Perinatal Stroke in Australian Children

University of Sydney

Faculty of Medicine and Health
Specialty of Child and Adolescent Health
Sydney Medical School

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A thesis submitted in fulfilment of the requirements for the degree of
Doctor of Philosophy
2023

Statement of Originality

This is to certify that, to the best of my knowledge, the content of this thesis is my own work. This thesis has not been submitted for any degree or other purposes.

I certify that the intellectual content of this thesis is the product of my own work and that all the assistance received in preparing this thesis and sources have been acknowledged.

Signature:

Bithi Roy

Date: 5/4/2023

Abstract

Background: More than half of all strokes in children occur during the perinatal period, with a global incidence of 25–40 per 100,000 live births. Aetiology is unclear, hampering prevention research. Diagnosis is often delayed, hindering intervention: first, because some risk factors are present in healthy infants, making at-risk differentiation difficult; second, because variable clinical presentations exist. The aims were to determine Australian birth prevalence, scrutinise risk factors, analyse clinical characteristics and outcomes.

Methods: Six studies were conducted. First**,** a systematic review examined the placental risk factors for perinatal stroke and second, a retrospective audit ascertained placental histopathology in neonatal practices. Third, a systematic review examined prematurity as a risk factor. Fourth study analysed aetiology; fifth study assessed prevalence; and sixth study examined outcomes of perinatal stroke from a longitudinal prospective population-based case-controlled study.

Results: Estimated birth prevalence of perinatal stroke in Australia was 9.6 cases per 100,000 live births per year. Independent risk factors were smoke exposure during pregnancy, 10-minute Apgar score <7, neonatal infection, and hypoglycaemia. Additional risk factors were emergency caesarean section, resuscitation at birth, and abnormal cord blood gas. Conclusive assumptions about the placenta were not possible, given the low frequencies of examination, inconsistent reporting, and the contribution of prematurity. However, thromboinflammatory placental changes were associated with perinatal stroke. Interestingly, 35% presented with pure respiratory symptoms including tachypnoea, apnoea, and cyanosis.

Conclusions: This perinatal stroke research identified rates, independent risk factors, clinical profiles and outcomes, with 48% having neurological impairment. A high index of clinical suspicion and prospective collaborative studies are needed to identify high-risk infants.

Author's Attribution Statement

This thesis includes six first author publications. Chapters 2, 3, 4 and 6 include published manuscripts. Chapter 5 includes one manuscript submitted for publication and Chapter 7 includes one manuscript under preparation for publication.

Chapter 2 of this thesis includes the publication:
Roy B, Arbuckle S, Walker K, Morgan C, Galea C, Badawi B, Novak I. The role of placenta in perinatal stroke: systematic review. Journal of Child Neurology. 2020; 35(11):773-783.

I was the lead and corresponding author on this paper. I contributed to conceptualization of the paper, data curation, analysis and writing the original draft, review, and editing. I responded to comments from reviewers during the publication process, with input from my supervisors.

Chapter 3 of this thesis includes the publication:
Roy B, Gonzalez J, Kwok A, Walker K, Morgan C, Webb A, Badawi N, Novak I. Routine placental histopathological examination: provides answers in neonatal management. American Journal of Pediatrics. 2022; 8(1):10-13.

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Chapter 4 of this thesis includes the publication:
Roy B, Walker K, Morgan C, Finch-Edmondson M, Galea C, Badawi N, Novak I. Epidemiology and pathogenesis of stroke in preterm infants: a systematic review. Journal of Neonatal-Perinatal Medicine. 2022; 15(1):11-18.

I was the lead and corresponding author on this paper. I contributed to conceptualization of the paper, data curation, analysis, and writing (original draft preparation, review and editing). I responded to comments from reviewers during the publication process, with input from my supervisors.

Chapter 5 of this thesis includes the manuscript submitted for publication:
Roy B, Webb A, Walker K, Morgan C, Badawi B, Nunez C, Eslick G, Kent AL, Hunt RW, Mackay MT, Novak I. Etiology of perinatal stroke: a population based study in Australia.

I was the lead and corresponding author on this paper. I contributed to conceptualization of the paper, ethics applications, methodology, data collection, data curation, analysis, project administration and writing (original draft preparation, review, and editing). I responded to comments from reviewers during the publication process, with input from my supervisors.

Chapter 6 of this thesis includes the publication:
Roy B, Webb A, Walker K, Morgan C, Badawi B, Novak I. Risk factors for perinatal stroke in term infants: a case-control study in Australia. 2023; doi:10.1111/jpc.16372.

I was the lead and corresponding author on this paper. I contributed to conceptualization of the paper, ethics applications, methodology, data collection, data curation, analysis, project administration and writing (original draft preparation, review, and editing). I responded to comments from reviewers during the publication process, with input from my supervisors.

Chapter 7 of this thesis includes the manuscript under preparation for publication:
Roy B, Webb A, Walker K, Morgan C, Badawi B, Novak I. Reports on perinatal stroke in Australian children: population-based longitudinal study.

I was the lead and corresponding author on this paper. I contributed to conceptualization of the paper, ethics applications, methodology, data collection, data curation, analysis, project administration and writing (original draft preparation, review, and editing). I responded to comments from reviewers during the publication process, with input from my supervisors.

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As supervisors for the candidature upon which this thesis is based, we can confirm that the authorship attribution statements above are correct.

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Signature:

Date: 5/4/2023

Supervisor: Professor Nadia Badawi
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Date: 5/4/2023

Auxillary Supervisor: Clinical Professor Karen Walker
Signature:

Date: 5/4/2023

Auxillary Supervisor: Dr Catherine Morgan
Signature:

Date: 5/4/2023

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Preface

FIRST AUTHOR PUBLICATIONS RELATED TO THIS THESIS

CHAPTER 2

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CHAPTER 5

Roy B, Webb A, Walker K, Morgan C, Badawi B, Nunez C, Eslick G, Kent AL, Hunt RW, Mackay MT, Novak I. Etiology of perinatal stroke: a population-based study in Australia; under review for publication.

CHAPTER 6

Roy B, Webb A, Walker K, Morgan C, Badawi B, Novak I. Risk factors for perinatal stroke in term infants: a case-control study in Australia. Journal of Paediatrics and Child Health. 2023; doi:10.1111/jpc.16372.

CHAPTER 7

Roy B, Webb A, Walker K, Morgan C, Badawi B, Novak I. A report on stroke in Australian children under the age of 2 years; under preparation for publication.

FIRST AUTHOR PAPER PRESENTATIONS RELATED TO THIS THESIS

2022

Roy B, Webb A, Walker K, Morgan C, Badawi N, Novak I. Presentation and outcomes of childhood stroke: national population-based study. In: Perinatal Society of Australia and New Zealand Annual Scientific Congress; May 15-18, 2022; Adelaide, Australia.

Roy B, Webb A, Walker K, Morgan C, Badawi N, Novak I. Types of childhood stroke: a population-based study in Australia. In: Academy of Child and Adolescent Health Conference; April 7-9, 2022; Doltone House – Darling Island, Pyrmont, Sydney, Australia.

2021

Roy B, Walker K, Morgan C, Badawi N, Novak I. Stroke in children under 2-years of age. In: Quebec American Academy of Cerebral Palsy and Developmental Medicine Conference, Virtual Meeting; October 6-9, 2021; Quebec, Canada.

2020

Roy B, Walker K, Morgan C, Badawi N, Novak I. Stroke in Australian children under 2-years of age. In: Perinatal Society of Australia and New Zealand Annual Scientific Congress; April 5-8, 2020; Convention Centre, Sydney, Australia.

2018

Roy B, Walker K, Morgan C, Badawi N, Novak I. Cerebral stroke in preterm infants: a systematic review. In: Perinatal Society of Australia and New Zealand Annual Scientific Congress; March 23-28, 2018; ANZ Viaduct Events Centre, Auckland, New Zealand.

2017

Roy B, Walker K, Morgan C, Badawi N, Novak I. The role of placenta in perinatal stroke – a systematic review. In: Perinatal Society of Australia and New Zealand Annual Scientific Congress; April 2-5, 2017; Canberra, Australian Capital Territory, Australia.

Roy B, Walker K, Morgan C, Badawi N, Novak I. The role of placenta in perinatal stroke – a systematic review. In: Women's and Newborn Health 8th Annual Academic Day; December 7, 2017; Kolling Auditorium, Royal North Shore Hospital, Sydney NSW Australia.

FIRST AUTHOR POSTER PRESENTATIONS RELATED TO THIS THESIS

2022

Roy B, Webb A, Walker K, Morgan C, Badawi N, Novak I. Aetiology of stroke in young Australian children: national population-based study. In: Perinatal Society of Australia and New Zealand Annual Scientific Congress; May 15-18, 2022; Adelaide, Australia.

2021

Roy B, Walker K, Morgan C, Badawi N, Novak I. Stroke in Australian children under 2-years of age. In: Pediatric Academic Societies Conference, Virtual Meeting: Phase 1; April 30-May 4, 2021.

CO-AUTHOR PRESENTATIONS RELATED TO THIS THESIS

2020

Gonzalez J, Kwok A, **Roy B.** Low birth weight babies: are we ignoring their placentas? In: Women and Newborn Health 11th Annual Academic Day; December 11, 2020; Kolling Auditorium, Royal North Shore Hospital, Sydney NSW Australia.

2021

Gonzalez J, **Roy B**, Kwok A, Walker K, Morgan C, Badawi N, Novak I. Placental histopathology in an at-risk population for cerebral palsy. In: Perinatal Society of Australia and New Zealand XXIV Annual Congress, Virtual Congress; March 29-31, 2021.

FUNDING RELATED TO THIS THESIS

6/04/2016	The Friends of the Mater Foundation
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25/09/2018	AusCP-CTN PhD Top-Up Scholarship progress
16/04/2019	The Friends of the Mater Foundation
14/06/2019	University of Sydney Postgraduate Research Support Scheme
31/07/2020	The Friends of the Mater Foundation

COMMUNITY AND PROFESSIONAL ENGAGEMENT

I have been involved in various formal and informal activities, engaging with key stakeholders and the wider community during the period of candidature. Locally, I have engaged with people with a lived experience of perinatal stroke. I have helped disseminate perinatal stroke research findings at a local level at the Northern Sydney Local Health District Academic Day and Academy of Child and Adolescent Health Conference in Sydney.

RELEVANT TRAINING

31/10/2016	Getting started with a Systematic Review: Interactive workshop, University of Sydney
1– 4/11/2018	Prechtl General Movement Assessment Advanced training course

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List of Abbreviations

AIHW	Australian Institute of Health and Welfare
APPIS	Arterial presumed perinatal ischaemic stroke
APSU	Australian Paediatric Surveillance Unit
ATIII	Antithrombin III
CHD	Congenital heart disease
CI	Confidence interval
CP	Cerebral palsy
CS	Caesarean section
CSVT	Cerebral sinovenous thrombosis
ECG	Electrocardiogram
ECHO	Echocardiogram
EEG	Electroencephalogram
GDM	Gestational diabetes mellitus
HSV	Herpes simplex virus
LBW	Low birth weight
MRI	Magnetic resonance imaging
n	frequency count
NAIS	Neonatal arterial ischaemic stroke
NHS	Neonatal haemorrhagic stroke
NVD	Normal vaginal delivery
PIH	Pregnancy induced hypertension
PPHS	Presumed perinatal haemorrhagic stroke
PVI	Periventricular venous infarction
SGA	Small for gestational age

Impact of COVID-19 on this Program of Research

The COVID-19 pandemic occurred shortly after recruitment commenced of perinatal stroke cases. Our recruitment numbers were approximately 35% below prevalence estimates based on the Australian Cerebral Palsy Register. This under-recruitment is likely due to differences in the ethical requirements of certain jurisdictions and busy clinicians, rather than the direct impact of the COVID-19 pandemic.

If there was a silver lining to this dreaded COVID-19 pandemic, it was that the University of Sydney's zoom sessions made it possible to participate in the PhD coaching and training sessions. Attending these invaluable onsite sessions would otherwise have been challenging for me as a busy clinical professional.

Chapter 1: INTRODUCTION

The problem: Identification of risk factors and clinical manifestations of perinatal stroke in Australian children

1.1 Perinatal Stroke

Definition

Stroke in children is the most common cause of unilateral spastic cerebral palsy (CP).(1) More than half of all strokes in children occur during the perinatal period (from 20 weeks of gestation to 28 days postpartum).(2, 3) The terms perinatal stroke and neonatal stroke (occurring from birth to 28 days postpartum) are used interchangeably. For consistency, this thesis uses the term perinatal stroke.

Perinatal stroke is defined as the clinical syndrome due to interruption of blood supply to a part of the brain in the perinatal period.(4) The vascular event may be blockage of a cerebral blood vessel due to a clot, or breakage of a blood vessel causing a bleed in the brain, or a structural defect of the blood vessel. Vascular events lead to hypoxia in affected areas of the brain, leading to potential brain damage, and its sequalae such as motor asymmetry(1), developmental delays, epilepsy(5, 6), and speech(7) or other sensory abnormalities.

Prevalence

The incidence of perinatal stroke varies, reflecting inconsistencies in single-centre small cohorts versus multi-centre large cohorts, and different terminologies for the stroke subtypes used by different authors. In addition, perinatal stroke is underdiagnosed in the absence of a typical presentation such as seizures. The reported incidence of perinatal stroke described in the literature varies from 1 in 2300 to 1 in 5000 live births(4, 8-10), with a systematic review finding that the highest pooled incidence of arterial ischaemic stroke was 24.6 per 100,000 live

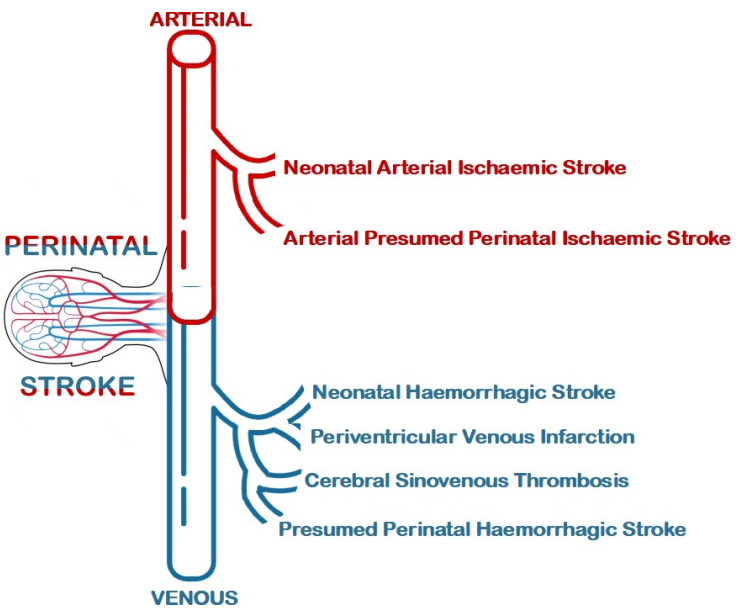
births.(11) When stratified by age, another study reported an incidence of 100/100,000 in preterm infants, 25–40/100,000 in term infants and 13/100,000 per year in children over one month of age.(12)

Classification

The terminology of perinatal stroke subtypes also varies within the literature and has limited the aggregation of data, thus impacting research progress. Stroke occurring at birth has been classified in studies based on pathology and the type of affected blood vessel (arterial ischaemic stroke and haemorrhagic stroke) or the age at which the stroke occurred (perinatal stroke occurring between 20 weeks of gestation and 28 days postnatal age; neonatal stroke occurring from birth to 28 days postnatal age; and paediatric stroke occurring from >28 days to 18-years of age).

A more comprehensive classification was developed by Dunbar and Kirton(13), which combines the above classification methods and was adopted within this thesis (Figure 1):

Figure 1: Perinatal stroke subtypes



a. Arterial stroke:

1. Neonatal Arterial Ischaemic Stroke (NAIS) – is defined as focal ischaemic infarction in one or more arterial territories, occurring more commonly in term infants with acute clinical presentation of stroke.

