

**Safety and effectiveness of colistin compared with tobramycin  
for multi-drug resistant *Acinetobacter baumannii* infections**

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Dr Ronald Gounden

E-mail: [gounden@sun.ac.za](mailto:gounden@sun.ac.za)

Student number: GNDRON001

Supervisors: Prof Gary Maartens

Dr Karen Cohen

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## **Declaration**

I, Ronald Gounden, hereby declare that the work on which this dissertation is based is my original work (except where acknowledgements indicate otherwise). Neither the whole work nor any part of it has been, is being, or is to be submitted for another degree in this or any other university.

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## **Statement of authors' role**

The following describes the contributions of the author and co-investigators:

- The author wrote the protocol and contributed to the design of the study. Professor Gary Maartens, Dr Richard van Zyl-Smit and Dr Colleen Bamford also contributed to the design of the study. Professor Maartens and Dr Karen Cohen gave input to the final draft of the protocol.
- The author created the data capture sheet with input from the co-investigators.
- The author was responsible for the data collection. Dr Colleen Bamford assisted in this regard.
- The author was responsible for the analysis of the data and was assisted by Dr Karen Cohen and Prof Gary Maartens.
- The author wrote the report which follows and was supervised by Professor Maartens and Dr Karen Cohen.

Signature: 

Signed by candidate
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Date: 31/05/2009

### **List of abbreviations**

APACHE	Acute Physiology, Age and Chronic Health Evaluation
BNF	British National Formulary
CI	Confidence Interval
$C_{max}$	Maximum serum concentration
CVVHD	Continuous veno-venous haemodialysis
GFR	Glomerular filtration rate
GSH	Groote Schuur Hospital
HR	Hazard ratio
ICU	Intensive care unit
IQR	Interquartile range
IU	International units
MCC	Medicines Control Council
MIC	Minimum Inhibitory Concentration
SIRS	Systemic Inflammatory Response Syndrome
VAP	Ventilator-associated pneumonia

## **Abstract**

**Background:** Nosocomial infections due to multi-drug resistant *Acinetobacter baumannii* are often treated with colistin, but there are few data comparing its safety and effectiveness with other antimicrobials, particularly the aminoglycosides.

**Methods:** A retrospective cohort study of patients treated with colistin or tobramycin for *A. baumannii* infections in intensive care units (ICUs) at Groote Schuur hospital was performed. Colistin was used for *A. baumannii* isolates which were resistant to all other available antimicrobials. Tobramycin was used when the organism was susceptible to this antimicrobial. We assessed and compared ICU mortality, nephrotoxicity and time to the first negative culture in the colistin and tobramycin groups.

**Results:** 32 patients, with similar admission APACHE scores and serum creatinine levels, were treated with each antimicrobial. There was no significant difference between the colistin and tobramycin groups in ICU mortality ( $p=0.54$ ) but in-hospital mortality was higher in the colistin group than the tobramycin group ( $p= 0.04$ ). There was no difference in time to death ( $p= 0.09$ ), nephrotoxicity ( $p=0.67$ ), change in creatinine from baseline to highest subsequent value ( $p=0.11$ ) and time to microbiological clearance ( $p=0.75$ ) between the colistin and tobramycin groups.

**Conclusion:** Our study suggests that colistin and tobramycin have similar risks of nephrotoxicity and are equally efficacious. Colistin is an acceptable antibiotic for the treatment of *A. baumannii* infections when the organism is resistant to other available antimicrobials.

## **Introduction and literature review**

### **Introduction**

Colistin (Polymyxin E) has been available since 1959 for the treatment of gram-negative infections, although the polymyxins were first discovered in 1947.<sup>1</sup> However, in the 1970's, colistin fell into disuse because it was perceived to be unacceptably nephrotoxic.<sup>1</sup> Other classes of antimicrobials, such as the aminoglycosides and later the fluoroquinolones, cephalosporins and carbapenems became the preferred treatment for gram-negative infections.

In the last decade, multi-drug resistant gram-negative infections (particularly *Acinetobacter* and *Pseudomonas*) have emerged in intensive care units (ICUs) worldwide<sup>2,3</sup>. "Multi-drug resistant" is generally defined as resistance to three or more classes of antimicrobials which are normally used to treat these infections<sup>4</sup>. More worryingly, pan-resistant isolates of *Acinetobacter* and *Pseudomonas*, which are resistant to all conventional antimicrobials, have emerged throughout the world.<sup>2</sup>

Groote Schuur Hospital (GSH) is an 867-bed tertiary referral centre in Cape Town, South Africa, with 56 ICU beds (respiratory, general surgery and neurosurgery). Since 2002, pan-resistant *Acinetobacter baumannii*, resistant to piperacillin-tazobactam, ceftazidime, cefepime, imipenem, meropenem, gentamicin, amikacin, tobramycin and ofloxacin/ ciprofloxacin, has been isolated from GSH ICU patients (Oliver S, personal communication 2006). These organisms were found to be susceptible only to colistin.

This increase in resistance has not been accompanied by the development of new antimicrobials with gram-negative activity.<sup>2</sup> Tigecycline, a tetracycline analogue, was developed in the 1990's and registered for use by the USA's Food and Drug Administration in June 2005. It is the only new agent effective against resistant gram-

negative organisms, but has substantial gram positive activity as well.<sup>5,6</sup> It has activity against pan-resistant *A baumannii*, but is not yet available in South Africa, and tigecycline-resistant *A baumannii* has already been found in phase 3 clinical trials.<sup>5</sup>

Colistin is therefore the only antimicrobial available to treat pan-resistant gram-negative infections in South Africa and is the last line of treatment for the ever-increasing number of pan-resistant isolates of *Acinetobacter baumannii*.

However, colistin continues to be used with a degree of reluctance. There is ongoing concern about its' potential for nephrotoxicity, particularly when administered to critically ill patients. Many of these patients have other risk factors for renal dysfunction such as sepsis, hypotension or exposure to other potentially nephrotoxic agents. In addition, there is uncertainty about the correct dose of colistin. There are few studies exploring the pharmacokinetics of colistin and to date, there is no worldwide consensus on the optimal dose of colistin required to ensure effectiveness, while avoiding toxicity. At GSH, colistin in the form of colistemetate sodium (Colimycine® Aventis, Bellon, France) is administered at a dose of 2 million international units (IU) 8 hourly in patients with normal renal function. The manufacturer recommends 50 000 IU/ kg / day in 2- 3 divided doses<sup>7</sup>, which for a 70 kg adult amounts to 1.2 million IU 8hrly. The GSH practise is based on studies that have used up to 3 million IU 8 hourly, with toxicity reported at these dosing levels not markedly increased compared with the lower recommended doses.<sup>8,9</sup>

Colistin is not registered for use in South Africa. Special permission has to be obtained from the Medicines Control Council (MCC) of South Africa to import and use this antimicrobial. Colistin has therefore not been submitted to the MCC regulatory processes. The regulatory requirement therefore is that informed consent must be obtained, if possible, prior to administering colistin to a patient.

At GSH we use colistin methanesulphonate (Colymicine®) which is imported from France. The practical implication is that clinicians unfamiliar with this drug may find the package insert, which is written in French only, unhelpful.

The clinical and microbiological outcomes of patients treated with colistin in South African ICUs have never been formally studied and documented.

The aminoglycoside tobramycin was previously regarded as standard therapy for *Acinetobacter baumannii* infections at GSH. However by 2006, 41% of *A baumannii* isolates were resistant to tobramycin (Whitelaw A, personal communication September 18 2007). It remains the conventional antimicrobial most commonly used to treat susceptible *A baumannii* infections, since it is associated with the least resistance, apart from colistin. In many countries, including the United States, the carbapenems are used to treat multi-drug resistant *A baumannii* infections, but carbapenem-resistance at our hospital was 70% in 2006 (Whitelaw A, personal communication September 18 2007)

To the author's knowledge, there are no published studies directly comparing the effectiveness and toxicity of colistin with the aminoglycoside antibiotics. Furthermore tobramycin, like all aminoglycosides, is nephrotoxic. Although it is widely believed that colistin is more nephrotoxic than the aminoglycosides, this has not been previously been explored in a comparative cohort study.

### Literature review

#### **Multi-drug resistant and pan-resistant *Acinetobacter baumannii* infections**

*A baumannii* is a gram-negative coccobacillus which colonises the skin, wounds, respiratory and gastrointestinal tracts. The most common clinical manifestations of infection with this organism are ventilator-associated pneumonia and bacteraemia. The mortality rate associated with *Acinetobacter* bacteraemia has been estimated to be

between 17 and 46%.<sup>4</sup> Genitourinary, soft tissue, intracranial (particularly meningitis), ophthalmic and skeletal infections can also occur.

