

CORPUS CALLOSUM SIZE ON MAGNETIC RESONANCE IMAGING, A SURROGATE FOR WHITE MATTER VOLUME LOSS IN CHILDREN IMAGED FOR THE CHRONIC SEQUELAE OF NEONATAL HYPOXIC ISCHAEMIC ENCEPHALOPATHY

Dr Tamarin Nell

A research report submitted to the Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, in partial fulfilment of the requirements for the degree of Master of Medicine in Diagnostic Radiology

Johannesburg, 2021

Declaration

I, Tamarin Nell, declare that this research report is my own work. It is being submitted for the degree of MMed (RadD) at the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at this or any other University.

A handwritten signature in black ink, appearing to be 'T. Nell', written in a cursive style.

Tamarin Chantal Nell

On this 15th day of November 2021

To Andre, completing this would not have been possible without you.

Publications and presentations

The article has been submitted for publication to *South African Journal of Radiology* (SAJR). Reference number: 2328.

It has never been presented at a congress.

Abstract

INTRODUCTION: The corpus callosum (CC) links the fibres of the cerebral hemispheres and is a reflection of the cerebral hemisphere volumes. Neonatal hypoxic ischaemic encephalopathy (HIE) results in injuries to the grey and white matter in the brain. MRI is the best imaging technique to assess these injuries, and closely correlates with the clinical findings.

AIM: To objectively assess the corpus callosum spatial parameters via MRI in children with and without reported features of Neonatal HIE as a possible surrogate to white matter volume loss.

METHOD: MRI images of children scanned for clinically suspected HIE were retrospectively reviewed and classified into MRI HIE patterns, by a blinded majority consensus reading of 3 Paediatric radiologists. Corpus callosum measurements in the non-HIE imaging pattern and HIE imaging patterns were compared. Thickness of the rostrum, genu, body and splenium were made, as well as the anteroposterior length and volume of the corpus callosum.

RESULTS: A total of 115 MRI scans were analysed and the corpus callosums were measured, where 41.7% demonstrated no MRI features of HIE and 58.% demonstrated an MRI pattern suggestive of HIE. The rostrum thickness, the genu thickness and CC volume between the categories showed a significant statistical difference ($p = 0.00016$, $p = 0.00011$, $p = 0.03754$ respectively).

CONCLUSIONS: CC thickness, particularly in the anterior regions, has shown promise of being a surrogate measure for white matter volume loss in children imaged for the chronic sequelae of hypoxic ischaemic encephalopathy. Further studies with larger sample sizes would be important in HIE.

KEYWORDS: Hypoxic ischaemic encephalopathy, corpus callosum, MRI

Acknowledgements

- My sincerest gratitude to Professor Nasreen Mahomed. Your unwavering support means the world to me. This accomplishment would not have been possible without your kindness and understanding.
- Special thanks to Dr Liam Lorentz for giving me permission to use his ready established database, as well as Professor Nasreen Mahomed, Dr Tanyia Pillay and Dr Halvani Moodley for their contributions to this database, as a part of the original study undertaken by Dr Liam Lorentz.
- Thank you to my amazing family for all your support along this journey.

Submission Format

This research report, for the purposes of Master in medicine, Department of Radiology has been drafted in the *Submissible Format* as per University of the Witwatersrand guidelines.

The original research article is written in the manuscript style required by the journal entitled *SA Journal of Radiology* with the intention to publish.

Requirements outlined by *SA Journal of Radiology* for original research include:

- Word count: 3000 words
- Tables/Figures: not more than 10 Tables/Figures
- Structured abstract: 250 words
 - Background
 - Objectives
 - Methods
 - Results
 - Conclusion
- Headings:
 - Introduction
 - Methods
 - Results
 - Discussion
 - Conclusion
 - Acknowledgments
 - References
- References: 40 or less
- Ethical statement should be included in the manuscript
- Compulsory supplementary file: ethical clearance letter/certificate
- The article was written with British English spelling
- Referencing done in Wits Vancouver style

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List of Abbreviations and Terminology

AP - Anteroposterior

APGAR - Activity, Pulse, Grimace, Appearance, Respiratory Score

CC - Corpus callosum

CP - Cerebral palsy

CT - Computed tomography

CMJAH - Charlotte Maxeke Johannesburg Academic Hospital

DICOM - Digital imaging and communication in medicine

HIE - Hypoxic ischaemic encephalopathy

HII - Hypoxic ischaemic injury

HIV - Human immunodeficiency virus

MR - Magnetic resonance

MRI - Magnetic resonance imaging

ROI - Region of interest

Study Protocol

Corpus Callosum Size on Magnetic Resonance Imaging, a surrogate for white matter volume loss in children imaged for the Chronic Sequelae of Neonatal Hypoxic Ischaemic Encephalopathy

Investigator: Dr. T. Nell

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1. Background Literature Review

1.1. Literature Review

The corpus callosum (CC) links the fibres of the cerebral hemispheres with each other. As such, CC thickness “mirrors the volumes of the cerebral hemispheres”, and responds to changes that occur to the cerebrum [1,2]. CC thickness can be useful to assess pathology in paediatric central nervous system disease. However, the thickness should be considered in the context of the age of the patient, as the CC changes with age as a part of a child’s normal development [1].

Luders et al emphasises that as a reliable marker for assessing the normal thickness of the CC during childhood, is by assessing thickness of the rostrum, as this is the portion of the CC least affected by the age of the patient [3].

From studies as early as 1988, MRI has been the imaging modality of choice in the assessment of the corpus callosum [4]. MR imaging in the sagittal plane, allows for the best visualisation of the corpus callosum [2].

There have been very few studies done on what is normal and abnormal CC measurements in the paediatric population [1].

The first study on CC size in the paediatric population in 2011 by Gavel et al., produced the first reference range for CC size in a normal paediatric population [5]. In 2014, Andronikou et al., did the first study using CC measurements on Magnetic Resonance Imaging (MRI) in South African children. In this early study, Andronikou and his team were the first to correlate CC thickness as a marker of brain volume [6].

Thereafter, in 2015, Andronikou et al. correlated CC thickness with Human Immunodeficiency Virus (HIV)-related brain disease and normal controls. In this pilot study, there were small patient numbers with only 48 patients reviewed [7].

These two studies, with albeit small patient numbers, did demonstrate that the thickness of the CC is affected by peripheral white matter loss, thus making it a possible surrogate for white matter atrophy [6,7].

There are many factors that may cause CC thinning. These include primary conditions such as hypomyelinating conditions, metabolic conditions and microcephaly, as well as, secondary causes such as hypoxic-ischaemic encephalopathy (HIE), HIV encephalopathy, hydrocephalus, dysmyelinating and demyelinating conditions [1,4,8].

HIE is a major contributor to death and cerebral palsy in children [9]. It is known to cause grey and white matter changes in the paediatric population. There are various MRI patterns of HIE depending on the duration and severity of the hypoxic insult, as well gestational age at the time of the insult [10]. MRI changes correlate closely to the clinical findings following HIE [10]. Kazi et al. described in 2013, that ischaemia due to hypoxia in the neonatal period results in a typical callosal infarction pattern, which when imaged in the chronic stages, is visualised as CC atrophy [11].

There have been studies done to assess the patterns of injury [9,10], as well as using total brain tissue volumes to prognosticate, and assessment post hypothermia treatment in HIE[12].

1.2. Tabulated Literature Review

Article Title	Journal, Year and Author	Purpose	Key Messages	Limitations	Relevance
“Biometry of the corpus callosum in children: MR imaging reference data” [5]	AJNR 2011 Garel, C., Cont, I., Alerti, C., Josserand, E., Moutard, M.L. and Ducou le Pointe, H.	To present a reference biometry of the corpus callosum in MRI of a large cohort of children	* Demonstrated the CC changes throughout infancy, childhood and up to adolescence * Provided biometric data on CC	Study done in France, Europe only	Provided the first reference of biometric data on CC in children. Predicted that in future that a possible correlation between CC biometry and morphology with the clinical status of children with disorders possibly associated with CC abnormalities
“Corpus callosum thickness in children: an MR pattern-recognition approach on midsagittal image” [1]	Pediatric Radiology 2015 An-dronikou, S., Pillay, T., Gabuza, L., Mahomed, N., Naidoo, J., Hlabangana, L.T., du Plessis, V. and Prabhu, S.P.	Demonstrate age-related development as well as primary and acquired causes of changes in the CC size	* Anatomically there are 4 parts * Thickness of the CC reflects the number of fibres or degree of myelination * Thickness reflects the volume of the cerebral hemispheres * Increasing thickness of the CC is an important feature of development * Failure to gain thickness or thinning of the CC may be the result of multiple diseases		Luders et al. - most reliable rule of thumb when assessing the normal thickness of the CC during childhood should be to assess the thickness of the rostrum as this is the portion of the CC least affected by age