2. Arterial Presumed Perinatal Ischaemic Stroke (APPIS) – is similar to NAIS, but presents later; that is, 6–12 months after the stroke, with motor deficit.

b. Venous stroke:

3. Neonatal Haemorrhagic Stroke (NHS) – is indicated by focal bleed within the brain parenchyma in term infants.

4. Periventricular Venous Infarction (PVI) – includes in-utero germinal matrix haemorrhage in term infants who present with motor deficit around 6 months of age.

5. Cerebral Sinovenous Thrombosis (CSVT) – is described as presence of thrombus in one or more cerebral veins or dural sinuses, or a parenchymal venous infarction in cerebral venous territory, occurring both in preterm and term infants.

6. Presumed Perinatal Haemorrhagic Stroke (PPHS) – is similar to NHS, but presents later; that is, after 28 days of age.

1.2 Risk Factors

The aetiology of perinatal stroke is unclear. Stroke associated with pregnancy has been described in the literature.(14) Risk factor studies have identified pre-eclampsia, oligohydramnios, instrumental deliveries, male gender, and hypoxic-ischaemic events(15) such as: foetal heart rate abnormalities; umbilical cord abnormalities; meconium-stained amniotic fluid; emergency caesarean section; umbilical arterial pH<7.10; 5-minute Apgar score <7, and resuscitation at birth.(16, 17)

Other factors that are described in the literature are hypoglycaemia(18), thrombophilia(19) and

genetic disorders(20) that cause bleeding, or clotting issues that cause haemorrhagic or thromboembolic phenomena.

The placenta is a highly vascular organ of pregnancy involved in blood flow to and from the foetus and could play an important role in the pathogenesis of perinatal stroke. Placental abnormalities such as thromboembolism, chorioamnionitis, or haemodynamic instability due to placental infarction or abnormal placental perfusion are known to be associated with unfavourable neonatal outcomes.(21, 22)

1.3 Clinical Presentation

The clinical presentation of perinatal stroke includes a variety of signs and symptoms, both specific and non-specific, and can vary between preterm and full-term infants or may be subtle and often go undiagnosed or misdiagnosed.(23) Full-term infants commonly present with seizures(24, 25) or lethargy, while preterm infants(26) may present with non-neurological symptoms such as apnoea, cyanosis, respiratory distress, and poor feeding.

Infants with non-specific symptoms therefore often present later, as much as 5–6 months after their stroke, with motor deficits such as hemiparetic cerebral palsy, and are termed ‘presumed perinatal stroke’ or late presenting perinatal stroke.(27) In the case of an intrauterine stroke, the convalescing newborn may not have acute stroke symptoms. Late diagnosis is problematic as it means that treatment starts later, potentially worsening outcomes.(28, 29)

1.4 Diagnosis

The diagnosis of perinatal stroke is based on ‘elimination’ and a high index of clinical suspicion of stroke.(3) Neuroimaging can reliably establish a diagnosis, define the anatomical location and timing of lesions, and indicate stroke aetiology.(30, 31) Magnetic resonance

imaging (MRI) is the preferred brain imaging modality and should be performed in all infants with suspected stroke.(32) Other investigations include cranial ultrasound (33), cardiac echocardiography and electroencephalogram (EEG) recordings of brain activity, which show the presence of seizure activity even when there is an electrical seizure and/or no clinical seizure activity.

Laboratory tests include sepsis workup, blood glucose level, and the coagulation profile (prothrombin time, activated partial thromboplastin time, fibrinogen). Placental histopathological examination is also an important part of investigating the causative pathways. Prothrombin markers (activated protein C resistance, antithrombin III, plasminogen, protein S, protein C, factor V Leiden, methyl tetrahydrofolate reductase, prothrombin gene, homocysteine, factors VIII IX XI, anticardiolipin antibody, lupus anticoagulant), hypercoagulable disorders and platelet aggregation are performed on both the mother and infant at approximately 6 months of age.

1.5 Outcome

Stroke in adults is a leading cause of mortality and morbidity with substantial economic costs for post-stroke care.(34) In comparison, there is a paucity of long-term follow-up data in children with perinatal stroke.(35) Outcomes are variable. Many factors such as gestational age (preterm or term infants), and type of perinatal stroke (ischaemic or haemorrhagic) determine the outcome, making it difficult to predict outcomes precisely. Studies have suggested combined motor, language, cognitive/behavioural deficits, and epilepsy are more significant with NAIS compared to PVI.(36, 37)

1.6 Research Questions

Studies in this program of research explore relevant issues and challenges regarding the early

and accurate diagnosis of perinatal stroke, risk factors, clinical presentations, and long-term outcome for infants.

The thesis addresses the following six questions based on who? what? and how? of perinatal stroke:

Who gets perinatal stroke? (**RISK FACTORS for perinatal stroke in published literature**)

1. Is it possible to detect whether placental abnormalities are involved in the causal pathway of perinatal stroke?
2. Do neonatologists routinely use placental histopathology to inform management?
3. Is there evidence of prematurity as a risk factor for perinatal stroke?

What are the risks for perinatal stroke in the Australian population? (**RISK FACTORS and EPIDEMIOLOGY of perinatal stroke in Australia**)

4. What is the epidemiology and birth prevalence of perinatal stroke in Australia?
5. Is it possible to precisely identify the risk factors of perinatal stroke?

How does perinatal stroke present? (**CLINICAL CHARACTERISTICS and OUTCOME of perinatal stroke in Australia**)

6. What are the clinical presentations and outcomes of perinatal stroke?

1.7 Aims of Research

In response to the six research questions, the six objectives of this thesis are:

1. To summarise the literature regarding the role of the placenta in perinatal stroke.
2. To audit the rates of placental histopathological examination in clinical practice.

3. To summarise the epidemiology and pathogenesis literature of stroke in preterm infants.
4. To examine the prevalence of perinatal stroke in Australia.
5. To conduct a well-powered comparative study to accurately isolate the risk factors for perinatal stroke from healthy infants.
6. To analyse the clinical characteristics and long-term outcomes of perinatal stroke.

1.8 Significance of the Research

Perinatal stroke is one of the leading causes of cerebral palsy in infancy. The lifelong disability impacts the quality of life of affected children and imposes a significant burden on families and national healthcare systems. The aetiology is not well understood. In addition, several risk factors also exist in healthy infants that may confound the diagnosis of stroke. This research examines historical (from the literature) and current (from Australian population-based studies) risk factors for perinatal stroke. Above all, the research aims to be future-facing – to enable discovery of life-changing preventative strategies, early diagnoses that facilitate earlier interventions to harness neuroplasticity, creating a brighter future for children with cerebral palsy.

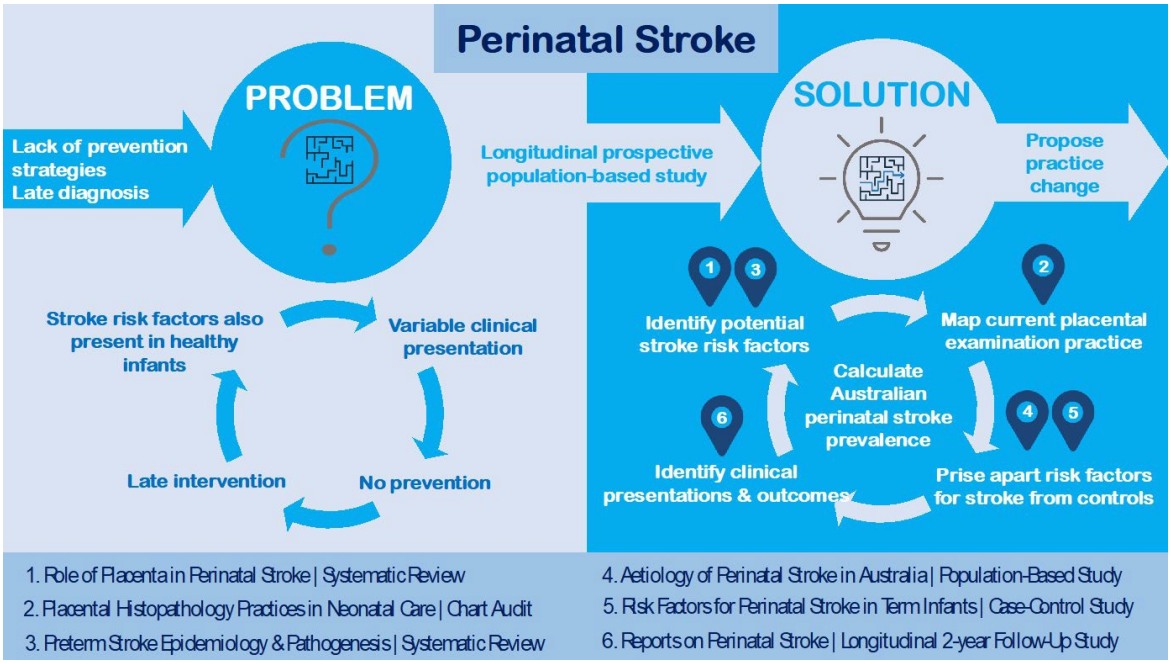
1.9 Outline of Studies

A summary of the six studies and how they fit together within this thesis is portrayed in Figure 2. Gaps in the literature exist about perinatal stroke risk factors, unquestionably because some of these risk factors are also present in healthy infants. For this reason, prevention strategies are lacking for neonates. This is compounded by the non-neurological manifestations of perinatal stroke. As a result, the overarching problem is a late diagnosis of perinatal stroke. For all those reasons there is delay in the initiation of multi-disciplinary therapy services, including physiotherapy, occupational

therapy, speech pathology, exercise physiology, and assistive technology and adaptive equipment that improve children's daily living skills.

The paucity of perinatal stroke pathogenesis evidence was addressed by reviewing the published literature to examine the potential risk factors, such as the placenta. The placenta is a highly vascular organ shared between the mother and foetus and is likely to cause a stroke when a thrombus or infection is present (Study 1). In addition, another systematic review was conducted to analyse the association between prematurity and perinatal stroke (Study 3). A retrospective audit was then conducted to map the current placental examination practices in neonatal management (Study 2). Resolving the information gap is the emphasis of the remaining studies. Study 4 was a prospective, population-based study to determine the birth prevalence rate and examine the risk factors of perinatal stroke in Australian children. Study 5 was a validation of risk factors in a case control comparative study. Study 6 prospectively analysed the clinical characteristics and outcomes of perinatal stroke, to ultimately expedite diagnosis and to enable timely intervention strategies. The final chapter summarises the results and limitations of the studies and discusses future research directions for perinatal stroke.

Figure 2: Thesis outline



Study 1: (Chapter 2)

A *systematic review* was conducted to summarise the literature regarding the role of the placenta in perinatal stroke (Aim 1). The review examined the frequency of placental examinations in perinatal stroke and assessed the role of the placenta within the aetiology of stroke. Placental examination was under-reported, hence the results should be interpreted with caution.

Study 2: (Chapter 3)

A 2-year *retrospective audit* was conducted to audit the rates of placental histopathological examination in clinical practice (Aim 2). The audit was conducted in a private hospital setting to determine the rates of placental screening in obstetric and neonatal management in the private sector. Overall, placental examination was performed in less than 10% of all live births. However, for high-risk pregnancies, placental collection rates were 6-fold higher.

Study 3: (Chapter 4)

A *systematic review* was undertaken to evaluate the literature regarding the epidemiology and pathogenesis of stroke in preterm infants, since not much is known about this (Aim 3). The vast majority of the publications pooled reports from preterm and full-term infants, precluding full understanding of the pathogenesis of stroke in the preterm population.

Study 4: (Chapter 5)

A *two-year population-based prospective study* was conducted to examine the prevalence of perinatal stroke cases in Australia (Aim 4). Stroke presenting in children under 2 years of age in Australia was studied. The Australian birth prevalence of acute symptomatic perinatal stroke was estimated to be 9.6 cases per 100,000 live births per year. When stroke cases were compared with the Australian population data (from the Australian Institute of Health and Welfare), caesarean section, low Apgar scores and resuscitation at birth were found to be significant risk factors for perinatal stroke.

Study 5: (Chapter 6)

A well-powered *prospective case-control study* was conducted to accurately isolate the risk factors for perinatal stroke in term infants (Aim 5). Maternal smoking, 10-minute Apgar score <7, neonatal infections, and hypoglycaemia were identified as independent risk factors for perinatal stroke. Emergency caesarean section, resuscitation at birth and abnormal cord blood gases were also identified as additional risk factors.

Study 6: (Chapter 7)

A *prospective longitudinal follow-up study* of stroke cases in Australia was conducted to analyse the clinical characteristics and outcomes of perinatal stroke (Aim 6). Perinatal stroke cases with acute and chronic symptoms in children up to 2 years of age were studied. Perinatal stroke was found to present with seizures, but also unexpectedly with non-neurological manifestations, including purely respiratory symptoms.

1.10 Ethics

Ethics approval was obtained from the Human Research Ethics Committee of the Children's Hospital at Westmead and Royal Prince Alfred Hospital
(HREC Reference: LNR/16/SCHN/449, 2019/ETH06281 Appendix A).

The systematic reviews were registered in the International Prospective Register of Systematic Reviews (PROSPERO Registration no: CRD42017081256; PROSPERO Registration no: CRD42018081466) in 2017.

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These studies were funded by:

1) University of Sydney Postgraduate Research Support Scheme

2) The Friends of the Mater Foundation

   The Mater Hospital Sydney

   PO Box 958, North Sydney NSW 2059 Australia

3) Cerebral Palsy Alliance Research Institute,

   PO Box 171, Forestville NSW 2087 Australia

Chapter 2: SYSTEMATIC REVIEW OF PLACENTA AND PERINATAL STROKE

2.1 Introduction

The placenta is a highly vascularized gestational organ that directly connects maternal and foetal circulation, and thus a clot or infection within the placenta increases the potential risk for perinatal stroke. Therefore, this systematic review was conducted to summarize what is known in the literature regarding the role of the placenta in the pathogenesis of perinatal stroke.

Placental pathology was available in 24% of perinatal stroke cases, of which 87% were abnormal, supporting circumstantial evidence and the biological plausibility of causality. However, due to the low frequency of placental examinations reported and the inconsistency in reported placental malformations, the results should be interpreted with caution.

A challenge within this evidence base was the inconsistent reporting of placental abnormalities. To enable uniform analysis and data aggregation, placental pathologies were objectively reclassified according to the Redline classification.(38) This classification included maternal and foetal vascular lesions, as well as infectious and immune/idiopathic inflammatory lesions. The most common placental pathology in perinatal stroke was the thromboinflammatory process (Redline's Category 2).

2.2 Published Manuscript

Roy B, Arbuckle S, Walker K, Morgan C, Galea C, Badawi N, Novak I. The role of the placenta in perinatal stroke: a systematic review. Journal of Child Neurology 2020; 35(11):773-783.

Topical Review Article

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SAGE

The Role of the Placenta in Perinatal Stroke: A Systematic Review

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Abstract

Context: Placental pathology may be an important missing link in the causal pathway of perinatal stroke. The study aim was to systematically review the literature regarding the role of the placenta in perinatal stroke. MEDLINE, Embase, Scopus, and Web of Science electronic databases were searched from 2000 to 2019. Studies were selected based on predefined criteria. To enable comparisons, placental abnormalities were coded using Redline's classification. Results: Ten studies met the inclusion criteria. Less than a quarter of stroke cases had placental pathology reported. Placental abnormalities were more common among children with perinatal stroke than in the control group. The most frequent placental abnormality was Redline's category 2 (thrombo-inflammatory process). Conclusions: Placental abnormalities appear to be associated with perinatal stroke, supporting additional indirect evidence and biological plausibility of a causative role. However, the results should be interpreted cautiously considering the low frequency of placental examination and lack of uniformity in placental pathology reporting. Clinical Trial Registration: PROSPERO Registration no: CRD42017081256.