*A baumannii* infections tend to occur in ICUs and outbreaks have been associated with contaminated respiratory equipment. Risk factors for colonisation and infection with *A baumannii* include central venous catheterisation, tracheostomy, mechanical ventilation and exposure to fluoroquinolones, third generation cephalosporins or carbapenems.<sup>10</sup>

Seven percent of healthy adults and infants may have cutaneous colonization with *A baumannii* and the organism is ubiquitous<sup>10</sup>. It is therefore often difficult to distinguish colonization from infection. The decision to treat with an antimicrobial when *A baumannii* has been cultured is therefore made on clinical grounds, but culture of the organism from a normally sterile site, such as the bloodstream, is clearly an indication to commence antimicrobial treatment. Signs of the Systemic Inflammatory Response Syndrome (SIRS) (two or more of temperature greater than 38°C, white cell count greater than  $12 \times 10^9/L$ , respiratory rate greater than 20 breaths per minute and heart rate greater than 90 beats per minute), new radiological infiltrates and the presence of purulent sputum at the time *A baumannii* is cultured in the case of pneumonia, support the necessity for antimicrobial treatment.<sup>11</sup>

#### ***A baumannii* resistance to antimicrobials**

There has been a worldwide emergence of multi-drug resistant *A baumannii* in the last decade.<sup>2</sup> Whereas in the 1970's many classes of antimicrobials could be used to treat infections due to this organism, most *A baumannii* are now resistant to ampicillin, cefotaxime and chloramphenicol, with increasing resistance to amikacin, tobramycin and the carbapenems observed.<sup>10</sup> In the United States, carbapenem-resistant *A baumannii* increased from 9% in 1995 to 40% in 2004<sup>4</sup>. *A baumannii* may acquire numerous mechanisms of resistance including beta- lactamases, alterations in penicillin-binding proteins, changes in cell wall porins and aminoglycoside-modifying enzymes.<sup>10</sup>

Tigecycline, a glycycline antimicrobial, may be a therapeutic alternative, but the drug is not yet available in South Africa and resistance to this antimicrobial has already been described.<sup>5</sup>

To date, there has been only one confirmed colistin-resistant *A baumannii* infection at Groote Schuur Hospital, but colistin resistance has been documented in other countries.<sup>12</sup> In one hospital in Greece 6 patients were found to have colistin-resistant *A baumannii* between January 2006 and March 2007.<sup>13</sup>

### Colistin

Colistin (polymyxin E) is an antibiotic derived from *Bacillus colistinus*. It is active against a wide variety of gram-negative organisms, including *A baumannii*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Enterobacter aerogenes* and *Klebsiella pneumoniae*. It is not indicated for the management of *Proteus* or *Neisseria* infections<sup>14</sup>.

The drug is administered as colistimethate sodium which is less toxic than colistin when administered intravenously. Colistimethate is then hydrolysed to colistin, which has been shown to possess greater antimicrobial activity than colistimethate. Colistin is a cationic detergent which destroys the cytoplasmic membrane, leading to bacterial lysis. It has been shown to display concentration-dependent killing.<sup>12</sup> This means that the higher the antimicrobial concentration, the greater the rate and extent of bacterial killing.<sup>15</sup>

Colistimethate is renally eliminated. The proportion which is not eliminated is available for hydrolysis to colistin. The clearance of colistin itself is predominantly via non-renal mechanisms, which remain to be elucidated, and a smaller proportion is eliminated renally. In renal dysfunction, the dosing interval of colistimethate must be extended because a high concentration of colistimethate is available for a greater period of time for hydrolysis to colistin.<sup>14</sup>

Colistin may also be administered intrathecally for the management of ventriculitis or meningitis. Intrathecal doses are 60 000 IU once or twice daily via an external ventricular drain which is then blocked for 3 hours.<sup>16,17</sup>

### **Adverse effects of colistin**

Adverse effects associated with the use of colistin include hypersensitivity reactions, neuromuscular blockade and central nervous system toxicity manifesting as ataxia, blurred vision, coma, confusion, psychosis and seizures.<sup>18</sup> These adverse reactions are rare and have not been detected in published cohort studies, but in post- marketing surveillance. The major concern is colistin's potential for nephrotoxicity.

Nephrotoxicity due to colistin may present as proteinuria, haematuria or acute tubular necrosis. Like aminoglycoside-induced nephrotoxicity, this condition is thought to be generally reversible<sup>11</sup>. Increased renal tubular cell permeability, with resultant influx of ions and water leading to cell lysis, is thought to be the mechanism of renal injury. Nephrotoxicity has been shown in two separate uncontrolled studies to occur in 14% of ICU patients treated with colistin, but an incidence of up to 24% has been reported.<sup>8,9,19</sup>

The first of these studies prospectively assessed 21 patients who had received at least 7 days of intravenous colistimethate in the ICU. No patient had acute renal failure on admission to hospital. The median daily dose of colistimethate used was 6 million IU (range 3-9 million IU). Nephrotoxicity (not defined in this paper) was observed in 3 patients. There was a significant correlation between the cumulative dose of colistimethate received and the difference in serum creatinine between the start and end of treatment ( $r=0.6$ ,  $p=0.004$  by Spearman's test).<sup>8</sup> This suggests that dose and duration of colistin therapy may be factors predisposing to nephrotoxicity.

In the second study, nephrotoxicity was defined as an absolute increase in serum creatinine of greater than 1mg/dl. Twenty- four patients with gram-negative infections were treated with colistin sulfomethate sodium at doses of 3 million IU 8 hourly. Dose

adjustment was allowed in renal dysfunction. Of the 21 patients who were not receiving continuous veno-venous haemodialysis (CVVHD) at baseline, 3 patients developed nephrotoxicity. One of these patients subsequently required CVVHD.<sup>9</sup>

The problem with these estimates is that researchers have not used a standard definition of nephrotoxicity.

In addition, these studies cannot control for other causes of renal failure, for example the effects of sepsis, hypotension and exposure to other nephrotoxic drugs on the renal function of patients in ICU. In this regard, comparative studies of colistin versus other antimicrobials are informative as the comparator groups are often similarly exposed to the above confounders.

Four studies compared colistin with the carbapenems in ICU patients and found that nephrotoxicity was not different in the colistin and carbapenem groups.<sup>20,21,22,23</sup> These studies used different definitions of nephrotoxicity. Rios et al compared the proportion of patients in the colistin and carbapenem group who had an increase in serum creatinine to greater than 1.4mg/dl or required renal replacement therapy.<sup>20</sup>

Garnacho-Montero compared the number of participants who developed “renal failure” during antimicrobial therapy. This was defined as a reduction in creatinine clearance of greater than 50% or the need for renal replacement therapy. In patients with normal renal function at baseline an increase in serum creatinine to greater than 2mg/dl during treatment was also regarded as renal failure.<sup>21</sup>

Reina compared the mean change in creatinine from the beginning to the end of treatment in the colistin and carbapenem group<sup>22</sup>, while Kallel et al compared the mean serum creatinine at the end of antimicrobial therapy in the colistin and carbapenem groups.<sup>23</sup>

There has been no study which directly compared the aminoglycosides with colistin. In the study by Reina et al, 48% of patients in the non-colistin group received combination therapy including an aminoglycoside.<sup>22</sup> These investigators found no significant differences in the increase in mean creatinine between the colistin and non- colistin groups.

### **Concerns about the effectiveness of colistin**

Although the in-vitro activity of colistin has been demonstrated against *A baumannii*, the clinical effectiveness of colistin remains an area of concern for a number of reasons.

The optimal dose of colistin has never been defined and there is no universal consensus on the dose which should be used. Worryingly, different manufacturers of the same product advise different doses. The total recommended daily dose of Colomycin® (Dumex- Alharma A/S, Copenhagen, Denmark) for a 60kg person with normal renal function is 1-2 million IU 3 times daily. The recommended dose of Coly-Mycin M Parenteral® (Parkedale Pharmaceuticals, Rochester, USA) is almost double this amount.<sup>1</sup>

A study of peak serum colistin concentrations determined by bioassay conducted in the United Kingdom showed that only 37.6% of patients between the ages of 30 and 39 years were in the target peak range for colistin (10-15mg/L) as prescribed by the British National Formulary (BNF) when dosed according to BNF recommendations.<sup>24</sup> The BNF recommends an intravenous dose of 1-2 million units 8 hourly in adults<sup>25</sup> In children between the ages of 10 and 19 the situation was even worse, with only 8.8% of samples falling within the target range. The mean concentrations in these groups were low (9.9mg/L and 5 mg/L respectively).

This uncertainty about the correct dose probably stems from the lack of studies of the pharmacokinetic-pharmacodynamic relationships of colistin, because the drug was registered in the 1950's. The pharmacokinetic disposition of the drug in critically ill

suspension or serial dilutions thereof were plated without or with varying concentrations of colistin sulphate. Colonies were then counted at 48 hours after incubation. These population analysis profiles revealed that in 15 of these isolates, heteroresistant subpopulations had emerged. The authors warn that the colistin concentrations achieved in clinical practice are often lower than required for the early eradication of resistant subpopulations.<sup>12</sup> This may give rise to resistance and treatment failure and underscores the necessity for optimal dosing to prevent resistance and treat infections appropriately. This is especially crucial since therapeutic options beyond colistin are not available and so the development of colistin resistance represents a grave concern.

Indeed a subsequent in-vitro study has shown that only minimal bacterial killing occurred after the second and subsequent doses of colistin in heteroresistant strains of *A baumannii*. This occurred regardless of the dosing schedule simulated, including 5mg/kg of body weight per day of colistin base activity (roughly 1.5 million IU eight hourly of colistin methanesulphonate), a dose recommended by at least one manufacturer.<sup>27</sup>

#### **Lack of data on the pharmacokinetic disposition of colistin in renal dysfunction**

Weighed against the need to achieve adequate serum concentrations and prevent resistance is the concern that increasing the dose may lead to greater nephrotoxicity.