“Correlating brain volume and callosal thickness with clinical and laboratory indicators of disease severity in children with HIV-related brain disease” [6]	Childs Nerv System 2014 Andronikou, S., Ackermann, C., Laughton, B., Cotton, M., Tomazos, N., Spottiswoode, B., Mauff, K. and Pettifor, J.M.	Determine if brain volume and midsagittal CC linear measurements on MRI in children with HIV-related brain disease correlate with the disease severity based on clinical and laboratory findings	* Brain volume did not show any significant correlations with clinical or laboratory findings * Linear CC measurements on MRI may be an early biomarker of HIV-related brain disease, prior to MRI evident white matter atrophy	Small patient numbers (33) Technical factors involved in measuring CC - section of the midline and point deposition are operator dependent	Thickness of the CC is affected by peripheral white matter loss, thus making it a possible useful surrogate for white matter atrophy
“Corpus callosum thickness on mid-sagittal MRI as a marker of brain volume: a pilot study in children with HIV-related brain disease and controls” [7]	Pediatric Radiology 2015 Andronikou, S., Ackermann, C., Laughton, B., Cotton, M., Tomazos, N., Spottiswoode, B., Mauff, K. and Pettifor, J.M.	Determine if thickness of CC on mid-sagittal MRI is a surrogate marker of brain volume in children with HIV-related brain disease and in controls with out HIV	* Anterior prefrontal portion of CC best marker of normality due to early development and stability with age * Based on controls - median prefrontal CC thickness in African patients of 9 mm * Many factors may cause CC thinning	Small patient numbers (48) Children < 5 years only Technical factors involved in measuring CC - section of the midline and point deposition are operator dependent	Thickness of the CC is affected by peripheral white matter loss, thus making it a possible useful surrogate for white matter atrophy
“MRI evaluation of pathologies affecting the corpus callosum: a pictorial essay” [8]	Indian J Radiol Imaging 2013 Kazi, A.Z., Joshi, P.C., Kelkar, A.B., Mahajan, M.S. and Ghawate, A.S.				Ischaemia due to hypoxia, especially in the neonatal period produces a typical diffuse callosal infarct pattern which in the chronic stages is seen as gliosis or atrophy

“The development of the corpus callosum in the healthy human brain” [3]	J Neurosci 2010 Luders,E. , Thompson, P.M. and Toga, A.W.		* Significant correlations between callosal thickness and age * Significant differences in callosal thickness between the youngest and the oldest groups * Temporally distinct groups * Handedness effects		Most reliable rule-of-thumb thickness measure of CC normality during childhood that is least affected by age would be the rostrum,
“MR imaging of the corpus callosum: normal and pathologic findings and correlation with CT” [4]	AJNR 1988 Reinartz, S.J., Coffman, C.E., Smoker, W.R.K., and Godersky, J.C.	To demonstrate that MR should be the primary imaging technique for the evaluation of corpus callosal disease	* MR has proved invaluable for evaluating the CNS * MR allows for the visualization and accurate measurement of midline sagittal structures such as the corpus callosum		MR should be the primary imaging technique for the evaluation of the corpus callosal
“MR imaging of the corpus callosum” [2]	AJR 1993 Georgy, B.A., Hesselink, J.R. and Jernigan, T.L.	To review the normal anatomy, development and process of demyelination of the corpus callosum			With MR imaging in the sagittal plane, the corpus callosum can be depicted in great detail

1.3. Limitations of the literature review

Although studies have been done on brain volume and CC thickness, there are no studies correlating CC volume with degree of brain volume loss in a paediatric setting.

Majority of the studies done, have only been studied with small patient numbers.

2. Rationale

The corpus callosum thickness “mirrors the volumes of the cerebral hemispheres”, and responds to changes that occur to the cerebrum.

There has been limited studies done in assessing the corpus callosum in the paediatric population.

There have especially been very limited studies done in a South African population. Quantitatively assessing a South African population for normal and abnormal CC sizes could be a useful parameter for South African Radiologists.

Currently CC measurements have been limited to thickness and anteroposterior length. However, volume assessment may be an easy adjunct to add to these measurements.

CC thickness can be useful to assess pathology in the CNS. To date, there has been limited research done in measuring the corpus callous in the paediatric population with known white matter abnormalities.

Can corpus callosum size be used as a surrogate for the assessment of white matter disease in the paediatric population?

Assessment of CC sizes in a paediatric population in whom MR imaging was done because of suspected HIE, which is a known cause of white matter abnormalities.

3. Aim

To describe the corpus callosum thickness, length and volume on MRI as a surrogate for white matter volume loss in children imaged for the chronic sequelae of neonatal hypoxic ischaemic encephalopathy.

4. Study Objectives

Primary Objective: To objectively assess the corpus callosum size on MRI in children with and without reported features of Neonatal Hypoxic Ischaemic Encephalopathy

Secondary Objective: To compare the 2 groups to see if there is a statistical significant difference

5. Methods

5.1. Research paradigm

The research paradigm was a retrospective cross-sectional study. Measuring, calculating and comparing corpus callosum sizes from a database of children who presented for an MRI based on a clinical history of HIE, whose MRI scans demonstrated normal findings or findings in keeping with HIE-related brain disease.

5.2. Population to be studied

The study population was from an established database principle investigator Dr Liam Lorentz for his MMed “A Descriptive Study of the Magnetic Resonance Imaging Findings of Hypoxic Ischaemic Injury at a Tertiary Academic Hospital”.

Permission has been obtained from the principle investigator to use this database.

The inclusion and exclusion criteria for this study were as follows:

Inclusion criteria (as applied in the original research):

- MRI examinations performed for children younger than 15 years of age during the period: 1 January 2014 to 31 December 2015.

- MRI examinations performed for children where the indication was for:
 - Suspected HIE or hypoxic ischaemic injury (HIE), including poor condition at birth: APGAR score (< 5), major resuscitation at birth, neonatal encephalopathy, altered level of consciousness
 - Cerebral palsy
 - Delayed milestones
 - Seizures
 - Cognitive or visual impairment
 - Focal neurological deficit, including tone, motor, reflexes, speech, feeding and movement abnormalities

Exclusion criteria (as applied in the original research)

- MRI examinations for children where the clinical history indicated:
 - Congenital neurological anomalies
 - Central nervous system tumours
 - Chromosomal abnormalities
 - Neurocutaneous syndromes
 - Head trauma
 - Infections
 - Mitochondrial and metabolic disorders
 - Toxin exposure

Principal investigator Dr Liam Lorentz collected the MRIs from January 2014 to December 2015, the clinical and laboratory data. Three Paediatric Radiologists (NM, HM, TP) interpreted the MRIs independently, blinded to all clinical data except age of child.

Myelination maps were provided for different age groups.

2 out of 3 consensus majority reading used at the data analysis phase.

5.3 Materials and Methods

This already established database of images was accessed using a DICOM viewer used on an Apple computer, Horos. Tools on Horos were used to measure corpus callosum thickness, length and volume for each patient.

Measurements of the CC thickness were done, in the mid-sagittal plane of the T1 weighted MR image, at predetermined anatomical levels - namely - rostrum, genu, body and splenium - using the caliper tool.



Fig 1. CC thickness measuring points

Measurements of the CC length were done, in the mid-sagittal plane of the T1 weighted MR image, from anterior to posterior, using the caliper tool.



Fig 2. CC length measurement from anterior to posterior

The volume of the CC was calculated using the pencil ROI Volume tools on the DICOM viewer. The outline of the CC was traced using the pencil tool, in the mid-sagittal plane, as well as 1-2 slices on either side of the mid-sagittal plane. Using the ROI volume tools, the volume was calculated, and averaged out, using the Compute Volume tool.

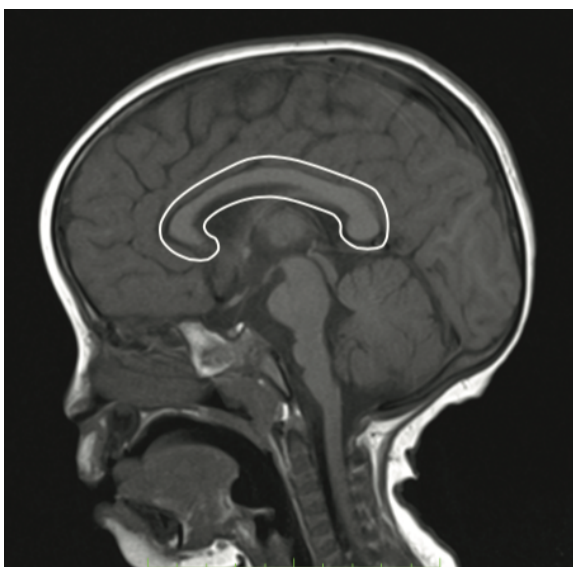


Fig 3. ROI online to calculate CC volume

Example of the final result after calculating the volume:

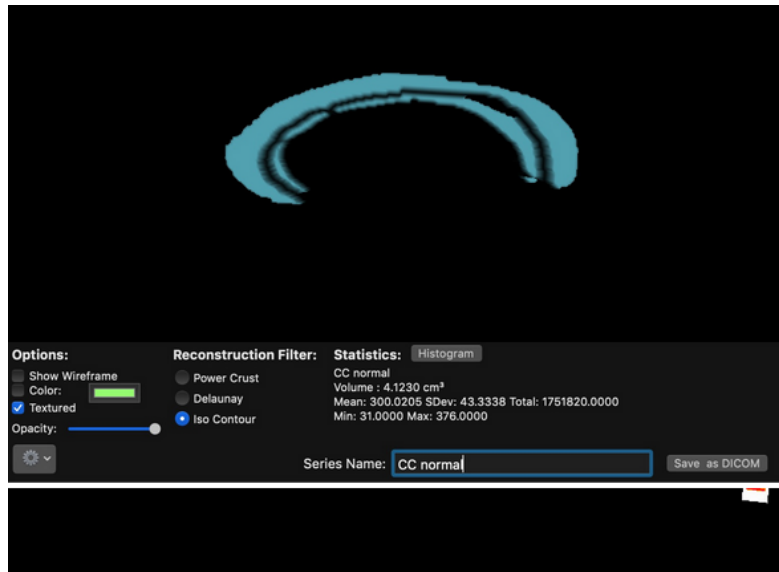


Fig 4. Example of completed volume calculation

Measurements were calculated using the sagittal T1-weighted imaging of each patient, as this is the best sequence for the visualisation of the corpus callosum.

