"COMPARISON BETWEEN SERUM THEOPHYLLINE LEVELS AFTER RECTAL AND INTRAVENOUS ADMINISTRATION OF AMINOPHYLLINE TO PRETERM NEONATES AT KNH NEWBORN UNIT"

A RESEARCH DISSERTATION SUBMITTED IN PART FULFILMENT OF MASTER OF MEDICINE DEGREE IN PAEDIATRICS AND CHILD HEALTH OF THE UNIVERSITY OF NAIROBI

BY

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DECLARATION

I declare that this dissertation in part fulfilment of my M.Med thesis (paediatrics and child health) is my original work and has not been presented to any other university or forum

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This dissertation has been submitted for consideration and with our approval as university supervisors

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DEDICATION

- To my family Mercy, Melanie and Margaret for being there for me.
- To my parents Mr. and Mrs. Chumba for their continued guidance and support.
- To the children of this country who are so dear to me.

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OPERATIONAL DEFINITIONS

NEONATE - A baby in the first 28 days of the postnatal life

PRETERM - A neonate delivered before 37 completed weeks of gestation.

LOW BIRTH WEIGHT - Neonate weighing less than 2500 grammes at birth

APNOEA OF PREMATURITY-

1) Cessation of respiratory airflow for more than 20 seconds in a stable preterm neonate.

OR Cessation of respiratory airflow for less than 20 seconds in a stable neonate with accompanying bradycardia, heart rate below 120 beats per minute and/or cyanosis.

2) For the purposes of the study, apnoea was significant if it occurred between 2nd and 7th day postnatal

PRIMARY APNOEA - This is apnoea without any identifiable Predisposing factor such that the apnoea can be Solely attributable to prematurity.

SECONDARY APNOEA - This is apnoea that occurs due underlying Disease/condition e.g. hypothermia, hypoglycemia and sepsis

TACHYCARDIA - Heart rate above 170 beats per minute in undisturbed preterm. (Normal 120-170 beats per minute).
Severe if above heart rate is above 200 beats per minute. [2]

BRADYCARDIA -Heart rate below 100 beats per minute in undisturbed preterm.[2]

NEONATAL SEPSIS

1) **Suspected sepsis-**This when a neonate is suspected to have sepsis On history and physical examination

2) **Confirmed sepsis** - This is when bacterial sepsis is confirmed by Isolating the causative organism in cultures of otherwise sterile fluids or area e.g. Csf. blood, urine or skin swabs.

HYPOTHERMIA - Neonate' s rectal temperature below 36^{0} c

HYPERTHERMIA - Neonate' s rectal temperature above 37.8^{0} c

SUB THERAPEUTIC-Serum theophylline levels: below5ug/ml

THERAPEUTIC –Serum theophylline levels: 5-15ug/ml

TOXIC - Serum theophylline levels: above15ug/ml

PEAK LEVELS - Serum theophylline levels: 1 hour after aminophylline loading dose

STEADY STATE LEVELS: Serum theophylline levels after 72 hours of aminophylline administration (3 hours after the seventh aminophylline dose)

LIST OF ABBREVIATION

KNH	-	Kenyatta National Hospital
Mgs	-	Milligrams
Kg	-	Kilograms
µg	-	Micrograms
µl	-	Micro litres
ml	-	Millilitres
NBU	-	New born Unit
CAMP	-	Cyclic Adenosine Monophosphate
AOP	-	Apnoea of Pre-maturity.
SHO	-	Senior House Officer
PROM	-	Premature Rupture of the Membranes
BBA	-	Born Before Arrival
EDTA	-	Ethylene Diamine Tetra acetic Acid
IV	-	Intravenous
CSF	-	Cerebrospinal fluid
>	-	Greater than.
<	-	Less than
Bwt	-	Birth weight
Hr(s)	-	Hour (s)

ABSTRACT

Background: Aminophylline IV formulation has traditionally been administered rectally at the Kenyatta National Hospital Newborn Unit (KNH- NBU) to prevent and treat apnoea of prematurity (primary apnoea).Apnoea of prematurity (AOP) affects premature neonates especially those with a gestational age of 32 weeks and below. Previous studies on rectal aminophylline administration as enema or suppositories in preterm neonates recommended rectal route as a good alternative to oral or intravenous routes. This study was conducted to compare serum theophylline levels after intravenous and rectal administration of the aminophylline IV formulation to preterm neonates at the KNH- NBU.

Objective: To compare serum theophylline levels after IV aminophylline infusion and rectal administration of the IV aminophylline formulation to preterm neonates at the KNH- NBU

Methods: Neonates with gestational age of 32 weeks and below were randomly assigned to receive IV or rectal aminophylline. Eligibility criteria included stable inborn preterm neonates of gestational age 32 weeks and below without sepsis, risk of sepsis or significant congenital anomalies. Consent was also obtained from the parent or guardian before a neonate was recruited into the study. Neonates who were on drugs that could interfere with theophylline metabolism were excluded. Aminophylline was administered at a loading dose of 5 mgs/kg bodyweight followed by 2 mgs /kg bodyweight per dose administered every12 hours in both arms of the study. Two mls (2) of blood were drawn at peak (1hour after loading dose) and at steady state, 72hours after initiation of aminophylline administration and 3 hours after the 7th aminophylline dose for analysis of serum theophylline levels. Serum theophylline levels were analysed using AxSYM Immunoassay Analyser and Theophylline Reagent Kit (Abbot Laboratories). Baseline heart rates were recorded at admission and twice daily during the 72 hour period of follow-up.

Results: There were 61 neonates in each arm of the study. There was no significant difference in median birth weights between the two arms of the study. The IV arm had a median birth weight of 1250g (range600-1600) compared to 1300g (range 800 -1600) in the rectal arm (p= 0.634). The IV arm had a median gestational age of 29weeks (range26-32) compared to 30 weeks (range 26 -32) in the rectal arm. This difference in median gestational ages were not significant (p=0.334). Rectal arm had significant lower peak theophylline levels (range 0.4-14.76) compared to 6.82 µg/ml (range 0.68-18.05) in the IV arm

(p=0.0004). The median steady state trough levels was still significantly lower at 6.6[µg/ml (range 0.41-16.46) in the rectal arm compared to 10.48 µg/ml (range 2.25-40) in the IV arm (p= 0.0001). Neonates in the rectal arm were two times more likely to attain sub therapeutic theophylline peak levels compared to the IV arm [OR 2.36(95%CI 1.06-5.27) p=0.02]. Neonates in the IV arm were five times more likely to have toxicity at steady state trough levels [OR5.06 (95%CI 1.21-24.29),p=0.01] and less likely to have sub therapeutic levels though this difference was not significant] OR 0.63 (95%CI 0.24-1.62), p= 0.29]. Tachycardia was more evident after 72 hours (steady state). Neonates in the IV arm were two times more likely to have tachycardia at 72 hours compared to the rectal arm, a difference that was significant [RR 1.85 (95%CI1.36-2.51), P=0.001].

Conclusion

Serum theophylline levels were sub therapeutic in most neonates one hour (peak levels) after rectal administration of IV aminophylline formulation. At steady state, rectal arm had more neonates with sub therapeutic levels while Intravenous arm had more neonates with toxic levels. Tachycardia was more prevalent after IV aminophylline infusion. The number of neonates with therapeutic serum theophylline levels at steady state was comparable in the two arms of the study. From this study result, further research is recommended to look at whether serum theophylline levels would improve to be within therapeutic range in most neonates at peak and steady state if higher doses of IV aminophylline formulation are administered rectally or aminophylline loading doses are infused followed by maintenance doses rectally at the same doses and schedule used in this study.

INTRODUCTION AND LITERATURE REVIEW

1.0 Apnoea of prematurity.

Apnoea of prematurity (AOP) also known as primary apnoea is defined as cessation of respiratory airflow for a period exceeding 20 seconds or if shorter, it is accompanied by either bradycardia or arterial oxygen desaturation [1, 2, 3, 4]. The occurrence of AOP is inversely related to gestational age (prevalent in neonates of 32 weeks and below) and directly related to active sleep[1, 2]

The prevalence of AOP has increased in the recent past due to the improvement in neonatal care[2, 3] which has enhanced survival of neonates with low gestational ages (below 34 weeks) and birth weights especially the extremely low (<1000g) and the very low (<1500g) birth weight babies. In the United States of America (USA), approximately 70% of preterm neonates born below 34 weeks gestation have at least one clinically significant apnoeic episode requiring hospital admission[3].

