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Vancomycin-associated nephrotoxicity in the Critically-Ill

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Abstract

In recent times there has been significant debate regarding which is the best dosing regimen for vancomycin. An increasing prevalence of invasive Methicillin resistant *Staphylococcus aureus* (MRSA) with reduced vancomycin susceptibility has led authors to advocate target serum trough concentrations be increased from 5-10 mg/L to 15–20 mg/L in an effort to curb microbial resistance. Higher serum concentrations, however, predispose the patient to an increased risk of nephrotoxicity placing the patient at risk of cardiovascular failure (secondary to fluid homeostasis disruption) and kidney failure. Minimising these risks is especially important in critically ill patients in order to reduce morbidity and mortality.

Although vast literature evaluating vancomycin-induced nephrotoxicity has been published, most analyses rely on small datasets with often inconclusive and/or conflicting results. A project with greater statistical power is thus necessary to better describe vancomycin and its relationship to nephrotoxicity as well as to evaluate whether the theoretical benefits of continuous infusion of vancomycin for minimizing nephrotoxicity translate to clinical practice. Not only will this serve to guide clinician prescribing practice, but it will also ensure vancomycin is not prematurely disregarded as a treatment option.

The aim of this thesis is to clarify what concentrations and dosing regimens of vancomycin are associated with nephrotoxicity. Specifically, the aims of this thesis are to:

1. Describe pharmacokinetic and clinically measured variables that are associated with vancomycin-associated nephrotoxicity
2. Determine whether intermittent or continuous infusion dosing of vancomycin is associated with greater nephrotoxicity.

We performed a series of retrospective analyses of over 1500 vancomycin recipients from two tertiary intensive care units. Furthermore, a meta-analysis comparing continuous infusion and intermittent infusion dosing and the respective incidence of vancomycin-associated nephrotoxicity was performed. Increased serum vancomycin concentrations and duration of therapy were identified as pharmacokinetic independent predictors of nephrotoxicity. Additionally, concomitant vasoactive therapy, increased illness-severity score and concomitant aminoglycoside use were identified as independent clinical predictors of nephrotoxicity. Furthermore, our data convincingly shows that administration of vancomycin by continuous infusion is associated with significantly less nephrotoxicity than intermittent infusion. Despite these hypothesis-generating findings, we believe that a large prospective randomised controlled trial is necessary to categorically determine

whether a mortality benefit can be achieved with continuous infusion of vancomycin.

Declaration by author

This thesis is composed of my original work, and contains no material previously published or written by another person except where due reference has been made in the text. I have clearly stated the contribution by others to jointly-authored works that I have included in my thesis.

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Timothy Hanrahan

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Publications during candidature

1. Hanrahan TP, Harlow G, Hutchinson J, Dulhunty JM, Lipman J, Whitehouse T, Roberts, JA. Vancomycin-associated nephrotoxicity in the critically ill: a retrospective multivariate regression analysis. Crit Care Med. 2014 Dec;42(12):2527–36.

2. Hanrahan TP, Whitehouse T, Lipman J, Roberts JA. Vancomycin nephrotoxicity: A meta-analysis of administration by continuous versus intermittent infusion; IJAA (Accepted for publication, April 2015)

3. Hanrahan TP, Kotapati C, Rowland J, Roberts M, Roberts JA, Lipman J, Udy A. Factors associated with vancomycin nephrotoxicity in the critically ill. AIC (Accepted for publication, May 2015)

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Contributor	Statement of Contribution
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Dr. Chaitanya Kotapati	Data Collection (33%) Statistical analysis (20%) Manuscript preparation (30%)
Dr. James Rowland	Data Collection (33%)
Dr. Matthew Roberts	Data Collection (33%)
Prof. Jason Roberts	Manuscript Review (25%)
Prof. Jeffrey Lipman	Study design (50%) Manuscript review (25%)
Dr. Andrew Udy	Study design (50%) Manuscript review (50%)

Contributions by others to the thesis

Analyses for all studies included in this thesis were performed by the MPhil candidate, Dr. Timothy P Hanrahan. All chapters and papers that constitute this Thesis were drafted by Dr. Timothy P Hanrahan, with the guidance of co-authors and degree supervisors, Prof. Jason A. Roberts, Dr. Joel Dulhunty and Prof. Jeffrey Lipman.

Data presented in Chapter 4 as the published article entitled: "Vancomycin-associated nephrotoxicity in the critically ill: a retrospective multivariate regression analysis" was generously donated by Dr. Tony Whitehouse. Co-authors Dr. Georgina Harlow and Dr. James Hutchinson were the primary data collectors. Dr. Joel Dulhunty oversaw and assisted with all aspects of statistical analysis whilst Dr. Tony Whitehouse and Prof Jeffrey Lipman aided in study design. Prof Jason A Roberts, as principal supervisor, oversaw all aspects of production.

Data presented in Chapter 4 as the submitted article entitled: "Vancomycin associated nephrotoxicity in the critically ill: a retrospective study" was collected by Dr. Chaitanya Kotapati, Dr. Matthew J Roberts, and Dr. James Rowland. Dr. Andrew Udy and Prof Jeffrey Lipman conceptualised the design of the study. Prof. Jason A Roberts, as principal supervisor, oversaw all aspects of production.

Study design, drafting and analysis of the meta-analysis presented in Chapter 4 entitled "Vancomycin-associated nephrotoxicity: a meta-analysis of administration by continuous versus intermittent infusion" was performed by the MPhil candidate Dr. Timothy P Hanrahan. Dr. Tony Whitehouse and Prof Jeffrey Lipman provided assistance with drafting of the article. Prof. Jason A Roberts, as principal supervisor, provided assistance and guidance of all aspects of the submission.

Statement of parts of the thesis submitted to qualify for the award of another degree

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Keywords

vancomycin, nephrotoxicity, sepsis, pharmacokinetics, acute kidney injury, glycopeptide, intensive care unit, infection

Australian and New Zealand Standard Research Classifications (ANZSRC)

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FoR code: 1103, Clinical Sciences – 25%

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LIST OF ABBREVIATIONS

ACEI – ANGIOTENSIN CONVERTING ENZYME INHIBITOR

AKI – ACUTE KIDNEY INJURY

AKIN – ACUTE KIDNEY INJURY NETWORK

APACHE II – ACUTE PHYSIOLOGY AND CHRONIC HEALTH EVALUATION II

ARB – ANGIOTENSION II RECEPTOR BLOCKER

AUC – AREA UNDER THE CURVE

BP – BLOOD PRESSURE

CAD – CORONARY ARTERY DISEASE

CI – CONTINUOUS INFUSION

CSF – CEREBROSPINAL FLUID

EGFR – ESTIMATED GLOMERULAR FILTRATION RATE

GCS – GLASGOW COMA SCALE

H&L – HOSMER AND LEMESHOW

ICU – INTENSIVE CARE UNIT

IHD – ISCHEMIC HEART DISEASE

II – INTERMITTENT INFUSION

IQR – INTERQUARTILE RANGE

ISDA – INFECTIOUS DISEASES SOCIETY OF AMERICA

MDRD – MODIFIED DIET IN RENAL DISEASE

MIC – MINIMUM INHIBITORY CONCENTRATION

MRSA – METHICILLIN RESISTANT *STAPHYLOCOCCUS AUREUS*

MSSA – METHICILLIN SENSITIVE *STAPHYLOCOCCUS AUREUS*

NPV – NEGATIVE PREDICTIVE VALUE

NSAID – NON-STEROIDAL ANTI-INFLAMMATORY DRUGS

OR – ODDS RATIO

PPV – POSITIVE PREDICTIVE VALUE

RCT – RANDOMISED CONTROLLED TRIAL

RIFLE – RISK, INJURY, FAILURE, LOSS OF FUNCTION, END-STAGE RENAL DISEASE

ROC – RECEIVER OPERATOR CHARACTERISTIC

SD – STANDARD DEVIATION

SOFA – SEQUENTIAL ORGAN FAILURE ASSESSMENT SCORE

TDM – THERAPEUTIC DRUG MONITORING

YI – YOUDEN INDEX

1 Introduction

Staphylococcus aureus is a Gram-positive facultative anaerobic bacterium that is considered normal skin flora in most parts of the world. It is though, an opportunistic pathogen and risk of sepsis, organ dissemination and abscess formation is high. The current treatment of choice for Methicillin resistant *S. aureus* (MRSA) is vancomycin, a glycopeptide antibiotic that inhibits bacterial cell-wall synthesis by binding d-alanyl-d-alanine. Although to date vancomycin has been effective, an increased prevalence of MRSA with reduced susceptibility to vancomycin has resulted in guidelines recommending increased serum concentrations from 5-10 mg/L to 15-20 mg/L to ensure better patient outcomes (1). A primary concern of this recommendation though, is that data strongly suggests vancomycin has a dose-dependent nephrotoxic effect (2-4).

The extent by which vancomycin is an independent nephrotoxic risk factor is currently disputed with literature largely forming inconclusive results. Further, definitive data that demonstrates the optimum dosing regimens to minimize nephrotoxicity is required. The failure to form definitive conclusions regarding dosing regimens and nephrotoxicity is largely related to previous studies relying on small samples sizes and a lack of prospective trials examining the controlled titration of vancomycin levels. A series of large studies that can categorically define the risk factors for nephrotoxicity and optimum dosing regimens are required to ensure vancomycin's prescribing practice is optimised and the clinical usefulness of this antibiotic is prolonged. This thesis will address this.

The following chapters have been provided. Chapter 2 will provide a literature review of the research field. It will discuss MRSA infection in the Intensive Care Unit (ICU) and the impact this has on mortality. It will provide a history of vancomycin, its use in ICUs and the clinical pharmacology. Furthermore, a review of vancomycin-associated nephrotoxicity will be provided. Chapter 3 outlines the aims of this thesis. Chapter 4 includes the 3 manuscripts accepted for publication during this thesis:

1. Vancomycin associated nephrotoxicity in the critically ill: A retrospective multivariate regression analysis
2. Factors associated with vancomycin nephrotoxicity in the critically ill
3. Vancomycin associated nephrotoxicity. Continuous versus intermittent infusion: A meta-analysis

Whilst Chapter 5 outlines the findings of the thesis and discusses potential avenues for future research.

2 Literature Review

2.1 Infection in Intensive Care Units

Infection is a common complication within ICU and accounts for a significant proportion of morbidity, mortality and economic burden. Progression to sepsis or septic shock is frequent with 11.8% of patients admitted to an Australian or New Zealand ICU receiving these diagnoses (5,6). Of these patients, 37.5% will die in-hospital – a figure at least double that of non-infected patients (6-8). Furthermore, the length of ICU stay increases from an average of 4 to 16 days, obviously adding significant strain to health systems worldwide.

2.2 Methicillin Resistant Staphylococcus aureus

A large number of pathogenic organisms are commonly identified in Australian and New Zealand ICUs. Fortunately, most are susceptible to standard antibiotics. It is concerning, however, that within this geographic area methicillin-resistant *Staphylococcus aureus* (MRSA) is responsible for 12.1% of all sepsis and septic shock, a figure exceeded only by methicillin-susceptible *S. aureus* (MSSA) (16%) (6,9). Data from the United States indicate that 25.8% of bacteraemias are due to MRSA (10,11) causing an attributable mortality of 23.4% (12,13). Additionally, mortality rates (both 30-day and in-hospital) have proven significantly higher in patients with MRSA bacteraemia when compared with MSSA, even when treated with appropriate antibiotics (13-15).

2.3 Vancomycin

2.3.1 History

Vancomycin is an amphoteric glycopeptide antibiotic that was isolated in 1953 from *Amycolatopsis orientalis*. The substance ("compound 05865") was found to be active against most gram-positive organisms (including penicillin-resistant *Staphylococci*), some anaerobic organisms and *Neisseria gonorrhoeae* (14,16-19). Serial passages of *Staphylococci* showed only a 4-8 fold increase in resistance to vancomycin with the same strains increasing their resistance 100000-fold to penicillin (20-22). Demand for vancomycin quickly increased and in 1958, vancomycin was approved by the US Food and Drug Administration for clinical use. This initial increase was short-lived though, as use of methicillin (also approved in 1958) and cephalothin shortly thereafter were favored secondary to less obtrusive adverse effect profiles. Vancomycin became a last resort treatment. Despite this, the emergence of MRSA and pseudomembranous enterocolitis saw a sharp increase in vancomycin prescribing during the early 1980's (23,24) as studies at the time suggested that vancomycin was equally effective against MRSA as standard therapy was against MSSA (25). Increased clinical use though, led to gradual resistance, with vancomycin-resistant enterococci (VRE) being reported in Europe by 1986 and the US by 1987 (26). Furthermore, MRSA has developed low-level resistance to vancomycin with Assadullah *et al.* (27) finding that 18% of strains have a minimum inhibitory concentration (MIC) greater than 4 mg/L. Additionally, a group at the Asan Medical Centre, South Korea, found that about half of MRSA isolates had vancomycin MICs $\geq$ 1.5 mg/L (28) a value often quoted as a high MIC when considering MRSA with reduced susceptibility to vancomycin. Given that an increased MIC is consistently associated with a higher mortality (29,30), curbing resistance and maintaining clinical efficacy is critical to the ongoing usefulness of vancomycin.

2.3.2 Pharmacokinetics of Vancomycin

Vancomycin is poorly absorbed by the oral route (31) and thus intravenous administration is required for systemic infections. Vancomycin has a volume of distribution ranging from 0.4-1.5 L/kg (32-36) with up to 50% being bound to plasma proteins (37). Although this is only moderate binding, *in vitro* studies have shown a 1 to 8-fold increase in MIC in the presence of albumin(38). As such, the MIC is likely increased *in vivo* where a vancomycin-protein interaction may occur. Vancomycin can be isolated from most body spaces although concentrations are variable and penetration into solid organs is traditionally considered poor (39). For example after 1g of intravenous (IV) vancomycin one study showed lung penetration of only 41% (40) whilst studies analysing cerebrospinal fluid (CSF) concentrations showed CSF-to-serum ratios ranging from 0 to

48%, the greater values being observed only in the presence of meningeal inflammation (34,41). Vancomycin is eliminated primarily via the renal route with 80-90% of that recovered being unchanged (34). Approximately 5.0-8.5% of vancomycin clearance is extra-renal (42).

