of colistin therapy pose a concern to patient outcome as it may indicate the inability of the antibiotic to resolve the initial MDR infection (possibly due to inappropriate dosing) or, it highlights the risks of probable acquisition of multiple MDR infections for patients with prolonged hospital admissions, compelling the need for colistin therapy. The exact reasons for repeat courses of colistin could not be definitively established, however, due to the absence of clinical notes and other important required data in the records reviewed for this study (refer to study limitations in Chapter 6, Section 6.2). In addition, often these courses were administered prior to the study data collection period.

According to colistin dosing guidelines a colistin loading dose of 9-12 MU should be administered to patients regardless of renal function to rapidly achieve the necessary MIC concentration of 2 mg/L and prevent regrowth of more resistant pathogens (Visser-Kift et al., 2014; Richards et al., 2015; Labuschagne et al., 2016; Nation et al., 2017). Furthermore, there is evidence to show that a maintenance dose of 4.5 MU administered 12 hourly rather than 3 MU administered 8 hourly resulted in more favourable patient outcomes at day 7 (Dalfino et al., 2012), however, both dosing strategies equating to a total dose of 9 MU per day are acceptable for patients with normal renal function (Richards et al., 2015; Labuschagne et al., 2016; Nation et al., 2017). Dose adjustments are also required for patients with compromised renal function based on available evidence due to the nephrotoxic nature of the drug (Falagas et al., 2009; Dalfino et al., 2012; Ortwine et al., 2014). The recommended frequency of colistin administration is either at eight or twelve- hourly intervals depending on the maintenance dose prescribed (Richards et al., 2015; Labuschagne et al., 2016). Longer dosing intervals, such as 24- hourly schedules, have demonstrated greater emergence of bacterial resistance to colistin compared to shorter intervals (Bergen et al., 2008).

Contrary to these guideline recommendations, the findings of this study suggests that both loading and maintenance dosing of colistin is variable and inconsistent, with adherence to available local dosing guidelines at best 50.0%. This reveals the extent of uncertainty associated with colistin utilisation in SA hospitals and the very urgent need for education so that our last-resort Gram-negative antibiotic can be preserved for as long as possible.

The poor compliance to appropriate colistin dosing demonstrated in this study is concerning. Since evidence has shown the importance of dosing colistin correctly to ensure favourable

patient outcomes, it is distressing to observe the huge variation in doses prescribed and the non-compliance to appropriate dose based on various categories of renal function and the incongruity in the prescription of a correct loading dose (Tables 3.5 and 3.7). The administration of a colistin loading dose is widely considered to be best practice, as it facilitates the rapid achievement of optimal bactericidal concentrations (Garoznick et al., 2011; Dalfino et al., 2012). Although compliance with the recommendation for a loading dose was high (93.9%), the actual loading doses ranged from 4 MU to 12 MU. This too demonstrates a lack of understanding regarding the need for appropriate loading doses.

There could be multiple factors that contribute to the poor dose compliance demonstrated by this study which could be established if physicians were interviewed to qualitatively assess and understand the exact reasons why to inform some behaviour change techniques in the future, however, some additional reasons are listed below;

a) The historic ambiguity of the appropriate colistin dose could contribute to the lack of confidence in accurate prescribing.
b) Prescribers may genuinely not know what dose to prescribe due to the complexity of the PK and PD of the drug in a critically ill patient.
c) In general, compliance to recommended guidelines is often poor with much evidence indicating the need for behaviour change in prescribers particularly those of older age (Tell et al., 2015; Levy et al., 2015). It is also most likely that most clinicians are unaware of the available South African colistin dosing guidelines.
d) Colistin is not commonly prescribed and only recently is re-emerging in South African hospitals as salvage therapy for critically ill MDR infections. As such, the prescription of colistin is not routine and may not be 'second nature' for prescribers as the prescription of other antibiotics may be.
e) The prescription of colistin should be done according to renal function and there is no standardized dosing schedule as for most other antibiotics. This may further contribute to the lack of clarity regarding selection of the correct dose per category of renal efficacy for prescribers.

The non-compliance demonstrated in this review indicates that although the tools and guidelines have been available to use in order to optimize colistin dosing, these have largely

been ignored or have not been widely disseminated. Regardless of the possible reasons stated above, since the pipeline for effective antibiotics is diminishing for serious MDR Gram-negative infections and colistin is currently the final option, it is critical that each time it is prescribed, it is done appropriately to achieve rapid therapeutic serum concentrations in order to maintain the efficacy of the antibiotic, prevent the emergence of resistance and offer the patient his/her best chance of survival.

In patients with severe sepsis and septic shock, the prompt administration of the right antibiotic can be lifesaving (Kumar et al., 2006; Rhodes et al., 2017). 'Hang time' is a colloquial term that describes the time lapsed from when an antibiotic is ordered/ prescribed to the time of actual IV administration which aims to be within one hour (Messina et al., 2015). The concept most commonly relates to the first dose of the first administered antibiotic as this is the highest impact opportunity for patient survival. Since according to best practices, colistin should only be prescribed following microbiological confirmation of its indication and is often escalation therapy (unlikely to be the first administred antibiotic), delays in colistin prescription and administration have been reported of up to 96 hours (Tigen et al., 2013).

Delays in the prompt administration of colistin could be attributed to the Section 21 approval process that is required prior to the procurement of colistin (Visser-Kift et al., 2014) amongst other factors. This process can delay the timely administration of the antibiotic since application to the MCC and authorization thereof is needed – a process which can take one to three days in itself. This could negatively impact patient outcome if stock of colistin takes two to five days post-approval to be delivered depending on the hospital location. The section 21 approval and procurement process of colistin was not included in the scope of this study and so metrics on the date of MCC application completion, date of MCC submission, date of approval, date of stock ordering and, date of stock delivery, were not evaluated. Additional factors that could have contributed to a delay in 'hang time' of the patients studied include (Messina et al., 2015):

- The use of paper based prescription charts as opposed to electronic prescription entries;
- Delays in the prescription evaluation, dispensing and processing time within pharmacies;
- Delays in delivery of medication from the pharmacy to the wards;

- Delays in the reconstitution of the medication by nursing staff as often these wait for standardised medication administration rounds;
- Often if 'stat' is not written on the prescription chart then the administration of the antibiotic is not considered urgent.

Combination therapy was prescribed to the majority of patients (98.6%) in this study. This practice, including duplicate and sometimes triplicate therapy, is recommended by local guidelines for the treatment of CPE, suggesting that combinations may improve efficacy and minimise the risk of resistant organism selection. Studies that have supported combination therapy for CPE have relatively low sample sizes, and concerns remain regarding the increased environmental burden of multiple antibiotic exposure, which may actually increase host colonisation with resistant organisms and increase the risk of *Clostridium difficile* infection (Paul et al., 2014). The spectrum of antibiotics listed as combination agents in Figure 3.12 is interesting but since there is no standard protocol of the appropriate agent of choice, the top five co-administered antibiotics as demonstrated by this study all have evidence for efficacy in combination with colistin and could be deemed appropriate.

For 69.9% (*n*=58) of the eligible patients, therapy was de-escalated to a narrower-spectrum antibiotic following the availability of sensitivity results. Although this is a somewhat low figure, it is in line with other studies indicating that de-escalation is not always possible for many reasons, including the limited number of effective antibiotics available to treat MDR infections, the limited understanding of how to de-escalate, and the fact that the practice has still not been widely accepted in critically ill patients (Garnacho-Montero et al., 2015).

Evidence pertaining to the appropriate duration of colistin therapy is lacking since treatment is often definitive and continues until resolution of the infection or clinical response. Table 3.9 compares the duration of therapy of this study to other international publications. As depicted, this study showed the shortest median duration of therapy when compared to others. The reasons for this are unknown but could possibly be due to higher doses used in some patients resulting in the faster attainment of therapeutic concentrations and perhaps quicker microbiological and or clinical cure (Dalfino et al., 2012).

Table 3.9. A comparison of the median duration of colistin therapy according to available published evidence

Number of patients	Median duration of colistin therapy (days)	Reference
24	13.5	Markou et al., 2003
12	14.7	Sabuda et al., 2008
258	17.9	Falagas et al., 2009
28	11	Dalfino et al., 2012
109	10	Tanita et al., 2013
127	10.4	Gibson et al., 2016
199	**9**	**This study**

Colistin-related nephrotoxicity remains an important concern and has been found to be influenced by elevated plasma drug concentrations (>2.5 mg/L) and longer duration of therapy (Hartzell et al., 2009). Similar insignificant effects as in this analysis of renal function have been demonstrated (Dalfino et al., 2012). Another study found that up to 43% of patients were at risk of or had acute kidney injury or renal failure according to the Risk, Injury, Failure, Loss and End-stage kidney disease (RIFLE) criteria after IV colistin therapy; however, this toxicity was reversed following discontinuation of treatment (Pogue et al., 2011). Recent results from the multicentre PK colistin study demonstrated that there was huge interpatient variability in the clearance of colistin (even at similar creatinine clearances), which is probably due to differences between individuals in conversion rates of the inactive prodrug to its active form (Nation et al., 2017). This adds to the complexity of providing optimal dosing, given the very narrow therapeutic window of the drug. Due to the critically ill nature of the patients in this study, many of whom required colistin as the only viable option for treatment of their MDR infections, the risk benefit ratio of renal toxicity versus prospect of survival was likely applied in an attempt to ensure better patient outcome.

The in-hospital mortality rate of the patients studied was 29.6%. It is difficult to compare the patient outcomes of this study to other publications as the definition of these outcomes, inclusion criteria and doses used vary between studies. In addition, a limitation of this study relates to lack of multivariate analysis of significant risk factors for mortality such as severity illness scores, percentage of patients receiving high risk indwelling therapies such as mechanical ventilation and renal replacement therapy, as well as, other co-morbidities.

The study patient's composite compliance to the proposed colistin stewardship bundle is at best 81.2% with the lowest compliance indicators relating to maintenance dose, de-escalation practices and 'hang time'.The findings of the study support the call for optimal colistin dosing to improve patient care and outcome and reiterates the need to ensure sufficient colistin dosing for critically ill patients. Since the results described prove that patients' who received higher colistin loading and maintenance doses had a shorter duration of therapy and more favourable overall in-hospital outcomes (Figures 3.23-3.25). This could be due to the concentration dependent nature of the drug and is in keeping with the concept that the optimal administration of antibiotics according to PK principles and rapid achievement of therapeutic concentrations would result in improved clinical cure (Dalfino et al., 2012; Vicari et al., 2013).

3.5. Summary and Conclusions

- The utilization of colistin spans both ICU and general wards in hospital settings.
- There is poor compliance to local guideline-based recommendations of colistin dosing prescribed in the four South African hospitals studied.
- Poor compliance relates to both loading and maintenance doses.
- Compliance to other antibiotic stewardship process measures such as, taking a culture prior to therapy, prescription of a loading dose and, prescription of colistin in combination with another Gram-negative antibiotic was good.
- In contrast, compliance to antibiotic 'hang time' and de-escalation once susceptibility results were available, is poor.

- Those patients with *P. aeruginosa* infection were found to be treated with colistin for longer durations compared to infections of other organisms.
- Effects on renal function of colistin therapy on the study patients were insignificant.
- Colistin loading and maintenance doses were found to be higher in the Pretoria Group of hospitals compared to the Johannesburg hospitals.
- Those patients who received higher colistin doses were found to also receive a shorter duration of treatment.
- Those patients who received higher colistin doses appear to have had better overall in-hospital outcomes.

CHAPTER FOUR

EVALUATION OF AEROSOLISED COLISTIN UTILIZATION ACROSS FOUR PRIVATE SECTOR HOSPITALS IN SOUTH AFRICA.

4.1. Introduction

Of the four hospitals studied, hospital one was the only institution which used inhaled colistin for the treatment of LRTI's. Possible reasons for this could be due to hospital one being a centre of excellence for CF patients (for which inhaled colistin therapy is common practice) and the prescription of inhaled colistin therapy is similarly done for other LRTI's. Also, the doctors that treat CF patients at this hospital may be the same as those attending to patients with LRTI's in ICU's and so similar drug delivery methodologies may be adopted across patients. Whereas, the other hospitals do not routinely treat CF patients and the concept of colistin inhalation therapy may not be common knowledge. As such, the lack of clinician and nursing experience may prohibit inhaled delivery in those settings. This chapter will review the results of 25 patients who received inhaled colistin therapy during the study period.

4.2. Results

4.2.1. General patient demographics

A summary of the demographics of the 25 patients who received inhaled colistin therapy for both CF and other nosocomial pulmonary infections is described in Table 4.1. The mean age and gender of patients between the two groups are the predominant differentiating factors; those with CF being considerably younger and predominantly females than the patients with nosocomial LRTI's.