At the KNH NBU, forty two percent of all low birth weight neonates admitted between January and December 2000 had apnoea though they were not classified as having primary or secondary apnoea.[5]

Aetiology, pathogenesis and management

The cause of primary apnoea (AOP) is thought to be due to abnormal breathing control caused by neuronal immaturity of the brain stem. It commonly occurs from the 2nd to the 7th day of life and rarely on the first day[1, 2, 3]. Primary apnoea (AOP) should be differentiated from secondary apnoea, which occurs due to an underlying pathology.[1, 2, 3, 4] Some of the known causes of secondary apnoea include sepsis, metabolic disorders, temperature instability, cardiovascular and respiratory problems, and intracranial haemorrhage.[1, 2, 4].

Apnoea is associated with a decrease in cerebral blood flow, hence a decrease in cerebral oxygen delivery. This decrease in cerebral blood flow may lead to an increase in the incidence of intraventricular haemorrhage, hydrocephalus and mechanical ventilation that may later progress to abnormal neurodevelopmental outcome especially after the first year of life[3, 6].

Depending on the underlying cause, apnoea can be managed by tactile stimulation, elevation of the infant's jaw, supplemental oxygen by bag, valve and mask ventilation, drugs (aminophylline, theophylline, caffeine citrate and doxapram) and continuous positive airway pressure ventilation when drugs fail to relieve the apnoea, using nasal prongs, nasal mask or facemask with three to six cm of water pressure.[1, 4, 5]

1.2 Aminopylline

Description.

Aminophylline is a methylxanthine derivative, a stable complex of theophylline and ethylenediamine that contains between 84% to 87.4% anhydrous theophylline and 13.5% to 15% anhydrous ethylenediamine. It is a white or yellowish powder, odourless with a slight ammoniacal odour[7, 8].

Indications and mode of action

Aminophylline is the drug of choice for prophylaxis and treatment of AOP and as an adjunct in weaning from mechanical ventilation[7, 8]. Its exact mechanism of action in relieving AOP is unknown but it is thought to be through a combination of stimulation of the respiratory centre, increased minute ventilation and increased muscle tone. At molecular level, it thought to act by inhibiting the enzyme phosphodiesterase with a resultant increase in cyclic AMP, translocation of intracellular calcium and adenosine receptor antagonism[7, 8]

Routes of administration

Ideally, aminophylline is administered as a slow intravenous infusion, orally as a solution or immediate release oral solid formulations and rectally as enema or suppository. Suppositories have been associated with toxicity in preterm neonates due to erratic and unreliable absorption[7,8]. It is well absorbed as enema [7, 8]. Following rectal administration, absorption may be slow, erratic and often incomplete.[7, 8]

Absorption, metabolism and elimination.

After aminophylline absorption, theophylline the active drug is readily released into circulation. It does not undergo any significant first pass effect. Peak theophylline levels are achieved within one hour after oral solid formulation and rectal enema[7, 8]

Approximately 40% of the drug is plasma protein bound. Unbound theophylline distributes throughout body fluids, but poorly into fat[7, 8]. The volume of distribution in preterm neonates is approximately 0.69 +/-0.095 litres per kilogram[9] After repeated dosing at equal intervals, serum steady state concentration is achieved 72 hours later .[7, 8]

The half-life of theophylline ranges from 10 to 45 hours in term and preterm neonates. The liver in children above one year and adults metabolizes it by biotransformation, catalyzed by different isoenzymes of cytochrome P450. In neonates, the N-demethylation pathway is absent while the functions of hydroxylation pathway is markedly deficient and approaches maximum levels at one year. Therefore, in neonates, fifty percent (50%) of the administered drug is excreted unchanged in urine. [7, 8]

Three separate studies on the pharmacokinetic profiles of aminophylline after IV infusion in preterm neonates recommended different loading and maintenance doses. These doses ranged from 5 to 7 mgs/kg bodyweight loading dose followed by 3 to 4 mgs/kg bodyweight per 24 hours maintenance dose.[10, 11,12] Currently at the KNH-NBU aminophylline is administered at a loading dose of 5-mgs/kg bodyweight followed by 2mgs /kg body weight maintenance doses every12 hours.

Adverse effects.

Theophylline has a narrow therapeutic margin. Serum levels between 5 and 15 µg/ml are required in the treatment and prophylaxis of AOP although some neonates may respond to lower levels. This is due to the wide inter individual variability in metabolism and elimination of theophylline [7, 8]. Serum levels above 15µg/ml are associated with a wide range of adverse effects, which include tachycardia, nausea, vomiting, insomnia, diarrhoea, irritability, gastro-paresis and transient diuresis. [7, 8, 13] while levels below 5ug/ml are associated with persistence of apnoea. Serum theophylline levels should be determined routinely for example (i) after initiating therapy to determine final dose adjustment, (ii) adjusting the dose in a neonate who continues to be symptomatic (iii) when there are signs of toxicity, (iv) when additional medication which may alter theophylline metabolism and exeretion is required.[7, 8, 13].

1.3 Practice at the KNH-NBU

At the KNH-NBU, neonates at risk of apnoea (gestational age of 32weeks and below) and neonates experiencing apnoeic episodes due to prematurity are started on aminophylline IV formulation administered rectally. Neonates with suspected secondary apnoea are investigated and managed according to the underlying cause.

STUDY JUSTIFICATION

Previous studies on rectal aminophylline administration in preterm neonates used either enema or suppositories and concluded that the rectal route is a therapeutically acceptable alternative to IV and oral route (as solid formulation or solution) but recommended further evaluation of this route[14, 15, 16]. The enema formulation is not easily available while suppositories being solid formulations are difficult to administer in mgs/kg body weight .No study has been done using the IV formulation administered rectally in this group of neonates. Though intravenous aminophylline formulation is widely available in most health facilities, its administration requires a syringe pump and close monitoring due to the risk of toxicity associated with this route. This becomes difficult in a resource poor setting where monitoring may not be adequate and the right equipment often unavailable.

At the KNH-NBU, IV aminophylline formulation is administered rectally yet the appropriateness of this formulation for rectal use has not been evaluated before.

Study utility

The findings of the study will guide on whether to continue using IV aminophylline formulation rectally at the current dose as used at the KNH-NBU or make necessary changes to the formulation used, route or dose given.

Null Hypothesis

There is a significant difference in serum theophylline levels after intravenous and rectal aminophylline administration using the IV formulation.

Research question.

Is rectal aminophylline administration of the IV formulation as used at the KNH-NBU to manage AOP in preterm neonates appropriate?

Main objective

1. To compare serum theophylline levels after rectal aminophylline administration with intravenous infusion of the IV formulation at the same dose and schedule of 5mgs/Kg bodyweight loading dose and 2mgs/Kg bodyweight per dose administered every 12 hours.

Specific objectives

1. To compare serum theophylline levels achieved one hour after IV and rectal aminophylline administration.

2. To compare serum theophylline levels achieved 72 hours after continued IV and rectal aminophylline administration.

3. To compare percentage of neonates with sub therapeutic, therapeutic and toxic levels between the IV and rectal arms.

4. To relate the route of administration with tachycardia during the 72-hour period of aminophylline administration.

METHODOLOGY

Study design

An open randomized comparative study.

Study area

The study was conducted in the newborn unit of KNH. This unit offers level one and two care to neonates and serves as the National Referral and Teaching Hospital. Neonates admitted are categorized and nursed according to their weights; neonates of the same weight category being nursed together in the same nursery. Clinical care is provided by Neonatologist, Paediatricians, senior house officers (SHOs) and Nurses. Monthly admissions average 160 of whom 60% are low birth weight with 76%of them being preterm neonates [17,18].

Reference Population.

Preterm neonates who were admitted in KNH-NBU during the study period

Inclusion Criteria

Neonates admitted in KNH-NBU during the study period and fulfilled the following criteria

1) Gestational age of 32 weeks and below

2) Born within the KNH

3) Consent given for the study by the parents/guardian.

Exclusion Criteria

The neonates who had any of the following conditions were excluded from the study

1. Gestational age above 32 weeks
2. Born before arrival or transfer in.
3. Sepsis/fever
4. Prolonged rupture of the membranes
5. Neonates who were on drugs capable of interfering with aminophylline metabolism such as phenobarbitone, erythromycin, cimetidine, diazepam, ephedrine, phenytoin and rifampin.
6. Neonates who had significant congenital anomalies.

