THE ASSOCIATION BETWEEN ANEURYSMAL SUBARACHNOID HAEMORRHAGE AND BIO MARKERS FOR RENAL DYSFUNCTION AT THE TIME OF ADMISSION.

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DECLARATION

I, Hlezikuhle Pethezinhle Nkala declare that this Research Report is my own work. It is being submitted for the degree of Master of Medicine at the University of the Witwatersrand, Johannesburg. It has not been submitted previously for any degree or examination at this or any other University.

(Signature of candidate)

8th day of October 2024

at Johannesburg.

DEDICATION

I dedicate this work to:

My parents: -

Bernard M. Nyoni

Nompumelelo Nyoni,

who have believed in and loved me since I was a child.

My husband: -

Walter Nkala,

who has supported all my dreams.

My daughters: -

Nkazimulo Muhle Nkala

Zoe Vuyelwa Nkala,

who have given me reasons to press on even when discouraged.

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I am also grateful to God Almighty, through His Son, Jesus Christ, who gave me the reason and purpose to perform the art of neurosurgery. To Him be glory.

ABBREVIATIONS AND NOMENCLATURE

aSAH	Aneurysmal subarachnoid haemorrhage
AKI	Acute Kidney Injury
AKIN	Acute Kidney Injury Network
CT-B	Computed Tomography of the Brain
KDIGO	Kidney Disease: Improving Global Outcome
GCS	Glasgow Coma Score
hs-CRP	highly sensitive C-reactive protein
MFS	Modified Fisher Scale
mg/ L	milligrams per litre
RIFLE	Risk, Injury, Failure, Loss, End-Stage
SAH	Subarachnoid haemorrhage
WFNS SAH	World Federation of Neurosurgical Societies Subarachnoid Hemorrhage Scale
µmol/ L	micromoles per litre
1SD	1 standard deviation

Nomenclature.

Time 0	0 hours: the time of admission of patient to hospital
Time 48	48 hours: 48 hours after admission of patient to hospital

ABSTRACT

INTRODUCTION

Rupture of a cerebral artery aneurysm is an event associated with intracranial and extracranial complications. Disturbance of renal function is said to be an extracranial complication but there is little information regarding incidence and time course of this problem. Changes in renal function may be assessed in several ways including change in serum creatinine levels over time.

AIM

The purpose of this study was to determine whether evidence of a risk of developing acute renal failure exists during the first 48 hours of hospitalisation in patients with aneurysmal subarachnoid haemorrhage (aSAH).

METHODS

Change in serum creatinine levels measured on admission and at 48 hours after admission in patients with documented aSAH was calculated. Patients were divided into two subgroups. Those in whom serum creatinine increased by more than 26.5 micromoles/litre (μmol/L) over 48hours were placed in Subgroup A. Those patients who did not demonstrate an increase of that magnitude were placed in Subgroup B.

RESULTS

Of the 23 patients who entered the study, 21 did not demonstrate a rise in serum creatinine of more than 26.5 μmol/L over 48 hours. Only 2 of 23 patients demonstrated such a change in serum creatinine. The proportion 21 of 23 patients is statistically significant ($p < 0.001$).

Serum creatinine levels, presented as mean ± 1 standard deviation (SD) was 63.0 ± 9.9 μmol/L and 65.8 ± 24.1 μmol/L, for Subgroups A and B, respectively. The difference is not significant. At 48 hours, the serum creatinine levels were 183.0 ±

36.8 µmol/L and 56.6 ± 17.0 µmol/, for Subgroups A and B, respectively. The difference is statistically significant ($p<0.05$).

For Subgroup B, the difference in serum creatinine on admission and at 48hours post admission was not statistically different.

CONCLUSION

Within the first 48hours of hospital admission 91.4% of patients with aSAH did not exhibit a rise in serum creatinine of such magnitude that they could be considered as at risk for developing acute renal failure.

KEY WORDS: aneurysmal subarachnoid haemorrhage aSAH), renal dyfunction risk, acute renal failure, creatinine: -admission, -48 hours

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1. INTRODUCTION

Within the cranial cavity the human brain is enclosed by 3 membranes:

- The pia mater which is intimately adherent to the surface of the brain.
- The arachnoid mater which lies superior to the pia mater.
- The dura mater which is the outermost membrane.

A potential space, termed the subarachnoid space, exists between the pia mater and arachnoid mater.

Blood flow is conducted to the brain via the left and right internal carotid arteries and the left and right vertebral arteries. These arteries combine within the cranial cavity to form a network of blood vessels that supply the various areas of the brain. The bulk of the intracranial blood vessels, from which smaller perforating branches conduct blood to brain tissue, lie within the subarachnoid space.

Vessels may have structural weak points of the wall. The arteries may also develop congenital or acquired area of malformation termed an aneurysm. Weak points and aneurysms have a propensity to rupture and consequently blood will escape under pressure from the blood vessel into the subarachnoid space and/ or into brain tissue.(1)

The term subarachnoid haemorrhage (SAH), indicates that bleeding has occurred from one or more of the intracranial blood vessels, and that free blood has accumulated within the subarachnoid space.(1)

SAH is divided into two categories:

- Spontaneous haemorrhage.
- Post-traumatic haemorrhage.

The most common cause of an intracranial spontaneous haemorrhage is rupture of a cerebral aneurysm.(2) SAH as a consequence of a ruptured cerebral aneurysm (aSAH) is a life-threatening event. The onset of haemorrhage is associated with many intra- and extra-cranial complications. Intracranial complications include ongoing or repeated episodes of bleeding, increase in intracranial pressure,

cerebral artery spasm, cerebral ischaemia, cerebral infarction, seizure activity and development of hydrocephalus. Extracranial complications, thought to be due to excessive systemic catecholamine release induced by the cerebral event, are intravascular volume depletion, acute hypertension, myocardial infarction, cardiac arrhythmia and pulmonary oedema. From about 30 years ago reports appeared suggesting that acute renal failure is an extracranial complication of SAH that has not been adequately recognized or emphasized.(3) Proponents admit that a causal link between acute renal failure and SAH has not been established.(4) However, the possibility of such a link has not yet been disproved. If the origin of systemic disturbances after cerebral aneurysm rupture is an excessive release of catecholamines, then the observation that acute renal failure is a sequel would make sense. However, the reports of renal failure following aneurysm rupture were not based on evidence gathered from prospective clinical studies. Thus, the suggestion that acute renal failure is an event directly caused by the presence of aSAH is not backed by objective information.

The purpose of this research project is to investigate whether or not patients with aSAH, who present within 96 hours of the onset of symptoms, exhibit - within 48 hours of admission to hospital - clinical or laboratory evidence that suggests they are at risk for subsequent development of acute renal failure.

2. LITERATURE REVIEW

2.1 Aneurysmal subarachnoid haemorrhage

The commonest cause of SAH is rupture of a cerebral artery aneurysm. This event accounts for about 85% of all spontaneous SAH.(2)

An extensive review of published literature on the incidence of aSAH was undertaken by de Rooij and co-workers.(5) This 2007 work reviewed 51 studies from 21 countries and concluded that the average global incidence of aSAH was 9 per 100,000 person-years. The evaluation noted regional differences in incidence throughout the world. The incidence in Japan was highest (22.7/ 100,000 person-years) while that of South

and Central America was low (4.2 per 100,000 person-years). The authors thought that these differences reflected the predominantly older population in Japan versus the predominantly young population found in South and Central America. Although aSAH may occur at any age the commonest group affected are those in the 60 -70 year age group.(5)

2.2 Renal dysfunction

"Renal dysfunction" is a shorthand medical term. Its use indicates that renal function is abnormal but does not specify the extent of the deterioration or the possible cause(s) of deterioration. It is a convenient term and is used in this report in the manner described above.

From 2004, various consensus definitions were proposed to standardize the usage of the phrase "renal dysfunction". Currently there are 3 widely accepted consensus statements:(6)

- Kidney Disease: Improving Global Outcome (KDIGO) criteria (10,14)
- Risk, Injury, Failure, Loss and End-Stage Renal Disease (RIFLE) criteria (8,10)
- Acute Kidney Injury Network (AKIN) criteria (7,10,13)

The precise criteria used by each proposal are detailed in Appendices 1, 2 and 3.