Table 4.1. Demographics and characteristics of the patients studied receiving aerosolised colistin (n=25)

Demographic	% (*n*)
Number of patients per hospital	
Hospital 1:	100.0 (25)
Hospital 2:	0 (0)
Hospital 3:	0 (0)
Hospital 4:	0 (0)
Number of patients according to treatment diagnosis	
Cystic fibrosis patients	52.0 (13)
Other LRTI's (including VAP)	48.0 (12)
Distribution of patients according to hospital location	
General wards	64.0 (16)
Intensive Care Units (ICU)	36.0 (9)
Age	
Cystic fibrosis patients mean **age**	31.8 years
Other LRTI's patients mean age	64.9 years
Gender cystic fibrosis patients	
Male	23.1 (3)
Female	76.92 (10)
Gender other LRTI patients	
Male	75.0 (9)
Female	25.0 (3)
Course of aerosolised colistin therapy	
First Course	72.0 (18)
Repeat Course	28.0 (7)
Indication for aerosolised colistin therapy	
Directed therapy	88.0 (22)
Salvage therapy	12 (3)

A breakdown of the number of patients who received aerosolised colistin therapy according to various categories of admitting diagnoses is summarised in Figure 4.1.

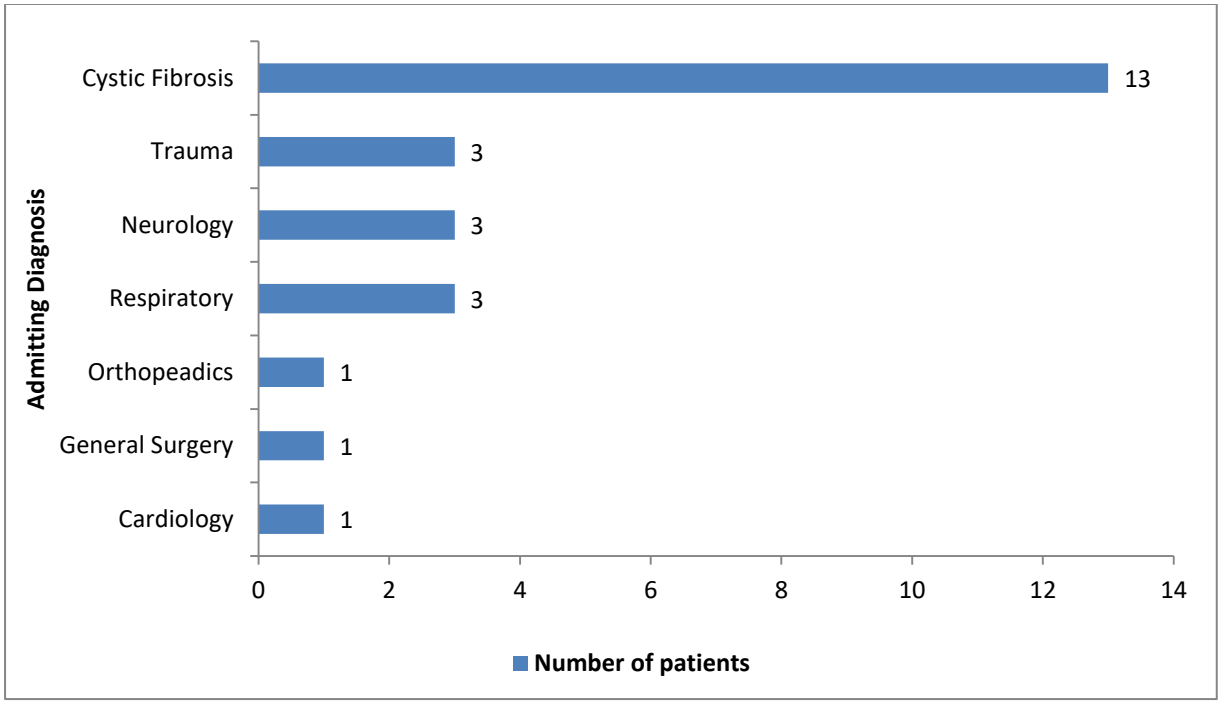

Figure 4.1. A breakdown of the number of patients per category of admitting diagnoses for patients included in the study on aerosolised colistin (n=25)

4.2.2. Microbial analysis for cystic fibrosis patients

For the patients who were admitted for acute exacerbations of CF (n=13), 100% had cultures taken of which all grew *P. aeruginosa*. This is not surprising as previously described- this organism is the most common opportunistic pathogen detected in patients with CF particularly those of advanced age. A breakdown of the specimen type in which *P. aeruginosa* was identified is shown in Figure 4.2, indicating sputum as the most common sample used for organism detection in these patients.

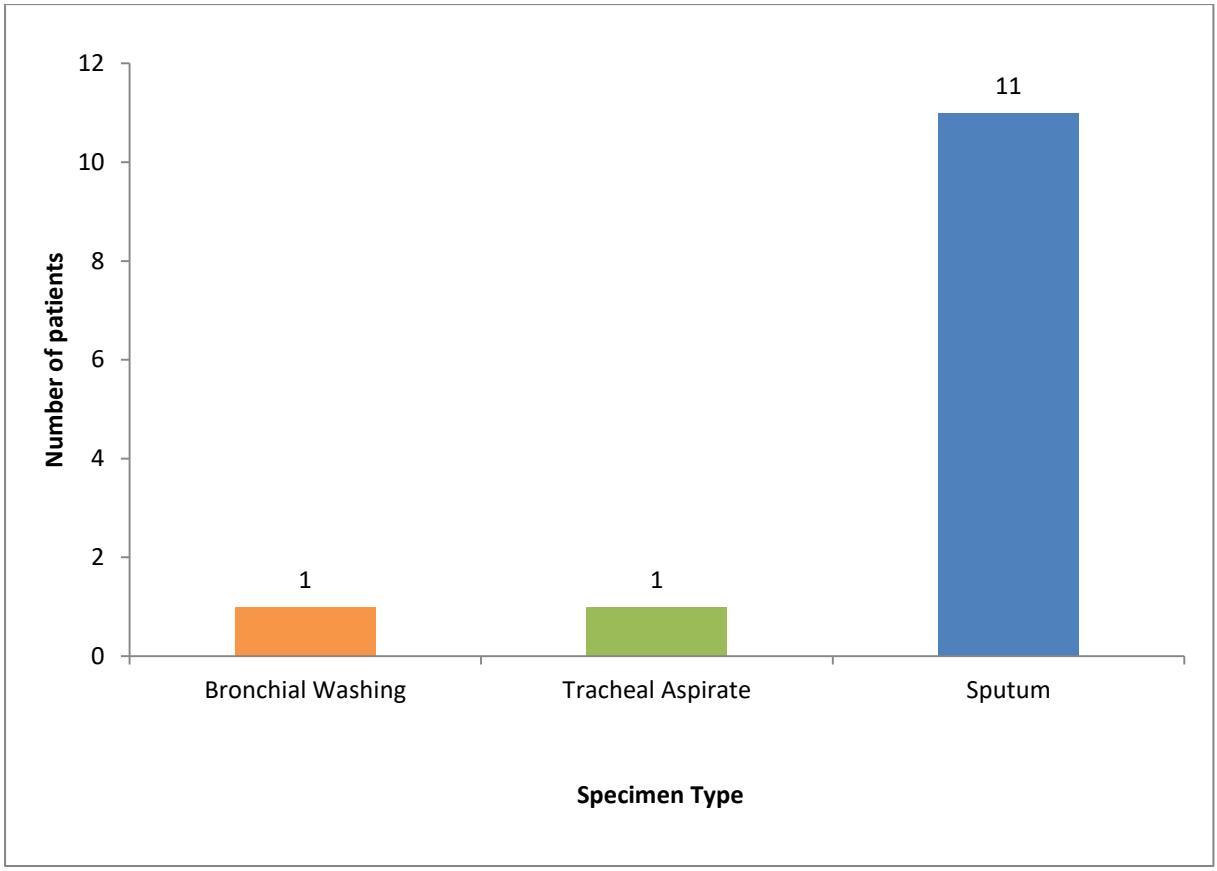

Figure 4.2. A breakdown of the number of patients per category of specimen type tested for organism growth for patients included in the study with cystic fibrosis (n=13)

4.2.3. Evaluation of *Pseudomonas aeruginosa* sensitivity profile of cystic fibrosis patients

The cumulative sensitivity profile of the thirteen *P. aeruginosa* isolates identified from the patients with CF in this study is summarised in Figure 4.3. Efficacy of the carbapenems was poor with only a third of isolates showing sensitivity to any of the carbapenems active against pseudomonal species. Due to the poor sensitivity of the organism demonstrated, colistin was the only viable option available for treatment in these patients.

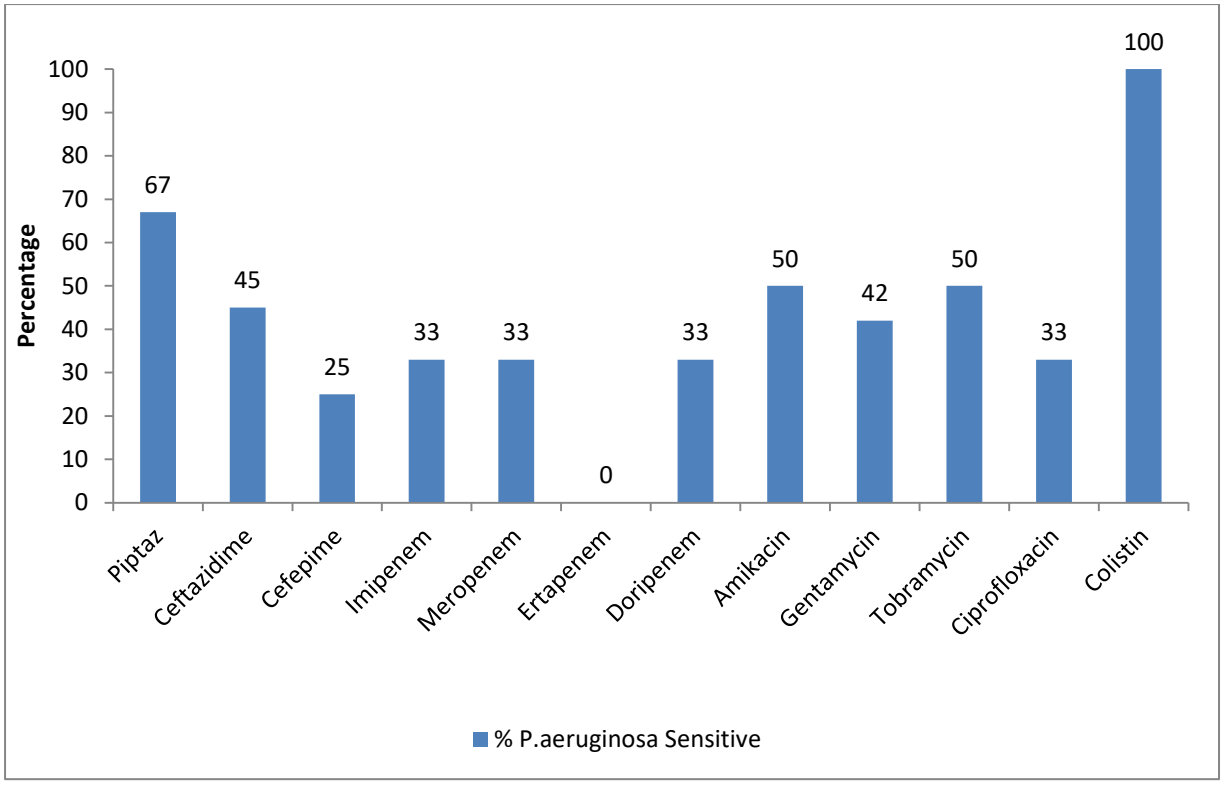

Figure 4.3. A cumulative antibiotic sensitivity profile of the thirteen *Pseudomonas aeruginosa* isolates detected in patients with cystic fibrosis in this study

4.2.4. Dose and duration of therapy for cystic fibrosis patients

The majority of CF patients (92.3%) received the appropriate dosing regimen of 2 MU inhaled 12 hourly (as recommended by available South African guidelines) whilst only one patient (7.7%) received an inhaled dose of 1 MU 24 hourly. This reflects overall good compliance to best practice inhaled colistin dosing recommendations for CF patients. The appropriate duration of treatment for acute pulmonary exacerbations has not been definitely established, however, recommendations of 14 days but not exceeding 21 days exist (Elborn., 2016; Stefani et al., 2017). The median duration of colistin treatment of CF patients studied was 10 days (range 4-21 days) indicating a shorter than recommended course of therapy. Furthermore, all treatment was discontinued in 84.6% (n=11) patients and for 15.4% (n=2) of patients, IV cefepime was continued following discontinuation of colistin inhalation therapy.