Sampling Method

Neonates admitted into the NBU were screened and those who met the inclusion criteria were enrolled into the study. They were randomly assigned to receive aminophylline rectally or intravenously after consent was obtained from parent/guardian. Randomization was done using random sampling tables (**Appendix... VI**) [19]. Three (3) columns were used. Starting at an arbitrary point in the random sampling tables, the researcher moved down the columns and continued to the next group of columns, writing down the numbers up to a total of 122. Numbers like 03 were counted as 3 while 00 and repetitions were ignored. The numbers were then written down on small cards. These cards were then divided equally into two, one group for rectal and the other IV then mixed thoroughly and kept in one container. From this container, they were picked randomly such that the probability of picking a number for either rectal or IV card was equal. Whenever a neonate was recruited, a card was picked from the container randomly and depending on the number written on the card, the neonates received either IV or rectal aminophylline.

Sample size determination

Sample size was calculated using the formula for comparing proportions from two independent groups[19].The sample size calculated was 122 subjects, 61 neonates in each arm of the study.

- $$n = \left\{ \frac{\not{Z}_{1-\alpha/2}\sqrt{\{2P(1-P)\}} + \not{Z}_{1-\beta}\sqrt{\{P_1(1-P_1) + P_2(1-P_2)\}}}{(P_1 - P_2)^2} \right\}^2$$

$$P = \frac{P_1 + P_2}{2}$$

P_1 = Percentage number of neonates who attained sub therapeutic concentration while on IV aminophylline from a previous study = 19 %[20]

P_2 = Percentage number of neonates who were expected to attain Subtherapeutic levels while on rectal aminophylline = 44 %(assuming a 25% difference is expected between the IV and rectal groups with most of the neonates on rectal aminophylline having sub therapeutic levels)

$n_1 : n_2 = 1:1$

$\not{Z}_{1-\alpha/2}$: 0.05 at 95% CI in a two-tail test=1.96

$\not{Z}_{1-\beta}$ = 80% power 0.842

n = 61

Total = 122

Clinical procedures and laboratory assays

Identification, training and the responsibilities of the Research team

The research team included the principal researcher, SHOs and nurses' working in NBU at the time the research was conducted.

The principal researcher carried out training of the research team. This was done prior to starting the study with the objective of minimising inter observer errors. The main members of staff trained were nurses and SHOs working in NBU at the time the research was being conducted. The content of this training was on the research to be conducted. This was done by holding a presentation and discussion followed by laying down a standard criterion to be used by all NBU staff to diagnose and grade the severity of apnoea. There was also a demonstration on how to assess respiratory airflow using chest wall movements or a tiny piece of cotton wool across the nostrils and how to identify a neonate with cyanosis clinically. Guidelines on how to assess a neonate with an apnoeic attack were provided. In cases of apnoea, other than resuscitating the neonate, they assessed respiratory airflow, checked for any signs of cyanosis and counted the heart rate for one minute then recorded the findings on the tally sheets provided. Success of sensitization was gauged by administering a questionnaire to the relevant staff after the sensitization (**Appendix V**). Follow up discussion were carried out every fortnight during the study period.

The SHOs were responsible for obtaining blood samples if the researcher was not available at the particular time a neonate was recruited. The principal researcher passed through the unit twice daily; every morning (at 8 am) and evening (at 8 pm) and at any other time as necessary, to review fresh and previous recruits and to ensure proper and adequate administration of the drug by checking on equipment, supplies and IV lines in each relevant units. The principal researcher also collected blood samples already drawn and took them to the laboratory. He was available on phone at all times.

Nurses working in NBU assisted the principal researcher administer aminophylline to the neonates and to monitor these neonates. They were responsible for checking the heart rates and rectal temperatures. They recorded the findings in the appropriate tally sheets that were provided by the principal researcher.

Identifying, recruiting and managing neonates

Neonates admitted who fulfilled the inclusion criteria were identified for the study. Once a potential neonate was identified, the parent/guardian was sought and an informed written consent obtained, then the neonate was recruited and randomly assigned to receive rectal or IV aminophylline. Neonates who exhibited any of the factors on the exclusion criteria were excluded. Sepsis was determined by antenatal and prenatal predisposition or clinical suspicion. Neonates with suspected sepsis were investigated with a full haemogram from which calculation of immature to total neutrophil ratio was done. A haemogram result obtained that was suggestive of infection, with neutropenia (absolute neutrophil count below 2.0 x10 ^{9}per litre) or neutrophilia (absolute counts above 7.5 x10 ^{9}per litre) or an elevated ratio of immature to total neutrophil counts above 0.2 were excluded. Also excluded were neonates confirmed to have sepsis with a positive blood culture. Neonates who developed apnoea while already in the study with features suggestive of sepsis were investigated as above and depending on the results obtained, were either excluded or allowed to continue in the study.

Neonates recruited into the study were examined daily, their rectal temperatures taken with a digital thermometer that read to an accuracy of 0.1^{o}c inserted approximately 2 centimetres from anal margin into the rectum and left in situ for one minute. In addition, their birth weights were taken at admission using a top bar spring scale model A1Z that weighed to an accuracy of 50 grammes. Baseline heart rates were taken using a stethoscope placed on the precordium and the heartbeat counted for one minute. Neonate's gestational age was assessed using the Dubowitz scoring system (**Appendix...VII**). Neonate's postnatal age was recorded in hours after birth.

Aminophylline administration

Aminophylline IV formulation obtained from Laborate Pharmaceutical (INDIA) was used. It is supplied as 10 mls ampoule containing 250 mgs of aminophylline. Aminophylline concentration in this ampoule is equivalent to 25 mgs per ml. To facilitate administration of small doses of aminophylline, a one ml insulin syringe was used. This syringe is calibrated into 100 units therefore one (1) unit in this syringe is equivalent to 0.01mls. Aminophylline was administered for the required duration (up to 36 weeks post conception age). Aminophylline loading and maintenance doses for weights between 600 to 1600 grammes were calculated in mls by the principal researcher and pinned on the walls of relevant units for accuracy, ease and uniformity of administration.

Rectal administration

The exact rectal aminophylline dose was withdrawn from the drug vial using the one-milliter insulin syringe then pushed into the rectum with the same syringe, the tip inserted approximately 1.5 centimetres from anal margin into the rectum.

IV administration

A Medivols Infusion Soluset (gravity feed measure volume fluid infusion set with a burette graduated to 150 mls) was used to administer IV aminophylline. To ensure the exact dose was administered, the one ml insulin syringe was used to withdraw the required dose from the drug vial then injected through the injection site of the medivols infusion set (position 3 in the drawing of soluset, **Appendix...VIII**) into the graduated burette and topped up to a volume of 20 mls by adding 5% dextrose into the burette. The drug was then allowed to flow from the burette into the drip chamber through gravity by releasing the flow regulator, while at the same time setting the flow rate to 40 drops per minute (equivalent to delivering a volume of 20 mls within half an hour). From the drip chamber, the infusion fluid was allowed to flow into the tubing and finally into the intravenous cannula of the patient through gravity by releasing the flow regulator. Since aminophylline does not adsorb onto the tubing there was no need to adjust the dose given or flush the tubing. For purposes of the study, the neonates were followed up for 72 hours. Thereafter they continued with their aminophylline treatment for the required duration as per the institution policy.

Blood sampling procedures

Venous blood samples were drawn one hour after loading dose in both groups. Once the vein for venipuncture was identified, sterile preparation of the skin was done using methylated spirit, followed by venipuncture with a butterfly needle. Two (2) mls of blood was drawn into a 2.5 ml syringe then transferred into a plain bottle, labelled and taken to the laboratory. The principal researcher or the Senior House Officer on duty removed these samples. Gentle pressure was applied on the venipuncture site after withdrawing the butterfly needle until bleeding stopped and there was no risk for further bleeding. These neonates were then followed up with twice-daily reviews by the principal researcher. A second blood sample was drawn 72 hours after initiation of the aminophylline administration, 3 hours after the 7th aminophylline dose for assay of theophylline steady state levels.