2.3.3 Pharmacodynamics of Vancomycin

By binding d-alanyl-d-alanine cell wall precursors, vancomycin inhibits peptidoglycan cross-linking and thus inhibits bacterial cell-wall synthesis. Vancomycin acts primarily in a time-dependent manner although some concentration-dependency has been noted in both animal models and human data (43,44).

Vancomycin concentration targets and dosing regimens to ensure optimal effects remain controversial. The Australian Therapeutic Guidelines recommend intermittent infusion (II) of 1.5 grams 12-hourly when patient creatinine clearance is greater than 90 mL/min (target trough concentration 15 ± 3 mg/L) or continuous infusion (CI) of 3 g over 24 hours with target concentrations of 20 ± 3 mg/L. A decreased dose is recommended as kidney function declines with dosing monitored and adjusted on the basis of trough and peak vancomycin serum concentrations. *In vitro,* animal, and limited human data suggest that an area under the curve (AUC)/MIC value ≥ 400 is optimal although no studies demonstrate a strong correlation between this and trough concentrations ≥ 15 mg/L (45).

Studies aiming to identify whether II or CI dosing protocols have optimum bactericidal activity and which has fewer adverse effects have yielded inconclusive results. As mentioned, the bactericidal activity of vancomycin is primarily time-dependent and higher concentrations do not correlate with better outcomes (39,46,47). Administration by II results in more variable concentrations than CI (48,49). As such, theoretically, CI should have greater clinical efficacy as peaks and troughs are minimised and time above MIC is maximised. Despite this, Wysocki *et al.* (50) were unable to demonstrate any significant advantage of CI over II when comparing microbiological outcomes in 119 critically ill patients. This may be explained by the fact that when compared with beta-lactam antibiotics vancomycin may have a longer post-antibiotic effect (bactericidal activity after concentrations fall below MIC) thus lessening the importance of maintaining serum concentrations above the MIC (51,52). Moreover, confirming these findings, Cataldo *et al.* (53) were unable to demonstrate a significant difference in mortality rates between both CI and II groups in a meta-analysis of six studies.

Despite a lack of data showing significance between CI and II patient mortality and clinical outcome, CI reaches target concentrations faster with fewer therapeutic drug monitoring (TDM)

samples, has less variability in the daily infused dose, and reduces costs (50). A large study of some 1,737 patients showed that vancomycin serum trough concentrations were higher in the CI group even after two to four II doses (54). Furthermore, 70% of patients dosed within the CI group achieved target concentrations, whilst only 34% of patients with II dosing achieved target (39,54). Given a 20% reduction in bactericidal response is noted when target drug concentrations are not met within 72 hours (55) surrogate outcome data suggests that CI should be beneficial.

Given this finding, Roberts et al. (56) have suggested that when vancomycin is administered by CI, the steady state concentrations should be five to six times the MIC of the infecting organism. Achieving these target exposures becomes problematic though when the MIC of the infective organism is ≥ 2 mg/L as high serum concentrations are necessary. Although CI of vancomycin is associated with a significantly lower risk of drug related nephrotoxicity (53) a clear exposure–toxicity relationship exists with Ingram et al. (2) demonstrating that a serum concentration ≥ 28 mg/L markedly increases nephrotoxic risk. Furthermore, Lodise et al. (4) found that serum trough concentrations > 20 mg/L are associated with an increased risk of nephrotoxicity with the predicted probability of nephrotoxicity being >20% amongst non-ICU patients. Panday et al. (57) thus suggested that CI should be restricted to *S. aureus* infections with an MIC < 1 mg/L. Clearly, when treating a patient infected with MRSA that has reduced susceptibility to vancomycin, achieving an exposure associated with good microbiological outcomes will be difficult without subjecting the patient to undue nephrotoxicity (4,57).

2.3.4 Nephrotoxicity

Early vancomycin research showed a significant nephrotoxic effect, although, this was largely associated with poor purification technique of the raw material (58,59). By the late 1970s, as purification techniques improved, the associated adverse effects were significantly reduced with most studies finding an average nephrotoxicity between 5-7% of patients (19,38,53,60-62). The reduced susceptibility of MRSA to vancomycin and the subsequent increase in vancomycin dose has renewed interest in nephrotoxicity as increased serum trough concentrations, daily doses >4 g/day and increased duration of treatment are independently associated with increased risk of vancomycin toxicity(63,64). Despite Pritchard *et al.* (63) evaluating 1504 courses of vancomycin to identify these factors, phase 2 of their study (which evaluated patient-specific risk factors differentiating nephrotoxic and non-nephrotoxic populations) analysed only 129 patients. Similarly, although nephrotoxicity rates as high as 43% have been reported, concomitant nephrotoxic agents and differences in baseline severity consistently skew the results (3,4,55,60,65,66). For example, Jeffres *et al.* (3) noted that the 43% of patients (40/94) who developed renal toxicity in their study had significantly greater baseline Acute Physiology and Chronic Health Evaluation (APACHE) II scores than those who did not. Further, a blood urea nitrogen to serum creatinine ratio >20 and administration of vasopressors was also significantly greater amongst patients who developed nephrotoxicity. Furthermore, in addition to the above mentioned risk factors, total body weight ≥101.4 kg, estimated creatinine clearance ≤86.6 ml/min and heart failure have all been independently associated with an increased risk (2,58,64). Given the heterogenous population of critically ill patients and the fact that by definition, they have higher morbidity, these confounders are particularly relevant and need to be accounted for in any future studies.

Critically ill patients themselves have increased vulnerability to nephrotoxic agents and subsequent renal failure due to severity of illness (67). Not only is baseline renal dysfunction more prevalent (64), but patients are more likely to have sepsis with organ failure, hypotension requiring treatment with vasopressors, diabetes mellitus with microalbuminuria and concomitant nephrotoxic treatment (67). Lodise *et al.* (64) have demonstrated that ICU stay at initiation of treatment is associated with increased risk of nephrotoxicity. Further, they note a difference between general patients and those in ICU receiving ≥ 4g vancomycin per day and < 4g per day of vancomycin with 35% vs. 39% and 10% vs 16% respectively developing nephrotoxicity. Complicating matters further, patients with sepsis (of which a vast majority are in the ICU) have larger vancomycin volumes of distribution (up to twice that of normal) and decreased vancomycin renal clearance (68). This has the two-fold effect of both increasing daily-dose requirements and increasing the time-of-exposure in patients already vulnerable. Huang *et al.* (69) confirmed that serum trough concentrations of vancomycin

can be higher in critically-ill patients, reinforcing the fact that they are at higher risk of nephrotoxicity than the general patient. Given vancomycin clearance is primarily renal, vancomycin clearance decreases in a linear fashion with reduced creatinine clearance. This results in a vancomycin half-life from 4 – 11 hours (in healthy adults) to 10 – 200 hours in patients with renal failure (34). Clearly, this increased exposure can be detrimental.

A number of nephrotoxicity definitions exist but recent studies refer to the RIFLE and Acute Kidney Injury Network (AKIN) classifications of acute kidney injury (AKI) (70,71). RIFLE (Risk; Injury; Failure; Loss of function; End-stage kidney disease) classifies three levels of severity, and two of clinical outcomes (Table 1). The advantage of these criteria is that severity of impairment is scaled and classified and has been validated for use in patients with pre-existing renal disease (72).

Table 1 Rifle Criteria Classification

	Serum creatinine concentration criteria	**Urine output criteria**
Risk	Serum creatinine increase to 1.5 fold OR GFR decrease >25% from baseline	<0.5 ml/kg/h for 6 hours
Injury	Serum creatinine increase to 2.0 fold OR GFR decrease >50% from baseline	<0.5 ml/kg/h for 12 hours
Failure	Serum creatinine increase to 3.0 fold OR GFR decrease > 75% from baseline OR serum creatinine ≥ 354 umol/L (≥ 4mg/dl) with an acute increase of at least 44 umol/L (0.5 mg/dl)	Anuria for 12 hours
Loss	Persistent acute renal failure = complete loss of kidney function > 4 weeks	
ESKD	Complete loss of kidney function for >3 months	

GFR = glomerulus filtration rate, ESKD = End-stage Kidney Disease

Although vancomycin-induced nephrotoxicity is considered reversible (60), AKI is associated with significantly worse clinical outcomes. Palmieri *et al*. (73) demonstrated that burn patients managed in an ICU who had AKI had significantly higher length of ICU stay ($P<0.0001$) than those without (43 vs 26 days, respectively). Further, those with AKI had 34% mortality whilst all patients who did not develop AKI during ICU admission survived. Of note, almost 85% of patients who progressed to higher RIFLE classes were on nephrotoxic antibiotics (aminoglycosides and vancomycin) - almost all having sepsis. This clearly demonstrates the significance of vancomycin in ICUs as a risk factor for nephrotoxicity. Furthermore, it demonstrates the need to provide clear evidence on how best to avoid the potential nephrotoxic consequences of vancomycin.

While several authors and guidelines have advocated increasing serum trough concentrations to >15

mg/L in efforts to maintain vancomycin efficacy in the face of rising MICs, there are no data to support improvements in clinical outcomes. Jeffres *et al.* (74) retrospectively analysed 102 MRSA health-care-associated pneumonia patients of which 31% died during their hospitalisation. Vancomycin serum trough concentrations and AUC values showed no correlation with hospital mortality. Hermsen *et al.* (75) similarly demonstrated no difference between hospital length of stay or mortality when comparing patients with higher and lower serum trough concentrations of vancomycin. However, they did demonstrate that higher serum trough concentrations were associated with consistently higher rates of nephrotoxicity. These findings raise questions surrounding the recommendation of high-trough concentrations given the potential negative sequelae.

Inconsistency is rife amongst vancomycin studies. To date, no published data have concluded categorically whether vancomycin is an independent risk factor for nephrotoxicity in the context of co-morbidities and drug co-administration in the ICU. Low sample sizes are a common theme with a search of the literature revealing only two large scale studies (n > 1000) (63,76). Pfeiffer *et al.* (76) identified that cancer, hypertension, and diabetes were the most common co-morbidities associated with nephrotoxicity but no conclusions regarding vancomycin as an independent risk factor for nephrotoxicity were drawn. Furthermore, as mentioned, small sample sizes in phase 2 of the analysis by Pritchard *et al.*(63) casts doubt as to the degree that vancomycin is an independent risk factor.

The safest mechanism of administration is not certain and as discussed, it is debatable if increasing serum trough concentrations does in fact improve clinical outcomes. Furthermore, although duration of treatment, elevated serum trough concentrations and total daily dose have been identified as risk factors, the extent by which each contributes is unknown. A large-scale study with the primarily goal of ascertaining whether vancomycin is an independent risk factor is clearly required.

3 Aims and Hypotheses

This thesis aims to define risk factors of vancomycin nephrotoxicity in an ICU population. Conclusions surrounding what serum concentrations and dosing regimens are most associated with nephrotoxicity will be sought. Specifically, the aims of this thesis were to:

1. Describe pharmacokinetic and clinically measured variables that are associated with vancomycin-associated nephrotoxicity
2. Determine whether II or CI dosing of vancomycin is associated with greater nephrotoxicity.

It is hypothesised that:

1. Vancomycin serum concentrations will correlate with incidence of vancomycin nephrotoxicity and thus be a factor predictive of nephrotoxicity, and
2. Given vancomycin, when administered by CI, has significantly less variability in serum concentrations, CI will have fewer adverse effects on renal function.

4 Vancomycin Associated Nephrotoxicity in the Critically Ill

4.1 Chapter Synopsis

The aim of this chapter is to identify variables predictive of nephrotoxicity in a population of critically ill patients. Furthermore, this chapter analyses the incidence and predictive power of dosing method in the same patient population.

4.2 Published manuscript entitled, "Vancomycin associated nephrotoxicity in the critically ill: A retrospective multivariate regression analysis"

The manuscript entitled, "Vancomycin associated nephrotoxicity in the critically ill: A retrospective multivariate regression analysis" has been published by Critical Care Medicine (2014; 42(12) 2527-2536) (77).

The co-authors contributed to the manuscript as follows:

1. Timothy Hanrahan: Statistical analysis and manuscript preparation
2. Georgina Harlow: Data collection
3. James Hutchinson: Data collection
4. Joel Dulhunty: Statistical analysis and manuscript review
5. Jeffrey Lipman: Manuscript review
6. Tony Whitehouse: Manuscript review
7. Jason Roberts: Manuscript review

The manuscript is presented as submitted: except figures and tables have been inserted into the text at slightly different positions. Also, the numbering of pages, figures and tables has been adjusted for overall Thesis continuity. The references are found alongside the other references of the Thesis, in the section 'References'.

Vancomycin associated nephrotoxicity in the critically ill: A retrospective multivariate regression analysis

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4.2.1 Abstract

Objectives**:** To evaluate the influence vancomycin dose, serum trough concentration and dosing strategy have on the evolution of acute kidney injury in critically ill patients.

Design**:** Retrospective, single-centre, observational study.

Setting**:** University Hospital Intensive Care Unit, Birmingham.

Patients**:** All critically ill patients receiving vancomycin from 1 December 2004 to 31 August 2009.

Intervention**:** None.