There is also great uncertainty with regard to dose adjustments of colistin in renal dysfunction. Again, several conflicting recommendations exist, but a definitive study of colistin pharmacokinetics in renal dysfunction is not available. As an example of this uncertainty, package inserts advise the extension of the dosing interval in patients receiving continuous veno-venous haemodialysis (CVVHD). This has led to dosing regimens of 36 hourly colistin, whereas a case report showed that the concentration of colistin had fallen below the MIC of the organism within 12 hours of administration of colistin in a patient receiving CVVHD.<sup>1</sup> This patient had cystic fibrosis and it is known that in patients with this disease the pharmacokinetics of antimicrobials are different, with lower blood concentrations and higher clearance rates (including non-renal

clearance) of antimicrobials. This is postulated to be due to the intracellular accumulation of antimicrobials in cystic fibrosis cells caused by an imbalance between endocytosis and exocytosis within these cells.<sup>28</sup>

Nevertheless, it is improbable that patients without cystic fibrosis will have pharmacokinetics so different to those of cystic fibrosis sufferers that colistin given 36 hourly could be appropriate. It is therefore clear that inappropriate dosing practises have arisen from the dearth of available pharmacokinetic data and this must be remedied.

One prominent antimicrobial therapy guideline advises the extension of the dosing interval according to the degree of renal impairment as follows: The dose is given 12 hourly if the GFR is between 50 and 90ml/min, 24 hourly if the GFR is between 10 and 50ml/min and 36 hourly if the GFR is less than 10ml/min. Anuric patients are to receive 1 million units after each episode of dialysis.<sup>29</sup> The origin of the information can be traced back to a package insert of a single manufacturer, with a supporting pharmacokinetic study unobtainable.

Ideally, therefore, therapeutic drug monitoring should be available for colistin. Several methods of determining colistin serum concentrations have been developed, including microbiological assays, thin layer chromatographic (TLC) methods and high performance liquid chromatography (HPLC).<sup>30,31,32</sup> Unfortunately HPLC methods require specialised equipment and expertise which are not available in many hospital laboratories. Furthermore, colistimethate is unstable and is hydrolysed in vivo and vitro into colistin but also a heterogenous mixture of various partially sulphated compounds, which possess some antimicrobial activity. This hydrolysis can occur even after the blood has been drawn. Thus the determination of colistin serum concentrations alone may not accurately reflect the serum concentrations of all antimicrobially active compounds. For these reasons as well as the absence of a commercially available reagent, therapeutic drug monitoring of colistin is not available in South Africa, whereas aminoglycoside monitoring is available and routinely used in patients with renal dysfunction.

The absence of therapeutic drug monitoring for colistin represents a major drawback when colistin is used, particularly in critically ill patients, many of whom are likely to have renal dysfunction.

Despite these theoretical concerns, comparative studies have not demonstrated a difference in mortality or poorer clinical outcomes associated with the use of colistin.<sup>33</sup> One study also compared the rates of microbiological eradication by colistin with imipenem-cilastatin and demonstrated no difference.<sup>21</sup>

### **Comparative studies of colistin**

A number of studies comparing colistin with other antimicrobials (mainly the carbapenems) have been published thus far.<sup>33</sup> A feature of this growing body of evidence is the heterogeneity of the definition of outcomes. The definition of nephrotoxicity, in particular, has been variable. The dose of colistin used also differs between the studies.

None of the studies report a sample size calculation or comment on the power of the study to detect a statistically significant difference in outcomes. It is therefore possible that a difference exists, but that the studies are underpowered to demonstrate this difference.

Furthermore, there has been no study which compared colistin monotherapy with aminoglycoside monotherapy for the treatment of gram-negative infections. A synopsis of comparative studies of colistin follows and table 1 summarises the study design and efficacy outcomes, while table 2 reviews the adverse effects reported in each trial.

**Table 1. Comparative studies of colistin and efficacy outcomes reported**

	<b>Study population</b>	<b>Colistin group</b>	<b>Comparator Group</b>	<b>Efficacy outcomes reported</b>
Garnacho-Montero et al <sup>21</sup>	35 patients with ventilator-associated pneumonia caused by <i>A baumannii</i>	21 patients  Up to 1.5 million units 8hrly in patients with normal renal function*	14 patients treated with imipenem-cilastatin  (6 of these also received sulbactam, amikacin or tobramycin)	Inpatient mortality was 61.9% in the colistin group and 64.2% in the control group (P=NS)  Clinical cure rates equal (57%)  Similar proportion of clinically cured patients tested became culture negative (66.7% colistin vs 50% control)
Rios et al <sup>20</sup>	61 patients with VAP due to <i>A baumannii</i> or <i>Pseudomonas</i>	31 patients received colistin at 1.5 million units 8hrly*	30 patients treated with imipenem or meropenem	Overall mortality was 51.6% in the colistin group and 45.1% in the control group (p=0.7)
Reina et al <sup>22</sup>	185 critically ill patients in ICU with an <i>Acinetobacter</i> or <i>Pseudomonas</i> infection	55 patients treated with colistin. Maximum dose 1.25 million units 8 hourly*	105 patients (81%) were treated with carbapenems and 25 (19%) with quinolones or piperacillin-tazobactam.  48% also received an aminoglycoside concurrently	The overall mortality in the colistin group was 34% versus 24% in the control group.  In patients with VAP, 2/29 in the colistin group had persistence of the microorganism after 7 days of therapy versus 5/86 in the tobramycin group.
Kallel et al <sup>23</sup>	<i>A baumannii</i> or <i>Pseudomonas</i> VAP and normal renal function	60 patients treated with colistin 2 million units 8hrly*	60 patients treated with imipenem-cilastatin	ICU mortality was 41.7% for colistin and 35% for imipenem (p=0.45).
Hachem et al <sup>34</sup>	Patients with malignancies and <i>Pseudomonas</i> infections. Outpatients were included	31 patients received colistin at 4.5 million units daily.* 8 patients received aminoglycosides concurrently	64 patients treated with other antipseudomonal antimicrobials	2.9 times greater clinical response rate in the colistin group (p=0.026) in the multivariate analysis

\*Colistin doses were reported as mg/kg of colistin methanesulphonate. Using a 70kg patient, the dose has been presented as international units, using the formula: 80mg= 1 million units of colistin

**Table 2. Adverse effects reported in comparative studies of colistin**

Study	Adverse effects reported
Garnacho-Montero et al <sup>21</sup>	Renal failure developed in 24% of colistin treated patients and 42% in the control group (p>0.05)  Neurophysiological studies in the colistin group revealed no neuromuscular blockade
Rios et al <sup>20</sup>	Two patients in each group with normal renal function at baseline developed abnormal renal function at day 5.
Reina et al <sup>22</sup>	In both groups, there was no significant increase in the mean creatinine after antimicrobial therapy.
Kallel et al <sup>23</sup>	No patients in either group developed renal failure.
Hachem et al <sup>34</sup>	The rates of nephrotoxicity was 23% in the control group and 22% in the non-colistin group

Garnacho- Montero compared 21 patients treated with colistin with 14 patients treated with imipenem-cilastatin for ventilator-associated pneumonia.<sup>21</sup> Inpatient mortality (62% in the colistin group and 64% in the imipenem-cilastatin group) and VAP-associated mortality rates were similar.

Clinical cure rates of VAP were 57% in each group. Of the 12 colistin treated patients regarded as clinically cured, 9 were tested for microbiological eradication. Six had become culture negative and 3 remained culture positive despite clinical improvement. In the imipenem- cilastatin arm, 8 were assessed as clinically cured. Of the 4 tested, 2 remained culture positive despite clinical improvement.

Four patients in the colistin group and 6 in the imipenem group developed renal failure. Renal failure was defined as an increase in serum creatinine to greater than 2mg/dL, a greater than 50% reduction in creatinine clearance or the need for haemodialysis.

Three patients in each group required haemodialysis. Electrophysiological studies did not detect neuromuscular blockade in the colistin group. This was a small study which failed to show a significant difference in any of the outcomes. Also 6 of the 14 patients who were treated with imipenem-cilastatin also received sulbactam, amikacin or tobramycin. The co-administration of aminoglycosides which are known to be nephrotoxic, in this group, should be borne in mind before concluding that colistin is no more nephrotoxic than imipenem-cilastatin.

Rios et al compared colistin with imipenem or meropenem for the treatment of VAP due to *Pseudomonas* or *Acinetobacter*.<sup>20</sup> The study included 31 colistin-treated patients and 30 carbapenem-treated patients. Overall 34 patients had normal renal function at baseline and of these, a similar proportion (2 per group) developed abnormal renal function at day 5 of treatment. The authors also found no difference in the overall mortality rate.

Reina et al studied 185 critically ill patients in ICU with an *Acinetobacter* or *Pseudomonas* infection, of which 55 were treated with colistin.<sup>22</sup> Of the 130 patients in the non- colistin group, 81% were treated with carbapenems, while some were treated with quinolones or piperacillin-tazobactam. Forty-eight percent of patients in the non-colistin group received adjunctive therapy with an aminoglycoside, although not as monotherapy. To the author's knowledge this study represents the only comparison of colistin-treated patients with a group in which many patients were receiving an aminoglycoside. The overall mortality in the colistin group was 34% versus 24% in the non- colistin group. This difference was not statistically significant. Of the 115 patients with VAP, 2/29 in the colistin group had persistence of the microorganism after 7 days of therapy versus 5/86 in the colistin group. Two patients in each group had renal failure at baseline and there was no deterioration in renal function in these patients. In neither the colistin nor the other-antimicrobial group was there a significant increase in the mean creatinine before compared to after antimicrobial therapy.

Kallel performed a matched case-control study of colistin versus imipenem for the treatment of VAP due to *A baumannii* or *P aeruginosa*.<sup>23</sup> Sixty colistin-treated patients were matched using their Simplified Acute Physiology Score (SAPS 2) and the PaO<sub>2</sub>/FiO<sub>2</sub> ratio to 60 controls. All included patients had to have normal renal function at baseline. ICU mortality was not different between the 2 groups, with rates of 42% for colistin and 35% for imipenem (p=0.45 for the difference). None of the participants in either group developed renal failure.

