EVALUATION OF INTRAVENOUS PRELOADING MAGNESIUM SUPPLEMENTATION AS A PREVENTIVE MEASURE OF CISPLATIN INDUCED NEPHROTOXICITY

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DECLARATION

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DEDICATION

I dedicate this work to God, for gracing me to do a Randomized Controlled Trial. To him be the glory, for great things He has done, to allow me health and strength to finish this work in a foreign country very far from home.

I dedicate this work to my wonderful father Jean Marie for his love and support.

I would like to dedicate this work to Professors Guantai and Yapi. They influenced my life and thinking in a very positive way.

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LIST OF ABBREVIATIONS

AKI	**:**	**A**cute **K**idney **I**njury
BSA	**:**	**B**ody **S**urface **A**rea
BUN	**:**	**B**lood **U**rea **N**itrogen
CrCl	**:**	**C**reatinine **C**learance
CI	**:**	**C**onfidence **I**nterval
CIN	**:**	**C**isplatin **I**nduced **N**ephrotoxicity
CRF	**:**	**C**ase **R**eport **F**orm
CTCAE	**:**	**C**ommon **T**erminology **C**riteria for **A**dverse **E**vent
CTR1	**:**	**C**opper **T**ransporter 1
ER	**:**	**E**strogen **R**eceptor
GFR	**:**	**G**lomerular **F**iltration **R**ate
HeR2	**:**	**H**uman **e**pidermal grow factor **R**eceptor **2**
IRMA	**:**	**R**enal **I**nsufficiency and **A**nticancer **M**edications
KNH	**:**	**K**enyatta **N**ational **H**ospital
LAHNC	**:**	**L**ocally **A**dvance **H**ead and **N**eck **C**ancer
MAPK	**:**	**M**itogen **A**ctivated **P**rotein **K**inase
NCCN	**:**	**N**ational **C**omprehensive **C**ancer **N**etwork
NSAIDS	**:**	**N**on-**S**teroidal **A**nti **I**nflammatory **D**rugs
NSCLC	**:**	**N**on-**S**mall **C**ell **L**ung **C**ancer
OCT2	**:**	**O**rganic **C**ation **T**ransporter 2
PCR	**:**	**P**athologic **C**omplete **R**esponse
PR	**:**	**P**rogesterone **R**eceptor

RMS : **R**habdo**m**yo**s**arcoma

ROS : **R**eactive **O**xygen **S**pecies

SCr : **S**erum **Cr**eatinine

TCC : **T**exas **C**ancer **C**entre

TNBC : **T**riple **N**egative **B**reast **C**ancer

UPSC : **U**terine **P**apillary **S**erous **C**arcinoma

ABSTRACT

BACKGROUND: Nephrotoxicity remains a problem for patients who receive cisplatin based chemotherapy. Magnesium depletion is known as a complication to chemotherapy with cisplatin and likely to enhance nephrotoxicity. The National Comprehensive Cancer Networkrecommended 8mEq intravenous magnesium supplementation as a preventive measure of cisplatin-induced nephrotoxicity. This intervention is not yet applied in our setting due to lack of strong evidence.

OBJECTIVE: To evaluate the effect of intravenous magnesium preloading supplementation on cisplatin-induced nephrotoxicity in cancer patients on cisplatin combination chemotherapy at Kenyatta National Hospital and Texas Cancer Centre, Kenya.

STUDY METHODOLOGY: 71 patients diagnosed with cancer and who were to receive their first cycle of cisplatin-based chemotherapy at Kenyatta National Hospital or Texas Cancer Center at a single dose 60 mg/m^2 or above of cisplatin on day 1 were randomly assigned to receive intravenous magnesium preloading supplementation or not as part of their chemotherapy regimen. Serum creatinine was measured, and creatinine clearance (CrCl) was estimated by the Cockcroft–Gault equation. The follow-up period was 17 days. The primary outcome measure was incidence of acute kidney injury grade 1 or higher as defined by Common Terminology Criteria for Adverse Event 4.03. Kaplan-Meier survival analysis and Cox regression analysis were also performed to enable comparison of nephrotoxicity-free survival times and identify predictor factors for cisplatin-induced nephrotoxicity.

RESULTS: There was a significant decrease in the incidence of CIN in the Magnesium Preloading Group, compared to the Non-Magnesium Preloading group (12.12 % vs 33.13%, respectively; P = 0.037). Intravenous Magnesium sulfate supplementation also reduced the severity of CIN as it significantly reduced the mean maximum change in serum creatinine (0.10 mg/dL (range: -0.090, 1.761) versus 0.19 mg/dL (range: -0.147, 1.86); P = 0.006) and the mean maximum change in creatinine clearance (-13.2 ml/min (range: -56.3, 17.9) versus -22.05 ml/min (range: -112.8, 16.5); P= 0.041) in the magnesium supplementation group compared to the non-magnesium supplementation group, respectively. Survival analysis showed that magnesium supplementation also increased the nephrotoxicity-free survival time (P=0.042). Esophageal

cancer and BUN>5 mmol/l were identified as significant predictive factors for cisplatin-induced nephrotoxicity.

CONCLUSION: The study has provided strong and direct evidence in support of the application of intravenous preloading magnesium supplementation at the dose of 8 mEq before administration of cisplatin as a preventive measure of cisplatin-induced nephrotoxicity in cancer patients treated with cisplatin-based regimen.

CHAPTER ONE: INTRODUCTION

1.1. Background

Cancer is a major health-related problem for many worlwide. The overall cancer burden is high and it is still expanding. Each year more than 11 million people are diagnosed with cancer [1].

Moreover, more than 8 million people die from the disease per year worldwide [2]. Cancer has moved from the third leading cause of death in 1990 to the second leading cause, right behind cardiovascular disease in 2013[1-3].

The problem is, however, far more seriousin the developing world. Indeed, more than 70% of all cancer deaths occur in developing world as resources available for prevention, diagnosis and treatment of cancer are limited or nonexistent [1]. According to WHO, deaths from cancer in the developing world are likely to grow to 6.7 million in 2015 and 8.9 million in 2030 if no action is taken [2].

Based on incidence rate, solid tumors are the most common type of cancer, and include prostate, breast, cervix, ovarian, head and neck and bladder cancers [2].At present, cisplatin is one of the most widely used chemotherapeutic agents for the treatment of numerous solids tumors and it is proved to be beneficial [4-12]. However severe side effects such as nausea, vomiting, nephrotoxicity, ototoxicity, neurotoxicity and bone marrow suppression have been found to accompany its administration [13; 14].

Of these various side effects, the nephrotoxicity is of particularly graveconcern. It is dose limiting, and can be life threatening to the patient. The more severe presentation of cisplatin-induced nephrotoxicity (CIN) is Acute Kidney Injury (AKI) which occurs in 20–30% of patients [15; 16]. The onset is typically seen after 10 days of cisplatin administration and it is clinically manifested by increased serum level of blood urea nitrogen (BUN), creatinine (Cr) and reduced serum magnesium and potassium levels [17;18].

Although cisplatin is still used as a first-line medication for solid tumors, nephrotoxicity on one hand limits its use and on the other hand, by restricting the applicable doses, limits its efficacy in cancer therapy [19].

As an alternative, development of cisplatin analogues with less nephrotoxicity but equal efficacy has been attemptedand led recently to the introduction of carboplatin and oxaliplatin, second- and third generation platinum drugs, into clinical use [20]. In spite of this improvement cisplatin still provides better survival rate in some cancers such as lung cancer and remains an important component of various chemotherapy protocols due to time tested efficacy, widespread availability and affordability [21]. This enduring clinical importance of cisplatin highlights the need for effective preventive strategies for CIN.

A number of measures have been developed over recent years to reduce or prevent the occurrence of CIN. Most approaches to date have involved reducing the maximum circulating concentration of cisplatin by fractionation of the dose, slower the rate of infusion, enforced diuresis with diuretics and/or intravenous hydration [22;23].

While these approaches have reduced the occurrence of CIN, they have not completely prevented it. The prevalence is still recognized to be high.The worldwide prevalence of CIN is between 28-36% in patients who received a single dose (>50 mg/m^2) [24; 25] while it is more prevalent in developing countries. A recent study in Kenya reported a percentage of occurrence of 88.7%among patients receiving the first cycle of cisplatin based regimen [26].

Research during the last few years has gained significant insight on the pathogenesis of cisplatin induced nephrotoxicity. There is a growing body of evidence linking magnesium deficiency and cisplatin nephrotoxicity. They showed that Magnesium deficiency enhances cisplatin nephrotoxicity in addition to the direct cytotoxic damage of cisplatin to renal cells [27].Following these findings,researcherssubsequently investigated on the protective effect of magnesium supplementation during cisplatin treatment. Although the results have been variable, growing data has demonstrated that magnesium supplementation added to volume hydration during cisplatin treatment represent a combinatory strategy to significantly reduced frequency and severity of renal toxicity [28;29]. Based on these findings, intravenous magnesium supplementation has been recommended by The National Comprehensive Cancer Network (NCCN) in January 2011 as a novel approach of prevention of cisplatin nephrotoxicity [30].

The current prospective, interventional study sought to establish any potential benefits of magnesium preloading supplementation in ameliorating CIN.

CHAPTER TWO: LITERATURE REVIEW

Introduction

There have been many studies on cisplatin based chemotherapy. However, since the focus of this research was on prevention of cisplatin induced nephrotoxicity (CIN) with preloading magnesium supplementation, this literature review examined peer-reviewed journal articles and other selectedpublished resources relevant to five sets of key questions in turn, as follows: what is the importance of chemotherapy in cancer treatment? What is the activity and efficacy of platinum based chemotherapy? What are the toxic manifestations of cisplatin? What are the current preventive measures of CIN? And finally, what is the potential of magnesium supplementation in the prevention of CIN?

2.1. Chemotherapy in cancer management

Surgery and radiotherapy dominated the field of cancer therapy until researchers revealed that chemicals can be used to treat cancer. As Paul Ehrlich, a German physician scientist coined the term "chemotherapy" to describe the use of drugs to treat a disease, treatment of cancer using this modality was named cancer chemotherapy [31].

Since the 1940s, chemotherapy of cancer has evolved over the years to modern targeted therapy, having today a real impact in the management of cancer. Regarding the treatment of solid tumors, it appears that the use of combined modalities, including chemotherapy, at early stages of disease as adjuvant therapy is effective in preventing growth of metastatic or recurrent disease [32; 33]

In order to demonstrate the utility of chemotherapy in cancer management, several studies have explored the effect of chemotherapy on the disease free interval. A study by Heyn et al [34] investigated on the use of chemotherapy in children who have localized primary disease. The sample consisted of randomized children less than 21 years of age with a histologic diagnosis of Rhabdomyosarcoma (RMS). The study reported a significant difference in the relapse rate between the control and intervention group (chi-square= 7.61, $p = 0.002$) and concluded that the use of chemotherapy as part of modalities of treatment of children with RMS is relevant.

The importance of chemotherapy in RMS has also been stressed by Wilbur et al [35], who reported that 63% of children with RMS treated by combination chemotherapy had a disease-free interval of 1 or more years. However most of the studies which investigated on the clinical relevance of chemotherapy in association with surgery in RMS treatment reported that the age of the patient, stage and the primary site of solid tumors are important variables in the prognosis of RMS [36;37].

While the increase of disease-free interval was demonstrated by using chemotherapy in association with surgery in solid tumors treatment, the confidence that chemotherapy might have the capacity to cure patients with metastatic cancer while not being excessively toxic was also important to open up the field of adjuvant chemotherapy.

Einhorn and Donohue [38;39] conducted several studies examining the effectiveness of cisplatin, vinblastine, and bleomycin combination chemotherapy in the advanced stages of testicular cancer. In their first study they reported that this chemotherapy regimen produced an overall 85% disease-free status [38]. Their subsequent efforts were aimed at demonstrating through a series of studies a high cure rate of metastasis testicular by use of this regimen. The results from these studies showed an increase in a cure rate of metastasis testicular cancer from 10% to 60% [38 40], thereby demonstrating that chemotherapy can be used as an adjunct to surgery or radiotherapy and the patients can be rendered free of disease by drugs and achieve a normal life span.

Although studies have demonstrated the concept of curing cancer and prevention of growth of metastatic or recurrent disease by adjunctive chemotherapy [41;42], many of these studies have not taken into consideration the tumor staging prior to surgery and body burden of metastatic tumor at time of drug treatment. It is equally important to know if adjunctive chemotherapy is effective despite the stages of cancer.

Frank and Schabel [42] examined the rationale for adjuvant chemotherapy. They logically and objectively demonstrated two concepts concerning adjuvant chemotherapy. First, they demonstrated that grossly evident primary tumors are generally not curable by drug treatment, and effective surgical adjuvant chemotherapy is both dose-responsive and related to the body burden of metastatic tumor at time of drug treatment. Secondly they showed that the effectiveness of surgical adjuvant chemotherapy decreases as the tumor staging is advanced prior

to surgery, as the interval from surgery to start of effective chemotherapy is increased, and as the drug doses are reduced.

Alternatively, neoadjuvant chemotherapy as a variation of adjuvant chemotherapy is frequently used for some type of cancer. Neoadjuvant chemotherapy refers to treatment given before primary therapy to shrink a tumor that is inoperable in its current state, so that it may be surgically removed [43;44].Chen et al [45] reported that neoadjuvant chemotherapy increases the rate of conserving breast surgeries in breast cancer. However, compared to postoperative therapy, we cannot assert its superiority in the treatment of advanced breast cancer with regard to local recurrence, distant recurrence, and overall survival [46].

2.2. Platinum based cancer chemotherapy

Platinum-based chemotherapy over the years has improved the disease-free and overall survival of patients since 1971 where it was applied to a cancer patient for the first time. Numerous studies using platinum based chemotherapy are available in the literature.

Boulikas And Vougiouka [47] in a review of recent clinical trials using platinum drugs reported in 2014 that for most advanced cancers the response rate to chemotherapy is about 50% in first line treatments and about 15% in second or third line treatments Additionally they reported that most platinum based chemotherapy used either cisplatin or carboplatin, mostly in combination with other different cytotoxic drugs such as paclitaxel, gemcitabine, doxorubicin, venorelbine, irinotecan, 5-fluorouracil, docetaxel, cyclophosphamide, pemetrexed or tanstazumab in accordance with the type of tumors. Numerous clinical studies have compared different combinations of platinum based chemotherapy favoring either bi-therapy or tri-therapy combination of a platinum salt and others cytotoxic agents with regard to the efficacy, toxicity and quality of life [48; 49].

In the study conducted by Kelly et al [50], researchers examined the efficacy of cisplatin based chemotherapy in patients with uterine papillary serous carcinoma (UPSC)an aggressive form of endometrial cancer characterized by a high recurrence rate and a poor prognosis. They found that patients with cancer in the hysterectomy specimen in stage IA and stage IB treated with

Platinum-based chemotherapy had no recurrences in contrast to those who did not receive chemotherapy

Another broad success of cisplatin based chemotherapy was in the treatment of Triple Negative Breast cancer (TNBC). Indeed, despite the fact that it is particularly complicated to find the optimal chemotherapy regimen for this type of tumor. In a longitudinal survey evaluating the correlation between cisplatin based regimen chemotherapy and the outcome in TNBC in its advanced stages, Byrski et al [51] observed a clinical gain with cisplatin salt chemotherapy in the treatment of TNBC compared to others tumors. Neo-adjuvant complete response rates were significantly higher for TN tumors (88%) than others (51%; P = 0.005). The 5-year overall survival (OS) for TN tumors following adjuvant/neo-adjuvant chemotherapy was 64% [95% confidence interval (CI) 44% to 79%] compared with 85% (95% CI 79% to 90%) for others. They concluded that TNBC is more sensitive to cisplatin based regimen compared to others. However, the sample was non-randomized, and the patients with Estrogen receptor (ER) positive breast cancer included in the study were not tested for progesterone receptor (PR) and human epidermal growth factor receptor (HeR2). These inconsistencies may constitute limitations for the study.

Further to support the hypothesis of the efficacy of cisplatin based chemotherapy, in 2005 D'Addario et al [52] in a meta-analysis of 37randomized trials, demonstrated that response is significantly higher with platinum-containing regimens compared to non–platinum-based chemotherapy in advanced non–small-cell lung cancer. This meta-analysis was one of the largest studies to evaluate the activity, efficacy and toxicity of platinum based versus non–platinum-based chemotherapy in patients with advanced non–small-cell lung cancer. The study included 7,633 patients. A 62% increase in the odds ratio (OR) for response was attributable to platinum-based therapy (OR, 1.62; 95% CI, 1.46 to 1.8; P <0.0001). The 1-year survival rate was increased by 5% with platinum-based regimens (34% v 29%; OR, 1.21; 95% CI, 1.09 to 1.35; P =0.0003). However, no significant difference was found in 1-year survival rate when platinum therapies were compared to third generation– based combination regimens (paclitaxel, docetaxel, gemcitabine, vinorelbine, and irinotecan) (OR, 1.11; 95% CI, 0.96 to 1.28; P = 0.17).

Moreover, an investigation on the efficacy of platinum based chemotherapy in patients with brain metastasis from non-small cell lung cancer (NSCLC) by Kim et al [53] found that the

median survival in patient receiving platinum based chemotherapy was longer than that of those who do not (58.1 vs. 19.0 weeks, $p<0.001$). One limitation of this study is that it was not randomized, thus selection bias may affect the external validity of the study.

In addition, Vermorken et al [54] evaluated the effectiveness of Cetuximab plus platinum based chemotherapy in head and neck cancer. In a randomized control study of 442 patients with untreated recurrent or metastatic squamous-cell carcinoma of the head and neck. They evaluated the efficacy of cetuximab plus platinum-based chemotherapy as first-line treatment and reported that the addition of cetuximab on platinum based chemotherapy prolonged the median progression-free survival time from 3.3 to 5.6 months (hazard ratio for progression, 0.54; $P<0.001$) and increased the response rate from 20% to 36% ($P<0.001$).

All of these studies on the activity and efficacy of platinum based regimens confirm that cisplatin is among the most effective broad-spectrum anti-tumors drugs, and support the conclusion that platinum based cancer chemotherapy significantly increases pathologic complete response (pCR) rates and survival. However, the study by D'Addario et al [52] contains one limitation. The authors consider cisplatin and carboplatin as equivalent agents while the therapeutic equivalence for NSCLC are still contradictory [55-56]. This methodology flaw may affect the interpretation of the result. Future research should replicate these findings with the use of the same cytotoxic agent.

2.3. Cisplatin toxicity

Cisplatin remains the leading chemotherapy agent for the treatment of solid tumors. However severe side effects that significantly restrict its clinical use and effectiveness have been reported in the research literature.

Several things are thought to be correlated with adverse effect of cisplatin administration such as dosage, the sites of solid tumors and interaction with other drugs. According to Laura et al [57], despite the fact that significant correlations were found with others variables, the main factor influencing the severity of the adverse effect was the dosage of cisplatin administered.

Laura and collaborators carried out a retrospective study and included 123 patients undergoing cisplatin based chemotherapy. The adverse effects recorded were by order of the most important: gastrointestinal disorder 72 % followed by hematological toxicity 54%, neurotoxicity 26 % nephrotoxicity 17 %, hepatic toxicity 11 % and ototoxicity 9%. In order to verify the correlation between the chemotherapy dosage and the incidence of adverse effects, they did a Spearman non-parametric correlation between daily or cumulative cisplatin dosage and adverse effects. They found that the cumulative amount of cisplatin was directly related to the number of adverse effects ($r2=0.3826$, $P<0.001$). Their results underline the cumulative dose toxicity of cisplatin chemotherapy.

While the dosage of cisplatin is correlated with his toxicity, its effectiveness is also known to be dose dependent. Due to this defect, platinum analogues e.g. carboplatin and oxaliplatin were synthesized to increase efficacy and reduce toxicity. Investigation on the efficacy and toxicity of platinum derivatives compared to cisplatin appeared fundamental to evaluate the alternative.

Lokich and Anderson [58]. conducted a systematic review of randomized clinical trials comparing carboplatin with cisplatin, both as single agents and in combination with other agents They identified five solid tumors within which comparative trials had been conducted: ovarian (10 trials), lung (2 trials), head and neck (2 trials) germ cell tumors (3 trials) and bladder cancer (1 trial). Effectiveness and toxicity were compared and cisplatin was found to be superior or equivalent to carboplatin in therapeutic efficacy in all five tumors- the superiority of cisplatin compared to carboplatin in terms of effectiveness was observed in germ cell tumors, bladder cancer, head and neck cancer while for others it was comparable. However, cisplatin was associated with an increased toxicity profile for gastrointestinal, renal and neurologic effects

A similar review by Go and Adjei [59] on pharmacology and clinical activity of cisplatin and carboplatin reported comparable findings. They examined 32 randomized control trials comparing cisplatin and carboplatin in five different tumors. They reported that carboplatin and cisplatin have equivalent efficacy only in sub-optimally debulked ovarian cancer and extensive-stage small-cell lung cancer. Concerning the toxicity, they found that while nephrotoxicity was known to be dose-limiting adverse effect for cisplatin in early clinical trials, myelosuppression, particularly thrombocytopenia, was the dose-limiting toxicity of carboplatin [60-61].