4.2.5. Co-administered antibiotic therapy for cystic fibrosis patients

All study patients with CF who received inhaled colistin therapy also received concomitant IV therapy with other antibiotics. A summary of additional IV antibiotics prescribed is depicted in Figure 4.4. For 23.1% (n= 3) of patients two additional antibiotics were prescribed and 15.4% (n= 2) of patients received three additional antibiotics. Interestingly ceftazidime was the most commonly prescribed concurrent IV antibiotic possibly due to its quorum sensing ability and moderate efficacy against mucoid *P. aeruginosa* strains in chronic infections.

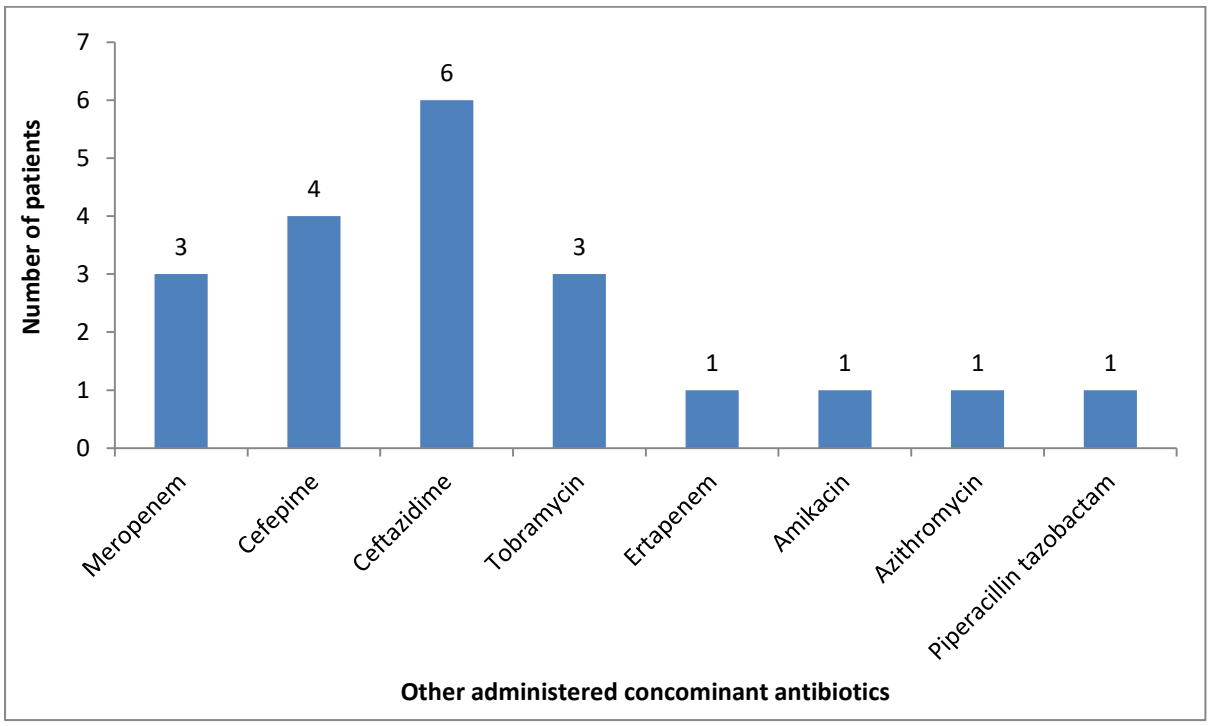

Figure 4.4. Additional antibiotic agents chosen as concomitant intravenous therapy to inhaled colistin for the studied cystic fibrosis patients

4.2.6. Cystic fibrosis patient outcome measures

Of the 13 CF patients studied, 100% were discharged from hospital and the median hospital LOS was 15 days (5-46 days). Similar utilization studies showing overall hospital LOS in CF patients on colistin inhalation therapy could not be found in the literature to draw comparisons.

4.2.7. Microbial analysis of patients with other lower respiratory tract infections on aerosolised colistin

In this study data regarding whether a patient was mechanically ventilated or not could not be established as this was not reliably recorded on the Bluebird® system. However, the aerosolised utilization of colistin in this subset of patients were due to an infection of the lower respiratory tract based on culture specimens taken and indication for treatment which could have included those of ventilator associated infection.

Of the 12 patients included in this study who received colistin inhalation therapy for a LRTI (other than CF), 100% had cultures taken that also demonstrated growth. Organisms were cultured from either tracheal aspirates 66.67% (n=8), or sputum samples 33.3% (n=4). The organisms identified in these patients included: *P. aeruginosa* 50% (n=6), *A. baumannii* 33.3% (n=4) and *K. pneumoniae* 16.7% (n=2). All organisms isolated were classified as MDR due to evidence of resistance to three or more antibiotic classes. A breakdown of the cumulative sensitivity per organism could not be accurately achieved due to the very small sample sizes of each of the identified organisms of this patient population; however, all isolates were sensitive to colistin.

4.2.8. Evaluation of aerosolised colistin dosing and duration of therapy for patients with other multidrug-resistant lower respiratory tract infections

A breakdown of the inhaled colistin dosing regimens prescribed for the 12 patients with LRTI's is summarised in Table 4.2. A large variation in prescribed inhaled colistin dose in these patients is evident and, no clear preference in appropriate dose regimen could be established from this evaluation.

The median duration of inhaled colistin therapy of this subset of patients was 9.5 days (3-23 days).

Table 4.2. Evaluation of the dosing regimens prescribed for patients with other multidrug-resistant lower respiratory tract infections on inhaled colistin therapy

Aerosolised dose prescribed in Million International Units (MU)	Frequency of administration prescribed in hourly (hrly) intervals	*%* of patients *(n)*
1	8	33.3 (4)
1	12	33.3 (4)
2	8	8.3 (1)
2	12	25.0 (3)

4.2.9. Compliance of study patients with multidrug-resistant lower respiratory tract infections to South African aerosolised colistin dosing guidelines.

The colistin dosing guideline by Labuschagne et al. (2016) included dosing recommendation for inhaled therapy. Compliance of the 12 patients' inhaled colistin dose evaluated in this study to the guideline is poor at 41.7%. This indicates that only five patients were prescribed an accurate and appropriate dose according to local guideline recommendations (Table 4.3).

Table 4.3. Study patients' compliance to the South African Society of Clinical Pharmacy colistin dosing guideline for inhaled colistin therapy in LRTI's (Labuschagne et al., 2016)

Body weight	Dosing recommendation
<40 kg	0.5 MU 12 hrly
> 40 kg	1 MU 12 hrly
Recurrent or severe pulmonary infections	2 MU 8 hrly
Study patients compliance to recommended dosing guidelines*	
	Compliant: 41.7% (n=5)
	Non-compliant: 58.3% (n=7)

*patients for which weight data was missing were assumed to be >40kg as the study only included adults patients.

4.2.10. Co-administered antibiotic therapy for patients with LRTI's.

None of the patients studied in this subset received dual inhalation and IV colistin and only inhaled colistin with IV therapy of other antibiotics was prescribed. The majority 83% (n=10) of patients, received such concomitant antibiotic therapy and the choice of antibiotic agent was varied and determined by the prescriber (Figure 4.5). For 20.0% (n= 2) of patients two additional IV antibiotics were received and, 80.0% (n= 8) of patients received one additional IV antibiotic. Two patients (17%) were prescribed colistin inhalation therapy alone.

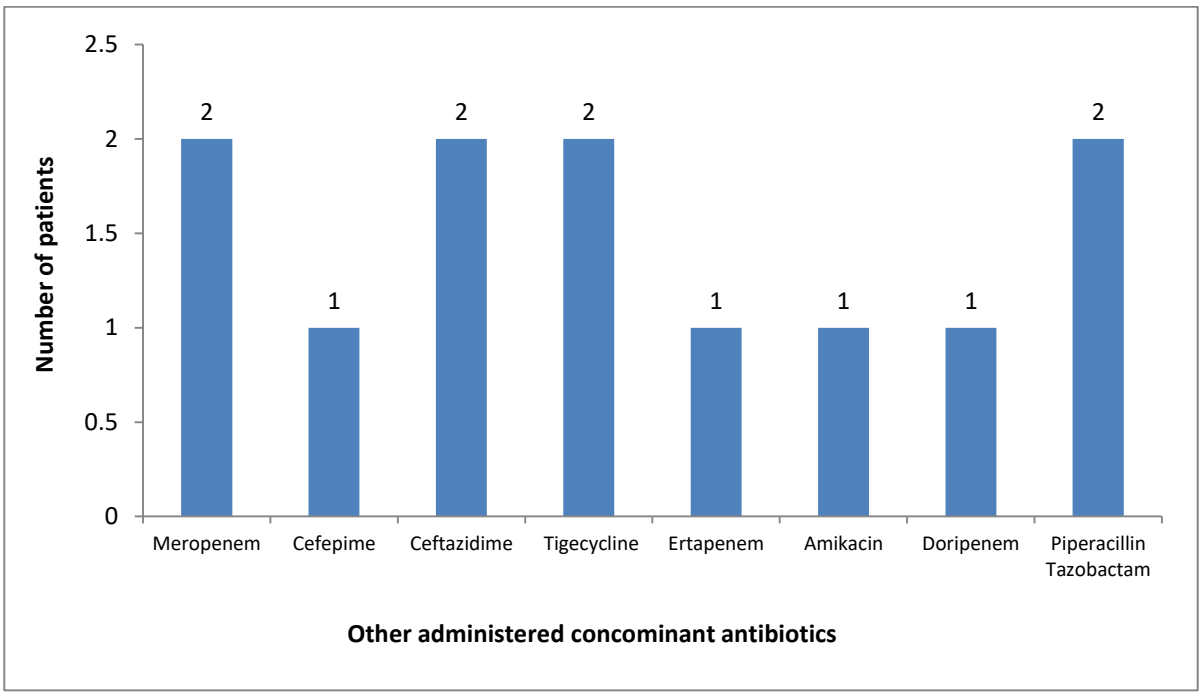

Figure 4.5. A breakdown of combination agents of choice prescribed intravenously with inhaled colistin therapy for patients with nosocomial lower respiratory tract infections

4.2.11. Lower respiratory tract infection patient outcome measures

Of the 12 patients with LRTI's on inhaled colistin treatment studied, 91.7% (n=11) of patients were discharged. The median ICU LOS of these patients was 38 days (8-145 days) and the median hospital LOS 74.5 days (8-195 days).

4.3. Discussion

This study found that inhaled colistin therapy was only prescribed in one hospital. For those patients with CF, for whom inhaled colistin therapy is common practice, as expected MDR *P. aeruginosa* was the only organism identified. Compliance to recommended dosing guidelines was good (92.3%) and appropriate antibitoic combination therapy was prescribed. The use of inhaled colistin has had to become a necessity for CF patients as a consequence of the escalating rates of MDR *P. aeruginosa* detected which is currently estimated at 18.1% (Yapa et al., 2014; Stefani et al., 2017). The risk of MDR *P. aeruginosa* has been shown to increase in the presence of diabetes, long term exposure to tobramycin and frequent acute exacerbations requiring IV therapy in hospital stays (Stefani et al., 2017). However, defining organism resistance according to conventional breakpoint concentrations achieved from IV therapy is not applicable when considered in the context of CF and the extremely high antibiotic concentrations achieved following inhaled administration (Stefani et al., 2017).

Reported colistin resistance following aerosolised exposure ranges from 19-34% in CF patients (Beringer, 2001) and evidence of the spread of colistin resistant *P. aeruginosa* strains in CF patients in both the United Kingdom and Denmark exists (Stefani et al., 2017). Some evidence suggests that the resistance may be transient and reversed when exposure to a particular agent is stopped or switched, however; anti-pseudomonal combination therapy (IV and inhalation) in the treatment of CF patients is still advised, to limit the development of resistance, add synergy and, offer additional symptomatic relief (Doring et al., 2000). In addition, inhaled antibiotic therapy may not reach all areas of the lungs in CF patients, due to obstruction of the airways, and so adjunct systemic therapy is recommended to enhance the prognosis of *P. aeruginosa* infection in an effort to preserve the health of the lung tissue for as long as possible (Ciofu et al., 2015; Emiralioglu et al., 2016).