Hachem et al studied 95 patients with malignancies who had a multi-drug resistant *Pseudomonas aeruginosa* infection.<sup>34</sup> Outpatients were included. Thirty-one patients received a colistin-containing regimen either because the organism was pan-resistant or because they were not responding to other antipseudomonals (quinolones or beta-lactams), despite proven susceptibility. At baseline, the control group was significantly older than the colistin group and there were significant differences in the types of malignancies in each group. In addition, aminoglycosides were used in the colistin and non-colistin group. Given these limitations, the finding of a 2.9 (95% confidence interval 1.1 to 7.6) times greater clinical response rate in the colistin group (p=0.026) in the multivariate analysis should be interpreted with caution. The patient population is very different to critically ill patients in ICU and colistin was used for infections which were not pan-resistant. It is therefore difficult to extrapolate the findings of this study to patients in ICU with pan-resistant organisms.

### **Tobramycin**

Tobramycin is an aminoglycoside antibiotic which is used to treat a wide range of gram-negative infections. It binds to the 30S-subunit of bacterial ribosomes and inhibits bacterial protein synthesis. It displays concentration-dependent killing with a significant post-antibiotic effect.<sup>15</sup>

Tobramycin is administered intravenously in a dose of 5-6mg/kg once daily in patients with normal renal function. Dose adjustment is required in renal dysfunction.

Antimicrobial guidelines recommend that in renal dysfunction an initial dose of 3-4mg/kg is used and therapeutic monitoring used to guide the timing of subsequent doses. The trough concentration is usually allowed to fall below 1mg/L before the next dose is given. However, another antimicrobial may be considered if it takes more than 48 hours for the concentration to decrease sufficiently, since concentration-dependent killing may be impaired.<sup>35</sup>

Tobramycin, like all aminoglycosides, is nephrotoxic. Nephrotoxicity has been shown to be associated with higher doses, increased duration of therapy and high peak and trough concentrations. Patient factors such as pre-existing renal dysfunction, increasing age, co-existent liver disease (particularly biliary obstruction), hypovolaemia, potassium and magnesium depletion and sepsis, predispose to nephrotoxicity. Loop diuretics, ACE-inhibitors, cephalosporins and non-steroidal anti-inflammatories potentiate aminoglycoside-induced nephrotoxicity.<sup>36</sup>

Aminoglycosides disrupt the membrane permeability of the proximal tubular cells. This leads to intracellular accumulation of aminoglycoside in the renal cortex. Direct toxicity to tubular epithelial cells is mediated by inhibition of lysosomal activity with accumulation of myeloid bodies within these cells.<sup>37</sup>

In a study of once daily administration of gentamicin and tobramycin in ICU patients, 14% of participants were reported to have a rise in creatinine of 45 µmol/mL or more.<sup>36</sup> This is consistent with the findings of a meta-analysis of 24 controlled trials which showed that the average rate of nephrotoxicity of tobramycin is 11.5%.<sup>36</sup> These estimates are not dissimilar to the estimated incidence of the nephrotoxicity of colistin.

Nephrotoxicity is usually reversible and may present as acute tubular necrosis or non-oliguric renal failure. It usually starts after a few days of treatment. Loss of tubular function usually manifests as a progressive increase in serum creatinine, but sudden deteriorations may occur.<sup>36</sup>

Tobramycin also causes neuromuscular blockade, ototoxicity, and, rarely, hypersensitivity reactions.<sup>36</sup>

## **Aims and objectives**

### **Aim**

To compare the safety and effectiveness of colistin and tobramycin for the treatment of *Acinetobacter baumannii* infections in ICUs

### **Objectives**

A. To determine whether colistin is more nephrotoxic than tobramycin in patients with *Acinetobacter baumannii* infections in ICUs.

### **Endpoints**

- The absolute increase in creatinine from baseline (the day before colistin or tobramycin was commenced) to the highest subsequent recorded value within 10 days of initiation of the antimicrobial, in those patients who were not receiving haemodialysis at baseline.
- The proportion of participants not receiving dialysis at the start of antimicrobial therapy who subsequently required dialysis in each group.
- The proportion of participants with normal renal function at baseline who had an increase in serum creatinine to greater than 50% above the upper limit of normal of the local reference values (100 µmol/ml in females, 120 µmol/ml males)

B. To determine whether there is a difference in mortality rates between patients who are treated with colistin and tobramycin for *Acinetobacter baumannii* infections in the ICU.

#### Endpoints

- The proportion of patients in each group who die while in the ICU.
- The proportion of patients in each group who die while in hospital

C. To determine whether the rates of eradication of *Acinetobacter baumannii* in patients treated with colistin and tobramycin are different.

#### Endpoints

- The proportion of patients in each group who achieve microbiological clearance (defined as two or more consecutive negative cultures for *Acinetobacter baumannii* from all sites sampled), within 10 days of initiation of the antimicrobial, with no subsequent positive cultures. The time to clearance was calculated as the time interval between antimicrobial initiation and the first negative culture.

### **Study design and research methods**

#### **Study design**

A cohort study of patients treated with colistin and tobramycin for the management of *A. baumannii* infections between 2003 and 2005.

A retrospective chart review was performed. The cohort was chosen based on exposure to colistin and tobramycin and their records followed prospectively to determine clinical outcomes. Thus, although the study sample was assembled retrospectively, the

fundamental criteria for a cohort study were met. These are: selection of the sample based on exposure, prospective follow up and assessment of outcomes in each exposure group.

### Population and sampling

Pharmacy records were used to identify all patients treated with colistin or tobramycin for *A baumannii* infections in Groote Schuur Hospital ICUs between January 2003 and December 2005. Any patient who received a dose of colistin or tobramycin was eligible for inclusion in the study.

### Study documentation

A standardised data capture form (Appendix A) was used to capture information about the hospital admission during which the patients received either tobramycin or colistin. Data was obtained from review of patient medical records, ICU records, prescription charts and nursing vital sign observation charts.

The Acute Physiology, Age and Chronic Health Evaluation II (APACHE II) score was calculated for each patient at the time of admission to ICU. This score is used to quantify the severity of a patient's illness and determine the prognosis of critically ill patients. It is widely used in clinical studies to compare the severity of illness of different groups.<sup>38</sup> The scoring system includes clinical and laboratory parameters, the age of the patient and the reason for admission to ICU (emergency or non-emergency).

Baseline demographics, chronic disease, clinical course in ICU and other antimicrobials used were also recorded. Microbiology, haematology and chemical pathology results were obtained from the Groote Schuur National Health Laboratory Services electronic database.

## Methods

### **Susceptibility testing**

At Groote Schuur Hospital, *A baumannii* is tested for susceptibility to piperacillin-tazobactam, ceftazidime, cefepime, imipenem, meropenem, gentamicin, amikacin, tobramycin, ofloxacin/ ciprofloxacin and colistin. For all antibiotics except colistin, susceptibility testing is performed using the Kirby-Bauer disc diffusion method on Mueller Hinton agar (Oxoid).

A disc diffusion test is not available for colistin. Minimum inhibitory concentrations for colistin are determined by E-test (AB Biodisk, Solna, Sweden) according to the manufacturer's instructions. Clinical Laboratory Standards Institute guidelines are used in the interpretation of susceptibility testing.

### **Distinguishing infection versus colonization with *A baumannii***

The following were regarded as criteria to distinguish infection from colonization with *A baumannii*: positive culture from a sterile site (blood or cerebrospinal fluid) or culture from other sites associated with a clinical sign of infection (a temperature greater than 38.0°C, white cell count greater than  $12 \times 10^9/L$ , new chest x-ray infiltrates, the presence of purulent sputum/tracheal aspirate or the necessity for inotropic support) documented within 48 hours of commencing the antimicrobial.<sup>11,39</sup>

Sepsis was defined as culture positivity accompanied by 2 or more of the following signs: temperature of greater than 38°C, heart rate greater than 90 beats per minute, respiratory rate greater than 20 breaths/ minute or white cell count greater than  $12 \times 10^9/L$ .<sup>39</sup>

At Groote Schuur Hospital, the decision to treat a culture of *Acinetobacter baumannii* is made by the ICU clinician in consultation with a clinical microbiologist.

#### **Dose of colistin and tobramycin used**

At Groote Schuur Hospital, colistin (Colimycine® Aventis, Bellon, France) is administered at a dose of 2 million units 8 hourly in patients with normal renal function. Colistin dosage is adjusted for renal failure as follows:<sup>40</sup>

Creatinine clearance (ml/min)	Dosing regimen
50-90	2 million units 12 hourly
10- 50	2 million units 24 hourly
<10	2 million units 36 hourly
Anuric	1 million unit after each episode of dialysis

Tobramycin is given in doses of 5-6 mg/kg daily in patients with normal renal function. In patients with a creatinine clearance of less than 60ml/minute, an initial dose of 3-4mg/kg was administered with further dosing according to tobramycin plasma concentrations.

#### **Statistical analysis**

Data analysis was performed using Intercooled STATA™ version 8.2 (Statacorp, College Station, Texas). Continuous variables were summarised using means and standard deviations if normally distributed, and medians and ranges if not normally distributed. The Shapiro-Wilk test was used to determine whether continuous data was normally distributed. Ninety-five percent confidence intervals (CIs) or interquartile ranges (IQRs) were calculated for all summary statistics and parameter estimates. Between-group comparisons of continuous data were performed using a Student's t test if normally distributed, and the Wilcoxon rank sum test if not normally distributed.

Proportions were compared using a two sample test of proportions when the observed frequencies were  $\geq 5$  in each group and the Fisher's exact test if the expected frequencies were  $< 5$ .