Furthermore, Sibon et al [62] investigated the toxicity of oxaliplatin-based regimen in first-relapsed or refractory Hodgkin lymphoma and showed that the treatment is relatively safe, with only a mild sensitive peripheral neuropathy not exceeding grade 2 that was subsequently reversible.

Although these results combined establish that cisplatin analogues represent an alternative by reducing toxicity, they also confirm that cisplatin remains the broadest spectrum platinum drugs in the treatment of solid tumors.

Research on the mechanism and risk factors of CIN observed that cisplatin induced nephrotoxicity has multiple pathways. Yao et al [63] reported in 2007a detailed study on the mechanism of nephrotoxicity induce by cisplatin. They reviewed clinical and experimental literature relevant to CIN and found that unbound platinum is mainly responsible of the injury. It is filtered at the glomerulus and taken up into tubular cells where it is partially metabolized into toxics species which in turn, through different intracellular effects, cause tubular damage and tubular dysfunction characterized by sodium, potassium, and magnesium wasting.

To gain further understanding on the mechanism of nephrotoxicity, especially on the signaling pathways leading to tubular cell death and inflammation, Taguchi et al [64] examined recent research and reported that exposure of tubular cells to cisplatin activates signaling pathways that are cell death promoting (Mitogen-Activated Protein Kinases [MAPK], p53, Reactive Oxygen Species [ROS]) or cytoprotective (p21). In the meantime, cisplatin induces TNF-α production in tubular cells, which triggers a robust inflammatory response, further contributing to tubular cell injury and death.

In recent years more studies have begun to look at the risk factors of cisplatin induced nephrotoxicity. For example, Kidera et al [65] in 2014found that the regular use of non-steroidal anti-inflammatory drugs (NSAIDs) were significantly associated with an increased risk for cisplatin nephrotoxicity (risk ratio, 1.357; $P = 0.047$). Additional observation showed that development of hypomagnesaemia during cisplatin treatment was significantly associated with a greater increase in serum creatinine level ($P = 0.0025$). This is consistent with observations by Yokoo et al [66] after injection of cisplatin to hypomagnesemic rats during a study. They reported an increase renal accumulation of cisplatin and the deterioration of acute kidney injury.

Moreover, de Jong et al [67] evaluated risk factors associated with CIN in a large cohort of 400 patients undergoing cisplatin based chemotherapy. They used logistic regression analysis to assess baseline parameters for independent prognostic factors. They found that older age, female gender, smoking, hypoalbuminaemia and paclitaxel administration were risk factors for nephrotoxicity. In addition, they discovered that there was a gradual increase in renal toxicity with increasing age at an OR of 1.03 year−1 (P=0.007) and Paclitaxel co-administration was strongly related to the development of nephrotoxicity (OR 4.0, CI1.8–8.8). Another finding from this study in accord with previous results [68-70]was that cisplatin-induced nephrotoxicity is related to the peak plasma concentration and/or the area under the plasma concentration–time curve of ultrafiltrable cisplatin.

Besides that, aminoglycosides co administration was incriminated as a significant risk factor of cisplatin induced nephrotoxicity by Haas et al [71]. The reported incidence of CIN was higher in patients receiving cisplatin in combination with aminoglycosides than in patients receiving cisplatin alone. On the other hand, looking at the factors such as cardiovascular disease and reduced baseline creatinine clearance, these studies agree that these factors were not associated with CIN.

Most recently, Prasaja et al [72] pushed further the investigation on CIN in order to understand its associated factors. They retrospectively reviewed the medical recordsof 88 adult cancer patients treated with cisplatin ≥60mg/m2. They revealed that age (OR=3.433, 95%CI= 1.363-8.645,p=0.008) and hypertension (OR=2.931, 95%CI=1.120-7.670,p=0.026) were both associated with development of CIN. Their result was not consistent with the observation of De Jong et al [73] regarding cardiovascular disease as a risk factor. Both studies were limited by a retrospective design, but the sample size in this study by Prasaja et al. was small as compared to that of the study by De Jong et al (400). The sample size was therefore less likely to be representative of the population.

There are various presentations of CIN but most of the available evidence describes the clinical features of cisplatin-induced Acute Kidney Injury (AKI) which is more common presentation of CIN [15-16]. According to Moon et al [74] cisplatin-induced AKI occurred more frequently during the 3rd- 4th cycle with a most common cumulative dose of 200-300 mg of cisplatin/BSA. Arany and Safirstein [24] and Gonzales-Vitale and Hayes [17] reported that it is most likely to

appear 10 days after cisplatin administration and is revealed by increase in the serum creatinine and blood urea nitrogen concentrations.

In summary, the literature surrounding cisplatin toxicity suggests that cisplatin toxicity is dose related and cumulative. Despite the introduction of platinum analogues, cisplatin remains the agent showing the broadest spectrum of antineoplastic activity. Factor such as NSAID, hypomagnesemia, age, hypoalbuminaemia and paclitaxel co-administration are strongly associated with development of cisplatin induced nephrotoxicity. Clinically it appears that the time of onset of cisplatin induced nephrotoxicity is unclear, underlining the need to have clearly defined diagnostic criteria for cisplatin injury before undertaking any study.

2.4. Preventive measures of cisplatin-induced nephropathy

During the course of exploring the mechanism of CIN, several strategies have been reported to provide renoprotection. These include pharmacologic, molecular or genetic approaches.

Pabla and Dong [14] identified in 2008 key primary targets for these approaches. The primary targets included cisplatin uptake by renal cell, cisplatin metabolism and bio-activation, cell death pathways, cell-cycle regulators, p53, MAPKs, oxidative stress and inflammation. Among those preventive measures, volume expansion plus saline diuresis was the first means accepted as standard prevention measure [23]. It is worth considering how since its first clinical application by Schilsky et al in 1976 [75], researchers have been seeking to find the best hydration protocol. Based on experimental studies results, it was postulated that the administration of diuretics and hypertonic saline might result in additional protection against CIN.

Pera et al [76] undertook in 1979 a study on the effects of mannitol or furosemide diuresis on the nephrotoxicity induced by cisplatin. They suggested that as tubular necrosis might be related to cumulative platinum uptake in the kidney, diuretics may have a protective effect by reduction of platinum concentration in the urine. Subsequently, two others studies supported the usefulness of diuretics in cisplatin nephrotoxicity prevention. First, a laboratory study by Heidemann et al [77] in 1985 concluded that both furosemide and acetazolamide attenuate the nephrotoxic response of cisplatin treatment. Secondly, a clinical trial in 1982 on cisplatin hydration with and without mannitol diuresis by Al-Sarraf et al [84]80confirmed the beneficial effect of mannitol. Both studies revealed that renal toxicity is less severe in patients treated with mannitol.

These positives results stand in contrast to the results reported by Santos et al and Yang et al. [79, 80]. In a randomized control trial, Santos and co-workers found in 2003 a significant difference in cisplatin nephrotoxicity between the saline + mannitol group and the saline group (P=0.02) or the saline + furosemide group (P=0.02). They concluded that hydration with saline or saline + furosemide appears to be associated with less cisplatin nephrotoxicity than saline + mannitol. This was consistent with the finding of Yang et al. [80]in 2014, who demonstrated a non-beneficial effect of mannitol administration on acute kidney injury (AKI) prevention. They carried out a meta-analysis of nine trials involving626 patients and compared the reduction in serum creatinine level with expansion of intravascular volume alone versus expansion plus mannitol. No significant difference was observed (Mean Difference: 1.63, 95% CI -6.02 to 9.28). Considering the results of these two studies, there is no reason to advocate for the use of diuretics in prevention of cisplatin induced nephrotoxicity.

While the protective effect of diuretics against CIN is still controversial, cisplatin along with vigorous intravenous hydration is recognized to be one of the most effective preventive strategies [85]. However, there is no consensus on the amount and duration of hydration. Whereas conventional long hydration has been recommended on labels for cisplatin, both long and short hydration are currently observed in clinical practice.

In 2011 a survey by Japan by Yamada et al. [82] to investigate the hydration methods used with cisplatin-containing regimens at various institutions in Japan revealed that hydration with 3000 ml intravenous saline was performed on day 1 for all institutions. 65% of the institutions performed hydration for up to 3 days whereas no more than 14 % of the institution did so only on day 1.

Additionally, a retrospective study conducted in 2014in Kenya by Mwai et al [26] revealed that the Kenyatta National Hospital, a referral institution in Kenya, performed hydration for just 2 days. They found that all patients were given pre- and post-treatment hydration using normal saline; 50.29 % were given a total of 1000 ml while 49.10 % received 2000ml. However, the investigators concluded that the doses of normal saline used was not statistically significantly associated with prevention of development of nephrotoxicity (p=0.487).

Researchers have investigated the efficacy and safety of this short hydration regimen as its use with outpatient chemotherapy containing cisplatin has been widespread in recent years. Horinouchi et al [83] undertook in 2013 a prospective study which included 44 patients. Cisplatin based chemotherapy was administered with pre- and post-treatment hydration with normal saline containing 10 mEq of potassium chloride in 500 ml of fluid over a 60-min period. Just before the administration of cisplatin, mannitol (20%, 200 ml) was administered as forced diuresis over 30 min. Magnesium sulfate (8 mEq) was added to pre-hydration. They found that 43 (97.8%) completed the cisplatin-based chemotherapy without Grade 2 or higher renal dysfunction. They concluded that short hydration with potassium and magnesium supplementation is safe without severe renal toxicities in regimens containing cisplatin (75 mg/m2) for patients with lung cancer.

To further support that hypothesis, a study with a larger sample sizes by Tiseo et al [84] in 2007 examined data of 107 outpatients previously enrolled in randomized studies. They aimed to assess the incidence of CIN in patient treated with cisplatin using a short hydration regimen, which included 2000 ml of fluids with control of diuresis. They found that out of 107 patients, 102patients had stable serum creatinine and creatinine clearance level around the normal values during treatment.

Whilst these findings have provided evidence for a more practical method of hydration, it is important to note that Horinouchi et al [83] was a non-randomized study and Tiseo et al [84] was retrospective. Therefore, these limitations may increase the uncertainty of their results. They remain insufficient to inform practice.

A useful comparison between long hydration and short hydration regimen was carried out in 2014 by Ouchi et al [85] in Japan. They found that short hydration regimen in outpatient chemotherapy containing intermediate- to high-dose cisplatin is as safe as the continuous hydration regimen and increased the efficacy of chemotherapy. Although this study confirms the efficacy of short hydration, it also has some limitations. It was a retrospective analysis of a small number of patients having different cisplatin based regimens. Therefore, other anticancer drugs may affect renal function and selection bias is also probable.

Considering these results, it appears that the protective effect of short hydration regimen against cisplatin induced nephrotoxicity is still questionable. A randomized control study with large patient numbers and a uniform cisplatin based regimen is needed to significantly bring to light the efficacy of short hydration regimen.

In addition to the above-mentioned approaches, researcher reported the efficacy of several pharmacologic agents. Ibrahim et al [86] in 2010 found Zerumbone a natural compound isolated from the fresh rhizomes of *Zingiber zerumbetto* to be strongly associated with reduced kidney damage in rat after cisplatin administration. A similar experimental study in 2014 by Abdel et al [87]suggested that *Azadirachta indica* attenuates cisplatin induce nephrotoxicity and oxidative stress. Unfortunately, these studies have only been done in rats, hence we cannot know whether these pharmacologic agents are effective in human.

In recent years, in order to improve the therapeutic index of cisplatin, researchers have investigated alternate methods of cisplatin administration. Indeed, Driessen et al [88] in 2014 examined the degree of nephrotoxicity after intermediate or high-dose cisplatin-based chemoradiotherapy in patients with locally advanced head and neck cancer (LAHNC). They compared different schedules - the standard treatment for LAHNC cisplatin 100 mg/m^2 on days 1, 22, 43 (cis 100) versus an alternative cisplatin schedule 40 mg/m^2 weekly during six weeks (cis 40). They observed that during treatment with cis 40, 17.3% developed an increase of ≥25% serum creatinine versus 77.5% treated with cis 100 ($p<0.05$). Reports on the degree of nephrotoxicity from this study revealed that according to Common Terminology Criteria for Adverse Events (CTCAE) version 4.03, while nephrotoxicity Grade 1 occurred more in cis 40 compared to cis 100 (68% vs 40%); grade 2, 3 and grade 4 were of that in cis 100 (respectively: 7%, vs 53%; 0% vs 5%; 0% vs2%). This result indicates that the fall in glomerular filtration rate is dependent of the amount given as single dose. This finding was in accord with a previous study [89]

Together the above mentioned studies on alternate methods of cisplatin administration demonstrate that fractionation of the dose may be a potential approach to attenuate cisplatin induced nephrotoxicity.

Clearly, numerous approaches to prevent cisplatin induced nephrotoxicity have been reported. While it emerges that hydration is the most effective and widely applied measure, there is not yet convincing evidence that the others means are successful or effective in clinical use.

Hydration which is known as the most effective preventive measure is widely recognized to be partially successful. According to Launay-Vacher et al [90] in the Renal Insufficiency and Anticancer Medications (IRMA) study carried out in 2010, the prevalence of renal insufficiency in cancer patients was high. Among 4684 solid tumor patients from different cancer centers, 50–60% had a stage 2 kidney disease. Furthermore, results of the lung cancer subgroup analysis revealed that among the population (445) of lung cancer patients, 62.1% had abnormal renal function - 85.8% were receiving anticancer drugs during the time period studied and cisplatin represented 23.63% of the prescribed anticancer drugs that were potentially toxic to the kidneys.

Additionally, the Kidera et al [65] study quoted previously looked at the incidence of cisplatin induce nephrotoxicity in 401 chemotherapy naïve patients who underwent chemotherapy including a high dose (>60 mg/m2) of cisplatin. Patients were hydrated pre- and post-treatment with isotonic saline containing 5% glucose, mannitol and furosemide. Nephrotoxicity was defined as an increase in the serum creatinine concentration of at least grade 2 during the first course of cisplatin chemotherapy. They found that the incidence of Cisplatin-induced nephrotoxicity was 32% (127 patients).

These results are consistent with Mwai et al [26] findings from a retrospective study carried out in KNH in 2014. They reported an incidence of at least grade 2 nephrotoxicity during the first course of cisplatin chemotherapy of 47 %. Together these studies revealed that despite the use of hydration and diuretics, prevention of cisplatin induced nephrotoxicity is still partial and the incidence is still high.

Although the study by Kidera et al [65] provided valuable information regarding the efficacy of hydration, the results are limited because of possible selection bias of treatment due to retrospective analysis.

In regard to the problem of partially successful approaches when preventive measures are used individually to prevent CIN, researchers recently investigated on the use of several agents together to achieve a clinically meaningful outcome. One of the current investigations is on the

use of both hydration and electrolyte treatment, specifically magnesium repletion as it is known that cisplatin induces magnesium depletion affecting up to 90% of patients if no corrective measures are initiated and magnesium deficiency itself may enhance cisplatin nephrotoxicity [27] [91].

2.5. Potential of Magnesium supplementation for prevention of CIN

Magnesium supplementation has been found to prevent cisplatin induced hypomagnesaemia in patient undergoing cisplatin based chemotherapy. Miguel et al [92] conducted in 1992 a clinical trial to evaluate the efficacy of intravenous and oral magnesium supplementation in the prevention of cisplatin induces hypomagnesaemia. The study recruited 41 patients treated with 100 mg/m^2 to participate in the study and randomly allocate them into 3 groups to receive different treatment: no magnesium, IV magnesium supplementation and oral magnesium supplementation. Results indicated that the patients on both the oral magnesium and IV magnesium supplementation arms presented significantly higher serum magnesium level than the control group from the second and third course of chemotherapy. Additionally, results showed that after the fourth course of chemotherapy 33% and 44 % of patient in oral and IV magnesium group respectively developed hypomagnesaemia compared to 99% in the control group (unsupplemented patients). No major side effect was reported. These findings indicate that both IV and oral magnesium supplementation are safe and efficacious to prevent cisplatin induced hypomagnesaemia.

Further investigation on IV magnesium supplementation by Anvari et al [93] in 2010 confirms this hypothesis. They conducted a prospective randomized study, which included 59newly diagnosed adult patients receiving cisplatin-based chemotherapy. Patients were randomly allocated to receive magnesium supplementation at a dose of 5 g IV per cycle (n=31) or to a control group (n=28). Serum magnesium levels <1.8 mg/dl were considered to indicate hypomagnesaemia. They reported that hypomagnesaemia was more frequent in the control group (38.7% vs. 60.7%, P=0.09). Therefore, the concluded that Magnesium supplementation at a dose of 5 g per cycle partially compensated for cisplatin- induced magnesium loss.

Although these studies showed that magnesium supplementation can prevent cisplatin induced hypomagnesaemia, it is also important to known if prevention of magnesium deficiency may be sufficient to overcome cisplatin-induced renal toxicity. In a study conducted by Ashrafi et al [94] in 2012, investigators examined the role of magnesium supplementation in CIN in a rat model. The study explored the nephroprotectant role of magnesium against cisplatin. Researchers randomly assigned Wistar rats to four experimental groups and administered magnesium sulfate at different dosages of 20, 80, 200mg/kg for group 1, 2, 3 respectively; group 4 received normal saline lacking magnesium supplementation. All four experimental groups received the same cisplatin regimen. They measured the levels of BUN and Cr in one hand and tissue damage scores in the others hand. They reported that the intensity of kidney toxicity in group 1 (low dose of Mg) was higher than those in other groups. Additionally, the study revealed that moderate and high doses of Mg supplementation did not provide a significantly better result when compared with the control group (group 4). The importance of these findings is that they suggest that magnesium supplementation alone does not have a nephroprotective effect against cisplatin-induced nephrotoxicity in rats, and that it may actually promote kidney toxicity under some conditions.

In further support of this finding, Oka et al [95] hypothesized that Mg infusion combined with low volume hydration may not be sufficient to overcome cisplatin-induced renal toxicity. They conducted in 2014 a historical prospective cohort study. 85 patients undergoing first cycle of cisplatin-based chemotherapy at the Osaka City University Hospital were included and classified into three groups: high volume hydration without Mg infusion (high-volume Mg-), high volume hydration with Mg infusion (high-volume Mg+), and with low volume hydration with Mg infusion (low-volume Mg+).

Researchers examined serum creatinine and creatinine clearance before and after cisplatin administration. It was found in the high-volume without magnesium group, a significant decrease of serum creatinine (Scr) and creatinine clearance (Crcl) post treatment compared to pretreatment ($p < 0.001$ and $p < 0.001$, respectively) while in the high-volume Mg+ group, there was no significant difference between pre- and post-treatment levels of Scr and Crcl ($p = 0.118$ and $p = 0.254$, respectively). The low volume magnesium group displayed a decrease of Scr and CrCl after treatment ($p = 0.068$ and $p = 0.055$, respectively). Additional result revealed that absence of

Mg infusion and low-volume hydration were both independent factors for decreased CrCl ($p < 0.001$ and $p = 0.001$, respectively). This study indicates the importance of combining both high volume hydration and magnesium supplementation to have a meaningful outcome. It also confirms the efficacy and safety of magnesium supplementation 8 mEq as recommended by the National Comprehensive Cancer Network (NCCN) [30].

Similarly, Muraki et al [96] examined in 2012 the effect of hydration with magnesium and mannitol without furosemide on CIN. In this study the outcomes of two different hydration regimens were retrospectively compared. An old hydration protocol included normal saline with mannitol and furosemide and a new one included normal saline with magnesium and mannitol without furosemide. They reported a significantly greater increase in creatinine clearance (P=0.0004) and a decrease in the serum creatinine level (P=0.0148) after the first course for new hydration protocol compared to old regimen. A multivariate analysis additionally revealed that the new hydration protocol was an independent factor for the protection against nephrotoxicity [HR 0.232 (95% CI: 0.055-0.986), P=0.039].

Although valuable information was gained from Oka et al [95], there were identified limitations. Because the cohort was constructed in past time and exposures documented in past, there was notable absence of data on potential confounding factors in the study, which may affect the internal validity of the study. In addition, in both Oka et al and Mukari et al studies mentioned above, investigators administered isotonic saline plus 8 mEq of magnesium sulfate before the administration of cisplatin in order to evaluate the protective effect of magnesium supplementation, Therefore the result of those studies also confirm the protective effect of magnesium preloading on CIN.