Prior to the acceptance of the consensus statements there were a number of studies, mostly retrospective, which drew attention to the fact that renal dysfunction occurred in patients with aSAH.(3,4) Comparing those studies was difficult because "renal dysfunction" does not have a precise meaning.

Once the consensus statements were published, studies using the various criteria began to appear in medical literature. Comparison of studies was now somewhat easier, but the use of different consensus proposals by different studies was still, to some degree, a hindrance.
The work of Gruber and co-workers (4) was followed by several more retrospective studies. The work of Mehta et al (7) - which used the AKIN criteria - examined the outcomes of AKI in groups of patients with aSAH. However, many published works

did not indicate when in the course of the illness, renal dysfunction appeared. (3,4,8,9,10,13) Thus, it is not possible to state whether renal dysfunction in patients with aSAH is an early complication or whether it appears in those patients who require long periods of support in the intensive care unit.

A retrospective study that evaluated the incidence and effect of renal dysfunction on survival and functional outcome of patients with aSAH was undertaken in by Zacharia *et al* in 2009.(8) Using RIFLE criteria, the study noted that 23% of patients developed risk for renal failure and that these patients had a poorer survival and functional outcome compared to those who did not exhibit risk. In contrast, the retrospective study of Rumalla et al demonstrated only a 4% incidence of acute renal failure.(9) Comorbidities such as diabetes mellitus, hypertension, congestive cardiac failure and obesity were associated with higher incidence of acute renal failure.(9) The strongest predictors of AKI included electrolyte disorders, diabetes mellitus, fluid status and the presence of a coagulopathy.(9)

More recently, some studies have tried to determine whether or not disturbance in renal function could be predicted from patient-related factors or laboratory investigations. The study of Eagles *et al*, (10) which defined renal dysfunction in terms of KDIGO criteria, indicated that cerebral angiography remains a risk factor for development of AKI in patients with pre-existing hypertension. A retrospective study by Sadan *et al* (11) suggested that the appearance of hyperchloraemia was associated with the development of renal dysfunction. However, the study could not explain whether the development of hyperchloraemia was the cause or the effect of renal dysfunction.

A prospective study in China was designed to determine whether the serum protein, highly sensitive C-reactive protein (hs-CRP) would be a reliable predictor of risk for acute renal dysfunction.(12) In a cohort of 164 patients, 13 patients out of 65 with a hsCRP blood level of 6.0 milligrams/ litre (mg/ L) or more developed acute renal dysfunction. Four of 99 patients with a blood level of 5.9 mg/ L or less developed acute renal dysfunction. These figures suggest that hs-CRP of 6 mg/ L or more has only a 20% positive predictive value for development of acute renal dysfunction.

In summary, various studies have examined, retrospectively, the incidence of renal dysfunction in aSAH patients. Other studies have evaluated possible patient-related

causes of renal dysfunction. Others have assessed the ability of serum markers to predict renal dysfunction.

A literature search for prospective studies that evaluated the risk for renal failure arising in the first 48 hours of hospitalization in patients with aSAH as made. No studies were found.

2.3 Renal dysfunction was a complication of subarachnoid haemorrhage

Many patients are admitted to Intensive Care Units because they have organ dysfunction.(4) However, some patients have no organ dysfunction on admission.(4) Nevertheless, all patients admitted to ICU have some degree of risk for organ dysfunction, even if organ function was normal at the time of admission.(4)
Organ dysfunction may be a consequence of the effects of the primary disease or of secondary physiological disturbances arising from complications such as hypovolaemia, infection, haemodynamic instability or hypoxaemia. Thus, if a patient admitted without evidence of organ dysfunction subsequently develops that problem it is not always possible to state whether the cause was a result of the primary disease or a result of secondary insults sustained while in ICU.(13)
Regardless of cause, the appearance of organ dysfunction may be abrupt and obvious or it may be initially silent and gradual. The threat and the consequences of organ dysfunction have prompted working groups to identify early warning signs of organ dysfunction.
In the case of renal dysfunction, the KDIGO group developed criteria that act as early warning signs for the possible onset of acute renal failure.(14) The working group defined the term “Acute Kidney Injury” (AKI). The definition states that AKI exists whenever there is an increase in serum creatinine of 26.5 micromoles/ litre (μmol/ L) or more from baseline over a period of 48 hours. Thus, a definition for kidney dysfunction in terms of a measurable quantity was created. Thus, change in serum creatinine of the magnitude mentioned above alerts clinicians to a possible risk of progression to acute renal failure if appropriate steps are not taken. Although AKI may be present, with appropriate medical intervention, the situation is potentially reversible.

2.4 Glasgow Coma Scale

The Glasgow Coma Scale is an accepted scale used in clinical practice for measuring the level of consciousness in a patient.(15) It provides an objective measure by which to describe depth of coma which in turn provides some insight into the severity of neurologic injury or dysfunction The scale utilises individual patient's responses to stimuli using responses to eye opening, motor response and verbal response. Appendix 4 summarises each of these responses and the numerical score assigned to each response. Finally, the 3 scores are summed to obtain what is called the Glasgow Coma Score (GCS). The latter is a range from 3 to 15 points; 15 out of 15 points indicating the best and 3 out of 15 points showing the worst possible scenario.

2.5 World Federation of Neurosurgical Societies Subarachnoid Hemorrhage Scale

The World Federation of Neurosurgical Societies Subarachnoid Hemorrhage (WFNS SAH) Scale was introduced in 1988.(16) The purpose of the scale was to provide an improved and objective description of severity of clinical presentations of patients with aSAH. The scale achieves this by combining GCS, which grades level of consciousness, with identification of the presence or absence of a focal motor deficit. As a result, the scale combines a predictor of mortality (GCS), with a predictor of future disability. The Scale has 6 grades, ranging from 0 to 5 with grade 5 indicating the most severe presentation. (See Appendix 5).

2.6 Fisher Scale and Modified Fisher Scale

The Fisher Scale was proposed in 1980.(17) The scale described the volume of blood in the subarachnoid space seen on the admission or first non-contrast computed tomographic images of the brain of patients with aSAH. The value of the scale was that it could be used to predict the chance of a patient developing cerebral artery vasospasm. The Fisher Scale was modified by Frontera *et al* (18) in 2006 and renamed Modified Fisher scale (MFS). The scale consists of 5 grades ranging from 0 to 4 with grade 4 indicating the highest risk of developing vasospasm. (See Modified Fisher Scale: Appendix 6)

3. JUSTIFICATION FOR THE RESEARCH

The following observations have been made in academic literature:

a) Renal dysfunction has been noted in patients with aSAH.

b) There is a lack of information as to whether the dysfunction is a direct result of the cerebral insult or of prolonged ICU stay.

c) Although predictive factors for risk of renal failure have been identified, those factors have not been used to identify whether or not the risk exists at the time of onset of aSAH.

The observations expressed in the literature review suggest that there is scope for prospective studies that seek to answer gaps in the medical knowledge surrounding the issue of renal dysfunction in patients with aSAH.

4. AIM OF THE RESEARCH

The purpose of this research project is to investigate whether or not patients with aSAH, who present within 96 hours of the onset of symptoms, exhibit - within the first 48 hours of their hospital admission – an increase in serum creatinine of more than 26.5 micromoles/ L (μmol/ L).

5. OBJECTIVES OF THE RESEARCH

The objectives of the research are:

Objectives:

- To investigate, in patients with documented aSAH and in whom symptoms of aneurysmal rupture have been present for 96 hours or less, the change in serum creatinine, the biomarker that indicates risk for renal dysfunction, over a 48-hour period commencing from the time of admission.
- To document GCS, WFNS SAH Scale grade and MFS grade in each study patient.

- To describe the haemorrhage from the responsible cerebral vessel(s) in terms of the anatomical location, size, and number of aneurysmal areas as evidenced by cerebral imaging.