To calculate 95% confidence intervals for the change in creatinine from baseline to the highest subsequently recorded value for the colistin and tobramycin groups, the difference in creatinine for each patient was first subjected to logarithmic transformation. This created a normal distribution of the log transformed data, allowing the calculation of geometric means and confidence intervals.

Kaplan-Meier curves were plotted for time to death in hospital, time to death in ICU and time to microbiological clearance. The date of initiation of the antimicrobial was used as the starting point for each patient and patients were censored when they were discharged from ICU for the analysis of death and for death in the case of microbiological clearance. Kaplan-Meier curves were compared with a log-rank test. Hazard ratios were calculated using a Cox proportional hazards model. For all statistical analyses, a p value of less than 0.05 was regarded as sufficient evidence to reject the null hypothesis.

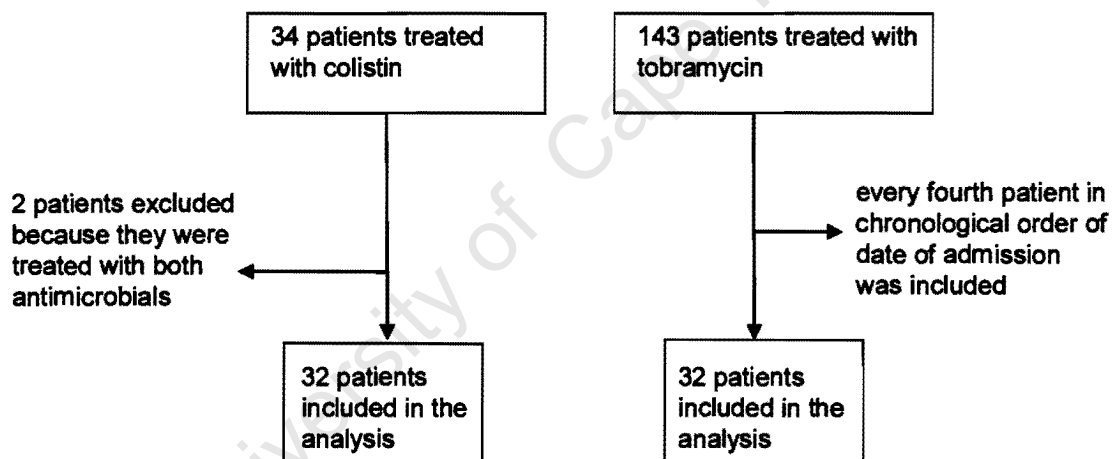
#### Ethics considerations

The study was approved by the Human Research Ethics Committee of the University of Cape Town. Only researchers involved in this study had access to data collection forms and the electronic database used for analysis. In the dissemination of results, no patient identifying features (hospital record numbers or patient names) were used.

## Results

Figure 1 shows the study profile. Pharmacy records identified 34 patients who had been treated with colistin and 143 with tobramycin for *A. baumannii* infections. Two patients had received both colistin and tobramycin during their ICU stay and were excluded from the analysis. Thirty-two patients remained who had only been treated with colistin.

Every fourth patient who received tobramycin, in order of date of admission to ICU, was then included to create a tobramycin group with an equal number of participants as the colistin group. We therefore included 64 patients (32 in each antimicrobial group).



**Figure 1. Study profile**

## Baseline characteristics

The baseline characteristics in the colistin and tobramycin groups were similar with respect to mean age, mean APACHE score at ICU admission and the number of patients with chronic diseases at the time of admission to ICU (table 1). Baseline chronic diseases included diabetes mellitus (2 patients), HIV (2 patients), atrial fibrillation (1 patient), ventricular septal defect (1 patient), hypertension (1 patient), and chronic

obstructive airways disease (2 patients) in the colistin group. In the tobramycin group patients had hypertension (4 patients), diabetes mellitus (2 patients), ischaemic heart disease (1 patient), previous nephrectomy (1 patient), rectal carcinoma (1 patient), liver cirrhosis (1 patient), epilepsy (1 patient) and HIV (one patient).

The median serum creatinine at the time the antimicrobial was commenced was higher in the colistin group (76  $\mu\text{mol/L}$ ) than in the tobramycin group (56  $\mu\text{mol/L}$ ), but this difference was not statistically significant (Wilcoxon rank sum test  $p=0.08$ ) Three patients in the colistin group and 5 in the tobramycin group were on haemodialysis at the time the antimicrobial was commenced and these proportions were not significantly different (Fishers exact test  $p=0.7$ ).

Patients received a median of 8 days of colistin (interquartile range (IQR) 5 to 13) and 7 days of tobramycin (IQR 6 to 10) (Wilcoxon rank sum test  $p = 0.72$ ).

Ten patients in each group (31%) had bloodstream infections with *A baumannii*. *A baumannii* was cultured in similar proportions from the sputum, wound pus swabs, central venous catheter tips and urine of the colistin and tobramycin groups (See table 1). The organism was cultured from the cerebrospinal fluid of one patient in the colistin group.

*Acinetobacter baumannii* was cultured from multiple sites in 17 (53%) patients in the colistin group and 20 (63%) patients in the tobramycin group ( $p=0.54$ ).

**Table 3: Baseline patient characteristics**

	Colistin (n=32)	Tobramycin (n=32)	P value
Number of patients with chronic diseases at the time of admission (%)	9 (28)	12 (37.5)	0.65
Median duration of treatment (IQR)	8 (5 to 13)	7 (6 to 10)	0.72
Mean age in years ( $\pm$ SD)	43.5 ( $\pm$ 15.6)	45.6 ( $\pm$ 18.2)	0.69 $\ddagger$
Mean APACHE score at ICU admission ( $\pm$ SD)	14.4* ( $\pm$ 5.1)	14.8** ( $\pm$ 5.4)	0.77 $\ddagger$
Dialysis at baseline (%)	3 (9.4)	5 (15.6)	0.60 $\dagger$
Median baseline creatinine (IQR) $\mu$ mol/L	76 (53 to 128)	56 (45 to 91)	0.08*
Site of <i>A. baumannii</i> isolation:			
Bloodstream infection	10 (31%)	10 (31%)	1.00=
Sputum/tracheal aspirate	22 (72%)	28 (88%)	0.13 $\dagger$
Wound pus swab	6 (18.8%)	11 (34.4%)	0.75 $\circ$
Cerebrospinal fluid	1 (3.1%)	0	NA
Central venous catheter tip	10 (31%)	9 (28%)	0.44=
Urine	5 (15.6%)	4 (12.5%)	1.00 $\dagger$

\*data available for 25 participants

\*\*data available for 21 participants

$\dagger$  Fisher's Exact Test

$\ddagger$  Two sample t- test

$\circ$  Wilcoxon rank sum test

= Two sample test of proportion

All, except one subject in each group, met at least one of the criteria for infection rather than colonization with *A. baumannii* documented in their records. Twenty patients (63%) in the colistin group and 22 (69%) patients in the tobramycin group met the criteria for sepsis (culture-positivity in conjunction with 2 or more components of the Systemic Inflammatory Response Syndrome)<sup>38</sup> at the time of initiation of the antimicrobial.

All the isolates in the colistin group were resistant to all other antimicrobials tested. All the isolates in the tobramycin group were susceptible to tobramycin. In 17 patients in the

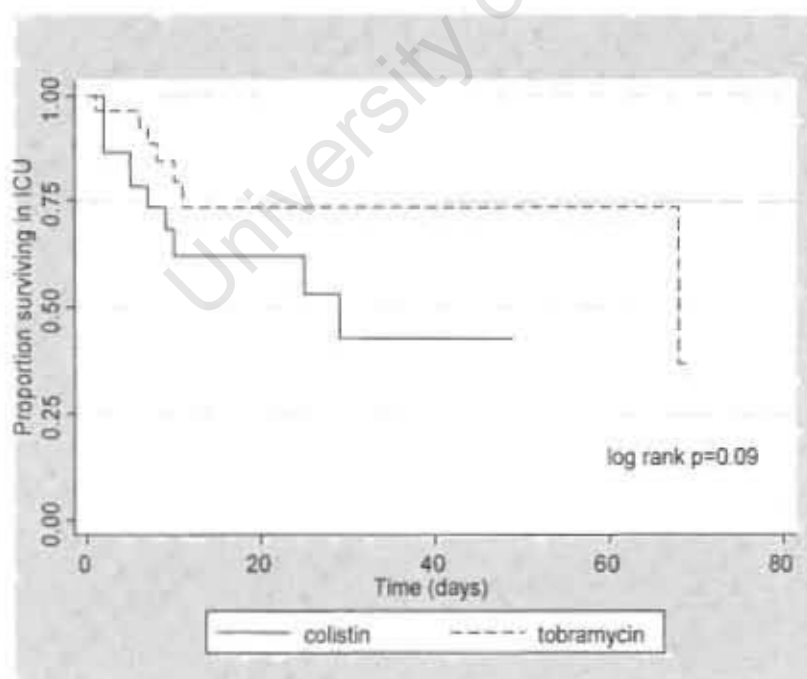
tobramycin group (53%), the organism was only susceptible to tobramycin and colistin. Carbapenem resistance was present in 24 (75%) of the tobramycin group.