A similar chemotherapy hydration regimen was evaluated in 2014 by Yoshida et al [97]. They retrospectively reviewed 496 thoracic malignancy patients treated with cisplatin (<60 mg/m^2)-containing regimens as a first-time chemotherapy. They compared the incidence of Grade 2 serum creatinine elevation between magnesium preloading group and non-magnesium preloading group during the first cycle and all cycles. The observed that the incidence of Grade 2 serum creatinine elevation in magnesium preloading group was significantly lower during both the first cycle and all cycles compared to non-magnesium preloading group (4.9 versus 19.1%

during the first cycle, and 14.2 versus 39.7% during all the cycles). The findings of this study reinforce the hypothesis on the protective effect of magnesium preloading against cisplatin induced nephrotoxicity. However, the study design is not adequate to inform practice because of the retrospective analysis.

In contrast to the available studies on the literature on magnesium supplementation before the administration of cisplatin which are retrospective, several prospective clinical trials have been done on magnesium supplementation before and after the administration of cisplatin. Bodnar et al [28] conducted in 2008 a double-blind, placebo-controlled, randomized study to examine the effect of magnesium supplementation on nephrotoxicity accompanying standard cisplatin-based chemotherapy in patients with epithelial ovarian cancer. They recruited 41 patients. Researchers administered magnesium supplementation before and after each course of chemotherapy with paclitaxel (135 mg/m2/24 h) plus cisplatin (75 mg/m2) every 3 weeks. Magnesium sulphate (5 g) was administered before and Magnesium sub-carbonate (500 mg) was administered three times per day orally during the treatment intervals. The control arm received a placebo instead of both magnesium salts. The observed that the control group showed a significantly greater decrease of GFR assessed by: serum creatinine ($p = 0.0069$), Clearance Cockcroft Gault equation ($p = 0.0077$) and Clearance Modification of Diet in Renal Disease equation ($p = 0.032$) formulae compared with the magnesium supplemented group. The study was limited by a small sample size.

In regard to the literature surrounding the role of magnesium supplementation on cisplatin induced nephrotoxicity, it appears that while strong evidence on the protective effect are available for high volume hydration plus magnesium supplementation before and after the administration of cisplatin, there is no similarly strong evidence yet available for magnesium preloading supplementation. Furthermore, the effect of magnesium supplementation on the pharmacokinetic of cisplatin has not yet been investigated. That may constitute a limitation to accept hydration plus magnesium preloading supplementation as a standard preventive measure for cisplatin induce nephrotoxicity while some studies have shown compelling results on the protective effect of magnesium preloading supplementation.

Conclusion

The above literature review indicates that several investigations have been done on cisplatin. Their results reinforce the need for more effective strategy for prevention of cisplatin nephrotoxicity. The review also identifies gaps and issues that have not so far been investigated. In order to gain strong evidence on the protective effect on magnesium preloading supplementation, it was necessary to conduct a prospective study that examines the causal relationship between magnesium supplementation and reduction of CIN. This is what the current study sought to examine by evaluating the association between the incidence of Grade 1 or more SCr elevation during first-time chemotherapy and the effect of Mg preloading.

Problem statement

Cisplatin has clinical benefit for several types of solid tumors such as lung, testicular, head and neck, ovarian, cervical and breast cancers. However, its clinical utility is limited by nephrotoxicity, the chief dose-limiting side effect.

Magnesium supplementation before the administration of cisplatin has been applied as a new hydration protocol in order to reduce the frequency and incidence of cisplatin induced nephrotoxicity in patient undergoing cisplatin based chemotherapy. In January 2011 The National Comprehensive Cancer Network (NCCN) also recommended the same intervention [30].

Since its application, several studies have reported on its efficacy and safety. For example, a recent study showed that volume hydration plus magnesium supplementation significantly reduced the incidence of Acute Kidney Injury [95]. A separate study also reported that magnesium preloading significantly lowered the incidence of severe nephrotoxicity [97].

However, there is strong methodological concern regarding the studies that currently provide the evidence that magnesium preloading has a preventive effect on cisplatin induced nephrotoxicity.Indeed, the previous studies on nephro-protective effect of magnesium preloading supplementation have been limited by small numbers of patients and retrospective study design.Consequently, there is need for more rigorous research [93;97].

To our knowledge, no randomized controlled trial (RCT) in our setting has been published to establish a causal relationship between Mg-preloading supplementation during cisplatin based chemotherapy and reduction of nephrotoxicity. Furthermore, there are no available studies on the nephron-protective effects of Mg-preloading supplementation in the literature with sufficient sample size or sample diversity to allow generalization of the findings.

All these factors limit the acceptance and application of Mg-preloading supplementation in our clinical setting while the need for more preventive measures is of such great importance. The current use of volume hydration and diuresis without magnesium supplementation at Kenyatta National Hospital (KNH) as a preventive measure against CIN has shown partial result. A retrospective study carried out in 2014 in KNH estimated a prevalence of CIN of 59% [26].

The potential benefits of magnesium preloading supplementation may not be realized unless strong, locally derived evidence of the potential protective effect of this intervention is provided, if any.

Justification

The current study sought to establish any potential benefits of magnesium preloading supplementation in ameliorating CIN. Currently, there are no reports in the literature of prospective interventional studies on the effects of Mg-preloading supplementation on CIN with a sufficient sample size. We sought to addressthese concerns by employing a randomized controlled trial and having a sample large enough to provide adequate statistical power.

Magnesium supplementation could potentially significantly reduce the incidence and degree of CIN. By diminishing this major dose-limiting toxicity of cisplatin and reducing the need for dose reduction, this intervention could serve to maintain the therapeutic index,enhance the dose-dependent antitumor efficacy of cisplatin and improve treatment outcomes. If magnesium supplementation does provide beneficial effect on the prevention of cisplatin induce nephrotoxicity, this study will be able to establish its efficacy and therefore offer direct, locally derived evidence for its introduction into practice.

In addition, through the publication of the findings, this study will add to the literature related to the protective effect of Mg-preloading supplementation against CIN.

Hypothesis:

We hypothesized, that the occurrence and the severity of nephrotoxicity among patients receiving the first course of standard cisplatin based chemotherapy will be significantly different amongpatients who also receive IV magnesium preloading supplementation compared to patients who do not receive IV magnesium preloading supplementation.

Objectives

General objective

The present study aimed to investigate the effect of IV magnesium preloading supplementation on cisplatin-induced nephrotoxicity in cancer patients receiving the first course of standard cisplatin based chemotherapy at Kenyatta National Hospital (KNH) and Texas Cancer Centre (TCC), Kenya.

Specific objectives

There werefour specifics objectives:

1. To determine the incidence of nephrotoxicity among patients undergoing the first course of cisplatin-based chemotherapy and receiving the current standard renoprotective intervention of volume hydration without magnesium supplementation.

2. To compare the incidence of nephrotoxicity among patients receiving the current standard renoprotective intervention of volume hydration without magnesium supplementation (control group) to those receiving magnesium preloading supplementation plus standard renoprotective intervention (intervention group), all of whom are undergoing the first course of cisplatin-based chemotherapy.

3. To compare the severity of nephrotoxicity between the intervention and control groups by comparing the average change in creatinine clearance and the difference in decrement between the two group of calculated creatinine clearanceat each time pointusing the trend curves.
4. To explore the influence of various baseline characteristics on the severity and degree of CIN in both comparison groups.

Significance of the study

A retrospective study on CIN carried out at KNH reported a prevalence 59% [26]. It revealed that preventive measures applied in the hospital have a limited result. In KNH, prevention still relies on decreases in drug dosage, hydration measures, and active screening for renal abnormalities.

Should the current study reveal the beneficial effects of IV Magnesium preloading supplementation, then this would provide direct evidence for the incorporation of this intervention into existing preventing measures. This in turn could reduce significantly the occurrence and severity of nephrotoxicity in patients receiving cisplatin-based chemotherapy, therefore improving chemotherapeutic efficacy of cisplatin in clinical use. Furthermore, the introduction of IV magnesium supplementation may encourage the use of cisplatin in preference to the other platinum compounds (such as the less toxic but more expensive Carboplatin) as part of the first line chemotherapy regimen for a range of solid tumors for which cisplatin remains the drug of choice. Subsequently this action may have an impact on further attenuation of the overall cost of chemotherapy.

CHAPTER THREE: METHODOLOGY

This chapter covers an overview of methodology used to conduct this study. The first part of the chapter contains a description of the research design, population sampling, and the randomization procedure. This is followed by an explanation of the experimental procedure and a detailed discussion of data collection, data management and data analysis methods. The chapter concludes with a discussion of the ethical considerations and measures to provide validity.

3.1 Study design

This study was a two armed, prospective, randomized, controlled, dual-center, double-blind, superiority trial to evaluate the effect of intravenous magnesium supplementation in reducing the incidence and severity of cisplatin induced nephrotoxicity among chemotherapy-naive cancer patients following the first course of standard cisplatin based chemotherapy.The experimental group in this study was cancer patients on standard cisplatin based chemotherapy who received magnesium preloading supplementation and the control group was the group of cancer patients on standard cisplatin based chemotherapy who do not receive magnesium preloading supplementation. This study was an investigator-initiated clinical trial.

3.2 Study setting

This study was performed in two centres in Kenya, Kenyatta National Hospital and Texas Cancer Centre between June 2015 and October 2015.

Kenyatta National Hospital (KNH) is 1800 bed capacity referral university hospital located in Nairobi serving a population of 4 million. It is the only public health facility in Kenya where patients can obtain advanced comprehensive treatment for cancer. It therefore has high demand for services. Records indicate that there are approximately 30 new cancer patients every week, while 50 inpatients and 100 outpatients are admitted and treated at the oncology wards/clinics weekly.

Texas Cancer Centre (TCC) is a leading private cancer care and treatment centre that has two branches located in Nairobi - one branch on Mbagathi Way for outpatients and the other in Hurlingham for inpatients. TCC provides screening, chemotherapy and radiotherapy for several

cancers. With a 25 -bed capacity and 30 chairs for delivery of chemotherapy regimens, the centre sees between 20 and 30 cancer patients per day. An average of 5 per day are new patients while an average of 10 patients are scheduled daily to receive their course of chemotherapy.
Each site followed the same protocol.

3.3 Participant selection

3.3.1 Target population

The target population for this study was chemotherapy-naive patients aged 18-70 years old, diagnosed with cancer attending KNH cancer treatment center (KNH-CTC) or TCC for their first cycle of chemotherapy and who receive cisplatin 60 mg/m^2 and above as part of their chemotherapy regimen on day 1.

Study eligibility criteria were set out to ensure that: 1) Cancer patients recruited were able to complete the first circle, 2) the Patient history was not such that the patient were likely to develop AKI. 3) The condition of the patient was not such that the patient was likely to have serious complications requiring urgent treatment.A number of previous studies were consulted when determining the inclusion and exclusion criteria. An extensive review of the literature was undertaken to ensure consistency with other studies.

The inclusion and exclusion criteria for this study are noted below:

3.3.2 Patient Inclusion Criteria

Patients were considered eligible for enrolment into this trial if they met the following criteria:

1. Patient aged between 18 and 70 years
2. Signed informed consent and ability and willingness to comply with the protocol
3. Patients that had confirmed diagnosis of a malignant solid tumor by histopathology and cytology investigation.
4. Patients who had not received any prior cancer chemotherapy and were to receive their first course of cancer chemotherapy that included cisplatin ($\geq$60 mg/m^2 on Day 1).

5. Adequate renal function prior to start of chemotherapy, as defined by:

    - Baseline serum creatinine (SCr) prior to start of chemotherapy < 1.5 mg/dl. (132 μmol/l)
    - Estimate Glomerular Filtration Rate (eGFR) ≥ 60ml/min. (no residual kidney disease)

    Baseline creatinine prior to start of chemotherapy was obtained and estimated creatinine clearance/ eGFR was computed using the Cockcroft-Gault formula, as described in Section 3.8.

6. Adequate liver function (within 28 days prior to randomization)

    - Total bilirubin ≤1.5 mg/dl
    - Serum transaminases (Aspartate amino transferase (AST) and / or alanine amino transferase (ALT)) ≤ 3 x the upper limit of normal (ULN) in the absence of parenchymal liver metastases or ≤5 x ULN in the presence of parenchymal liver metastases
    - Serum Albumin Level between 3.5 to 5.5 g/dl

7. Adequate bone marrow function
    - Absolute Neutrophil Count (ANC) ≥1.5 x 10^9/l
    - Platelets count (Plt) ≥100 x 10^9/l
    - Hemoglobin (Hb) ≥9g/dl (can be post transfusion)

8. Adequate Electrolytes balance
    - Serum potassium < 5.0 mmol/L

3.3.3 Patient Exclusion Criteria

The exclusion criteria for the trial were as follows:

- Patient with more than one cancer.
- Medical signs and/or symptoms of active infectious disease
- Evidence of any other disease/metabolic dysfunction that in the opinion of the investigator would have put the participant at high-risk of treatment-related complications or prevented compliance with the trial protocol.
    - Uncontrolled diabetes mellitus (random blood sugar level >200 mg/dl).
    - Uncontrolled hypertension
    - Unstable angina, congestive heart failure, myocardial infarction within the previous year, or evidence of pre-existing peripheral neuropathy
- Patients with exposure to contrast media in the two weeks prior to cisplatin administration.
- Patients who had used potentially nephrotoxic drugs (non-steroidal anti-inflammatory drugs, aminoglycosides, amphotericin B, angiotensin-converting-enzyme inhibitors such as captopril and enalapril and angiotensin receptor blockers such as losartan) in the two weeks prior to cisplatin therapy.
- Patients taking oral magnesium-containing agents (Magnesium oxide, Magnesium hydroxide, Magnesium citrate, Magnesium gluconate, Magnesium chloride).
- Patients on drugs that falsely elevated serum creatinine such as sulfonamides
- Pregnancy
- Patient who required procedures such as administration of radiocontrast media for medical imaging or concomitant medications prohibited by the protocol prior to the completion of the study follow-up

3.4 Sample size estimation

The sample size was determined on the basis of the primary hypothesis that the occurrence of CIN among patients receiving the first course of standard cisplatin based chemotherapy and receiving IV magnesium preloading supplementation will be significantly different from that among patients on standard cisplatin based chemotherapy and who do not receive IV magnesium preloading supplementation. In this regard, occurrence of CIN as manifested by the development of AKI grade 1 or higher (NCI CTCAE, version 4.0) was chosen as the primary outcome of interest.

In order to calculate the target sample size, estimated proportions of CIN occurrence in control and intervention group was needed.These figures were obtained from previous studies on IV magnesium supplementation and CIN. The study from Yoshida et al [40] involving administration of preloading IV magnesium supplementation for reduction of CIN showed that the incidence of grade 1 or higher nephrotoxicity in the non-magnesium group was 81.5% compared to 50.9 % in the intervention group following the first cycle, representing a 30 % absolute reduction in the occurrence of CIN. A study in KNH (30) revealed that 88.7% of the patients on the standard hydration preventive measure who underwent cisplatin based chemotherapy develop AKI grade 1 or higher after the first cycle.
Assuming an absolute reduction in the occurrence of CIN of at least 30% with IV magnesium supplementation, the estimated proportion of participants in the proposed magnesium intervention group who would develop CINwas estimated at approximately 58%.

With regard to type I and type II errors, a two-tailed αof 0.05 and a 1-β of 0.8 was chosen. This level of potential error and statistical power are conventionally considered acceptable in routine health care research. The sample size for this study was calculated using the formula below described by Chan [98] for estimating sample sizes for superiority trials with a dichotomous outcome of interest:

$$m \text{ (size per group)} = C \times \frac{\pi 1\,(1-\pi 1) + \pi 2\,(1-\pi 2)}{(\pi 1 - \pi 2)^2}$$

Where: c = 7.9 for 80% power

$\pi 1$ and $\pi 2$ are the proportion estimates, $\pi 1 = 0.88$ and $\pi 2 = 0.58$.

Therefore, for an 80% power:

$$m \text{ (size per group)} = 7.9 \times [0.88 (1 - 0.88) + 0.58 (1 - 0.58)] / (0.88-0.58)^2$$

$$m = 30.65$$

Hence a minimum of 31 x 2 = 62 patients were required for this study, 31 in each comparison group.

This number wasadjusted upwards to 70 to account for an expected 10% loss to follow up rate [99].

3.5 Recruitment of study participants

Recruitment strategies

Prior to starting recruitment, an announcement of the study was made to the oncology physicians and oncology pharmacists at both study sites.Following this announcement, sensitization meetings were held with study staff from both trial centers to improve their understanding of the study aims and protocols, thus ensuring the cooperation and buy-in of the investigators. In addition, banner advertisements placed at strategic sites within the trial centers were used to inform patients about the study.

Recruitment process

Candidates for the study were initially approached regarding enrollment at one of two different venues.

The first venue was at the radio-oncology clinics at both sites where the attendingphysicians initially identified potential participantswho were scheduled to receive their first course of chemotherapy and who would be receiving a 3-weekly cisplatin as part of their regimen. At this point, the initial offer for information regarding the study to the patient was made by the attending physician with subsequent counselling by the researcher.

The second venue was at the research office. After a list of chemotherapy naïve patient was obtained from the Records Department, and potentially suitable patients for the study were selected by the researcher based on patient files as having a confirmed diagnosis of cancer and a documented cisplatin based regimen prescribed. These patients were telephoned and invited to come to the hospital. They were approached and counselled at this time regarding enrollment in the study.

Patients were then given time to consider the issues and discuss it privately with their relatives. Participants were made aware that study involvement was voluntary and that they could refuse to participate or withdraw at any time without consequence.If the patient was willing to take part in the study, then they were invited to sign the consent form. A signed copy of the consent form wasalso given to each participant.

Any patient who expressed any misgivings about enrollment was not enrolled.

Consenting participantswere provided with a study information sheet and the details of the study. They were then assessed for eligibility by the researcher who filled the eligibility sheet. Regardless of venue of initial counselling, all recruited patients were seen again for clinical assessment by the physician. Following clinical assessment, blood samples were drawn and sentto Lancet laboratory for screening (to assess liver function, blood counts and blood chemistry) prior to enrollment.

When all inclusion and exclusion criteria were addressed and the eligibility of the participant confirmed, the participant was randomly assigned to one of the two comparison groups as described below (Section 3.6).

3.6 Randomization Procedures

Randomization took place after written consent was obtained from the study patients and baseline information was gathered. Patients who fulfilled the eligibility criteria, who agreed to participate and who could financially afford the treatment were then randomly allocated into one of the two treatment groups.

3.6.1 Allocation—sequence generation

The study biostatistician used a computer program to generate the random allocation sequence. Block randomization was applied, whereby random numbers were generated and allocated in a one-to-one ratio to a sequence of permuted blocks, with stratification for gender and center. The resultant randomization schedule ensured that both centers contributed a comparable number of subjects, and that the gender distribution was balanced in the final sample of patients.

3.6.2. Allocation-concealment mechanism

Once the randomization schedule was generated, the biostatistician used this schedule to create a sequentially numbered, sealed, opaque envelopes each containing a slip which contained a code indicating which of the two different interventions an assigned patient was to receive. The envelopes were provided securely to the research coordinator at each centercarrying out eligibility screening and recruitment.The coded allocation schedule was held by the hospital pharmacist at each trial center while the envelopes were held by the research coordinator.

3.6.3. Allocation—implementation

Following successful recruitment and assessment for eligibility, on day 1 of patient treatment the next appropriate envelope in sequence was taken and the patient's name and ID number was written beside the number of the envelope. The envelope was then handed over to the hospital pharmacist by the research coordinator. Once the envelope had been opened bythe hospital pharmacist at each trial centre, the allocation was made and the details of the patient was noted on the allocation slip contained in the envelope. The envelope and allocation slip were then kept under lock and key by the pharmacist.

No envelopes were opened out of sequence, and no envelopes were skipped.The randomization number and the treatment allocation were kept concealed from the patient, investigator, laboratory and study personnel till completion of the study.

Oncethe group allocation for each patient was made, the pharmacist (not involved in care of the trial patients and independent of the investigator) prepared the corresponding pre-hydration infusion as described in section 3.7.2, and labeled the infusion bags with the corresponding patient’s ID, date and hour. The infusion bags bore no indication of whether the contained magnesium supplementation or not, and were made available to the research nurse immediately

before administration. The pre-hydration solutions were administered at the same rate for intervention and control group by the research nurse at each trial center.The pharmacist confirmed that the administration of the pre-hydration infusions was done as planned.

3.7. Interventions and Treatments

3.7.1. Supply of Study Medication at the site

Drugs and solutes were supplied by the hospital pharmacy in each trial center in their commercially available forms. They comprised:

- Granisetron 3mg and dexamethasone 4mg/ml)
- Ondansetron 8mg tablet
- Dexamethasone 4mg tablet
- Mannitol 20 % (100ml)
- Potassium chloride 15% (10ml).