The concept of antibiotic quorum-sensing inhibition has been studied in an attempt to break through the biofilm for the treatment of chronic mucoid *P. aeruginosa* infection. Quorum-sensing is a mechanism that inhibits the production of certain elements that are involved in the development of the biofilm (such as extracellular DNA) to weaken the matrix and allow for antibiotic penetration into the organism (Ciofu et al., 2015). Antibiotics such as azithromycin, ciprofloxacin and ceftazidime have shown to inhibit quorum-sensing in *P.*

aeruginosa and are recommended co-administered agents for advanced stage CF patients (Koerner-Rettberg and Ballmann, 2014; Ciofu et al., 2015).

The duration of inhaled colistin therapy for these patients, however, was found to be shorter than recommended (Elborn. 2016; Stefani et al., 2017). These patients could have received additional doses of inhaled colistin therapy as part of the take home medication prescriptions to continue therapy following hospital discharge.

For patients with other LRTI's in this study the large disparity of inhaled colistin doses prescribed and the variation in other co-adminstered IV antibiotics indicates a lack of consistency and understanding of the suitable dosing regimen to select in these patients. This could be due to the lack of consensus and limited guidelines available on appropriate inhaled colistin treatment regimens.

The only available South African guideline that provides dosing recommendations for inhaled colistin in LRTI's is that by Labuschagne et al. (2016). Even though the sample size is limited for this subset of patients, it is the first evaluation of inhaled colistin therapy conducted in a South African private hospital for the treatment of LRTI's. The evaluation, however, reveals that there is much improvement required in increasing compliance to the prescription of an appropriate inhaled colistin dose for nosocomial LRTI's as currently compliance is at 41.7%.

Furthermore, these patients all received inhaled colistin therapy without concomitant IV colistin therapy. Direct comparisons cannot be made relating to duration of therapy due to limitations of the literature in describing duration of inhaled colistin monotherapy without adjunctive IV colistin. The findings of this study (9.5 days) are comparable though to durations described in other studies where median durations of inhaled and concurrent IV colistin was administered for 7.5 days (Michalopoulos et al., 2005), 13 days (Kofteridis et al., 2010) and 11.23 days (Demirdal et al., 2016).

The in-hospital LOS demonstrated for patients with LRTI's on inhaled colistin therapy is considerably longer than that documented in previous studies which revealed ICU LOS of 20.5 days (Michalopoulos et al., 2005; Kofteridis et al., 2010). It is difficult, however, to make direct comparisons of patient outcomes on inhaled colistin therapy in this study to others due to

differences in treatment regimens, drug resistance profiles, organisms detected, and mean patient ages.

4.4. Summary and conclusions

- Inhaled colistin is not common practice across South African private hospitals as only one of the hospitals was found to prescribe colistin via the inhalation route.
- Compliance to dosing recommendations of inhaled colistin for patients with CF is good at 92.3% with only one patient evaluated deemed to have received an inappropriate dose.
- All patients with CF were discharged from hospital and were found to receive a shorter median duration of colistin treatment compared to that noted in previously conducted studies.
- There is poor compliance (41.7%) to guideline-based recommendations of inhaled colistin dosing for patients with MDR LRTI's.
- Patients with LRTI's in this study received inhaled colistin therapy in combination with other IV antibiotics and none received both IV and inhaled colistin, which some authors suggest to be best practice.
- The total hospital LOS of patients with MDR LRTI's was prolonged compared to CF patients and all but one patient was discharged.

CHAPTER FIVE

REVIEW OF ORGANISM SENSITIVITY PROFILES AND RESISTANCE PATTERNS

5.1. Introduction

A review of the cumulative antibiotic sensitivity profiles (antibiograms) of the major organisms identified in this study, including *K. pneumoniae, P. aeruginosa* and *A. baumannii,* was obtained from analysis of the actual laboratory reports for which the results were issued per patient as available on the Bluebird® system. Antibiograms are used in clinical practice to select appropriate empiric and directed therapy for patients in a hospital; based on the bacterial environment and resistance patterns established as a result. Regrettably, due to the large proportion of missing data from susceptibility reports evaluated for many of the isolates, antibiograms according to their strict criteria could not be accurately produced for this analysis (ACSQHC, 2013). As such, syndromic antibiograms were developed, and will be evaluated, based on the specimen types of all patients included in the study that had more than 30 isolates identified per source. Blood (n=66) and sputum (n=31) study sample results met this criteria. In addition, the susceptibility of the three major Gram-negative organisms cultured in patients who received IV colistin treatment is reviewed.

5.2. Results and discussion

5.2.1. The syndromic antibiogram established from the study blood samples

A distribution of the organisms identified from the blood samples (n=66) of all study patients is depicted in Figure 5.1. In total, nine species were identified, however, *K. pneumoniae* was the most predominant organism followed by *P. aeruginosa* and *A. baumannii*.

The extent of resistance of the three major Gram-negative organisms identified in blood samples is described in Figure 5.2. The classification of MDR was defined as organisms that showed resistance to three or more classes of antibiotics and XDR as those only showing susceptibility to colistin. It is important to note that three isolates of *K. pneumoniae* from

blood samples were reported as colistin resistant, however, sensitivity to the carbapenems was described in these instances. All isolates of each of the three predominant Gram-negative species identified were classified as either MDR or XDR. Of the other *Enterobacteriaceae* identified in Figure 5.1, four were not classified as MDR or XDR for which the prescription of colistin could be deemed inappropriate. However, in most of these instances, and as discussed in previous chapters, colistin was prescribed empirically and then evidently de-escalated in 69.9% of patients when sensitivity reports became available. The remaining 11 isolates for the other organisms identified were also classified as MDR and therefore compelled the use of colistin for the study patients.

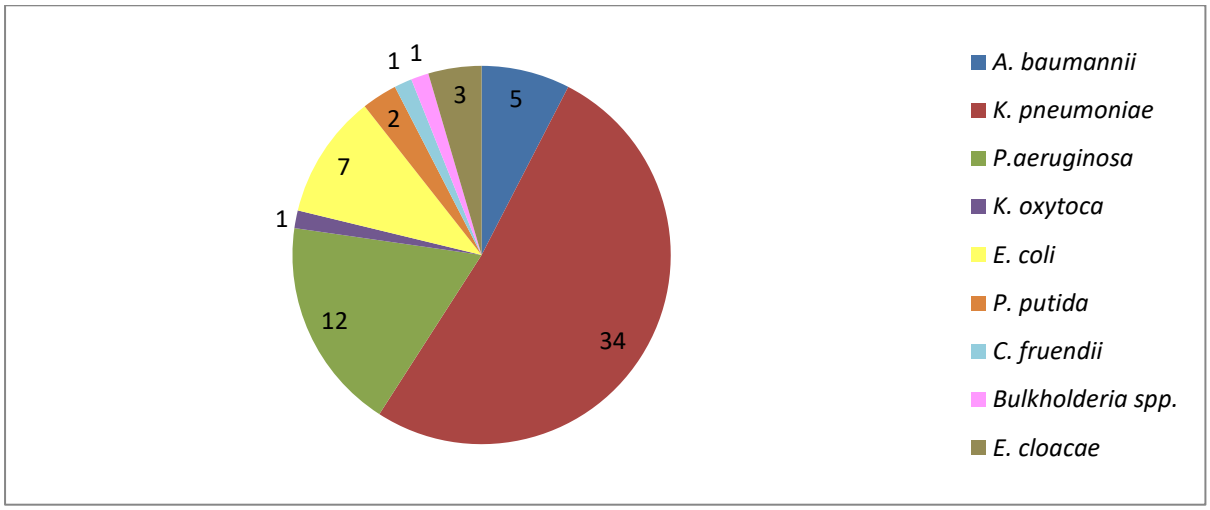

Figure 5.1. Species distribution identified from blood samples (n=66) of patients included in this study

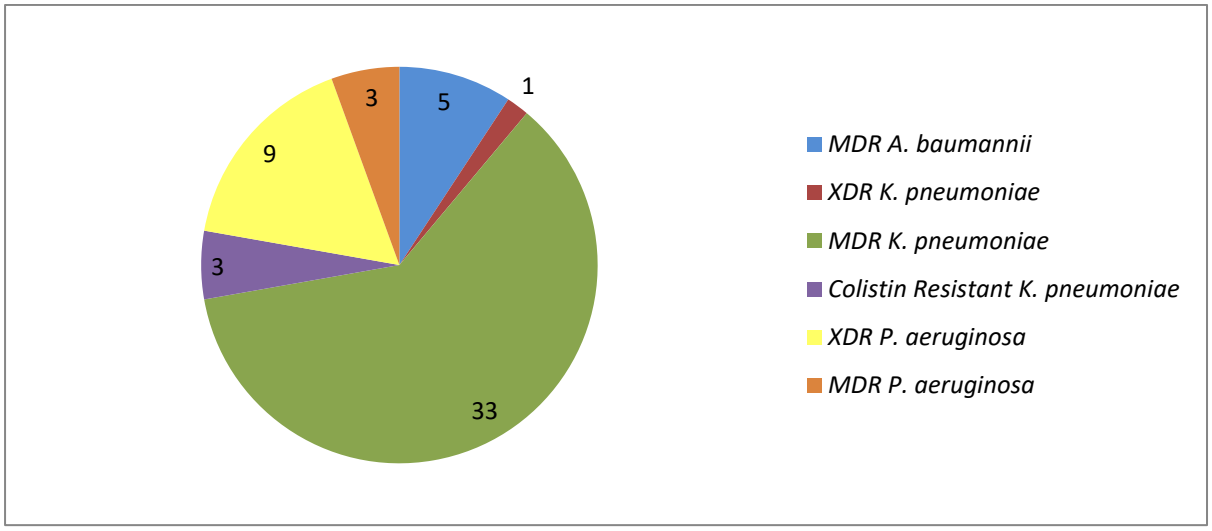

Figure 5.2 Major Gram-negative organisms as a function to resistance in bacteraemia of patients included in the study

The cumulative antibiogram for bacteraemic pathogens is illustrated in Figure 5.3. This indicates the percentage resistant versus susceptible for all the organisms identified from the blood samples studied. Percentage calculations were based on the total number of samples tested for susceptibility per antibiotic. As can be noted, substantial resistance of the bacteraemic pathogens for many of the antibiotics is depicted, in particular for the β-lactams such as cephalosporins and those with additional enzyme inhibitors. Furthermore, the sensitivity of the carbapenems: ertapenem (56.0%), doripenem (59.0%), imipenem (61.0%) and meropenem (62.0%) suggest their role as empiric monotherapy for sepsis in this setting is limited. This is because only those antibiotics that exhibit sensitivities greater than 90.0% are usually selected for therapy of bacteraemia and then, it is essential that the drug delivery is optimised to give the patient the best chance of survival (Deresinski, 2007). Therefore, for the bacteraemic pathogens in this study, colistin was the most active agent considering 95.0% susceptibility to all organisms was observed. The evaluation of organism sensitivity and the extent of resistance identified in these pathogens highlights the magnitude of the threat of antibiotic resistance facing patients currently in South African hospitals.

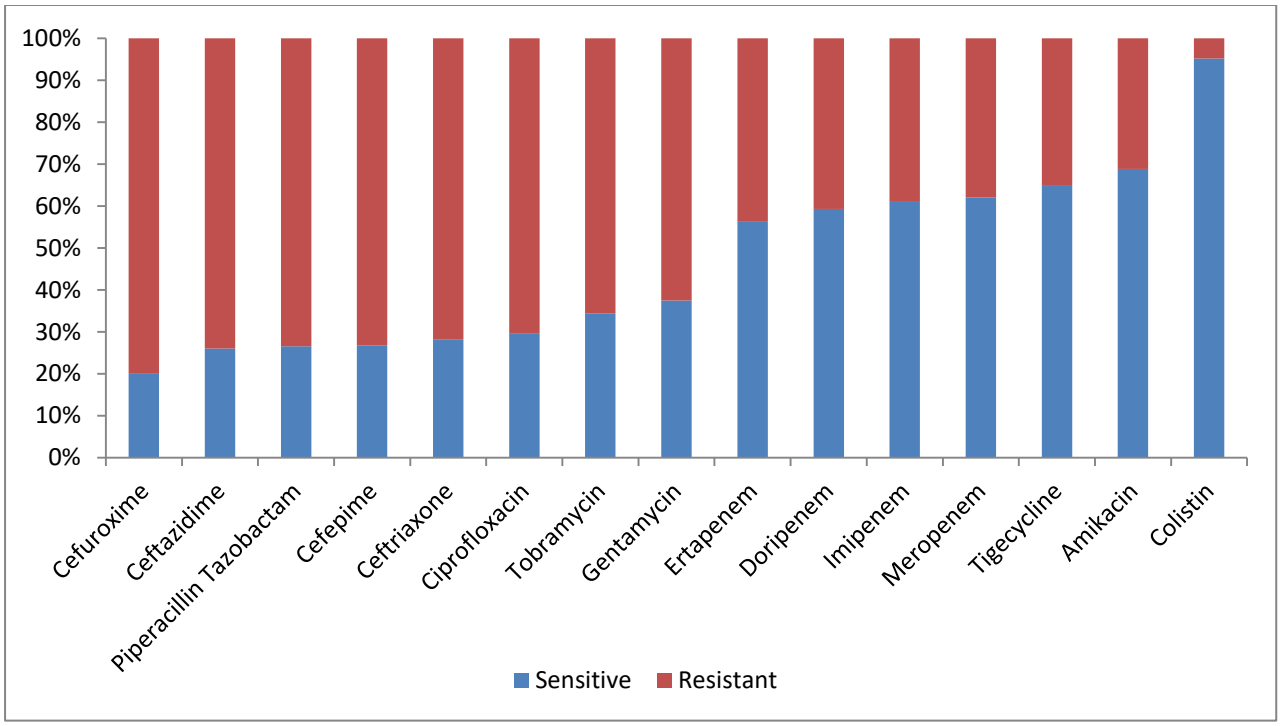

Figure 5.3. Cumulative antibiogram from bacteraemic pathogens (n=66) identified in the study

5.2.2. Syndromic antibiogram established from the study of sputum samples

A distribution of the organisms identified from the sputum samples (n=31) of all study patients is depicted in Figure 5.4. Fifteen of these pathogens were cultured in patients who received inhalation colistin therapy. Only four species were isolated from the respiratory tract with *P. aeruginosa* the most predominant pathogen. This is to be expected due to the inclusion of patients with CF in the study (Chapter four). *Klebsiella pneumoniae* was the second most commonly identified species followed by *A. baumannii* and *E. cloacae*.