### Clinical and bacteriological outcomes

#### **Duration of hospitalization and survival**

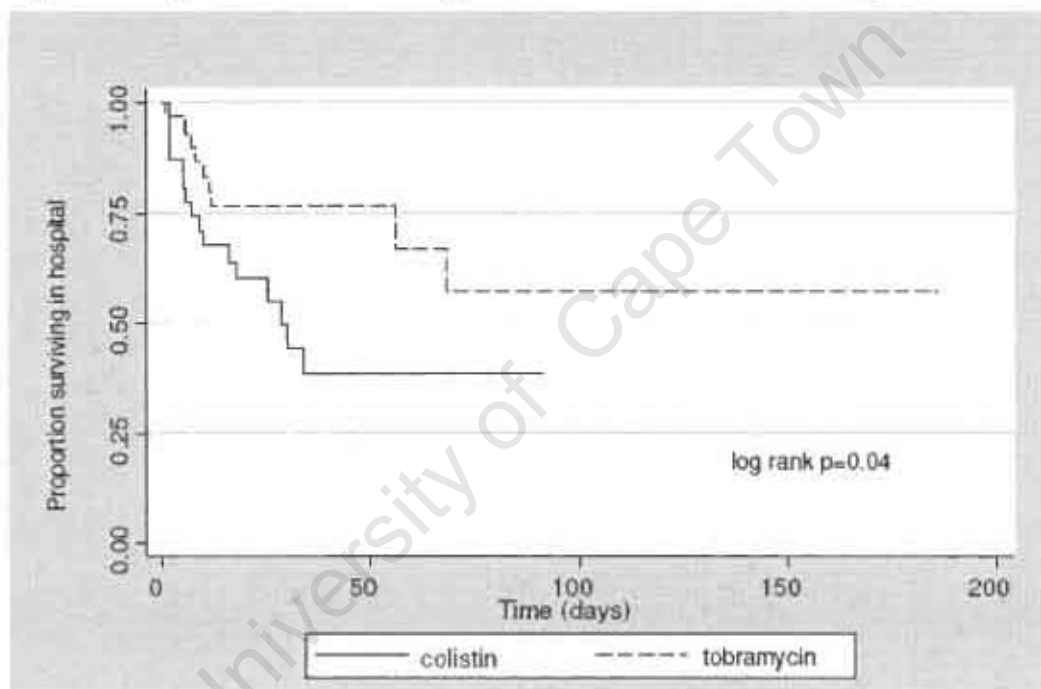
The median length of ICU stay after initiation of the antimicrobial was 6 days (IQR 4 to 21) in the colistin group and 9 days (IQR 4 to 21) in the tobramycin group (Wilcoxon rank sum test  $p = 0.06$ ). Eleven colistin-treated patients (34.4%) and 7 tobramycin-treated patients (21.9%) died in ICU ( $p = 0.54$ ). There was no significant difference in time to death in ICU by Kaplan–Meier survival analysis (log rank  $p = 0.09$ ) (figure 2). The hazard ratio for ICU survival in patients treated with colistin compared with tobramycin was 0.44 (95% CI 0.16 to 1.19).

**Figure 2. Kaplan- Meier ICU survival curves by antibiotic.**



The median length of total hospital stay after initiation of the antimicrobial was 18 days (IQR 7 to 34) in the colistin group and 27 days (IQR 12 to 56) in the tobramycin group ( $p= 0.08$ ) Sixteen patients treated with colistin (50%) and 9 patients treated with tobramycin (28.1%) died in hospital. The hazard ratio for total in-hospital survival in patients treated with colistin compared to tobramycin was 0.43 (95% CI 0.19 to 0.99) (logrank  $p=0.04$ ) (see figure 3).

**Figure 3. Kaplan- Meier curves by antimicrobial for total in-hospital survival**

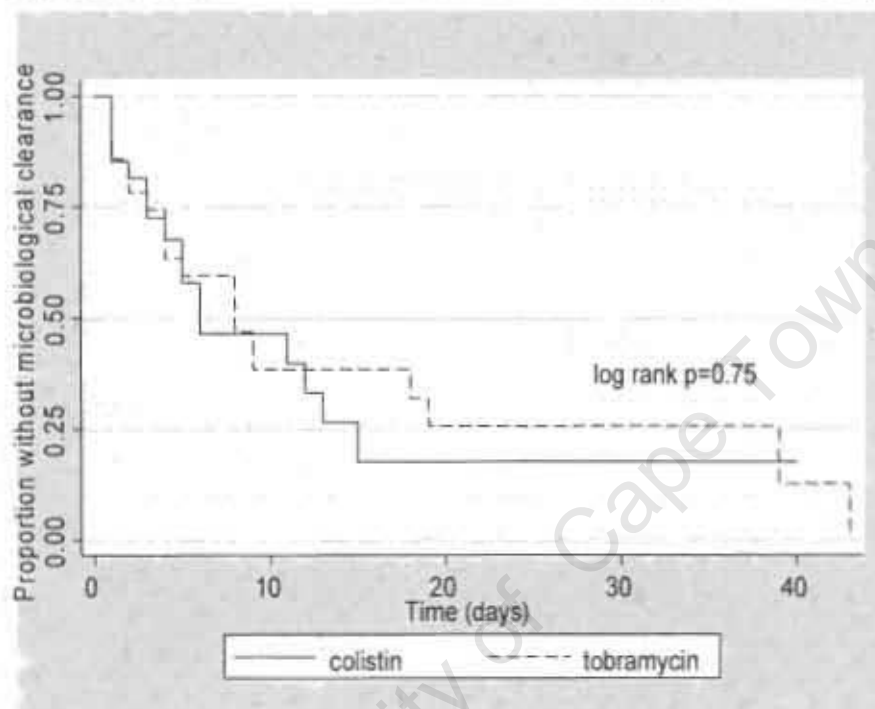


### Bacteriological clearance

Bacteriological eradication was documented in 16 (50%) patients treated with colistin and 17 (55%) treated with tobramycin (two sample test of proportion  $p= 0.79$ ). The median time to clearance of *A baumannii* was 3 days for colistin and 4 days for tobramycin (Wilcoxon rank sum test  $p = 0.46$ ). There was no significant difference in time to microbiological clearance by Kaplan-Meier analysis (figure 4) between the

colistin and tobramycin groups (log rank  $p = 0.75$ ). The hazard ratio for microbiological persistence in patients treated with colistin compared with tobramycin was 0.90 (95% CI 0.46 to 1.76).

**Figure 4. . Kaplan- Meier curves for microbiological clearance by antimicrobial.**



#### **Adverse effects**

There was a modest increase in creatinine from baseline to highest recorded within 10 days of antimicrobial commencement in both groups, with a median increase of 28  $\mu\text{mol/L}$  (IQR 11 to 135) in those on colistin and 17  $\mu\text{mol/L}$  (IQR 6 to 22) in those on tobramycin (Wilcoxon rank sum test  $p = 0.11$ ). The geometric mean change in creatinine in the colistin group was 42  $\mu\text{mol/L}$  (95% CI 24.4 to 74.9) and 19.6  $\mu\text{mol/L}$  (12.1 to 31.6) in the tobramycin group.

Only one participant in the cohort required initiation of haemodialysis after antimicrobial therapy was commenced, a male patient with a baseline serum creatinine of 118  $\mu\text{mol/L}$ . He started dialysis 6 days after colistin initiation.

The proportion of patients with normal renal function at baseline who increased their creatinine concentrations to greater than 50% above the upper limit of normal were similar in the two groups: 2 of 23 (8.7%) in the tobramycin group and 4 of 21 (19%) in the colistin group (Fishers exact test  $p=0.67$ ).

No other adverse effects were documented.

### **Outcomes in patients with bloodstream infections**

Ten patients in each group had bloodstream infections. Colistin was used for a mean of 13 days (IQR 2 to 12.5) and tobramycin 7.5 days (IQR 3.5 to 11.5) for bloodstream infections (Wilcoxon rank sum  $p=0.25$ ). Five patients in the tobramycin group (50%) and 3 patients (30%) in the colistin group with bloodstream infections died in ICU (two sample test of proportion  $p=0.58$ ).

Six patients with bloodstream infections in each group (60%) were demonstrated to have achieved sustained microbiological clearance.

## **Discussion and conclusion**

### **Summary of findings**

#### **Nephrotoxicity**

The primary safety endpoint was the change in creatinine concentration from the start of antimicrobial therapy to the highest value recorded within 10 days. In both the colistin and tobramycin group, there was a modest increase in serum creatinine concentrations. This is despite the fact all patients were treated with colistin and tobramycin in ICU and likely to have multiple risk factors for renal dysfunction. The proportion of patients with normal renal function at baseline who increased their creatinine concentrations to greater than 50% above the upper limit of normal was 8.7% in the tobramycin group and 19% in the colistin group. The incidence of nephrotoxicity of colistin in our study is consistent with the 14-24% incidence reported in previous studies.<sup>21</sup>

We found that there was no difference in the median increase in creatinine concentrations between the colistin and tobramycin groups. However, our sample size may have been too small to detect a difference in this outcome. This will be discussed in more detail in the section which deals with limitations of the study.

#### **Mortality**

We found no significant difference in ICU survival between colistin and tobramycin treated patients. This is in keeping with the results of other studies which have compared colistin with other antimicrobials, mainly the carbapenems. However, patients treated with colistin had statistically significantly higher in-hospital mortality than those treated with tobramycin. This is the first study to demonstrate higher mortality in colistin treated patients than in patients treated with other antimicrobials.

There are several possible explanations for this finding. The study was not powered to demonstrate a difference in the effectiveness of colistin and tobramycin in terms of microbiological clearance. Thus, colistin may have been less effective than tobramycin and this may have contributed to the higher mortality rate seen in this group.

The mean APACHE II scores at the time of admission to ICU were similar in the colistin and tobramycin groups and there were no significant differences in the baseline characteristics we have recorded. However, the study population was heterogenous as all patients admitted to ICU for any reason were included. It is possible that the groups were not identical in all prognostically important respects, which may have not been accounted for. This is a problem with studies which rely on medical records where not all possible prognostic factors may have been recorded by attending doctors.