The main study drugswere supplied by the oncology pharmacy of Kenyatta National hospital for use at both centers to ensure uniformity of treatment. These were:

- 10 ml of 50 % Magnesium sulfate solution and
- Cisplatin 50 mg /50 ml

3.7.2. Preparation of the intervention and control pre-hydration solutions

The pre-hydration solutionwas prepared by the hospital pharmacist and contained either:

- Potassium chloride (KCl) 20 mmol (1.5g) plus Magnesium sulfate 8 mEq (1g) diluted in 1litre of normal saline for the intervention group
- Potassium chloride (KCl) 20 mmol only diluted in 1litre of normal saline for the control group

The solutions were prepared on the day of administration and fully inverted approximately 10 times to ensure proper mixing.

3.7.3. Delivery of treatments and interventions

Intervention group

The experimental manoeuvre in this trial was the administration of Magnesium sulfate as part of the pre-hydration solution. As per the NCCN recommendation 8 mEq (1g) diluted in 1litre of normal saline was administered for the intervention group as preloading supplementation before administration of cisplatin.

The total volume of pre-hydration solution administered to the patients was equal both in experimental and control group. Each patient received a total volume of 3050 ml of normal saline 0.9%. Besides pre-hydration, participants were encouraged to drink a minimum of 500 ml of water daily, following administration of cisplatin and were given a patient information card to monitor their fluid intake.

The pre-hydration solutions were administered to the participants in both intervention and control group by the research nurse at each trial center.

Participants randomized to the intervention arm were given the following treatment:
On Day 1

1. Antiemetic prophylaxis

Prior to commencing chemotherapy, standard antiemetic prophylaxis was administered. A 5-HT_3 receptor antagonist (granisetron) 3 mg, and dexamethasone (9.9 mg) mixed together with 50 mL of Normal Saline was administered by 15-minute I.V. infusion as a single dose at least 30 minutes before initiation of chemotherapy.

2. Pre-hydration with $MgSO_4$ and KCl supplementation

Following the antiemetic prophylaxis, Potassium chloride (KCl) 20mmol (1.5g) plus Magnesium sulfate 8 mEq (1g) diluted in 1litre of normal saline as described previously (section 3.7.3) was administered by IV infusion over 2 hours.

3. Diuresis

Following the prehydration before the administration of cisplatin, 200ml of 20% mannitol was administered as forced diuresis by IV infusion over 30 minutes.

4. Cisplatin and Other cytotoxic drugs

The patient-specific cisplatin-based chemotherapy regimens varied from patient to patient as prescribed by the medical oncologist. Some patients received only cisplatin while others were prescribed a cisplatin-based regimen that contained two or three cytotoxic drugs. The cytotoxic drugs were prepared as per manufacturer's instructions and administered as per prescription. Immediately after diuresis Cisplatin dose was diluted in 500 ml of 0.9% NaCl solution (N/saline) and administered by IV infusion over 90 minutes. All patients received a cisplatin dose > 60 mg/m^2.

5. Post hydration

Following cisplatin administration, 1 liter 0.9% sodium chloride + 20mmol KCl was administered by IV infusion over 2hrs.

On day 2

Delayed antiemetic prophylaxis was started with Dexamethasone tablet 4 mg orally twice daily plus ondansetron tablet 8 mg twice a day.

24 hoursafter cisplatin administration,an additional IV hydration of 500ml 0.9% sodium chloride was administer by IV infusion over 1hr.

On day 3-5

Delayed antiemetic prophylaxis was continued until day 5 after cisplatin administration: Dexamethasone tablet 4 mg orally twice a day plus ondansetron tablet 8 mg twice daily.

The treatment received by patients in the intervention group is summarized in Table 3.1**.**

Table I: Intervention treatment (Mg- supplementation Group): Drugs administered, day, route and rate of administration.

Day	Treatment	Drug & solute	Administration	Time
Day 1	Acute Antiemetic prophylaxis	Normal saline 50ml Dexamethasone 9.9 mg Granisetron 3mg	iv	30 mn
	Pre-hydration	Normal saline 1000 ml Potassium chloride (KCl) 20mmol (1.5g) Magnesium sulfate 8 mEq (1g)	iv	120mn
	Diuresis	200 ml of mannitol 20%	iv	30 mn
	Other Cytotoxic drugs	Per prescription	Per prescription	Per prescription
	CDDP	Normal saline (NaCl 0.9%) 500ml Cisplatin as prescribed	iv	90mn
	Post hydration	Normal saline 1000ml Potassium chloride (KCL) 20mmol		120mn
Day 2	Delay antiemetic prophylaxis	Dexamethasone 4mg tablet Ondansetron 8mg tablet	Orally twice a day Orally twice a day	
	Additional hydration	Normal saline 500 ml	iv	60mn
Day 3 to Day 5	Delay antiemetic prophylaxis	Dexamethasone 4mg tablet Ondansetron8mg tablet	Orally twice a a day Orally twice a day	

Control Arm

Patients who were allocated in the control arm (non-Mg preloading group) did not receive Magnesium sulfate. Theyreceived the same treatment as the intervention group (magnesium preloading group), except that the pre-hydration solution whichcontained 20mmol of Potassium chloride (10ml of KCl 15%) but NOT magnesium sulfate.

Standardization of Interventions

All patients in all two groups were reviewed by the investigator, who remained blind to group allocation and their outcome measures. They were given standardised advice and instructed by the principal investigator about the purpose and use of antiemetic drugs for the prevention of delayed emesis due to cisplatin administration. This was reinforced in an information booklet given to each patient, i.e. the ‘’Patient Information Card’’ (Appendix 2) which also contained hydration instructions to protect kidney and emergency medical conditions that required calling the investigator.

3.8.Participant follow-up

Following administration of the treatment on day 1 and day 2, patients attended visits for review on day 2, day 6, day 10 and day 17, which entailed:

1. Recording any Adverse Experiences
2. Reviewing of antiemetic drugs compliance
3. Recording changes to concomitant medications.
4. Performing physical examination.
5. Performing and recording vital signs.
6. Collecting blood for serum creatinine and eGFR determination

On day 17, a full laboratory work up (hematology and chemistry) was added to the review to prepare the patient for the second cycle. On day 21 the off study form (Appendix 6) was filled by

the investigator and the patient was officially handed over by the principal investigator to the oncology physician for continuation of chemotherapy.

3.9 Outcomes

3.9.1. Primary Outcome Measures

The primary outcome measure was the incidence of at least grade 1 AKI, as defined by the Common Terminology Criteria for Adverse Events (CTCAE) version 4.03 (Appendix 3), after the first cycle of cisplatin based chemotherapy. According to these criteria, AKI grade 1 is present when an abrupt reduction in kidney function results in an absolute increase in SCr level by ≥0.3 mg/dL (26.5 µmol/L), or an increase of 1.5-fold in the baseline SCr level known or presumed to have occurred within prior 7 days.

The serum creatinine concentration was determined before the first course of cisplatin chemotherapy (baseline value) and on Day 2, 6, 10, and 17 after cisplatin administration. The sample for this procedure wasdrawn by the research nurse then labeled, stored and transported and analyzed as described in section 3.13 below.

The increase in the serum creatinine concentration was calculated as the difference between the serum creatinine values obtained and the baseline value. The maximum value of serum creatinine level during follow up visit was used to assess nephrotoxicity after the first course of chemotherapy. This outcome measure was performed as a double blind assessment.

3.9.2. Secondary Outcome Measures

1. Estimated creatinine clearance before and after cisplatin infusion between the magnesium supplementation group and non-magnesium supplementation group after the first chemotherapy course. The estimated creatinine clearance was computed using the Cockcroft-Gault formula [103], and was determined at baseline and after cisplatin infusion.
2. Grading of AKI according to CTCAE version 4.03 (Appendix3). According to the increase in the serum creatinine concentration calculated as the difference between the maximum value

after the first course of chemotherapy and the baseline value the number of patient with each grade of AKI by group was determined.

3.10. Blinding Procedures

This study implemented a double-blind design. Six groups of individuals involved in the trial (patients, investigators, caregivers/clinicians, research associates, outcome assessors and laboratory technicians) were kept unaware of what treatment arm participants had been randomized to.All the outcome measurements taken at baseline and during follow up at day 2, day 6 day 10 and day 17 were measured by the laboratory that remained blinded to group allocation throughout the study.The outcome assessor who received and assessed the laboratory resultswas not involved in any other aspect of the study. The pharmacist did not reveal to the investigator, the patient and the research team which group thepatients had been allocated to.

3.11. Data collection

3.11.1. Data Collection Instruments

A Case Report Form (CRF) available in Appendix 4 was used as data collection tool. It was designed to record all observations and other pertinent data for each participant. The CRF contents wereconsistent with the FDA's CDASH (Clinical Data Acquisition Standards Harmonization) standards [100].

Study personnel at each site entered data from source documents corresponding to a participant's visit into the protocol-specific Case Report Form (CRF) when the information corresponding to that visit was available. The data collected in a case report form are as follows:

- Concomitant Medications

All concomitant medication and concurrent therapies were documented at baseline/screening on the CRF and review at day 2, 6, 10 and 17. Dose, route, unit frequency of administration, and indication for administration and dates of medication was captured.

- Demographics data

Demographic information (date of birth, gender, race etc.) were recorded at screening on the CRF.

- Medical History

Relevant medical history, including history of current disease, and information regarding underlying diseases were recorded at screening on the CRF.

- Physical Examination

A complete physical examination was performed by the Clinical Research Associate who was physician during all visits. New abnormal physical examination findings were documented and were followed.

- Vital Signs

Body temperature, blood pressure, pulse and respirations were performed after resting for 5 minutes on screening and study day 2, 6, 10, 17 by the research nurse and recorded on the CRF.

- Adverse Events

Information regarding occurrence of adverse events was captured throughout the study. Duration (start and stop dates and times), severity/grade, outcome, treatment and relation to study drug was recorded on the CRF.

- Laboratory Measurements

Hematology test, Blood Chemistry Profile test as detailed in section 3.3were performed at screening and at day study 17. Only serum creatinine level was performed at baseline,day 2, 6 and 10.

3.11.2. Determination of the Primary outcome

3.11.2.1. Sample collection, Handling and Transport

At baseline, mid-point and post-intervention participants were assessed on the primary outcome measures.The research nurse collected at each visit a blood specimen of at least 3-5 mL by venipuncture into a vacuum blood collection tube, specifically a serum separating tube (yellow top tube). The tube was inverted about 5 times to mix the sample with the silica and separator. After collection, the specimens were labelled with date of collection and the participant's study

code number, and immediately sent for analysis at pathologists Lancet Kenya Limited, an ultramodern laboratory located in Upper Hill Nairobi which provides a range of routine, specialized and referral services.

For transportation of the specimens to the laboratory, blood sample tubes were placed in a Styrofoam container and a cooler box with ice to maintain temperature between 2-8 ˚C. A maximum limit of two hours wasensured by the administrative assistant at each trial center and the laboratory courier for the transfer of the specimens from the study site to the laboratory.

3.11.2.2. Sample analysis

The blood samples were analyzed within 24 hours after collection. Measurement of creatinine in serum was analyzed using Jaffe's method, which is based on the Jaffe reaction. Creatinine reacts with picrate ion formed in alkaline medium to develop a red-orange colour. The colour produced from the sample is then compared in a colorimeter at wave length of 505 nm with that produced by a known amount of creatinine under the same condition [101].

3.11.3. Determination of Secondary outcomes

Estimated creatinine clearance (ml/min) was determined using baseline serum creatinine and maximum post serum creatinine levels for each patient. The Cockcroft–Gault formula [100] was used for calculation:

CrCl [ml/min] = (140 – age [years] × weight [kg]) × 0.85 [if female] / (72 × sCr [mg/dl]).

ΔCrCl was calculated using the formula: ΔCrCl [ml/min] = (CrCl [ml/min] before chemotherapy) – (CrCl [ml/min] after chemotherapy)

3.12. Data management

3.12.1. Data Forms and Data Entry

Pathologist Lancet Kenya laboratory provided email electronic reports to the outcome assessor to securely view the results. The patient's results were kept by the outcome assessor and only day 17 results were printed and attached to the patient file. All changes to the study database were documented after information had been captured using Case Report Form, all data was entered electronically into a Microsoft Access (2013) computer database. Completed Case Report Forms

were checked for completeness and accuracy by the investigator and administrative assistant at each trial center against the source data. Checks was applied at the time of data entry into a specific field and/or before the data was written (committed) to the database.
The Investigator was responsible for all information collected on patients enrolled in this study.

3.12.2. Security and Back-Up of Data

Access to the data base was limited by the use of passwords and only the investigators and outcome assessor and administrative assistant were allowed access. The database was backed up on separate media, once the updates were done. All forms related to study data was stored in a locked cabinet. Only the administrative assistant at each trial center and the principal investigator had access to these cabinets

3.13. Data analysis

Statistical analyses were conducted on modified intention-to treat (mITT) bases. All participants who were enrolled and randomized to one of the two groups and received the allocated intervention as prescribed by the study protocol were included in the evaluation of the primary and secondary outcomes. Any subjects who did not receive the allocated intervention as prescribed by the study protocol were excluded from the analysis.
The baseline characteristics and laboratory data were presented as the means and standard deviations or mean and range according to the normality of the distribution for continuous variables and as frequencies and percentages for categorical variables. The incidence of AKI was compared between the two groups using a $\chi 2$ test. The differences in changes in SCr levels and in the eGFR were analyzed using Student's t-test or the Mann-Whitney U test.
In addition, the survival analysis was conducted using the Kaplan Meier and the difference in time to event was analyzed using the log-rank test. The difference in the creatinine clearance trend was analyzed using generalized linear model. A value of $P < 0.05$ was considered statistically significant. All analyses were performed using Stata statistical software, version 10.0 (StataCorp LP, College Station, Tex., USA)

3.14. Quality assurance

3.14.1. Quality Control of the Pathologists Lancet Kenya Limited

Pathologists Lancet Kenya Limited applied stringent internal quality control procedures. They ran quality control checks daily, with review of results and appropriate action was taken when necessary. Weekly internal audits by the Head of Department and Laboratory Manager were undertaken. In addition, an External Quality Assurance (EQA) programme was running on a monthly basis.

3.14.2. Training and certification plans

Each center's personnel were trained centrally in the study requirements, standardized measurement of height, weight, and blood pressure, requirements for laboratory specimen collection, counseling for adherence and the eliciting of information from study participants in a uniform reproducible manner. Entering data forms, responding to data discrepancy queries and general information about obtaining research quality data was also covered during the training session.

3.15. Ethical considerations

3.15.1. Informed Consent

Researchers at each centre ensured that the patients were given full and adequate oral and written information about the nature, purpose, possible risks and benefits of the trial. Patients were also notified that they are free to discontinue from the trial at any time. The patientswere given the opportunity to ask questions and were allowed as much time as they required to consider the information provided.

3.15.2. Confidentiality

Participant confidentiality was strictly held in trust by the research staff. This confidentiality was extended to cover testing of biological samples in addition to the clinical information relating to participants. The study protocol, documentation, data and all other information generated were held in strict confidence. No information concerning the study or the data was released to any unauthorized third party. All laboratory specimens, evaluation forms, reports and other records related to the trial that left the site were identified only by the Participant Identification Number

(PID) to maintain participant confidentiality. Clinical information was not released without written permission of the participant, except as necessary for monitoring by KNH/UON-ERC.

3.15.3. Independent Human Research Ethics Committee Approval

Ethics approval for this study was granted by the Kenyatta National Hospital/University of Nairobi Ethics and Research Committee (KNH/UON-ERC).
Approval number: KNH-ERC /A /245 (appendix5).

3.15.4. Participant Reimbursement

Participants were given 400 Kenya shillings on day 6 and day 10 as reimbursement for transport costs. This was because the follow up sessions on thesedays following cisplatin infusion were not part of the routine visitsfor patients on cisplatin chemotherapy.

3.15.5. Financial Disclosure and Conflicts of Interest

There was no conflict of interest to declare. A declaration confirming the absence of any conflict of interest was signed.

CHAPTER 4: RESULTS

4.1. Participant enrollment, allocation and follow up

Between June 2015 and October 2015, 104 patients were screened of whom 33 were excluded for various reasons: 29 did not fulfil the inclusion criteria, one declined to participate and 3 were not enrolled due to financial constraints. Seventy-one patients (71) were randomized to receive either IV magnesium supplementation (n=35) or no IV magnesium supplementation (n=36). Two patients did not receive the complete intervention as prescribed by the study protocol; of these, one died and the other did not show up on day 2. These two subjects were considered as significant deviations from the protocol, and were discontinued from the study. Therefore, a total of 69 patients were followed up, and all 69 patients (100%) completed the prescribed 17 days of follow up and their data was available for safety analysis. This is summarized in the Consort Flow Diagram (Figure 1).

4.2. Participants baseline characteristics

The baseline sociodemographic and clinical characteristics of the enrolled subjects are presented in Table II and Table III, respectively. The mean age of all the patients was 49 years (range 18 – 70), with more females (n=42, 60.87%) than males (n=27, 39.13%). The most common malignancies were cervical cancer (42.03%), esophageal cancer (15.94%), and nasopharyngeal cancer (10.14%). All patients were of African origin and 84 % were married.

Analysis of the distribution of the baseline characteristics revealed that a higher proportion of males were allocated to the Non Magnesium group (59.26%) than the magnesium group (40.74%). However, the differences in the gender composition between the two groups were not statistically significant (p=0.345). In addition, the mean dose of cisplatin did not differ significantly among treatment arms (mean dose (mg): 123.85 ± 2.39 in the magnesium group and 122.18 ± 2.52 in the Non Magnesium group, p=0.634).

Overall, the two treatment arms were comparable with respect to sociodemographic (Table II) and clinical (Table III) characteristics thus providing reasonable assurance that the randomization was performed successfully.

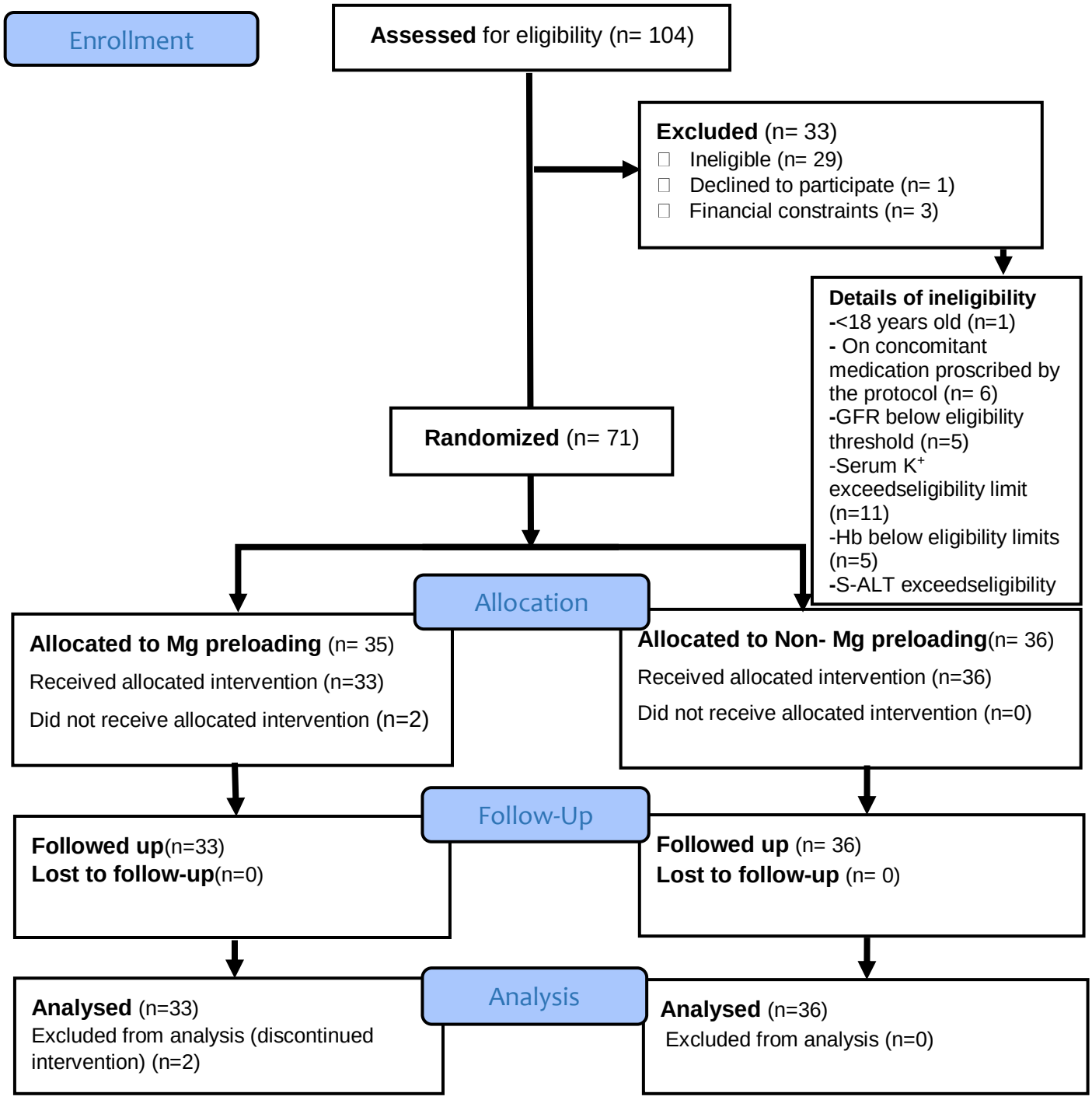

Figure 1: Consort Flow Diagram outlining participant enrollment, allocation and follow up

Table II: Participant Demographics Characteristicsaccording to Magnesium exposure.