Keywords

placental pathology, placental disease, stroke in children

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Perinatal stroke is the most common type of vascular stroke in children.[1] It is defined as a cerebrovascular event occurring from 20 weeks' gestation up to 28 postnatal days due to ischemia (secondary to embolism or anatomical narrowing of the blood vessels) or hemorrhage in a blood vessel of the brain. Perinatal stroke is classified by the timing and presentation of stroke: acute symptomatic perinatal stroke presenting in the neonatal period and presumed perinatal stroke presenting in infancy or childhood. Acute symptomatic perinatal stroke includes (1) neonatal arterial ischemic stroke, (2) neonatal cerebral sinovenous thrombosis, and (3) neonatal hemorrhagic stroke. Presumed perinatal stroke includes (1) arterial presumed perinatal ischemic stroke, (2) periventricular venous infarction, and (3) presumed perinatal hemorrhagic stroke.[2]

The true incidence of perinatal stroke is likely to be higher with better detection from advanced diffusion-weighted magnetic resonance imaging (MRI), especially in the case of hyperacute and acute stroke.[3] The estimates suggest an incidence of perinatal stroke between 1 in 1600 and 1 in 3000 live births.[4-6]

The pathogenesis of perinatal stroke is complex and multifactorial and includes an array of possible associations and putative risk factors. Neonatal infection such as meningitis may cause acute inflammatory processes in the cerebral arteries of the neonatal brain and may be an independent risk factor for arterial ischemic stroke.[7] Other likely independent risk factors include maternal smoking during pregnancy, intrapartum maternal fever (>38ºC), low Apgar score at 5 minutes (<7), and neonatal hypoglycemia (blood glucose level <2 mmol/L).[8,9] Maternal use of cocaine during pregnancy is another risk factor for arterial stroke due to vasoconstriction and vasospasm in the fetus.[10]

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Hematologic conditions such as the physiological prothrombotic state during pregnancy prevents fetal, maternal, and neonatal bleeding at the time of delivery by increasing clotting factors V, VII, VIII, IX, X, and XII, von Willebrand factor, and plasma fibrinogen levels that could predispose to thromboembolism, thus elevating the risk of neonatal arterial ischemic stroke.[1] However, a case-control study (n ¼ 182 cases and controls each) showed no difference in thrombophilia markers at 12 months of age, but the possible role of coagulopathy could not be excluded at the time of the perinatal stroke.[11] In addition, inherited prothrombotic conditions from genetic mutations like methylenetetrahydrofolate reductase and factor V Leiden gene mutations, and deficiency in protein C or S, are possible risk factors for neonatal arterial ischemic stroke but the evidence is weak and inconsistent.[12-14]

A direct link between congenital heart disease and perinatal stroke is not clear, although a cardioembolic event secondary to cardiac catheterization or cardiac surgery is a plausible cause of ischemic stroke.[15] A small population-based case-control study (n ¼ 40) could not confirm a link between congenital heart disease in infants with perinatal arterial stroke, but the small sample size means the results should be interpreted cautiously.[16] Consensus exists that it is important to evaluate neonates with congenital heart disease for risk of perinatal stroke.[17]

The placenta is a vascular gestational organ carrying blood to and from the fetus and could have a significant role in the etiology of perinatal stroke via (1) placental blood flow abnormalities inducing clots, (2) sudden catastrophic events such as placental abruption creating acute hemodynamic instability or hypoperfusion in the fetus resulting in stroke, or (3) chorioamnionitis. The precise mechanism of placental abnormality and perinatal stroke is not well understood, partly because the placenta is commonly discarded before the clinical presentation of perinatal stroke.[18] Other placental abnormalities such as small or hypoplastic placenta, large placental infarcts, thrombosis, knot in the umbilical cord, narrow umbilical cord diameter, or abnormal insertion of the cord may trigger perinatal stroke because of placental vascular malperfusion.[19]

Aim

The aim of this study was to conduct a systematic review of published literature to explore the frequency of placental examination in perinatal stroke and to evaluate the role of the placenta in perinatal stroke within the studies.

Method

Study Design

A systematic review was conducted using the Cochrane recommendations for conducting a review, and the findings were reported using the Meta-analyses Of Observational Studies in Epidemiology (MOOSE) checklist.[20]

Data Source

MEDLINE, Embase, Scopus, and Web of Science (Science and Social Science Citation Index) electronic databases were systematically searched. A further hand search was conducted of publications using a sensitive methodological filter for placental pathology studies. Search terms were (placental pathology*) OR (aetiol* OR etiol*) AND perinatal stroke, as well as MeSH (Risk Factor OR Aetiology) AND perinatal stroke with limits of English language and human studies. A full list of search terms can be found in the Supplementary File and PROSPERO registration.

Study Selection

Studies from January 2000 to August 2019 were included for analysis if they met all the following criteria:

1. Perinatal stroke was the primary outcome (diagnosis of stroke was based on the reporting paper and not viewing of the original neuroimaging).
2. Risk factors were identified in the fetus and/or all live births.
3. Placental pathology data were available for any perinatal stroke case within the study.
4. All outcomes (including deaths) were reported.
5. All types of study designs
6. The articles were published in English.

The exclusion criteria were as follows:

1. Review papers
2. Abstracts and conference abstracts

Figure 1 summarizes the study flow using a PRISMA diagram.[21]

Data Extraction

Publications were reviewed to identify perinatal stroke cases that met the study selection criteria for stroke in a fetus, or an infant born preterm or term, or in a baby post term age. The principal reviewer (BR) and an independent second reviewer (IN) devised the search strategy. The reference lists of these selected studies were hand searched for further relevant articles, but no additional publications were identified.

An a priori designed data extraction tool was used based on recommendations from the Cochrane group. Extraction included study design, country of the study, gestational age, perinatal stroke types, risk factors of stroke, numbers of cases with placental examination, specific placental abnormality, clinical presentation and outcome of stroke and unadjusted and adjusted odds ratios if reported. Extraction of data from each of the 10 studies was carried out independently by both extractors (BR and IN). The quality of included studies was assessed using the Oxford Centre for Evidence-Based Medicine 2011 Levels of Evidence.[22]

Stroke Types

The article specifically sought to identify cases with a vascular cause of infarction to elucidate cases at higher risk of placental abnormality. Inconsistent terminology has been used in the litera- ture to classify types of perinatal stroke. To enable aggregation of data, we recoded the strokes uniformly using Dunbar and Kirton's contemporary classifications.[2] The following variables were

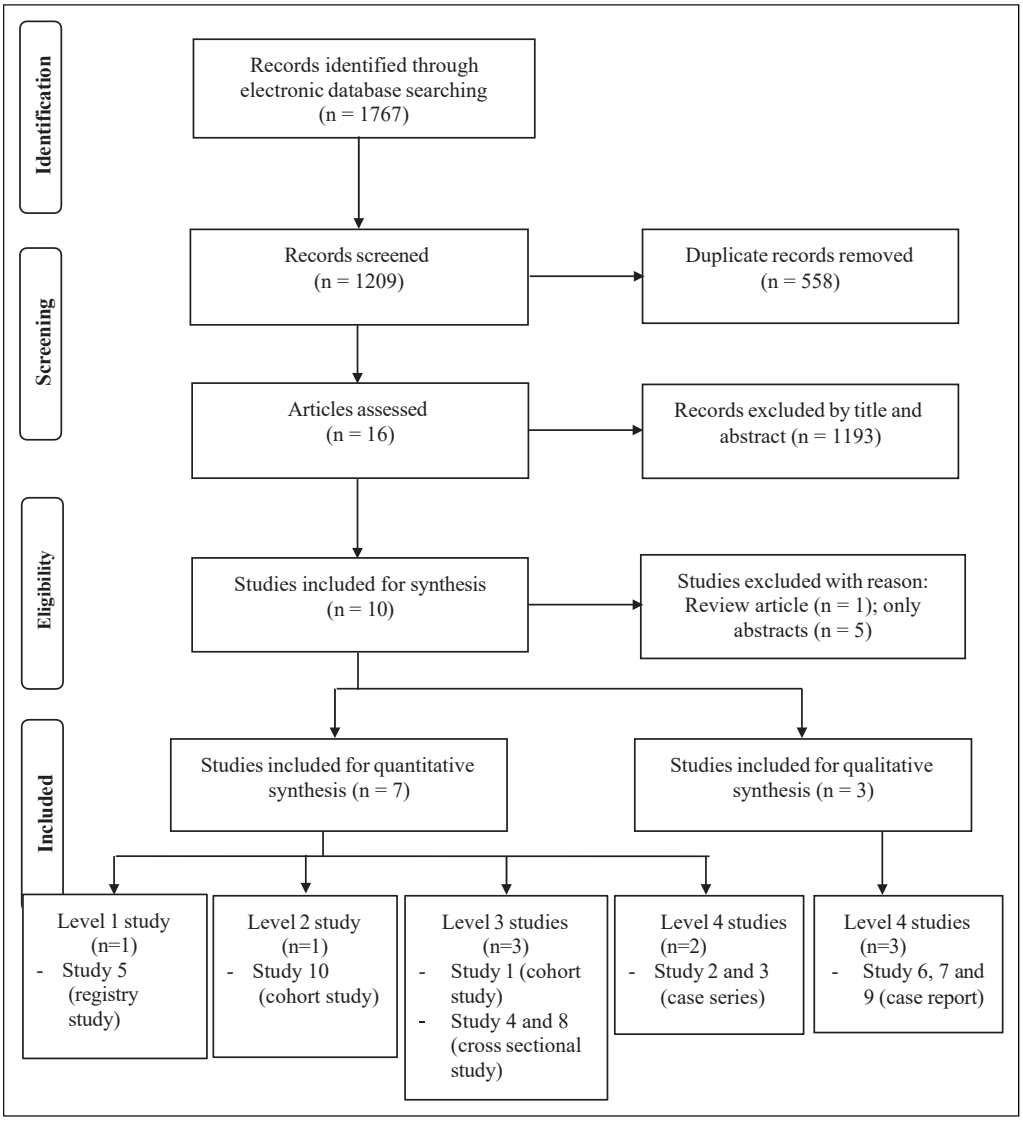

Figure 1. Study selection: Placental pathology in perinatal stroke.

extracted to name the different types of perinatal stroke: (1) age when stroke happened (fetal or in utero, neonatal from 0 to 7 days of age, and perinatal from 0 to 28 days of age), (2) type of vessel affected (arterial or venous), and (3) etiology (ischemic or hemor- rhagic). Arterial ischemic stroke and cerebral stroke nomenclatures were also used interchangeably. It is important to note that some authors used the term *cerebral infarction* to describe a type of stroke. We acknowledge that cerebral infarction is not a reliable indicator of ischemic stroke as it is also used to describe cerebral infarction due to hypoxic-ischemic encephalopathy. After careful analysis of the cases from the reporting papers where authors used the term *cerebral infarction*, it could be confirmed that all cases had indeed experienced ischemic stroke. Discrepancies regarding classification of stroke and any potential chance of systematic bias were discussed and a final decision was made without requiring further adjudication (Table 1).

Risk Factors

Some of the included studies analyzed clusters of risk factors, which cannot be considered synonymous with true causation. The plausible causative factor among the variety of risk factors identified by the placental pathologist author (SA) are underlined for each of the perinatal stroke cases (Table 2). It is important to note that the term *fetal thrombotic vasculopathy*, which was used in some of the studies has now been superseded in the literature by the more accurate term *fetal vascular malperfusion*.[34,35] These terminologies were coded as interchangeable events.

Statistics

Descriptive statistics, n (%), were used for the etiologic and clinical variables extracted from papers. We intended to conduct a meta-

Table 1. Characteristics of Studies Included in the Systematic Review.

Study no:	Author (publication year)	Study origin	Study design	Study population	Study period	Sample size	Placental availability (percentage)	Placental abnormality (percentage)	Level of evidence as per OCEBM[22]
Study 1	Bernson-Leung (2018)[23]	USA	Retrospective Cohort study Case-control study	Preterm ¼ 7	2005-2015	Cases ! 46 Controls ! 99	Cases ! 46/46 Controls ! 99/99	Cases ! 41/46 (89%) Controls ! 61/99 (62%) Reported *P* value (cases vs controls) *0.0007*	Level 3
Study 2	Fluss (2016)[24]	Switzerland (2 cases); France (3 cases)	Case series	Term; male ¼ 3, female ¼ 2	NR	5	2/5	2/2	Level 4
Study 3	Magnetti et al (2014)[25]	Italy	Retrospective Case series	Term ¼ 3, Preterm ¼ 2; male ¼ 4, female ¼ 1	2009-2013	5	5/5	3/5	Level 4
Study 4	Takenouchi et al (2012)[26]	USA	Retrospective Cross sectional study	Term	2004-2009	4	4/4	3/4	Level 3
Study 5	Elber et al (2011)[27]	Canada	Retrospective Registry study	34 wk to 28 postnatal days	1992-2006	186	12/186 (6%)	10/12 (83%)	Level 1
Study 6	Dueck (2009)[28]	Canada	Case report	Term	NR	1	1/1	1/1	Level 4
Study 7	Das (2008)[29]	USA	Case report	36 wk	NR	1	1/1	1/1	Level 4
Study 8	Curry (2007)[30]	USA	Prospective Cross-sectional study	NR	1997-2005	60	8/60 (13%)	7/8 (88%)	Level 3
Study 9	Ghidini (2006)[31]	USA	Case report	Fetus	NR	1	1/1	1/1	Level 4
Study 10	Lee (2005)[32]	USA	Retrospective Cohort study Case-control study	NR	1997-2002	Cases ! 40 Controls ! 120	Cases ! 3/40 (8%) Controls ! 11/120 (9%)	Cases ! 3/3 (100%) Controls ! 5/11 (45%)	Level 2
Total	10 studies		Prospective ¼ 1 Retrospective ¼ 9	Fetus to 28 postnatal days		n ¼ 349	83/349 (24%)	72/83 (87%)	Level I ¼ 1 Level II ¼ 1 Level III ¼ 3 Level IV ¼ 5

*NR – Not Reported.
OCEBM – Level 1 (study 5): 'Local and current random sample survey'; Level 2 (study 10): 'Observational study with dramatic effect'; Level 3 (studies 1, 4 and 8): 'Local non-random sample'; Level 4 (studies 2, 3, 6, 7, 9): 'case-series or historically controlled studies'.

analysis; however, the data could not be extracted at the individual case level, precluding meaningful meta aggregation. Fisher exact associations were conducted for type of stroke and Redline's category classification of placental histopathology. Significance was set at $P < .05$. All analysis was conducted using Stata, version 14 (StataCorp, College Station, TX).