In addition, a significant p-value does not guarantee that a finding is not due to chance. There is a small (less than 5%) probability that the finding of higher mortality in the colistin group was due to chance.

ICU mortality may be a more important measure than hospital mortality as the ICU admission was the time period within which an *A baumannii* infection was acquired and patients were exposed to colistin or tobramycin. In-hospital mortality may be more likely than ICU mortality to be influenced by non-*Acinetobacter* infection related factors, such as the initial reason for admission to ICU. Our primary effectiveness outcome was therefore ICU mortality, with a secondary exploration of total in-hospital mortality.

The Kaplan-Meier curves for ICU mortality showed a trend towards higher mortality in the colistin group (hazard ratio for ICU survival 0.44, 95% CI 0.16 to 1.19, logrank  $p=0.09$ ) which was non-significant. Some patients who survived the ICU died in the wards and it is possible that the greater number of mortality events may have led to a significant result, which was not seen in the ICU-only analysis.

We can therefore not discount the finding of higher mortality in the colistin group and further studies may cast light on the reason for our finding.

### **Microbiological clearance**

We found no difference in the time to microbiological clearance and the proportion of patients in whom microbiological clearance was achieved.

### **Limitations of the study**

#### **Small sample size**

The major limitation of this study is the small sample size. We included all patients treated with colistin between 2003 and 2005 and compared them with an equal number of patients treated with tobramycin. As colistin is only prescribed when *A baumannii* is resistant to all other available antimicrobials, the number of patients treated in this time period was small.

This will have influenced the power of the study to detect a statistically significant difference in the study endpoints. The primary safety outcome was the magnitude of the change in creatinine concentration in those patients who were not receiving dialysis. There was a high degree of variability in the change in creatinine in the colistin group, which was demonstrated by a very high standard deviation.

A power calculation requires standard deviations for each group and therefore assumes normal distribution of the data.<sup>41</sup> The change in creatinine was not normally distributed. Nonetheless, we performed a power calculation for our study, cognizant of the fact that a power calculation is likely to underestimate power when data is non-parametric.

We therefore estimated that our study had only 35% power to demonstrate, at the 5% level of significance, a difference in the mean change in creatinine between the colistin and tobramycin groups. The implication therefore is that a difference in the creatinine change may exist but that we were underpowered to detect this difference.

Published studies are of a similar size to ours and are therefore also likely to be underpowered.

### **Limitations of performing a retrospective study**

We obtained clinical information from patient medical records. This resulted in some missing data. Laboratory information could reliably and consistently be obtained from the NHLS electronic database, which meant that serum creatinine and culture results were available for all patients. However, information such as the presence of new chest x-ray changes or purulent sputum may not always have been documented in the medical records.

As a result of the incompleteness of medical records we could not report on all clinical outcomes of infection with *A baumannii*. For example, we could not reliably ascertain whether or not a patient had clinical or radiographic resolution of ventilator-associated pneumonia. We were therefore restricted to comparing rates of microbiological eradication.

In an observational cohort study, both known and unknown confounders may influence results. The investigator is able to adjust for these variables only if he is able to measure them. In a retrospective study, the investigator is limited in what he can measure and therefore adjust for.

In the ICU setting, sepsis, hypotension and the use of other nephrotoxic drugs contribute to impairment of renal function. We were not able to exclude the confounding impact of

these variables because of our reliance on sometimes incomplete medical records and the complex nature of treating patients in ICU.

### Strengths of the study

This is the first study which directly compares the efficacy and safety of colistin with tobramycin as monotherapy for *A baumannii* infections. It is therefore relevant to our hospital where patients are most likely to receive colistin or tobramycin for *A baumannii* infections.

The comparative nature of the study is a strength. The colistin and tobramycin groups were treated in the same ICU's over the same time period and the groups were similar with regard to baseline patient characteristics. The groups only differed with regard to susceptibility to tobramycin and therefore exposure to tobramycin or colistin. This design allows a meaningful comparison of the efficacy and safety of the drugs.

This is a cohort study and therefore has the strengths inherent in this type of study. The most important of these is the clarity of the temporal relationship between exposure to colistin or tobramycin and outcomes which follow. This temporal sequence is an important tenet of the assumption of causality. Thus, an increase in creatinine after commencing colistin or tobramycin is likely to be attributable to the exposure to the antimicrobial.

### Implications for clinical practise

Our data suggests that colistin is an effective and reasonably safe treatment for infections due to multi-drug resistant nosocomial organisms like *A baumannii* when the organism is resistant to other available antimicrobials. A therapeutic drug monitoring service with a standardised reference range for peak and trough concentrations would be of benefit in patients with renal dysfunction.

### Implications for future research

Further studies comparing colistin with aminoglycosides are still needed, particularly in light of our finding of higher in-hospital mortality in the colistin group.

A larger study of colistin versus an aminoglycoside would be ideal, but most comparative studies thus far have been small. Thus, ultimately, a meta-analysis when similar studies have been performed may provide the required statistical power.

Comparative studies have employed a variety of endpoints and ensuring that the outcomes are sufficiently homogenous to conduct a sound meta-analysis will be important. A prospective multi-centre study may be the solution since harmonization of study procedures and outcomes will ensure homogeneity.

An added advantage of a prospective study would be the ability to reliably record all pertinent patient exposures and outcomes, so that all possible confounders may be recorded and accounted for.

In the meantime, the reality is that regardless of our beliefs about the toxicity of colistin, we are forced to use this agent for the treatment of pan-resistant *A baumannii* infections. We therefore need to learn to use the drug more safely and effectively. To this end, studies of the pharmacokinetics of colistin in renal dysfunction are required to rationalise dosing and dose adjustments in renal impairment.

Further pharmacokinetic-pharmacodynamic studies in which serum concentrations achieved by varying doses of colistin are compared to the MIC of the infecting organism are required so that the optimal dose of colistin can be determined. This is important not only for optimal effectiveness of the drug but also to prevent colistin resistance.

### Conclusion

Our data suggests that colistin is not significantly different to tobramycin in terms of efficacy or nephrotoxicity and that it is an acceptable treatment of *A baumannii* infections when the organism is resistant to other available antimicrobials. However we demonstrated higher in-hospital mortality in the colistin group and further studies with larger sample sizes, or the application of meta-analytic techniques to other studies of aminoglycosides versus colistin, are required to definitively demonstrate non-inferiority of colistin.

University of Cape Town

## **References**

1. Li J, Nation R, Turnidge J, Milne R, Coulthard K, Rayner C, Paterson D. Colistin: the re-emerging antibiotic for multi-drug resistant Gram-negative bacterial infections. *Lancet Infect Dis* 2006; 6:589-601
2. Siegel R. Emerging Gram- Negative Antibiotic Resistance: Daunting Challenges, Declining Sensitivities, and Dire Consequences. *Respir Care* 2008;53(4):471-9
3. Berlana D, Llop J, Fort E, Badia M, Jodar R. Use of colistin in the treatment of multiple- drug –resistant gram- negative infections. *Am J Health – Syst Pharm* 2005; 62: 39-47
4. Munoz-Price L, Weinstein R. Acinetobacter infection. *N Engl J Med* 2007; 358:1271 -81
5. Livermore D. Tigecycline: what is it and where should it be used. *Antimicrob. Agents Chemother* 2005; 56: 611-14
6. Breedt J, Teras J, Gardofskivs J, Maritz F, Vaasna T, Ross D et al. Safety and efficacy of tigecycline in treatment of skin and skin-structure infections: Results of a double- blind phase 3 comparison study with vancomycin-aztreonam. *Antimicrob Agents Chemother* 2005;49:4658-66
7. Colimycine package insert information. Colimycine 1 000 000IU. Bellon, France: Aventis; 1997 Nov
8. Falagas M, Fragoulis K, Kasiakou S, Sermaidis G, Michalopoulos A. Nephrotoxicity of intravenous colistin: a prospective evaluation. *Int J Antimicrob Agents* 2005; 26: 504-7

9. Markou N, Apostolakos H, Koumoudiou C, Athanasiou M, Koutsoukou A, Alamanos I et al. Intravenous colistin in the treatment of sepsis from multiresistant Gram-negative bacilli in critically ill patients. *Critical Care* 2003; 7: R78-83
10. Allen D, Hartman B. *Acinetobacter* Species. In: Mandell G, Bennet J, Dolin R, eds. *Principles and practice of infectious disease*. Sixth Edition. Pennsylvania: Elsevier Inc. 2005; 2632-6
11. Jackson W, Shorr A. Update in ventilator-associated pneumonia. *Curr Opin Anaesthesiol* 2006; 19: 117-21
12. Jian Li, Rayner C, Nation R, Owen R, Spelman D, Tan K, Liolios L. Heteroresistance to Colistin in Multidrug-Resistant *Acinetobacter Baumannii*. *Antimicrob. Agents Chemother* 2006; 50: 2946-50
13. Matthaïou D, Michalopoulos A, Rafailidis P, Karageorgopoulos D, Papaïanou V, Ntani G et al. Risk factors associated with the isolation of colistin-resistant gram-negative bacteria: a matched case control study. *Crit Care Med* 2008; 36: 807-11
14. Li J, Nation R, Milne R, Turnidge J, Coulthard K. Evaluation of colistin as an agent against multi-resistant Gram-negative bacteria. *Int J Antimicrob Agents* 2005; 25: 11-25
15. Rea R, Capitano B. Optimizing use of aminoglycosides in critically ill patients. *Semin Resp Crit Care Med* 2007;28:596-603
16. Vasen W, Desmery P, Ilutovich S, Di Martino A. Intrathecal use of colistin. *J Clin Microb* 2000; 38: 3523