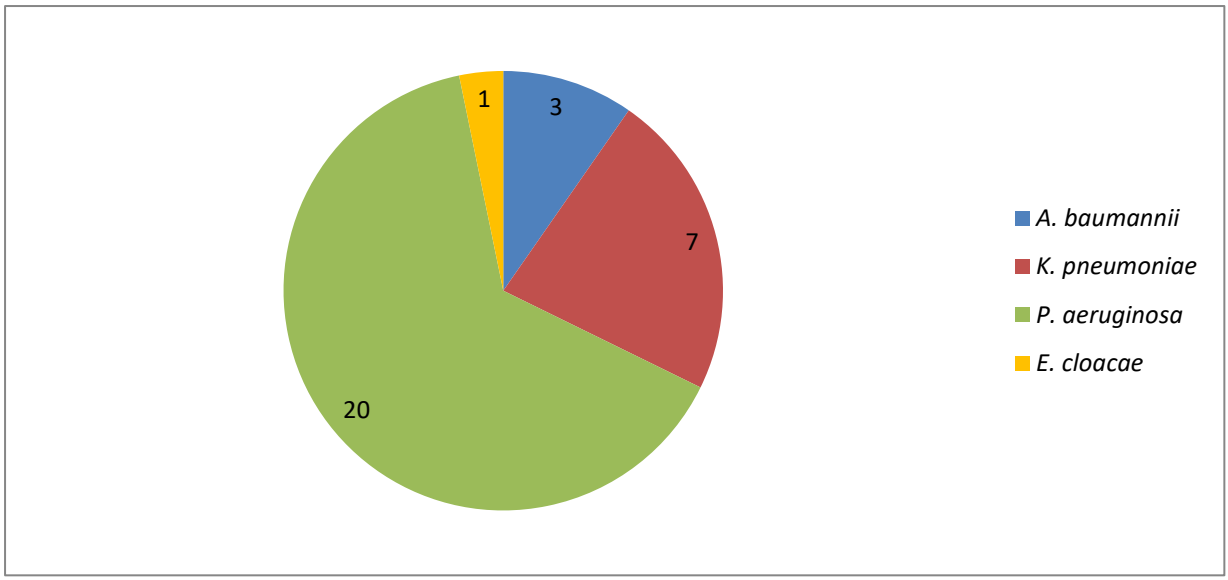

Figure 5.4. Species distribution identified from sputum samples (n=31) of patients included in this study

Evaluation of the three major Gram-negative pathogens identified from sputum samples as a function of resistance is illustrated in Figure 5.5. All (n=3) the *A. baumannii* pathogens were classified as MDR. The extent of resistance amongst *K. pneumoniae* (n= 7) was demonstrated with the majority classified as MDR (n=6) and one isolate XDR. Furthermore, only two *P. aeruginosa* isolates were not MDR or XDR. The one *E. cloacae* isolate was shown to be colistin resistant; however, sensitivity to numerous other classes of antibiotics was reported. This could possibly indicate hetero-resistance of this strain of *E. cloacae* to colistin which was recently described by Napier et al. (2014).

The cumulative antibiogram for pathogens identified from sputum samples is described in Figure 5.6. Due to *P. aeruginosa* being the principal organism in this subset, only those antibiotics for which data was available and which demonstrate pseudomonal activity were included in the figure. Similar to that discussed in Section 5.2.1 for blood samples, the sensitivity of the β-lactam antibiotics including the cephalosporins and piperacillin-tazobactam, as well as, the quinolones (ciprofloxacin) and aminoglycosides (gentamicin and tobramycin) was shown to be ≤ 35% for the *P. aeruginosa* isolates obtained from sputum. The poor activity of the carbapenems: imipenem (35%), meropenem (42%), doripenem (44%) is concerning and demonstrates the species' inherent ability to accumulate resistance. Amikacin demonstrated susceptibility greater than 50% but even this is too low to support the empirical use of this agent as monotherapy. Once again, colistin was the most active agent demonstrating 97% sensitivity amongst Gram-negative bacteria cultured from the respiratory tract.

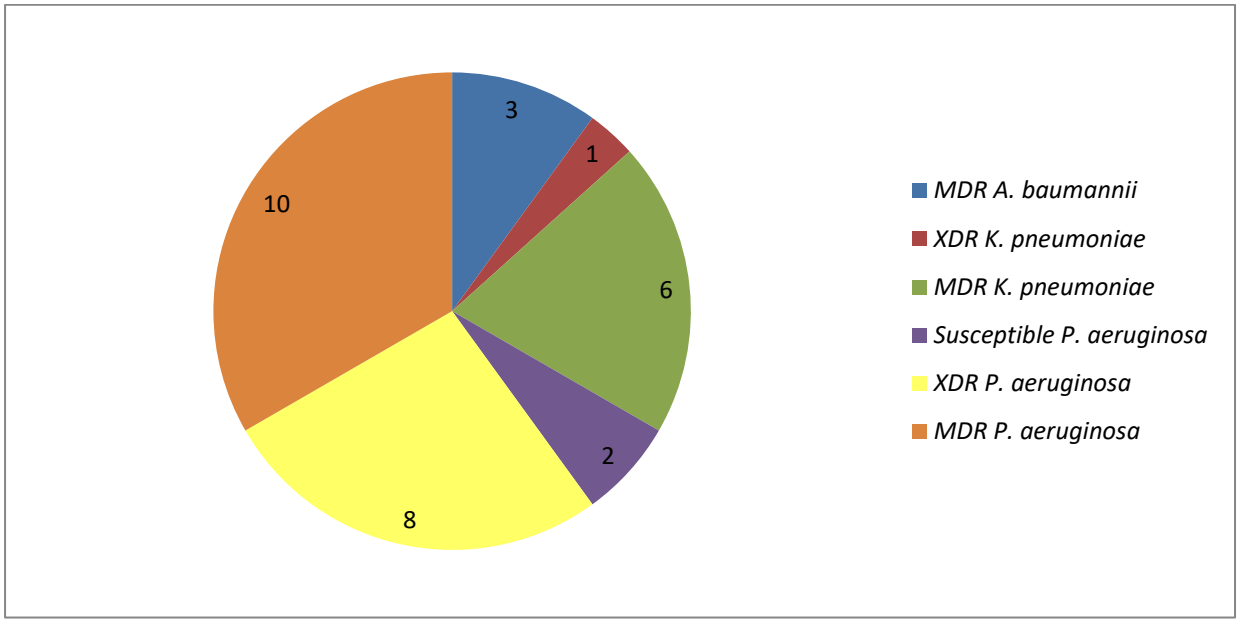

Figure 5.5. Major Gram-negative organisms as a function to resistance in sputum sources of patients included in the study

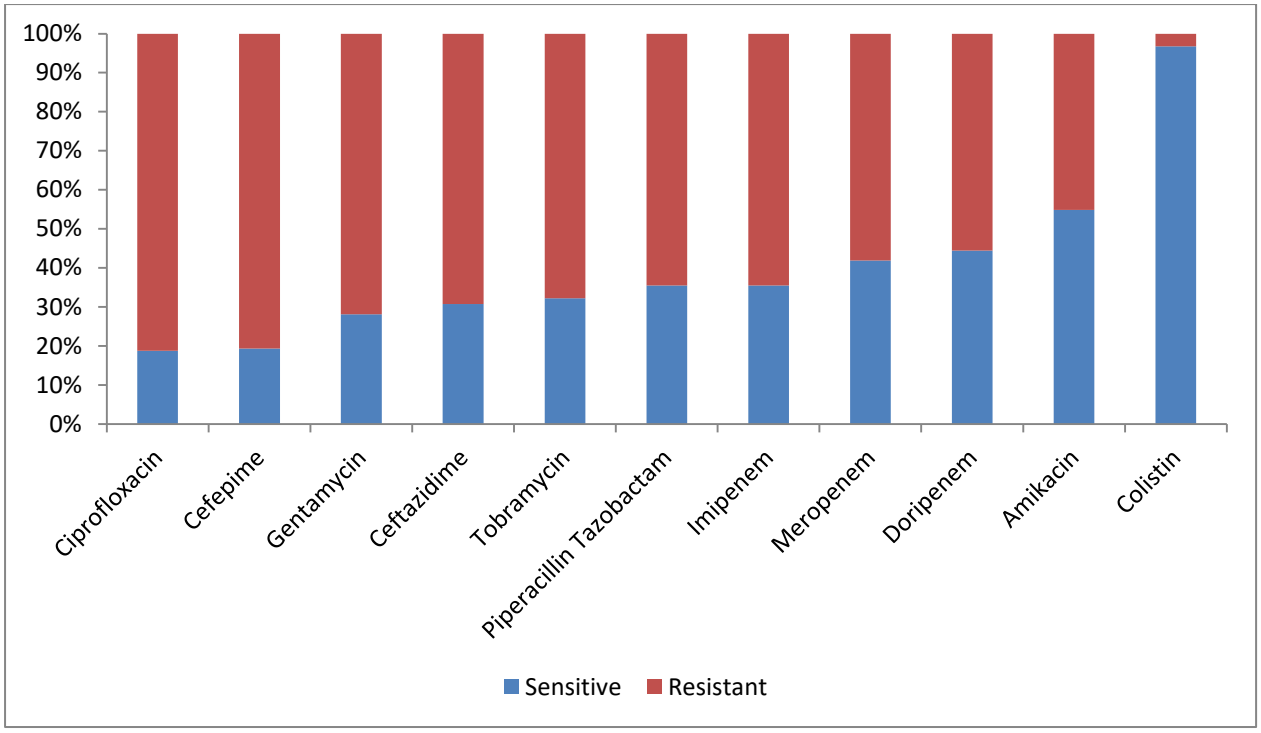

Figure 5.6. Cumulative antibiogram from sputum pathogens (n=31) identified in the study

5.2.3. Cumulative susceptibility of the three major Gram-negative organisms in patients who received intravenous colistin (all sources)

The susceptibility profile of the three predominant Gram-negative organisms cultured from patients who received IV colistin therapy were combined and also evaluated. The respective cumulative sensitivity profiles are depicted in Table 5.1 for *K. pneumoniae*, *P. aeruginosa* and *A. baumannii*. The results of *A. baumannii* need to be interpreted with caution since there are less than 30 isolates tested for this organism and therefore deductions regarding susceptibility cannot reliably be made based on the rules for antibiograms (ACSQHC, 2013). With this in mind colistin was shown to be 100% sensitive to all the *A. baumannii* isolates (n=18) for which it was tested and tigecyline was the next most susceptible agent (77.0%; n=13).

The resultsfor *K. pneumoniae* and *P. aeruginosa*, however, meet the criteria for validity and as such can be evaluated. Of the *K. pneumoniae* (n=67) and *P. aeruginosa* (n=39) isolates tested for colistin susceptibility, 93.0% and 97.0% were reported as colistin sensitive respectively. For *P. aeruginosa*, the susceptibility of the carbapenems was extremely low: only

10.0%, 8.0% and 6.0% of isolates were sensitive to imipenem, meropenem and doripenem, respectively. For *K. pneumoniae*: susceptibility to imipenem, doripenem, meropenem and ertapenem was 72.0%, 71.0%, 68.0% and 38.0% respectively. These results highlight the inefficacy of the carbapenem class of antibiotics as single therapeutic options for either of these two organisms, which is not surprizing given the fact that colistin is often used for suspected CPE. The 2nd most active antibiotic against both organisms was amikacin although cumulative susceptibility was only 30.0% for *P. aeruginosa* and 80.0% for *K. pneumoniae*.