Blood cultures were done using the same aseptic technique, 2 mls of blood was drawn into a 2ml syringe using the butterfly needle then transferred into a sterile blood culture bottle

containing aerobic culture media with utmost care to avoid contamination and taken immediately to the microbiology laboratory for cultures and sensitivity. For the haemogram, 1 ml of blood was drawn into the 2ml syringe using the butterfly needle and transferred quickly to an EDTA vacutainer, shaken gently to mix with the anticoagulant and taken to haematology laboratory where a Coulter counter was used to analyze the sample and a print out of a full blood picture obtained. At the same time a peripheral blood film was prepared and immature to total neutrophil ratio was calculated. The investigation results were brought to the unit by the clerk and handed over to the SHO for appropriate action.

Monitoring heart rate and rectal temperature

Prior to starting aminophylline administration, baseline heart rate and rectal temperature were obtained. The heart rate was counted for one full minute. The rectal temperature was taken using IN4 care digital clinical thermometer [IN4 care technology Taiwan, model INA-B162A] that reads body temperature to an accuracy of ± 0.1°C. The thermometer was placed approximately two centimeters from anal margin into the rectum and left in situ for one minute, after which the reading was recorded. Subsequently, apart from the routine three hourly temperatures and heart rate recording there were two extra daily recording of heart rate and rectal temperature, in the morning (8am) and evening (8pm) one hour after every aminophylline dose. The heart rates that were taken after aminophylline doses (at 8 am and 8 pm) were used during result analysis. This were done by the nurses and recorded on the data collection sheets attached to the patient notes.

Assaying of aminophylline blood levels

Blood samples for assay of serum theophylline levels were labelled using study numbers, put in a plain safe bag and taken to the laboratory within one hour of collection. Once a sample was received in the laboratory, it was centrifuged and serum separated and labelled using the same study number as the blood samples then frozen at a temperature of negative 4 °C. Assay of Serum theophylline levels were done in batches of one hundred using AxSYM Immunoassay analyzer (Abbot) and theophylline II assay reagent kit (Abbot Laboratories) that utilizes Fluorescence polarization immunoassay technology. The minimum amount of serum required was 150 µl.[21] Results obtained were printed out in µg/ml.

DATA MANAGEMENT

Data collection

A standard data collection form was used to collect all the data generated in the study. (**Appendix...III & IV**)

Data analysis

Data was analyzed using statistical package for social sciences (SPSS) version 9 and EPI info. Mann-Whitney u test was used to compare the baseline characteristics and serum theophylline levels in the two-study population. Chi square and Odds ratio (using EPI info) was used to compare neonates who had sub therapeutic, therapeutic and toxic serum theophylline levels between the two-arms. P values below 0.05 were considered significant. Results are presented in graphs and tables

Ethical Consideration.

The research proposal for this study was presented to the KNH ethics and research committee (ERC) for review and approval prior to starting the study. Informed written consent was obtained from the parent/guardian for each patient recruited into the study .The signed consent forms were kept safely by the principal researcher. (**Appendix I &II**).

Aseptic techniques during procedures were followed strictly. Pressure was applied on the venipuncture site to ensure no bleeding. Used material especially sharps were disposed off carefully to avoid accidental injuries.

Recruited neonates were identified using study numbers for confidentiality. Resuscitation and emergency care took priority during the study period and the principal researcher participated in the management of these neonates.

Relevant clinical and laboratory finding were passed on to the primary care clinician during the study period to help with decision making during patient management.

RESULTS

Study population

Over a four-month period, one hundred and twenty two (122) neonates had their blood samples analyzed for serum theophylline levels. Sixty one (61) neonates in each arm of the study. The IV arm had 34 males and 27 females while the rectal arm had 29 males and 32 females. Gestational age ranged from 26 to 32 weeks in both arms with a median of 29 weeks in the IV arm and 30 weeks in the rectal arm. Median birth weight was 1250g [range 600-1600 g] in the IV arm and 1300g [range 800 - 1600g] in the rectal arm. (Table 1)

Table1

Baseline characteristics of the study population.

Characteristics	**Route of administration. Median (range)**		**P value**
	IV arm (n=61)	Rectal arm (n=61)	
Birth weight (grammes)	1250(600-1600)	1300 (800-1600)	0.634
Gestational age (weeks)	29(26-32)	30(26-32)	0.335
Heart rate at admission (beats/min)	144 (120-168)	142(100-164)	0.657
Rectal temperature (°centigrade)	36.3(33.2-37.8)	36.4(33.4-37.0)	0.80

The Birth weights and gestational ages of the two study populations were comparable. No significant difference was found between the two populations. P = 0.634 and 0.335 respectively (Table 1).

Table 2

Pattern of serum theophylline levels at peak and steady state levels after Rectal and IV aminophylline administration to preterm neonates

serum theophylline levels	Route of aminophylline administration		P value
	IV arm (n=61) Median (range)	Rectal arm (n 61) Median (range)	
Peak theophylline levels in ug/ml (1hr)	6.82(0.68-18.05)	4.32(0.40-14.76)	0.0004
Steady state theophylline level in ug/ml (after 72 hrs)	10.48(2.25-40)	6.61(0.41-16.46)	0.0001

Peak serum theophylline levels were higher in the IV arm compared to the rectal arm. This difference was significant (P=0.0004).

Steady state trough serum theophylline levels were significantly higher in the IV arm compared to the rectal arm. (P =0.0001, Table 2).

Table 3

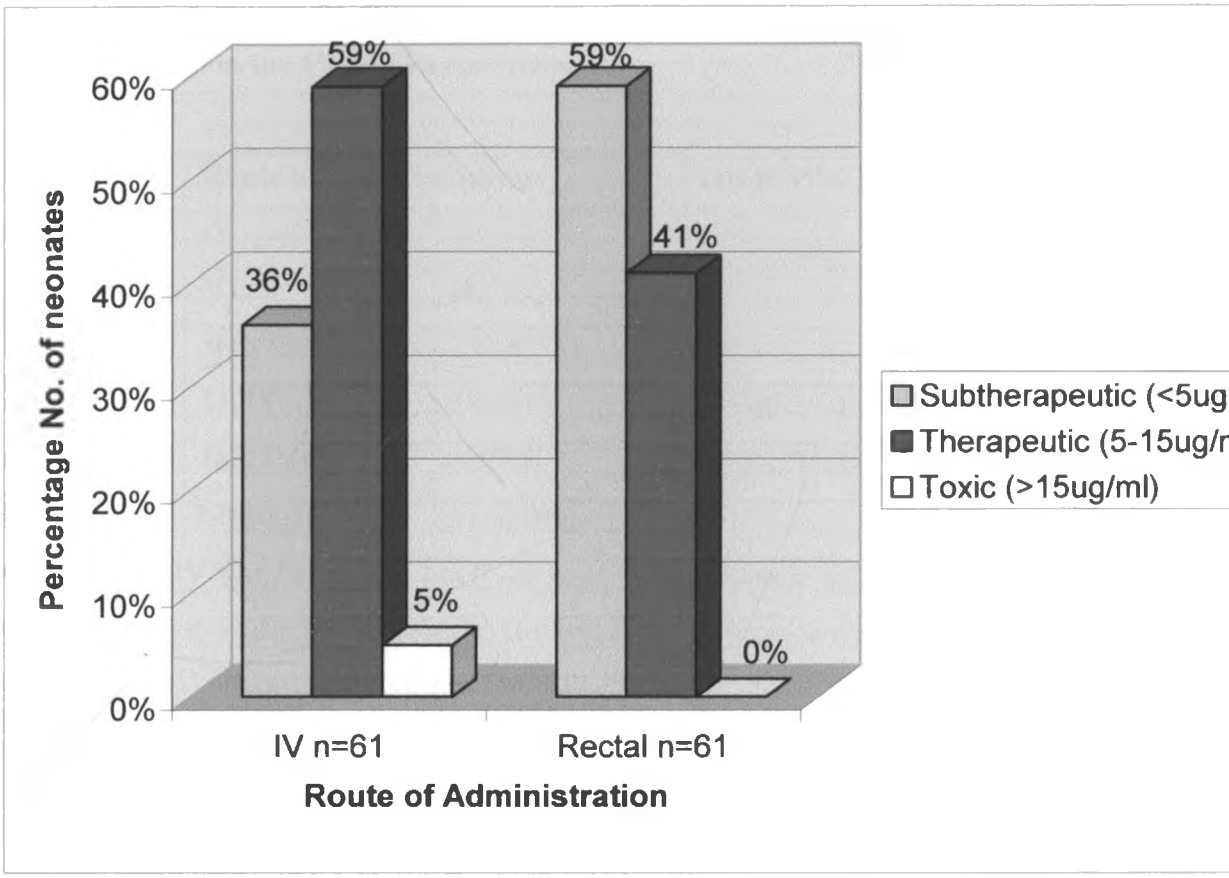
Preterm neonates who achieved Sub therapeutic and Therapeutic Peak serum theophylline levels in the IV and Rectal arms.