Measurements and Main results: The prevalence of new onset nephrotoxicity was reported using RIFLE criteria and independent factors predictive of nephrotoxicity were identified using logistic regression analysis. Complete data were available for 1430 patients. Concomitant vasoactive therapy (OR = 1.633; $p < 0.001$), median serum vancomycin (OR = 1.112; $p < 0.001$) and duration of therapy (OR = 1.041; $p = <0.001$) were significant positive predictors of nephrotoxicity. II was associated with a significantly greater risk of nephrotoxicity than CI (OR = 8.204; $p = <0.001$).

Conclusions**:** In a large dataset, higher serum vancomycin concentrations and greater duration of therapy were independently associated with increased odds of nephrotoxicity. Furthermore, CI was associated with a decreased likelihood of nephrotoxicity compared with II. This large dataset supported the use of CI of vancomycin in critically ill patients.

4.2.2 Introduction

MRSA is associated with significant morbidity and mortality in the ICU. MRSA is responsible for 10% of all infections (8) and 14% of all instances of sepsis (78). Furthermore, MRSA is associated with a 50% greater likelihood of mortality than MSSA (79). Given that between 19-25% of patients colonised with MRSA develop infection, with an overall mortality rate as high as 6.3 per 100 000 infections (80) effective antibiotic treatment is critical to treatment success.

Vancomycin is the antibiotic most widely used for the treatment of infections mediated by MRSA (81). Of concern, MRSA with reduced susceptibility to vancomycin is increasing in prevalence with studies suggesting trough serum concentrations <10 mg/L are associated with the emergence of vancomycin-resistant *S. aureus* (82,83). Subsequently, clinical practice guidelines now advocate targeting trough serum concentrations of 15-20 mg/L, which is much higher than the previous target of 5-10 mg/L (1,84,85). This increase in the target exposure is considered likely to increase the likelihood of concentration-related adverse effects, including nephrotoxicity.

Some authors have proposed that doses >4 g/day, high serum trough concentrations and an increased duration of vancomycin therapy are associated with nephrotoxicity (8,63,64). To date though, there is a relative paucity of large-scale data able to measure the significance of vancomycin exposure as an independent risk factor for nephrotoxicity.

This study aimed to evaluate the influence vancomycin dose, serum trough concentration and dosing strategy have on the evolution of acute kidney injury in critically ill patients.

4.2.3 Materials and Methods

A retrospective cohort study was conducted on data from the University Hospital Birmingham, a tertiary referral and university affiliated hospital. This ICU treats up to 80 critically ill patients at any one time and manages approximately 4500 patients annually. The ICU provides local and tertiary care for all adult specialties including heart, lung, liver, kidney and bone marrow transplantation. The data of all patients who received intravenous vancomycin from 1 December 2004 to 31 August 2009 was extracted from a central database. The data of patients receiving vancomycin by non-intravenous routes were not included in the primary database.

Local protocol dictated that patients with a central venous catheter receive vancomycin by CI. No criteria were established as to which patients should receive vancomycin by II, but typically, this would occur if 1) the clinician was not compliant with the protocol or 2) no central line was present. Data of those patients by which the dosing method was unknown or those patients who received vancomycin by both continuous and II were included in interests of maximising available data. If a patient was the recipient of an II, serum concentrations were measured within 30-minutes of the next dose. If the patient was on CI, the samples were taken randomly, but at least 18 hours after the preceding dose change.

The study was approved by the South Birmingham Research Ethics Committee (09/H1207/140). Data extracted from the hospital's electronic database included sex, weight (where available), date of birth, ethnicity, hospital and ICU admission dates, ICU and hospital discharge dates, hospital discharge status, time of vancomycin prescription, administration start times, rate of infusion, dosage, serum creatinine concentration at admission, serum creatinine concentration during vancomycin therapy, trough serum vancomycin concentration and MRSA status. If multiple trough serum vancomycin concentrations were available, the median and maximum measured concentrations were recorded.

As rifampicin's pulmonary penetration is often considered superior to vancomycin (86) any concomitant prescription was included in the analysis to measure effects it may have on clinical outcome. Furthermore, given reports of rifampicin renal toxicity (87,88), inclusion allowed analysis of its influence on renal function when prescribed simultaneously with vancomycin. Given patients included in the analysis were admitted to the ICU, inotrope data were collected to account for potential confounding effects on renal function. Sequential Organ Failure Assessment (SOFA) (79,89) data were also collected at the start of treatment. With the exception of Glasgow Coma Scale (GCS) and blood pressure (BP), all components (ventilation status, worst daily PO_2/FiO_2 ratio, highest inotrope use, liver function, platelet count and creatinine concentrations) of the SOFA

score were calculated using data collected from the same electronic database.

Estimated glomerular filtration rate (eGFR) was calculated using the Modification of Diet in Renal Disease (MDRD) formula (80,90) for all serum creatinine concentrations obtained throughout the ICU stay. The primary endpoint, new onset nephrotoxicity, was defined as an increase in serum creatinine concentration ≥50%, a decrease in eGFR ≥25% or a serum creatinine concentration ≥350 μmol/L (in the setting of an acute increase ≥ 44μmol/L) as per the RIFLE acute kidney injury classification system (70). Secondary endpoints were death within 72 hours of the last recorded vancomycin dose (irrespective of treatment modality), all-cause mortality and a combined endpoint of either death within 72 hours of vancomycin administration or nephrotoxicity.

The prevalence of new onset nephrotoxicity was reported and univariate analysis was performed to determine data distribution and the prevalence of missing data. Data for which no serum vancomycin concentration, dosing amount or creatinine concentration were available (n = 755) or, which had incomplete SOFA score availability (n = 356) were excluded from analysis. Furthermore, where unique patients had multiple ICU admissions during the study period (n = 151), only data from the first episode were used. Continuous variables with a normal distribution are reported as mean ± standard deviation (SD); non-normal variables are reported as median and inter-quartile range (IQR). The Pearson product-moment correlation coefficient was used to identify highly correlated potential predictive variables ($r > 0.8$) with the variable most predictive of nephrotoxicity included in further analysis. Predictive variables associated with the primary and secondary endpoints were explored using logistic regression analysis. Manual and backward stepwise techniques were used to identify the model with best fit. Interactions between predictive variables were included where multivariate and bivariate findings differed and inclusion of the interaction improved goodness of fit. Independent predictive variables with a p-value <0.05 were considered statistically significant. Goodness of fit was assessed by the Hosmer and Lemeshow (H & L) statistic and the Nagelkerke R^2 index. Receiver operating characteristic (ROC) curves were used to explore thresholds for nephrotoxicity at different highest measured and median serum vancomycin concentrations. Youden's index was used to identify the optimal threshold for maximising sensitivity and specificity at specific threshold values. Statistical analysis was performed in SPSS (Version 20.0, IBM Corp. Armonk, NY).

4.2.4 Results

During the study period, 2359 patients were prescribed vancomycin therapy in line with the study inclusion criteria. Of these, 2208 were primary admissions, of which 1430 had complete datasets (65%). Univariate analysis comparing excluded and included patients showed no significant differences between age at admission (p = 0.055), Day 1 MDRD (p = 0.319) or weight (p = 0.349) (Table 2).

Table 2 Comparison of baseline characteristics between patients included and excluded from final analysis

	Excluded N = 778 (35%)	**Included N = 1430 (65%)**	**p-value**
Age at admission	57.95	56.48	0.055
Day 1 MDRD	72.22	71.01	0.319
Weight	77.46	76.78	0.349

MDRD = Modified Diet in Renal Disease

Median age was 60.0 (45-70) years with 65% (935/1430) male. Median weight was 75.0 (67.0-86.0) kg. Vasoactive therapy was used in 62% (885/1430) of patients, whilst 6% (92/1430) received simultaneous rifampicin therapy. Furthermore, 11% (150/1430) were identified as MRSA positive. The median trough serum vancomycin concentration was 15.3 (9.6-19.6) mg/L whilst the median length of vancomycin therapy was 4.4 (2.3-8.6) days. The median average dose was 1.7 (1.1-2.1) grams of vancomycin per day. The predominant method of administration was CI (46% or 653/1430), followed by II (28% or 390/1430); 16% (221/1430) received vancomycin by both continuous and II, whilst the mode of administration was not described in 11% (150/1430) of patients. The median SOFA score (not inclusive of GCS) was 6.0 (4.0-8.0). The prevalence of nephrotoxicity in the study population during ICU admission was 21% (300/1430); ICU mortality for the study population was 20% (288/1430). Patient demographics are summarised in Table 3. Table 4 summarises differences in clinical and demographic variables between patients who did and did not develop nephrotoxicity during ICU admission.

Table 3 Demographic data (n = 1430)

Factors		N (%)
Sex (Male)		935 (65%)
Age (median (IQR))		60.0 (45-70)
Weight (median (IQR))		75.0 (67.0-86.0)
SOFA[1] score (median (IQR))		6.0 (4.0-8.0)
Median serum vancomycin concentration (mg/L; median (IQR))		15.3 (9.6-19.6)
Average vancomycin dose daily (grams; median (IQR))		1.7 (1.1-2.1)
Length of vancomycin therapy (days; median (IQR))		4.4 (2.3-8.6)
ICU Mortality		288 (20%)
Nephrotoxicity		300 (21%)
Death within 72 hours of last vancomycin		224 (16%)
Nephrotoxicity or died within 72 hours of cessation		469 (32%
Infusion Type	Continuous Infusion	653 (46%)
	Intermittent Dosing	390 (28%)
	Mixed Dosing	221 (16%)
	Unknown	166 (12%)
Simultaneous vasoactive therapy		885 (62%)
MRSA[2] Positive		150 (11%)
Simultaneous rifampicin therapy		92 (6%)

[1]Glasgow coma scale (GCS) values were not available for inclusion thus SOFA total is SOFA minus GCS.
[2]Methicillicin resistant Staphylococcus-aureus
SOFA = Sequential Organ Failure Assessment, MRSA = Methicillin resistant Staphylococcus aureus, IQR = Interquartile range

Table 4 Summary of nephrotoxic and non-nephrotoxic groups

		Nephrotoxicity (Median (IQR)) N = 300	Non-nephrotoxic (Median (IQR)) N = 1130	Significance (p-value)
Age (years)		62.0 (51.0-71.0)	59.0 (44.0-70.0)	0.004
Sex (Male)		191 (63.7%)	744 (65.8%)	0.482
Weight (kg)		75.0 (66.0-85.0)	75.0 (67.0-85.7)	0.366
SOFA score		7.0 (5.0-9.0)	6.0 (4.0-8.0)	<0.001
Median vancomycin serum concentration (mg/L)		18.9 (13.8-22.2)	14.2 (9.2-18.4)	<0.001
Duration of treatment (days)		8.0 (4.0-15.8)	4.0 (2.0-6.9)	<0.001
Average vancomycin daily (grams/day)		1.1 (1.6-0.6)	1.8 (1.3-2.3)	<0.001
Total vancomycin exposure (grams)		8.1 (4.8-13.9)	6.8 (4.0-11.0)	<0.001
Infusion Method	Continuous	161 (53.7%)	492 (43.5%)	0.001
	Intermittent	77 (25.7%)	313 (27.7%)	0.001
	Mixed	44 (14.7%)	177 (15.7%)	0.001
	Unknown	18 (6.0%)	148 (13.1%)	0.001
Simultaneous rifampicin		23 (7.7%)	69 (6.1%)	0.327
MRSA		38 (12.7%)	112 (9.9%)	0.166
Simultaneous vasoactive prescription		234 (78.0%)	651 (57.6%)	<0.001

[1] Calculated by Mann-Whitney U statistic as variables fail Kolmogorov-Smirnov normality testing
[2] GCS values were not available for inclusion thus SOFA total is SOFA minus GCS.
SOFA = Sequential Organ Failure Assessment, MRSA = Methicillin resistant *Staphylococcus aureus*, IQR = Interquartile range

Patients who received vancomycin by II received a significantly lower median average daily dose (1.5 [0.9-2.2] grams) than those who received vancomycin by CI (1.7 [1.2-2.1] grams; p = 0.003), mixed method administration (1.7 [1.2-2.1] grams; p = 0.020) or unknown method of administration (2.0 [1.0-2.1] grams; p = 0.005). Furthermore, patients who received vancomycin by II (8.8 [6.5-11.2] mg/L) had a significantly lower median serum vancomycin concentration than those who received it by CI (18.4 [15.6-21.2] mg/L; p = <0.001). Table 5 summarizes group differences by method of vancomycin administration.

Table 5 Summary of patients' data receiving vancomycin categorised by infusion method type.