17. Yagmur R, Esen F. Intrathecal colistin for the treatment of *Pseudomonas aeruginosa* ventriculitis: report of a case with successful outcome. *Critical Care* 2006;10:428
18. Polymyxins. In: McEvoy G, Snow E, Miller J, Kester L, Litvak K, Welsh O et al, eds. *AHFS Drug Information 2008*. Bethesda: American Society of Health System Pharmacists 2008; 490-5
19. Falagas E, Kasiakou S. Toxicity of polymyxins: a systematic review of the evidence from old and recent studies. *Critical Care* 2006 ;10(1):R27
20. Rios F, Luna C, Maskin B, Valiente A, Lloria M, Gando S et al. Ventilator-associated pneumonia due to colistin susceptible-only microorganisms. *Eur Resp J* 2007;30: 307-313
21. Garnacho- Montero J, Ortiz- Leyba C, Jiménez- Jiménez A, Barrero-Almodóvar L, Garcia- Garmendia L, Bernabeu Wittell M et al. Treatment of Multidrug-Resistant *Acinetobacter Baumannii* Ventilator-Associated Pneumonia (VAP) with Intravenous Colistin: A Comparison with Imipenem-Susceptible VAP. *Clin Infect Dis* 2003; 36: 1111-8
22. Reina R, Estenssoro E, Saenz G, Canales H, Gonzalvo R, Vidal G et al. Safety and Efficacy of Colistin in *Acinetobacter* and *Pseudomonas* infections: a prospective cohort study. *Intensive Care Med* 2005; 31: 1058-65
23. Kallel H, Hergafi L, Bahloul H, Hakim A, Dammak H, Chelly H et al. Safety and efficacy of colistin compared with imipenem in the treatment of ventilator-associated pneumonia: a matched case-control study. *Intensive Care Med* 2007; 33: 1162-7

24. Bowker K (2005) Therapeutic drug monitoring of colistin- a seven year review from a UK clinical antibiotic assay service. PhD Thesis, Bristol University.
25. Antibacterials. In British Medical Association. British National Formulary 50 September 2005. London: BMJ Publishing Group Ltd 2005; 295
26. Markou N, Markantonis S, Dimitrakis E, Panidis D, Boutzouka E, Karatsas S et al. Colistin serum concentrations after intravenous administration in critically ill patients with serious multi-drug, gram-negative bacilli infections: a prospective, open-label, uncontrolled study. Clin Ther 2008;30:143-51
27. Tan C, Li J, Nation R. Activity of colistin against heteroresistant *Acinetobacter baumannii* and emergence of resistance in an in vitro pharmacokinetic and pharmacodynamic model. Antimicrob Agents Chemother 2007; 51: 4313-5
28. Quesnel L, Jaran A, Braganza J. Antibiotic accumulation and membrane trafficking in cystic fibrosis cells. J Antimicrob Chemother 1998; 41: 215-21
29. Gilbert D, Moellering R, Eliopoulos G, Sande M. The Sanford Guide to Antimicrobial Therapy 2008. Thirty-Eighth Edition. Sperryville: Antimicrobial Therapy Inc. 2008; 178-84
30. Li J, Milne W, Nation R, Turnidge J, Coulthard K, Valentine J. Simple method for assaying colistin methanesulfonate in plasma and urine using high-performance liquid chromatography. Antimicrob. Agents Chemother 2002; 46 :3304-7
31. Wootton M, Holt H, McGowan A. Development of a novel assay method for colistin sulphomethate. Clin Microbiol Infect 2005; 11: 243-44

32. Li J, Milne W, Nation R, Turnidge J, Coulthard K, Johnson D. A simple method for the assay of colistin in human plasma, using pre-column derivization with 9-fluoronelmethyl chloroformate in solid phase extraction cartridges and reversed-phase high-performance liquid chromatography. *J Chromatograph B* 2001; 761: 167-75
33. Zavascki P, Li J. Intravenous colistemethate for multidrug-resistant gram-negative bacteria. *Lancet* 2008;8:403-5
34. Hachem R, Chemaly R, Ahmar C, Jiang Y, Boktour M, Rjaili G. Colistin is effective in treatment of infections caused by multi-drug resistant *Pseudomonas aeruginosa* in cancer patients. *Antimicrob Agents Chemother* 2007;51:1905-11
35. General anti-infectives for systemic use. In: Gibbon C, Blockman N, eds. *South African medicines formulary 2008*. Eighth edition. Cape Town: Health and medical publishing group 2008; 264-333
36. Blaser J, Cerny A, Cottagnoud P, Fux C, Hoigne R, Imhof A et al. Aminoglycoside antibiotics. In: Aronson J, ed. *Meyler's side effects of drugs*. Fifteenth edition. Amsterdam: Elsevier BV 2006; 118-35
37. Bjuik S, Mouton J, Gyssens I, Verbrugh H, Bruining H. Experience with a once-daily dosing program of aminoglycosides in critically ill patients. *Intensive Care Med* 2002; 28: 936-942
38. Intensive care medicine. In Kumar P, Clark M eds. *Clinical Medicine*, fourth edition 1998; 829-59. WB Saunders, Edinburgh
39. American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference: definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. *Crit Care Med* 1992 (6): 864-74.

40. Oliver S. Western Cape academic hospitals antimicrobial recommendations and wound care management 2008

41. Sayed R. (2007). Introduction to biostatistics. [Handout]. Cape Town, South Africa: University of Cape Town: Masters in public health

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**Appendix A: Data Capture Sheet, Colistin Study , GSH**

**Patient:** \_\_\_\_\_ **Folder no.**.....

**Date of Birth:** ..... **Ward:** ..... **Sex:**.....

**Co – morbidity**.....

**Date of admission to GSH**.....

**Reason for ICU admission and date:** .....

**Markers of sepsis 48 hours prior to commencing tobramycin/ colistin**

White Cell Count.....Highest Temperature Recorded.....

Inotropic Support Needed within 24 hours of starting the drug...Y.../....N.....

New documentation of purulent sputum?      Y / N

Newly documented CXR infiltrate                      Y / N

**APACHE Score:** ICU admission.....      At start of antibiotics:.....

**SOFA SCORE on commencing therapy**.....

**Pre – Treatment Creatinine:** .....

**Site of Acinetobacter identification:** .....

**Follow Up Cultures, Microscopy ( with dates)** .....

**Antibiotic Used:**                      COLISTIN                      /                      TOBRAMYCIN

**Duration and date commenced**.....

Reason for stopping: .....

Dose used and levels obtained .....

Adverse effects experienced? .....

Other antimicrobials used during therapy .....

.....

**Discharged from ICU?(date)**.....**Death in ICU (date)**.....

**Ventilated?**      Y / N      **Dates** .....

**Discharged from GSH / death at GSH...(date)**.....

**Follow- up Creatinine Levels (Until treatment completed or if abnormal, up until 10 days)**

<b>Date</b>	<b>Creatinine Level</b>

**SOFA SCORES- Day 5**.....

**Day 10**.....

**Daily PaO<sub>2</sub> from blood gas**

<b>Date</b>	<b>PaO<sub>2</sub></b>

**Apache II score**

Initials

Date

Time

High abnormal range

Low abnormal range

4      3      2      1      0      1      2      3      4      **Score**

<b>Temperature</b>	° C	>40	39-40.9		38.5-38.9	36-38.4	34-35.9	32-33.9	30-31.9	<30		
<b>MAP</b>	mmHg	>159	130-159	110-129		70-109		50-69		<50		
<b>Heart rate</b>	bpm	>179	140-179	110-139		70-109		55-69	40-54	<40		
<b>Respiratory rate</b>	bpm	>49	35-49		25-34	12-24	10-11	6-9		<6		
<b>Oxygenation</b>												
<b>FiO<sub>2</sub>&gt;0.5 AaDO<sub>2</sub></b>	mmHg	>499	350-499	200-349		<200						
<b>FiO<sub>2</sub>&lt;0.5 PaO<sub>2</sub></b>	mmHg				<b>PaO<sub>2</sub></b>	>70	61-70		55-60	<55		
<b>Arterial pH</b>		>7.69	7.6-7.69		7.5-7.59	7.33-7.49		7.25-7.32	7.15-7.24	<7.15		
<b>Serum sodium</b>	mmol/L	>179	160-179	155-159	150-154	130-149		120-129	111-119	<111		
<b>Serum potassium</b>	mmol/L	>6.9	6-6.9		5.5-5.9	3.5-5.4	3-3.4	2.5-2.9		<2.5		
<b>Serum creatinine *</b>	mg/100ml	> 3.5	2 - 3.4	1.5 - 1.9		0.6 - 1.4		< 0.6				
<b>Haematocrit</b>	%	>59.9		50-59.9	46-49.9	30-45.9		20-29.9		<20		
<b>Total WCC</b>	x 10 <sup>9</sup> /L	>39.9		20-39.9	15-19.9	3-14.9		1-2.9		<1		

**GCS points**      **Neurologic points=15-GCS**     

\* (mg/100ml x 88.402 = umol/L)      NOTE: Double point score for ARF

**Total of APS all of the above 12 variables**      **Total APS =**     

**Se HCO<sub>3</sub> if no ABG (venous)**      mmol/L      >51.9      41-51.9      32-40.9      22-31.9      18-21.9      15-17.9      <15     

**Age points**      <44      0      45-54      2      55-64      3      65-74      5      >74      6     

**Chronic health points**      Non-operative or emergency      5      Elective post-op      2      Neither      0     

**Total APACHE II**      **APS + Age points + Chronic Health points**

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