Peak serum theophylline levels	Route of aminophylline administration.		OR (95%CI)	P value
	IV arm N (%)	Rectal arm N (%)		
Subtherapeutic	22(38%)	36(59%)		
Therapeutic	36(62%)	25(41%)	2.36(1.06-5.27)	0.02
Total	58(100%)	61(100%)		

Neonates on rectal aminophylline arm were two times more likely to have sub therapeutic peak (1 hour after loading dose) theophylline levels compared to the IV infusion arm (Table3). Three neonates (3) in the IV arm had toxic levels and none in the rectal arm of the study. These numbers were too few hence not included in the analysis.

Figure 1

Peak Serum Theophylline Levels after Rectal and IV administration of aminophylline in preterm neonates.



The IV arm had more neonates with therapeutic peak theophylline levels compared to the rectal arm.

The rectal arm had more neonates with sub therapeutic levels compared to the IV arm.

Three neonates (5%) in the IV arm had toxic levels and none in the rectal arm (Figure 1).

Table 4

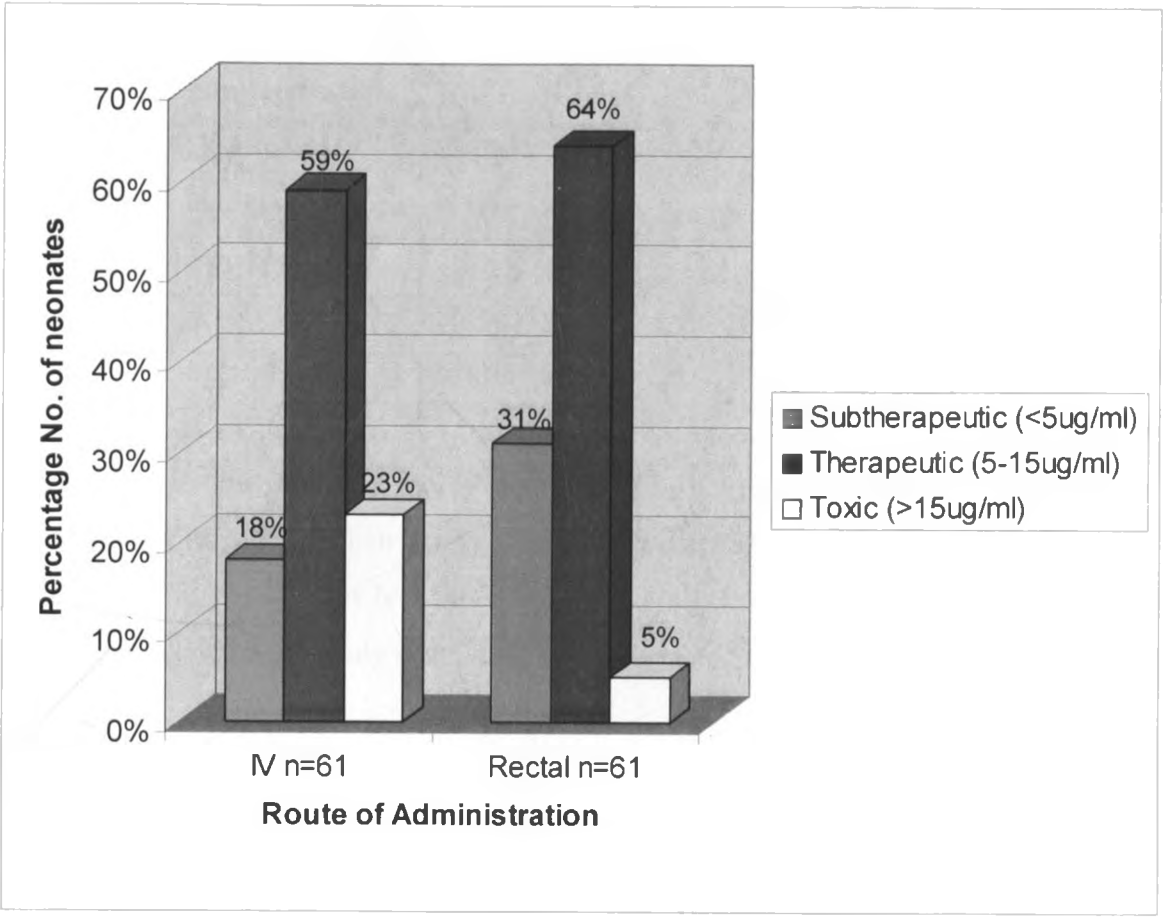
Preterm neonates who achieved Sub therapeutic, Therapeutic and Toxic steady state theophylline levels in the IV and Rectal arms

Steady state theophylline levels	Route of administration		OR (95%CI)	P value
	IV arm N (%)	Rectal N (%)		
Therapeutic	36(59%)	39(64%)	1.0	
Sub therapeutic	11(18%)	19(31%)	0.63(0.24-1.62)	0.29
Toxic	14(23%)	3(5%)	5.06(1.21-24.29)	0.01
Total	61(100%)	61(100%)		

Neonates in the IV arm were less likely to have subtherapeutic serum theophylline level compared to the rectal arm at steady state. However, this difference was not significant. Neonates in the IV arm were five times more likely to have toxic serum theophylline level compared to the rectal arm. This difference was statistically significant ($p< 0.05$, table 4)

Figure 2

Pattern of steady state serum theophylline levels in preterm neonates in the IV and Rectal arms.



The two arms had an almost equal percentage of neonates with therapeutic levels at steady state trough levels. The rectal arm had more neonates with sub therapeutic levels compared to the IV arm.

The IV arm had more neonates with toxic levels compared to the rectal arm (Figure 2).

Table 5

Neonates with tachycardia after 72 hrs of Rectal and IV aminophylline administration to preterm neonates

Heart rate after 72 hours of aminophylline administration.	**Route of aminophylline administration.**		**RR (95%CI)**	**P value**
	IV arm N (%)	Rectal arm N (%)		
Tachycardia	19(31%)	5(8%)	1.85(1.35-2.51)	0.001
No tachycardia	42(69%)	56(92%)		
Total	61(100%)	61(100%)		

Neonates in the IV arm were two times more likely to have tachycardia after 72 hours of aminophylline administration when steady state had been achieved (p=0.001, table 5).

Note: 8 neonates in the IV arm had more than one episode of tachycardia compared to 2 neonates in the rectal arm at steady state.

DISCUSSION

This was an open randomized comparative study that compared serum theophylline levels after intravenous and rectal aminophylline administration of the IV formulation to stable preterm neonates of gestational age 32 weeks and below at the KNH-NBU.

Recruited neonates were randomised into the two groups, IV and rectal using random sampling numbers. Statistical analysis of birth weights and gestational ages in the two study populations were comparable therefore randomization worked. Analysis of serum theophylline showed a difference in median theophylline levels at peak (after 1 hr) and steady state (after 72 hrs) between the two arms. Since birth weights and gestational ages were comparable in the two arms of the study, this difference could be attributed to the different routes used to administer aminophylline.

The results of this study indicate that intravenous aminophylline formulation is absorbed rectally. Median peak level after rectal aminophylline administration was 4.32ug ml [less than the minimum therapeutic level of 5ug/ml]. Furthermore, fifty-nine percent (59%) of the neonates in this arm of the study had sub-therapeutic levels. This suggests that the rate of absorption of this formulation rectally might be slow hence the lower peak levels obtained, alternatively, the one-hour cut off used to assay for peak theophylline levels in this study missed the actual peak after rectal aminophylline administration. However, theophylline levels improved with continued rectal aminophylline administration to reach a median steady state level of 6.61ug/ml which was marginally above the minimum therapeutic level of 5ug/ml. Sixty- four percent (64%) of neonates in this arm of the study had steady state levels that were within the therapeutic range (5-15ug/ml), which was an improvement from the forty- one percent (41%) seen at peak levels.