Variable	Continuous Infusion (n = 653)	Intermittent Infusion (n = 390)	Mixed (n = 221)	Unknown (n = 166)	P[1]
Sex (Male)(%)	417 (63.9%)	260 (66.7%)	145 (65.6%)	113 (68.1%)	0.685
Age (median (IQR))	59 (44-69)	61 (47.8-71)	59 (45-70)	63 (46.8-72)	0.060
Weight (median (IQR))	75 (66.1-85)	75 (67.8–88)	75 (65-84.5)	75 (65-87.9)	0.331
SOFA score (median (IQR))	7.0 (5.0-9.0)	5.0 (3.0-7.0)	6.0 (3.0-8.0)	6.0 (4.0-8.0)	<0.001
Median serum vancomycin concentration (mg/L; median (IQR))	18.4 (15.6-21.2)	8.8 (6.5-11.2)	15.5 (12.1-19.1)	11.9 (8.2-17.7)	<0.001
Average vancomycin dose daily (grams; median (IQR))	1.7 (1.2-2.1)	1.5 (0.9-2.2)	1.7 (1.2-2.1)	2.0 (1.0-2.1)	0.003
Length of vancomycin therapy (days; median (IQR))	5.3 (3.4-10.3)	4.4 (2.5-7.3)	5.0 (2.9-9.2)	0.8 (0.4-1.2)	<0.001
ICU Mortality (%)	172 (26.3%)	49 (12.6%)	31 (14.0%)	36 (21.7%)	<0.001
Nephrotoxicity (%)	161 (24.7%)	77 (19.7%)	44 (19.9%)	18 (10.8%)	0.001
Death within 72 hours of last vancomycin (%)	130 (19.9%)	36 (9.2%)	25 (11.3%)	33 (19.9%)	<0.001
Nephrotoxicity or died within 72 hours of cessation (%)	253 (38.7%)	101 (25.9%)	66 (29.9%)	49 (29.5%)	<0.001
Simultaneous vasoactive therapy (%)	469 (71.8%)	177 (45.4%)	151 (68.3%)	88 (53.0%)	<0.001
MRSA Positive (%)	64 (9.8%)	56 (14.4%)	20 (9.0%)	10 (6.0%)	0.014
Simultaneous rifampicin therapy (%)	35 (5.4%)	38 (9.7%)	14 (6.3%)	5 (3.0%)	0.009
Highest measured serum vancomycin concentration (mg/L; median (IQR))	24.7 (18.7-28.5)	11.8 (8.4-17.2)	19.9 (15.8-26.0)	12.7 (9.0-19.1)	<0.001
Cumulative vancomycin dose (grams; median (IQR))	9.0 (6.0-14.4)	5.8 (4.0-9.0)	8.0 (4.7-13.8)	2.0 (1.5-3.0)	<0.001

[1]Calculated by Kruskall-Wallis statistic where variable is linear as variables fail Kolmogorov-Smirnov normality testing)

SOFA = Sequential Organ Failure Assessment, MRSA = Methicillin resistant *Staphylococcus aureus,* IQR = Interquartile range, ICU = Intensive Care Unit

ROC analysis indicated that the threshold for development of nephrotoxicity for median vancomycin concentration was 17.8 mg/L (sensitivity = 0.60, specificity = 0.71, Youden's Index = 0.31, AUC = 0.677) whilst the threshold for highest measured serum vancomycin concentration during admission was 23.7 mg/L (sensitivity = 0.65, specificity = 0.74, Youden's Index = 0.39, AUC = 0.727). Table 6 summarises the risk of nephrotoxicity, sensitivity and specificity for incremental increases in trough serum vancomycin concentration.

Table 6 Precision of predicting nephrotoxicity and incremental risk increase of different threshold values for highest measured vancomycin serum concentrations

Threshold level (mg/L)	Nephrotoxicity (%)	Relative Risk Increase[1]	Sensitivity	Specificity	YI	PPV	NPV
10	21.7%	-	1	0.043	0.043	0.217	1
15	23.2%	1.069	0.936	0.178	0.115	0.232	0.914
20	26.2%	1.207	0.84	0.372	0.212	0.262	0.898
25	33.1%	1.525	0.747	0.600	0.346	0.331	0.899
30	41.5%	1.912	0.603	0.774	0.377	0.415	0.880
>30	47.9%	2.207	0.303	0.912	0.216	0.478	0.831

[1]Relative to first threshold level (10 mg/L)
YI = Youdens index, PPV = Positive predictive value, NPV = Negative predictive value

4.2.4.1 Predictors of nephrotoxicity

The most parsimonious logistic regression model identified duration of therapy in days (OR = 1.041; $p < 0.001$), simultaneous vasoactive therapy (OR = 1.633; $p < 0.001$) and median trough serum vancomycin concentration (OR = 1.112; $p < 0.001$) as independent positive predictors of nephrotoxicity (Table 7). II was associated with a significantly greater risk of nephrotoxicity than CI (OR = 8.204; $p < 0.001$). There was however a significant interaction between median serum vancomycin concentration and infusion method. A 1 mg/L increase in the median serum vancomycin concentration had lower odds of nephrotoxicity in the II group compared with the CI group (OR = 0.92; $p = 0.013$). There was adequate goodness of fit (Hosmer and Lemeshow test $X^2 = 13.31$, df = 8, $p = 0.102$; Nagelkerke $R^2 = 0.192$).

4.2.4.2 Predictors of nephrotoxicity or death within 72 hours (combined endpoint)

Independent positive predictors of nephrotoxicity or death within 72 hours of vancomycin treatment (Table 7) SOFA (OR = 1.128; p < 0.001), positive MRSA status (OR = 1.696; p = 0.008), simultaneous vasoactive therapy (OR = 1.501 p = 0.008), median vancomycin serum concentration (OR = 1.094; p = <0.001) and duration of therapy (OR = 1.032; p = <0.001). There was a significantly greater odds of nephrotoxicity or death within 72 hours of dosing in those who received vancomycin by II compared to CI (OR = 1.645; p = 0.007). Goodness of fit was adequate (Hosmer and Lemeshow test X^2 = 14.553, df = 8, p = 0.068; Nagelkerke R^2 = 0. 208).

4.2.4.3 Predictors of death within 72 hours

Independent predictors of death within 72 hours of the last vancomycin dose showed SOFA (OR = 1.190; p < 0.001), simultaneous rifampicin therapy (OR = 2.075; p = 0.010) and median trough serum vancomycin (OR = 1.034; p = 0.009) as positive predictors (Table 8). The odds of death from mixed method dosing (OR = 0.619; p = 0.047) was less than that for patients receiving CI; II was non-significantly different to CI (OR = 0.726; p = 0.167). Goodness of fit was adequate (Hosmer and Lemeshow test X^2 = 5.469, df = 8, p = 0.706; Nagelkerke R^2 = 0.113).

Table 7 Logistic regression analysis with nephrotoxicity and nephrotoxicity OR death endpoints

Factors		Nephrotoxicity				Nephrotoxicity OR Death[1]			
		All factors		Final Model		All factors		Final Model	
		OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value
Age[2]		1.032 (0.957-1.114)	0.413	–	–	1.072 (0.996-1.153)	1.031	–	–
Weight		0.998 (0.990-1.007)	0.708	–	–	1.000 (0.992-1.008)	0.725	–	–
Sex[3]		0.900 (0.679-1.193)	0.464	–	–	1.148 (0.878-1.502)	0.411	–	–
SOFA score[4]		1.044 (0.997-1.092)	0.065	–	–	1.134 (1.085-1.185)	<0.001	1.128 (1.080-1.179)	<0.001
Infusion Method[5]	Intermittent	1.022 (0.625-1.671)	0.932	8.204 (2.875-23.411)	<0.001	1.525 (1.059-2.195)	0.308	1.645(1.149-2.356)	0.007
	Mixed	2.139 (1.251-3.657)	0.005	2.781 (0.661-11.705)	0.163	0.924 (0.645-1.325)	0.504	0.945 (0.660-1.352)	0.755
	Unknown	1.267 (0.732-2.194)	0.398	8.050 (2.403-26.967)	0.001	1.431 (0.935-2.191)	0.356	1.487 (0.973 – 2.274)	0.067
Simultaneous rifampicin prescription		1.029 (0.602-1.757)	0.918	–	–	1.305 (0.788-2.162)	0.604	–	–
MRSA Positive		0.865 (0.564-1.326)	0.505	–	–	1.644 (1.096-2.466)	0.066	1.696 (1.145-2.511)	0.008
Simultaneous vasoactive prescription		0.683 (0.496-0.940)	0.019	1.633 (1.226-2.174)	<0.001	1.445 (1.066-1.957)	0.003	1.501 (1.111-2.029)	0.008
Median serum vancomycin (mg/L)		1.104 (1.077-1.132)	<0.001	1.112 (1.085-1.139)	<0.001	1.088 (1.063-1.114)	<0.001	1.094 (1.069-1.120)	<0.001
Median serum vancomycin*Intermittent		–	–	0.924 (0.868-0.983)	0.013				
Median serum vancomycin*Mixed		–	–	0.961 (0.889-1.039)	0.314				
Median serum vancomycin*Unknown		–	–	0.891 (0.838-0.947)	<0.001				
Duration of therapy (days)		1.040 (1.027-1.053)	<0.001	1.041 (1.028-1.054)	<0.001	1.031 (1.019-1.043)	0.187	1.032 (1.020-1.044)	<0.001
Goodness of fit:									
Hosmer and Lemeshow test		$X^2 = 10.666$, df = 8	0.221	$X^2 = 13.307$, df = 8	0.102	$X^2 = 21.489$, df = 8	0.006	$X^2 = 14.553$, df = 8	0.068
Nagelkerke R^2		0.184		0.192		0.212		0.208	

[1] Death during vancomycin dosing or within 72 hours of cessation
[2] Age was re-categorised as an ordinal scale in 10-year increments: Odds >1 is the increase in odds of the outcome within a 10-year increase in the factor
[3] Odds ratio compares female relative to male
[4] GCS values were not available for inclusion thus SOFA total is SOFA minus GCS
[5] Odds ratio is relative to continuous infusion
SOFA = Sequential Organ Failure Assessment, MRSA = Methicillin resistant *Staphylococcus aureus*, GCS = Glasgow coma scale

Table 8 Logistic regression analysis with death within 72 hours of vancomycin dosing and all-cause mortality as end-points

Factors		Death within 72 hours of vancomycin dosing				All-Cause Mortality[1]			
		All factors		Final Model		All factors		Final Model	
		OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value	OR (95% CI)	*P*-value
Age[2]		1.047 (0.957-1.145)	0.318	–	–	1.069 (0.984-1.161)	0.116	–	–
Weight		0.997 (0.987-1.006)	0.495	–	–	0.994 (0.985-1.003)	0.186	0.992 (0.984-1.001)	0.081
Sex[3]		1.203 (0.867-1.670)	0.269	–	–	1.197 (0.884-1.619)	0.245		–
SOFA score[4]		1.189 (1.126-1.255)	<0.001	1.190 (1.133-1.249)	<0.001	1.166 (1.110-1.226)	0.000	1.172 (1.121-1.226)	<0.001
Infusion Method[5]	Intermittent	0.699 (0.438-1.116)	0.134	0.726 (0.461-1.143)	0.167	0.682 (0.447-1.040)	0.076	0.735 (0.488-1.107)	0.141
	Mixed	0.620 (0.386-0.997)	0.049	0.619 (0.386-0.994)	0.047	0.545 (0.353-0.884)	0.006	0.554 (0.358-0.855)	0.008
	Unknown	1.443 (0.906-2.296)	0.122	1.379 (0.878-2.164)	0.163	1.219 (0.781-1.903)	0.384	1.218 (0.784-1.892)	0.380
Simultaneous rifampicin		1.816 (1.009-3.266)	0.047	2.075 (1.190-3.619)	0.010	1.650 (0.952-2.859)	0.074	1.793 (1.055-3.046)	0.031
MRSA Positive		1.370 (0.834-2.251)	0.214	–	–	1.373 (0.868-2.171)	0.176	–	–
Simultaneous vasoactive prescription		1.045 (0.707-1.543)	0.826	–	–	1.134 (0.795-1.618)	0.488	–	–
Median serum vancomycin (mg/L)		1.030 (1.002-1.058)	0.032	1.034 (1.008-1.060)	0.009	1.030 (1.005-1.056)	0.020	1.038 (1.014-1.063)	0.002
Duration of vancomycin therapy (days)		1.006 (0.997-1.015)	0.215	–	–	1.014 (1.005-1.023)	0.003	1.015 (1.006-1.024)	0.001
Nephrotoxicity		0.932 (0.644-1.350)	0.709	–	–	1.199 (0.863-1.668)	0.279	–	–
Goodness of fit:									
Hosmer and Lemeshow test		$X^2 = 2.969$, df = 8	0.936	$X^2 = 5.469$, df = 8	0.706	$X^2 = 10.586$, df = 8	0.226	$X^2 = 7.560$, df = 8	0.478
Nagelkerke R^2		0.120		0.113		0.146		0.139	

[1] Death during ICU admission
[2] Age was re-categorised as an ordinal scale in 10-year increments: Odds >1 is the increase in odds of the outcome within a 10-year increase in the factor
[3] Odds ratio compares female relative to male
[4] GCS values were not available for inclusion thus SOFA total is SOFA minus GCS
[5] Odds ratio is relative to continuous infusion
SOFA = Sequential Organ Failure Assessment, MRSA = Methicillin resistant *Staphylococcus aureus*, GCS = Glasgow coma scale

4.2.4.4 Predictors of all cause mortality

SOFA (OR = 1.172; p < 0.001), simultaneous rifampicin therapy (OR = 1.793; p = 0.031), median trough serum vancomycin (OR = 1.038; p = 0.001) and duration of vancomycin therapy (OR = 1.015; p = 0.001) were significant positive predictors of all-cause mortality (Table 8). Weight was non significantly negatively predictive of mortality (OR = 0.992; p = 0.081). II (OR = 0.735; p-value = 0.141), and mixed method dosing (OR = 0.554; p = 0.008) had lower odds of death than participants receiving vancomycin by CI. There was adequate goodness of fit (Hosmer and Lemeshow test $X^2 = 7.560$, df = 8, p = 0.478; Nagelkerke $R^2 = 0.139$).

4.2.5 Discussion

There are few large-scale studies examining the influence of vancomycin therapy on nephrotoxicity in critically ill patients. The need to better understand the vancomycin exposure-toxicity relationship is important given recent guidelines advocating higher serum trough concentrations to counter the decreasing susceptibility of MRSA (84,85). In this study of 1430 critically ill patients, we found that elevated median trough serum vancomycin concentration is associated with a significant increase in risk of nephrotoxicity with each 1 mg/L increase in concentration associated with a 11.2% increase in the odds of nephrotoxicity. Duration of therapy was also positively predictive of nephrotoxicity with every 1 day increase in the duration of therapy being associated with a 4.1% increase in the odds of nephrotoxicity.

These findings are in concordance with other studies that show duration of vancomycin therapy to have a significant positive association with nephrotoxicity (3,55,64). Of interest, Pritchard et al. (63) noted a significant rising trend in vancomycin serum concentrations (p < 0.001) without an increase in the incidence of nephrotoxicity during the same period. This finding, however, may be confounded by the association of increasing serum trough concentrations with a decreasing duration of therapy during the same period. We hypothesise that if organisms with reduced susceptibility to vancomycin continue to become more prevalent, then the potential combination of increased duration of treatment and higher trough serum concentrations may result in a further increased incidence of nephrotoxicity.