Table 5.1 Cumulative antibiotic sensitivity profile of *K. pneumoniae, P aeruginosa and A. baumannii* isolates from patients who received intravenous colistin therapy

Antibiotic	*Klebsiella pneumoniae* (n=78)			*Pseudomonas aeruginosa* (n=40)			*Acinetobacter baumanii* (n=18)		
	n	% R*	% S*	n	% R*	% S*	n	% R*	% S*
Amikacin	65	20	80	40	70	30	17	59	41
Cefepime	73	95	5	32	78	22	n/a	n/a	n/a
Ceftazidime	54	94	6	33	79	21	n/a	n/a	n/a
Ceftriaxone	68	96	4	n/a	n/a	n/a	n/a	n/a	n/a
Cefuroxime	73	96	4	n/a	n/a	n/a	n/a	n/a	n/a
Ciprofloxacin	74	84	16	40	75	25	n/a	n/a	n/a
Colistin	67	7	93	39	3	97	18	0	100
Doripenem	52	29	71	34	94	6	15	93	7
Ertapenem	61	62	38	n/a	n/a	n/a	n/a	n/a	n/a
Gentamicin	74	80	20	38	74	26	18	89	11
Imipenem	60	28	72	39	90	10	17	94	6
Meropenem	60	32	68	40	93	8	17	94	6
Piperacillin-Tazobactam	73	96	4	38	82	18	n/a	n/a	n/a
Tigecycline	57	33	67	n/a	n/a	n/a	13	23	77
Tobramycin	65	92	8	39	69	31	17	71	29

*R= resistant; S= sensitive, n/a= antibiotic does not demonstrate activity against this organism

The analysis of the sensitivities of these three predominant Gram-negative organisms identified in the study for patients on IV colistin indicate that no therapeutic alternatives were available as treatment options for the drug-resistant Gram-negative infections observed in the study.

5.2.4. Evaluation of the Minimum inhibitory concentrations (MIC) of the three major Gram-negative organisms

The MIC is defined as the minimum antibiotic concentration required to impede the observable growth of an organism (Andrews, 2001). In this study, only 30.2% (n=71) of isolates were submitted for MIC determination. No MIC tests were performed on *A. baumannii* isolates.

Of the *K. pneumoniae* isolates tested, 67.9% (n=53) had MIC results available on the laboratory reports (Table 5.2). The median MICs for this organism were lower than the MIC breakpoint of < 2.0 mg/L for the carbapenems except for ertapenem (range 0.032-32 mg/L) and for colistin was 0.5 mg/L (range 0.125-32 mg/L) (The European Committee on Antimicrobial Susceptibility Testing, 2017). Minimum inhibitory concentrations were available for only 30% (n=12) of the *P. aeruginosa* isolates, cultured from patients on IV colistIn. The median MIC of colistin for *P. aeruginosa* (n=12) was 1.0 mg/L (range 0.5 – 2.0 mg/L) (Table 5.3). This data should be interpreted with caution due to the small sample size.

The analysis of the available MIC data for this organism indicates the level of carbapenem resistance of these isolates, which would require antibiotic doses at unsafe and toxic levels, if colistin were not available as a treatment option.

In this regard, the MIC_{50} and MIC_{90} (minimum antibiotic concentration required to inhibit 50.0% and 90.0% growth of each species) was calculated for both *K. pneumoniae* and *P. aeruginosa* and described in Table 5.4. The present colistin MIC_{50} (0.5 mg/L) and MIC_{90} (1 mg/L) for MDR and XDR *K. pneumoniae* infections in the study of IV colistin patients was shown to be below breakpoint and thus currently a viable option for treatment. For *P. aeruginosa* the MIC_{50} and MIC_{90} of all tested antibiotics were multiple folds above their respective breakpoints, except for colistin where this was determined to currently be 0.5

mg/L and 1 mg/L respectively. The MIC data indicates the extreme levels of resistance reported for *P. aeruginosa* isolates in this cohort of patients where colistin is certainly the only remaining therapeutic agent available with achievable and safe MIC targets.

Table 5.2. Analysis of the Minimum inhibitory concentrations (MIC) of various antibiotics tested against *K. pneumoniae* (n=53) isolates identified in patients who received intravenous colistin therapy

	MIC					
Variable (breakpoint mg/L)	**n**	**Median**	**Interquartile range**		**Minimum**	**Maximum**
Colistin (2)	24	0.5	0.50	0.5	0.125	32.0
Doripenem (1)	33	0.5	0.25	4.0	0.032	32.0
Ertapenem (0.5)	50	3.0	0.50	8.0	0.032	32.0
Imipenem (2)	51	1.0	0.50	4.0	0.25	32.0
Meropenem (2)	52	1.0	0.50	6.0	0.064	32.0

Table 5.3. Analysis of the Minimum inhibitory concentrations (MIC) of various antibiotics tested against *P. aeruginosa* (n=12) isolates identified in patients who received intravenous colistin therapy

	MIC					
Variable (breakpoint mg/L)	**n**	**Median**	**Interquartile range**		**Minimum**	**Maximum**
Amikacin (8)	9	64.0	64.0	64.0	16.0	64.0
Cefepime (8)	8	40.0	12.0	64.0	8.0	64.0
Ceftazidime (8)	9	32.0	16.0	64.0	4.0	64.0
Colistin (2)	11	1.0	0.5	2.0	0.5	2.0
Imipenem (4)	10	16.0	16.0	16.0	2.0	16.0
Meropenem (2)	10	16.0	16.0	16.0	1.0	16.0
Piperacillin Tazobactam (16)	9	128.0	128.0	128	8.0	128.0

Table 5.4. The MIC_{50} and MIC_{90} of antibiotic agents tested agaisnt *K. pneumoniae* and *P. aeruginosa* obtained following evaluation of all specimens tested for MIC in this study

***Klebsiella pneumoniae* (n=53)**							
	Amikacin	**Doripenem**	**Ertapenem**	**Imipenem**	**Meropenem**	**Piptaz**	**Colistin**
MIC_{50} (mg/L)	2.0	0.5	2.0	1.0	1.0	128.0	**0.5**
MIC_{90} (mg/L)	16.0	32.0	32.0	32.0	32.0	128.0	**1.0**
***Pseudomonas aeruginosa* (n=12)**							
	Amikacin	**Cefepime**	**Ceftazidime**	**Imipenem**	**Meropenem**	**Piptaz**	**Colistin**
MIC_{50} (mg/L)	64.0	64.0	64.0	16.0	16.0	128.0	**0.5**
MIC_{90} (mg/L)	64.0	64.0	64.0	16.0	16.0	128.0	**2.0**

5.3. Summary and conclusion

- Evaluation of the syndromic antibiograms revealed substantial resistance to the majority of antibiotics for the bacteraemic pathogens with colistin showing the greatest susceptibility (95%) to all organisms.
- Similarly, for organisms obtained from sputum, the sydromic antibiotic demonstrated colistin as the only viable therapeutic option since 97% sensitivity was observed. The next most sensitive antibiotic agent in this cohort was amikacin showing an activity of only 55%.
- Cumulative sensitivity of the *K. pneumoniae* and *P. aeruginosa* isolates from all sources in patients who received IV colistin was 93.0% and 97.0% respectively with the inadequacy of the carbapenem class of antibiotics highlighted for both organisms.
- For *P. aeruginosa* the MIC_{50} and MIC_{90} of all tested antibiotics were multiple times above their respective breakpoints except when considering colistin (0.5 mg/L ; 1 mg/L). This reveals the extent of resistance of this organism and highlights colistin as the only likely antibiotic possibility.
- Antibiotic resistance patterns of the three major Gram-negative organisms identified from this study were similar regardless of specimen site (blood versus sputum).

- The extensive antibiotic resistance displayed by *P. aeruginosa, K. pneumoniae and A. baumannii* by this study, indicates the limited therapeutic options available against these organisms.
- The current dependence on colistin as last and only feasible therapeutic alternative is emphasized against these organisms.

CHAPTER SIX

THESIS SUMMARY, FUTURE RECOMMENDATIONS AND STUDY CONCLUSION

6.1. Thesis Summary

For the first time in South Africa, the utilization of colistin was studied across four private sector hospitals including 237 patients in order to establish how and why colistin is being used. In addition, patient outcomes following administration of this last resort antibiotic were established and evaluated. The pertinent study findings of IV and aerosolised colistin are summarised accordingly in Chapters three and four and the evaluation of the organism sensitivity profiles identified is described in Chapter five. This study aimed to achieve a baseline of colistin utilization in South Africa with the following objectives as outlined in Chapter one, to provide insight to enhance the appropriate use of this antibiotic in the future.

Objective 1: To ascertain colistin utilization including: dose, dose frequency, route of administration and duration of treatment.

Although many experts believe that colistin should be restricted for use in high level care units due to the toxic nature of the drug and need for enhanced patient safety monitoring in these units, the findings of this study suggest that colistin is used in both the intensive care and general wards of hospitals. This is necessitated due to the need to treat MDR and XDR pathogens outside of ICU's which reflects on the extent and spread of antibiotic resistance across various patients. More patients in the ICU received IV colistin compared to those who received inhaled colistin. The study also found extremely poor compliance to local available colistin dosing guidelines and a large variation in colistin doses prescribed in both IV and aerosolised routes of administration for critically ill patients. This may be due to the complexity of appropriate colistin dosing and the lack of awareness of prescribers to optimise doses according to PK and guideline recommendations. These findings require urgent attention in order to improve compliance to colistin dose according to different renal function categories to enhance patient safety and successful treatment outcome. Furthermore, the

duration of treatment was found to be shorter in patients who received higher colistin IV loading and maintenance doses which further reiterates the need to ensure optimal colistin dosing.

The dosing compliance of patients with CF, however, was found to be good and in accordance with guideline recommendations for these patients. The duration of inhaled colistin therapy for patients with cystic fibrosis was slightly shorter than that noted by other studies internationally.

Objective 2: To ascertain which were the most prevalent infecting organism and source of infections that necessitate the use of colistin.

The three major organisms for which colistin was prescribed in this study was *P. aeruginosa, K. pneumoniae* and *A. baumannii*, all of which demonstrated extremely high levels of resistance to all antibiotics including the carbapenems. Due to the high resistance rates noted in the study colistin was most often the only viable therapeutic option as salvage treatment for these patients. The most common source of organism growth of the patients studied included; blood, sputum, urine and tracheal aspirate samples.

Objective 3: To establish if appropriate antimicrobial stewardship principles are practiced during colistin therapy.

A colistin stewardship bundle was devised in this study (see Section 3.2.11) to measure compliance to possible stewardship related principles for the IV administration of colistin. Compliance to eight stewardship process measures was evaluated including: (1) obtaining an appropriate culture prior to the commencement of colistin therapy; (2) prescription of a loading dose; (3) prescription of an appropriate loading dose; (4) prescription of appropriate maintenance dosing including adjustment according to renal insufficiency; (5) compliance to antibiotic 'hang time'; (6) prescription of colistin in combination with another Gram-negative antibiotic; (7) de-escalation of colistin therapy and (8) median duration of therapy. Following the assessment of the colistin utilization according to these principles, compliance was good although the lowest compliance noted was due to administration 'hang time', inappropriate

loading and maintenance doses prescribed (which were proven critical to patient outcomes) and deficiency of de-escalation practices. Therefore, improvement in several colistin process measures particularly maintenance dosing warrants immediate consideration. Similar findings relating to inappropriate doses were noted for patients who received inhaled colistin therapy for LRTI's. Compliance to duration of therapy could not be established as recommendations are lacking in South African and international guidelines. Overall, composite compliance to the stewardship bundle proposed in this study for patients on IV colistin therapy was found to be 81.2% at best which warrants improvement in the future.

Objective 4: To establish patient outcomes while on colistin therapy including effects on renal function, hospital length of stay and overall mortality.

A 29.6% in-hospital mortality rate was observed in patients who received IV colistin with a median duration of ICU LOS of approximately a month and median overall hospital LOS of approximately six weeks. The effects of renal function following IV colistin administration were negligible and insignificant as no difference in renal function was noted pre and post colistin administration. This is in line with recent findings that colistin may not be as toxic as originally thought and that renal toxicity may be short-lived and reversed once treatment is stopped (Dalfino et al., 2012). The results of the outcome measures evaluated echo what is expected for critically ill patients with MDR infections including, prolonged hospital LOS and poor patient outcomes (Deresinski, 2007). Of importance to this study, patients who received higher colistin loading and maintenance doses were shown to have better in-hospital overall outcomes. All of the patients who received inhaled colistin for cystic fibrosis were discharged and their hospital LOS was considerably shorter than median hospital LOS of ten and a half weeks recorded for patients who received inhaled colistin for a MDR LRTI's.