6. ETHICS APPROVAL

- The Ethics Committee or Research on Human Subjects of the University of the Witwatersrand approved the study prior to commencement.
- Ethics Committee Approval Certificate No. M230628 is attached (see Appendix 8).
- Consent to carry out the study and for the use of data (clinical, laboratory and calculated) obtained during the study was sought from each patient or next of kin. Consent was requested from individual patients if they were compos mentis. However, for the comatose or confused patient, consent was obtained from the next of kin and also from the patient if he or she later recovered consciousness.
- An explanatory leaflet was prepared and given to each patient or next of kin and recovered patient.
- A consent form was prepared. Patients or next of kin were requested to sign the form if they agreed to give permission for the use of the information collected.
- Patients or next of kin were advised that they were under no obligation to give consent.
- Patients or next of kin were advised that confidentiality and anonymity would be preserved at all times during the study and in the future.

Additional approval for the research was obtained from

- Head of Department of Neurosurgery, University of Witwatersrand.
- Chief Executive Officer of each participating hospital.

7. METHODOLOGY

7.1 Study description

This study was conducted on a group of patients with a clinical diagnosis of SAH due to a ruptured cerebral artery aneurysm admitted to the Neurosurgical Units of 3 participating hospitals.

7.2 Participating Hospitals

The 3 participating hospitals were:

a) Charlotte Maxeke Johannesburg Academic Hospital

b) Chris Hani - Baragwanath Academic Hospital

c) Helen Joseph Hospital

These hospitals are the teaching hospitals associated with the University of Witwatersrand. Geographically, they are located within a 30-kilometre (km) radius of the centre of the City of Johannesburg, Republic of South Africa.

7.3 Study design:

- Prospective, observational, cross-over study.

7.4 Inclusion criteria:

- Patients 18 years of age or older who present with symptoms or signs of subarachnoid haemorrhage.
- Presentation within 96 hours of the onset of signs or symptoms.

7.5 Exclusion criteria:

- Patients in whom aSAH co-exists with another central nervous system pathology such as cerebral neoplasia, other cerebrovascular anomalies, or traumatic injury.
- Patients with known chronic kidney disease presenting with aSAH
- Patients who died within 48hours of admission to hospital.

7.6 Patients

7.6.1 General

Any patient with signs or symptoms of aSAH admitted to the Neurosurgical Units at the participating hospitals is managed according to a specific protocol. The protocol includes drawing of blood for a battery of routine blood tests on admission and then daily and arrangement for computerized tomographic imaging of the brain (CT-B) and/ or digital subtraction angiography by the duty doctor.

For the purpose of project described in this report the duty doctor notified the project investigator of the admission of any patient suspected to have aSAH.

Each patient or a family member of each patient with suspected aSAH was interviewed with a view to inclusion in the study. Those patients who agreed to participate and who met inclusion criteria comprised the initial study group. Only those in whom subsequent radiological confirmation of aSAH was obtained formed the final study cohort.

Once a patient entered the final study cohort the admission (Time 0) and 48-hour post-admission (Time 48) serum creatinine results were retrieved from the Hospital Laboratory Records System.

7.6.2 Subgroup Allocation

Once results of serum creatinine levels were known, each patient was allocated to a study subgroup as follows:

a) Subgroup A consisted of those patients who demonstrated an increase in serum creatinine of at least 26. 5 μmol/ L within the first 48 hours of admission to hospital.
b) Subgroup B consisted of those patients who did not demonstrate an increase in serum creatinine of more than 26.5 μmol/ L within the first 48 hours of admission to hospital.

7.6.3 Additional Patient Information

Additional information collected on each patient who entered the final study cohort was as follows:

- Age (years)
- Gender
- Social history
- Comorbidity history
- Anatomical site(s) of aneurysm(s)
- Number of aneurysms
- GCS
- WFNS
- MFS
- Definitive aneurysm treatment (clipping or coiling/ stenting)
- Total number of CT-B procedures performed per patient.

7.7 Definitions

7.7.1 Definition of “risk for development of acute renal failure”

A “risk for development of acute renal failure” was deemed present if the serum creatinine increased by more than 26.5 µmol/ L over a period of 48 hours.

7.8 Statistical Analysis

The primary purpose of the current project was to determine the proportion of patients who display a specific change in serum creatinine within 48 hours of admission to hospital. Based on published study information suggesting that the incidence of renal dysfunction risk is approximately 10 %, the required study sample size was calculated to be 11.5 patients. It was considered prudent to double the sample size to 23 patients. However, it is acknowledged that, because sample size

is small, statistical calculations of other patient characteristics that are not related to the primary purpose, may be prone to type 2 errors.

To describe patient demographic features, either numbers or the median and interquartile range (IQR) have been used to report categorical values. Continuous variables have been reported as the mean ± 1 standard deviation (1SD).

The statistical significance of the number of patients who demonstrated an increase in serum creatinine of more than 26.5 µmol/ L versus the number of patients who did not was determined from the z-test for a single proportion.

Since the number of patients in the study's two subgroups is very different (2 versus 21 patients) it was assumed that data is not normally distributed. Therefore, a decision to use a non-parametric test, namely the Wilcoxon Rank Sum Test, for comparison of patient age, GCS and between-group serum creatinine levels was made. When comparing serum creatinine levels of Subgroup B at Time 0 and Time 48, with 21 values at each time period, it could be argued that there would be normal data distribution. However, for consistency, and because 21 data points is a small number of values, use of the Wilcoxon Rank Sum Test was continued. The various calculations required to obtain a result from the Wilcoxon Rank Sum Test are set out in Appendix 7.

No attempt to statistically evaluate Subgroup A serum creatinine levels at Time 0 and Time 48 was made since there are only 2 data points for each time interval. Two samples containing two data points each is too small for meaningful statistical analysis.

The distribution of males and females between the two subgroups was compared using the Fisher exact test. The result should be accepted with caution since one cell of the calculation contains a zero value.

For WFNS scale and MFG scale, the number of patients per grade in each subgroup was documented but no statistical comparisons of differences were attempted.

Statistical significance was set at 5% (that is, a p-value equal to or less than 0.05).

8. **RESULTS**

During the period 23rd of October 2023 to the 22nd of January 2024, a total of 32 patients with clinical diagnosis of aSAH were reviewed. Nine patients did not meet the inclusion criteria, leaving a total of 23 patients who entered the final study cohort. These patients were, on the basis of change in serum creatinine over 48 hours, allocated to one of 2 subgroups. Those patients who exhibited a rise in serum creatinine of more than 26.5 µmol/ L over 48 hours were placed in Subgroup A (2 patients). Those who did not exhibit an increase in serum creatinine of the specified magnitude were placed in Subgroup B (21patients).

Table 1 shows the number of patients, the male: female distribution, patient age and admission GCS for each subgroup.

The age of patients (presented as mean ± 1SD) in Subgroup A was 67.5 ± 0.7 years and for Subgroup B was 43.7 ± 13.1 years. Although analysis indicated a statistically significant difference in age ($p \leq 0.05$) between the two subgroups of patients, this finding was not borne out clinically since patients of a similar age decade (60+ years) exist in both subgroups (see Appendix 7.1 for a breakdown of individual patient age). There was no statistically significant difference between the subgroups for male: female distribution nor for GCS.