All findings were recorded on a data collection sheet.

These were then charted on to a Microsoft Excel spreadsheet.

The data collection sheet and spreadsheet represent measurements and corpus callosum volumes in the normal and HIE-related brain disease paediatric population for pre-established ages categories.

5.4 Data Collection

Data collected as follows:

- Age of the patient

Age Stratification
Neonate (< 6 months)
6 - 23 months
2-5 years
> 5 years

- Clinical status
- MRI findings: with/without HIE MRI findings
- Corpus callosum measurements:
 - Thickness at different levels:
 - Rostrum
 - Genu
 - Body
 - Splenium
 - Length
 - Total volume

See Appendix I for “Data Collection Sheet”

5.5. Reliability and validity

All corpus callosum measurements and volume calculations were performed using the same DICOM viewer and tools, using the same MRI sequence, in the same manner, by the Principal investigator in a reproducible manner to ensure reliable results.

The number of patients available in the database is higher than in any other previous studies done on corpus callosum sizes in a paediatric population.

5.6. Bias

Personal bias is a possible concern as all measurements will be performed by a single investigator. However, all measurements are quantifiable and objective, and do not make use of any subjective variables.

5.7 Limitations

The study cohort to be used is limited. However, this study will lay the groundwork for further studies in this field.

HIE causes grey and white matter loss. Grey matter loss can be measured with the assessment of the thalamus and basal ganglia, however, this is not the purpose of this study.

6. Data analysis and statistics

Data from the “Data Collection Sheet” was captured on a Microsoft Excel spreadsheet by the Primary Investigator. Statistical Analysis using SAS Analytics University Edition version 9.2 were performed.

Means and standard deviation or medians and percentages were calculated for the numerical data.

A Two-sample *t*-test such as the Mann-Whitney test was used.

A significance level (*p*-value) of less than 0.05 was used.

7. Ethics

The study population is from an established database that principal investigator Dr Liam Lorentz compiled for his MMed “A Descriptive Study of the Magnetic Resonance Imaging Findings of Hypoxic Ischaemic Injury at a Tertiary Academic Hospital” - Certificate Number M170516. The supervisor of this study was Prof Nasreen Mahomed. Ethics approval was obtained as a sub-study of M170516 and approved in August 2020, by the Human Ethics Committee (Medical), of the University of the Witwatersrand - Ethics certificate M200781.

See Appendices II and III.

7.1. Data safety

All data had already been collected and anonymised by allocating a random number code to each patient by Dr Liam Lorentz. Permission was obtained from Dr Liam Lorentz for the use of this database. Only the age of the patient, the final report of the imaging findings and the actual images were made available to the primary investigator and supervisor. Data continues to be stored on two separate and secure devices, the location of which will only be known by the primary investigator.

8. Timing

Month of the Year	1	2	3	4	5	6
Literature search	Dec 2019					
Reading literature	Dec 2019					
Summarising literature	Dec 2019					
Preparing Protocol		Jan 2020				
Protocol Assessment			Feb 2020			
Ethics application			Feb 2020			
Collecting data				March 2020		
Data analysis				March 2020		
Writing up thesis					April 2020	
Submit: marking					30 Apr 2020	
Writing up paper						May 2020

9. Budget

Printing	R200
Stationary	R100
Travel	R500
Total	R800

This project will be self-funded.

10. Anticipated problems

No problems are anticipated.

11. References

- (1) Andronikou, S., Pillay, T., Gabuza, L., Mahomed, N., Naidoo, J., Hlabangana, L.T. *et al* 2014. Corpus callosum thickness in children: an MR pattern-recognition approach on the midsagittal image. *Pediatr Radiol* 45:258-272 DOI 10.1007/s00247-014-2998-9
- (2) Georgy, B.A., Hesselink, J.R. & Jernigan, T.L. 1992. MR imaging of the corpus callosum. *AJR* 1993; 160: 949-955 DOI 0361-803X/93/1605-0949
- (3) Luders, E., Thompson, P.M. & Toga, A.W. 2010. The development of the corpus callosum in the healthy human brain. *J Neurosci.*, August 18,2010; 30(33):10985-10990 DOI 10.1523/JNEUROSCI.5122-09.2010
- (4) Reinarz, S.J., Coffman, C.E., Smoker, W.R.K., & Godersky, J.C. 1988. MR imaging of the corpus callosum: normal and pathologic findings and correlation with CT. *AJNR* 9: July/August 1988; 649-656 DOI 0195-6108/88/0904-0649
- (5) Garel, C., Cont, I., Alberti, C., Josserand, E., Moutard, M.L. & Ducou le Pointe, H. 2011. Biometry of the corpus callosum in children: MR imaging reference data. *AJNR* 32:1436-1443 <http://dx.doi.org/10.3174/ajnr.A2542>
- (6) Andronikou, S., Ackermann, S., Laughton, B., Cotton, M., Tomazos, N., Spottiswoode, B. *et al* 2014. Correlating brain volume and callosal thickness with clinical and laboratory indicators of disease severity in children with HIV-related brain disease. *Childs Nerv Syst* 30:1549-1557 DOI 10.1007/s00381-014-2434-3
- (7) Andronikou, S., Ackermann, S., Laughton, B., Cotton, M., Tomazos, N., Spottiswoode, B. *et al* 2015. Corpus callosum thickness on mid-sagittal MRI as a marker of brain volume: a pilot study in children with HIV-related brain disease and controls. *Pediatr Radiol* 45:1016-1025 DOI 10.1007/s00247-014-3255-y
- (8) Krupa, K. & Bekiesinska-Figatowska, M. 2013. Congenital and acquired abnormalities of the corpus callosum: a pictorial essay. *Biomed Research International Volume 2013*, Article ID 265619 <http://dx.doi.org/10.1155/2013/265619>
- (9) Schroff, M.M., Soares-Fernandes, J.P., Whyte, H. & Raybaud, C. 2010. MR imaging for diagnostic evaluation of encephalopathy in the newborn. *RadioGraphics* 2010; 30:763-780 DOI 10.11.48/rg.303095126
- (10) Krishan, P. & Strof, M. 2016. Neuroimaging in neonatal hypoxic ischemic encephalopathy. *Indian J Pediatr* 2016; 83(9):995-1002 DOI 10.1007/s12098-016-2042-1

- (11) Kazi, A.Z., Joshi, P.C., Kelkar, A.B., Mahajan, M.S. & Ghawate, A.S. 2013. MRI evaluation of pathologies affecting the corpus callosum: a pictorial essay. *Indian J Radiol Imaging Oct-Dec*; 23(4):321-332 DOI 10.4103/0971-3026.125604
- (12) Parikh, N.A., Lasky, R.E, Garza, C.N., Bonfante-Mejia, E., Shankaran, S. & Tyson, J.E. 2009. Volumetric and anatomic MRI for hypoxic-ischemic encephalopathy: relationship to hypothermia therapy and neurosensory impairments. *J Perinatol* 2009 February; 29(2):143-149 DOI 10.1038/jp.2008.184

12. Appendices

Appendix I

Data Collection Sheet

Study number: _____

Patient age at time of MR:

Neonate

Less than 6 months

7 to 23 months

2 to 5 years

More than 5 years

Clinical Status:

MRI findings as per Dr Lorentz study:

No HIE related changes OR HIE related changes

Corpus callosum measurements:

Thickness (in cm)

- Rostrum _____

- Genu _____

- Body _____

- Splenium _____

Length (in cm) _____

Volume (cm³) _____

Appendix II



R14/49 Dr Liam Lorentz

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)

CLEARANCE CERTIFICATE NO. M170516

NAME: Dr Liam Lorentz
(Principal Investigator)
DEPARTMENT: Radiology
Charlotte Maxeke Johannesburg Academic Hospital

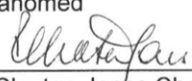
PROJECT TITLE: A Descriptive Study of the Magnetic Resonance Imaging Findings of Hypoxic Ischaemic Injury at a Tertiary Academic Hospital

DATE CONSIDERED: 26/05/2017

DECISION: Approved unconditionally

CONDITIONS:

SUPERVISOR: Dr Nasreen Mahomed

APPROVED BY: 
Professor P. Cleaton-Jones Chairperson, HREC (Medical)

DATE OF APPROVAL: 13/07/2017

This clearance certificate is valid for 5 years from date of approval. Extension may be applied for.

DECLARATION OF INVESTIGATORS

To be completed in duplicate and **ONE COPY** returned to the Research Office Secretary in Room 10004, 10th floor, Senate House/3rd floor, Phillip Tobias Building, Parktown, University of the Witwatersrand. I/We fully understand the conditions under which I am/we are authorised to carry out the above-mentioned research and I/we undertake to ensure compliance with these conditions. Should any departure be contemplated, from the research protocol as approved, I/we undertake to resubmit to the Committee. **I agree to submit a yearly progress report.** The date for annual re-certification will be one year after the date of convened meeting where the study was initially reviewed. In this case, the study was initially reviewed May and will therefore be due in the month of May each year. Unreported changes to the application may invalidate the clearance given by the HREC (Medical).