We found that CI was significantly less likely to cause nephrotoxicity in multivariate analysis than all other infusion types despite patients on CI receiving greater daily doses than those receiving II of vancomycin. Patients who received II had an 8.2 times higher odds of nephrotoxicity than those who received CI and this effect was independent of baseline renal function and serum vancomycin

concentration. This confirms the conclusion reached in a recent meta-analysis that suggested CI is associated with a significantly reduced risk of nephrotoxicity compared with II (RR = 0.6, 95% CI 0.4-0.9; p = 0.02)(53). Moreover, mixed and unknown dosing strategies were associated with lower odds of nephrotoxicity than II. This is expected, due to the fact that the latter categories likely consist of a large proportion of patients dosed by CI in accordance with unit protocol. Furthermore, it has been shown that up to 70% of patients dosed within the CI group achieved target concentrations, whilst only 34% of patients with II dosing achieved target (54). Given the rise in trough concentration recommendations over the study period (85), the II group may have simply been undertreated.

The higher prevalence of nephrotoxicity in the CI group compared with the II group (24.7% vs. 19.7%) in bivariate analysis deserves mention as this suggests the possibility of confounding. As described above, the median serum vancomycin concentration was significantly higher in patients receiving CI and this was identified as the main factor hypothesised to be responsible for this confounding effect (Table 5). In addition, there was a significant interaction between median serum vancomycin concentration and infusion method in multivariate analysis, such that an increase in median serum concentration was associated with a higher odds of nephrotoxicity in those who received vancomycin by CI compared with those with II. As discussed previously, 66% of patients receiving II do not reach target concentrations. Therefore, it could very well be that an increasing serum vancomycin concentrations is associated with increasing nephrotoxicity in the CI group. Again, AUC would be ideal to study this relationship.

To our knowledge this is the first large-scale study that has shown vancomycin administration by CI is associated with decreased nephrotoxicity. Unfortunately, the decrease in acute kidney injury associated with CI does not translate to improved mortality. II was associated with a non-significant lower odds of mortality than CI (p = 0.141). A greater percentage of the cohort received CI (Table 3) and the duration of treatment and median SOFA score were both higher in the CI group (Table 5), alluding to potential non-measured factors confounding the result. Given local protocol dictates that prescription of vancomycin by CI can only be administered by central line, and inherently, a patient requiring central access is likely to have greater morbidity, CIs being more predictive of mortality than II in this cohort is not surprising. A large prospective study is required to categorically determine the effect of treatment method on mortality.

In addition to being associated with nephrotoxicity, duration of vancomycin therapy also appears positively predictive of all-cause mortality. We are, however, unable to speculate on why this is the

case as information on the indication for vancomycin therapy, infection site and sensitivities of the targeted organism are unknown. These factors may all contribute to extended vancomycin regimens in the context of greater morbidity. It is interesting to note that although nephrotoxicity was positively associated with mortality in the enter model, it was not included in the final logistic regression model due to poor significance. We hypothesise that follow up at 28 days, or later, would identify this trend as significant.

The analysis included in this study provided interesting results. In phase 1 of their study, Pritchard et al. (63) showed that a median trough vancomycin serum concentration of 14 mg/L was the threshold for development of nephrotoxicity. Here we have shown that maximum sensitivity and specificity for nephrotoxicity occurred at a median concentration of 17.8 mg/L. Though not a large increase in concentration compared with Pritchard et al. (63), our study suggests that the lower spectra of recommended serum concentrations are relatively safe. In clinical practice, the median concentration is not prospectively useful. We found the threshold for nephrotoxicity is 23.7 mg/L when considering the highest measured serum concentration observed for a single patient. A prudent clinician, with the aid of therapeutic drug monitoring (TDM), thus has the potential to negate significant risk of nephrotoxicity by ensuring measured concentrations do not surpass these values. Furthermore, it is clear that greater concentrations do have an association with nephrotoxicity (Table 6) and this must be considered when targeting high serum concentrations to circumvent the challenge of a high MIC. As MRSA MICs continue to rise it will be necessary to look to other agents for therapeutic purposes, particularly if the clinical context deems the risk of acute kidney injury not tolerable to the patient.

It must be recognised that this study is limited by its retrospective nature and as such causality cannot be demonstrated. An inherent flaw of retrospective data analysis is the difficulty to account for all potential confounding variables and simultaneous treatment agents. A prospective randomised controlled trial is necessary to confirm these results. While we are able to explore factors associated with nephrotoxicity in patients receiving vancomycin, we are unable to quantify the overall risk of nephrotoxicity associated with vancomycin use in a general ICU population. We also acknowledge that SOFA has not been validated for tracking the severity of illness in ICU. Despite this, inclusion of this SOFA score allowed for the degree of morbidity to be partially accounted for in the multivariate analysis. Furthermore, the titration of vancomycin dosing based on MDRD determinations of eGFR is not validated and may not be optimal. Finally, generalisability to other ICU population groups needs to be ensured by validation with an independent dataset.

In summary, we have shown that trough serum vancomycin concentrations and duration of therapy are associated with increased risk of nephrotoxicity. Further, baseline organ function (SOFA) and simultaneous vasoactive therapy are predictive of nephrotoxicity. Given recommendations to increase serum vancomycin concentrations to 15-20 mg/L to combat rising MICs, these data reinforce the valuable role that TDM plays in optimising safe vancomycin therapy. We have also demonstrated that CI is associated with significantly less nephrotoxicity than dosing by II. Despite this, there is still a lack of data showing whether the method of administration impacts on all-cause mortality and resolution of infection, despite our results showing a small non-significant trend towards survival advantage in the II cohort. Given that CI is associated with decreased nephrotoxicity, reaches target concentrations faster with fewer samples (when loading doses are used) (32,56), has less variability in the daily infused dose, reduces costs (50) and has less variability in serum concentrations (48), this large dataset supports use of CI of vancomycin in critically ill patients.

4.3 Published manuscript entitled, "Factors associated with vancomycin nephrotoxicity in the critically ill"

The manuscript entitled, "Factors associated with vancomycin nephrotoxicity in the critically ill" has been accepted by Anaesthesia and Intensive Care for publication.

The co-authors contributed to the manuscript as follows:

1. Timothy Hanrahan: Statistical analysis and manuscript preparation
2. Chaitanya Kotapati: Manuscript preparation and data collection
3. Matthew J Roberts: Data Collection
4. James Rowland: Data collection
5. Jeffrey Lipman: Manuscript review
6. Jason A Roberts: Manuscript review
7. Andrew A Udy: Manuscript review

The manuscript is presented as submitted: except figures and tables have been inserted into the text at slightly different positions. Also, the numbering of pages, figures and tables has been adjusted for overall Thesis continuity. The references are found alongside the other references of the Thesis, in the section 'References'.

Vancomycin associated nephrotoxicity in the critically-ill: A retrospective multivariate analysis

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4.3.1 Summary

Vancomycin is a glycopeptide antibiotic commonly used in the management of MRSA infection. The recent increase in prevalence of MRSA with reduced susceptibility to vancomycin has prompted experts to advocate for higher target trough serum concentrations. This study aimed to evaluate the potential consequences of more aggressive vancomycin therapy, by examining the association between higher serum concentrations and AKI in a population of critically ill patients. We collected data for all patients who received vancomycin over a 5-year period, and evaluated the prevalence of new onset AKI using the RIFLE kidney disease criteria. One-hundred and fifty-nine patients provided complete data, with 8.8% manifesting new onset AKI while receiving vancomycin. The median age was 57 (44 - 68) years, whilst the median trough serum concentration was 16 (10 – 19) mg/L. Multivariate logistic regression analysis identified mean trough concentration (OR = 1.174; p = 0.024), APACHE II score (OR = 1.141; p = 0.012) and simultaneous aminoglycoside prescription (OR = 18.896; p = 0.002) as significant predictors of AKI. These data suggest higher trough vancomycin serum concentrations are associated with greater odds of AKI in the critically ill.

4.3.2 Introduction

Vancomycin is a glycopeptide antibiotic commonly used to treat MRSA and coagulase negative staphylococci infections in the critically ill (91,92). Recently, there has been an increase in the prevalence of MRSA with reduced susceptibility to vancomycin, with serum concentrations <10 mg/L associated with the emergence of vancomycin-resistant *Staphylococcus aureus.* (85). Subsequently, current consensus guidelines now recommend a target serum vancomycin trough concentration between 15 and 20 mg/L(84). Given the historical target was 5 to 10 mg/L (1), there is a relative paucity of data examining the effect of more aggressive drug exposures on the incidence of vancomycin associated adverse effects - especially in the critically ill.

Vancomycin associated nephrotoxicity has been reported in up to 40% of recipients, although the exact factors predictive of AKI in this setting are still debated (91). Current data suggest that higher daily doses, higher trough serum concentrations, and increased duration of therapy, are associated with nephrotoxicity (63,64). Given the baseline risk of developing nephrotoxicity is between 36 and 67% in the critically ill (depending on the definition employed) (93), there is an imperative to more specifically identify which factors are linked with vancomycin associated AKI, in order to optimise clinical efficacy and safety when prescribing this agent.

As such, our aim was to examine the incidence over a five year period, of new onset AKI in critically ill patients admitted to a tertiary-referral hospital, who were receiving vancomycin. Furthermore, we aimed to evaluate independent risk factors predictive of AKI in this cohort, with a view to informing future dosing practice.

4.3.3 Methods

Data were obtained from the institutional database of a large tertiary referral ICU. This facility admits upwards of 2500 patients annually, providing services to all subspecialties excluding cardiothoracic and solid organ transplant surgery. Data concerning all patients who received intravenous vancomycin therapy, irrespective of indication, for greater than 96hrs, between 1 January 2004 and 31 December 2008, were extracted for analysis. The Institutional Review Board for Low and Negligible Risk Research approved this study, without the requirement for individual patient consent, as per the National Statement on Ethical Conduct in Human Research (2007).

Baseline variables included; age, sex, weight, date of admission, admission diagnosis, APACHE II score (94), and treatment received whilst in ICU. Information pertaining to patient comorbidities

was also collected. Patients were excluded from further analysis if they were less than 18 years of age, had a baseline serum creatinine concentration >176.8 μmol/L (2 mg/dl) (4), or were receiving renal replacement therapy at the time of commencing vancomycin therapy. Vancomycin doses, serum concentrations, and baseline renal function for at least two days prior to commencement of therapy, were also required for inclusion in analysis. Local protocol dictates that vancomycin dose is determined by actual body weight on admission. Given weight was not recorded for all patients; it was assumed in such cases that protocol was followed.

In those patients receiving vancomycin by II, trough serum concentrations were obtained within 48 hours of commencing therapy, after a minimum of three doses (85). Thereafter, trough serum concentrations were collected 30 minutes prior to the next dose. If a patient received vancomycin by CI, concentrations were collected at random intervals, but not before 18 hours had elapsed since prior dose alteration. The choice of infusion method was ultimately dictated by individual clinician preference, though the standard unit protocol was II during the study period. Dosing was subsequently adjusted to achieve a vancomycin concentration in the desired range. Where multiple serum vancomycin concentrations were available, mean trough concentrations were calculated for use in subsequent analysis.

AKI was determined using the RIFLE (70) criteria, such that a patient was deemed to have AKI if they showed a sustained increase in serum creatinine (>1.5 times baseline) for greater than 48 hours duration. Urine output, eGFR and calculated creatinine clearance were not used as a measure of renal function, as changes from baseline creatinine are adequately sensitive in detecting AKI (95,96). Creatinine serum concentrations were recorded for the duration of ICU admission, though only values obtained during vancomycin administration were included in the assessment of vancomycin-associated AKI. Vasopressor data, and simultaneous nephrotoxic agent administration (including angiotensin converting enzyme inhibitors, diuretics, aminoglycosides, amphotericin, iodinated contrast agents, acyclovir, cyclosporin, and nonsteroidal anti-inflammatory drugs) were collected as potential confounding variables.

Continuous data are reported as mean +/- SD where normally distributed, and median, +/- IQR where non-normally distributed. Categorical data are presented as counts (%). Univariate comparisons were made between those with and without new-onset AKI, utilising parametric and non-parametric tests of significance as appropriate. Backwards, stepwise logistic regression analysis was used to ascertain a parsimonious model identifying predictors of AKI. Goodness of fit was assessed by the Hosmer and Lemeshow (H & L) statistic and the Nagelkerke R^2 index. ROCs

were used to identify thresholds for AKI. A $p < 0.05$ was considered as statistical significance. Analysis was performed in SPSS (Version 20.0, IBM Corp. Armonk, and NY).

4.3.4 Results

During the study period, 303 patients were prescribed vancomycin, of whom 251 received this for greater than 96 hours. A further 93 patients were excluded from data extraction, reasons for which are presented in Figure 1. One hundred and fifty-eight patients were included in the final analysis.