Characteristics	Overall	Mg Preloading	Non-Mg Preloading	P value
Age; Mean (years)	49.2 (18 – 70)	50.33 (21 – 67)	48.17 (18 – 70)	0.485
Sex; n (%)				
Male	27 (39.13)	11 (40.74)	16 (59.26)	0.345
Female	42 (60.87)	22 (52.38)	20 (47.62)	
Marital Status n (%)				
Married	57 (82.61)	28 (49.12)	29 (50.88)	0.680
Single	7 (10.14)	2 (28.57)	5 (71.43)	
Widowed	3 (4.35)	2 (66.67)	1 (33.33)	
Divorce	2 (2.90)	1 (50)	1 (50)	
Occupation n (%)				
Unknown	2 (2.90)	0 (00)	2 (100)	0.702
Homemaker	8 (11.59)	5 (37.5)	3 (62.50)	
Farmer	19 (27.54)	12 (63.16)	7 (36.84)	
Student	3 (4.35)	1 (33.33)	2 (66.67)	
Business	12 (17.39)	5 (41.67)	7 (58.33)	
Other	17 (24.64)	6 (35.29)	11 (64.71)	
Teacher	5 (7.25)	2 (40)	3 (60)	
Driver	2 (2.90)	2 (100)	0 (00)	
Retired	1(1.45)	0 (00)	1(100)	
Ethnicity n (%)				
Luhya	5 (7.25)	3 (60)	2 (40)	0.068
Kikuyu	19 (27.54)	9 (47.37)	10 (52.63)	
Kamba	15 (21.74)	5 (33.33)	10 (66.67)	
Kisii	3 (4.35)	2 (66.67)	1 (33.33)	
Meru	9 (13.04)	5 (55.56)	4 (44.44)	
Luo	4 (5.80)	3 (75)	1 (25)	
Others	13 (18.84)	5 (38.46)	8 (61.54)	
Trial center n (%)				
Kenyatta National hospital	36 (57.17)	17 (47.22)	19 (52.78)	0.916
Texas Cancer Centre	33 (47.83)	16 (48.48)	17 (51.52)	

Table III: Participant baseline Clinical characteristics according to magnesium exposure

Characteristics	Overall	Mg Preloading	Non- Mg Preloading	P value
Weight ;mean ± SD (kg)	62.13 ± 11	63.69 ± 12.43	60.72 ± 9.46	0.267
Height; mean ± SD (cm)	164.53 ± 7.96	165.03 ± 7.76	164.08 ± 8.24	0.625
BSA ; mean ± SD (m 2)	1.67 ± 0.17	1.69 ± 0.19	1.65 ± 0. 17	0.380
Type of cancer n (%)				
Cervical	29 (42.03)	16 (55.17)	13 (44.83)	0.264
Nasopharyngeal	7 (10.14)	0 (00)	7 (100)	
Esophageal	11 (15.94)	4 (36.36)	7 (63.64)	
Oral	6 (8.70)	3 (50)	3 (50)	
Hypopharyngeal & laryngeal	6 (8.70)	4 (66.67)	2(33.33)	
Stomach	4(5.80)	2 (50)	2 (50)	
Sarcoma	2(2.90)	1 (50)	1(50)	
Gastroesophageal	1 (1.45)	1 (100)	0 (00)	
Others	3 (4.35)	2 (66.67)	1 (33.33)	
Metastatic at presentation				
No	61 (88.41)	29 (47.54)	32 (52.46)	0.896
Yes	8(11.59)	4 (50)	4 (50)	
Renal function status (%)				
Hb ; mean ± SD	12.82 ± 1.76	12.73 ± 1.74	12.90 ± 1.79	0.703
BUN (mmol/l); mean ± SD	3.6 ± 1.07	3.59 ± 1.03	3.61 ± 1.12	0.947
Cr (mg/dl); median(range)	0.7 (0.40;1.39)	0.68 (0.45 ; 1.39)	0.72 (0.40 ; 1.11)	0.327
CrCl; mean ± SD	104.15 ± 27.9	106.81 ± 29 .25	101.70 ± 26.83	0.451
Sodium (Na) ; mean ± SD	137.04 ± 2.95	137.03 ± 3.26	137.05 ± 2.67	0.972
Potassium (K) ; mean ± SD	4.29 ± 0.42	4.32 ± 0.37	4.25 ± 0.47	0.511
Chloride (Cl); mean ± SD	100.46 ± 3.5	100.63 ± 3.61	100.30 ± 3.61	0.705
Albumin (Alb); mean ± SD	40.22 ± 4.47	39.90 ± 4.23	40.52 ± 4.73	0.574

Table III continuation

Comorbidity n (%)				
No comorbidity	63 (91.30)	29 (46.03)	34 (53.97)	0.659
hypertension	3 (4.35)	2 (66.67)	1 (33.33)	
Diabetes	1 (1.45)	1 (100)	0 (00)	
Other	2 (2.90)	1 (50)	1 (50)	
Smoking status n (%)				
No	62 (89.86)	29 (46.77)	33 (53.23)	0.603
yes	7 (10.14)	4(57.14)	3(42.86)	
Alcohol consumption n (%)				
No	61 (88.41)	28 (45.90)	33(54.10)	0.377
Yes	8(11.59)	5 (62.50)	3 (37.5)	
Chemotherapy regimens n (%)				
Cisplatin only	40 (57.97)	18 (45)	22 (55)	0.581
Cisplatin combination regimen	29(42.03)	15(51.72)	14 (48.28)	
Cisplatin dose (mg) ; mean ± SD	122. 97 ± 1.73	123.85 ± 2.39	122.18 ± 2.52	0.634
Combination drugs n (%)				
Paclitaxel	22 (75.86)	12 (54.55)	10 (45.45)	0.853
5-Fluorouracil	3(10.34)	1 (33.33)	2 (66.67)	
Other drugs	4(13.79)	2 (50)	2 (50)	
Concurrent radiation n (%)				
No	40 (57.97)	18 (45)	22 (55)	0.581
Yes	29(42.03)	15 (51.72)	14 (48.28)	

4.3. Incidence of Cisplatin-Induced Nephrotoxicity

On the basis of the SCr data collected, all patients who developed cisplatin-induced nephrotoxicity (CIN) were identified. These were defined as all patients who developed Grade 1 or higher SCr elevation following the first cycle of cisplatin-based chemotherapy (as defined by the CTCAE, version 4.0).

The incidence of a Grade 1 or higher SCr elevation was 12.12 % (n= 4) in the Mg preloading group and 33.33% (n=12) in the non-Mg preloading group (Figure 2).Intravenous Mg preloading supplementation significantly reduced the incidence of CIN following the first cycle of cisplatin-based chemotherapy [risk difference = -0.21, 95% CI: -0.40, -0.02; P = 0.037].

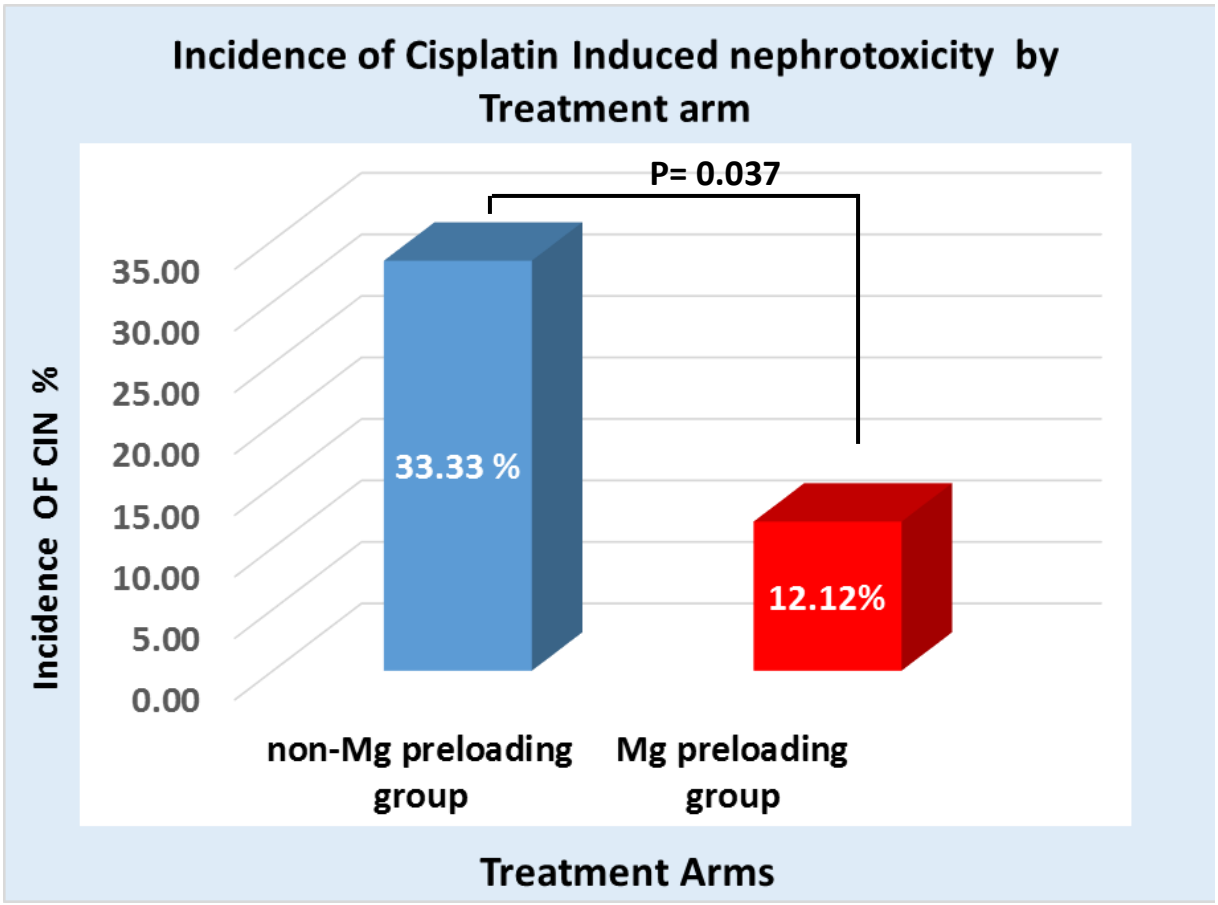

Figure 2: Incidence of CIN (CTCAE Grade 1 and above) in the Mg preloading group and the non-Mg preloading group.

4.4. Severity of nephrotoxicity between the treatment arms

4.4.1. Comparison of the median maximum serum creatinine level in the Mg preloading group and the non-Mg preloading group

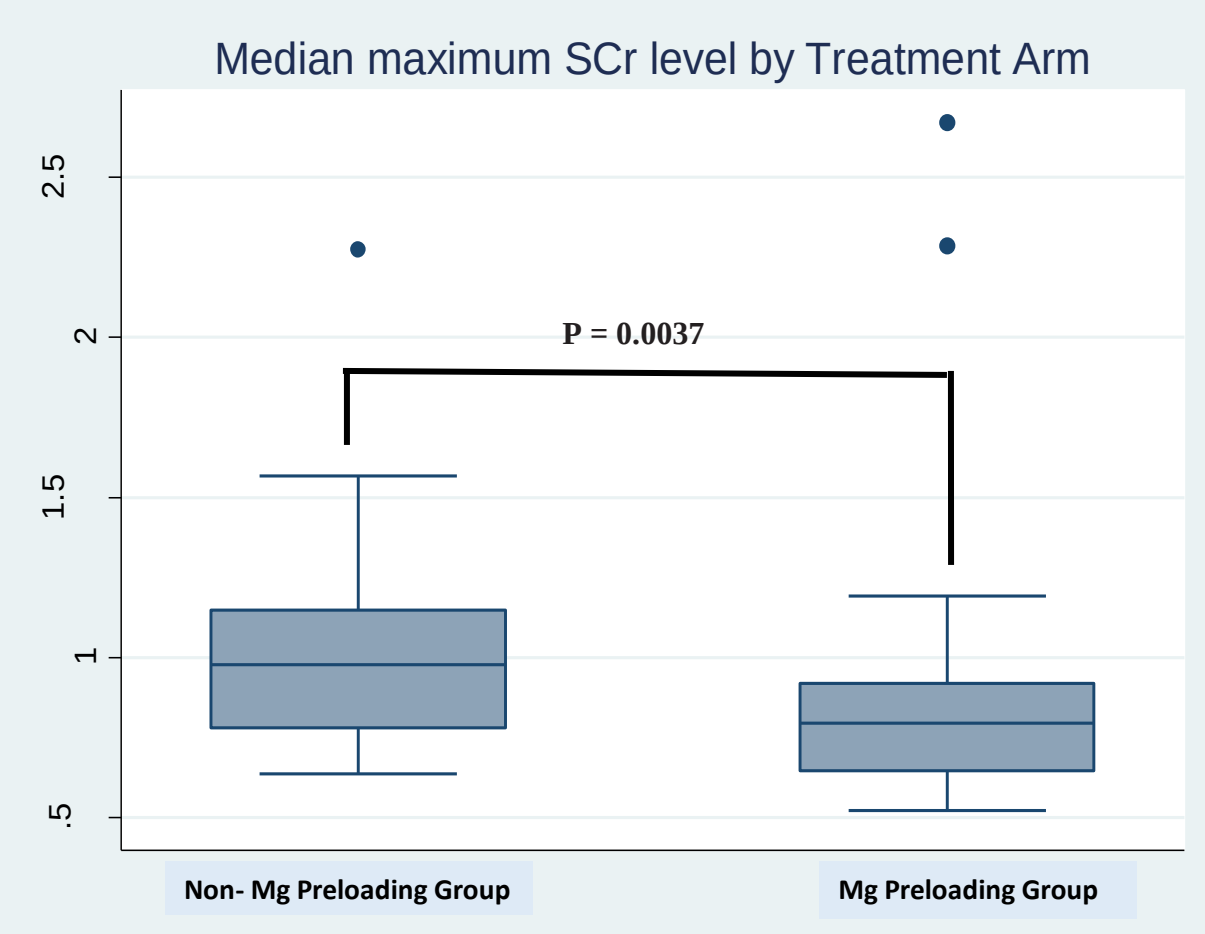

Figure 3: Box-and-whisker plot comparing the median maximum serum creatinineby treatment groups

The maximum serum creatinine level observed during follow up was determined for each patient and used to calculate the median maximum serum creatinine for each treatment arm.

The median maximum serum creatinine level was 0.80 mg/dl (range: 0.52–2.67 mg/dl) in the Mg preloading groupand 0.98 mg/dl (range: 0.64–2.27 mg/dl) in the non-Mg preloadinggroup (Figure 3). The median maximum serum creatinine level in the Mg preloading group was significantly lower than that in the non-Mg preloading group (P=0.0037)

4.4.2. Change of serum creatinine from baseline

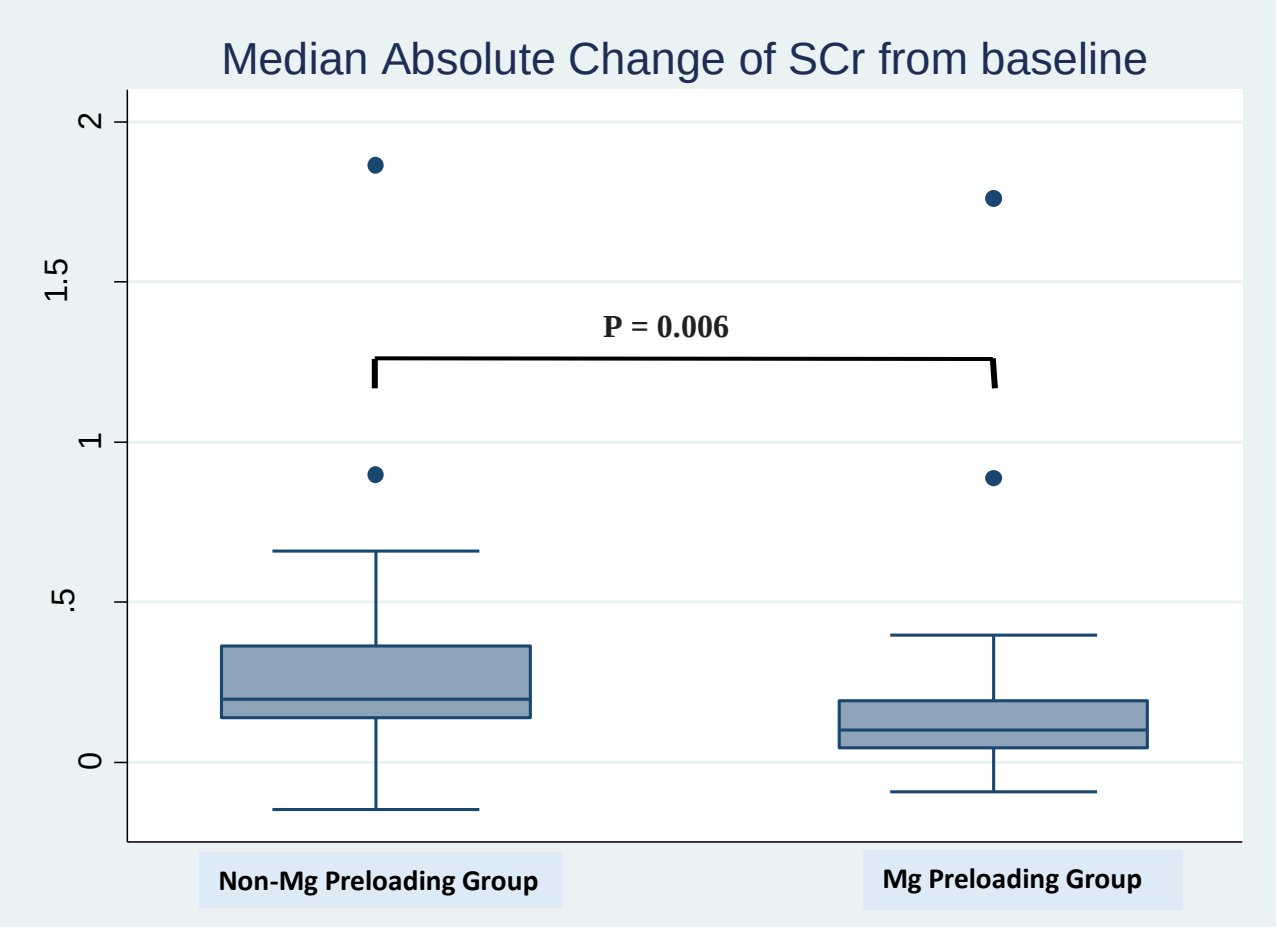

Figure 4: Box-and-whisker plot comparing the Median Maximum change of Serum Creatinine concentrations from baseline by treatment groups.

The maximum absolute SCr level change from baseline for each patient was calculated as the difference between the most deviant (increase or decrease) SCr level observed and the SCr at baseline. This was calculated for each patient and used to determine the median maximum change in SCr level for each treatment arm (Figure 4).

Patients who received intravenous magnesium preloading supplementation (n = 33) showed a median maximum change in serum creatinine level of 0.10 mg/dL (range: -0.090, 1.761), whereas those who did not receive magnesium (n = 36) showed a median maximum change of 0.19 mg/dL (range: -0.147, 1.86). An overall elevation in SCr was observed in both groups

following cisplatin-based chemotherapy, with the results indicating that magnesium supplementation significantly reduced the maximum elevation of SCr induced by cisplatin (P = 0.006).