Results

Both reviewers (BR and IN) independently reviewed all 1209 titles/abstracts after excluding the duplicate studies. Sixteen studies were selected for full-text review. Of these, 6 were excluded (1 review paper and 5 abstracts or conference

Table 2. Etiologic Risk Factors and Relationship of Placental Pathology vs Types of Perinatal Stroke.[a]

Study no:	Cases	Nonplacental risk factors/associations	Placental abnormalities	Redline's categories[b] 1	2	3	4	Other[c]	Type of stroke	Outcome
Study 2	Case 1	Severe perinatal asphyxia, emergency CS, MSL, high serum lactate, maternal and infant protein C deficiency	Placental gross hypotrophy with chronic ischemic lesions, retroplacental hematoma			P		Placental hypotrophy, Retroplacental hematoma	NAIS	Left spastic CP, epilepsy, mildintellectual disability
	Case 2	Neonatal protein C deficiency	Severely abnormal with multiple thrombosis, ischemic areas, fetal vasculopathy		P	P			NAIS	CP
Study 3	Case 3	Maternal hypodysfibrinogenemia	FTV, inflammatory lesions		P				CSVT þ NAIS	NR
	Case 4	Maternal infection, PROM, preeclampsia	Placental infarction, focal edema villi			P		Focal edema of villi	CSVT þ NAIS	NR
	Case 5	Neonatal sepsis, patent foramen ovale	Necrotizing chorioamnionitis		P				NAIS	NR
Study 4	Case 6	NR	FTV		P				NAIS	NR
	Case 7	NR	FTV		P				NAIS	NR
	Case 8	NR	FTV		P				NAIS	NR
Study 5	Case 9	IUGR, nuchal cord, oligohydramnios, polycythemia, CHD	Chronic villitis, distal villous immaturity, placental infarct, placental weight <3rd percentile			P			NAIS	Mild motor dysfunction
	Case 10	IUGR, emergency CS, anaemia, CHD	Chronic villitis, chronic intervillositis, villous edema, positive immunostaining for CD68[þ] cells		P				NAIS	Lost to follow-up
	Case 11	GBS positive, abnormal FHR, emergency CS, feto-maternal hemorrhage, anaemia	Chorionic thrombosis, avascular fibrotic villi, increased nucleated red blood cells		P				NAIS	Mild motor dysfunction
	Case 12	Abnormal FHR, emergency CS, anaemia, systemic thrombosis, CHD	Cord overcoiling, distal villous immaturity	P					NAIS	Mild motor dysfunction
	Case 13	Infertility, placental abruption, emergency CS, DIC	Velamentous cord insertion, cord venous congestion, chronic villitis, chronic intervillositis, placental infarction, distal villous immaturity, placental weight <10th percentile	P	P	P			NAIS	Mild motordysfunction, language delay

(continued)

Table 2. (continued)

Study no:	Cases	Nonplacental risk factors/associations	Placental abnormalities	Redline's categories[b] 1	2	3	4	Other[c]	Type of stroke	Outcome
	Case 14	GBS positive, PROM, sepsis, DIC	Severe and diffuse funisitis, acute cord thrombosis, cord venous congestion, severe chorioamnionitis, chorionic thrombosis, stem villous thrombosis, distal villous immaturity, villous chorangiosis		P			Villous chorangiosis	CSVT	Language delay
	Case 15	APH, abnormal FHR, emergency CS, chorioamnionitis, sepsis, anaemia	Retroplacental hematoma, cord hemangioma, moderate chorioamnionitis, avascular fibrotic villi, distal villous immaturity		P			Retroplacental hematoma	CSVT	Normal
	Case 16	Abnormal FHR, emergency CS	Marginal cord insertion, acute stem vessel thrombosis, acute chorionic thrombosis	P	P				CSVT	Mild motor dysfunction, language delay
	Case 17	CHD, postcardiac surgery	True cord knot and stricture, acute chorionic thrombosis, acute cord thrombosis, retro membranous hematoma	P					CSVT	Language delay
	Case 18	Abnormal FHR, emergency CS	Cord stricture, stem villous thrombosis, chorionic thrombosis, placental infarct, distal villous immaturity, placental weight <10th percentile		P	P			CSVT	Mild motor dysfunction, language delay
Study 6	Case 19	Vacuum delivery	Chorioamnionitis, funisitis, vasculitis, layered deposition of fibrin, inflammatory material (lines of Zahn) within the umbilical vein confirmed thrombosis within the vein	P	P				NAIS	NR
Study 7	Case 20	MSL, anemia, thrombocytopenia	Large chorangioma with thrombosis, hemorrhage and infarction					Chorangioma	NAIS	NR

(continued)

Table 2. (continued)

Study no:	Cases	Nonplacental risk factors/associations	Placental abnormalities	Redline's categories[b] 1	2	3	4	Other[c]	Type of stroke	Outcome
Study 8	Case 21	NR	Fibrous plaques with increased intervillous fibrin					Fibrous plaques with increased intervillous fibrin	NAIS	NR
	Case 22	NR	Small and 20% infarcted			P			NAIS	NR
	Case 23	NR	Circumvallate insertion					Circumvallate insertion	NAIS	NR
	Case 24	NR	Small extensive >25% infarcted			P			NAIS	NR
	Case 24	NR	Small abnormal placenta			P		Abnormal placenta	NAIS	NR
	Case 26	NR	Extensive dystrophic calcifications >35% with prominent syncytial knots			P			NAIS	NR
	Case 27	NR	Chorioamnionitis with prominent decidual abscesses		P				NAIS	NR
Study 9	Case 28	Reduced fetal movement, MSL, elevated nucleated red blood cells	Multiple thrombosed chorangiomas, small hemangiomas					Chorangiomatosis	NAIS	CP
Study 10	Case 29	NR	Staphylococcus positive on fetal side of placenta					Staphylococcus positive on fetal side of placenta	NAIS	NR
	Case 30	Preterm	Funisitis		P				NAIS	NR
	Case 31		Acute chorioamnionitis, placental infarction		P	P			NAIS	NR
Total	31			5/31 ¼ 16%	17/31 ¼ 55%	11/31 ¼ 35%	0/31 ¼ Nil	8/31 ¼ 26%	NAIS¼24 CSVT¼5 Combination (NAISþCSVT)¼2	

Abbreviations: APH, ante partum hemorrhage; CHD, congenital heart disease; CS, cesarean section; CSVT, cerebral sinovenous thrombosis; CTG, cardiotocography; DIC, disseminated intravascular coagulopathy; FHR, fetal heart rate; FM, fetal movement; FTH, fetal thrombotic vasculopathy; FTV, fetal thrombotic vasculopathy; GBS, group B *Streptococcus*; GDM, gestational diabetes mellitus; IUGR,

intrauterine growth restriction; MSL, meconium-stained liquor; NAIS, neonatal arterial ischemic stroke; NR, not reported; PE, pre-eclampsia; PROM, prolonged rupture of membrane; US, ultrasonography.
[a]Study 1 could not be classified as per Redline's classification as the placental findings were not reported on case basis, hence not included in Table 2. The underline denotes a potentially causative risk factor identified by the placental pathologist author (SA).
[b]Redline's placental lesion has 4 categories[27,33]:
Category 1: Sudden catastrophic event: retroplacental hematoma and acute umbilical cord occlusion by thrombosis, true cord knots, cord overcoiling or abnormal cord insertion sites.
Category 2: Thrombo-inflammatory process: chorioamnionitis, villitis, chorionic vessel and stem vessel thrombi, and avascular fibrotic villi.
Category 3: Decreased placental reserve: defined by 2 or more of either multiple placental infarcts, distal villous immaturity, and placental weight <10th percentile for gestational age.
Category 4: Adaptive response to stressful intrauterine environment: increased fetal nucleated red blood cells or villous chorangiosis.
[c]Other: Placental pathology which could not be included in the Redline's categories.

abstracts). Both reviewers unanimously agreed on the 10 studies that met the inclusion criteria for analysis; thus, no arbitration of disputes was required (Figure 1). Ten studies published between 2000 and 2019 met the inclusion criteria (Table 1). All of the studies were conducted in high-income countries, and only 1 study was prospective (study 8). The studies consisted of 349 perinatal stroke cases from the period 1992-2015. Placental pathology reports were available for 24% (83/349) of the perinatal stroke cases. Of these, 87% (72/83) cases had abnormal placental pathology. In 31 cases, the type of stroke and placental pathology was reported on an individual case basis (Table 2).

Stroke

The most common type of perinatal stroke was neonatal arterial ischemic stroke (24/31; 77%). Category 2 of Redline's placental lesion was the most frequently reported placental abnormality (17/31; 55%). Most studies primarily focused on perinatal stroke cases. Perinatal stroke types were inconsistently classified. Studies 3 and 4 reported cases of fetal thrombotic vasculopathy and types of perinatal brain injuries, respectively. These 2 studies included perinatal stroke cases that were extracted and analyzed as per the study design. The characteristics of the selected studies are shown in Table 1. The studies used a standard definition of perinatal stroke applicable to the year they were published, although various nomenclatures were used to describe the type of stroke.[2] The studies included fetus and preterm and term infants up to 28 postnatal days. Some studies only reported data at the group level and not on an individual case basis.

Risk Factors

Of the 31 cases of stroke where the patients could be identified on an individual case-by-case basis (Table 2), 19 cases suggested both non-placental and placental associations related to perinatal stroke. The common nonplacental associations were caesarean section (n ¼ 8/31; 26%), abnormal fetal heart rate (n ¼ 5/31; 16%), anemia (n ¼ 5/31; 16%), and congenital heart disease (n ¼ 4/31; 13%).

Placenta

Placental findings (Table 2) were described at the individual case level for studies 2 to 10. Findings were also categorized using Redline's classification of placental histology, by BR and an independent expert pathology reviewer (SA). We intended to classify by the more contemporary and uniform comprehensive placental histopathology classification—the Amsterdam Placental Workshop Group Consensus Statement[34]—but could not, because the included studies were conducted and coded in the Redline classification era and data were insufficient for recoding.

For studies 5, 8, and 10, the frequency of placental histopathologic examination was 6% (n ¼ 12/186), 13% (n ¼ 8/60), and 8% (n ¼ 3/40), respectively. Abnormal placental pathology for study 5 was 83% (n ¼ 10/12), study 8 was 88% (n ¼ 7/8), and study 10 was 100% (n ¼ 3/3).

Of the 2 case-control studies (studies 1 and 10), study 1 found 89% (n ¼ 41/46) abnormal placental pathology among the cases, compared with 62% (n ¼ 61/99) in controls with the reported *P* value of <.001 (cases vs controls). Of note, study 10 found 100% abnormal placental pathology among the cases (n ¼ 3/3) compared with 45% in controls (n ¼ 5/11).

Placental Classifications

Studies 3 and 5 used Redline's placental histology classification[27,33] and study 1 adapted predefined categories of placental abnormality based on a local protocol. There was little uniformity in describing placental pathology across the other studies. To enable comparisons within this review, the placental reports were classified for studies 2 to 10 using Redline's classification (Table 2), and author SA had the final authority in case of any discrepancy.[36] Study 1 cases could not be accommodated within Redline's classification as the placental findings were not reported on an individual case basis.

Some of the placental abnormalities did not meet any of the Redline categories or were lacking sufficient information to permit any coding and thus these events were coded as "other" (Table 2) within our systematic review. The "other" category included (1) placental hypotrophy (case 1); (2) small abnormal placenta (case 25); (3) retroplacental hematoma (cases 1 and 15), which does not necessarily represent abruption and without clinical history or information regarding the chronicity is a common nonspecific finding post placental delivery; (4) chorangioma or chorangiomatosis (cases 20 and 28); (5) fibrous plaques with increased intervillous fibrin (case 21); (6) circumvallate insertion (case 23), which is more commonly associated with a chronic rather than an acute catastrophic event; and (7) Staphylococcal positive on the fetal side of the placenta (case 29) with no inflammatory response, which is likely to be a contaminant.

We also present a detailed description of the association between the type of placental pathology and type of stroke in Table 2. There were no statistically significant associations between the types of stroke and Redline's placental classification codes: category 1 (*P* ¼ .404), category 2 (*P* ¼ .102), category 3 (*P* ¼ .441), or "other" (*P* ¼ .494). Category 4 could not be analyzed statistically as there were no placental data coded with this placental abnormality.

Discussion

Perinatal stroke occurs because of a constellation of risk factors rather than an isolated event and can also be viewed as a cascading causal pathway.[37,38] The placenta is a very vascular gestational organ that is responsible for blood flow to and from the fetus and is likely to have a pivotal role in the causal pathway of perinatal stroke.[25,39]

Our review investigated the relationship between placental pathology and perinatal stroke among the multitude of possible associations and putative risk factors. We identified 10 studies (n ¼ 349) of varying levels of evidence and potential sources of bias (levels 1, 2, 3, and 4). We found that there was a low frequency of placental examinations within the included studies, even for developed countries, 6%-13% (studies 5, 8, and 10). Nevertheless, when pathology was ordered, the placental pathology was often abnormal. The findings need to be interpreted cautiously as the frequency of placental histopathologic examination was low (studies 2, 5, 8, and 10), in contrast to the studies that directly sought cases of placental pathology (studies 1, 3, 4, 6, 7, and 9).

Plausible reasons for not ordering pathology were nonspecific and may include delayed clinical presentations of perinatal stroke, cost of placental storage and examination, failure of treating physicians to request placental examination, and the common practice of discarding the placenta soon after birth before it is recognized as an important pathology. Furthermore, we found that of the total placental examinations, 87% (Table 1; 72/83) had abnormal placental pathology. The abnormal yield was as follows: 55% (Table 2; 17/31) had a thrombo-inflammatory process at work, 32% (10/31) had decreased placental reserve, 16% (5/31) had a sudden catastrophic event and 19% (6/31) had associated placental risk factors.

We surmise that placental pathologies such as a sudden catastrophic event causing blood flow obstruction to the fetus (Redline's category I), or placental thrombo-inflammatory process (Redline's category II), or decreased placental reserve (Redline's category III) may have caused interruption of cerebral blood flow, thus giving rise to perinatal stroke. In our view, adaptive response to a stressful intrauterine environment (Redline's category IV) may be a fetal stress response to categories I, II, or III and therefore should be considered as a related pathology rather than an independent pathologic entity.

We sought to differentiate between the etiologic and circumstantial risk factors in order to identify the true causal pathway of stroke; for example, perinatal stroke due to thrombus from protein C deficiency (a protein that prevents blood clotting) or due to thrombus and fibrin formation in case of chorioamnionitis. The pathologist author proposed plausible causative factors (see Table 2) but these require confirmation in further research using rigorous methodologies.

It was also noted that, across the studies, perinatal stroke was seldom reported in the same manner. For example, neonatal arterial ischemic stroke was used as a synonym for arterial ischemic stroke, ischemic perinatal stroke, perinatal arterial stroke, and middle cerebral artery stroke. Future research should aim to harmonize terminology to enable aggregation of data.

Limitations and Recommendations:

Reporting quantity and quality. The major limitations of this review were sparse placental examinations, lack of standardization, and nonhomogenous reporting of placental pathology, which precluded aggregation of data. Comparisons between studies could be advanced if standardized methods of reporting key placental lesions such as a catastrophic event, placental infection, or placental adaptive response to stressful intrauterine environment were implemented within clinical care.

Reporting processes. Chorioamnionitis, a known risk factor for cerebral palsy, was the most prevalent risk factor identified in this systematic review, although more clarification of clinical, histologic, associated funisitis, villitis, or more importantly intravascular thrombi abnormalities would have been clinically and etiologically informative.[40] Thus, moving forward, we recommend that placental findings are reported synergistically for expert pathologists and clinicians to further elucidate the etiology and for clinical management of perinatal stroke. We also recommend that all placentas should be stored for 72 hours to create the opportunity for expert histopathologic examination.

Classification and nomenclature. We note that Redline's placental histology classification describes the actual effect rather than the etiology of the placental pathology.[36] For example, cate- gory I describes both the event and the cause, whereas category IV codes increased nucleated red blood cells, which may be a secondary downstream event to any of the abnormalities described in categories I, II, or III. In future, widespread adop- tion of the placental histopathology classification as per the Amsterdam Placental Workshop Group Consensus Statement is likely to yield a clear correlation between placental abnorm- ality and perinatal stroke. Another limitation was the hetero- geneity in the nomenclature of stroke type, which made it difficult to make definitive comparisons between the type of stroke and type of placental pathology. We therefore recom- mend adoption of the contemporary stroke nomenclature.[2]

Etiology and prevention. Proactive prevention of perinatal stroke is a research priority of both parents and clinicians. To make the paradigm shift toward identifying ways to prevent perinatal stroke, we recommend that the field makes a systematic and rigorous effort to elucidate the etiologic role of the placenta in cases of acute symptomatic perinatal stroke, ultimately aiming to identify novel treatment targets. An adequately powered case-control study, with standardized classification of placental pathology, while reporting abnormal placental pathology as well as a standardized nomenclature for stroke type, is needed to confirm a meaningful association between abnormal placental pathology and specific types of perinatal stroke.