Principal Investigator Signature _____

Date _____

PLEASE QUOTE THE PROTOCOL NUMBER IN ALL ENQUIRIES

Appendix III

UNIVERSITY OF THE
WITWATERSRAND
JOHANNESBURG



HUMAN RESEARCH ETHICS
COMMITTEE (MEDICAL)

Office of the Deputy Vice-Chancellor (Research & Post Graduate Affairs)

TO: Dr T Nell
School of Clinical Medicine
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Division of Diagnostic Radiology
Medical School
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CC: Supervisor: Professor N Mahomed <Nasreen.Mahomed@wits.ac.za>
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FROM: Iain Burns
Human Research Ethics Committee (Medical)
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E-mail: Iain.Burns@wits.ac.za

DATE: 2020/08/25

REF: R14/49

PROTOCOL NO: **M200781** (*This is your ethics application study reference number. Please quote this reference number in all correspondence relating to this study*)

PROJECT TITLE: *Corpus callosum size on Magnetic Resonance Imaging, a surrogate for white matter volume loss in children imaged for the chronic sequelae of neonatal hypoxic ischaemic encephalopathy*

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

MSWorks2000/Iain0007/Clearscan.wps



R14/49 Dr T Nell

**HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)
CLEARANCE CERTIFICATE NO. M200781**

NAME: Dr T Nell
(Principal Investigator)

DEPARTMENT: School of Clinical Medicine
Department of Radiation Sciences
Division of Diagnostic Radiology
Medical School
University

PROJECT TITLE: Corpus callosum size on Magnetic Resonance Imaging, a surrogate for white matter volume loss in children imaged for the chronic sequelae of neonatal hypoxic ischaemic encephalopathy

DATE CONSIDERED: Ad hoc

DECISION: Approved unconditionally

CONDITIONS: Sub-study under M170516

SUPERVISOR: Professor N Mahomed

APPROVED BY: 
Dr CB Penny, Chairperson, HREC (Medical)

DATE OF APPROVAL: 2020/08/25

This clearance certificate is valid for 5 years from the date of approval. Extension may be applied for.

DECLARATION OF INVESTIGATORS

To be completed in duplicate and **ONE COPY** returned to the Research Office Secretary on the 3rd Floor, Phillip Tobias Building, Parktown, University of the Witwatersrand, Johannesburg.
I/we fully understand the conditions under which I am/we are authorized to carry out the above-mentioned research and I/we undertake to ensure compliance with these conditions. Should any departure be contemplated, from the research protocol as approved, I/we undertake to submit details to the Committee. **I agree to submit a yearly progress report.** When a funder requires annual re-certification, the application date will be one year after the date when the study was initially reviewed. In this case, the study was initially reviewed in **July** and will therefore reports and re-certification will be due early in the month of **July** each year. Unreported changes to the application may invalidate the clearance given by the HREC (Medical).


Principal Investigator Signature

Date

Article for submission

1. Title page

Corpus Callosum Size on Magnetic Resonance Imaging, a surrogate for white matter volume loss in Children Imaged for the Chronic Sequelae of Neonatal Hypoxic Ischaemic Encephalopathy

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2. Abstract

INTRODUCTION: The corpus callosum (CC) links the fibres of the cerebral hemispheres and is a reflection of the cerebral hemisphere volumes. Neonatal hypoxic ischaemic encephalopathy (HIE) results in injuries to the grey and white matter in the brain. MRI is the best imaging technique to assess these injuries, and closely correlates with the clinical findings.

AIM: To objectively assess the corpus callosum spatial parameters via MRI in children with and without reported features of Neonatal HIE as a possible surrogate to white matter volume loss.

METHOD: MRI images of children scanned for clinically suspected HIE were retrospectively reviewed and classified into MRI HIE patterns, by a blinded majority consensus reading of 3 Paediatric radiologists. Corpus callosum measurements in the non-HIE imaging pattern and HIE imaging patterns were compared. Thickness of the rostrum, genu, body and splenium were made, as well as the anteroposterior length and volume of the corpus callosum.

RESULTS: A total of 115 MRI scans were analysed and the corpus callosums were measured, where 41.7% demonstrated no MRI features of HIE and 58.% demonstrated an MRI pattern suggestive of HIE. The rostrum thickness, the genu thickness and CC volume between the categories showed a significant difference ($p = 0.00016$, $p = 0.00011$, $p = 0.03754$ respectively).

CONCLUSIONS: CC thickness, particularly in the anterior regions, has shown promise of being a surrogate measure for white matter volume loss in children imaged for the chronic sequelae of hypoxic ischaemic encephalopathy. Further studies with larger sample sizes would be important in HIE.

KEYWORDS: Hypoxic ischaemic encephalopathy, corpus callosum, MRI

3. Introduction

The corpus callosum (CC) links the fibres of the cerebral hemispheres with each other. As such, CC thickness “mirrors the volumes of the cerebral hemispheres”, and responds to changes that occur to the cerebrum [1,2]. CC thickness can be useful to assess pathology in paediatric central nervous system disease. However, the thickness should be considered in the context of the age of the patient, as the CC changes with age as a part of a child’s normal development [1].

Luders et al emphasises that a reliable marker for assessing the normal thickness of the CC during childhood, is by assessing thickness of the rostrum, as this is the portion of the CC least affected by the age of the patient [3].

From studies as early as 1988, MRI has been the imaging modality of choice in the assessment of the corpus callosum [4]. MR imaging in the sagittal plane, allows for the best visualisation of the corpus callosum [2].

There have been very few studies done on what is normal and abnormal CC measurements in the paediatric population [1].

The first study on CC size in the paediatric population in 2011 by Garel et al., produced the first reference range for CC size in a normal paediatric population [5]. In 2014, Andronikou et al., did the first study using CC measurements on Magnetic Resonance Imaging (MRI) in South African children. In this early study, Andronikou and his team were the first to correlate CC thickness as a marker of brain volume [6].

Andronikou et al. correlated CC thickness with Human Immunodeficiency Virus (HIV)-related brain disease and normal controls. In this pilot study, there were small patient numbers with only 48 patients reviewed [7].

These two studies, with albeit small patient numbers, did demonstrate that the thickness of the CC is an important marker for peripheral white matter loss, thus making it a possible surrogate for white matter atrophy [6,7].

There are many factors that may cause CC thinning. These include primary conditions such as hypomyelinating conditions, metabolic conditions and microcephaly, as well as, secondary causes such as hypoxic-ischaemic injury (HII) or encephalopathy (HIE), HIV encephalopathy, hydrocephalus, dysmyelinating and demyelinating conditions [1,4,8].

HII/HIE is a major contributor to death and cerebral palsy in children [9]. It is known to cause grey and white matter changes in the paediatric population. There are various MRI patterns of HII depending on the duration and severity of the hypoxic insult, as well as gestational age at the time of the insult [10]. MRI changes correlate closely to the clinical findings following HIE [10].

Kazi et al. described in 2013, that ischaemia due to hypoxia in the neonatal period results in a typical callosal infarction pattern, which when imaged in the chronic stages, is visualised as CC atrophy [11].

There have been studies done to assess the patterns of injury [9,10], as well as using total brain tissue volumes to prognosticate, and assessment post hypothermia treatment in HIE[12].

The objective assessment of the corpus callosum may be an important marker in children with sequelae of HIE.

There is a high burden of disease associated with HII, with a current incidence of HIE in a sub Saharan African population being 14.9 per 1000 live births, versus high income regions only have incidences as low at 1.5/1000 live births [13, 14].

Previous studies of the corpus callosum done, have only been studied with small patient numbers [1, 6, 7].

There have especially been very limited studies done in a South African population. Quantitatively assessing a South African population for normal and abnormal CC sizes could be a useful parameter for South African Radiologists.

Presently CC measurements have been limited to thickness and anteroposterior length. However, volume assessment may be an easy adjunct to add to these measurements.

There are currently no studies on the use of the corpus callosum as a surrogate for sequelae of HIE in a sub Saharan African setting.

4. Methods

The University of the Witwatersrand Research Ethics Committee granted ethics approval Clearance Certificate Number: M200781 as a sub-study under M170516.

These certificates are attached as Appendix A.

Corpus callosum measurements were performed on MRI scans from an already established database created by another radiologist for his MMed “A Descriptive Study of the Magnetic Resonance Imaging Findings of Hypoxic Ischaemic Injury at a Tertiary Academic Hospital”.

All MRI brain examinations performed for children younger than the age of 15 years, from January 2014 to December 2015, with the following clinical indications, were included in the study: suspected HIE/HII, cerebral palsy, delayed milestones, seizures, cognitive or visual impairment, or other focal neurological deficit (such as tone, motor, reflexes, speech, feeding and movement abnormalities). A clinical history indicating congenital neurological anomalies, central nervous system tumours, chromosomal abnormalities, neurocutaneous syndromes, head trauma, infections, mitochondrial and metabolic disorders and toxin exposure, were excluded.

A total of 128 MRI scans were analysed and the corpus callosum measured. MRI images of children scanned for clinically suspected HIE were retrospectively reviewed and classified into MRI HIE patterns, by a blinded majority consensus reading of 3 Paediatric radiologists. All clinical information provided on the MRI request form was collated and also provided.

After considering and excluding outliers there were a total of 115 MRI brain examinations available. The criteria for identifying outliers were as follows: <1 month of age, hydrocephalus, corpus callosum dysgenesis, imaging discordance (from the initial study), abnormal anterior CC or no CC to measure, no sagittal images provided and a significant outlier in the <6 months category (See Figure 1).

The database of MRI images was accessed using a free DICOM viewer on an Apple computer called Horos. Tools available on Horos were used to measure the corpus callosum thickness (in 4 anatomical points), length and volume for each patient.