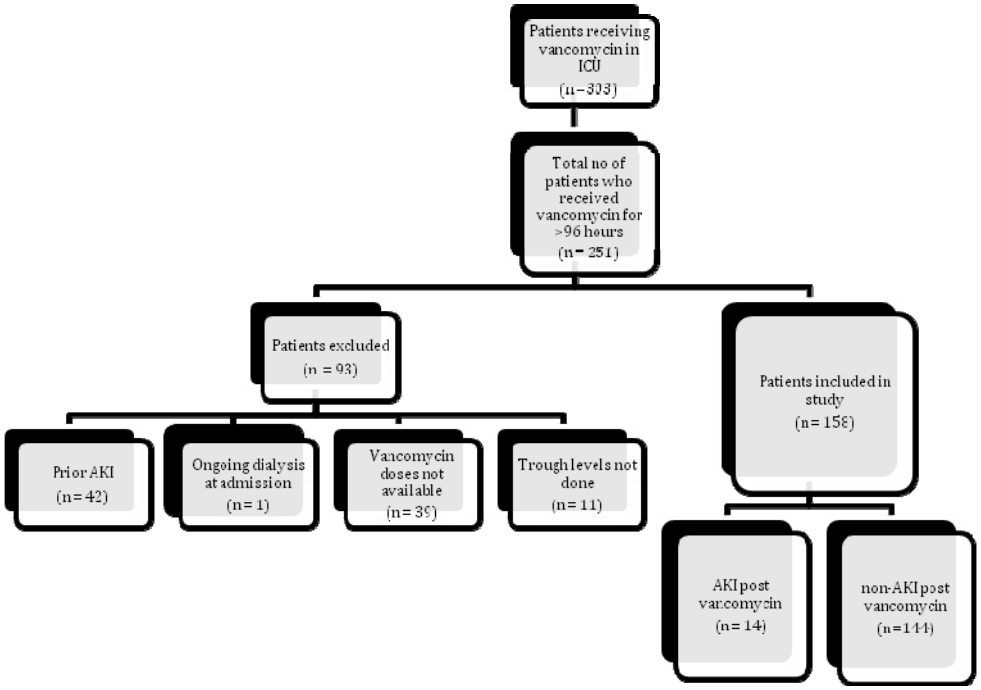

Figure 1 Consort diagram outlining patients selected and omitted from further analysis
AKI = acute kidney injury, ICU = intensive care unit.

The median age was 57 (43.75 – 68.25) years, mean APACHE II score 21.32 ± 7.40, and 65.8% (104/158) were male. Median vancomycin treatment time was 158 (120 - 234) hours. The primary method of vancomycin administration was intermittent dosing (145/158; 91.8%), whilst two patients (1.3%) received vancomycin by CI. Eleven patients (7%) received vancomycin by both CI and II. 8.9% (14/158) developed new onset AKI following vancomycin exposure. Twelve (7.6%) patients died in ICU. There was simultaneous use of nephrotoxic agents in 70.9% (112/158). These characteristics are summarised in Table 9.

Table 9 Summary of demographic data of those patients included in final analysis

Factors		(n = 158)
Sex (Male)		104 (65.8%)
Age (median (IQR))		57 (43.75 – 68.25)
APACHE (mean ± SD)		21.32 ± 7.40
Duration of vancomycin therapy (hours; median (IQR))		158 (120 - 234)
ICU Mortality		12 (7.6%)
AKI		14 (8.9%)
Infusion Type	Continuous Infusion	2 (1.3%)
	Intermittent Dosing	145 (91.8%)
	Mixed Dosing	11 (7.0%)
Simultaneous nephrotoxic agent		112 (70.9%)

AKI = acute kidney injury, APACHE = acute physiology and chronic health evaluation, ICU = intensive care unit, IQR = Interquartile range, SD = standard deviation.

There was no significant difference in the duration of vancomycin therapy amongst those patients who developed AKI (175.5 (127.75 – 374.75)) compared to those with did not (158 (117.75 – 213.75))**.** However, patients with AKI were more likely to have sepsis (64.3% vs 36.1%; p = 0.047), or ischemic heart disease (35.7% vs. 11.1%; p = 0.023), as their primary admission diagnosis. Table 10 summarises the key differences between the AKI and non-AKI groups.

The serum trough concentration threshold with the greatest sensitivity and specificity for AKI was 16.5 mg/L by ROC analysis (sensitivity = 0.93, specificity = 0.60, Youden's index = 0.53, AUC = 0.815). Backwards logistic regression analysis identified mean trough serum vancomycin concentration (OR = 1.174; p = 0.024) and APACHE II score (OR = 1.141; p = 0.012) as significant independent positive predictors of AKI. In addition, simultaneous administration of an aminoglycoside was the only nephrotoxin identified as being predictive of new onset AKI (OR = 18.896; p = 0.002). Goodness of fit was adequate (Hosmer & Lemeshow X^2 = 1.583, df = 8, p = 0.991; Nagelkerke R^2 = 0.492 summarises the regression analysis (Table 11).

4.3.5 Discussion

In this cohort of critically ill patients treated with vancomycin for greater than 96 hours, new-onset AKI was noted in approximately one out of every ten patients. Higher peak, mean, and initial trough vancomycin concentrations were associated with AKI in univariate analysis, although only the mean concentration was an independent predictor in regression modelling (OR = 1.174; p = 0.024). This means that for every 1 mg/L increase in mean trough serum vancomycin concentration, there is a 17.4% increase in the odds of new-onset AKI (Table 11).

These findings are consistent with Lodise et al. (4) who noted that the mean initial vancomycin trough value was significantly higher amongst patients who had nephrotoxicity. Similarly, Pritchard et al. (63) identified an increased prevalence of vancomycin toxicity in patients with higher serum vancomycin trough values. Interestingly, Pritchard et al. (63) also identified duration of therapy > 7 days as an independent risk factor for nephrotoxicity. In our study, duration of therapy was non-significantly greater in the new-onset AKI group, although the sample-size is not sufficient to further explore this observation.

Interestingly, all 14 patients who had new-onset AKI in this cohort had mean trough serum vancomycin concentrations > 15 mg/L. Furthermore, ROC analysis identified 16.5 mg/L as the concentration with the greatest sensitivity and specificity for new onset AKI. This suggests that although greater trough serum vancomycin concentrations may be more efficacious (55,85), this may be at the expense of more AKI in a small number of patients. Prospective studies evaluating patients with serum vancomycin concentrations above 15mg/L, compared with those below 15 mg/L would further elucidate this finding.

Our study also suggests that concurrent use of aminoglycosides and vancomycin increases the risk of new-onset AKI (55,60,85,97). Specifically, there was an 18.89 (p = 0.002) times greater odds of developing AKI in those patients who received aminoglycosides and vancomycin simultaneously. Although it is standard practice to use single daily doses at our institution, we did not collect data on the type or duration of aminoglycoside therapy in this cohort. Rather, we only determined if the patient had received aminoglycosides of any type, at any time, during their admission. As such, the precise impact of aminoglycosides cannot be determined accurately, although a relationship appears to exist, mandating further research.

We included APACHE II in the multivariate model to account for illness severity (94). The mean APACHE II score in patients who developed AKI was non-significantly higher in univariate

analysis (25.86 vs 20.88; p = 0.107). Nevertheless, APACHE II was identified as a significant independent risk factor in regression modeling. In this case, every one-figure increase in APACHE II score is associated with a 14.1% increase in the odds of AKI. This is perhaps not that surprising, although it does suggest that those patients with greater illness severity are at higher risk of AKI in the setting of vancomycin therapy. Moreover it highlights the value of careful TDM and tracking of renal function in such patients.

Similarly, there was a significantly higher rate of AKI in patients that were diagnosed with sepsis at the time of admission. However, this was not an independent predictor of AKI in our multivariate model, a finding contrary to other studies (98). Patients with sepsis have a larger vancomycin volume of distribution (up to twice that of normal) and decreased vancomycin renal clearance, as compared with the general population (68). This has the two-fold effect of both increasing daily-dose requirements, and increasing the duration-of-exposure in patients already vulnerable to AKI. Given vancomycin clearance is primarily renal (34), vancomycin clearance decreases in a linear fashion with reduced creatinine clearance. This serves to increase the half-life from 3-9 hours (in healthy adults) (34) to up to 180 hours in those with severe renal failure (99). As such, we postulate that having a diagnosis of sepsis would likely contribute to new-onset AKI with vancomycin therapy, notwithstanding the results of our multivariate analysis.

No significant increase in AKI was seen amongst patients receiving vancomycin and inotropes, a finding disparate with previous reports (3,100). Furthermore, despite AKI being an established independent risk factor for increased mortality (73,101), we were not able to demonstrate this. There was however a non-significant trend towards increased mortality in those with new-onset AKI in univariate analysis. These observations, seemingly at odds with current literature, are likely attributable to the low incidence of AKI in our study cohort.

This study is limited by its retrospective design, in that causality cannot be determined. Additionally, we cannot account for all potential confounders, only those that were collected and included in multivariate analysis. Ideally, a prospective trial would confirm these results. We also acknowledge that the sample size may not be sufficient to identify all predictive factors with absolute certainty, especially as we have reported a lower rate of AKI compared with previous literature (91). It should be noted that we have also excluded patients with a baseline creatinine greater than 176.8 μmol/L (4); a group at high risk of developing AKI. As such, these data cannot be generalised to all patients in the ICU. Furthermore, we did not include any long-term morbidity and mortality data in the study, so we are unable to determine the lasting significance of these

findings.

In conclusion, these data confirm higher trough serum vancomycin concentrations are associated with greater odds of new-onset AKI in the critically ill, although absolute cause and effect remains uncertain. Our findings also underscore the importance of strict TDM and dose adjustment of vancomycin (102-104), particularly with the significant alterations in pharmacokinetics commonly encountered in these patients. This is especially important in those with higher baseline illness severity, as this appears to be an important risk factor for new onset AKI in this setting. As this sub-group often require empirical antibacterial therapy, use of vancomycin TDM should be considered mandatory, with the avoidance of supra-therapeutic concentrations. Future prospective research should now systematically evaluate the impact of aggressive vancomycin exposure on both clinical efficacy, and toxicity.

Table 10 Comparison of patients who developed AKI versus those who did not whilst receiving vancomycin in the ICU

		AKI (median (IQR)) n = 14[1]	Non AKI (Median (IQR)) n = 144	p-value
Age (years)		63 (54.25 – 72.50)	55.5 (43.00 – 68.00)	0.083
APACHE (mean ± SD)[2]		25.86 ± 10.58	20.88 ± 6.91	0.107
Length of ICU stay (days)		11 (4.75 – 27.25)	16 (9 – 22)	0.317
Diagnoses	Trauma	0 (0%)	24 (16.7%)	0.130
	Sepsis	9 (64.3%)	52 (36.1%)	0.047
	Postop	6 (42.9%)	44 (30.6%)	0.374
	Respiratory Failure	10 (71.4%)	66 (45.8%)	0.093
Highest trough concentration (mg/L)		28.00 (25.00 – 37.25)	22.00 (13.00 – 27.00)	<0.001
First trough concentration (mg/L)		21.00 (14.00 – 27.50)	10.00 (6.00 – 17.00)	<0.001
Mean trough concentration (mg/L)		20.00 (17.00 – 29.00)	15.00 (10.00 – 19.00)	<0.001
Duration of vancomycin therapy (hours)		175.50 (127.75 – 374.75)	157.50 (117.75 – 213.75)	0.268
Sex (Male)		10 (71.4%)	94 (65.3%)	0.773
Infusion Method	Continuous Infusion	0 (0%)	2 (1.4%)	0.502
	Intermittent Dosing	14 (100%)	131 (91%)	
	Mixed	0 (0%)	11 (7.6%)	
ICU Mortality		2 (14.3%)	10 (6.9%)	0.280
Diabetes Mellitus		2 (14.3%)	19 (13.2%)	1.000
Hypertension		5 (35.7%)	47 (32.6%)	0.775
IHD/CAD		5 (35.7%)	16 (11.1%)	0.023
Aminoglycosides		10 (71.4%)	52 (36.1%)	0.019
Aciclovir		2 (14.3%)	12 (8.3%)	0.358
Cyclosporin		0 (0%)	3 (2.1%)	1.000
Steroids		3 (21.4%)	36 (25%)	1.000
IV Contrast		1 (7.1%)	35 (24.3%)	0.193
NSAIDS		0 (0.0%)	5 (3.5%)	1.000
Amphotericin		2 (14.3%)	6 (4.2%)	0.150
ACEi or ARB		2 (14.3%)	19 (13.2%)	1.000
Vasopressors		12 (85.7%)	96 (66.7%)	0.228

[1] Reported as median (IQR) as data was non-normally distributed as per significant Shapiro-Wilk normality

[2] Reported as mean ± SD as data was normally distributed as per non-significant Shapiro-Wilk normality test

ACEi = angiotensin-2 converting enzyme inhibitor, APACHE = acute physiology and chronic health evaluation, ARB = angiotensin-2 receptor blocker, CAD = coronary artery disease, ICU = Intensive care unit, IHD = ischaemic heart disease, IQR = Interquartile range, IV = intravenous, NSAID = non-steroidal anti-inflammatory drug, SD = standard deviation.

Table 11 Multivariate analysis identifying risk-factors associated with vancomycin associated AKI.

Variables		AKI			
		All factors		Backwards LR[1]	
		OR (95% CI)	*P*-value	OR (95% CI)	*P*-value
Age[2]		1.368 (0.720 - 2.599)	0.338	–	–
Sex[3]		1.198 (0.222 – 6.476)	0.834		—
APACHE II		1.146 (0.999 – 1.314)	0.052	1.141 (1.029-1.265)	0.012
Highest Trough Concentration		1.035 (0.928 - 1.154)	0.540	–	–
First Trough Concentration		1.100 (0.973 - 1.245)	0.129	1.103 (0.979-1.243)	0.106
Mean Trough Concentration		1.127 (0.916 - 1.387)	0.257	1.174 (1.021-1.349)	0.024
Duration of therapy		0.999 (0.993 - 1.005)	0.774	–	–
Vasoactives[4]		1.265 (0.179 - 8.913)	0.814	–	–
Sepsis[4]		1.495 (0.272 - 8.230)	0.644	–	–
Concomitant nephrotoxic agents[4]	Aminoglycosides	21.312 (2.082 – 218.133)	0.010	18.896 (2.980-119.809)	0.002
	Aciclovir	1.119 (0.072-17.447)	0.936	–	–
	Steroids	1.421 (0.168-12.058)	0.747	–	–
	IV Contrast	0.563 (0.040 – 7.871)	0.670	–	–
	Amphotericin	2.755 (0.079 - 95.812)	0.576	–	–
	ACEi or ARB	2.152 (0.209 - 22.138)	0.519	–	–
Goodness of Fit:					
Hosmer & Lemeshow test		$X^2 = 0.739$, df = 8	0.999	$X^2 = 1.583$, df = 8	0.991
Nagelkerke R^2		0.522		0.492	

[1]Logistic Regression

[2]Age was re-categorised as an ordinal scale in 10-year increments: Odds >1 is the increase in odds of the outcome within a 10-year increase in the factor.