6.2. Limitations of this study

These included:

- The retrospective nature and the need to collect data from electronic prescription records which were completed and captured by frontline pharmacists.

- The electronic system which did not have the facility to record parameters such as renal replacement therapy and patient weight.
- The inability to access and review clinical notes and other clinical parameters including but not limited to: other medication prescribed, patient temperature, blood pressure and other indwelling devices.
- The high numbers of patients that did not have their weight recorded made it difficult to calculate creatinine clearances using the Cockcroft-gault equation and thus compliance with the SASOCP dosing guidelines based on creatinine clearance may be skewed.
- Although not an aim of the study, the illness severity score such as the APACHE II score were not recorded and therefore patient risk in relation to mortality or outcome could not be corrected for.
- Duration of therapy could not be used as a stewardship process indicator due to the limited guidance available as to what an appropriate duration of IV colistin should be.
- Finally, data on side effects of colistin other than nephrotoxicity were not actively investigated.

6.3. Future recommendations

This study has identified recommendations to improve the utilization of colistin going forward. As such and optimistically within five to ten years, the introduction of colistin therapeutic drug monitoring (TDM) would be useful to individualize and optimise dosing, given the variability of PK and PD parameters displayed in critically ill patients and the growing need to refine therapy and maintain the efficacy of the antibiotic for the future. However, this may not be a practically viable option in the current context of healthcare in South Africa. This is due to the additional infrastructure required by the laboratories, the anticipated associated costs and the limited amount of TDM currently in place which is largely reserved for the aminoglycoside class of antibiotics and vancomycin.

In addition, the package insert of colistin should be updated and amended accordingly with the latest accurate PK and PD data. This will better inform and assist pharmacists and prescribers of the appropriate dosing strategies required to achieve therapeutic efficacy for critically ill patients, and serve as a quick and reliable reference.

As an immediate solution, however, the design and implementation of a national intravenous colistin antibiotic stewardship bundle (Table 6.1), to be implemented in all hospitals in which colistin is prescribed, is strongly recommended. The implementation of care bundles in antibiotic stewardship can assist in enhancing compliance to evidence-based quality measures which could in turn improve the utilization of the antibiotic agents. Bundles include a set of evidence-based measures that when implemented together are shown to produce better outcomes and have a greater impact than that of the isolated implementation of individual measures (Reser et al., 2012). Bundles also help to create reliable and consistent care systems in hospital settings since they are clear and concise, in addition to promoting multi-disciplinary collaboration (Reser et al., 2005; Reser et al., 2012).

Table 6.1. Summary of the proposed intravenous colistin process measures as a stewardship bundle

Process measures
• Documentation and record of the patient weight.
• Obtaining an appropriate culture prior to the commencement of colistin therapy.
• Prescription of a loading dose.
• Prescription of an appropriate loading dose.
• Prescription of an appropriate maintenance dose including adjustment according to renal insufficiency.
• Compliance to antibiotic 'hang time'.
• Prescription of colistin in combination with another Gram-negative antibiotic.
• De-escalation of colistin therapy.
• Duration of therapy.

The goal of such a recommendation would be to collectively improve colistin utilization by increasing compliance to targeted stewardship principles - in particular to selection of the appropriate dose - which could have a marked impact on improving the appropriate use of colistin. As a set of audit measures, such an intervention would aim to minimize the risk of colistin resistance emerging. Once implemented, it would be of paramount importance to

compare compliance to process and outcome measures in an interrupted time series study (pre and post implementation of such a bundle) at individual hospitals or at a national level. Furthermore, changes in colistin susceptibility could be longitudinally monitored over time.

6.4. Conclusion

This study identified that stewardship opportunities for improving colistin prescription and utilization exist (for both administration routes), and recommends the implementation of a colistin stewardship care bundle to preserve colistin efficacy in the foreseeable future (Messina et al., 2017). Results of the study showed that those patients who received higher IV colistin doses demonstrated shorter durations of treatment and better overall in-hospital outcomes. The appropriate dosing of colistin is complex but this should not be reason enough to not get it right. As a result of the escalating rates of MDR and XDR Gram-negative organisms currently being experienced in South Africa, and evidenced by this study, pharmacists along with prescribers should take up the challenge and work collectively as a team to always ensure the appropriate use of colistin. The consequences of not doing so could have devastating consequences for public health in South Africa and beyond.

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APPENDIX A

PP04 Abstract submitted (Podium): All Africa Congress on Pharmacology and Pharmacy 2016

Evaluation of colistin utilization in patients with multidrug- and extensive drug-resistant Gram-negative infections in four private hospitals in South Africa.

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[2]Department of Quality Leadership, Johannesburg, South Africa
[3]Department of Clinical Microbiology, Ampath Laboratories, Johannesburg

Purpose: The emergence of life-threatening multidrug-resistant (MDR) and extremely drug resistant (XDR) bacteria has been widely documented as a global threat to society. These 'super-bugs' have gained resistance mechanisms to almost all antibiotics currently available necessitating the use of older, more toxic drugs such as colistin, as salvage therapy. Whilst emergence and spread of colistin resistance was recently documented in South Africa, compliance to current dosing guidelines is unknown and no local information of why and how colistin is prescribed is available. The primary purpose of this study was therefore to evaluate the current utilization of colistin, in order to develop an antimicrobial stewardship intervention to improve compliance to colistin process measures with the goal of enhancing outcomes.

Methods: Electronic patient records of all adult patients in four Johannesburg hospitals with colistin were retrospectively reviewed over a five month period. The following data was collected: patient demographics, organism results and antibiotic susceptibility profiles, diagnosis, indication for colistin use as well as - dose, duration of therapy, route, administration of a loading dose, prescription in combination or as monotherapy, de-escalation of therapy following organism results and effects on renal function. Evaluation of outcome measures included: overall mortality, Intensive Care Unit length of stay (LOS), and hospital LOS while on colistin therapy.

Results: Evaluation of study results demonstrated that the mean age of the patient population (n=64) was 50 years of which 60.9% were male. Administration occurred mostly in the intensive care units (76.6%). Therapy was regularly administered intravenously (IV) (90.6%) followed by nebulisation (7.8%). The mean duration of colistin therapy was 13,6 days. The compliance rate of administration of a loading dose (95.8%) and as combination therapy (98.3%) was high, although daily dosing regimens in million units (MU) of colistin varied considerably from 1MU, 1,5MU, 2MU and 3MU IV 8 hourly to 1,5, 2MU, 3MU, and 4.5 MU IV 12 hourly. Colistin was prescribed as directed or definitive therapy in 73.4% of patients, with 26.6% of treatment being initiated empirically. Organisms justifying the need for colistin use include; *Klebsiella pneumoniae* (35.9%), *Pseudomonas aeruginosa* (26.6%) and *Acinetobacter baumannii* (15.6%). Outcomes measures reflect a 29.7% overall mortality rate and an average LOS for hospital (55.9 days) and ICU (37.4 days).

Conclusions: The data suggests that several opportunities to improve appropriate colistin use exist particularly regarding the dose and duration of therapy. In contrast, compliance to loading dose administration was >90%.

SAASP 15 Abstract submitted (Podium): 7th Federation of Infectious Diseases Society of South Africa (FIDSSA) Conference, 2017

Evaluation of colistin utilization across multiple South African private hospitals: Indeed it's time for colistin stewardship.

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Introduction: The increased prevalence of multidrug-resistant Gram-negative infections in critically ill patients has resulted in the re-introduction of colistin, a previously considered toxic antibiotic, as rescue therapy. Although colistin resistance has been documented in South Africa, there is no local evidence as to why and how colistin is used in hospitals. This study aimed to evaluate the utilization of colistin in order to identify stewardship opportunities regarding its' appropriate use in the future.

Method: A retrospective electronic record review of adult patients on intravenous colistin therapy for more than 72 hours in four Gauteng private hospitals was conducted between 1 September 2015- 30 June 2016. Evaluation of six colistin stewardship process measures (colistin bundle) were reviewed; obtaining a culture prior to therapy, administration of a loading dose, administration of the correct loading dose, maintenance dose modifications based on renal function, whether colistin was administered in combination and if de-escalation following culture and sensitivity results occurred. Outcome measures included; effects on renal function, overall hospital mortality, intensive care unit length of stay (LOS), and hospital LOS.

Results: Results of 199 patients demonstrated a 75.9% composite compliance to the colistin stewardship bundle. Non-compliance was mainly due to incorrect loading and maintenance doses prescribed and inappropriate dose adjustment according to renal function. Compliance to local, current colistin dosing guidelines was at best 48.2%. Significantly shorter durations of treatment were found in patients who received higher loading doses (p=0.040) and in those that received maintenance doses of 4.5 MU twice daily vs 3MU three times daily (p=0.0027). In addition, more of the patients that demised received the 3 MU three times daily maintenance doses, compared to those who survived (p=0.0037; phi coefficient=0.26).

Conclusion: This study demonstrated that many stewardship related opportunities to improve appropriate colistin utilization exist in particular relating to dose. Colistin stewardship should be implemented as a matter of urgency to preserve the efficacy of this last resort antibiotic.

SAPJ Invited article as winner of the Academy of Pharmaceutical Sciences Young Scientist award.

Cum Laude

Academy of Pharmaceutical Sciences

Our collective contribution matters: Pharmacists unite in tackling antibiotic resistance for South Africa

A P Messina[a,b] BPharm, **S Van Vuuren**[a] PhD, **A J Brink**[c,d] MBChB, MMed

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[c] Ampath National Laboratory Services, Milpark Hospital, Johannesburg, South Africa
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Angelliki Messina is the Boehringer Ingelheim Young Scientist winner in the category Pharmaceutical Practice, which was awarded for a presentation on colistin utilisation

The relentless misuse (including under dosing, inappropriate duration and incorrect indications) of antibiotics globally has over time hastened the natural process of antibiotic resistance which Sir Alexander Fleming warned us of in 1945. Many bacteria today have gained resistance mechanisms to combat antibiotics causing experts to caution that we are currently at the dawn of a post antibiotic era. As a result, the advancements in modern medicine including organ transplants, chemotherapy and joint replacement surgery could one day be a memory of the past since these procedures may be too risky to perform without effective antibiotic prophylaxis.

It is safe to say that antibiotic resistance is currently one of the biggest global public health threats. It is predicted to be the leading cause of mortality by 2050 (one person dying every 3 seconds) if the existing status quo continues[1]. In this overview, we describe the state of antibiotic resistance in South Africa and how pharmacists can work together to tackle antibiotic resistance.

Multi-drug resistant (MDR) bacteria, only sensitive to last line therapy or pan resistant are escalating at a distressing rate in South Africa. These organisms, particularly MDR Gram-negative pathogens, pose a specific threat to hospital environments necessitating the use of older, more toxic drugs such as colistin, as a final option to help treat severe infections[2,3,4].

An editorial entitled "Wake up, South Africa! The antibiotic horse has bolted" indicated that the rise in MDR Gram-negative bacteria including the carbapenem-resistant Enterobacteriaceae (CRE) was as a result of a home grown multifaceted problem including the abuse of all antibiotics[5].

A study by Kift et al. (2014) indicated that carbapenem susceptibility decreased by 18% over a four year period in South African public sector hospitals[6].

A local prevalence study conducted by Paruk et al. (2012), evaluated antibiotic prescription practices in the intensive care units (ICU) of both public and private sector hospitals in five provinces. This study found that unsuitable antibiotics were initiated in over 50% of patients reviewed and 72% of these patients received antibiotic therapy for an inappropriate duration[7].

Alarmingly, van Boeckel et al. (2014) noted that antibiotic consumption increased in proportionately to population growth during 2000-2010 in the BRICS countries, of which South Africa is one[8].

The problems experienced currently with drug-resistant tuberculosis and non-*albicans Candida* infections which are resistant to first line antifungal therapy further enhances the crisis South Africa is facing with MDR organisms[9].

Carbapenem resistance among the Gram-negative Enterobacteriaceae can occur through various mechanisms; however, the most common is through the production of beta-lactamases, a bacterial enzyme which hydrolyses carbapenems and all other beta lactam antibiotics including penicillins and cephalosporins. These CRE's often contain additional mechanisms of resistance to the aminoglycosides and to some extent the fluoroquinolone class of antibiotics[10]. Risk factors for CRE organisms include previous antibiotic exposure, prolonged hospitalization, severe illness and surgery, to name a few[10].