Conclusion

In conclusion, placental abnormality appeared more common among children with perinatal stroke than controls. This result must be interpreted cautiously because of the low frequency of placental examination in published cases and the lack of uniformity in placental histopathology reporting. More research is required to verify the precise role of the placenta in the multifactorial etiology of perinatal stroke.

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Author Contributions

BR conceptualized and designed the study, designed the data collection instruments, collected data (first extractor), carried out the initial analyses, and drafted the initial manuscript. IN collected data (second extractor), assisted with the analyses, and critically reviewed the manuscript for important intellectual content. SA (Pathologist) assisted with the designing of data collection instruments, and assisted with interpretation and analyses of placental pathology. CG assisted with statistical analysis and interpretation of data, contributed to drafting and revising it critically for final approval. CM, KW, and NB contributed to the analysis and interpretation of data and critically reviewed the manuscript for important intellectual content.
All authors reviewed and revised the manuscript and approved the final version for publishing.

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Ethical Approval

Ethical approval was not required as this study entailed review of published literature.

References

1. Raju TN, Nelson KB, Ferriero D, Lynch JK, Participants N-NPSW. Ischemic perinatal stroke: summary of a workshop sponsored by the National Institute of Child Health and Human Development and the National Institute of Neurological Disorders and Stroke. *Pediatrics*. 2007;120(3):609-616.
2. Dunbar M, Kirton A. Perinatal stroke: mechanisms, management, and outcomes of early cerebrovascular brain injury. *Lancet Child Adolesc Health*. 2018;2(9):666-676.
3. Krishnamoorthy KS, Soman TB, Takeoka M, Schaefer PW. Diffusion-weighted imaging in neonatal cerebral infarction: clinical utility and follow-up. *J Child Neurol*. 2000;15(9):592-602.
4. Nelson KB, Lynch JK. Stroke in newborn infants. *Lancet Neurol*. 2004;3(3):150-158.
5. Laugesaar R, Kolk A, Tomberg T, et al. Acutely and retrospectively diagnosed perinatal stroke: a population-based study. *Stroke*. 2007;38(8):2234-2240.
6. Chabrier S, Husson B, Dinomais M, Landrieu P, Nguyen The Tich S. New insights (and new interrogations) in perinatal arterial ischemic stroke. *Thromb Res*. 2011;127(1):13-22.
7. Giraud A, Guiraut C, Chevin M, Chabrier S, Sebire G. Role of perinatal inflammation in neonatal arterial ischemic stroke. *Frontiers Neurol*. 2017;8:612.
8. Harteman JC, Groenendaal F, Kwee A, Welsing PMJ, Benders MJNL, De Vries LS. Risk factors for perinatal arterial ischaemic stroke in full-term infants: a case-control study. *Arch Dis Child Fetal Neonat Ed*. 2012;97(6):F411-F416.
9. Darmency-Stamboul V, Chantegret C, Ferdynus C, et al. Antenatal factors associated with perinatal arterial ischemic stroke. *Stroke*. 2012;43(9):2307-2312.
10. Heier LA, Carpanzano CR, Mast J, Brill PW, Winchester P, Deck MDF. Maternal cocaine abuse: the spectrum of radiologic abnormalities in the neonatal CNS. *Am J Neuroradiol*. 1991; 12(5):951-956.
11. Nelson KB. Thrombophilias, perinatal stroke, and cerebral palsy. *Clin Obstet Gynecol*. 2006;49(4):875-884.
12. Govaert P, Ramenghi L, Taal R, Dudink J, Lequin M. Diagnosis of perinatal stroke II: mechanisms and clinical phenotypes. *Acta Paediatr*. 2009;98(11):1720-1726.
13. Simchen MJ, Goldstein G, Lubetsky A, Strauss T, Schiff E, Kenet G. Factor V Leiden and antiphospholipid antibodies in either mothers or infants increase the risk for perinatal arterial ischemic stroke. *Stroke*. 2009;40(1):65-70.
14. Kenet G, Lutkhoff LK, Albisetti M, et al. Impact of thrombophilia on risk of arterial ischemic stroke or cerebral sinovenous thrombosis in neonates and children: a systematic review and meta-analysis of observational studies. *Circulation*. 2010;121(16):1838-1847.
15. McQuillen PS, Miller SP. Congenital heart disease and brain development. *Ann N Y Acad Sci*. 2010;1184:68-86.
16. Lee J, Croen LA, Lindan C, et al. Predictors of outcome in perinatal arterial stroke: a population-based study. *Ann Neurol*. 2005; 58(2):303-308.
17. Roach ES, Golomb MR, Adams R, et al. Management of stroke in infants and children: a scientific statement from a special writing group of the American Heart Association Stroke Council and the Council on Cardiovascular Disease in the Young. *Stroke*. 2008; 39(9):2644-2691.
18. Schlatterer SD, Harris KN, Scafidi J, Tsuchida T, Chang T. Risk factors and etiology of perinatal strokes-a single center ten year retrospective series. *Ann Neurol*. 2015;78:S216.
19. Mineyko A, Kirton A. The black box of perinatal ischemic stroke pathogenesis. *J Child Neurol*. 2011;26(9):1154-1162.
20. Stroup DF, Berlin JA, Morton SC, et al. Meta-analysis of observational studies in epidemiology: a proposal for reporting. Meta-analysis of Observational Studies in Epidemiology (MOOSE) group. *JAMA*. 2000;283(15):2008-2012.
21. PRISMA. PRISMA 2009 checklist. http://prisma-statement.org/documents/PRISMA%202009%20flow%20diagram.pdf. Published 2009. Accessed January 13, 2020.
22. OCEBM levels of Evidence Working group. The Oxford 2011 Levels of Evidence. https://www.cebm.net/wp-content/uploads/2014/06/CEBM-Levels-of-Evidence-2.1.pdf. Published 2013. Accessed January 13, 2020.
23. Bernson-Leung ME, Boyd TK, Meserve EE, et al. Placental pathology in neonatal stroke: a retrospective case-control study. *J Pediatr*. 2018;195:39-47.e5.

24. Fluss J, Garcia-Tarodo S, Granier M, et al. Perinatal arterial ischemic stroke related to carotid artery occlusion. *Eur J Paediatr Neurol*. 2016;20(4):639-648.
25. Magnetti F, Bagna R, Botta G, et al. Fetal thrombotic vasculopathy and perinatal thrombosis: should all placentas be examined? *Am J Perinatol*. 2014;31(8):695-700.
26. Takenouchi T, Kasdorf E, Engel M, Grunebaum A, Perlman JM. Changing pattern of perinatal brain injury in term infants in recent years. *Pediatr Neurol*. 2012;46(2):106-110.
27. Elbers J, Viero S, MacGregor D, DeVeber G, Moore AM. Placental pathology in neonatal stroke. *Pediatrics*. 2011;127(3): e722-e729.
28. Dueck CC, Grynspan D, Eisenstat DD, Caces R, Rafay MF. Ischemic perinatal stroke secondary to chorioamnionitis: a histopathological case presentation. *J Child Neurol*. 2009;24(12):1557-1560.
29. Das S, Ankola P, Chiechi M, Sandhu J. Perinatal cerebral arterial infarction associated with a placental chorioangioma. *Am J Perinatol*. 2008;25(6):381-383.
30. Curry CJ, Bhullar S, Holmes J, Delozier CD, Roeder ER, Hutchison HT. Risk factors for perinatal arterial stroke: a study of 60 mother-child pairs. *Pediatr Neurol*. 2007;37(2):99-107.
31. Ghidini A, Locatelli A. Diffuse placental chorioangiomatosis causing multiple fetal cerebral embolism: a case report. *J Reprod Med*. 2006;51(4):321-324.
32. Lee J, Croen LA, Backstrand KH, et al. Maternal and infant characteristics associated with perinatal arterial stroke in the infant. *JAMA*. 2005;293(6):723-729.
33. Redline RW. Placental pathology and cerebral palsy. *Clin Perinatol*. 2006;33(2):503-516.
34. Khong TY, Mooney EE, Ariel I, et al. Sampling and definitions of placental lesions: Amsterdam Placental Workshop Group Consensus Statement. *Arch Pathol Lab Med*. 2016;140(7):698-713.
35. Naqvi M, Scourtas A, Hanmer K, Khattaby I, Roberts D. Placental pathology and ischemic perinatal stroke. *Reproduct Sci* 2016;1: 325A-326A.
36. Redline RW. Classification of placental lesions. *Am J Obstet Gynecol*. 2015;213(4)(suppl):S21-S28.
37. Kraus FT.Fetal thrombotic vasculopathy. Perinatal stroke, growth restriction, and other sequelae. *Surg Pathol Clin*. 2013;6(1): 87-100.
38. Riel-Romero RMS. Neonatal stroke. *Neurol Res*. 2008;30(8): 839-844.
39. Redline RW. Placental pathology: a systematic approach with clinical correlations. *Placenta*. 2008;29(Suppl A): S86-S91.
40. Shi Z, Ma L, Luo K, et al. Chorioamnionitis in the development of cerebral palsy: a meta-analysis and systematic review. *Pediatrics*. 2017;139(6):e20163781.

Chapter 3: THE REAL STORY OF THE PLACENTA IN NEONATAL CARE

3.1 Introduction

The placenta is an important source of information about many neonatal conditions, but it is underutilised in clinical practice and research. Some of the reasons are the lack of awareness among physicians and the cost to the healthcare system of routine examinations.

This second study examines the frequency of placental examinations in real-world neonatal practice. An audit over a 2-year period found placental examination was performed for 8% of the total live births in a single hospital setting. Of these, 59% were for high-risk pregnancies such as those resulting in low birth weight (< 2500 grams). Low birth weight is a known cause of neonatal morbidity and this study revealed 78% of the low-birth-weight babies had abnormal placental histopathology. This was an important clinical finding for exploring the pathogenesis and prognosis of babies with low birth weight.

This study was the focus of a quality improvement project to increase the frequency of placental examination through the development of a hospital policy on placental screening.

3.2 Published Manuscript

Roy B, Jeannette G, Adrian K, Karen W, Catherine M, Webb A, Nadia B, Iona N. Routine placental histopathlogical examination: provides answers in neonatal management. American Journal of Pediatrics 2022; 8(1):10-13.

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Routine Placental Histopathological Examination: Provides Answers in Neonatal Management

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Abstract: Placental abnormality may affect the fetus adversely. The purpose of this study was to identify the prevalence of placental histopathological examination in a private hospital setting and analyse the placental histopathology findings for high-risk pregnancies. A retrospective cross-sectional study was conducted at the Mater Hospital in Sydney from January 2018 to June 2020. The placental histopathology was classified as per the 2014 Amsterdam Placental Workshop Group criteria, enabling uniformity for analysis. There were 5594 live births during the study period. Of these, 5% (256/5594) were low birth weight (LBW). Placental histopathology was conducted for 8% (443/5594) of the live births and 59% (152/256) of LBW births. The LBW group was subclassified into small for gestation (SGA) (n=66) and non-SGA (n=86) to analyse differences in placental abnormalities between the two groups. Of SGA, 82% (54/66) had placental abnormality compared to 76% (65/86) for non-SGA. Intervillous fibrin deposits ($p=0.013$) and smaller placental weight ($p=0.008$) were more common in the SGA; whereas the placental inflammatory-immune process was more common in the non-SGA. Original placental histopathology reports did not employ the objective Amsterdam classification system, thereby risking subjective or variable interpretation by clinicians. In conclusion, placental histopathology plays an important role in neonatal management. A quality improvement project may improve the prevalence of placental histopathological examination.

Keywords: Low Birth Weight (LBW), Placental Histopathology, Placental Vascular Malperfusion

1. Introduction

Placental abnormality may adversely affect the fetus, as the placenta provides oxygen and nutrients, as well as removes waste products from the growing fetus [1, 2]. Therefore, investigations into placental abnormality may be important in the etiology of neonatal sepsis, birth asphyxia, preterm delivery, intrauterine growth restriction (IUGR) and cerebral palsy [3-6]. They could also offer vital clues to the cause of fetal and neonatal deaths and provide physical evidence for medico-legal assessments [7]. Despite these benefits, a comprehensive examination of the placenta is not part of standard practice in many hospitals. This could be due to challenges such as limited storage facilities for placentas, burden on anatomical pathologists, cost to the healthcare

system or simply a lack of specific guidelines for individual hospitals.

Placental examination has been standardised by the 2014 Amsterdam Placental Workshop Group criteria and includes: a) macroscopic examination of placental shape, placental weight, umbilical cord insertion, umbilical blood vessels, placental abruption and umbilical membrane; b) histopathological examination to identify placental infarction, calcification, and signs of inflammation; and c) microbiological examination to isolate specific bacterial or viral infections in neonates [8].

We undertook this study at the Mater Hospital in Sydney, Australia, with the aim of identifying the prevalence of placental histopathological examination for live births in a private hospital setting. We particularly focused on those with low birth weight (LBW) since LBW is one of the most common pregnancy complications and is associated with increased risk of still birth and cerebral palsy (CP) [9]. The Mater Hospital is a private multispecialty hospital with a Level 5 maternity service that allows preterm deliveries up to 32 weeks gestation [10]. There are 2500–3000 deliveries annually. At the time of the study, the placental histopathological examinations were conducted at the discretion of the clinicians (obstetrician and/ or neonatologist) at the hospital's affiliated pathological laboratory.

2. Method

2.1. Study Design

The retrospective cross-sectional study was conducted from January 2018 to June 2020 for all live birth deliveries that had placental histopathological examination. The study also included an itemised placental histopathology analysis of the LBW infants. The LBW infants were subclassified into small for gestational age (SGA) and non-SGA. Low birth weight is defined as birth weight less than 2500g [11]. Small for gestational age is a birth weight<10th percentile and non-SGA is a birth weight>10th percentile.

2.2. Data Source

We retrieved data from the hospital electronic database that stores patient records.

2.3. Study Selection

Cases of all gestational ages, including both singletons and multiples, were included if following criteria were met:

1. Live birth at the Mater Hospital;
2. Placental histopathology report available.

2.4. Data Extraction

Data was extracted using the investigator-designed extraction form based on key variables in the published literature, which included: neonatal birth weight; gestational age; diagnosis; and a placental histopathology report. The placental histopathology reports were categorized by the primary investigator into the 2014 Amsterdam Placental Workshop Group criteria, to enable uniformity and standardisation of the analysis [8, 12].

2.5. Statistics

The demographics of the SGA and non-SGA groups were compared using chi-squared tests of independence and independent sample *t*-tests. The placental histopathological classifications of the SGA and non-SGA groups were compared using chi-squared tests of independence or Fisher's exact tests where subcategories had $n<5$. A p-value of<0.05 was considered statistically significant.

3. Results

There were 5594 live births in the 30-month study period, of which 5% (256/5594) were LBW infants (Figure 1). Placental histopathological examination was available for 8% (n=443/5594) of live births and 59% (n=152/256) of LBW infants. Of the 152 LBW infants, 66 were SGA and 86 were non-SGA. The patient characteristics are shown in Table 1. Both SGA and non-SGA groups were homogenous except for the gestational age; average gestation for SGA was 37 weeks and for non-SGA it was 34 weeks ($p<0.001$).

Seventy eight percent (119/152) of the LBW infants had placental abnormality and, of these, 82% (54/66) were SGA and 76% (65/86) were non-SGA (Table 1). As per the 2014 Amsterdam Placental Workshop Group criteria, the placental abnormalities fell into three broad categories: vascular lesions; inflammatory process; and other placental processes [8, 12]. Twenty nine percent (34/119) had one type of placental abnormality while 71% (84/119) had more than one type (Table 2).