Table 1. Characteristics of study patients for each study subgroup

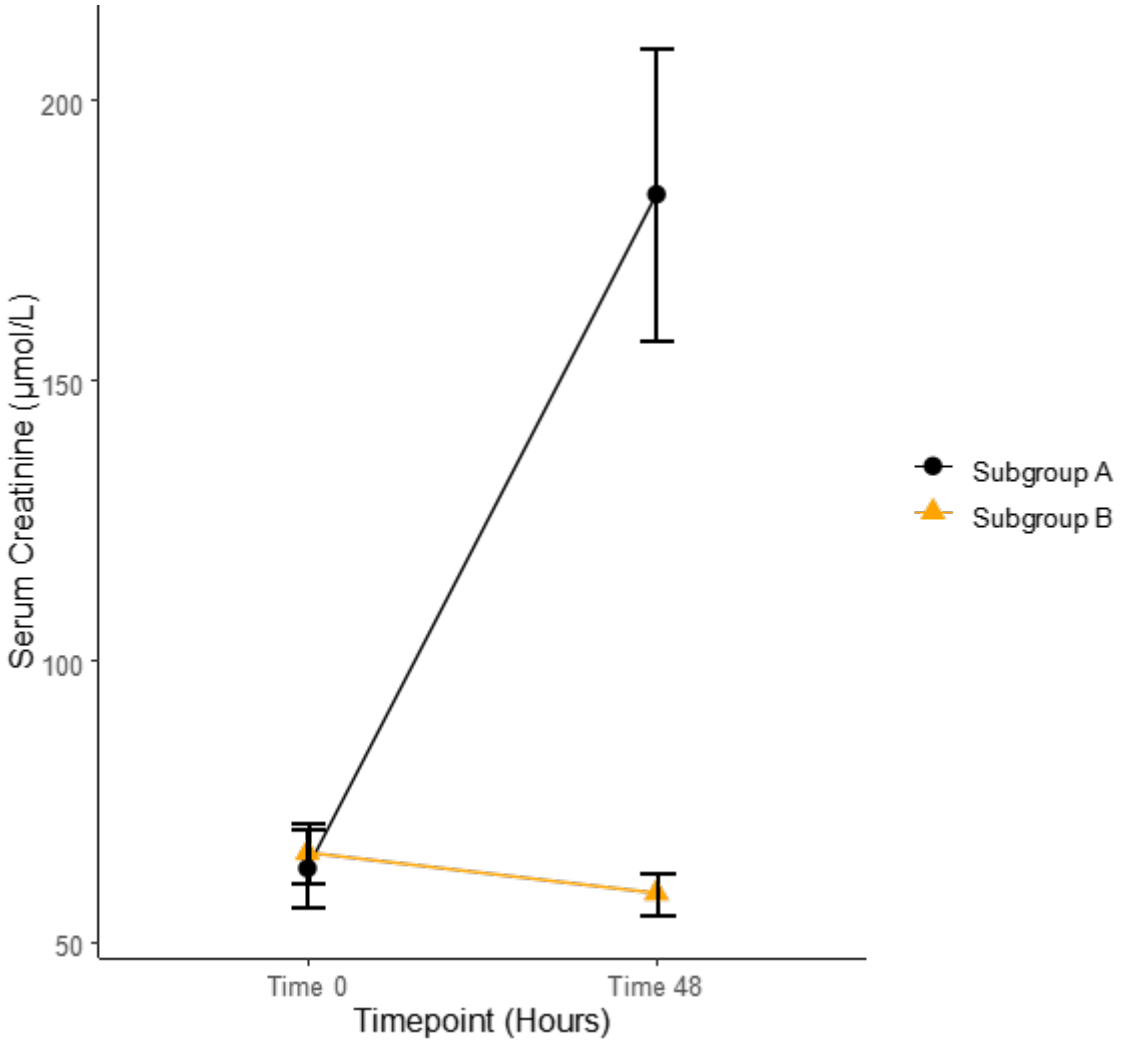
	Subgroup A	Subgroup B	Total
Number of patients	2	21	23
Male : Female distribution, n	2 : 0	13 : 8	15 : 8
Age (in years), mean ± 1SD	67.5 ± 0.7	43.7 ± 13.1	
Glasgow Coma Score, median (IQR)	7 (-,-)	15 (13,15)	

n = number. 1SD = 1 Standard Deviation. IQR = Interquartile Range. For Subgroup A patients an IQR for GCS cannot be calculated because there are only 2 data points.

A z-test proportional analysis of the number of patients in Subgroup A (2 patients) versus the number of patients in Subgroup B (21 patients) indicated that the difference was statistically significant ($p < 0.001$). In clinical terms, a significant number of patients with aSAH do not exhibit a rise in serum creatinine in excess of 26.5 μmol/ L within 48hours of admission to hospital. This does not mean such patients will not develop renal dysfunction at some later time in their hospital stay. However, it does suggest that a disturbance in renal function is not an immediate or early complication of aSAH.

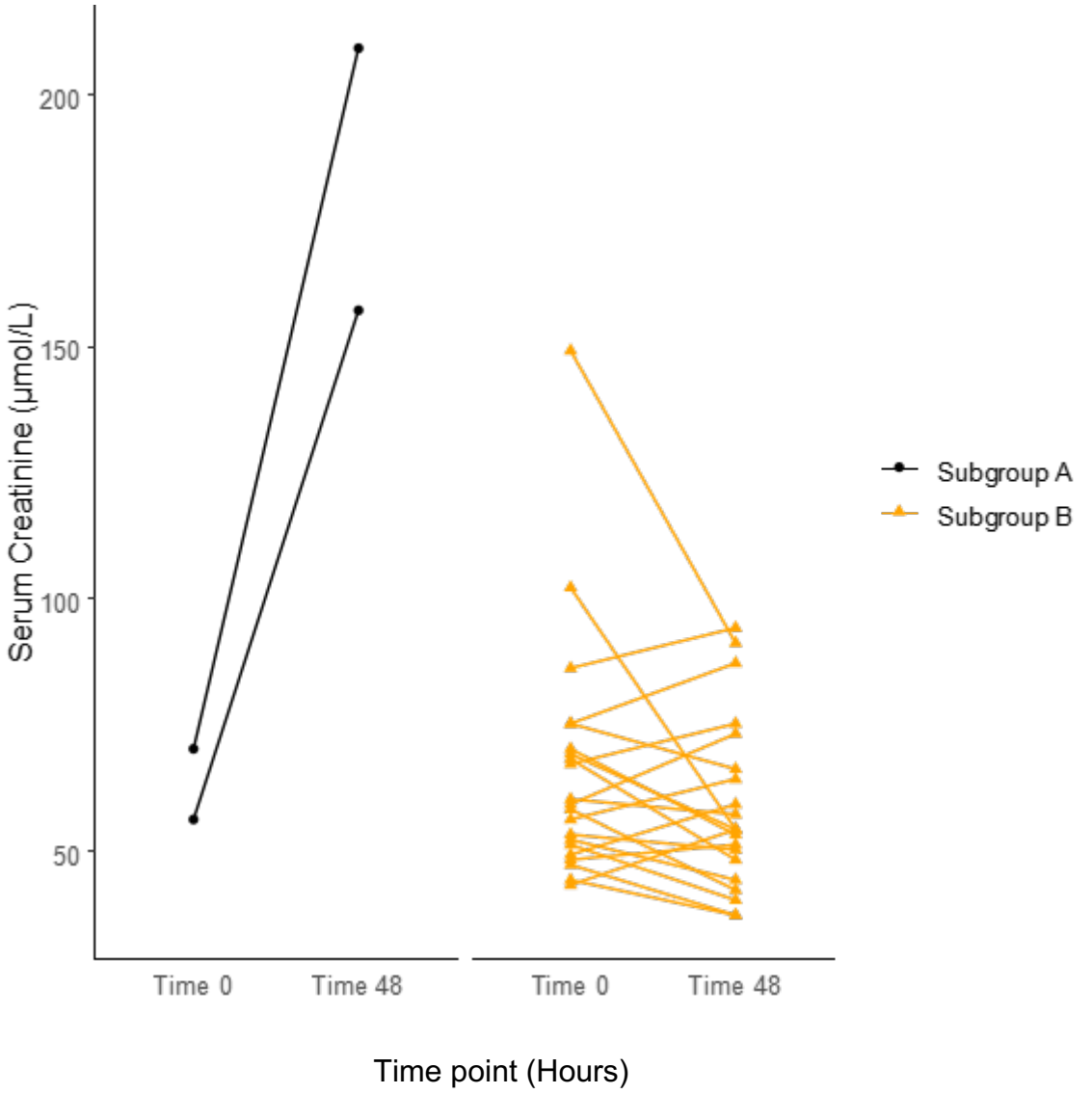
Serum creatinine (presented as mean ± 1SD) at Time 0 was 63.0 ± 9.9 μmol/ L and 65.8 ± 24.1 μmol/ L for Subgroup A and Subgroup B, respectively. This difference was not statistically significant. At Time 48 the serum creatinine levels for Subgroup A had increased to 183.0 ± 36.8 μmol/ L while that for Subgroup B had decreased to 58.6 ± 17.0 μmol/ L. This difference was statistically significant ($p \leq 0.05$). The levels of serum creatinine for Subgroup B at Time 0 and Time 48 (65.8 ± 24.1 versus 58.6 ± 17.0 μmol/ L) were not statistically different. Change in serum creatinine between Time 0 and Time 48 for Subgroup A were not analysed statistically because there are only two data points for each time period. However, the change in serum creatinine does show that a clinically important deterioration in renal function has occurred in both of those patients.