The mid-sagittal plane of the T1-weighted MR image was used to perform these measurements as this is the best sequence to visualise the corpus callosum. Specific tools used include line caliper, pencil ROI and Volume tools.

Measurements of the CC thickness were done in the mid-sagittal plane at the pre-determined anatomical levels - namely - rostrum, genu, body and splenium - using the caliper tool. CC length from anterior to posterior were measured using the pencil ROI.

The volume of the CC was calculated using the pencil ROI and Volume tools on the DICOM viewer. The outline of the CC was traced using the pencil tool, in the mid-sagittal plane, as well as the edge slices on either side of the mid-sagittal plane. Using the ROI volume tools, the missing volumes were pre-populated and then the volume calculated using the Compute Volume tool.

Statistical analysis using SAS Version 9.2 was performed. A p -value < 0.05 was considered significant.

CC measurements were categorised for age and MRI pattern. The MRI pattern of injury was based on a prototype created by the primary investigator for the original study and thus already divided into these categories. The MRI imaging patterns were divided into 5 categories based on the type of injury visualised: no MRI features to suggest HIE (1), punctate periventricular white matter injury (2), watershed injury (3), central injury involving deep grey matter nuclei (4) or diffuse injury (5). Where MRI patterns demonstrating HIE (2-5) were compared to those that did not (1).

5. Results

A total of 115 corpus callosums were measured, of which 65.2% were male and 35.8% female. The largest age categories were the 7-23 months and 2-5 years categories, where there were 44 (38.3%) and 45 (39.1%) respectively as summarised in Table 1.

According to the request form at the time of the MRI, there was a varying distribution of reported developmental delay and the vast majority, 112 (97.4%), had no clinical features of HIE as indicated in Table 1. Forty-eight (41.7%) were classified in the category of MRI pattern 1 - no HIE findings, 25 (21.7%) showed punctate periventricular white matter injury (MRI pattern 2), 4 (3.5%) watershed injury (MRI pattern 3), 10 (8.7%) central injury (MRI pattern 4) and 28 (24.4%) diffuse/mixed injury (MRI pattern 5) as summarised in Table 2.

According to the Mann-Whitney test performed before the study to determine a minimum sample, a minimum sample size needed to be at least 23. MRI pattern 3 and 4, both had sample sizes that fell short of this number, with 4 and 10 in each category, respectively. Therefore, MRI patterns 2-5 were grouped together into a new category, HIE imaging features on MRI and compared to MRI pattern 1 (no HIE imaging findings). Forty-eight (58.3%) of the sample fell in the former category and 67 (41.7%) in the latter as summarised in Table 3. The same categorisation was applied to the age categories, as the sample sizes for the <6 months and >5 years were also well below the needed amount, with 13 in each category as summarised in Table 1.

In the non-HIE rostrum thickness group, the mean was 5.701 mm (SD 1.464) and in the HIE rostrum thickness group the mean was 4.018 mm (SD 0.1834), and demonstrated a statistically significant difference when using the Mann-Whitney test ($p = 0.00016$ - greater than 0.05). See Figure 3.

In the non-HIE splenium thickness group, the mean was 6.830 mm (SD 2.128) and in the HIE splenium thickness group the mean was 3.989 mm (SD 0.1883), and although there is a difference in these measures, this was found to be not statistical-

ly significant when using the Mann-Whitney test ($p=0.951 > 0.05$). See Figure 3. In the non-HIE CC length group, the mean was 13.7412 mm (SD 1.4344) and in the HIE CC length group the mean was 13.03 mm (SD 1.918), and although there is a difference in these measures, this was found to be not statistically significant when using the Mann-Whitney test ($p=0.11577 > 0.05$). See Figure 4.

In the non-HIE genu thickness group, the mean was 8.008 mm (SD 2.27) and in the HIE genu thickness group the mean was 5.444 mm (SD 2.49), and demonstrated a statistically significant difference when using the Mann-Whitney test ($p = 0.000011 < 0.05$). See Figure 3.

In the non-HIE body thickness group, the mean was 4.171 mm (SD 1.297) and in the HIE body thickness group the mean was 2.3865 mm (SD 0.1282), and demonstrated a statistically significant difference when using the Mann-Whitney test ($p=0.025 < 0.05$). See Figure 3.

In the non-HIE CC volume group, the mean was 4.4317 cm³ (SD 2.0673) and in the HIE CC volume the mean was 2.114755 cm³ (SD 1.4768), and demonstrated a statistically significant difference when using the Mann-Whitney test ($p=0.03754 < 0.05$). See Figure 5.

Despite there being an absolute difference in the mean thickness of the splenium and the length of the CC in the HIE and non-HIE groups, there was no statistical significance and the null hypothesis was not rejected with a confidence of 95 % as demonstrated by p values being greater than 0.05 when using the Mann-Whitney test. In contrast the p values for the thickness of the rostrum, the thickness of the genu, thickness of the body and volume of the CC were less than 0.05 indicating that there was a statistically significant difference in these means with a confidence interval of 95%. These findings are summarised in Table 3.

6. Discussion

The best regions to measure thickness of the CC are the anterior portions, specifically the rostrum and genu [3,7]. In our dataset there was a statistically significant difference between the non-HIE MRI pattern and HIE MRI patterns for the thickness of the rostrum, the thickness of the genu and CC volume.

Andronikou et al [5] established that the normal mean genu thickness of a 3 year old African population was comparable to a European reference dataset by Garel et al [7], 9 mm compared to 9.1 mm respectively. In this study, the non-HII MRI pattern genu thickness mean was 8.974 mm (age category 2-5 years), which is comparable to the controls in the Andronikou study of 2015 [7]. The mean thickness of the genu in the HII sample was 5.444 mm.

As such the following comparisons and deductions can be drawn. In this study there was significant difference in both the rostrum and the genu thickness of the non-HIE pattern and HIE patterns, possibly indicating that anterior CC thickness is a radiological imaging finding to include with classic HIE imaging pattern findings when supported by clinical findings of existing HIE. Furthermore this is supported by the significant difference in both the non-HIE and HIE genu thickness in comparison to the non-HIV controls in Andronikou et al 2015.

Volume of the CC also showed significant results with the mean volume in the MRI features of no HIE being 4.43 cm³ and the mean for HIE being 2.11 cm³. CC volume has previously only been studied in an adult populations. This positive finding gives hope that CC volume may be a significant finding when compared to normal population reference when controlled for those factors that may affect CC volume that are extrinsic to HIE.

CC volume may be an ideal surrogate and target for machine learning algorithms , since volume accounts for variation in both thickness and length of the CC as an aggregated function, which means any small changes in thickness of an area of the CC may be accounted for in function by a thicker adjacent region or longer CC.

Currently and historically, measurements done by Radiologists although quantitative, are subjective in nature and lack a certain level of repeatability. With modern advances in Artificial Intelligence (AI) such as deep machine learning, it is possible to overcome these flaws and provide a higher level of accuracy and efficiency.

Medicine progresses because of the implementation of technology to make better diagnosis's and decisions that allow treatments to achieve better outcomes for patients. This has been the case for all technology applied to medicine in the pursuit of providing better care for our patients. Artificial intelligence applies modalities of collating and processing large amounts of data and creating useful insights by leveraging modern computing efficiencies. This allows clinicians to achieve better outcomes.

Application of AI is dependant on providing parameters that are of significance in our pursuit of achieving the aforementioned efficiencies. These parameters are typically quantitative measures that can be related to diagnostic, prognostic and clinical outcomes, ie. can we correlate the various measures in our study in the context of HIE, at various ages to provide improved diagnostic, prognostic and management of patients well being [15].

Based on our findings, we would suggest that the genu, the rostrum and the volume be considered as possible surrogates for white matter volume loss and ideal parameters for the application of machine learning algorithms in the interpretation of suspected HIE or other white matter volume loss conditions.

Limitations

There weren't enough samples within the different age categories and HIE pattern subsets to do comparative statistical analysis in all of the age categories.

“Control” patients in the no HIE imaging findings category (MRI pattern 1) could not be classified as totally normal. All scans that formed part of the study were done in children imaged for known HIE and most had some developmental delay.

Other contributing factors relating to CC thinning may have been at play but were not accounted for clinically such as mental retardation, low intelligence quotient (IQ), attention deficit hyperactivity disorder (ADHD) and HIV [1,4,8]. Although the study was to differentiate HIE from non-HIE MRI findings, there were no true clinical non HIE patients, as none of the children in the study were scanned as true controls with no suspected HIE.

As this is a retrospective study, it is difficult to truly look at CC changes without follow up scans to observe the change in the ascribed CC measurement as the subjects develop to full expected CC maturity at the age of five.

Recommendation

A larger sample in the specific age categories and a sample of MRI scans in children with no clinical HIE as a control, is advised. A planned prospective study investigating HIE imaging findings, controlling for HIV, mental retardation, low IQ, ADHD and other possible causes for CC thinning, should be considered.

It would be suggested that a comparative analysis of correlation between the developmental changes and changes in the ascribed CC measurements be performed as a longitudinal study, controlling for the above mentioned confounding factors.

7. Conclusion

CC thickness, particularly in the anterior regions, has shown promise of being a surrogate measure for white matter volume loss in children imaged for the chronic sequelae of hypoxic ischaemic encephalopathy, however, in consideration of the limitations and recommendations in this study, a larger sample size study would be prudent to verify and support these initial findings.

CC volume findings showed significant differences in out HIE and non-HIE MRI patterns, but fall victim to the same limitations as the CC thickness. Further investigations to specifically assess normal and abnormal CC volumes in a wide variety of conditions known to cause CC changes, should be investigated.