4.4.3. Change in creatinine clearance from baseline.

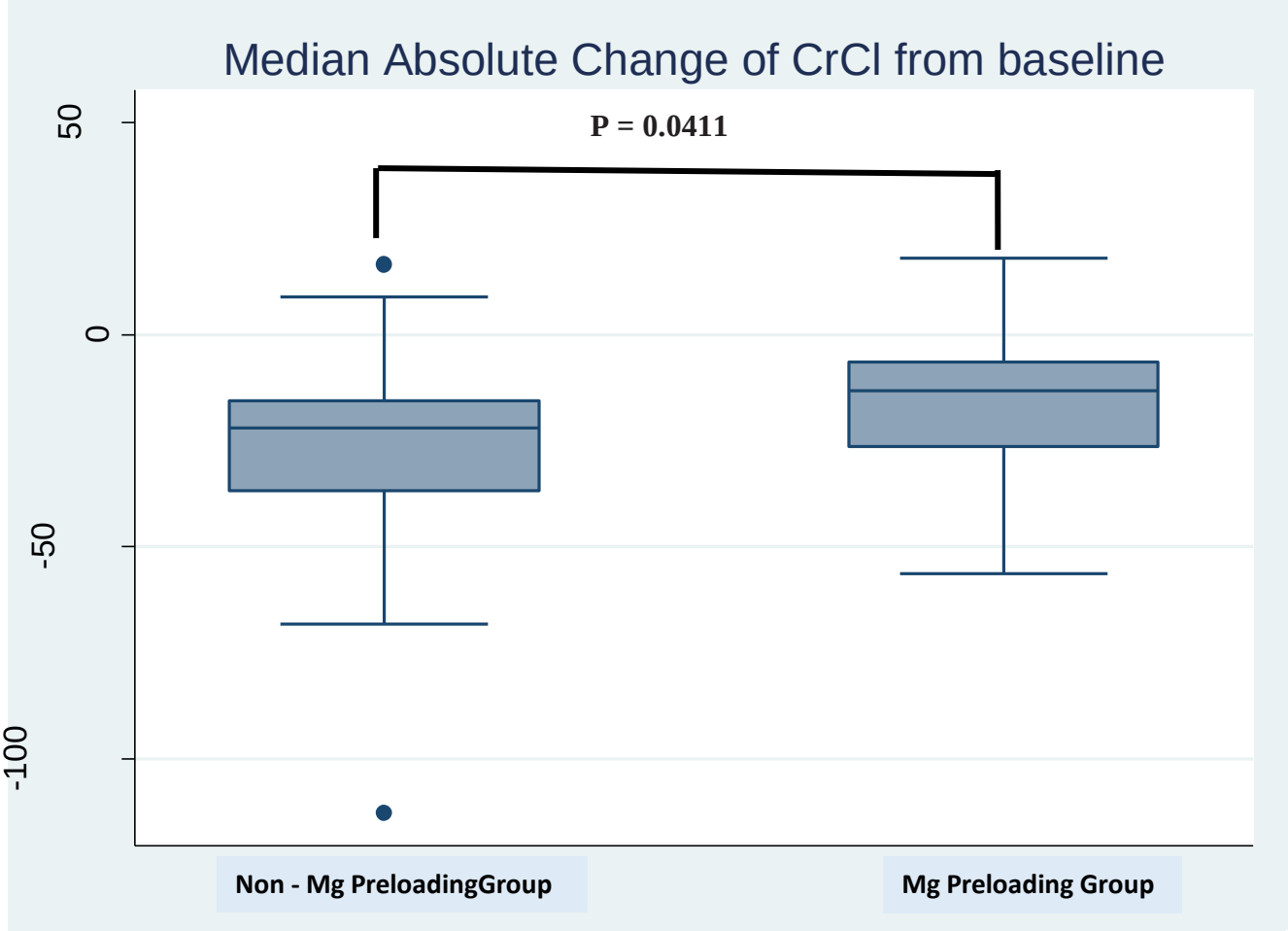

Figure 5: Box-and-whisker plot comparing the Median Maximum change of Creatinine Clearance (CrCl) to baseline by treatment groups

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Creatinine clearance (CrCl) was calculated by The Cockcroft–Gault formula. The maximum absolute change in CrCl from baseline for each patient was therefore determined as the difference between the most deviant (increase or decrease) CrCl observed and the CrCl at baseline. This was calculated for each patient and used to determine the median maximum change in CrCl for each treatment arm (Figure 5). Patients who received intravenous magnesium

preloading supplementation (n = 33) showed a median maximum change in CrCl of -13.2 ml/min (range: -56.3, 17.9), whereas those who did not receive magnesium (n = 36) showed a median maximum change of -22.05 ml/min (range: -112.8, 16.5). An overall reduction in CrCl was observed in both groups following cisplatin-based chemotherapy, with the results indicating that magnesium preloading supplementation therapy significantly limited the decline in CrCl induced by cisplatin (P = 0.0411).

4.5. Comparison of Time to event (Time from treatment to CIN) between the two treatments Arms.

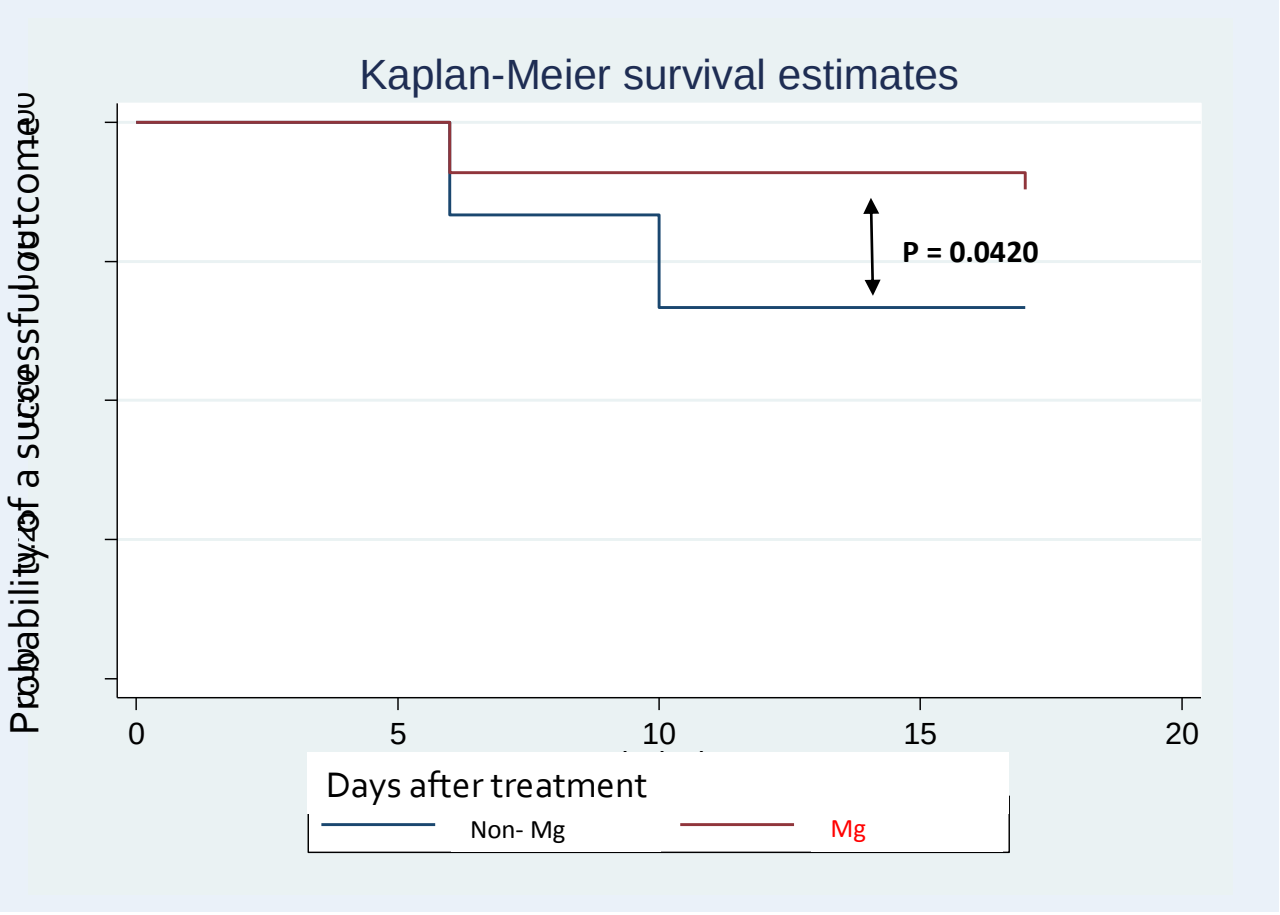
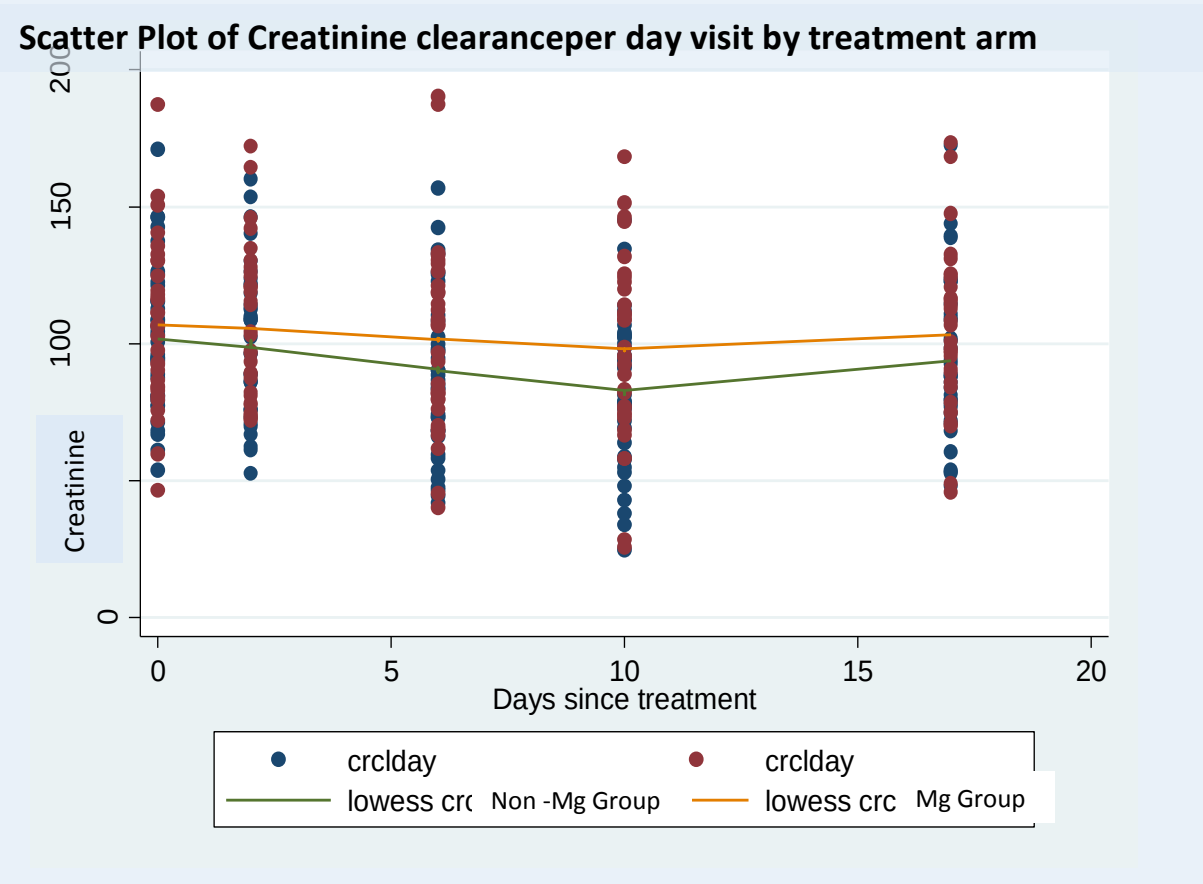


Figure 6: Kaplan Meier survival curves showing the comparison of time to CIN between the two treatments groups. The time from treatment with cisplatin-based chemotherapy to development of CIN (i.e. time-to-event) was compared for the patients in the two treatment arms using the Kaplan-Meier analysis and the log-rank test. The Kaplan-Meier survival curves are presented in Figure 6.

The Kaplan-Meier survival analysis showed that survival rate for the Mg group was consistently higher than that for the Non Mg group at each point of analysis: day 6 [(0.90 95% CI: (0.74-0.96) **vs** (0.83 95% CI: 0.66 - 0.9214)]; day 10 [(0.90 95% CI: (0.74- 0.96) vs 0.66 95 % CI (0.48 - 0.79)]; day 17 [0.8788 95% CI (0.70 - 0.95) vs 0.66 95% CI (0.48 - 0.79)].The log rank test revealed that magnesium supplementation was associated with extended survival without CIN (P= 0.0420).

4.6. Evolution of calculated creatinine clearance over time between the two treatment arms



*The trend lines were generated as locally weighted least squares (LOWESS) curves

Figure 7: Calculated creatinine clearance (CrCl) over time by treatment groups. The continuous orange line represents the trend line for the Magnesium Preloading group, the continuous green line represents Non-Magnesium group.

To obtain an overall picture of the change in renal function with time, the trend of CrCl from baseline to day 17 was evaluated for both treatment groups. A global decrease of CrCl was

demonstrated from baseline to day 10 for both treatment groups. Both trend curves hit an inflection point corresponding to the day 10 evaluation of CrCl. This is followed up by an increase in CrCl toward the end of the follow up period, suggesting an overall initial deterioration followed by recovery of renal function (Figure 7). The difference in the CrCl trends between the two groups was assessed using generalized linear model and the trends were found to be significantly different (P = 0.003).

4.7. Predictors of Cisplatin Induced Nephrotoxicity

4.7.1 Bivariate analysis of potential risk factors

Different statistical tests were used to investigate on the association between individual baseline variables (demographics and clinical characteristics) and CIN. This bivariate analysis revealed that baseline hemoglobin level (P=0.0371) and serum urea level (P= 0.0075) were significantly associated with cisplatin nephrotoxicity (Table IV). A positive association was also detected between CIN and a diagnosis of cervical cancer (P= 0.031). Examination of the possible impact of the others baseline variables on the occurrence of Cisplatin induced Nephrotoxicity revealed no significant association (Table IV).

4.7.2 Cox regression analysis

Univariate and multivariable Cox regression analysis were used to determine which factors were associated with development of CIN following the first course of chemotherapy (Table V). The demographic and baseline clinical characteristics used as covariates were included in the model. Baseline serum urea level greater than 5 mmol/l (HR: 8.51, 95% CI: 2.51 - 28.85; P= 0.001) and esophageal cancer (HR: 3.98, 95% CI: 1.11-17.24; P= 0.033) both emerged as the only parameters independently associated with CIN.

Table IV: Bivariate analysis exploring possible associations between baseline variables and Cisplatin Induced Nephrotoxicity.

Characteristics	Overall	Cisplatin Induced Nephrotoxicity		P value
		Yes (n = 16)	No (n = 53)	
Age; Mean n (%)				
<60	54	11(20.37)	43(79.63)	0.293
>60	15	5(33.33)	10(66.67)	
Gender ; n (%)				
Male	27	9(33.33)	18(66.67)	0.109
Female	42	7(16.67)	35(83.33)	
Weight ;mean ± SD (kg)	62.13 ± 11	59.5 ±10.66	62.93 ± 11.07	0.277
Height; mean ± SD (cm)	164.53 ± 7.9	166.12 ± 6.8	164.05 ± 8.2	0.366
BSA ; mean ± SD (m 2)	1.67± 0.17	1.62 ± 0.18	1.69 ± 0.17	0.2122
Type of cancer n (%)				
Cervical				
no	40	13 (32.50)	27 (67.50)	**0.031**
yes	29	3	26	
Nasopharyngeal				
no	62	1524.19	47 (75.81)	0.556
yes	7	1(14.29)	6 (85.71)	
Esophageal				
no	58	11(18.97)	47(81.03)	0.056
yes	11	5 (45.45)	6 (54.55)	
Oral				
no	63	14 (22.22)	49 (77.78)	0.538
yes	6	2(33.33)	4 (66.67)	
Hypopharyngeal & laryngeal				
no	63	13	50(79.37)	0.103
yes	6	3 (50)	3 (50)	
Stomach				
no	65	15(23.08)	50 76.92	0.93
yes	4	1(25)	4(75)	
Sarcoma				
no	67	15(22.39)	52(77.61)	0.362
yes	2	1(50)	1(50)	

Table IV: continuation

Gastroesophageal				
no	68	16(23.53)	52 (76.47)	0.58
yes	1	0(00)	1(100)	
Metastatic at presentation n (%)				
no	61	15(24.59)	46(75.41)	0.446
yes	8	1(12.50)	7(87.50)	
Kidney status				
Hb ; mean ± SD	12.82 ± 1.76	13.62 ± 1.87	12.58 ± 1.66	**0.0371**
BUN; mean ± SD	3.6 ± 1.07	4.21 ± 1.41	3.41 ± 0.47	**0.0075**
SCr (mg/dl) ; median(range)	0.71(0.40;1.39)	0.75 (0.40;1.39)	0.69 (0.45;1.03)	0.3894
CrCl; mean ± SD	104.14 ±27.93	96.4 ± 27.38	106.48 ± 27.92	0.4512
Sodium (Na) ; mean ± SD	137.04 ± 2.94	136.18± 2.18	137.3 ± 3.11	0.1871
Potassium (K) ; mean ± SD	4.28 ± 0.42	4.1 ± 0.51	4.34 ± 0.38	0.0523
Chloride (Cl); mean ± SD	100.46 ± 3.59	99.25 ± 2.32	100.83 ± 3.83	0.1238
Albumin (Alb); mean ± SD	40.22 ± 4.47	40.26 ± 4.66	40.20 ± 4.46	0.9612
Hypertension (n (%)				
no	66	15	51	0.67
yes	3	1	2	
Smoking n (%)				
no	62	14 (22.58)	48 (77.42)	0.722
yes	7	2 (28.57)	5 (71.43)	
Alcohol consumption n (%)				
no		13 (21.31)	48 (78.69)	0.308
yes		3 (37.50)	5 (62.50)	
Chemotherapy regimens n (%)				
cisplatin only	40	10 (25)	30(75)	0.675
combination regimen	29	6(20.29)	23 (79.31)	
Cisplatin dose (mg) ;mean±SD	122.97±14.40	121.03 ± 13.17	123.56 ± 14.81	0.541
Use of paclitaxel				
no	47	12 (25.53)	35(74.47)	0.50
yes	22	4(18.18)	18(81.82)	
Use of 5FU				
no	66	15 (22.73)	51(77.27)	0.67
yes	3	1 (33.33)	2 (66.67)	
Concurrent radiation n (%)				
no	40	10 (25)	30(75)	0.675
yes	29	6(20.29)	23 (79.31)	
Trial center n (%)				
Kenyatta National hospital	36	8(22.22)	28(77.78)	0.843
Texas cancer Centre	33	8(24.24)	25(75.76)	

Table V: Cox regression analysis of potential risk factors (demographic and clinical characteristics) for the development of and Cisplatin Induced Nephrotoxicity.

Covariates	Univariate model		Multivariate model	
	Hazard Ratio (95% CI)	P-Value	Hazard Ratio (95% CI)	P - Value
Age	1.009(0.97 - 1.05)	0.633		
<60	1.00			
>60	1.72(0.59 - 4.95)	0.314		
sex				
Male	0.48(0.18 - 1.30)	0.150	0.66(0.23 - 1.87)	0.445
Female				
Weight	0.97(0.93-1.02)	0.331		
Height				
BSA	0.23(0.01 - 3.49)	0.290	0.24(0.017-3.37)	0.290
Type of cancer	1.24(1.00 - 1.54)	**0.048**	1.21 (0.95 - 1.55)	0.120
Cervical				
no	1.00			
yes	0.30(0.087 - 1.07)	0.065	0.44(0.11 - 1.16)	0.222
Nasopharyngeal				
no	1.00			
yes	0.59(0.077 - 4.46)	0.610		
Esophageal				
no	1.00			
yes	2.53(0.87 - 7.26)	0.087	3.98(1.11-17.24)	**0.033**
Oral				
no	1.00			
yes	1.54(0.35- 6.78)	0.567		
Hypopharyngeal & laryngeal				
no	1.00			
yes	2.49(0.70 - 8.74)	0.154	1.20(0.30 - 4.74)	0.794
yes	1.14(0.15 - 8.70)	0.893		
Sarcoma				
no	1.00			
yes	2.15(0.28 - 16.36)	0.456		
Gastroesophageal				
no	1.00			
yes	1.24 e-14	1.000		

Table V: Continuation

IV Mg preloading	0.34(0.11 - 1.06)	0.065	0.27(086 - 0.89	**0.031**
Metastatic at presentation				
No	1.00			
Yes	0.50(0.06 - 3.80)	0.506		
kidney status				
Hb	1.32(1.00 - 1.74)	**0.044**	3.55(0.78 - 16.19)	0.101
BUN	1.77 (1.15 - 2.73)	**0.009**		
BUN <5	1.00			
BUN> 5	5.96(2.14 - 16.59)	**0.001**	8.51(2.51 - 28.85)	**0.001**
SCr	1.025 (0.99 - 1.05)	0.103	1.007(0.97 - 1.04)	0.647
CrCl	0.98 (0.96 -1.00)	0.216	0.99(0.97 - 1.01)	0.631
Sodium (Na)	0.90(0.77 - 1.05)	0.204	0.93(0.78 - 1.11)	0.445
Potassium (K)	0.31 (0.10-0.98)	**0.047**	0.49(0.17 - 1.42)	0.193
Chloride (Cl)	0.91 (0.80 - 1.03)	0.142	0.92(0.80 - 1.06)	0.261
Albumin (Alb)	1.002(0.89 - 1.11)	0.0971	0.97(0.87 - 1.08)	0.588
Ethnicity	1.03(0.81 - 1.31)	0.795		
Occupation	1.05(0.81 - 1.37)	0.676		
Comorbidity n (%)	1.13(0.68 - 1.88)	0.611		
Smoking status n (%)				
No	1.00			
Yes	1.28 (0.29-5.63)	0.743		
Alcohol consumption status				
no	1.00			
Yes	2.03(0.57 - 7.14)	0.270	1.71(0.47- 6.17)	0.411
Cisplatin combination regimen				
no	1.00			
Yes	0.8(0.28 - 2.22)	0.683		
Cisplatin dose	0.99(0.95-1.02)	0.624		
Combination drugs	0.96(0.52 - 1.76)	0.906		
Concurrent radiation				
No	1.00			
Yes	0 .81 (0.29 - 2.22)	0.68		
Trial center	1.08 (0.40 - 2.88)	0.875		

CHAPTER 5: DISCUSSION, CONCLUSION AND RECOMMENDATIONS

5.1 Discussion of results

In the current study, we analyzed the preventive effect of intravenous Mg preloading supplementation on cisplatin-induced nephrotoxicity (CIN) in cancer patients who received cisplatin dose of at least 60 mg/m^2. We found that 12.12 % (4/33) of patients who received intravenous magnesium preloading supplementation developed CIN whereas 33.33 % (12/ 36) was found among those who did not receive IV magnesium preloading supplementation. As a result, intravenous preloading magnesium supplementation significantly reduced the incidence rate of nephrotoxicity (P=0.037), consistent with previous observations [28; 29,65, 92;97].