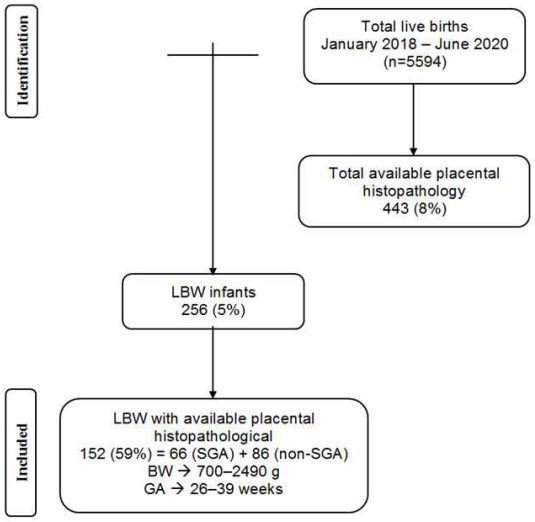

Figure 1. *Patient demographics.*

BW – birth weight; GA – gestational age;
LBW – low birth weight; SGA – small for gestation

Table 1. *Demographics – SGA vs non-SGA.*

	SGA	non-SGA	*p* value
n (%)	66 (43%)	86 (57%)	
Gestational age – average (range; weeks)	37 (26–39)	34 (27–36)	<0.001
Birth weight – average (range; grams)	2224 g (700–2490 g)	2133 g (1020–2490 g)	0.052
male (%): female (%)	26, 40 (39%: 61%)	37, 49 (43%: 57%)	0.653
Placental abnormalities	54/66 (82%)	65/86 (76%)	0.355
one abnormality	15/54 (28%)	19/65 (29%)	0.861
>1 abnormality	39/54 (72%)	46/65 (71%)	

SGA – small for gestation

Table 2. *Placental histopathological classification: 2014 Amsterdam Placental Workshop Group criteria.*

		SGA	non-SGA	*p* value
1	Placental vascular processes			
	a. Maternal stromal-vascular lesions			
	I. Developmental	2/66 (3%)	3/86 (4%)	1.000
	II. Malperfusion	21/66 (32%)	25/86 (29%)	0.715
	1) Placental infarction	10/21 (48%)	12/25 (48%)	0.979
	2) Syncytial knots	10/21 (48%)	6/25 (24%)	0.094
	3) Distal villous hypoplasia	2/21 (10%)	6/25 (8%)	0.260
	4) Fibrin deposition	0	2/25 (8%)	0.493
	III. Loss of integrity	6/66 (9%)	10/86 (12%)	0.613
	b. Fetal stromal-vascular lesions			
	I. Developmental	3/66 (5%)	1/86 (1%)	0.580
	II. Malperfusion	31/66 (47%)	32/86 (37%)	0.226
	1) Intervillous fibrin deposit	8/31 (26%)	1/32 (3%)	0.013
	2) Intervillous thrombus	5/31 (16%)	8/32 (25%)	0.384
	3) Dystrophic calcification	12/31 (39%)	8/32 (25%)	0.243
	III. Loss of integrity	2/66 (3%)	0	0.187
2	Placental inflammatory-immune processes	8/66 (12%)	21/86 (24%)	0.108
3	Other placental processes			
	a. Abnormal cord insertion	5/66 (8%)	6/86 (7%)	0.888
	b. Single umbilical artery	2/66 (3%)	3/86 (4%)	1.000
	c. Small placental weight	23/66 (35%)	14/86 (16%)	0.008
	d. Large placental weight	0	4/86 (5%)	0.133

4. Discussion

Placental histopathology is integral to diagnosis and management in obstetric and neonatal care [13, 14]. However, the discretionary logistics and costs of placental storage and histopathological examination places a burden on the healthcare system, which limits the number of placentas clinicians send to the pathologists. At the Mater Hospital there was no provision for routine storage of the placenta and the decision to order placental histopathological examination was by the clinician on a case-by-case basis. However, in the case of an obvious risk factor, such as low birth weight, preterm delivery, multiple births etc., a higher number of placental histopathology tests were ordered, suggesting cases were missed if there wasn't an apparent risk factor.

The placental histopathological examination was much higher for the LBW infants compared to overall infants (59% versus 8%). Low birth weight could be due to prematurity or growth retardation, or both. Placental analysis was reported separately for SGA and non-SGA LBW infants because growth-retarded infants have a worse prognosis due to poor catch-up growth rate [15]. Incidentally there were more preterm infants in the non- SGA group ($p<0.001$).

Placental vascular malperfusion was the most common placental histopathological abnormality in both groups. Small for gestation and non-SGA infants had similar placental histopathological abnormality, except that intervillous fibrin deposit was more common in the SGA group ($p=0.013$), suggesting increased placental vascular flow abnormality as one of the causes of growth retardation [16]. Likewise, more SGA infants had smaller placental weight compared to non-SGA infants ($p=0.008$) [17, 18].

The limitations of this study include an inherent selection bias, as the request for placental examination was based on individual clinician discretion rather than on standardised criteria. Another limitation was that the placental histopathology report did not precisely follow the 2014 Amsterdam Placental Workshop Group criteria classification system, which left the interpretation open to the clinicians.

The results of this study led to significant knowledge translation strategies and practice change being considered at

the Mater Hospital in Sydney. This includes developing a placental storage and management policy that outlines specific criteria for ordering placental pathological examination. In addition, the hospital is considering routine storage of placentas for 48 hours, which is particularly important in neonatal sepsis as it allows the placenta to be sent for histopathological examination, to explore the possibility of chorioamnionitis.

5. Conclusion

Information regarding placental abnormality could be a missing link in neonatal management. Therefore, placental histopathological examination should be included as standard practice where clinically indicated.

This study was the subject of a quality improvement project that aimed to improve the incidence of placental examination. The next step in continuous quality improvement would be to devise and implement a standard placental histopathology report, to facilitate correct interpretation and clinical application of the placental histopathological findings by the clinician.

Conflict of Interest

The authors have no conflicts of interest to disclose.

Financial Disclosure

There are no financial relationships relevant to this article to disclose.

References

[1] Redline RW. Placental pathology: a systematic approach with clinical correlations. Review. *Placenta*. Mar 2008; 29 Suppl A: S86-91. doi: https://dx.doi.org/10.1016/j.placenta.2007.09.003.

[2] Jaiman S, Romero R, Pacora P, et al. Disorders of placental villous maturation are present in one-third of cases with spontaneous preterm labor. *Journal of Perinatal Medicine*. May 26 2021; 49 (4): 412-430. doi: https://dx.doi.org/10.1515/jpm-2020-0138.

[3] Spinillo A, Gardella B, Muscettola G, Cesari S, Fiandrino G, Tzialla C. The impact of placental massive perivillous fibrin deposition on neonatal outcome in pregnancies complicated by fetal growth restriction. *Placenta*. 11 2019; 87: 46-52. doi: https://dx.doi.org/10.1016/j.placenta.2019.09.007.

[4] Shannon P, Hum C, Parks T, et al. Brain and Placental Pathology in Fetal COL4A1 Related Disease. Case Reports. *Pediatr Dev Pathol*. May-Jun 2021; 24 (3): 175-186. doi: https://dx.doi.org/10.1177/1093526620984083.

[5] Chalak L, Redline RW, Goodman AM, et al. Acute and Chronic Placental Abnormalities in a Multicenter Cohort of Newborn Infants with Hypoxic-Ischemic Encephalopathy. Clinical Trial, Phase III Multicenter Study Randomized Controlled Trial. *J Pediatr*. Oct 2021; 237: 190-196. doi: https://dx.doi.org/10.1016/j.jpeds.2021.06.023.

[6] Han X, Du H, Cao Y, et al. Association of histological and clinical chorioamnionitis with perinatal and neonatal outcome. *J Matern Fetal Neonatal Med*. Mar 2021; 34 (5): 794-802. doi: https://dx.doi.org/10.1080/14767058.2019.1618824.

[7] Mittal N, Byard RW, Dahlstrom JE. A practical guide to placental examination for forensic pathologists. Review. *Forensic Sci Med Pathol*. 06 2020; 16 (2): 295-312. doi: https://dx.doi.org/10.1007/s12024-019-00214-2.

[8] Khong TY, Mooney EE, Ariel I, et al. Sampling and Definitions of Placental Lesions: Amsterdam Placental Workshop Group Consensus Statement. *Arch Pathol Lab Med*. Jul 2016; 140 (7): 698-713. doi: https://dx.doi.org/10.5858/arpa.2015-0225-CC.

[9] Nkwabong E, Kamgnia Nounemi N, Sando Z, Mbu RE, Mbede J. Risk factors and placental histopathological findings of term born low birth weight neonates. *Placenta*. Feb 2015; 36 (2): 138-41. doi: https://dx.doi.org/10.1016/j.placenta.2014.12.005.

[10] NSW Ministry of Health. NSW Maternity and Neonatal Service Capability Framework. Accessed 13 August 2021, https://www1.health.nsw.gov.au/pds/ActivePDSDocuments/GL2016_018.pdf.

[11] World Health O. ICD-10: international statistical classification of diseases and related health problems: tenth revision. 2nd ed ed. Geneva: World Health Organization; 2004.

[12] Redline RW. Classification of placental lesions. Review. *Am J Obstet Gynecol*. Oct 2015; 213 (4 Suppl): S21-8. doi: https://dx.doi.org/10.1016/j.ajog.2015.05.056.

[13] Ravishankar S, Redline RW. What Obstetricians Need to Know About Placental Pathology. Review. *Obstet Gynecol Clin North Am*. Mar 2020; 47 (1): 29-48. doi: https://dx.doi.org/10.1016/j.ogc.2019.10.007.

[14] Fouks Y, Many A, Shulman Y, Bak S, Shinar S. The Contribution of an Infectious Workup in Understanding Stillbirth. *Am J Perinatol*. 03 2021; 38 (4): 377-382. doi: https://dx.doi.org/10.1055/s-0039-1697668.

[15] Tachibana M, Nakayama M, Ida S, et al. Pathological examination of the placenta in small for gestational age (SGA) children with or without postnatal catch-up growth. *J Matern Fetal Neonatal Med*. Mar 2016; 29 (6): 982-6. doi: https://dx.doi.org/10.3109/14767058.2015.1029911.

[16] Stergiotou I, Bijnens B, Cruz-Lemini M, Figueras F, Gratacos E, Crispi F. Maternal subclinical vascular changes in fetal growth restriction with and without pre-eclampsia. Research Support, Non-U.S. Gov't. *Ultrasound Obstet Gynecol*. Dec 2015; 46 (6): 706-12. doi: https://dx.doi.org/10.1002/uog.14815.

[17] Benton SJ, McCowan LM, Heazell AE, et al. Placental growth factor as a marker of fetal growth restriction caused by placental dysfunction. Research Support, Non-U.S. Gov't. *Placenta*. 06 2016; 42: 1-8. doi: https://dx.doi.org/10.1016/j.placenta.2016.03.010.

[18] Strand KM, Andersen GL, Haavaldsen C, Vik T, Eskild A. Association of placental weight with cerebral palsy: population-based cohort study in Norway. *Bjog*. Dec 2016; 123 (13): 2131-2138. doi: https://dx.doi.org/10.1111/1471-0528.13827.

Chapter 4: SYSTEMATIC REVIEW OF PREMATURITY AND PERINATAL STROKE

4.1 Introduction

The pathogenesis of perinatal stroke in preterm infants is unclear given the differences in brain maturation and injury mechanisms unique to the immature brain. This study investigated the epidemiology and aetiology of stroke in preterm infants.

The systematic review confirmed the lack of information regarding the aetiology and the precise causal pathway of stroke in preterm infants. The review identified that seizure was not the most common clinical presentation of stroke in preterm infants (unlike full-term infants) and cranial ultrasound was the key screening tool for diagnosis, followed by brain MRI. The three most common types of strokes in preterm infants were: periventricular haemorrhagic infarction (PVHI); perinatal arterial ischemic stroke (PAIS); and cerebral sinovenous thrombosis (CSVT).

Amalgamated reports of preterm and full-term infants presented challenges in isolating the preterm data and consequently interpreting the results.

4.2 Published Manuscript

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Review

Epidemiology and pathogenesis of stroke in preterm infants: A systematic review

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Abstract.
BACKGROUND: Perinatal stroke is one of the principal causes of cerebral palsy (CP) in preterm infants. Stroke in preterm infants is different from stroke in term infants, given the differences in brain maturation and the mechanisms of injury exclusive to the immature brain. We conducted a systematic review to explore the epidemiology and pathogenesis of periventricular hemorrhagic infarction (PVHI), perinatal arterial ischemic stroke (PAIS) and cerebral sinovenous thrombosis (CSVT) in preterm infants.
METHODS: Studies were identified based on predefined study criteria from MEDLINE, EMBASE, SCOPUS and WEB OF SCIENCE electronic databases from 2000 – 2019. Results were combined using descriptive statistics.
RESULTS: Fourteen studies encompassed 546 stroke cases in preterm infants between 23 – 36 weeks gestational ages and birth weights between 450 – 3500 grams. Eighty percent (436/546) of the stroke cases were PVHI, 17% (93/546) were PAIS and 3% (17/546) were CSVT. Parietal PVHI was more common than temporal and frontal lobe PVHI. For PAIS, left middle cerebral artery (MCA) was more common than right MCA or cerebellar stroke. For CSVT partial or complete thrombosis in the transverse sinus was universal. All cases included multiple possible risk factors, but the data were discordant precluding aggregation within a meta-analysis.
CONCLUSION: This systematic review confirms paucity of data regarding the etiology and the precise causal pathway of stroke in preterm infants. Moreover, the preterm infants unlike the term infants do not typically present with seizures. Hence high index of clinical suspicion and routine cUS will assist in the timely diagnosis and understanding of stroke in this population.

Keywords: Arterial ischemic stroke, cerebral sinovenous thrombosis, periventricular hemorrhagic infarction
Subject terms: neonatal stroke, perinatal stroke, stroke in preterm infants

1. Introduction

Stroke in preterm infants occurs from the 20th to 37th gestational week. Stroke may happen either before birth (known as fetal or in-utero stroke) or

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after birth, up to 37 weeks postmenstrual age [1]. Long-term morbidities include motor impairment, cognitive and speech disorders, developmental delay, cerebral palsy and/or epilepsy [2, 3]. The estimated prevalence of stroke in preterm infants varies widely in published literature; ranging from 1:1600 to 1:8000 [4, 5]. Stroke has been more commonly studied in term infants, therefore the global burden of perinatal stroke in preterm infants is undetermined. These gaps in knowledge are due to paucity of published data and small single centre studies [6, 7].

In literature, the common types of stroke described in preterm infants are periventricular hemorrhagic infarction (PVHI), perinatal arterial ischemic stroke (PAIS) and cerebral sinovenous thrombosis (CSVT). PVHI is a periventricular venous congestion due to the obstruction of the terminal vein and impaired blood flow in the medullary vein following germinal matrix hemorrhage. PAIS is disruption of bl- ood flow to the cerebral artery due to thrombosis, embolism, vasospasm or hypoxic-ischemic encephalopathy. CSVT is a clot in the venous sinuses of the brain causing blockade of the blood draining from the brain. This causes blood cells to break and leak into the brain tissue causing hemorrhage.

The diagnosis of stroke is established from neuroradiological investigations [8]. Neuroimaging is routinely ordered in full-term infants presenting with seizures or unexplained apneas. While, in preterm infants, symptoms like apneas can also be linked to problems of prematurity rather than to stroke alone. In addition, the pathophysiology of stroke in preterm infants is different to the other brain injuries that are common to preterm infants like intraventricular hemorrhage (IVH) and periventricular leukomalacia (PVL) [9].