The low serum theophylline levels observed in the rectal arm at peak and steady state could be attributed to some of the local factors that are known to reduce rectal aminophylline absorption. These factors include the formulation, dose, rectal faecal loading and defecation or expulsion of aminophylline post administration.[7,8]

Intravenous aminophylline administration achieves therapeutic theophylline levels faster. Fifty- nine percent (59%) of the neonates on IV aminophylline infusion arm achieved these levels within an hour (at peak). Unfortunately, it was associated with an increased risk of toxicity. Twenty-three percent (23%) of neonates in this arm had toxic theophylline levels at

steady state, which was significantly higher than the 5% seen in the rectal arm after the same period of aminophylline administration at the same dose and schedule.

At steady state, the percentage number of neonates with therapeutic theophylline levels was comparable in rectal and the IV arms (64%compared to 59%). However, the actual serum theophylline levels showed the rectal arm had a lower median of 6.61ug/ml compared to10.48ug/ml seen in the IV arm at steady state. This implies the bioavailability of theophylline is higher after IV aminophylline infusion.

Available literature recommends 5-15ug/ml as the therapeutic serum theophylline levels required in the management of AOP[1, 3, 7, 8]. Based on this recommendation, it would appear that the rectal route using this formulation might not be ideal in a neonate who already has apnoeic episodes and requires urgent relief of apnoea. However, it may be useful for prophylaxis of AOP in a neonate at risk. Apnoea of prematurity is known to occur from the 2nd to the 7th day of life and rarely on the first day[1,2,3,4] therefore starting rectal aminophylline administration using the IV formulation before apnoea occur might be able to prevent its occurrence. Furthermore, the rectal route is known to be safe and convenient in neonates[7, 8]. This becomes important especially where there is acute shortage of staff and unavailability of resources for IV aminophylline administration.

Intravenous aminophylline infusion is the ideal route[1, 2, 7, 8] however; this study shows there is an increased risk of toxicity associated with this route, which may restrict its use. Intravenous aminophylline administration requires proper equipment that can be used to effectively monitor serum theophylline in relation to resolution of apnoea or otherwise to guide on the dose adjustments. This is especially so in neonates who continue to be symptomatic despite being on aminophylline infusion or neonates who show signs of theophylline toxicity such as tachycardia, food intolerance and agitation to enable proper and appropriate dose adjustment therefore avoid undue toxicity. [7,8,20]

The relationship between serum theophylline levels achieved in this study and control or resolution of apnoea was not established because of limitation of resources. Establishment of this relationship is important especially in African preterm neonates since none of the studies that recommended 5-15ug/ml as serum levels required to relieve and prevent AOP was local. It is possible that this group of neonates may show a different response to theophylline given the wide interindividual variation in response, metabolism and elimination of theophylline that is thought to be both genetic and environmental[7, 8, 20].Therefore, there is a need for further evaluation of this relationship in African neonates to define levels that are able to relieve and control apnoea effectively in this group.

Other studies on rectal aminophylline administration include that of Lyon who used suppositories at 10 mgs /kg bodyweight loading and maintenance doses in forty-one preterm neonates with a mean gestational age of 28.3 weeks. He obtained a median steady state theophylline level of 8.0 µg/ml, which was higher than 6.96 µg/ml seen in this study. The higher dose together with formulation used by Lyon could explain the difference observed between his study and this study. [19]

Cooney used a specially prepared rectal enema in a small number of preterm neonates at a dose of 8.45 mgs /kg bodyweight loading and maintenance doses respectively. He obtained a median steady state theophylline level of 11 µg/ml[15], which was almost twice higher than the 6.96 µg/ml seen in this study. This marked difference observed between these two studies could be attributed to the different formulations and doses used. The higher median level Cooney obtained in his study may be attributed to the higher dose together with the formulation he used. It is possible the gel was better retained in the rectum with minimal expulsion therefore more aminophylline was available for absorption thus improving its bioavailability compared to the IV formulation used in this study, which is a solution and therefore, could be expelled easily.

These studies including present study suggest that aminophylline administered rectally as enema, suppository including the IV formulation is absorbed and that the rectal route may be useful in the management of AOP. However, more studies are required in this area to try to identify the best rectal formulation as well as the dose required to achieve better and effective serum theophylline levels that are able to control apnoea adequately.

Intravenous aminophylline infusion is well distributed and achieves therapeutic levels faster within an hour. However regular monitoring and adjustment of doses is necessary to minimise toxicity due to the wide individual variation in responds, metabolism and elimination of theophylline.[7, 8, 20] Hugo J.O in his study on therapeutic monitoring of serum theophylline after IV infusion in 106 term and preterm neonates with apnoea found 21 % of the neonates had toxic levels, 60% had therapeutic levels and a further 18% had sub therapeutic levels [20]. This finding was comparable to that seen in this study where 23 % of the neonates on IV aminophylline had toxic serum theophylline levels, 59% had therapeutic levels and another 18% had sub therapeutic levels. This shows the need to monitor neonates on IV aminophylline infusion closely for clinical signs of toxicity such as tachycardia, intolerance to feeds, agitation and if need be serum theophylline levels to avoid undue toxicity.

CONCLUSIONS

- Serum theophylline levels are sub therapeutic in most neonates 1 hr after rectal administration of the IV aminophylline formulation. Rectal arm had more neonates with sub therapeutic levels at steady state (after 72 hrs)
- Fifty-nine (59%) of the neonates in the IV arm had therapeutic serum theophylline levels, which was comparable to the 64% in the rectal arm after 72 hours of aminophylline administration.
- Neonates on aminophylline infusion were five times likely to have toxic serum theophylline levels at steady state compared to rectal aminophylline arm [OR 5.06 (95%CI1.21-24.29)]. Tachycardia was more prevalent after IV aminophylline infusion.

RECOMMENDATION

- From the findings of this study that showed IV aminophylline is absorbed rectally, a study is recommended to see whether increasing the doses of IV aminophylline formulation administered rectally would lead to an improvement in the number of neonates with therapeutic theophylline levels at peak and steady state.
- In view of the findings of this study that showed most neonates achieved therapeutic peak theophylline after aminophylline infusion, a study is recommended to see whether infusing loading doses followed by maintenance doses rectally (using the IV formulation) at the same doses and schedule as used in this study would lead to an improvement in the number of neonates would achieve therapeutic theophylline levels at peak and steady state
- Neonates on aminophylline infusion require serum theophylline levels need to be monitored frequently.

STUDY LIMITATIONS

- Inability to do serial aminophylline levels due to cost.
- The large amount of blood needed for serial serum theophylline determination could not be ethically obtained from the neonates.
- Micro techniques that could have been used to analyse the available small volumes were not available.
- Heart rates were taken twice-daily morning and evening after aminophylline administration due to the inability to monitor heart rates continuously as initially set out due to the absence of cardiac monitors.

REFERENCE

1. Stoll J.B. and Kliegman, R.M. Respiratory tract disorders. In: Behrman R.E. Kliegman R.M and Hal B.J. (eds.) - Nelson s textbook of paediatries 16th ed.W.B Saunders co 2000, pp 497-498.

2. Vyas H. and Milner D A,. Other respiratory diseases in the newborn. In: Roberton N.R.C (ed.)-Roberton textbook of neonatology 2nd ed. Churchill Livingstone co 1992pp329-333.

3. h.t.t.p://www.emedicine.com/paed/topic1157.htm

4. Papageorgiou A. and Bardin C. L,. Extremely low birth weight infant. In: Avery B.G,. Fletcher M. A,. Macdonald G.M. *(eds.)*-Neonatology: The Pathophysiology and management of the newborn 5th ed. Lippincott Williams and Wilkins co 1999 pp464-466

5. Simiyu D.E. Morbidity and Mortality of low birth weight infants in the newborn unit of KNH, Nairobi. EAMJ. 2004; **81**:367-374.

6. Pichler G., Urlesberger, B.and Muller W. Impact of bradycardia on cerebral blood oxygenation and cerebral blood volume during apnoea in preterm infants. PhysiolMeas: 2003; **24**: 671-680.

7. Parfitt K. .Bronchodilators and other anti-asthmatic drugs. In: Martindale, the complete drug reference 32nd ed. Pharmaceutical Press co 1999 pp748-754

8. Undem J.V.and Lichtenstein M.C,. Theophylline.In: Hardman G.J,. Limbird E.L. and Gilman A.G. (eds.) - The pharmacological basis of therapeutics 10th ed. McGraw hill co 2001 pp743-745.