8. References

- (1) Andronikou, S., Pillay, T., Gabuza, L., Mahomed, N., Naidoo, J., Hlabangana, L.T. *et al* 2014. Corpus callosum thickness in children: an MR pattern-recognition approach on the midsagittal image. *Pediatr Radiol* 45:258-272 DOI 10.1007/s00247-014-2998-9
- (2) Georgy, B.A., Hesselink, J.R. & Jernigan, T.L. 1992. MR imaging of the corpus callosum. *AJR* 1993; 160: 949-955 DOI 0361-803X/93/1605-0949
- (3) Luders, E., Thompson, P.M. & Toga, A.W. 2010. The development of the corpus callosum in the healthy human brain. *J Neurosci.*, August 18, 2010; 30(33):10985-10990 DOI 10.1523/JNEUROSCI.5122-09.2010
- (4) Reinartz, S.J., Coffman, C.E., Smoker, W.R.K., & Godersky, J.C. 1988. MR imaging of the corpus callosum: normal and pathologic findings and correlation with CT. *AJNR* 9: July/August 1988; 649-656 DOI 0195-6108/88/0904-0649
- (5) Garel, C., Cont, I., Alberti, C., Josserand, E., Moutard, M.L. & Ducou le Pointe, H. 2011. Biometry of the corpus callosum in children: MR imaging reference data. *AJNR* 32:1436-1443 <http://dx.doi.org/10.3174/ajnr.A2542>
- (6) Andronikou, S., Ackermann, S., Laughton, B., Cotton, M., Tomazos, N., Spottiswoode, B. *et al* 2014. Correlating brain volume and callosal thickness with clinical and laboratory indicators of disease severity in children with HIV-related brain disease. *Childs Nerv Syst* 30:1549-1557 DOI 10.1007/s00381-014-2434-3
- (7) Andronikou, S., Ackermann, S., Laughton, B., Cotton, M., Tomazos, N., Spottiswoode, B. *et al* 2015. Corpus callosum thickness on mid-sagittal MRI as a marker of brain volume: a pilot study in children with HIV-related brain disease and controls. *Pediatr Radiol* 45:1016-1025 DOI 10.1007/s00247-014-3255-y
- (8) Krupa, K. & Bekiesinska-Figatowska, M. 2013. Congenital and acquired abnormalities of the corpus callosum: a pictorial essay. *Biomed Research International Volume 2013*, Article ID 265619 <http://dx.doi.org/10.1155/2013/265619>
- (9) Schroff, M.M., Soares-Fernandes, J.P., Whyte, H. & Raybaud, C. 2010. MR imaging for diagnostic evaluation of encephalopathy in the newborn. *RadioGraphics* 2010; 30:763-780 DOI 10.11.48/rg.303095126
- (10) Krishan, P. & Strof, M. 2016. Neuroimaging in neonatal hypoxic ischemic encephalopathy. *Indian J Pediatr* 2016; 83(9):995-1002 DOI 10.1007/s12098-016-2042-1

- (11) Kazi, A.Z., Joshi, P.C., Kelkar, A.B., Mahajan, M.S. & Ghawate, A.S. 2013. MRI evaluation of pathologies affecting the corpus callosum: a pictorial essay. *Indian J Radiol Imaging Oct-Dec*; 23(4):321-332 DOI 10.4103/0971-3026.125604
- (12) Parikh, N.A., Lasky, R.E, Garza, C.N., Bonfante-Mejia, E., Shankaran, S. & Tyson, J.E. 2009. Volumetric and anatomic MRI for hypoxic-ischemic encephalopathy: relationship to hypothermia therapy and neurosensory impairments. *J Perinatol* 2009 February; 29(2):143-149 DOI 10.1038/jp.2008.184
- (13) Lee, A.C.C., Kozuki, N., Blencowe, H., Vos, T., Bahalim, A., Darmstadt, G.L. *et al* 2013. Intrapartum-related neonatal encephalopathy incidence and impairment at regional and global levels for 2010 with trends from 1990. *Pediatr Res* 2013;74 (Suppl 1):50-72. DOI 10.1038/pr.2013.206
- (14) Ballot, D.E., Rakotsoane, D., Cooper, P.A., Ramdin, T.D., Chirwa, T. & Pepper, M.S. 2020. A prospective observational study of developmental outcomes in survivors of neonatal hypoxic ischaemic encephalopathy in South Africa. *S Afr Med J* 2020;110(4):308-312. DOI 10.7196/SAMJ.2020.v110i4.14311
- (15) Brady, A.P. & Neri, E. 2020. Artificial Intelligence in Radiology - Ethical Considerations. *Diagnostics* 2020;10,231 DOI 10.3390/diagnostics10040231

9. Legend of Figures

Figure 1. Sample size and number of MRI studies evaluated high-lighting the methodology and clinical relevant exclusions criteria to determine the cohort of cases within the study parameters.

Figure 2. Length, thickness and volume measurements of the corpus callosum in the mid sagittal plane of a T1 weighted MRI sequence

Figure 3. Box and whisker graph illustrating the comparison of the maximum, minimum, median and interquartile ranges of the thickness of the rostrum, genu, body and splenium of the corpus callosum in non-HIE and HIE imaging data cohorts

Figure 4. Box and whisker graph illustrating the comparison of the maximum, minimum, median and interquartile ranges in length of the corpus callosum in the non-HIE and HIE imaging data cohorts

Figure 5. Box and whisker graph illustrating the comparison of the maximum, minimum, median and interquartile ranges in volume of the corpus callosum in the non-HIE and HIE imaging data cohorts

10. Tables and Figures

Table 1 Clinical Characteristics at time of MRI Examination (n = 115)

Male		75	65.2%
Female		40	35.8%
Age at time of MRI			
	< 6 months	13	11.3%
	7-23 months	44	38.3%
	2-5 years	45	39.1%
	> 5 years	13	11.3%
Developmental delay			
	None	40	34.8%
	Present, but no specified	27	23.5%
	Motor	8	7%
	Speech	5	4.3%
	Combination	33	28.7%
	Not known	2	1.7%
Clinical HIE			
	Mild	0	
	Moderate	0	
	Severe	3	2.6%
	None	112	97.4%

Table 2 Description of Final MRI Findings

MRI Pattern of injury			
	1 - Normal	48	41.7%
	2 - Punctate periventricular white matter injury	25	21.7%
	3 - Watershed	4	3.5%
	4 - Central	10	8.7%
	5 - Diffuse/mixed	28	24.4%

Table 3 Results of non-HIE and HIE measurements with p values

	Non HIE	HIE	
n = 115	48 (41.7%)	67 (58.3%)	
Thickness (in mm)	Mean +/- standard deviation (minimum - maximum)		
Rostrum	5.701 +/- 1.464 (1.89 - 8.69)	4.018 +/- 0.1834 (0.89 - 8.82)	<i>p</i> = 0.00016
Genu	8,008 +/- 2.27 (2.34 - 12.7)	5.444 +/- 2.49 (1.21 - 12)	<i>p</i> = 0.000011
Body	4.171 +/- 1.297 (1.13 - 10.48)	2.3865 +/- 0.1282 (0.644 - 6.079)	<i>p</i> = 0.025
Splenium	6.830 +/- 2.128 (1.994 - 10.48)	3.989 +/- 0.1883 (0.857 - 8.305)	<i>p</i> = 0.0951
Length (in cm)	13.7412 +/- 1.4344 (10.305 - 15.909)	13.03 +/- 1.918 (7.299 - 17.65)	<i>p</i> = 0.11507
Volume (in cm3)	4.4317 +/- 2.0673 (0.2379 - 8.4872)	2.114755 +/- 1.4768 (0.0641 - 4.9411)	<i>p</i> = 0.03754

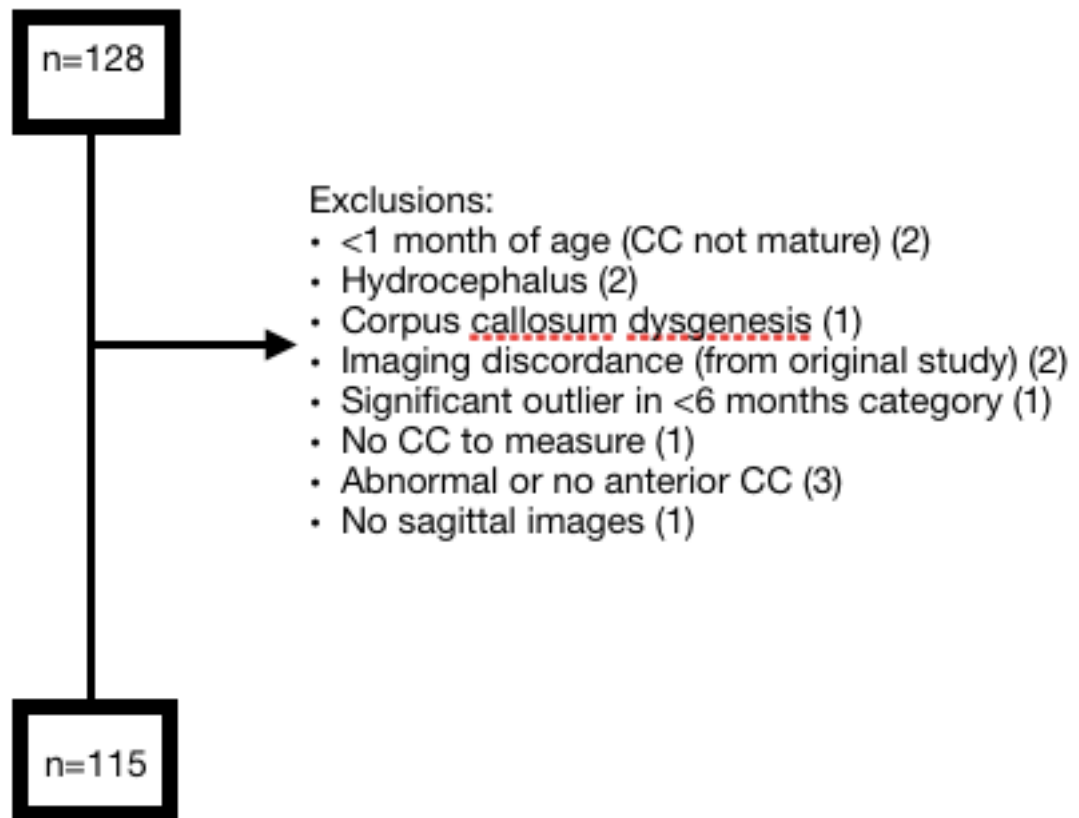


Fig 1 Sample size and number of MRI studies evaluated high-lighting the methodology and clinical relevant exclusions criteria to determine the cohort of cases within the study parameters