Literature suggests the likely causal pathway of stroke in preterm infants involves a multiplicity of risk factors rather than any single risk factor. Some of the known independent risk factors for PAIS include: Fetal heart rate abnormality (p = 0.008), neonatal hypoglycemia (p = 0.02), twin to twin transfusion syndrome (p = 0.005), dehydration, infection, patent ductus arteriosus and prothrombotic disorders [10, 11]. The presence of more than three potential risk factors is known to increase the risk of stroke more than any of these single factors in isolation: Maternal age, race, infertility, miscarriage, intrauterine growth restriction and gestational diabetes [11]. In the scant literature available, placental abnormalities such as placental weight < 10th percentile (p = 0.00), placental infarction (p = 0.00), malperfusion (p = 0.01), and chorioamnionitis (p = 0.04) are commonly associated with PVHI [12]. Plus, infection, prothrombotic disorders and dehydration are risk factors associated with CSVT [13, 14].

Therefore, we conducted a systematic review to explore the epidemiology and pathogenesis of PVHI; PAIS; and CSVT in preterm infants. To our knowledge, this is the first comprehensive systematic review of these three types of stroke in preterm infants.

2. Materials and methods

2.1. Literature search and selection

A systematic search was conducted from Jan- uary 2000 to December 2019 using MEDLINE, EMBASE, SCOPUS and WEB OF SCIENCE (Science and Social Science Citation Index) databases based on the Cochrane recommendations for conducting reviews. Search terms included (perinatal stroke*) or (cerebral stroke*) or (neonatal stroke*) and (preterm* or < 37 weeks*), as well as MeSH (Medical Subject Headings) (risk factor or etiology or outcome) with limits of English language and human studies applied. In addition, the reference list of the included studies for further relevant articles was hand-searched. The findings were reported according to the Meta-analyses Of Observational Studies in Epidemiology (MOOSE) checklist [15].

Studies were included in the systematic review if PVHI, PAIS and CSVT in preterm infants met the following criteria: (1) All original group study designs or case series; (2) studies published from January 2000 to December 2019 and (3) studies published in English. The exclusion criteria were: (1) studies where analysis of stroke in preterm and term infants could not be differentiated; (2) single case reports; (3) review papers and (4) abstracts and conference abstracts. If data were duplicated in > 1 study, then we included the study with the largest number of stroke cases.

2.2. Data extraction

An a priori designed data extraction tool based on the Cochrane recommendations was used. The following data were extracted from each study: First author's last name, publication year, country where the study was performed, study design, study period, type of stroke, gestational age, birth weight, risk

factors of preterm stroke, clinical presentation and outcome of each of the stroke types, as well as unadjusted and adjusted odds ratios, if reported. The quality of included studies were assessed using the Oxford centre for evidence-based medicine 2011 levels of evidence [16]. Study selection and data extraction were conducted independently by two investigators (BR and IN or MFE), with disagreements resolved by consensus.

2.3. Statistical analysis

All the findings were summarised and descriptive statistics used to describe the etiological and clinical variables extracted from papers. If the included studies reported on an individual case basis, proportions of the clinical variables were evaluated across the three stroke categories. A meta-analysis could not be conducted due to the heterogeneity of the reported data in the selected studies; explicitly, mismatched aims of the studies, discordant outcomes of stroke, and the small number stroke cases in each of the subgroups for analysis. The statistician author (CG) deemed the conduct of any inferential analyses to be inappropriate. Associations were investigated for case by case where data were available. All analyses were conducted using STATA V.14 (STATA, Version 14, Stata Corp, College Station, TX, USA).

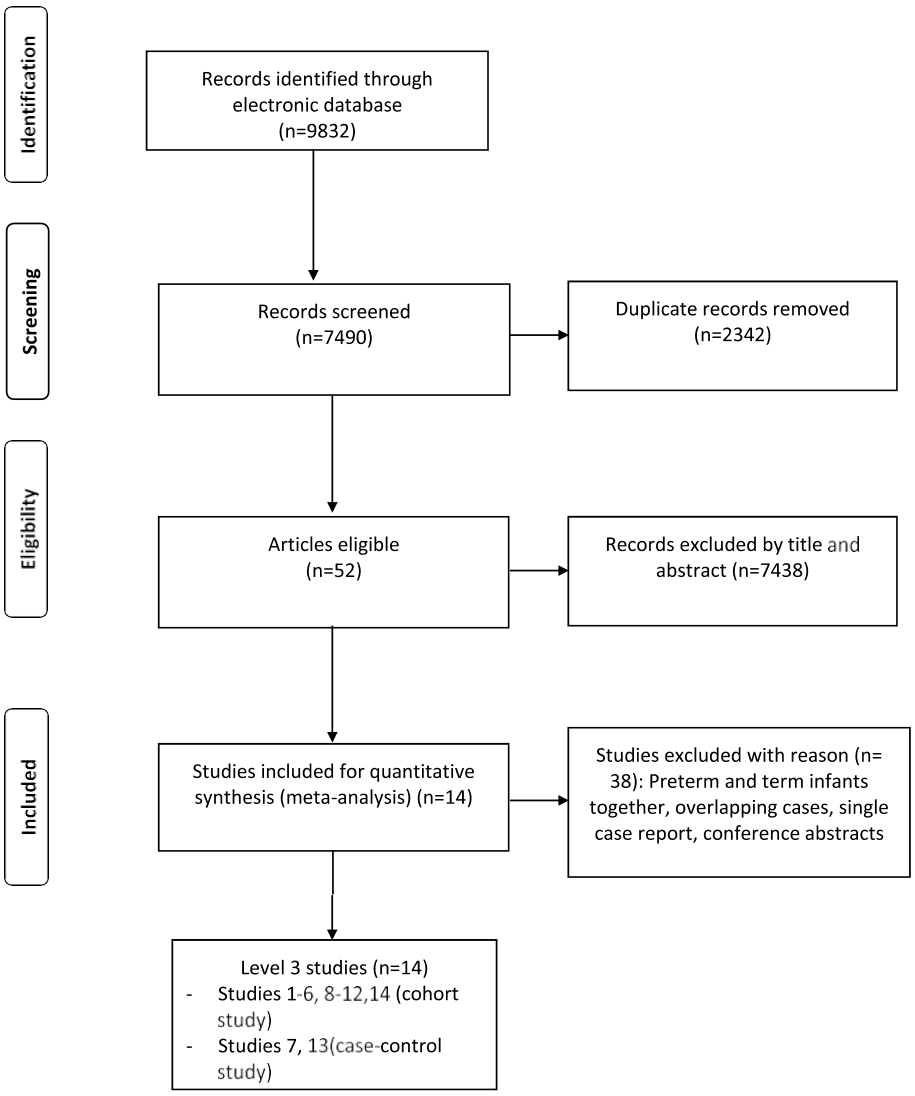

Fig. 1. Flow chart of study selection using the PRISMA diagram [40].

3. Results

3.1. Study characteristics

The search strategy is summarised in the PRISMA flow diagram (Fig. 1). 7490 studies were identified and after removing the duplicates and examining the titles and abstracts, 52 studies were eligible for full-text review. Of these, 14 studies met the predetermined criteria and were included (Table 1). All the included studies were from high income countries. The studies were analysed based on the type of stroke (Table 1): PVHI [17–23], PAIS [24–28] and CSVT [29, 30] in the chronology of the publication year (starting with the most recent study).

The eligible studies published between 2000 and 2019 included a total of 546 stroke cases from the period 1989 to 2015. The gestational ages and birth weights of the preterm infants were from 23–36 weeks and 450–3500 grams respectively.

3.2. Types of preterm strokes

3.2.1. Periventricular hemorrhagic infarction

Eighty percent ($n = 436/546$) of the stroke cases in preterm infants resulted in PVHI. These infants were between 23–35 weeks of gestation and birth weights of 450–2965 grams. The studies included only unilateral PVHI cases. All 436 cases were diagnosed using cranial ultrasound (cUS). In addition, 21% (90/436) of the cases had brain magnetic resonance imaging (MRI) and 26% (23/90) had Diffusion Tensor Imaging (DTI) to predict the likelihood of unilateral spastic cerebral palsy (USCP) [17, 18, 20]. Serial EEG was conducted in 5% (23/436) to differentiate PVHI from PVL. Brain injury timing was determined using a combination of cUS and EEG to be post-natal in PVHI, but in utero in PVL [19].

The potential risk factors put forward in the literature for PVHI were genetic mutations of methylenetetrahydrofolate reductase (MTHFR) gene 88%, factor V Leiden gene 41%, COL4A1 gene 12% and prothrombin gene 6% [20]. However, many of these propositions have been disproven, specifically MTHFR [31] and more generally the minimal association between neonatal stroke and thrombophilia [32]. Some of the common clinical associations were emergency caesarean section in 30%, patent ductus arteriosus 19%, thrombocytopenia 18%, pneumothorax 12%, seizures 11%, intrauterine growth restriction and/or pre-eclampsia 8% [17–19, 21–23].

Studies [19, 22] focused on different factors to predict the outcomes of PVHI (e.g., anatomical location of brain injury versus timing of the injury versus types of cerebral veins involved), which precluded aggregation of findings across studies. Soltirovska Salamon et al. [18] ($n = 213$) classified PVHI into anatomical subtypes based on the location of the lesion: Parietal PVHI (84%), frontal PVHI (10%) and temporal PVHI (6%). This study was the only study to report the overall mortality of 33% based on anatomical subtypes, which included 99% for parietal PVHI and 1% for temporal PVHI. Thirty six percent of the infants with PVHI developed cerebral palsy (CP) and 75% of these had USCP. Ninety eight percent of the infants with CP had a parietal PVHI and only 2% had a temporal PVHI. Infants in Soltirovska Salamon et al. study [18] with temporal or frontal PVHI were deemed to be at higher risk for long-term cognitive, behavioural and visual problems. Harteman et al. [20] ($n = 62$) was the only study to classify PVHI based on the timing of the lesion, which was centred on their unit's protocol for performing cUS. Typical PVHI (73%) was described when the onset of PVHI was within 6– 96 hours of age whereas atypical PVHI (27%) was when the onset of PVHI was < 6 hours or > 96 hours of age. Thrombophilia due to factor V Leiden mutation was more commonly associated with atypical PVHI. This study predicted higher prevalence of CP with atypical PVHI; 85% versus 12% with atypical and typical PVHI respectively.

Dudink et al. [22] ($n = 20$) classified PVHI into venous subtypes based on the types of cerebral vein affected. This study predicted 45% prevalence of CP in the cases with PVHI involving the terminal veins and normal outcome with involvement of the veins of the temporal and caudate area.

3.2.2. Perinatal arterial ischemic stroke

Seventeen percent ($n = 93/546$) of the preterm infants included in this review were diagnosed with PAIS. These infants were between 23–36 weeks of gestation with birth weights of 535–3500 grams. More than 50% were middle cerebral artery (MCA) stroke; including left MCA and lenticulostriate branch [25, 27, 28]. The other types of PAIS were posterior cerebral artery (PCA) stroke, cerebellar infarcts involving posterior inferior cerebellar arteries and perforator stroke [26, 28]. All 93 cases were diagnosed by MRI.

Multivariate analysis by Benders et al. [27] ($n = 31$) indicated hypoglycemia, fetal hart rate

Table 1
Characteristics of studies included in the systematic review

Study no:	Author (year)	Stroke type	Study origin	Study design	Study period	*n*	GA (weeks)	BW (grams)	OCEBM 2011 Levels of evidence [16]
Study 1	Roze et al. (2015) [17]	PVHI	Netherlands	Retrospective Cohort study	2007–2012	23	25–34	650–1950	Level 3
Study 2	Soltirovska Salamon et al. (2014) [18]	PVHI	Netherlands	Retrospective Cohort study	1990–2012	213	28–34	910–2965	Level 3
Study 3	Tsuji et al. (2014) [19]	PVHI	Japan	Retrospective Cohort study	1997–2005	22	23–31	538–1240	Level 3
Study 4	Harteman et al. (2012) [20]	PVHI	Netherlands	Retrospective Cohort study	2005–2010	62	25–34	580–2210	Level 3
Study 5	Roze et al. (2009) [21]	PVHI	Netherlands	Prospective Cohort study	1995–2003	38	25–35	700–2430	Level 3
Study 6	Dudink et al. (2008) [22]	PVHI	Netherlands	Retrospective Cohort study	2000 – 2005	20	24–34	775–2800	Level 3
Study 7	Bassan et al. (2006) [23]	PVHI	USA	Retrospective Case-control study	1997 – 2002	58	23–33	450–2375	Level 3
Study 8	Portale et al. (2019) [24]	PAIS	Italy	Retrospective Cohort study	January 1996–February 2015	2	35–36	3360–3500	Level 3
Study 9	Van der Aa et al. (2016) [25]	PAIS	Netherlands	Retrospective Cohort study	1990–2015	12	28–35	NA	Level 3
Study 10	Ecury-Goossen et al. (2013) [26]	PAIS	Belgium	Retrospective Cohort study	1999–2011	25	24–36	NA	Level 3
Study 11	Benders et al. (2009) [27]	PAIS	Netherlands	Prospective Cohort study	1990–2005	31	27–36	NA	Level 3
Study 12	Golomb et al. (2008) [28]	PAIS	USA	Retrospective Cohort study	1989–2006	23	23–35	535–2786	Level 3
Study 13	Raets et al. (2013) [29]	CSVT	Netherlands	Prospective Cross-control study	2009–2013	11	24–28	615–1185	Level 3
Study 14	Kersbergen et al. (2011) [30]	CSVT	Netherlands	Retrospective Cohort study	2002–2010 2006–2009	6	30–35	NA	Level 3
Total	2000–2009 = 5 studies 2010–2020 = 9 studies	PVHI = 7 studies PAIS = 5 studies CSVT = 2 studies		Prospective = 3 studies Retrospective = 11 studies	1989 – 2015	2000–2009 = 170 2010–2020 = 376 Total = 546	23–36 weeks	450–3500 grams	Level 3 = 14 studies

BW – birth weight, CSVT – cerebral sinovenous thrombosis, GA – gestational age, n – population number, NA – not available, PAIS – perinatal arterial ischemic stroke, PVHI – periventricular hemorrhagic stroke. Oxford Centre for Evidence-based Medicine (OCEBM) – Level 3 (studies 1–14): 'Local non-random sample'.

abnormality and twin-to-twin transfusion syndrome as independent risk factors. Common clinical presentation was apnoea (83%) whilst seizures was an uncommon presentation amongst the preterm infants (10–30%) [27, 28]. The outcome of PAIS was dependent on gestational age; cortical sparing was noted in preterm infants compared to term infants (58% versus 4%) [25]. Golomb et al. [28] reported disability/neurological abnormality in 100% (22/22) and CP in 77% (17/22) of the surviving infants; 52% (12/23) had MCA stroke. Van der Aa et al. [25], Ecury-Goossen et al. [26] and Benders et al. [27] reported cumulative outcomes for preterm and term infants; impeding separate outcome analysis in the preterm infants.

3.2.3. Cerebral sinovenous thrombosis

Three percent (17/546) of the stroke cases in preterm infants were CSVT [29, 30]. Theses infants were between 24–35 weeks of gestation. The study by Raets et al. [29] was a prospective case control study ($n = 11$ with CSVT versus $n = 238$ without CSVT) of preterm infants < 29 weeks of gestation. All 11 cases had partial or complete thrombosis in the transverse sinus, including one case with multiple sinus thrombosis. All the cases were diagnosed by cUS, although 64% (7/11) were confirmed by MRI. None of the potential risk factors such as sepsis, patent ductus arteriosus and Apgar scores were significantly different between the CSVT and non-CSVT groups. The outcomes at 6 months to 2 years included: One neonatal death; 50% (5/10) were typically developing and 50% had abnormal Bayley scales of infant development or mental or psychomotor developmental assessment scores suggesting long-term disability.

The study by Kersbergen et al. [30] was a retrospective cohort study ($n = 6$ preterm infants and $n = 24$ full term infants) of preterm infants ≥ 30 weeks of gestation. In this study, all six cases had straight sinus thrombosis and 83% (5/6) had multiple sinus thrombosis and only 33% (2/6) had transverse sinus thrombosis. The preterm infants had severe white matter lesions ($p < 0.001$) in contrast to the term infants who had predominant punctate white matter lesions ($p < 0.001$). All six preterm infants were diagnosed using MRI or magnetic resonance venography (MRV). Outcomes in these cases at 3 years were: 83% (5/6) mortality and in the one surviving child CP with epilepsy.