Figure 1 depicts the changes in serum creatinine measured in μmol/ L and displayed as mean ± 1SD at the two study time intervals (Time 0 and Time 48). Figure 2 shows the individual changes in serum creatinine at the study time intervals for each patient in the two study subgroups (see Appendix 8 for a breakdown of individual patient results).



("Time 0" indicates time of admission to hospital, "Time 48" indicates 48 hours post-admission).

Figure 1: Change in serum creatinine in micromoles per litre presented as mean ± 1 standard deviation for subgroup A and subgroup B patients over the 48-hour study period.



("Time 0" indicates time of admission to hospital, "Time 48" indicates 48 hours post-admission).

Figure 2: Change in serum creatinine in micromole per litre for individual study patients in subgroup A and subgroup B over the 48-hour study period.

Only 2 out of 23 study patients (8.6%) demonstrated an increase in serum creatinine over 48 hours. Both these patients were located in subgroup A. In terms of KDIGO criteria, those 2 patients can be categorized as having AKI and being at risk for the development of acute renal failure. The remaining 21 patients, who were located in subgroup B, did not demonstrate an increase in serum creatinine over 48 hours. These patients are not at risk for AKI within the first 48 hours of their hospitalisation. Of importance is the observation that 13 of the 21 subgroup B (61.9%) patients

demonstrated a decrease in serum creatinine over 48 hours. This most likely reflects in-hospital intravascular volume replenishment prescribed by attending clinicians.

Tables 2 and 3 indicate, for each subgroup, the number of patients associated with various social habits and comorbidities per study subgroup, respectively. Of interest is the finding that there were more patients in Subgroup B, the subgroup not associated with deterioration in renal function, presenting with hypertension, insulin-dependent diabetes mellitus, dyslipidaemia, confirmed presence of retroviral disease and history of cigarette smoking than there were in Subgroup A.

Table 2: Distribution of patients between study subgroups according to aspects of social history

	Subgroup A	**Subgroup B**	**Total**
Social History:			
Alcohol Consumption:			
Yes	1	1	2
No	0	5	5
Unknown	1	15	15
Recreational Drug Use:			
Yes	0	2	2
No	0	6	6
Unknown	2	13	15
Cigarette Smoking:			
Yes	1	7	8
No	0	3	3
Unknown	1	11	12

Table 3: Distribution of patients between study subgroups according to presence or absence of co-morbidity

	Subgroup A	Subgroup B	Total
Co-Morbidity:			
Yes	2	11	13
No	0	10	10
Hypertension:			
Yes	2	8	10
No	0	13	13
Diabetes mellitus:			
Yes	0	2	2
No	0	21	21
Retroviral disease:			
Yes	0	5	5
No	0	10	10
Unknown	2	6	8
Asthma:			
Yes	0	1	1
No	2	20	22
Obesity:			
Yes	0	1	1
Not measured	1	21	22
Dyslipidaemia			
Yes	1	3	4
No	0	0	0
Unknown	1	18	19

Table 4 shows the total number of contrast-associated CT-B procedures required to obtain a conclusive diagnosis of aneurysm rupture. Every study patient received at least 1 contrast-associated CT-B procedure within the first 48 hours of hospital admission. Although not part of the initial 48 hours of hospitalisation the total number of contrast-associated CT-B procedures is documented here to emphasise the fact that patients with aSAH may be exposed to several doses of radiographic contrast media. During the entire hospitalisation period 9 and 5 Subgroup B patients received 2 and 4 contrast-associated interventions, respectively.

Table 4: Number of contrast-associated computed tomographic brain imaging procedures per study patient

Number of contrast – associated brain imaging procedures.	**Subgroup A**	**Subgroup B**	**Total**
0	0	0	0
1	2	4	6
2	0	9	9
3	0	3	3
4	0	5	5

The anatomical locations of the ruptured cerebral aneurysms are shown in Table 4.

Table 5: Anatomical location of ruptured aneurysm: Distribution of patients amongst the various anatomical locations of identified ruptured aneurysms for each study subgroup

Cerebral Blood Vessel	Subgroup A	Subgroup B	Total
Anterior Communicating Artery	0	6	6
Basilar Tip Artery Aneurysm	1	2	3
Distal Anterior Cerebral Artery Aneurysm	0	5	5
Left Middle Cerebral Artery (M1 segment)	1	1	2
Left Middle Cerebral Artery (M4 segment)	0	1	1
Left Posterior Communicating Artery	0	1	1
Right Middle Cerebral Artery (M1 segment)	0	3	3
Right Middle Cerebral Artery (M2 segment)	0	1	1
Right Posterior Communicating Artery	0	1	1

The anatomical location of cerebral aneurysm was widespread.

Figure 3 shows the distribution of patients amongst the various grades of the WFNS SAH Scale. With the exception of Grade 0, Subgroup B patients were distributed throughout all 5 grades of the scale although the majority (13 patients) were Grade 1 patients. There were 4 Grade 4 patients: 2 in Subgroup A and 2 in Subgroup B.