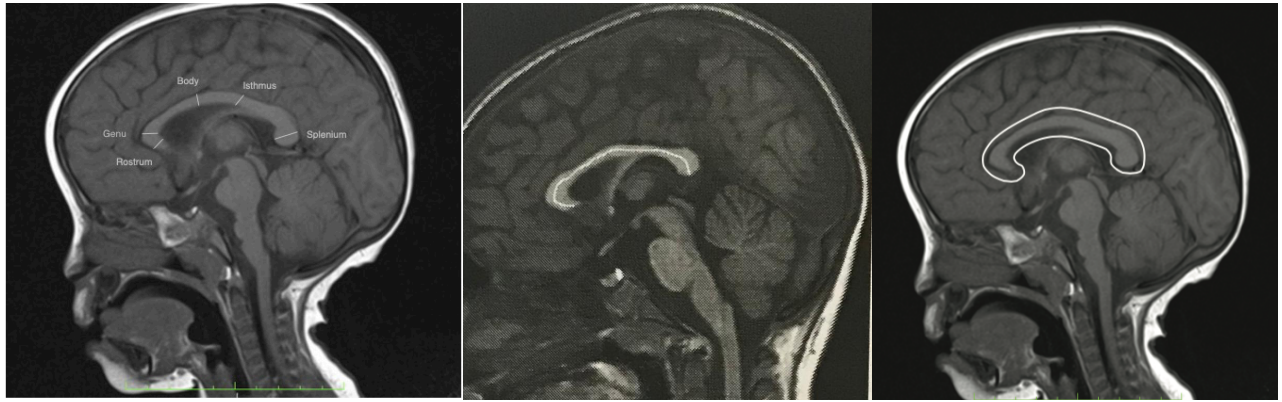


Fig 2 Length, thickness and volume measurements of the corpus callosum in the mid sagittal plane of a T1 weighted MRI sequence

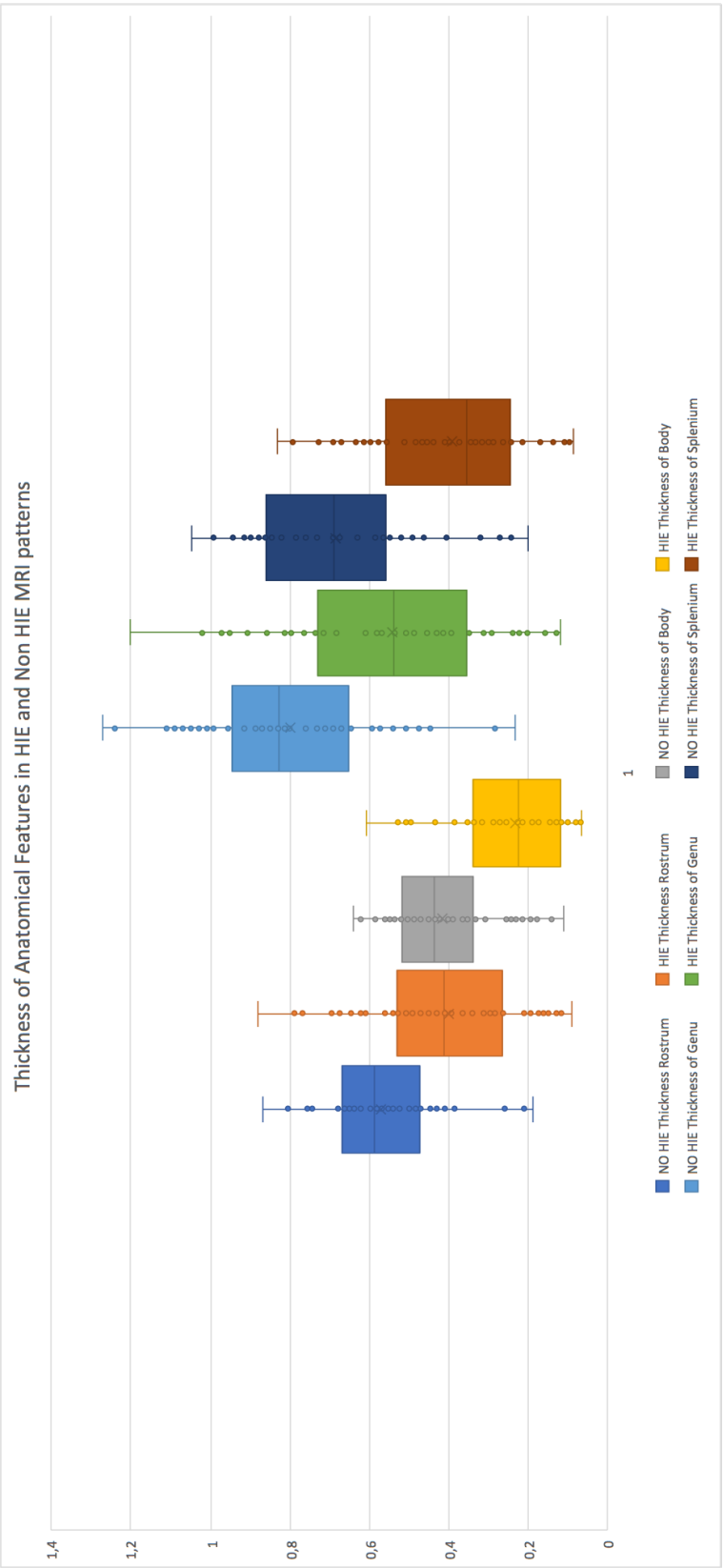


Fig 3 Box and whisker graph illustrating the comparison of the maximum, minimum, median and interquartile ranges of the thickness of the rostrum, genu, body and splenium of the corpus callosum in non-HIE and HIE imaging data cohorts

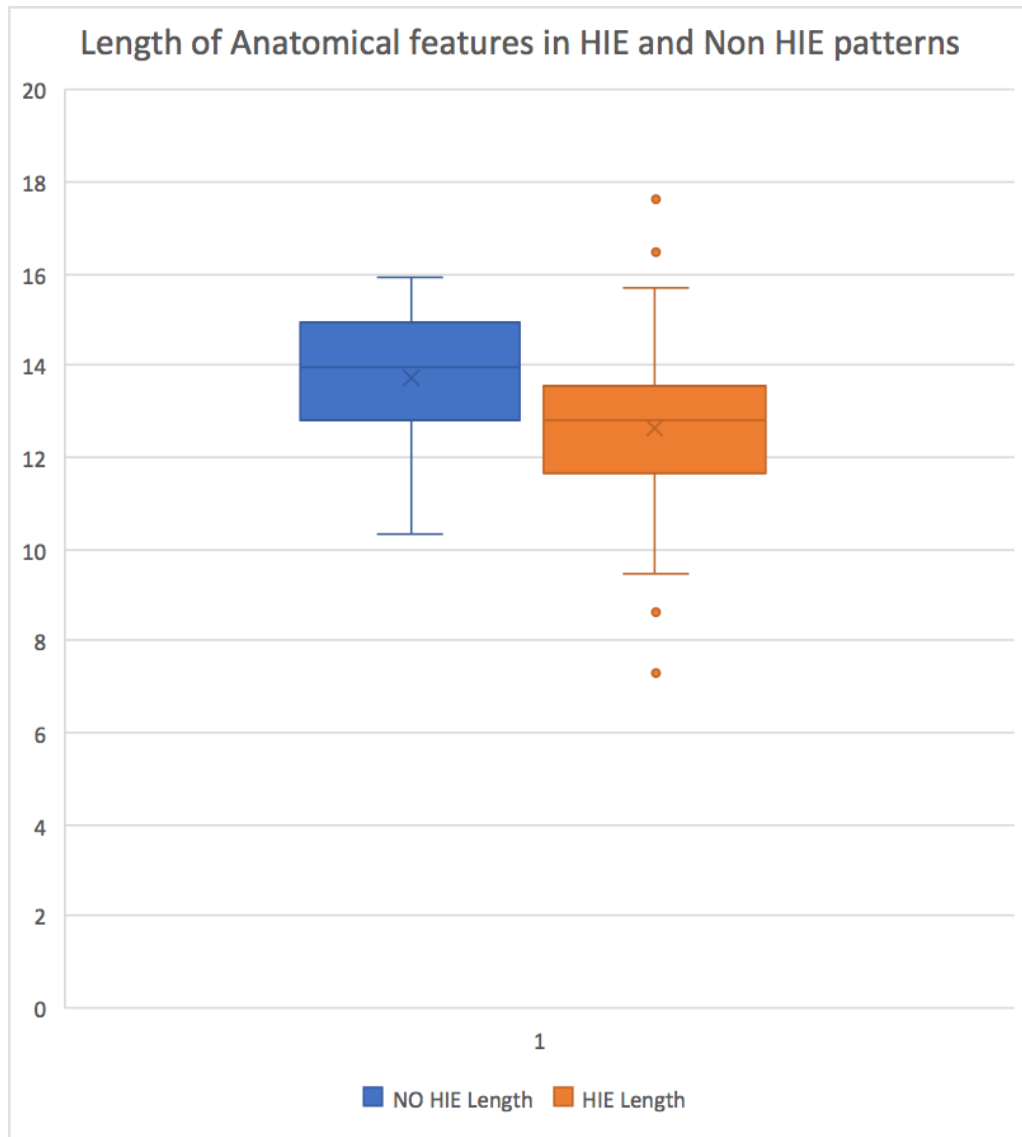


Fig 4 Box and whisker graph illustrating the comparison of the maximum, minimum, median and interquartile ranges in length of the corpus callosum in the non-HIE and HIE imaging data cohorts