[3] The OR is the odds of AKI in a female versus that of a male i.e. Females have a 0.165 lower odds of AKI than that of males

[4] Dichotomous variables are expressed as the OR of having or receiving the nominated variable versus not

ACEi = angiotensin-2 converting enzyme inhibitor, APACHE = acute physiology and chronic health evaluation, ARB = angiotensin-2 receptor blocker, CAD = coronary artery disease, ICU = Intensive care unit, IHD = ischaemic heart disease, IQR = Interquartile range, IV = intravenous, NSAID = non-steroidal anti-inflammatory drug, SD = standard deviation.

4.4 Published manuscript entitled "Vancomycin associated nephrotoxicity. Continuous versus intermittent infusion: a meta-analysis"

The manuscript entitled, "Vancomycin associated nephrotoxicity. Continuous versus intermittent infusion: a meta-analysis" has been accepted by the International Journal of Antimicrobial Agents for publication.

The co-authors contributed to the manuscript as follows:

1. Timothy Hanrahan: Data collection, statistical analysis and manuscript preparation
2. Tony Whitehouse: Manuscript review
3. Jeffrey Lipman: Manuscript review
4. Jason Roberts: Manuscript preparation and review

The manuscript is presented as submitted: except figures and tables have been inserted into the text at slightly different positions. Also, the numbering of pages, figures and tables has been adjusted for overall Thesis continuity. The references are found alongside the other references of the Thesis, in the section 'References'.

Vancomycin-associated nephrotoxicity: a meta-analysis of administration by continuous versus intermittent infusion.

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4.4.1 Abstract

Vancomycin is a glycopeptide antibiotic widely used in the management of methicillin-resistant *Staphylococcus aureus*. Guidelines currently recommend vancomycin be administered by II, despite recent research suggesting CI may be associated with fewer rates of vancomycin-associated nephrotoxicity. In 2012, Cataldo et al presented a meta-analysis supporting the use of CI. Here, we present an updated meta-analysis, inclusive of a recently published large-scale retrospective study. Pubmed, EMBASE and Cochrane review databases were searched using keywords 'vancomycin' and 'continuous' or 'intermittent' or 'infusion' or 'discontinuous' or 'administration. Seven studies were included in final analysis. Using a random effects model, a non-significant trend of reduced nephrotoxicity in those who received vancomycin by CI (RR = 0.799, 95% CI 0.523 – 1.220, p = 0.299) was identified. A large randomised controlled trial is necessary to confirm these results.

4.4.2 Introduction

Vancomycin is an antibiotic widely used in the treatment of MRSA infections. In 2009, the Infectious Diseases Society of America (ISDA) advocated vancomycin serum trough concentration targets be increased from 5 to 10 mg/L to a now accepted norm of 15 to 20 mg/L to ensure that sufficient drug exposures are achieved for less susceptible strains (85). The recommendation for higher troughs is potentially problematic with reports of increased nephrotoxicity with increased serum trough concentrations >15 mg/L (91). Consequently, the focus has shifted to identifying modifiable risk factors associated with nephrotoxicity and subsequently optimising vancomycin therapy to reduce the incidence of nephrotoxicity.

Recent literature has suggested that vancomycin administered by CI is associated with reduced rates (48,77,105) and slower onset (106) of nephrotoxicity when compared with that of II. Furthermore, in 2012, a systemic review by Cataldo et al (53) meta-analysed the available published data and showed a clear trend towards reduced nephrotoxicity when vancomycin was administered by CI. Despite this, guidelines continue to recommend II of vancomycin. Recently, we performed a large retrospective multivariate analysis of 1430 patients who received vancomycin in a tertiary hospital wherein II was associated with a significantly greater risk of nephrotoxicity than CI (OR = 8.204; $p \leq 0.001$) in multivariate analysis. Here we present an updated meta-analysis, inclusive of all papers included in Cataldo et al's review of nephrotoxicity in CI versus II and, an additional two papers.

4.4.3 Materials and Methods

4.4.3.1 Search Strategy, selection criteria and study Selection

The meta-analysis by Cataldo et al (53) was used as a baseline, with all papers assessable for nephrotoxicity included in this study being selected. Furthermore, a search using Pubmed, EMBASE and Cochrane review that studied vancomycin and administration method from January 2012 was also performed. Keyword searches included 'vancomycin' and 'continuous' or 'intermittent' or 'infusion' or 'discontinuous' or 'administration. In view of being consistent, these were the same keywords utilised by Cataldo et al (53). Observational studies or RCTs were the only publication type included in analysis. Studies were excluded if they were conducted on a paediatric population. Abstracts were reviewed and papers assessed for consideration of eligibility.

4.4.3.2 Analysed outcomes

The primary outcome measure was incidence of nephrotoxicity (Table 13). Incidence was compared between those who received vancomycin by II and those who received vancomycin by CI method.

4.4.3.3 Analysis

To compare results, dichotomous variables were expressed as risk ratios with 95% confidence intervals using comprehensive meta-analysis (107). Data was pooled using the random-effects model and a summary of the risk ratios of the effects with 95% confidence intervals were calculated. The chi-square test was performed to assess heterogeneity with I^2 statistic assessing the extent. A p-value <0.05 was considered significant.

4.4.4 Results

In addition to our recently published study (77), our literature search identified only one additional study comparing CI and II to those identified by Cataldo. In total, seven studies were included in meta-analysis (48,50,77,105,106,108,109).

4.4.4.1 Demographics and setting

All studies were performed in adult tertiary centres with the target pathogen being MRSA and/or other gram-positive infections. One study (48) was performed in patients with osteomyelitis and thus received vancomycin for extended periods (up to 6 weeks). A proportion of treatment was on an outpatient basis (once pharmacokinetic steady-state had been reached). One study (106) analysed all patients receiving vancomycin as outpatients, whilst the remainder (50,77,105,108,109) were performed in patients admitted to tertiary intensive-care units (ICU). Overall the mean age was 56.58 years and the mean duration of treatment was 24.6 days. 70% of patients were male. Of 1534 vancomycin courses across all contributing studies, 946 (62%) were delivered by CI, whilst 588 (38%) were delivered by intermittent dosing. 330 (22%) patients developed nephrotoxicity after vancomycin administration. A summary of data is included in Table 12.

4.4.4.2 Methodology used in included studies

Two studies were RCTs (50,109), two were prospective cohort (48,108) and the remainder were retrospective cohort studies (77,105,106)(Table 13). Three studies (48,50,105) targeted serum trough concentration of 20-25 mg/L, one targeted 20-30 mg/L (108), one targeted 15-20 mg/L

(109), and the remainder did not specify. Vancomycin administration regimens and AKI definitions are outlined in Table 13.

4.4.4.3 Study Quality and Design

The quality of the cohort studies included in the analysis were quantified using the Newcastle-Ottawa scale as developed by Wells et al (110)(Table 14). Three studies were multi-centre (48,50,106) whilst the remainder were single-centre, thus there is reasonable external generalizability. The RCT performed by Wysocki et al. (50) was well designed with effort to avoid bias. As blinding was not possible at the time of administration, a committee, blinded to the infusion method, extracted data from charts for analysis. Furthermore, demographic characteristics, severity of underlying disease, site of infection, and pathogens were similar in the CI and II groups. Similarly, the RCT by Schmelzer et al (109) was well designed with computer-generated randomisation however; there is no indication as to whether data extraction and analysis was blinded.

4.4.4.4 Summary of Findings

Effect sizes of the seven studies are summarised in Table 15 with Figure 2 showing the Forest plot for nephrotoxicity. A fixed effects model found a moderate degree of heterogeneity (I^2 = 43.67) confirming the correct use of random effect model for final analysis. Random effects modelling found a non-significant trend of reduced nephrotoxicity in those who received vancomycin by CI (RR = 0.799, 95% CI 0.523 – 1.220, p = 0.299).

Including only the studies that were performed in ICUs, a fixed effects model found an I^2 of only 15.62. For consistency however a random effect model is reported. Random effects modelling found a non-significant higher risk of nephrotoxicity in patients receiving vancomycin by CI (RR = 1.024, 95% CI 0.765-1.371, p = 0.872). Effect sizes are summarised in Table 16 with Figure 3 showing the Forest plot for nephrotoxicity in ICU patients.

4.4.4.5 Discussion

This meta-analysis has shown that when using a random effects model, CI of vancomycin is associated with a non-significant reduced risk of nephrotoxicity when compared to that of II. This is despite the largest included study (77) showing a greater percentage of nephrotoxicity in the CI group during the univariate analysis. Interestingly, Hanrahan et al (77) showed that II was associated with greater odds of nephrotoxicity in the multivariate analysis. This suggests that the

univariate analysis of this study was likely influenced by confounders. Of note, SOFA was significantly higher in the CI group suggesting pre-existing organ dysfunction was contributing to the nephrotoxicity. Given multivariate analysis partially corrects for confounding; it is likely that the multivariate analysis from our recent study (77) is a truer representation of effect. Despite this, we included the univariate analysis result in meta-analysis for consistency and highlight this as a potential limitation of this meta-analysis.

All 3 studies that reported daily dose noted greater total daily doses in the CI group, yet those who report AUC had greater exposure in the II. This is likely due to II having greater maximum serum concentrations than CI, perhaps an underlying variable contributing to CIs overall reduced risk of nephrotoxicity. In only ICU patients, there was a non-significant trend towards increased risk of nephrotoxicity in II. Of note however, given the much larger sample size, our recent study (77) had a significantly greater weighting (64.12%) than the other included studies (Table 16). As such the abovementioned confounders are likely influencing this result and we acknowledge this to be a significant limitation. Furthermore we acknowledge that three of the seven included studies are retrospective in nature, which may expose the analysis to confounders.

4.4.4.6 Conclusion

There are few large-scale studies that compare the incidence of nephrotoxicity after CI and II. This meta-analysis, in addition to recent papers supporting CI, have shown CI should be considered as the preferential administration method for vancomycin to reduce nephrotoxicity risk. Large prospective RCTs ultimately need to confirm these results.

Table 12 Characteristics of studies included in meta-analysis

Reference	Age (years, mean)		Sex (male, n)		Infusion Method		Vancomycin Duration(days, mean)		Daily Dose (mean)		Nephrotoxicity (n)	
	CI	II	CI	II	CI	II	CI	II	CI	II	CI	II
Wysocki, 1995 (108)	61	67	10	10	13	13	16	16	24 mg/kg/day	12 mg/kg/day	2	3
Wysocki, 2001 (50)	64	62	45	35	61	58	13	14	NA	NA	10	11
Vuagnat, 2004 (48)	NA	NA	NA	NA	23	21	101	66	33.9 mg/kg/day	31.9 mg/kg/day	0	4
Hutschala, 2009 (105)	59	59	21	72	119	30	9	9	1935 mg/day	1325 mg/day	33	11
Ingram, 2009 (106)	51	55	25	24	40	40	22	20	NA	NA	4	10
Schmelzer, 2013 (109)	40	41	32	33	37	36	NA	NA	NA	NA	1	3
Hanrahan, 2014 (77)(median)	59	61	417	260	653	390	5.3	4.4	1.7 g/day	1.5 g/day	161	77

CI = Continuous infusion, II = Intermittent infusion, NA = Not available

Table 13 Comparison of studies including vancomycin administration method, study type, target infection and nephrotoxicity definitions

Reference	Target Concentration		Loading Dose		Vancomycin Dose		Study Type	Target Infection	Hospital Unit	Nephrotoxicity Definition
	CI	II	CI	II	CI	II				
Wysocki 1995 (108)	20-30mg/L	Peak: 20-40mg/kg Trough 5-10mg/kg	15 mg/kg for 1 hour	–	30 mg/kg/day	15 mg/kg infused over 1 hour BD[11]	Prospective Cohort	• Bacteraemia • Pneumonia	Intensive Care Unit	Rise in serum creatinine of 44.2 umol/L or more, OR a rise of 88.4 uumol /L if the initial creatinine was 265.2umol/L or above.
Wysocki 2001 (50)	20-25 mg/L	10-15 mg/L	15 mg/kg for 1 hour	_	30 mg/kg/day	15 mg/kg infused over 1 hour BD	Randomised Control Trial	• Severe Hospital acquired infections	Intensive Care Unit	50 % increase in serum creatinine from the day treatment was started to the end of treatment
Vuagnat, 2004 (48)	Trough 20-25 mg/L	Peak: <50 mg/L Trough: 20-25 mg/L	20 mg/kg over 1 hour	_	40 mg/kg/day	20 mg/kg over 1 hour BD	Prospective Cohort	• Osteomyelitis	Inpatients until vancomycin reached steady state, then managed as outpatients	50 % increase in serum creatinine from the day treatment was started to the end of treatment
Hutschala, 2009 (105)	20-25 mg/L	Trough: 15 mg/L	20 mg/kg	20 mg/kg	0.025mg/kg/min	Adjusted according to trough concentration	Retrospective Cohort	• Infection post Cardiac-surgery	Intensive Care Unit	Increase in SCr of more than or equal to 0.3 mg/dL or a percentage increase in creatinine of at least 50%, or a reduction in urine output (<0.5ml/kg/hour) for more than 6 hours.
Ingram, 2009 (106)	–	–	–	–	Physician guided	Physician guided	Retrospective Cohort	• All	Outpatient parenteral antimicrobial therapy unit	50 % increase in serum creatinine from the day treatment was started to the end of treatment
Schmelzer, 2013 (109)	15-20 mg/L	15-20 mg/L	20 mg/kg	–	0.9 to 2.4 ug/kg/hr Altered according to renal clearance	15 mg/kg BD	Randomised Control Trial	• Ventilator-associated pneumonia	Intensive Care Unit	50% increase in serum creatinine from baseline during treatment.
Hanrahan, 2014 (77)	NA	NA	NA	NA	NA	NA	Retrospective Cohort	• All	Intensive Care Unit	An increase in serum creatinine concentration more than or equal to 50%, a decrease in eGFR more than or equal to 25%, or a serum creatinine concentration more than or equal to 350 μmol/L (in the setting of an acute increase ≥ 44 μmol/L)