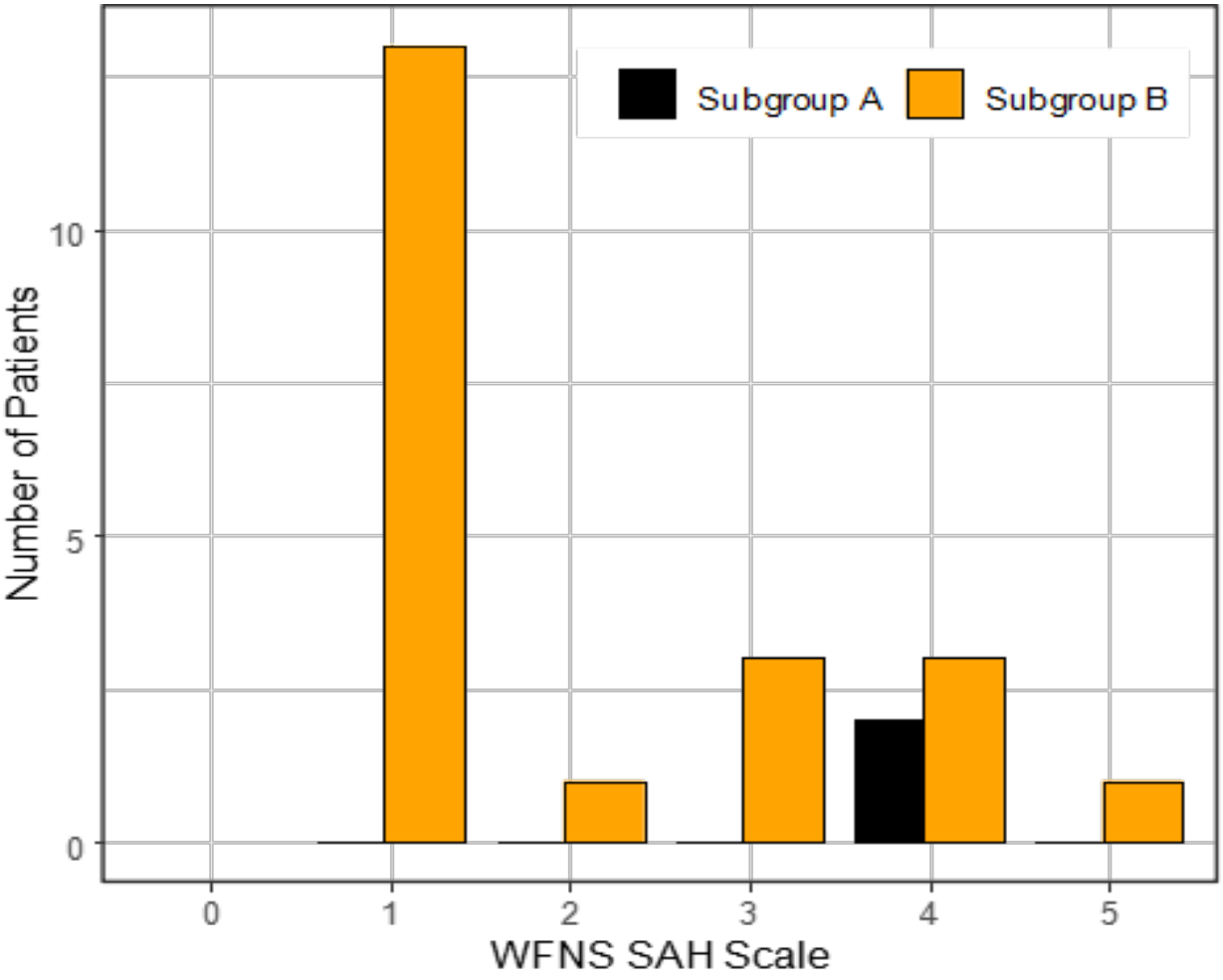


Fig3: World Federation of Neurosurgical Societies Subarachnoid Haemorrhage (WFNS SAH) Scale: Distribution of patients amongst the various grades for each study subgroup

Figure 4 shows the distribution of patients amongst the MFS grades. Both patients in Subgroup A were classified MFS Grade 4. There were 12 Subgroup B patients with a MFS Grade 4 classification. With the exception of Grade 0, Subgroup B patients were distributed throughout all of the MFS grades.

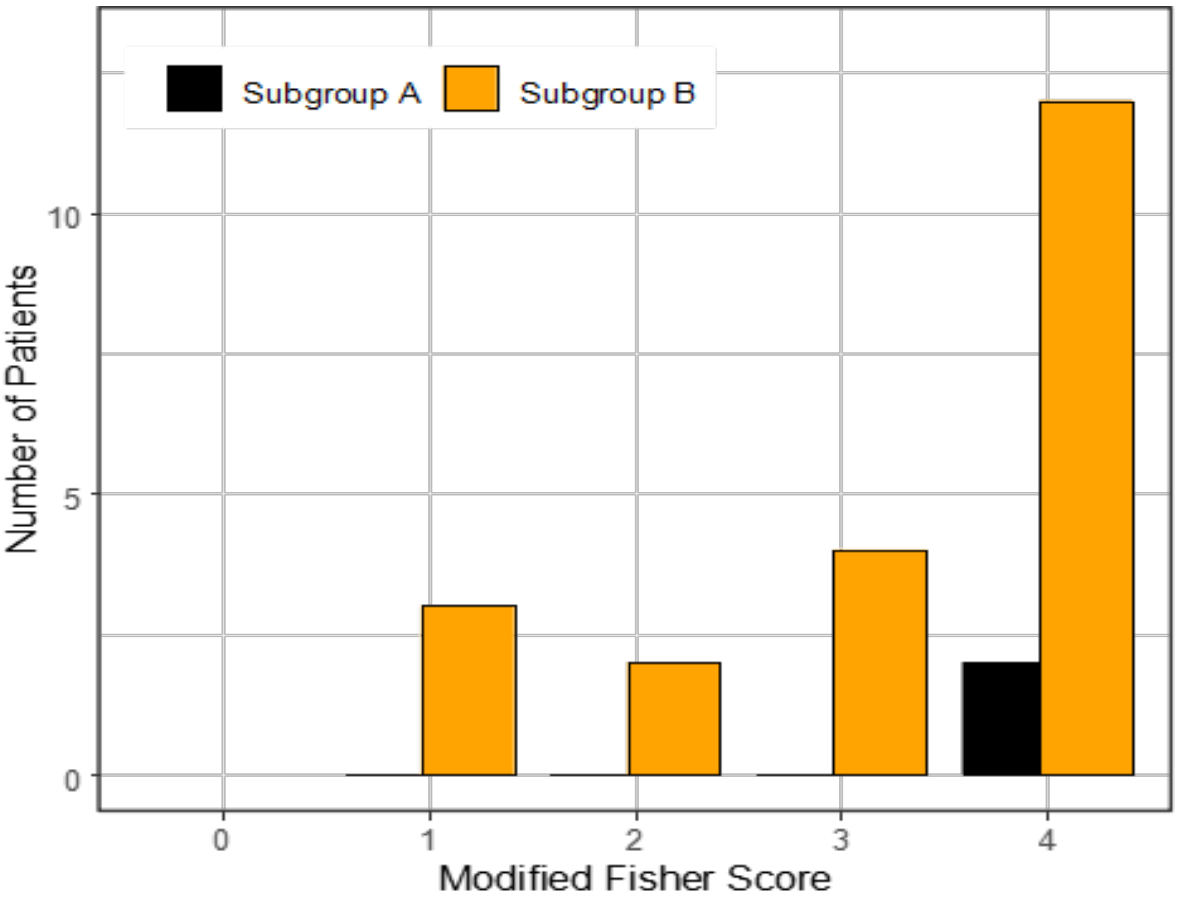


Fig 4: Modified Fisher Scale. Distribution of patients amongst the various grades for each study subgroup

These results show that patients with aSAH have a wide range of clinical presentations in terms of the WFNS SAH Scale and the MFS.

The definitive treatment received by the study patients is shown in Table 6. The majority of patients underwent a radiological procedure, namely angiography with digital subtraction imaging, and endovascular coiling of the aneurysmal defect.

Table 6: Definitive treatment

Definitive Treatment of the ruptured aneurysm	Subgroup A	Subgroup B	Total
Clipping	0	1	1
DSA and Coiling	0	11	11
DSA Only	0	3	3
Not Done	2	4	6
Missing/Not applicable	0	2	2

9.DISCUSSION

Spontaneous SAH is a consequence of vascular diseases of the central nervous system. Rupture of a cerebral artery aneurysm, a life-threatening event, is the most frequent cause of spontaneous SAH – accounting for about 85% of presentations.(2) The global incidence of aSAH is about 9/ 100,000 person-years.(5) Early mortality from aSAH is mostly related to the amount of blood released at the time of rupture - the larger the volume the greater the chance of early demise. About 30% of patients die, with or without medical care, in the first 24 hours following rupture of the aneurysm.(19) For alert patients who are alive at 24 hours post-rupture and undergo an appropriately timed surgical procedure, the mortality rate at 6 months post-intervention is about 8%. In contrast, comatose patients, alive at 24 hours post-rupture who undergo appropriately timed surgery have about a mortality rate of about 45% by 6 months post-intervention.(19)

The emphasis of management of the patient with aSAH is surgical exclusion of the aneurysm or aneurysms from the cerebral circulation. The suitability of a patient for surgical intervention and the timing of surgical intervention is largely governed by four clinical concerns:

a) The presence of intracranial complications
b) The depth of coma at presentation
c) The duration of time since the start of cerebral haemorrhage
d) The presence of extracranial disturbance of major organ function

As a generalization, early mortality is more often related to intracranial complications of aSAH and later mortality is more likely to be due either to extracranial complications of aSAH or to an unrelated secondary intercurrent illness or a pre-morbid condition.

Prior to 1995, most medical literature describing extracranial complications of aSAH did not mention renal dysfunction as a disturbance of note. The 1995 work of Solenski *et al* (3) drew attention to the fact that renal dysfunction was a medical complication seen in patients with aSAH. The 1999 work of Gruber *et al* (4) suggested that, not only was renal failure a medical complication associated with the presence of aSAH, but its presence was also associated with an increase in the mortality rate of such patients. This study was a retrospective analysis and the authors agreed that their work did not establish a causal link between aSAH and the appearance of renal failure. The authors also agreed that most of the deaths in patients with renal failure could be ascribed to the presence of a Systemic Inflammatory Response Syndrome with and without bacterial infection.