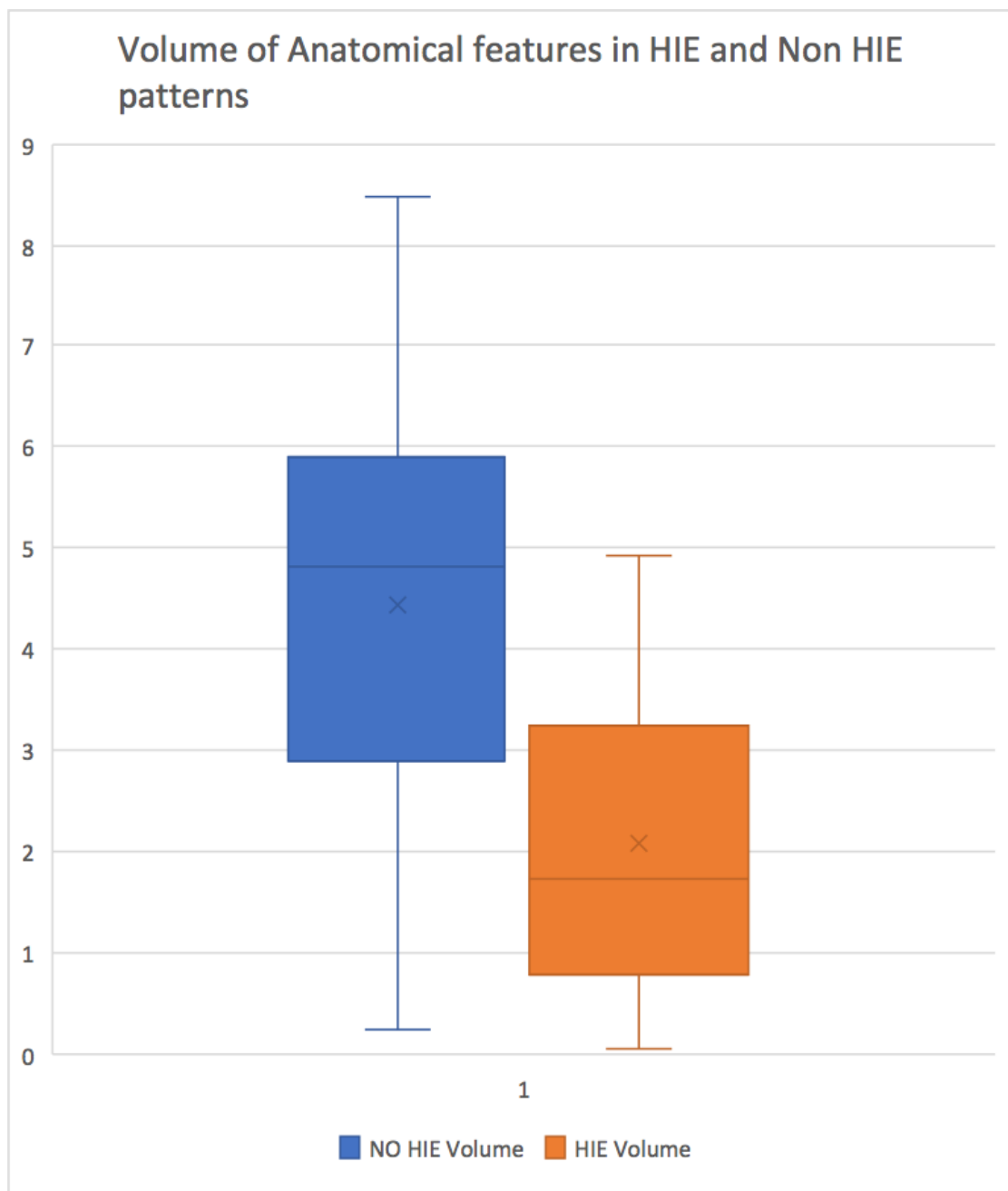


Fig 5 Box and whisker graph illustrating the comparison of the maximum, minimum, median and interquartile ranges in volume of the corpus callosum in the non-HIE and HIE imaging data cohorts

APPENDIX A - ETHICS CLEARANCE CERTIFICATES



R14/49 Dr Liam Lorentz

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)

CLEARANCE CERTIFICATE NO. M170516

NAME: Dr Liam Lorentz
(Principal Investigator)
DEPARTMENT: Radiology
Charlotte Maxeke Johannesburg Academic Hospital

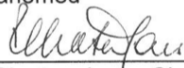
PROJECT TITLE: A Descriptive Study of the Magnetic Resonance Imaging Findings of Hypoxic Ischaemic Injury at a Tertiary Academic Hospital

DATE CONSIDERED: 26/05/2017

DECISION: Approved unconditionally

CONDITIONS:

SUPERVISOR: Dr Nasreen Mahomed

APPROVED BY: 
Professor P. Cleaton-Jones Chairperson, HREC (Medical)

DATE OF APPROVAL: 13/07/2017

This clearance certificate is valid for 5 years from date of approval. Extension may be applied for.

DECLARATION OF INVESTIGATORS

To be completed in duplicate and **ONE COPY** returned to the Research Office Secretary in Room 10004, 10th floor, Senate House/3rd floor, Phillip Tobias Building, Parktown, University of the Witwatersrand. I/We fully understand the conditions under which I am/we are authorised to carry out the above-mentioned research and I/we undertake to ensure compliance with these conditions. Should any departure be contemplated, from the research protocol as approved, I/we undertake to resubmit to the Committee. **I agree to submit a yearly progress report.** The date for annual re-certification will be one year after the date of convened meeting where the study was initially reviewed. In this case, the study was initially reviewed May and will therefore be due in the month of May each year. Unreported changes to the application may invalidate the clearance given by the HREC (Medical).

Principal Investigator Signature _____

Date _____

PLEASE QUOTE THE PROTOCOL NUMBER IN ALL ENQUIRIES



Office of the Deputy Vice-Chancellor (Research & Post Graduate Affairs)

TO: Dr T Nell
School of Clinical Medicine
Department of Radiation Sciences
Division of Diagnostic Radiology
Medical School
University

E-mail: tam.nell@gmail.com

CC: Supervisor: Professor N Mahomed <Nasreen.Mahomed@wits.ac.za>
and <HREC-Medical.ResearchOffice@wits.ac.za>

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

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

DATE: 2020/08/25

REF: R14/49

PROTOCOL NO: **M200781** (*This is your ethics application study reference number. Please quote this reference number in all correspondence relating to this study*)

PROJECT TITLE: *Corpus callosum size on Magnetic Resonance Imaging, a surrogate for white matter volume loss in children imaged for the chronic sequelae of neonatal hypoxic ischaemic encephalopathy*

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

MSWorks2000/Iain0007/Clearscan.wps



R14/49 Dr T Nell

**HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)
CLEARANCE CERTIFICATE NO. M200781**

NAME: Dr T Nell
(Principal Investigator)

DEPARTMENT: School of Clinical Medicine
Department of Radiation Sciences
Division of Diagnostic Radiology
Medical School
University

PROJECT TITLE: Corpus callosum size on Magnetic Resonance Imaging, a surrogate for white matter volume loss in children imaged for the chronic sequelae of neonatal hypoxic ischaemic encephalopathy

DATE CONSIDERED: Ad hoc

DECISION: Approved unconditionally

CONDITIONS: Sub-study under M170516

SUPERVISOR: Professor N Mahomed

APPROVED BY: 
Dr CB Penny, Chairperson, HREC (Medical)

DATE OF APPROVAL: 2020/08/25

This clearance certificate is valid for 5 years from the date of approval. Extension may be applied for.

DECLARATION OF INVESTIGATORS

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

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


Principal Investigator Signature

Date

Appendix B: Note on referencing style

Please note that the referencing in this thesis is a modification of the Vancouver Referencing style, done according to the Faculty of Health Sciences Style Guide as set out by the Wits Health Sciences Library.

The information on this WHSL Vancouver Citation Style Guide for Theses, Dissertations and Research Reports is available from <http://libguides.wits.ac.za/whsl-vancouver> updated on 8 August 2017.

APPENDIX C - TURN IT IN REPORT