BD = Bolus Dosing, sCR = Serum creatinine, NA = Not available, eGFR = estimated Glomerular filtration rate

Table 14 Newcastle-Ottawa scale study quality analysis for cohort studies

	Selection				Comparability	Exposure/Outcome		
	Representativeness of the exposed cohort	Selection of the non-exposed cohort	Ascertainment of exposure	Demonstration that outcome of interest was not present at start of study	Comparability of cohorts on the basis of design or analysis	Assessment of outcome	Was follow-up long enough for outcomes to occur	Adequacy of follow up of cohorts
Wysocki 1995 (108)	*	*	ND	*	**	ND	*	*
Vuagnat, 2004 (48)	SG	*	*	*	–	B *	*	*
Hutschala, 2009 (105)	SG	*	–	*	**	*	*	*
Ingram, 2009 (106)	SG	*	*	*	–	*	*	*
Hanrahan, 2014 (77)	SG	*	*	*	**	*	*	*

SG = Selected group of users; ND = No description

Table 15 Comparison of continuous infusion and intermittent infusion method in all patients included in meta-analysis

	Risk Ratio	Lower Limit	Upper Limit	Z-Value	p-value	Relative Weight
Wysocki 1995 (108)	0.667	0.133	3.354	-0.492	0.623	5.88
Wysocki 2001 (50)	0.864	0.397	1.881	-0.367	0.713	17.19
Vuagnat, 2004 (48)	0.102	0.006	1.785	-1.563	0.118	2.07
Hutschala, 2009 (105)	0.756	0.435	1.314	-0.991	0.322	24.11
Ingram, 2009 (106)	0.400	0.137	1.170	-1.673	0.094	11.27
Schmelzer, 2013 (109)	0.324	0.035	2.975	-0.996	0.319	3.35
Hanrahan, 2014 (77)	1.249	0.981	1.589	1.808	0.071	36.13
Overall	0.799	0.523	1.220	-1.039	0.299	–

A risk ratio >1 is the increase in risk of the outcome when using intermittent infusion compared with continuous infusion.

Table 16 Comparison of continuous infusion and intermittent infusion method in all intensive-care unit patients included in meta-analysis

	Risk Ratio	Lower Limit	Upper Limit	Z-Value	p-value	Relative Weight
Wysocki 1995 (108)	0.667	0.133	3.354	-0.492	0.623	2.93
Wysocki 2001 (50)	0.864	0.397	1.881	-0.367	0.713	11.73
Hutschala, 2009 (105)	0.756	0.435	1.314	-0.991	0.322	21.22
Schmelzer, 2013 (109)	0.324	0.035	2.975	-0.996	0.319	1.70
Hanrahan, 2014 (77)	1.249	0.981	1.589	1.808	0.071	64.12
Overall	1.024	0.765	1.371	0.161	0.872	–

A risk ratio >1 is the increase in risk of the outcome when using intermittent infusion compared with continuous infusion

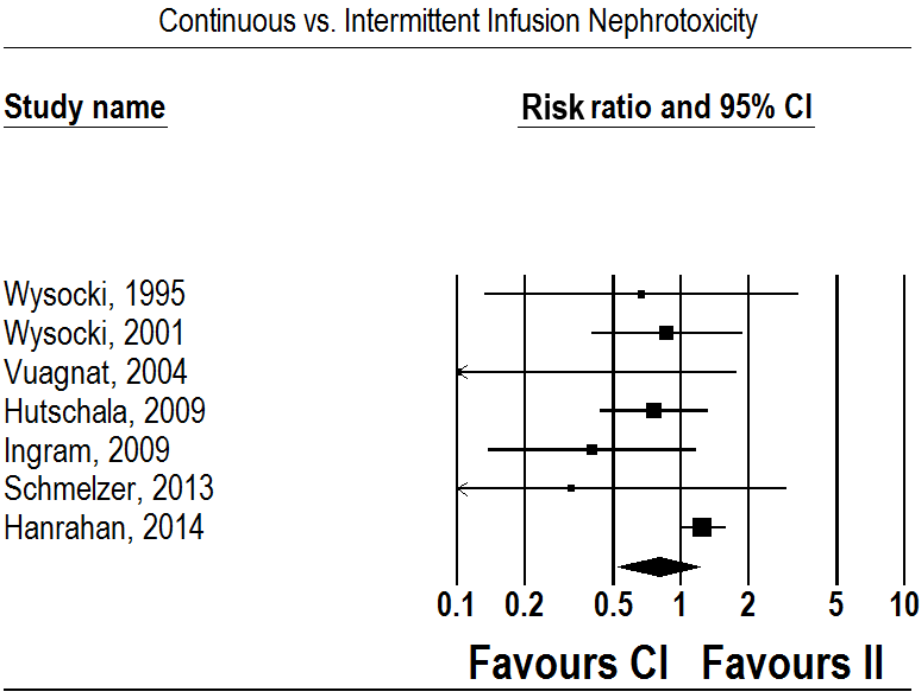

Figure 2 Forest plot comparing continuous infusion and intermittent infusion's influence on vancomycin-induced nephrotoxicity.
Continuous infusion, in all but Hanrahan et al (77) was associated with reduced odds of nephrotoxicity. Overall RR = 0.799 (0.523-1.220); p = 0.299
CI = Continuous infusion, II = Intermittent infusion

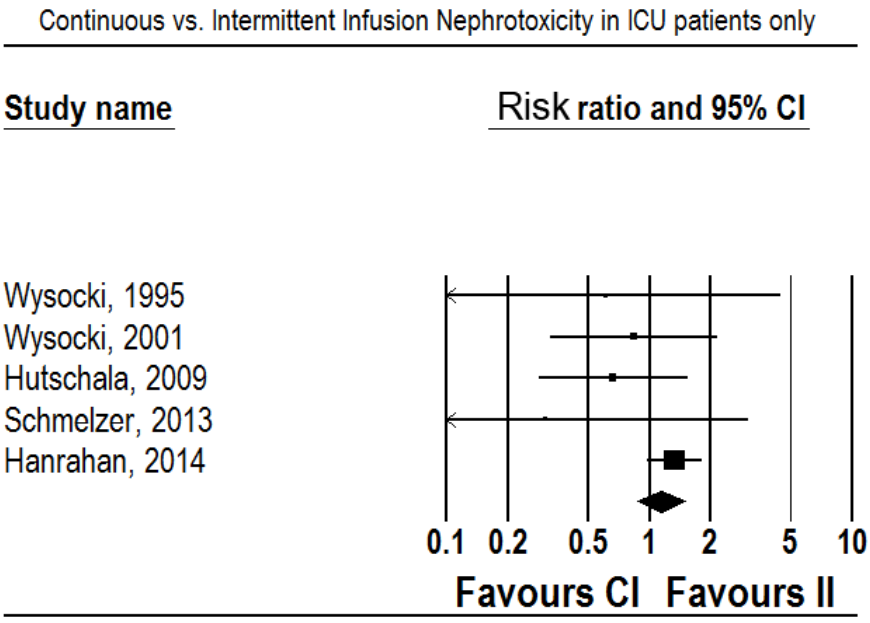

Figure 3 Forest plot comparing continuous and intermittent infusion's influence on vancomycin-induced nephrotoxicity in only those patients who were admitted to an intensive care unit.
Continuous infusion, in all but Hanrahan et al (77), was associated with reduced odds of nephrotoxicity. Overall RR = 1.024 (0.765 – 1.371); p = 0.872
CI = Continuous infusion, II = Intermittent infusion

5 Summary of findings, general discussion, future directions and conclusion

Despite being in use for more than 60 years, the optimal dosing regimen of vancomycin is still disputed. With rising resistance to vancomycin it is pertinent that dosing strategies be optimised to ensure antibiotic longevity and minimize adverse events. The aim of this thesis was to identify independent factors associated with vancomycin-associated nephrotoxicity.

Two patient populations were studied, and one meta-analysis was performed. The findings are summarised below.

5.1 Summary of results

Retrospective analyses of two critically ill patient populations were performed. The first analysis (77) included 1430 patients who were prescribed vancomycin at any time whilst admitted to the University Hospital Intensive Care Unit, Birmingham, UK. The prevalence of vancomycin-associated nephrotoxicity was 21. Concomitant vasoactive therapy (OR = 1.633), median serum vancomycin concentration (OR = 1.112) and duration of therapy (OR= 1.041) were significant positive predictors of nephrotoxicity. Furthermore, it was identified that patients who received vancomycin by II had significantly greater risk of nephrotoxicity (OR = 8.204) compared with those who received vancomycin by CI.

The second analysis included 159 patients who were prescribed vancomycin whilst admitted to the ICU of a tertiary hospital in Brisbane, Queensland, Australia. The prevalence of vancomycin-associated nephrotoxicity in this cohort was 8.8. Multivariate analysis identified mean trough concentration (OR = 1.174), APACHE II score (OR = 1.141), and simultaneous aminoglycoside prescription (OR = 18.896) as significant predictors of nephrotoxicity.

Finally, a meta-analysis and systematic review compare the incidence of vancomycin-associated nephrotoxicity between II and CI. A random effects model identified a non-significant trend of reduced nephrotoxicity in those who received vancomycin by CI (RR = 0.799).

5.2 General Discussion

Previously, robust data identifying predictors of vancomycin-associated nephrotoxicity were lacking. Despite studies identifying increased serum trough, daily doses >4 grams per day and increased duration of therapy as independent risk factors for toxicity (3,63,64), results were often skewed by failure to account for baseline illness severity and small patient cohorts. This thesis aimed to provide a large-scale study, to complement literature, and provide categorical conclusions.

Here, we have confirmed the results of previous studies with increased serum vancomycin concentrations and duration of therapy being identified as independent risk factors for nephrotoxicity. Furthermore, concomitant vasoactive therapy, increased illness-severity score (APACHE II) and concomitant aminoglycoside use were identified as independent predictors of nephrotoxicity.

The retrospective analysis reported in Chapter 4, to our knowledge, is the largest dataset to suggest CI is associated with reduced risk of nephrotoxicity. The meta-analysis confirms these results. Interestingly though, the multivariate analysis is at odds with the presented univariate analysis. On one hand, multivariate analysis has identified II as being associated with significantly more nephrotoxic consequences than CI (OR = 8.204). Conversely, the univariate analysis notes increased rates of nephrotoxicity in those treated by CI (24.7% vs. 19.7%). There is however a likely explanation for this result. Firstly, hospital policy dictated that administration of vancomycin by CI was limited to those with a central line. Immediately, the implication is that those receiving vancomycin by CI have greater morbidity, or a central line would not be necessary. Indeed, this is reflected by the significantly greater SOFA scores in those receiving vancomycin by CI (7 vs. 5 respectively; $p = <0.001$). Furthermore, those in the CI group received greater daily doses (1.7 vs. 1.5; $p = 0.003$), were more likely to receive simultaneous vasoactive therapy (71.8% vs. 45.4%; $p = <0.001$), were more likely to die in ICU (26.3% vs. 12.6%; $p <0.001$) and had a significantly greater duration of vancomycin therapy (5.3 vs. 4.4 days; $p = <0.001$). Ultimately, when controlling for these confounders in a logistic regression model, the results above are certainly conceivable.

Unfortunately, these results did not translate to mortality with II being non-significantly less predictive of mortality than CI (OR = 0.735; $p = 0.141$). This certainly suggests unmeasured confounders are present, an unfortunate limitation of retrospective analyses. Alternatively, a substantial proportion of patients receiving vancomycin therapy are treated empirically. Thus, it is plausible these patients did not have infections responsive to vancomycin. Ultimately, prospective randomised controlled trials are necessary to consolidate these results.

The abovementioned discordance provided an interesting result for the meta-analysis. Given meta-analyses are formulated using the incidence of events, the abovementioned confounders are not accounted for. Ultimately the largest study, and subsequently that with the greatest weighting included in analysis (77) was at odds with the remainder. Despite this, though non-significant, using a random effects model the final overall RR was 0.799 ($p = 0.299$). This, in conjunction with the multivariate analysis suggests CI should be the recommended administration method in critically ill patients.

5.3 Future Directions

This thesis provides substantial evidence that serum trough concentrations are indeed associated with vancomycin-associated nephrotoxicity. Furthermore, we have convincingly shown that CI is associated with significantly less nephrotoxicity than II. There are however, a number of limitations identified that call for further research, with possible avenues outlined below.

1. Both original studies included in this thesis study the relationship of vancomycin and nephrotoxicity by including the mean (or median) serum concentration in a regression model. Ultimately, by comparing II with CI, we are comparing serum trough concentrations with steady-state concentrations, respectively. This is indeed a limitation as the AUC is likely to be significantly different, and perhaps, introduce confounders. A retrospective study comparing the calculated area under the curve between continuous and II would be ideal.
2. A retrospective analysis cannot demonstrate causality. A well-designed, large prospective randomised controlled trial comparing CI and II with respect to both incidence of nephrotoxicity and mortality would provide irrefutable conclusions.

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