9. Aranda J.V.and Sitar D.S. Pharmacokinetic aspects of theophylline in premature newborns. N.ENGL J. MED .1976; **295**:413-416

10. Jone R.A. and Baille E. Dosage schedule for intravenous aminophylline in apnoea of prematurity based on pharmacokinetic studies. Arch Dis Child.1979; **54**:190-193

11. Islam S.I. and Ali A.S.Pharmacokinetics of theophylline in preterm neonates during first month of life. Saudi Med J. 2004; **25**:459-465

12. Henderson-Smart D.J. Methylxanthine treatment for apnoea in preterm infants. Cochrane Database syst review **-01**; 2001:CD000140

13. Barbara Schmidt. Methylxanthine therapy in premature infants: Sound practise, disaster, or fruitless byway? J paed. 1999; **135**:367-374.

14. Lyon A.J. and McIntosh. Rectal aminophylline in the management of apnoea of prematurity. Arch Dis Child.1985; **60**:38-41

15. Cooney S. Rectal aminophylline gel in treatment of apnoea of prematurity. Lancet. 1991; **337**:1351.

16. Karlson M.O. Population pharmacokinetics of rectal theophylline in neonates. Ther Drug Monit.1991; **13:**195-200

17. Meme J.S .Low birth weights babies and neonatal mortality at KNH. *Masters of medicine in paediatrics and child health dissertation UON 1976.*

18. Kasirye B. Neonatal morbidity and mortality at KNH: A prospective study. Master *of medicine in paediatrics and child health dissertation UON 1984*

19. Armitage B.and Berryl G.The size of a statistical investigation. In: Statistical methods in medical research 2nd ed. Blackwell scientific publication1994 pp 357.

20. Hugo J.O. and Janett F.P.Therapeutic monitoring of theophylline in newborns with apnoea. Pharmacology and therapeutics. 2004; **29**:322-324.

21. Chang J.M., Goteher S.and Gushaw J.B. Homogenous enzyme immunoassay for theophylline in serum and plasma. Clinical Chem.1982; **28**:361-367

APPENDIX

APPENDIX I

STUDY SUBJECT CONSENT INFORMATION

Introduction

I am Dr Chumba, a postgraduate student, currently doing postgraduate studies in paediatrics and child health at University of Nairobi .As part of my postgraduate studies; I am required to do a research project. I intend to do a study on aminophylline, a drug that is used to manage breathing problems. The study requires comparing serum levels of aminophylline, after rectal administration and intravenous infusion. The doses and schedules to be used are the standard recommended doses. This drug is commonly used to manage apnoea of prematurity, a condition common in neonates weighing 1500 grammes and below or gestational age of 32 weeks and below. Your child weight and/or gestational age fall within this category. With your permission, I may need to include your child into the study.

About the study

Neonates delivered with a birth weight of 1500 grammes and below or gestational age of 32 weeks and below are prone to developing apnoea of prematurity (baby stopped breathing) a condition that is fairly common in this age group and birth weight. This condition is managed by giving the child aminophylline either through the rectal route, as intravenous infusion or as oral solution. At our newborn unit, it is administered as rectal enema since it is a safe and convenient way but can be given in through any of the other routes safely.

This study will involve removing from your baby a sample of 2 mls of blood on three occasions, the first, third and the fifth day after starting aminophylline, from the neonate. This blood will be used to assay serum aminophylline levels and a small amount stored for further analysis. Removal of blood may be associated with mild occasional discomfort such as pain, bleeding or infection. However some of these, are very rare complication e.g. infection. To minimize risk, extreme care will be taken when doing this procedure to ensure safety of the neonate. A sterile procedure will be performed with thorough cleaning and sterilization of the skin using spirit, prior to removing blood. After removal, gentle pressure will be applied to prevent/ stop any bleeding. Additionally a qualified person will do the procedure. The child's rights will be respected and confidentiality will be maintained at all times. No names will be mentioned in the study documents. Any useful information, which will improve the

quality of care and outcome of the baby, will be shared with the caregiver for appropriate action. It is important that you understand that participation in this study is voluntary and you can withdraw from the study at any time without losing any of your rights to care. You are now free to ask any questions relating to this study to get clarification on any issues that may not be clear to you. You may therefore decide to participate or not to participate in the study.

APPENDIX II

CONSENT FORM

Study no---------- date---------- time----------

I, have been adequately, explained about the study by Dr Chumba. I understand that my rights and the rights of my child will be respected and confidentiality maintained at all times. I also understand that this consent is voluntary and that I can withdraw from the study at any time without any penalties or loss of my child's rights to proper care. I therefore consent for my neonate to be recruited into the study

Mother's signature ---------- date----------

I have adequately explained to the mother of the neonate all the issues touching on this study and she has accepted the child to be recruited into the study.

Dr. Chumba

Investigators signature---------- date----------

For any issues you may contact

Dr. Chumba John at Tel 0720-430752

cc: subject file
Investigators file

APPENDIX III

DATA FORM

Neonatal Details.

1) Study no----------

2) Date of birth---------- time-of birth----------

3) Age (hours) ----------

4) Sex male---------- female----------

5) Weight grammes ----------

6) Date of admission ----------

7) Heart rate- baseline……day1……day 2……..day 3……

8) Rectal temperature – at admission…day1……day 2…..day 3…

9) Theophylline blood levels day1---------- day3.....

10) No apnoeic episodes recorded day 1----------day2---------- day3----------

APPENDIX IV

DAILY DATA COLLECTION SHEET

Study no----------

1hour After Morning dose	1hour after Evening dose
8.00AM	8.00PM

Day 1

Date----------

Rectal Temperature ^{0} C

Heart rate

Day2

Date----------

Rectal temperature ^{0} C

Heart rate

Day3

Date----------

-Rectal temperature ^{0} C

Heart rate

APPENDIX V

QUESTIONNAIRE FOR ASSESSING EFFECTIVENESS OF STAFF SENSITIZATION ON APNOEA IN NBU.

1) What is AOP?

2) What are the possible causes of apnoea?

3) What groups of babies are likely to be affected by apnoea?

4) What are the signs and symptoms of apnoea?

5) How do we manage apnoea episodes associated with AOP?

6) What are the likely complications of AOP?