The dosage and method of magnesium sulfate supplementation therapy has varied widely in previous studies, ranging from 8 to 60 mEq (28; 30; 102;103; 104) and including administration before and/or after cisplatin. To our knowledge, this is the first randomized controlled trial to report the protective effect of magnesium supplementation given at the dose of 8 mEq and just before the administration of cisplatin. the dose, method and findings in this study are consistent with those of a retrospective study design by Yoshida et al [97], where the magnesium group received 8 mEq before administration of cisplatin and saw a significant decrease in the incidence of cisplatin nephrotoxicity (P< 0001). These findings support the NCCN Clinical Practice Guidelines in Oncology which recommends that 8 mEq Mg preloading before cisplatin should be included in the treatment protocol as prophylaxis of CIN [30]

As previously reported [65] CIN generally manifests as an increased of serum creatinine level (SCr) and reduction in creatinine clearance (CrCl) due to renal tubular dysfunction. Intravenous magnesium preloading, when evaluated in the current study on the basis of the SCr level and the CrCl, was shown to be associated with the decrease of renal toxicity induced by cisplatin. The median maximum change of SCr level in the Mg preloading group 0.10 mg/dl (range: -0.090, 1.761) was significantly lower than that in the non-Mg preloading group 0.19 mg/dl (range: -0.147, 1.86) (P=0.0037) suggesting that magnesium supplementation therapy limited the elevation of serum creatinine level induced by cisplatin. Our findings are consistent with the result of a prospective non randomized study by Oka et al [95].Patients who received 8 mEq before cisplatin administration showed no significant difference between pre and post treatment SCr levels [(p = 0.118).

As there is nonlinear relationship between SCr and CrCl [105], it was therefore important that renal function be evaluated on the basis of CrCl as calculated by the Cockcroft & Gault equation. The median maximum change in CrCl from baseline was therefore compared between two arms. The results demonstrated a significantly larger decrease in CrCl from baseline in the non-magnesium supplementation group compared to the magnesium supplementation group (P = 0.012), suggesting that magnesium preloading supplementation therapy limited renal function decline induced by cisplatin.

Although the current study could not demonstrate significance of the difference in magnesium sulfate concentration between the two groups, Evans et al [102], in a randomized study to determine whether routine intravenous magnesium supplements are necessary in patients receiving cisplatin chemotherapy with continuous infusion of 5-fluorouracil revealed that the mean serum magnesium level was significantly lower in the patient who do not receive magnesium compared to those who received intravenous magnesium supplementation with each cycle($P < 0.05$). This was consistent with previous studies who reported that the majority of cisplatin-treated patients develop polyuria, hypomagnesaemia and renal Mg^{2+}, Ca^{2+}, Na^{+} and K^{+} wasting [106-109]. These defects likely arise from impaired functionality of the renal proximal convoluted Tubule (PCT) and distal convoluted tubule (DCT) segments (k The relevance between hypomagnesemia and cisplatin-induced nephrotoxicity remain to be completely elucidated. The results of an experimental rat model study suggested that hypomagnesemia could cause dehydration and up-regulation of the Organic Cation Transporter 2 (OCT2) and the Copper Transporter 1 (CTR1), both identified to contribute to the uptake of cisplatin, and thereby enhancing renal accumulation of cisplatin and then deterioration of AKI [66] This hypothesis seems valid as it is well known that the nephrotoxic effect of cisplatin is proportional to the amount of drug accumulated [22; 31;110].

The Kaplan Meier survival curves by treatment was built to compare the pattern of survival rates over time from the day of cisplatin administration to the end of the follow up period between two group. The pattern of survival between the two groups was found highly significant using the log rank test (P= 0.0420). As we ensured in this study comparability of the patient and disease characteristics of the two treatment groups, this statistical significance found between the 2

survival curvesindicates the beneficial effect of magnesium supplementation in extendingthe time to development of CIN, as report by a previous study [97]. In addition, the survival curves in both treatment arms revealed that CIN mostly occurs within the 10 day following cisplatin administration, in agreement with prior studies[24].

The evolution of calculated CrCl from baseline to day 17 revealed a global decrease of CrCl from baseline to day 10 for both groups. Both trend curves hit an inflection point on day 10 and increased toward the end of the follow up period. This suggested an initial decline in renal function, and an average length of recovery time less than 2 weeks in both treatment arms. This is consistent with the retrospective study by Hyung et [74]which examined the pattern of nephrotoxicity in 552 patients who received cisplatin combination chemotherapy study and found that most patients had an average recovering time of 2 weeks.

To assess the potential risk factors for CIN, univariate and multivariate cox regression analyses were performed. Consistent with previous results[65] esophageal cancer was found to be associated with an increased risk for cisplatin nephrotoxicity (HR = 3.98, 95% CI: 1.11-17.24; P= 0.033).Evidence supporting our finding are very scarce. Yashiro et al demonstrated that a difference in dosage or in the combination of chemotherapeutic agents could not account for the difference in nephrotoxicity among the malignancies. Further research is therefore advocated to understand the mechanism of renal toxicity apparent selectively in patients with esophageal cancer.

With regard to the laboratory variables, unlike number of large studies[65; 67;111],we found BUN greater than 5 mmol/l to be a predictor factor of CIN (HR = 8.51, 95% CI: 2.51 - 28.85; P= 0.001).

To understand this result we compared the median baseline SCr in both BUN subgroups. As expected, the group with the BUN > 5 had likewise an elevated baseline SCr compared to the group with BUN < 5 (0.69 mg/dl vs 0.91 mg/dl). In addition, the difference in mean SCr between the BUN subgroups was statistically significant P = 0.0065, suggesting that BUN was directly proportionate to SCr. As both BUN and SCr vary inversely with the glomerular filtration rate (GFR) [112], the elevated BUN could reflect a decrease in GFR which increases the risk of

developing CIN [113]. The result of BUN as a predictive factor of CIN therefore could be interpreted in this context.

Alternative explanation for high BUN > as predictor factor of CIN could be found from the study of Steward and al. [114] They identified BUN level as a factor that correlated positively with kidney cortex platinum concentrations. Meanwhile they also revealed that the hydration volume does not affect kidney cortex platinum concentrations. They concluded that any effect of the hydration volume is not mediated by reduction of kidney cortex platinum concentrations. Their findings therefore suggest that patient at high level of baseline serum urea receiving high dose of platinum drugs are less likely to benefit from the protective effect of hydration thereby are at high risk of developing cisplatin induced nephrotoxicity.

It was reported by de Jongh et al. [67] that smoking may be a risk factor for CIN. However, no association was found between smoking and nephrotoxicity in the present study.This is likely because of the small number of smokers (7/69) in the study sample.

Admitting that further study is warranted to determine the mechanism of hypomagnesemia induced decline in kidney function in patient treated with Cisplatin, our results demonstrate that supplementation of magnesium in cancer patients receiving cisplatin appears to be beneficial with reduced renal tubular damage.

Limitations of the present study include the fact that renal function was assessed only after the first course of chemotherapy due to time constraints while damage by cisplatin may be cumulative, and the assessment of renal function in the subsequent cycles would have been important.

In addition, Magnesium status was not assessed prior chemotherapy and at the end of the follow up period. The studies therefore was not able to associate hypomagnesemia and cisplatin induced nephrotoxicity.

5.2 Conclusion

Intravenous preloading Magnesium supplementation administered at a dose of 8 mEq before cisplatin administration was significantly associated with both a reduced frequency and reduced severity of renal toxicity in cancer patients treated with cisplatin based regimen. Our study demonstrates that the protective effect of magnesium supplementation can be seen by limitation of serum creatinine level increased and reduction of the slope of decline in creatinine clearance. Magnesium supplementation therefore appears to have a protective mechanism that limits renal tubular injury induced by cisplatin. Our findings also confirm that the dose and method of supplementation are quite appropriate and therefore support the recommendation of the NCCN guideline in oncology.

This treatment added to the current strategies may be quite beneficial for reducing the nephrotoxicity profile of cisplatin in patients treated with cisplatin based regimen, resulting in increased chemotherapeutic efficacy of cisplatin in clinical practice. This in turn will have a very highly favorable impact on the treatment of solid tumors as it is well known that Cisplatin still remains the drug of choice for a number of solid tumors.

Finally, the improvement of the use of cisplatin in clinical practice will be more beneficial for patients in developing countries as cisplatin is cost-effective compared to other platinum derivatives

The data from this study constitute strong and direct evidence in support of the application of intravenous preloading magnesium supplementation at the dose of 8 mEq before administration of cisplatin as a preventive measure of cisplatin-induced nephrotoxicity.It is therefore recommended that magnesium be routinely supplemented during cisplatin based treatment for cancer patient.

5.3 Recommendations

Recommendations for practice

As part of preventive strategy against cisplatin induced nephrotoxicity, the following should be routinely supplemented during each cycle of Cisplatin based regimen:

- 8 mEq of intravenous magnesium sulfate administered before administration of cisplatin.
- 20 mmol of potassium chloride administered before and after administration of cisplatin (after serum potassium has been determined and confirmed to be within normal range).
- intravenous post hydration with one-liter normal saline
- antiemetic prophylaxis for 4-days minimum following cisplatin administration to prevent delayed vomiting thus dehydration in patients treated with cisplatin based regimen

A routine laboratory workup of bone marrow, liver and renal function tests should also be ordered less than 2-day prior chemotherapy specifically cisplatin based regimen.

Recommendations for future research

Based on the difference in the prevalence of CIN in our control group (33%) with the previously reported prevalence of CIN in KNH (89%). we strongly recommend a randomized controlled trial comparing the hydration protocol used as the control in this study to the routinely used hydration protocol at KNH.

Regarding the high risk of cisplatin nephrotoxicity associated with esophageal cancer, further research is advocated to understand the mechanism of renal toxicity apparent selectively in patients with esophageal cancer.

Some patients treated with Mg preloading regimen still developed CIN, supporting the need for studies aimed at identifying complete preventive measures against CIN.

Finally, we strongly recommend larger studies that can allow for adequate sub-group analysis for all the variables identified as predictor factors of CIN, in order to assess for any differences in the treatment effect within these subpopulations.

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APPENDIX 1: Research Participant Information Statement/Research Participant Consent form

Research Study Title:

EVALUATION OF INTRAVENOUS PRELOADING MAGNESIUM SUPPLEMENTATION AS A PREVENTIVE MEASURE OF CISPLATIN INDUCED NEPHROTOXICITY

KNH/UoN ERC Approval Number: KNH- ERC/A/245

This is a clinical trial, a type of research study. Your study doctor will explain the clinical trial to you. Clinical trials include only people who choose to take part. Please take your time to make your decision about taking part. You may discuss your decision with your friends and family. You can also discuss it with your doctor.

You are being asked to take part in this study because you have cancer and as part of your management, you are going to receive treatment that includes the drug cisplatin.

Why is this study being done?

The purpose of this study is to compare the effects of a new protocol of hydration containing magnesium sulfate with a standard hydration protocol whichdoes not contain magnesium sulfate. Magnesium sulfate is given as a supplement to correct or prevent magnesium deficiency which is a side effect of cisplatin administration.

This study is being done to find out if giving magnesium supplementation before the administration of cisplatin can reduce the occurrence and the degree of nephrotoxicity (damage to the kidneys) in patients with cancer who are undergoing cisplatin-based chemotherapy.In this study, in addition to you cancer chemotherapy, you will get either the hydration regimen with magnesium sulfate or the hydration regimen without magnesium sulfate to prevent nephrotoxicity. You will not get both.

How many people will take part in the study?

About 62 people will take part in this study

Who is carrying out the study?

Institution:Department of Pharmaceutics and Pharmacy Practice, School of Pharmacy; University of Nairobi. P.O BOX 30197-0400 Nairobi.**Investigator**: Dr Marius Beniet Youan Bi, Pharm.D. , Master Student in Clinical Pharmacy, University of Nairobi.Contact 0719641397; email:mariusdebeniet@gmail.com

Supervisors: **Dr. David G. Nyamu**: Department of Pharmaceutics and Pharmacy Practice, School of Pharmacy

Dr Eric M. Guantai: Department of Pharmacologyand Pharmacognosy, School of Pharmacy
Dr IreneWeru: Cancer Treatment Center, Kenyatta National Hospital.

Ethical approval: The study has been approved by the Ethics and Research Committee of Kenyatta National Hospital/University of Nairobi P.O BOX 20723-00100, Nairobi.Tel.no. 2726300/2716450.Ext 44102.

What will happen if I take part in this research study?

Before you begin the study, you will need to have the following exams, tests or procedures to find out if you can be in the study. These exams, tests or procedures are part of regular cancer care and may be done even if you do not join the study. If you have had some of them recently, they may not need to be repeated. This will be decided by your study doctor.

1. History and physical exam and an assessment of your ability to carry out activities of daily living (which will include questions such as whether you are able to feed, bathe, and dress yourself)
2. Blood tests to measure the adequate functionof liver, kidney and bone marrow.
3. You will be asked to give information about any other medications that you may be taking.

During the study

If the exams, tests and procedures show that you can be in the study, and you choose to take part, you will be randomly assigned into one of two study groups.Random assignmentmeans that you will have an equal chance of being placed in either of the groups.Neither you nor your study doctor will be ablechoose the group you will be in.
The patients inone two study groups the hydration regimen with magnesium sulfate, while the other group will receive the hydration regimen without magnesium sulfate. The treatments that both groups will receiveare described in detail below.

After randomization.
On day 1
Before the treatment both group will have a blood test for serum creatinine analysis. This is to assess the level of kidney function before receiving treatment.

If you are in group 1 (often called “Intervention Arm "),

- **On day one** you will receive (granisetron) 3 mg, and dexamethasone (9.9 mg) mixed together with 50 mL of Normal Saline and administered by 15-minute i.v.infusion as a single dose to prevent emesis (vomiting). Then a cytotoxic agent will be administered. Potassium chloride (KCl) 20mmol/L plus Magnesium sulfate $MgSO_4$ 8 mEq (1g) diluted in 1litre of Normal Saline will follow by IV infusion over 2 hours. Immediately before the administration of cisplatin, 200ml of 20% mannitol will be administered by IV infusion over 30 minutes. Then Cisplatin diluted in 500 ml of 0.9% NaCl solution (N/saline) will be administered by IV infusion over 60 minutes. After the administration of cisplatin you will receive by IV infusion over 2 hours, one liter of Normal Saline + 20mmol KCl.
- **On day 2** you will receive an additional dose of anti-emetics for control of delay emesis (Dexamethasone 4mg tablet orally twice in a day plus Ondansetron 8mg tablet orally twice in a day) and 500ml of normal saline IV infusion over 1hour.
- **On day 3 to 5 you will continue** Dexamethasone 4mg tablet orally twice in a day plus Ondansetron 8mg tablet orally twice in a day.

If you are in group 2(often called “control Arm "),
You will receive the same treatment as the group1 (magnesium preloading group), except the pre-hydration solution which will contain 20 mmol of Potassium chloride (10ml of KCl 15%) but will NOT contain magnesium sulfate.
After the start of treatment, you will need the following tests and procedures

<u>On day 2, day 6, day 10 and day 17 after administration of cisplatin</u>

- You will be asked to give information about any medications that you may betaking
- You will be asked about any side effects that you may be experiencing
- Vitals signs will be monitored to detect any abnormality.
- Blood tests to measure creatinine level and electrolytes will be performed to evaluate renal function

How long will I be in the study?

The treatment will be administered over 5 days for both group 1 andgroup 2. The study doctor will ask you to visit the office for follow-up examination and to collect the blood for creatinine analysis at day 2, day 6, day 10 and day 17 after the start of treatment.

Can I stop being in the study?

Yes. You can decide to stop at any time. Tell the study doctor if you are thinking about stopping or decide to stop. He or she will tell you how to stop safely.
The study doctor may stop you from taking part in this study at any time if he/she believes it is in your best interest, if you do not follow the study rules, or if the study is stopped.

What side effects or risks can I expect from being in the study?

You may have side effects while on the study. Everyone taking part in the study will be watched carefully for any side effects. However, researchers don't know all the side effects that may happen. Side effects may be mild or serious. Your health care team may give you medicines to help lessen side effects. Magnesium side effects occur rarely at the dose that areadministered topatients who undergoes cisplatin chemotherapy. This is because hypomagnesaemia (low levels of magnesium in the blood) is a frequent complication to chemotherapy with cisplatin affecting up to 90% of patients who do not receive prophylactic magnesium supplementation [3].

Risks and side effects related to the pre-hydration solution containing magnesium sulfate plus potassium chloride or potassium chloride without magnesium sulfate.

- **Rare and minor and include**:

1- Sweating
2- Flushing
3- Dizziness

- **Rare, but serious**
- difficulty breathing,
- low pulse rate,
- bradycardia (abnormally low heart rate)
- hypotension (abnormally low blood pressure)
- depressed reflexes

For more information about risks and side effects, ask your study doctor.

Are there benefits to taking part in the study?

Taking part in this study may or may not make your health better. There is proof that preloading magnesium supplementation can decrease nephrotoxicity (kidney damage) but strong evidence in our setting isnot available yet. We do know that the information from this study will help researchers learn more about magnesium preloading supplementation as an additional treatment for preventing nephrotoxicity in patient undergoing cisplatin based chemotherapy for cancer. This information could help doctor to prevent nephrotoxicity.

What other choices do I have if I do not take part in this study?

Your other choice will be to get treatment or care for cancer without being in thestudy.
Talk to your study doctor about your choices before you decide if you will take part in this study.

Will my medical information be kept private?

Information will be kept in a password-protected database. We will do our best to make sure that the personal information in your medical record will be kept private. Your personal information may be given out only if required and authorized by the Kenyatta National Hospital/University of Nairobi Ethics and Research Committee (KNH/UON-ERC). If information from this study is published or presented at scientific meetings, your name and other personal information will not be used.

What are the costs of taking part in this study?

You and or your health insurance company will need to pay for some of the costs of treating your cancer in this study. Taking part in this study may or may not cost you or your insurance company more than the cost of getting regular cancer treatment.

The research will supply for magnesium supplementation and perform additional laboratory test at no charge while you take part in this study. You will not be paid for taking part in this study but transport reimbursement will be considered for follow up session at Days 6 and Day 10.

What are my rights if I take part in this study?

Taking part in this study is your choice. You may choose either to take part or not to take part in the study. If you decide to take part in this study, you may leave the study at any time. No matter what decision you make, there will be no penalty to you and you will not lose any of your regular benefits. Leaving the study will not affect your medical care. You can still get your medical care from our institution. A copy of the signedInformed Consent form will be given to you

What can I do if I have a complaint or a concern?

Any concerns or complaints about the conduct of this study should be directed to:

KNH/UoN-ERCSecretary
University of Nairobi, School of Pharmacy
P.O BOX 20723-00100, Nairobi.
Tel.no. 2726300/2716450.Ext 44102
Email: uonknh-erc@uonbi.ca.ke.

Any complaint will be investigated promptly and you will be informed of the outcome

This information sheet is for you to keep.

Research Participant Consent form

Research Study Title	**EVALUATION OF INTRAVENOUS PRELOADING MAGNESIUM SUPPLEMENTATION AS A PREVENTIVE MEASURE OF CISPLATIN INDUCED NEPHROTOXICITY**
KNH/UoN-ERC Approval Number :	
Researcher's Name :	Dr Marius Beniet Youan Bi
Researcher's Relationship to UoN/KNH:	Postgraduate Student in Clinical Pharmacy. University of Nairobi.