Often, colistin is the last resort antibiotic used to combat severe CRE infections. Devastatingly, documented reports have emerged globally indicating instances of colistin-resistance within Gram-negative pathogens including: *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Acinetobacter baumannii*[3]. In such instances the consequences of MDR and pan-resistant organisms are dire and

SAMJ Publication accepted: Abstract submitted for publication to the South African Medical Journal

Opportunities to optimize colistin stewardship in hospitalized patients in South Africa: Results of a multi-site utilization audit

AP Messina,[1,2] *BPharm;* ***AJ Brink,***[3,4] *MB BCh, MMed (Clin Micro);* ***GA Richards,***[5] *MB BCh, PhD, FCP (SA) FRCP;* ***S van Vuuren***[1] *PhD*

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[4] *Division of Infectious Diseases and HIV Medicine, Department of Medicine, University of Cape Town, Cape Town, South Africa*
[5] *Division of Critical Care, Faculty of Health Sciences, University of the Witwatersrand, Johannesburg and Charlotte Maxeke Johannesburg Academic Hospital, Johannesburg, South Africa.*

Background. Colistin is an old antibiotic which has been reintroduced as salvage therapy in hospitalized patients as it is frequently the only agent active against Gram-negative bacteria. Various guidelines for colistin administration have led to confusion in establishing the appropriate dose which has potential for adverse consequences including treatment failure or toxicity. The emergence and spread of colistin resistance has been documented in South Africa (SA), but no local information exists as to how and why colistin is used in hospitals, and similarly compliance with current dosing guidelines is unknown.

Objectives. To evaluate the current utilization of colistin in SA hospitals, in order to identify stewardship opportunities that could enhance the appropriate use of this antibiotic.

Methods. Electronic patient records of adult patients on intravenous (IV) colistin therapy for more than 72 hours in four private hospitals were retrospectively audited over a ten month period (1 September 2015- 30 June 2016). The following data was recorded; patient demographics, culture and susceptibility profiles, diagnosis and indication for use. Compliance with six colistin process measures were audited; obtaining a culture prior to initiation, administration of a loading dose, administration of the correct loading dose, adjustments to maintenance dose according to renal function, whether it was administered in combination with another antibiotic and whether de-escalation following culture and sensitivity results occurred. Outcome measures included; effects on renal function**,** overall hospital mortality, intensive care unit length of stay (LOS), and hospital LOS.

Results. Records of 199 patients on IV colistin were reviewed. Compliance with obtaining a culture prior to antibiotic therapy was 99%, prescription of a loading dose (93.5%), and prescription of colistin in combination with another agent (98.5%). However, overall composite compliance to the six colistin stewardship process measures was 75.9%. Non-compliance related to inappropriate loading and maintenance doses, lack of adjustment according to renal function, and lack of de-escalation was evident in two-thirds of cases. Significantly shorter durations of treatment were found in patients who received higher loading doses (p=0.040) and in those that received maintenance doses of 4.5 MU twice daily vs 3MU three times daily (p=0.0027). In addition, more of the patients that demised received the 3 MU three times daily maintenance doses, compared with those who survived (p=0.0037; phi coefficient=0.26).

Conclusion. The study identified multiple stewardship opportunities to optimize colistin therapy in hospitalized patients. The urgent implementation of a stewardship bundle to improve colistin utilisation is warranted.

APPENDIX B

RESEARCH OPERATIONS COMMITTEE FINAL APPROVAL OF RESEARCH

Approval number. UNIV-2015-0054

Ms Angeliki Messina

E mail. Angeliki.Messina@netcare.co.za

Dear Ms Messina

RE: EVALUATION OF PATIENTS WITH MULTIDRUG-RESISTANT GRAM-NEGATIVE INFECTIONS TREATED WITH COLISTIN IN FOUR PRIVATE SECTOR HOSPITALS IN SOUTH AFRICA

The above-mentioned research was reviewed by the Research Operations Committee's delegated members and it is with pleasure that we inform you that your application to conduct this research at private Hospitals, has been approved, subject to the following:

i) Research may now commence with this FINAL APPROVAL from the Committee.

ii) All information regarding the Company will be treated as legally privileged and confidential.

iii) The Company's name will not be mentioned without written consent from the Committee.

iv) All legal requirements regarding patient / participant's rights and confidentiality will be complied with.

v) The research will be conducted in compliance with the GUIDELINES FOR GOOD PRACTICE IN THE CONDUCT OF CLINICAL TRIALS IN HUMAN PARTICIPANTS IN SOUTH AFRICA (2006)

vi) The Company must be furnished with a STATUS REPORT on the progress of the study at least annually on 30th September irrespective of the date of approval from the Committee as well as a FINAL REPORT with reference to intention to publish and probable journals for publication, on completion of the study.

vii) A copy of the research report will be provided to the Committee once it is finally approved by the relevant primary party or tertiary institution, or once complete or if discontinued for any reason whatsoever prior to the expected completion date.

viii) The Company has the right to implement any recommendations from the research.

ix) The Company reserves the right to withdraw the approval for research at any time during the process, should the research prove to be detrimental to the subjects/ Company or should the researcher not comply with the conditions of approval.

x) APPROVAL IS VALID FOR A PERIOD OF 36 MONTHS FROM DATE OF THIS LETTER OR COMPLETION OR DISCONTINUATION OF THE TRIAL, WHICHEVER IS THE FIRST.

We wish you success in your research.

Yours faithfully

19/8/2015

Prof Dion du Plessis
Full member: Research Operations Committee & Medical Practitioner evaluating research applications as per Management and Governance Policy

Shannon Nell
Chairperson: Research Operations Committee
Date: 1/9/2015

This letter has been anonymised to ensure confidentiality in the research report. The original letter is available with author of research

APPENDIX C

R14/49 Ms Angeliki Phroso Stephanou and Dr Adrian Brink

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)

CLEARANCE CERTIFICATE NO. M150404

NAME: (Principal Investigator)	Ms Angeliki Phroso Stephanou and Dr Adrian Brink
DEPARTMENT:	Pharmacy and Pharmacology Netcare Milpark, Netcare Unitas and Netcare Pretoria and East Hospitals
PROJECT TITLE:	Evaluation of Patients with Multi-Drug Resistant Gram Negative Infections Treated with Colistin in Four Private Sector Hospitals in South Africa
DATE CONSIDERED:	24/04/2015
DECISION:	Approved unconditionally
CONDITIONS:	
SUPERVISOR:	Ass Prof Sandy van Vuuren
APPROVED BY:	Professor P Cleaton-Jones, Chairperson, HREC (Medical)
DATE OF APPROVAL:	05/06/2015

This clearance certificate is valid for 5 years from date of approval. Extension may be applied for.

DECLARATION OF INVESTIGATORS

To be completed in duplicate and **ONE COPY** returned to the Secretary in Room 10004, 10th floor, Senate House, University.

I/we fully understand the conditions under which I am/we are authorized to carry out the above-mentioned research and I/we undertake to ensure compliance with these conditions. Should any departure be contemplated from the research protocol as approved, I/we undertake to resubmit the application to the Committee. I **agree to submit a yearly progress report**.

Principal Investigator Signature Date

PLEASE QUOTE THE PROTOCOL NUMBER IN ALL ENQUIRIES

APPENDIX D

Hospital: _____ Gender: _____ Admitting Diagnosis: _____

Patient Study Number: _____ Age: _____ _____

Date: _____ Weight: _____ _____

Ward: _____ APACHE II Score: _____

Reason for starting Colistin Therapy: ☐ Failure of alternative therapy ☐ Infection with MDR organism ☐ Used empirically ☐ No clinical reason ☐ Other

Colistin Start Date _____ Colistin End Date _____ Total days of Colistin Therapy _____ Route of Administration: IV Nebs

Colistin dose and frequency prescribed/day: _____ First Course ☐ Repeat Course ☐

Hang Time Compliant:* Y N Loading dose prescribed: Y N

If yes, what dose? _____

Were cultures done? Y N If yes, Specimen type: Urine Blood Tracheal Aspirate Other If Yes, Infecting Organism Cultured: _____

Organism Sensitivity Profile:	Other antibiotic(s) prescribed with colistin:	MIC:
		MIC:
		MIC:
		MIC:
		MIC:
		MIC:

SCr Prior*: _____ Creatinine Clearance Day 1: _____ Total LOS in ICU (Days)*: _____

SCr Post: _____ Creatinine Clearance on final day: _____ Total LOS in Hospital (Days): _____

eGFR Prior*: _____

eGFR Post: _____ Overall Patient Outcome:

Was therapy de-escalated? Y N If yes, to which antibiotic? ☐ Discharged ☐ Demissed

Was Therapy stopped? Y N _____

*hang time compliance is defined as the time of antibiotic prescription to the time of administration within one hour. LOS= Length of Stay. SCr=Serum Creatinine. eGFR=Estimated Glomerular Filtration Rate

APPENDIX E

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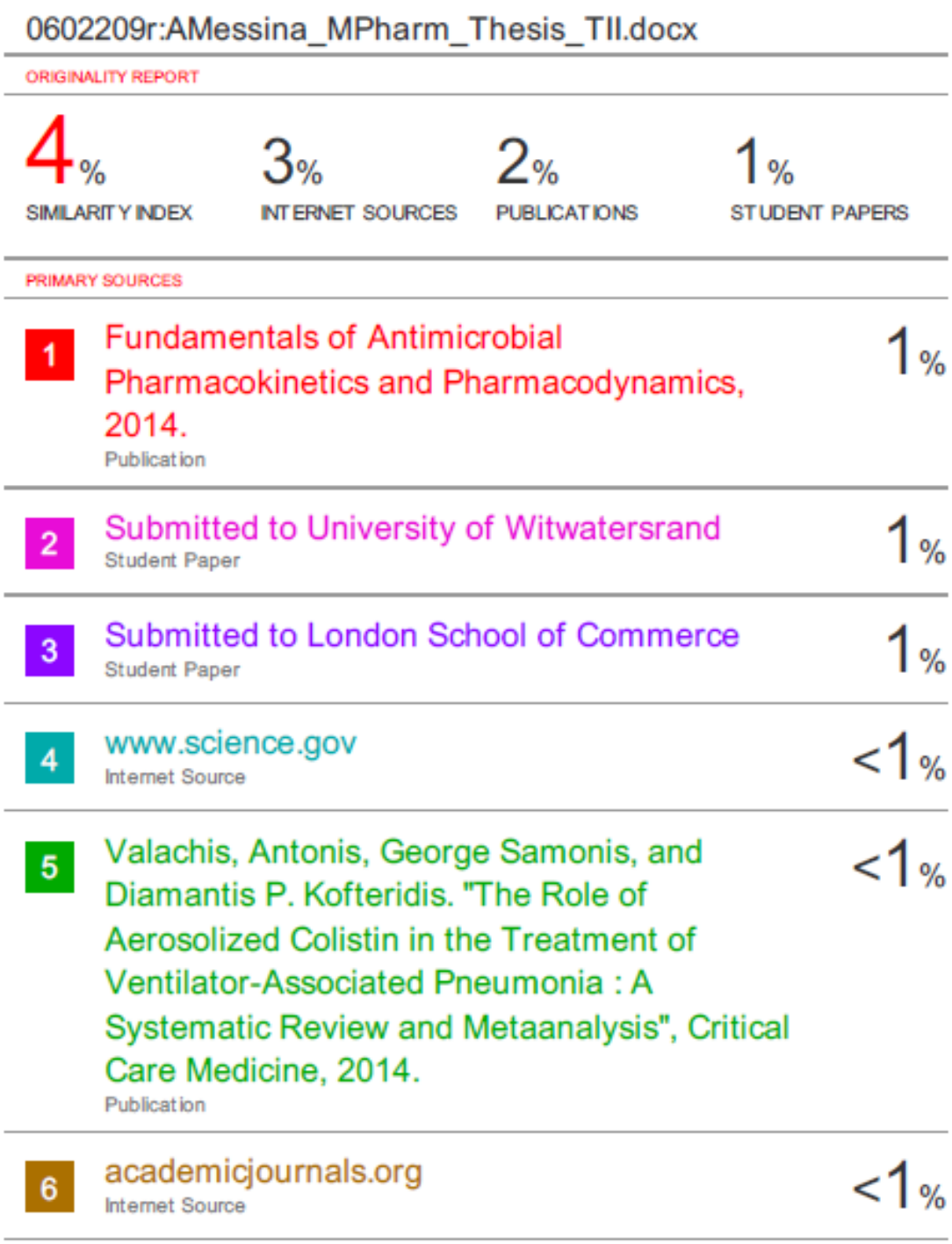

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