Gruber and co-workers' study (4) was later complimented by several studies that examined the incidence of renal disturbance and its effect on the outcome of patients with aSAH. At this stage evaluating, comparing and contrasting literature becomes complicated because authors used a variety of definitions to describe the degree of renal function disturbance. Some studies discussed "renal dysfunction" while others discussed the incidence of "renal failure". To complicate matters further some articles document "risk of renal failure" and others document "risk of renal injury". For example, Solenski *et al* (3) reported an incidence of "renal dysfunction" in up to 7% of patients with aSAH. Zacharia and co-workers (8), who used both RIFLE criteria and the calculated creatinine clearance as indicators of renal disturbance, documented a 23.1% "risk for renal failure" in patients with aSAH.

More recently consensus statements have provided nomenclature and definitions for various stages of renal function disturbance.(6, 14) In particular, the definition of AKI identified the kidney that was at risk of developing acute renal failure. This situation was said to be present when the serum creatinine increased by more than 26.5 µmol/ L within a period of 48-hours.(6, 14) Consequently, a defined and measurable change by which patients could be consistently and accurately identified was now in place.

Using the AKIN criteria to identify the presence of AKI, Tujjar and co-workers (13) undertook a retrospective analysis of 243 patients with non-traumatic SAH admitted to an Intensive Care Unit. The authors noted a 12% incidence of AKI in their patient group. Since this study evaluated the patients throughout their stay in the Intensive Care Unit the authors were able to identify the median time to appearance of AKI as 8 days after admission to the Unit. As a result, the authors concluded that AKI was not a complication of the early phase of non-traumatic SAH. The study also showed that older patients and those with higher WFNS SAH and MFS grades had poor neurological outcome but, most importantly, renal dysfunction was not an independent predictor of either poor neurological outcome or of mortality.

The project presented in this document (hereafter referred to as “the current project”) differs from the work of Tujjar et al.(13) In contrast to Tujjar’s work, the current project used the KDIGO criterion of change in serum creatinine over a 48-hour period beginning at the time of admission to determine the incidence of risk of developing acute renal failure in a group of patients with aSAH. In terms of patient numbers, the current project is smaller in numbers and patients were only followed for 48 hours from admission. With these differences in mind, the current study found that a change in serum creatinine of more than 26.5 µmol/ L in the first 48 hours of hospitalization occurred in only 8.6% of patients with aSAH.

Co-morbid conditions such as diabetes mellitus, hypertension and obesity and acquired critical illnesses such as infection and septic shock are known to increase the risk of developing acute renal failure in any critical ill patient.(8,9) As a consequence, if dysfunction of any major organ system arises during the course of a patient’s stay in intensive care, determining whether the precipitating cause is the primary illness or a result of a co-morbid condition or a new secondary event is

difficult. In the current project various co-morbid conditions and social habits that influence health were found in both subgroups of the study patients. This suggests that the co-existing conditions did not have an immediate influence on renal function in the first 48 hours of the study patients' hospitalisation. However, this situation does not exclude the possibility of an influence on patient progress at a later time in the course of the illness.

10. CONCLUSION

The current project determined that the percentage of patients with aSAH who, within 48 hours of admission, did not display a risk for developing acute renal failure was 91.4%. This suggests that the haemorrhagic event is not the cause of early disturbance in renal function in patients with aSAH. This does not mean that such patients are immune from developing acute renal failure at a later stage of their illness. Consequently, regular evaluation of renal function of patients with aSAH during their hospitalization remains appropriate medical care.

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12. APPENDICES

Appendix 1

Kidney Disease: Improving Global Outcome criteria:

For acute kidney dysfunction:

Stage 1: Serum creatinine increase by 26.5 micromoles per litre, or 1.5 - 2 times increase from baseline in 48hours; urine output of <0.5ml/kg/h for >6hours.

Stage 2: >2 - 3 times increase from baseline of serum creatinine; urine output <0.5ml/kg/h for 12hours.

Stage 3: >3times serum creatinine increase from baseline or >/= 350 micromoles per litre or initiation of renal replacement therapy or in patients younger than 18years, decrease of eGFR to < 35ml/min per 1.73m^2; or urine output of <0.3ml/kg/h for 24 hours or anuria for 12 hours.

Appendix 2

Risk, Injury, Failure, Loss and End-Stage Renal Disease criteria:

Risk: 1.5 times increase of baseline serum creatinine, eGFR decreased >25%; urine output of <0.5ml/kg/h for 6-12hours.

Injury: 2 times increase of baseline serum creatinine, eGFR decreased >50%; <0.5ml/kg/h for 12hours.

Failure: 3 times increase of baseline serum creatinine, or serum creatinine >/= 350 micromoles per litre, or its acute rise by 44 micromoles per litre, eGFR >75% or initiation of renal replacement therapy; or urine output of <0.3ml/kg/h for 24 hours or anuria for 12 hours.

Loss: requirement of renal replacement therapy for > 4weeks.

End-Stage: requirement of renal replacement therapy for > 3months.

Appendix 3

Acute Kidney Injury Network criteria:

Risk: Serum creatinine of 1.5 -1.9 times increase from baseline value or increase of 26.5 micromoles per litre within 48hours; urine output of <0.5ml/kg/h for >/= 6 hours. Injury: >2 - 3 times increase of baseline serum creatinine; urine output <0.5ml/kg/h for 12hours.

Failure: >3 times increase of baseline serum creatinine; urine output of <0.3ml/kg/h for 24 hours or anuria for 12 hours.

Appendix 4

Glasgow Coma Scale

Variable:	Point Score:
Eye Opening:	
Spontaneous	4
To speech	3
To pain stimulus	2
None	1
Best Motor Response:	
Obeys commands	6
Localises to stimulus	5
Normal Flexion	4
Abnormal Flexion	3
Extension to stimulus	2
None	1
Verbal Response:	
Orientated	5
Confused	4
Inappropriate words	3
Sounds	2
None	1

The Glasgow Coma Score is the summation of the individual points obtained for each Variable. Minimum score is 3 points. Maximum score is 15 points.

Appendix 5

World Federation of Neurosurgical Societies Subarachnoid Hemorrhage Scale

Grade:	Description:
0.	Intact aneurysm
1.	GCS 15/15, absent motor deficit.
2.	GCS 13-14/15, absent motor deficit.
3.	GCS 13-14/15, motor deficit present.
4.	GCS 7-12/15, motor deficit present or absent.
5.	GCS 3-6/15, motor deficit present or absent.

Appendix 6

Modified Fisher Scale

Grade:	Description:
0	No visible blood.
1	Thin SAH, focal or diffuse, with no intraventricular haemorrhage.
2	Thin SAH, focal or diffuse, with intraventricular haemorrhage.
3	Thick SAH, focal or diffuse, with no intraventricular haemorrhage.
4	Thick SAH, focal or diffuse, with intraventricular haemorrhage.

Appendix 7

Statistical Information

Explanation: The Wilcoxon Rank Sum Test is a non-parametric statistical test. The test ranks individual results obtained from study samples in ascending numerical order. When two or more results are the same the ranks for those results are averaged. Ranks are then summed and the total score of the smaller of the two samples is evaluated using a table of critical values (if two samples are of equal size then either of the rank sums can be chosen for evaluation). If, for a two group study with 2 patients in one group and 21 patients in the other, the numerical sum of the ranks of the smaller sample score falls outside the range 7 – 41, then a statistically significant difference at the 5% level is present. For two samples, each of 21 patients, the rank sum must fall outside the range 374 – 529 to be statistically significant.

The table of critical values was obtained from a publication by Kirkwood (20) who, in turn, acknowledged reproduction of the table (with permission) from the work of Colton.(21)

7.1 Calculation of the Wilcoxon Rank Sum for Age (in years) for study patients in Subgroup A and Subgroup B

	Subgroup A		Subgroup B	
Arbitrary Patient Number	Age (in years)	Rank	Age (in years)	Rank
1	67	21		
2	68	22		
3			23	1
4			25	2.5
5			25	2.5
6			28	4
7			32	5
8			35	6.5
9			35	6.5
10			39	8
11			40	9
12			42	10
13			43	11
14			44	12
15			45	13
16			47	14
17			50	15.5
18			50	15.5
19			55	17
20			60	18
21			65	19
22			66	20
23			69	23
Rank Sum		**43**		**233**

The Rank Sum for Subgroup A is greater than 41. Therefore, a statistical difference at the 5% level exists between the two study subgroups in terms of age.