APPENDIX VI

Random sampling Numbers

03	47	43	73	86	36	96	47	36	61	46	98	63	71	62	33	26	16	80	45	60	11	14	10	95
97	74	24	67	62	42	81	14	57	20	42	53	32	37	32	27	07	36	07	51	24	51	79	89	73
16	76	62	27	66	56	50	26	71	07	32	90	79	78	53	13	55	38	58	59	88	97	54	14	10
12	56	85	99	26	96	96	68	27	31	05	03	72	93	15	57	12	10	14	21	88	26	49	81	76
55	59	56	35	64	38	54	82	46	22	31	62	43	09	90	06	18	44	32	53	23	83	01	30	30
16	22	77	94	39	49	54	43	54	82	17	37	93	23	78	87	35	20	96	43	84	26	34	91	64
84	42	17	53	31	57	24	55	06	88	77	04	74	47	67	21	76	33	50	25	83	92	12	06	76
63	01	63	78	59	16	95	55	67	19	98	10	50	71	75	12	86	73	58	07	44	39	52	38	79
33	21	12	34	29	78	64	56	07	82	52	42	07	44	38	15	51	00	13	42	99	66	02	79	54
57	60	86	32	44	09	47	27	96	54	49	17	46	09	62	90	52	84	77	27	08	02	73	43	28
18	18	07	92	46	44	17	16	58	09	79	83	86	19	62	06	76	50	03	10	55	23	64	05	05
26	62	38	97	75	84	16	07	44	99	83	11	46	32	24	20	14	85	88	45	10	93	72	88	71
23	42	40	64	74	82	97	77	77	81	07	45	32	14	08	32	98	94	07	72	93	85	79	10	75
52	36	28	19	95	50	92	26	11	97	00	56	76	31	38	80	22	02	53	53	86	60	42	04	53
37	85	94	35	12	83	39	50	08	30	42	34	07	96	88	54	42	06	87	98	35	85	29	48	39
70	29	17	12	13	40	33	20	38	26	13	89	51	03	74	17	76	37	13	04	07	74	21	19	30
56	62	18	37	35	96	83	50	87	75	97	12	25	93	47	70	33	24	03	54	97	77	46	44	30
99	49	57	22	77	88	42	95	45	72	16	64	36	16	00	04	43	18	66	79	94	77	24	21	90
16	08	15	04	72	33	27	14	34	09	45	59	34	68	49	12	72	07	34	45	99	27	72	95	14
31	16	93	32	43	50	27	89	87	19	20	15	37	00	49	52	85	66	60	44	38	68	88	11	80
68	34	30	13	70	55	74	30	77	40	44	22	78	84	26	04	33	46	09	52	68	07	97	06	57
74	57	25	65	76	59	29	97	68	60	71	91	38	67	54	13	58	18	24	76	15	54	55	95	52
27	42	37	86	53	48	55	90	65	72	96	57	69	36	10	96	46	92	42	45	97	60	49	04	91
00	39	68	29	61	66	37	32	20	30	77	84	57	03	29	10	45	65	04	26	11	04	96	67	24
29	94	98	94	24	68	49	69	10	82	53	75	91	93	30	34	25	20	57	27	40	48	73	51	92
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31	96	25	91	47	96	44	33	49	13	34	86	82	53	91	00	52	43	48	85	27	55	26	89	62
66	67	40	67	14	64	05	71	95	86	11	05	65	09	68	76	83	20	37	90	57	16	00	11	66
14	90	84	45	11	75	73	88	05	90	52	27	41	14	86	22	98	12	22	08	07	52	74	95	80
68	05	51	18	00	33	96	02	75	19	07	60	62	93	55	59	33	82	43	90	49	37	38	44	59
20	46	78	73	90	97	51	40	14	02	04	02	33	31	08	39	54	16	49	36	47	95	93	13	30
64	19	58	97	79	15	06	15	93	20	01	90	10	75	06	40	78	78	89	62	02	67	74	17	33
05	26	93	70	60	22	35	85	15	13	92	03	51	59	77	59	56	78	06	83	52	91	05	70	74
07	97	10	88	23	09	98	42	99	64	61	71	62	99	15	06	51	29	16	93	58	05	77	09	51
68	71	86	85	85	54	87	66	47	54	73	32	08	11	12	44	95	92	63	16	29	56	24	29	48
26	99	61	65	53	58	37	78	80	70	42	10	50	67	42	32	17	55	85	74	94	44	67	16	94
14	65	52	68	75	87	59	36	22	41	26	78	63	06	55	13	08	27	01	50	15	29	39	39	43
17	53	77	58	71	71	41	61	50	72	12	41	94	96	26	44	95	27	36	99	02	96	74	30	83
90	26	59	21	19	23	52	23	33	12	96	93	02	18	39	07	02	18	36	07	25	99	32	70	23
41	23	52	55	99	31	04	49	69	96	10	47	48	45	38	13	41	43	89	20	97	17	14	49	17
60	20	50	81	69	31	99	73	68	68	35	81	33	03	76	24	30	12	48	60	18	99	10	72	34
91	25	38	05	90	94	58	28	41	36	45	37	59	03	09	90	35	57	29	12	82	62	54	65	60
14	50	57	74	37	98	80	33	00	91	09	77	93	19	82	74	94	80	04	04	45	07	31	66	49
15	22	04	39	43	73	81	53	94	79	33	62	46	86	28	08	31	54	46	31	53	94	13	38	47
49	79	13	77	48	73	82	97	22	21	05	03	27	24	83	72	89	44	05	60	35	80	39	94	88
38	75	80	18	14	22	95	75	42	49	39	32	82	22	49	02	48	07	70	37	16	04	61	67	87
40	96	23	70	00	39	00	03	06	90	55	85	78	38	36	94	37	30	69	32	90	89	00	76	33

APPENDIX VII

Dubowitz scoring system for gestational age assessment.

Physical maturity	-1	0	1	2	3	4	5
Skin	Sticky, friable, transparent	Gelatinous, red, translucent	Smooth pink, visible veins	Superficial peeling &/or rash, few veins	Cracking, pale areas, rare veins	Parchment, deep cracking, no vessels	Leathery, cracked, wrinkled
Lanugo	None	Sparse	Abundant	Thinning	Bald areas	Mostly bald	
Plantar surface	Heel–toe 40–50 mm:-1 <40 mm:-2	<50 mm, no crease	Faint red marks	Anterior transverse crease only	Creases ant. 2/3	Creases over entire sole	
Breast	Imperceptible	Barely perceptible	Flat areola, no bud	Stippled areola, 1–2 mm bud	Raised areola, 3–4 mm bud	Full areola, 5–10 mm bud	
Eye/ear	Lids fused loosely (-1), tightly (-2)	Lids open, pinna flat, stays folded	Slightly curved pinna soft; slow recoil	Well-curved pinna, soft but ready recoil	Formed & firm, instant recoil	Thick cartilage, ear stiff	
Genitals male	Scrotum flat, smooth	Scrotum empty, faint rugae	Testes in upper canal, rare rugae	Testes descending, few rugae	Testes down, good rugae	Testes pendulous, deep rugae	
Genitals female	Clitoris prominent, labia flat	Prominent clitoris, small labia minora	Prominent clitoris, enlarging minora	Majora & minora equally prominent	Majora large, minora small	Majora cover clitoris & minora	

Neuromuscular maturity

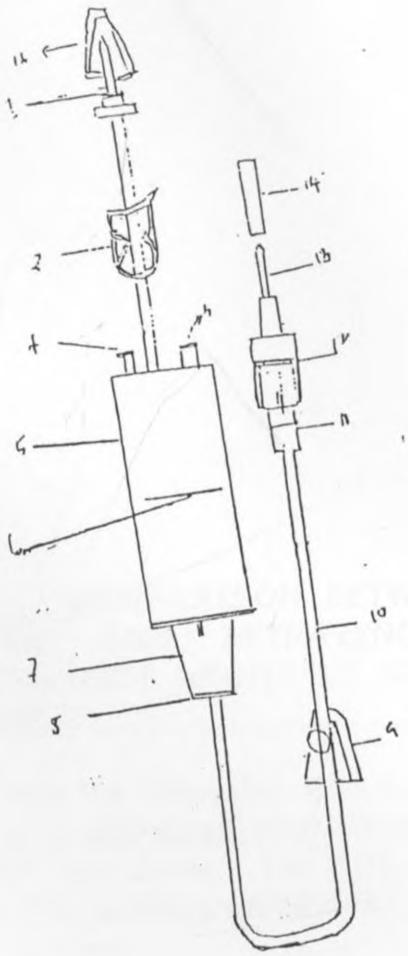
	-1	0	1	2	3	4	5
Posture							
Square window (wrist)	<90°	90°	60°	45°	30°	0°	
Arm recoil		180°	140–180°	110–140°	90–110°	<90°	
Popliteal angle	180°	160°	140°	120°	100°	90°	<90°
Scarf sign							
Heel to ear							

Maturity rating

Score	Weeks
-10	20
-5	22
0	24
5	26
10	28
15	30
20	32
25	34
30	36
35	38
40	40
45	42
50	44

APPENDIX VIII

Medivols infusion soluset (Measure volume fluid infusion set)



Labelling

1. Closure-piercing device
2. Clamp
3. Injection site into the burette
4. Air filter shut off device
5. Graduated burette
6. Shut-off valve
7. Drip chamber
8. Fluid filter
9. Flow regulator
10. Tubing
11. Injection site.
12. Male fitting
13. Insertion needle
14. Protective cap

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Ref: KNH-ERC/01/2806

Date: 16th June 2005

Dr. John Chumba
Department of Paediatrics
Faculty of Medicine
University of Nairobi

Dear Dr. Chumba

RESEARCH PROPOSAL: "COMPARISON BETWEEN SERUM THEOPHYLLINE LEVELS AFTER RECTAL AND INTRAVENOUS ADMINISTRATION OF AMINOPHYLLINE IN PRETERM NEONATES AT THE KENYATTA NATIONAL HOSPITAL NEWBORN UNIT" (P82/6/2005)

This is to inform you that the Kenyatta National Hospital Ethics and Research Committee has reviewed and **approved** your above cited research proposal for the period 16th June 2005 to 15th June 2006. You will be required to request for a renewal of the approval if you intend to continue with the study beyond the deadline given.

On behalf of the Committee, I wish you fruitful research and look forward to receiving a summary of the research findings upon completion of the study.

This information will form part of database that will be consulted in future when processing related research study so as to minimize chances of study duplication.

Yours sincerely,

PROF. A. N. GUANTAI
SECRETARY – KNH-ERC

c.c: Prof. K. M Bhatt, Chairperson, and KNH-ERC
The Deputy Director (C/S), KNH
The Dean, Faculty of Medicine, UON
The Chairman, Department of Paediatrics, UON
The Supervisors: Prof. A. Wasunna, Kemri (UON)
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