Participant Consent

I have read the above consent form and understood it. The nature of the study has been explained to me. I voluntarily agree to participate in the study.

_____ _____ ____ / _____ / ___

Signature of Participant **Name (***First name and Surname)* **Year Month Day**

Address: _____ **Telephone:** _____

Investigator's statement

I, the undersigned, have explained to the participant the procedures to be followed in the study and the risks and benefits involved.

_____ _____ ____ / _____ / ___

Signature of Person Conducting **Name (***First name and Surname)* **Year Month Day**

Consent Discussion

_____ _____ ____ / _____ / ___

Signature of Investigator **Name (***First name and Surname)* **Year Month Day**

Department of Pharmaceutics and Pharmacy Practice, School of Pharmacy, University of Nairobi.
P.O BOX 30197-0400 Nairobi. Tel: 0719641397; email: mariusdebeniet@gmail.com

_____ _____ _____/ _____ / ___

Signature of Witness **Name (***First name and Surname)* **Year Month Day**

Relationship of Witness to Research Participant/investigator: _____

UTANGULIZI 1: TAARIFA YA MSHIRIKI UTAFITI/FOMU YAIDHINI ARIFU

MADA YA UTAFITI: KNH-ERC/A/ 245

NAMBARI YAIDHINISHO:KNH/UON ERC: ……..

Hili ni jaribio la kikliniki na aina ya uchunguzi wa kiutafiti. Daktari wako wa utafiti atakueleza kuhusu jaribio la kikliniki. Jaribio hili hujumuisha tu wale watu wanaochagua kushiriki. Tafadhali tafakari kuhusu kushiriki kwako katika utafiti. Waweza kujadili na marafiki, familia yako au na daktari wako wa kibinafsi kuhusu uamuzi wako. Unaombwa kushiriki utafiti kwa maana unaugua saratani, na kama mojawapo ya matibabu yako, utapewa tiba ya saratani yaani kemotherapia iliyo na chembechembe za platini.

KWA NINI UTAFITI UNAFANYWA?

Madhumuni ya utafiti ni kulinganisha athari (iwapo ni kubwa au chache ama sawia) za mfumo mpya wa uvuvio(hydration) wenye madini ya magnesia na mfumo kawaida wa uvuvio usio na magnesia. Magnesia hutolewa kama kiambatisho kurekebisha au kuzuia ukosefu wa magnesia ambao ni athari upande ya upeanaji wa kemotherapia iliyo na Platini. Uchunguzi unafanywa kubaini ikiwa upeanaji wa kiambatisho cha magnesia kabla ya kemotherapia yenye platini huweza kupunguza matukio na kiwango cha sumu ya figo (nephrotoxicity) kwa wagonjwa wenye saratani katika kemotherapia iliyo na Platini. Katika utafiti huu,utapata pamoja na kemotherapia, utaratibu wa matibabu ya uvuvio(hydration) yaliyo na kidini cha magnesia au yasiyo na kidini cha magnesia ili kuzuia sumu ya figo(nephrotoxicity). Hutapata yote mawili.

NI WATU WANGAPI WATASHIRIKI UTAFITI?

Takriban watu sitini na wawili (62)watashiriki katika utafiti huu.

NANI ANAENDESHA UTAFITI?

Chuo: Idara Ya Famasia Na Mazoezi Ya Ufamasia, Kitivo Cha Ufamasia; Chuo Kikuu Cha Nairobi. S.L.P 30197-0400 Nairobi.

Mtafiti: Dkt Marius Beniet Youan Bi, Pharm.D. , Mwanafunzi Wa Uzamifu Katika Matibabu, Chuo Kikuu Cha Nairobi.Simu: 0719641397; Barua Pepe:mariusdebeniet@gmail.com

Msimamizi: **Dkt. David G. Nyamu**: Idara Ya Famasia Na Mazoezi Ya Ufamasia. Kitivo Cha Ufamasia

Dkt. Eric M. Guantai: Idara Ya Famakolojia Na Ufamaknosia ., Kitivo Cha Ufamasia

Dkt. Ireneweru: Kituo Cha Matibabu Ya Saratani, Hospitali Kuu Ya Kenyatta

IDHINISHO LA MAADILI:

Utafiti umeidhinishwa na Kamati ya Maadili na Utafiti ya Hospitali Kuu ya Kenyatta pamoja na Chuo Kikuu cha Nairobi.S.L.P 20723-00100, Nairobi.SIMU: 2726300/2716450.Ext 44102.

MATUKIO NI YEPI IWAPO NITASHIRIKI UTAFITI?

Kabla ya utafiti, unahitaji kuwa na yafuatayo: Uchunguzi,majaribio na taratibu ili kujua iwapo unafaa kushiriki utafiti. Uchunguzi,majaribio na taratibu ni huduma za kawaida za saratani na huweza zikafanywa hata kama hutajiunga kushiriki utafiti. Iwapo umekuwa na huduma hizi hivi karibuni basi si lazima zirudiwe. Hili litategemea na daktari wako wa utafiti.

1. Historia na uchunguzi wa kimwili, na tathmini ya uwezo wako kushiriki shughuli za kila siku(maswala kama; iwapo unaweza kujilisha,kuoga na kuvaa nguo).
2. Uchunguzi wa damu ili kupima nguvu kazi ya ini, figo na ombwe la mifupa.
3. Utaulizwa kutoa habari kuhusu matibabu yoyote uliyo nayo kea sasa.

WAKATI WA UTAFITI

Katika uchunguzi,majaribio na taratibu, onyesha kwamba una ari ya kushiriki na uamue kuhusika. Utanasibishwa katika makundi. Kunasibishwa ina maana kuwa utawekwa katika kundi kupitia bahati nasibu. Programu ya kompyuta itatumiwa kukutia katika mojawapo ya vikundi. Sio wewe wala daktari mtaamua kundi lako. Utakuwa na nafasi sawa ya kutiwa baina ya vikundi baada ya kunasibishwa. Siku ya kwanza kabla ya matibabu, makundi yote mawili yatafanyiwa uchunguzi wa damu dhidi ya sumu ya misuli inayopatikana kwa damu (serum creatinine).

UKIWAKUNDI LA 1 (Kundi Zuizi):

- **Siku ya 1:** Utapewa kinga ya kutapika (granisetron 3mg) na kinga ya nyenge(dexamethasone 9.9mg) zikichanganywa na chumvi(Normal Saline 500Ml). Hii itapeanwa kwa mmiminiko wa dakika 15 kama kipimo kimoja kuzuia kutapika. Kisha ejenti za kuharibu seli(cytotoxics) zitapeanwa. Potashi (KCL 20mmol/L) na Magnesia(MgS04 mEq 1g) zikichanganywa na chumvi(N/Saline 0.9 NaCl) zitafuatilizwa kama mmiminiko kwa zaidi ya masaa mawili. PUnde tu kabla ya kupeana chembe za platini, asilimia ishirini ya (200mL ya 20% mannitol) itamiminiwa kwa zaidi ya dakika thelathini. Kisha mchanganyiko wa chembe za platini na chumvi (500mL ya 0.9% NaCl) utamiminiwa kwa zaidi ya dakika sitini. Mwisho utapewa mmiminiko wa chumvi (1L ya 0.9% NaCl) pamoja na Potashi(200mmol KCl).

- **Siku ya 2:** Utapewa kipimo ziada cha dawa za kuzuia kutapika ili kudhibiti kuchelewa kwa tapiko.(Tembe za Dexamethasone kumezwa mara mbili kwa siku,

tembe za Odansetron kumezwa mara mbili kwa siku. Kisha mmiminiko wa chumvi (500mL 0.9% ya NaCl) kwa zaidi ya lisaa limoja.

- **Siku ya 3 - 5**: Utaendelea na tembe za Dexamethasone kumezwa mara mbili kwa siku na tembe za Odansetron kumezwa mara mbili kwa siku.

UKIWA KUNDI LA 2 (KUNDI DHIBITI)

Utapata matibabu sawa na kundi la kwanza,

Utapokea matibabu sawa na kundi la kwanza (magnesia preloading kikundi), ila kabla ya taratibu ufumbuzi ambayo yana 20 mmol ya Potassium chloride (10ml ya KCl 15%) lakini sio vyenye magnesium sulfate.

Baada ya mwanzo wa matibabu, utahitajika uwe na majaribio na taratibu zifuatazo:
Siku ya 2, 6, 10 na 17 baada ya upeanaji wa dawa za chembe za platini (cisplatin)

- Utaulizwa kutoa taarifa kuhusu matibabu yoyote uliyo nayo kwa sasa.
- Utaulizwa kuhusu athari upande zozote.
- Dalili za mapigo ya moyo,nyuzi joto mwilini,mkimbio wa damu na kiwango cha kupumua zitaangaliwa kutambua ubatilifu wowote.
- Uchunguzi wa damu kupima kiwango cha sumu itokayo kwa misuli(creatinine) na vimelea vya nishati mwilini(electrolytes) ili kutathmini uwezo wa figo.

NI KWA MUDA UPI NITASHIRIKI UTAFITI?

Matibabu yatachukua siku sita. Wakati wa matibabu na baada ya matibabu ya kundi la 1 na la 2, daktari katika zoezi atakuuliza kutembelea ofisi kaa minajili ya jaribio la ufuatilizi na kuchukua damu ili kufanya uchunguzi wa sumu itokanayo na misuli au kretini. Hii itakuwa siku ya pili, sita, kumi na kisha kumi na saba baada ya mwanzo wa matibabu.

NAWEZA KUJIONDOA NA KUTOKA UTAFITI?

Naam, waweza kuamua kujionda wakati wowote. Mwambie daktari wa utafiti ikiwa una fikra za kujiondoa au kuacha. Atakueleza jinsi ya kujiondao kea usalama. Daktari wa utafiti yuwaweza kukusimamisha dhidi ya kushiriki wakati wowote iwapo anaamini ni kwa minajili ya manufaa yako, ikiwa huzingatii sharia au ikiwa utafiti umesimamishwa.

NI ATHARI UPANDE AU HATARI ZIPI NITARAJIE NIKISHIRIKI UTAFITI?

Waweza kupata athari upande ukishiriki utafiti huu. Kila mmoja anayeshiriki ataangaliwa kea makini iwapo kuna athari upande. Hata hivyo watafiti hawafahamu aina zote zaathari upande zinazoweza kuibuka. Athari upande zaweza kuwa ni kidogo au kubwa sana. Wahudumu wako wa afya wanaweza kupa madawa ili kupunguza athari upande. Hata hivyo athari upande za magnesia ni nadra kujitokeza katika vipimo vya (8meq) vikipewa wagonjwa wanaoelekezwa katika kemotherapia iliyo na chembe za platini. Sababu ni kuwa, haipomagnesia ni tatizo la kila mara kea tiba ya saratani iliyo na kidini cha platini. Hii huathiri asilimia tisin i(90%) ya wagonjwa wasiopea kizuizi cha magnesia ambatisho.

Hatari na athari upandezinazohusiana na mchanganyiko wa umiminiaji wenye magnesia na potash au ile isiyo na vimelea hivi vya magnesia ni kama ifuatavyo:

- **ATHARI UPANDE NADRA NA NYEPESI**
  - Jasho
  - Usafishaji
  - Kizunguzungu
- **ATHARI UPANDE/HATARI NADRA NA YENYE UZITO**
    - Pumu
    - Mapigo duni ya mishipa
    - Mapigo hafifu ya moyo
    - Mkimbio wa damu batili
    - Mmemenyuko duni

Kwa habari zaidi kuhusu hatari na athari upande, uliza daktari wako wa utafiti huu.

JE KUNA FAIDA ZA KUSHIRIKI UTAFITI?

Kushiriki kwako kwaweza kuboresha au kutoboresha hail yako ya afya. Thibitisho lipo kwamba upakiaji mapema wa vidonge vya magnesia huweza kupunguza tukio la sumu ya figo lakini uhalali huu haupo katika mfumo wetu. Data kutokana na huu itawezesha watafiti kujua mengi kuhusu vidonge vya kiambatisho cha magnesia kama tiba zidadi ya kinga dhidi ya sumu ya figo kea wagonjwa walio kwa matibabu ya kemotherapia iliyo na dawa za kiuongo cha chembe za Platini. Ujumbe huu ni muhimu sana kwa daktari ili kuzuia sumu ya figo.

NI CHAGUO LIPI LINGINE NINALO KWA KUSHIRIKI UTAFITI?

Uteuzi wako mwingine ni kama:

Kupata tiba ama huduma za saratani pasi na kushiriki zoezi la utafiti. Ongea na daktari wa utafiti kuhusu hiari zako kabla ya uamuzi wa kushiriki zoezi la utafiti.

JE TAARIFA KUHUSU AFYA YANGU ITAHIFADHIWA KWA SIRI?

Ujumbe huu utahifadhiwa katika hifadhidata iliyo na nambari ya siri(nywila). Tutahakikisha taarifa ya kibinafsi katika rekodi zako za matibabu imewekwa kwa kisiri. Hta hivyo hatuna hakikisho la siri kamilifu maana ujumbe wako wa kibinafsi waweza kuhitajika na Kamati ya Maadili na Utafiti ya Hospitali au Chuo Kikuu cha Nairobi. Iwapo taarifa ya utafiti huu imechapishwa au kuwasilishwa mbele ya makongamano ya kisayansi basi jina lako na ujumbe mwingine wa kibinafsi havitatumika.

GHARAMA NI ZIPI KATIKA UTAFITI?

Wewe na au kampuni yako ya bima ya afya mtagharamia baadhi ya malipo ya kemotherapia. Kushiriki utafiti huu hakutakugharimu wewe au kampuni ya yako ya bima ya afya zaidi ya malipo ya kawaida ya kutibu saratani.

Utafiti utashughulikia kuwepo kwa vidonge vya kiambatisho cha magnesia na kutekeleza uchunguzi zidadi wa maabara bila malipo. Aidha hutalipwa kwa kushiriki uatafiti huu.

HAKI ZANGU NI ZIPI IKIWA NITASHIRIKI KEA UTAFITI?

Kushiriki utafiti ni chaguo lako. Una uamuzi wa ama kushiriki au kutoshiriki. Ukiamua kushiriki pia waweza kujiondoa wakati wowote. Mbali na uamuzi unaochukua, hakutakuwa na adhabu kwako na hutapoteza mojawapo ya faida za kawaida.

Kujiondoa katika utafiti hautaathiri huduma zako za kimatibabu. Utaweza kupokea huduma zako za kimatibabu kutoka kwa chuo chetu.

NITAFANYA NINI IKIWA NINA MALALAMISHI?

.Malalamishi yoyote kuhusu mfumo wa utafiti huu yaelekezwe kupitia anwani ifuatayo:

KatibuKNH/UoN-ERC

Chuo kikuu cha Nairobi, Kitivo cha Famasia

S.L.P 20723-00100, Nairobi.

Simu: 2726300/2716450.Ext 44102

Barua pepe: uonknh-erc@uonbi.ca.ke.

Lalamishi lolote litachunguzwa kwa haraka na utaarifiwa kuhusu uamuzi.

Kartasi hii ya taarifa ni yako kuihifadhi/kuiweka.

FOMU YA IDHINI YA MSHIRIKI UTAFITI

MADA YA UTAFITI	[Titre du document]
Nambari ya Idhinisho: KNH/UoN-ERC.	KNH-ERC /A/ 245
Jina la mtafiti :	Daktari Marius Beniet Youan Bi
Uhusiano wa mtafiti na Chuo kikuu cha Nairobi au Hospitali kuu ya Kenyatta:	Mwanafunzi wa Uzamifu katika kozi ya Matibabu ya Famasia. Chuo kikuu cha Nairobi.

IDHINI YA MSHIRIKI UTAFITI.

Nimesoma na kuelewa fomu ya idhini iliopo hapo juu. Mfumo na sura ya utafiti imeelezwa kwangu ipasavyo. Kwa hivyo nakubali kujitolea na kushiriki utafiti kwa hiari bila kushurutishwa.

_____ _____ _____ / _____ / ____

Sahihi ya mshiriki utafiti **Jina (***la kwanza na la familia)* **mwakamwezisiku**

Anwani: _____Simu: _____

TAARIFA YA MCHUNGUZI

Mimi mwenye sahihi hapo chini, nimemweleza mshiriki katika utafiti kuhusu mbinu ambazo zitafuatwa katika uchunguzi na hata athari na manufaa husika.

_____ _____ / _____ / ____

Sahihi ya msimamizi wa mazungumzo ya idhini. **Jina (***la kwanza na la familia)***Mwaka Mwezi Siku**

_____ _____ /_____ / ____

Sahihi ya Mchunguzi **Jina (***la kwanza na la familia)* **Mwaka MwakaSiku**

Idara ya Famasia na Mazoezi ya Ufamasia, Kitivo Cha Famasia, Chuo Kikuu Cha Nairobi. S.L.P 30197-0400 Nairobi. Simu: 0719641397; Barua Pepe: mariusdebeniet@gmail.com

_____ _____ / _____ / _____

Sahihi Ya Shahidi **Jina(***la kwanza na la familia)* **MwakaMweziSiku**

Uhusiano wa shahidi na mshiriki utafiti au mchunguzi:_____

APPENDIX 2: PATIENT INFORMATION CARD.

PATIENT INFORMATION CARD

Research Study Title	**EVALUATION OF INTRAVENOUS PRELOADING MAGNESIUM SUPPLEMENTATION AS A PREVENTIVE MEASURE OF CISPLATIN INDUCED NEPHROTOXICITY**
KNH/UoN ERC Approval Number :	
Researcher's Name :	Dr Marius Beniet Youan Bi

Today, you received high-dose cisplatin. To prevent damage to your kidneys, you need to drink plenty of fluids.

- Drink all types of fluids such as:
  - Water.
  - Milk.
  - Juices.
  - Decaffeinated soft drinks (soda).
- Do NOT drink water only. Avoid fluids with caffeine and alcohol.
- If you have diabetes or problems with your blood sugar, drink fluids with no sugar. Otherwise add water to sweet drinks (half water and half juice).
- Starting tomorrow: Drink **two to three quarts (eight to twelve 8-ounce glasses)** of fluid every day for a week.
- Take your antinausea medicines as the research nurse told you to.

• **PLEASE KEEP TRACK OF YOUR FLUID INTAKE.**

• **This may help you to see that you are reaching your goal.**

Write the time and number of ounces that you drink today and the following day.

Time	Number of Ounces/glasses	Time	Number of Ounces/glasses

Call Your Nurse or Doctor if you:

- Are urinating less frequently or in smaller amounts than normal.
- Have nausea, vomiting, or diarrhea.
- Have dizziness.
- Are unable to eat or drink for more than 24 hours after getting high-dose cisplatin.
- Have a fever of 100.4° F (38° C) or higher.
- Have heartburn.
- Have any unexpected, or unexplained problems.
- Have any questions or concerns.

The information on this card is selective and does not cover all possible side effects; others may occur.

Please report any problems to the investigator.

APPENDIX 3: Common Terminology Criteria for Adverse Events (CTCAE) Version 4.03

Research Study Title	**EVALUATION OF INTRAVENOUS PRELOADING MAGNESIUM SUPPLEMENTATION AS A PREVENTIVE MEASURE OF CISPLATIN INDUCED NEPHROTOXICITY**
KNH/UoN-ERC Approval Number :	
Researcher's Name :	Dr Marius Beniet Youan Bi

Renal and urinary disorders					
	Grade				
Adverse Event	**1**	**2**	**3**	**4**	**5**
Acute kidney injury	Creatinine level increase of >0.3 mg/dL; creatinine 1.5 - 2.0 x above baseline	Creatinine 2 - 3 x above baseline	Creatinine >3 xbaseline or >4.0 mg/dL; hospitalization indicated	Life-threatening consequences; dialysis indicated nto kidney), renal (kidney dama	Death
Bladder perforation	-	Extraperitoneal perforation, indwelling catheter indicated	Intraperitoneal perforation; elective radiologic, endoscopic or operative intervention indicated	Life-threatening consequences; organ failure; urgent operative intervention indicated	Death
Definition: A disorder characterized by a rupture in the bladder wall.					
Bladder spasm	Intervention not indicated	Antispasmodics indicated	Hospitalization indicated	-	-
Definition: A disorder characterized by a sudden and involuntary contraction of the bladder wall.					
Chronic kidney disease	eGFR (estimated Glomerular Filtration Rate) or CrCl (creatinine clearance) <LLN 60 ml/min/1.73 m2 or proteinuria 2+ present; urine protein/creatinine >0.5	eGFR or CrCl 59 - 30 ml/min/1.73 m2	eGFR or CrCl 29 - 15 ml/min/1.73 m2	eGFR or CrCl <15 ml/min/1.73 m2; dialysis or renal transplant indicated	Death