7.2 Calculation of the Wilcoxon Rank Sum for the Glasgow Coma Score for study patients in Subgroup A and Subgroup B

	Subgroup A		Subgroup B	
Arbitrary Patient Number	GCS	Rank	GCS	Rank
1	7	3.5		
2	7	3.5		
3			4	1
4			7	3.5
5			7	3.5
6			8	6
7			10	7
8			13	8
9			14	9.5
10			14	9.5
11			15	16
12			15	16
13			15	16
14			15	16
15			15	16
16			15	16
17			15	16
18			15	16
19			15	16
20			15	16
21			15	16
22			15	16
23			15	16
Rank Sum		**7**		**256**

The Rank Sum for Subgroup A does not fall outside the range 7 – 41. Therefore, there is no statistical difference between the two study subgroups in terms of Glasgow Coma Score.

7.3 Calculation of the Wilcoxon Rank Sum for serum creatinine at T0 for study patients in Subgroup A and Subgroup B

	Subgroup A: Time 0		Subgroup B: Time 0	
Arbitrary Patient Number	Serum Creatinine (micromole/ L)	Rank	Serum Creatinine (micromole/ L)	Rank
1	56	9.5		
2	70	17.5		
3			43	1
4			44	2
5			47	3
6			48	4
7			49	5
8			51	6
9			52	7
10			53	8
11			56	9.5
12			58	11
13			59	12
14			60	13
15			67	14
16			68	15
17			69	16
18			70	17.5
19			75	19.5
20			75	19.5
21			86	21
22			102	22
23			149	23
Rank Sum		**27**		**229.5**

The Rank Sum for Subgroup A does not fall outside the range 7 – 41. Therefore, there is no statistical difference between the two study subgroups in terms of serum creatinine levels at Time 0.

7.4 Calculation of the Wilcoxon Rank Sum for serum creatinine at Time 0 and Time 48 for study patients in Subgroup B

	Subgroup B: Time 0		Subgroup B: Time 48	
Arbitrary Patient Number	Serum Creatinine (micromole/ L)	Rank	Serum Creatinine (micromole/ L)	Rank
1	43	5	37	1.5
2	44	6.5	37	1.5
3	47	8	40	3
4	48	9.5	42	4
5	49	11	44	6.5
6	51	13.5	48	9.5
7	52	14	50	12
8	53	15.5	51	13.5
9	56	20	53	15.5
10	58	22	54	18
11	59	23.5	54	18
12	60	25	54	18
13	67	28	57	21
14	68	29	59	23.5
15	69	30	64	26
16	70	31	66	27
17	75	34	73	32
18	75	34	75	34
19	86	36	87	37
20	102	40	91	38
21	149	41	94	39
Rank Sum		**476.5**		**398.5**

The Rank Sum for Subgroup B does not fall outside the range 374 – 529. Therefore, there is no statistical difference between the serum creatinine at Time 0 and Time 48 for study patients in Subgroup B.

7.5 Calculation of the Wilcoxon Rank Sum for serum creatinine at Time 48 for study patients in Subgroup A and Subgroup B

	Subgroup A: Time 48		Subgroup B: Time 48	
Arbitrary Patient Number	Serum Creatinine (micromole/ L)	Rank	Serum Creatinine (micromole/ L)	Rank
1	157	22		
2	209	23		
3			37	1.5
4			37	1.5
5			40	3
6			42	4
7			44	5
8			48	6
9			50	7
10			51	8
11			53	9
12			54	11
13			54	11
14			54	11
15			57	13
16			59	14
17			64	15
18			66	16
19			73	17
20			75	18
21			87	19
22			91	20
23			94	21
Rank Sum		**45**		**231**

The Rank Sum for Subgroup A is greater than 41. Therefore, a statistical difference at the 5% level exists between the two study subgroups in terms of serum creatinine levels at Time 48.

Appendix 8

Individual and summed results for serum creatinine at Time 0 and Time 48 for each patient in Subgroup A and Subgroup B

	Serum creatinine (micromoles/ L)			
	Subgroup A		Subgroup B	
Arbitrary Patient Number	Time 0	Time 48	Time 0	Time 48
1	70	209		
2	56	157		
3			67	75
4			69	54
5			149	91
6			75	66
7			49	59
8			52	44
9			56	64
10			48	51
11			43	54
12			59	73
13			60	57
14			70	53
15			86	94
16			44	37
17			47	37
18			51	40
19			75	87
20			58	42
21			53	50
22			68	48
23			102	54
Mean ± 1 SD	**63.0 ± 9.9**	**183.0 ± 36.8**	**65.8 ± 24.1**	**58.6 ± 17.0**

SD = standard deviation

Appendix 9

Ethics Clearance Certificate

UNIVERSITY OF THE WITWATERSRAND, JOHANNESBURG

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)

Office of the Deputy Vice-Chancellor (Research and Innovation)

TO: Dr HP Nkala
School of Clinical Medicine
Department of Surgery
Division of Neurosurgery
Medical School
University

E-mail: hlezi@yahoo.co.uk

CC: Supervisor: Professor J Ouma
<jmno@mweb.co.za>
and <HREC-Medical Research Office@wits.ac.za>

FROM: Mr Iain Burns
Human Research Ethics Committee (Medical)
Tel: 011 717 1252

E-mail: Iain.Burns@wits.ac.za

DATE: 2023/10/23

REF: R14/49

PROTOCOL NO: **M230628** (This is your ethics application reference number. Please quote it in all enquiries, oral or written, relating to this study.)

PROJECT TITLE: *The association between aneurysmal subarachnoid haemorrhage and bio markers for renal dysfunction at the time of admission*

Please find attached the Clearance Certificate for the above project. I hope it goes well and that an article in a recognized publication comes out of it. This will reflect well on your professional standing and contribute to Government funding of the University.

MSWorks2000/iain0007/Clearscan.wps

UNIVERSITY OF THE WITWATERSRAND JOHANNESBURG

R49 Dr HP Nkala

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)
CLEARANCE CERTIFICATE NO. M230628

NAME: Dr HP Nkala **DEGREE:** MMed
(Principal and Co-Investigators)

DEPARTMENT: School of Clinical Medicine
Department of Surgery
Division of Neurosurgery
Medical School
University

PROJECT TITLE: *The association between aneurysmal subarachnoid haemorrhage and bio markers for renal dysfunction at the time of admission*

DATE CONSIDERED: 2023/06/30

DECISION: Approved unconditionally

CONDITIONS:

NOTE: If contact information regarding student study participants is required, please contact the Registrar's office - <Nicoleen.Potgieter@wits.ac.za>

SUPERVISOR: Professor J Ouma

APPROVED BY:
Ms M van Niekerk, Co-Chairperson, HREC (Medical)

DATE OF APPROVAL: 2023/10/23 **EXPIRY DATE:** 2028/10/22

This Clearance Certificate is valid for 5 years from the date of approval. An extension may be applied for.

DECLARATION OF INVESTIGATORS

To be completed in duplicate and **ONE COPY** returned to the Research Office secretariat on the 3rd floor, Phillip Tobias Building, Parktown, University of the Witwatersrand, Johannesburg.

I/we fully understand the conditions under which I am/we are authorized to carry out the above-mentioned research and I/we undertake to ensure compliance with these conditions. Should any departure be contemplated from the research protocol as approved, I/we undertake to submit details to the Committee. **I agree to submit a yearly progress report**. When a funder requires annual re-certification, the application date will be one year after the date when the study was initially reviewed. In this case, the study was initially reviewed in **June** and therefore reports and re-certification will be due in the month of **June** each year. Unreported changes to the study may invalidate the clearance given by the HREC (Medical).

Signature of Principal Investigator

23/10/2023
Date