4. Discussion

Stroke in infants is an important cause of CP and other neurological disabilities [33]. Strategies to improve the overall outcome of preterm infants should include timely diagnosis of stroke, which may be challenging due to the coexisting morbidities of prematurity that confound identification of stroke [34]. This systematic review confirms the paucity of epidemiologic data and outcome data for the three most common types of stroke in preterm infants; namely, PVHI, PAIS and CSVT. In this review, approximately 70% of the stroke cases were reported in the second epoch of publications (2010 – 2019) compared to 30% in the previous decade. This may be due to increased awareness of stroke in this population and the emergence of more sophisticated neuroimaging techniques such as brain MRI with DTI, magnetic resonance angiography (MRA) and MRV coupled with the routine use of cUS [35].

The three types of stroke in preterm infants included in this review were PVHI, PAIS and CSVT. Different studies used different techniques to classify PVHI to accurately predict the long-term outcome based on the anatomical location of the PVHI, type of cerebral venous involvement and on the timing of the occurrence of PVHI [18, 20, 22]. Parietal PVHI was more common than temporal and frontal lobe PVHI [18].

Data regarding PAIS were scarce in preterm infants [27, 28, 36]. Left MCA stroke was most common compared to right MCA, PCA, and cerebellar strokes [27, 28]. For infants between 28–32 weeks' gestation, lenticulostriate branch of the MCA was most often involved. Whereas, for infants > 32 weeks' gestation, main branches of the MCA were more often involved [27]. Perforator stroke was under-recognised because of the lack of clinical symptoms [26]. CSVT was the least reported type of preterm stroke in the literature [13, 37].

A clear understanding of the mechanism and identification of specific risk factors may aid in devising future novel preventative strategies for preterm stroke. PVHI and CSVT are due to obstruction in the venous drainage following germinal matrix hemorrhage and blockage of a dural sinus by thrombus respectively. PAIS is secondary to lack of blood supply following a thrombus in the cerebral artery. The potential risk factors were hemodynamic instability, cerebral blood flow abnormalities and genetic triggers or septicemia causing thrombophilia [20, 23, 24, 27]. The factors associated with flow abnormalities

in the fetal brain were emergency caesarean section [18–20], fetal distress/low Apgar scores/resuscitation [23] and pneumothorax [21].

cUS with doppler technology was widely used to diagnose PVHI MRI with diffusion-weighted imaging (DWI) sequences and MRV is the gold standard [38, 39]. Diffusion tensor imaging (DTI) identified the microstructural properties of the white matter in the posterior limb of the internal capsule, elucidating the risk for motor impairment [17]. Asymmetry visible on the DTI was a reliable predictor of the development of USCP [17].

Early diagnosis of stroke assists in the recovery process through neuroplasticity though the scope of healing also depends on the gestational age vis-a´-vis the embryological development of the preterm brain. Parietal PVHI had a poorer prognosis with 40% mortality rate and 50% risk of CP, while, temporal PVHI and frontal PVHI had increased risk of cognitive, behavioural and visual problems [18, 22].

Amalgamated reporting of stroke in preterm and term infants was the major limitation of this review, preventing disaggregation of data across studies into a meta-analysis. There was also a potential risk of duplication of cases because of multiple reporting of the same stroke cases across some studies. The results should be interpreted cautiously because the individual case data were often reported retrospectively and as a secondary end-point. The review also included a heterogenous studies eg not same aims of the studies (aimed for etiology, clinical presentation, investigations and or outcome of stroke) and not same outcomes of stroke (outcomes at 6 months to 3 years and anytime in between).

Stroke in preterm infants is unlikely to present with focal seizures and may be asymptomatic or have nonspecific clinical presentation. Therefore, appropriate neuroimaging cUS in presence of three or more risk factors of stroke should become part of routine screening in all preterm babies. In addition, analysis of stroke in preterm and term infants separately is likely to yield a clearer understanding of the pathophysiology unique to preterm infants.

5. Conclusion

In conclusion, this systematic review confirms, that, the etiology of stroke in preterm infants is not known and the precise causal pathway is not always very well understood and more research is required. The outcome of stroke was likely to be complicated by the co-morbidities of prematurity, though, it was outside the scope of this review. Early identification and the reported prevalence rate of stroke may increase with greater recognition of the clinical presentation of stroke, routine cUS and brain MRI in high-risk preterm groups.

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Disclosures

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Contributors' statement

Dr Bithi Roy conceptualized and designed the study, designed data collection instruments (first extractor), carried out the initial analyses, drafted, reviewed and revised the manuscript. Prof Iona Novak assisted with data extraction (additional extractor), analyses, critically reviewed the manuscript for important intellectual content and revised the manuscript. Dr Megan Finch-Edmondson assisted with data extraction (additional extractor), reviewed and revised the manuscript. Claire Galea assisted with statistical analysis, reviewed and revised the manuscript. Dr Catherine Morgan, A/Prof Karen Walker and Prof Nadia Badawi critically reviewed and revised the manuscript for important intellectual content. All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

References

[1] Engle WA, American academy of pediatrics committee on fetus and newborn. Age terminology during the perinatal period. Pediatrics. 2004;114(5):1362-4.
[2] Kirton A, Deveber G. Life after perinatal stroke. Stroke. 2013;44(11):3265-71.
[3] Dunbar M, Kirton A. Perinatal stroke: Mechanisms, management, and outcomes of early cerebrovascular brain injury. Lancet Child Adolesc Health. 2018;2(9):666-76.
[4] Collaborators GBDS. Global, regional, and national burden of stroke, 1990-2016: A systematic analysis for the Global Burden of Disease Study 2016. Lancet Neurol. 2019;18(5):439-58.

[5] Dunbar M, Mineyko A, Hill M, Hodge J, Floer A, Kirton A. Population Based Birth Prevalence of Disease-Specific Perinatal Stroke. Pediatrics. 2020;146(5):11.
[6] Laugesaar R, Kolk A, Tomberg T, Metsvaht T, Lintrop M, Varendi H, et al. Acutely and retrospectively diag- nosed perinatal stroke: A population-based study. Stroke. 2007;38(8):2234-40.
[7] Chabrier S, Husson B, Dinomais M, Landrieu P, Nguyen The Tich S. New insights (and new interrogations) in perinatal arterial ischemic stroke. Thromb Res. 2011;127(1):13-22.
[8] Medley TL, Miteff C, Andrews I, Ware T, Cheung M, Monagle P, et al. Australian clinical consensus guideline: The diagnosis and acute management of childhood stroke. Int J Stroke. 2019;14(1):94-106.
[9] Volpe JJ. Brain injury in the premature infant–from pathogenesis to prevention. Brain Dev. 1997;19(8):519-34.
[10] Benders MJ, Groenendaal F, Uiterwaal CS, Nikkels PG, Bruinse HW, Nievelstein RA, et al. Maternal and infant characteristics associated with perinatal arterial stroke in the preterm infant. Stroke. 2007;38(6):1759-65.
[11] Lee J, Croen LA, Backstrand KH, Yoshida CK, Henning LH, Lindan C, et al. Maternal and infant characteristics associated with perinatal arterial stroke in the infant. JAMA. 2005;293(6):723-9.
[12] Harteman JC, Nikkels PG, Kwee A, Groenendaal F, de Vries LS. Patterns of placental pathology in preterm infants with a periventricular haemorrhagic infarction: Association with time of onset and clinical presentation. Placenta. 2012;33(10):839-44.
[13] deVeber G, Andrew M, Adams C, Bjornson B, Booth F, Buckley DJ, et al. Cerebral sinovenous thrombosis in children. N Engl J Med. 2001;345(6):417-23.
[14] Javed I, Sultan T, Rehman ZU, Yaseen MR. Clinical spectrum and outcome of cerebral venous sinus thrombosis in children. J Coll Physicians Surg Pak. 2018;28(5):390-3.
[15] Stroup DF, Berlin JA, Morton SC, Olkin I, Williamson GD, Rennie D, et al. Meta-analysis of observational studies in epidemiology: A proposal for reporting. Meta-analysis Of Observational Studies in Epidemiology (MOOSE) group. JAMA. 2000;283(15):2008-12.
[16] OCEBM levels of Evidence Working group. The Oxford 2011 levels of evidence. 2011.
[17] Roze E, Benders MJ, Kersbergen KJ, van der Aa NE, Groenendaal F, van Haastert IC, et al. Neonatal DTI early after birth predicts motor outcome in preterm infants with periventricular hemorrhagic infarction. Pediatr Res. 2015;78(3):298-303.
[18] Soltirovska Salamon A, Groenendaal F, van Haastert IC, Rademaker KJ, Benders MJNL, Koopman C, et al. Neuroimaging and neurodevelopmental outcome of preterm infants with a periventricular haemorrhagic infarction located in the temporal or frontal lobe. Dev Med Child Neurol. 2014;56(6):547-55.
[19] Tsuji T, Okumura A, Kidokoro H, Hayakawa F, Kubota T, Maruyama K, et al. Differences between periventricular hemorrhagic infarction and periventricular leukomalacia. Brain Dev. 2014;36(7):555-62.
[20] Harteman JC, Groenendaal F, van Haastert IC, Liem KD, Stroink H, Bierings MB, et al. Atypical timing and presentation of periventricular haemorrhagic infarction in preterm infants: the role of thrombophilia. Dev Med Child Neurol. 2012;54(2):140-7.
[21] Roze E, Van Braeckel KN, van der Veere CN, Maathuis CG, Martijn A, Bos AF. Functional outcome at school age of preterm infants with periventricular hemorrhagic infarction. Pediatrics. 2009;123(6):1493-500.
[22] Dudink J, Lequin M, Weisglas-Kuperus N, Conneman N, Van Goudoever JB, Govaert P. Venous subtypes of preterm periventricular haemorrhagic infarction. Arch Dis Child Fetal Neonatal Ed. 2008;93(3):F201-F6.
[23] Bassan H, Feldman HA, Limperopoulos C, Benson CB, Ringer SA, Veracruz E, et al. Periventricular hemorrhagic infarction: risk factors and neonatal outcome. Pediatr Neurol. 2006;35(2):85-92.
[24] Portale A, Fiumara A, Scalora L, Greco F, Smilari P, Venti V, et al. Arterial ischemic stroke (AIS) in childhood: Clinical report from a single control center. Childs Nerv Syst. 2019;35(2):283-93.
[25] van der Aa NE, Benders MJ, Nikkels PG, Groenendaal F, de Vries LS. cortical sparing in preterm ischemic arterial stroke. Stroke. 2016;47(3):869-71.
[26] Ecury-Goossen GM, Raets MM, Lequin M, Feijen-Roon M, Govaert P, Dudink J. Risk factors, clinical presentation, and neuroimaging findings of neonatal perforator stroke. Stroke. 2013;44(8):2115-20.
[27] Benders MJ, Groenendaal F, De Vries LS. Preterm arterial ischemic stroke. Semin Fetal Neonatal Med. 2009;14(5):272-7.
[28] Golomb MR, Garg BP, Edwards-Brown M, Williams LS. Very early arterial ischemic stroke in premature infants. Pediatr Neurol. 2008;38(5):329-34.
[29] Raets MM, Sol JJ, Govaert P, Lequin MH, Reiss IK, Kroon AA, et al. Serial cranial US for detection of cere- bral sinovenous thrombosis in preterm infants. Radiology. 2013;269(3):879-86.
[30] Kersbergen KJ, Groenendaal F, Benders MJ, van Straaten HL, Niwa T, Nievelstein RA, et al. The spectrum of associated brain lesions in cerebral sinovenous thrombosis: relation to gestational age and outcome. Arch Dis Child Fetal Neonatal Ed. 2011;96(6):F404-9.
[31] Hickey SE, Curry CJ, Toriello HV. ACMG practice guideline: Lack of evidence for MTHFR polymorphism testing. Genet Med. 2013;15(2):153-6.
[32] Curtis C, Mineyko A, Massicotte P, Leaker M, Jiang XY, Floer A, et al. Thrombophilia risk is not increased in children after perinatal stroke. Blood. 2017;129(20):2793-800.
[33] Greenham M, Gordon A, Anderson V, Mackay MT. Outcome in childhood stroke. Stroke. 2016;47(4):1159-64.
[34] Badawi N, McIntyre S, Hunt RW. Perinatal care with a view to preventing cerebral palsy. Dev Med Child Neurol. 2021;63(2):156-61.
[35] Kirton A, de Veber G. Stroke in the fetus and neonate. Future Cardiol. 2006;2(5):593-604.
[36] Benders MJ, Groenendaal F, Uiterwaal CS, de Vries LS. Perinatal arterial stroke in the preterm infant. Semin Perinatol. 2008;32(5):344-9.
[37] Berfelo FJ, Kersbergen KJ, van Ommen CH, Govaert P, van Straaten HL, Poll-The BT, et al. Neonatal cerebral sinovenous thrombosis from symptom to outcome. Stroke. 2010;41(7):1382-8.
[38] Dudink J, Mercuri E, Al-Nakib L, Govaert P, Counsell SJ, Rutherford MA, et al. Evolution of unilateral perinatal arterial ischemic stroke on conventional and diffusion-weighted MR imaging. AJNR Am J Neuroradiol. 2009;30(5):998-1004.
[39] Kuker W, Mohrle S, Mader I, Schoning M, Nagele T. MRI for the management of neonatal cerebral infarctions: importance of timing. Childs Nerv Syst. 2004;20(10):742-8.
[40] PRISMA. PRISMA 2009 checklist 2009.

Chapter 5: AETIOLOGY OF PERINATAL STROKE: A POPULATION-BASED STUDY IN AUSTRALIA

5.1 Introduction

Perinatal stroke (occurring from 20 weeks of gestation to 28 days postnatal age) is a major cause of neurodevelopmental disability in infants, although the incidence in Australia is unknown. The aetiology of perinatal stroke is poorly understood and several of the known risk factors are also present in healthy infants.

A 2-year population-based prospective study of perinatal stroke was conducted in Australia between 2017 and 2019 and included 60 term infants. The estimated birth prevalence rate of perinatal stroke was 9.6 cases per 100,000 live births per year.

Comparison of the perinatal stroke cases with the Australian population data from the Australian Institute of Health and Welfare (AIHW), showed caesarean section ($p=0.04$), 5-minutes Apgar score <7 ($p<0.01$) and neonatal resuscitation at birth ($p<0.01$) were significant risk factors for perinatal stroke.

5.2 Manuscript Under Review

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Etiology of Perinatal Stroke: A Population Based Study in Australia

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Short Title: Perinatal Stroke in Australia

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Abbreviations:

AIHW	Australian Institute of Health and Welfare
APPIS	Arterial Presumed Perinatal Ischemic Stroke
APSU	Australian Paediatric Surveillance Unit
AT III	Antithrombin III
CHD	Congenital Heart Disease
CI	Confidence Interval
CS	Cesarean Section
CSVT	Cerebral Sinovenous Thrombosis
GDM	Gestational Diabetes Mellitus

HSV	Herpes Simplex Virus
LBW	Low Birth Weight
MRI	Magnetic Resonance Imaging
n	Number
NAIS	Neonatal Arterial Ischemic Stroke
NHS	Neonatal Hemorrhagic Stroke
NVD	Normal Vaginal Delivery
PIH	Pregnancy Induced Hypertension
PPHS	Presumed Perinatal Hemorrhagic Stroke
PVI	Periventricular Venous Infarction
SGA	Small for Gestational Age

Article Summary

Unlike adults, perinatal stroke may not have specific risk factors. Identifying at-risk infants is difficult because some risk factors are present in healthy infants.

CONTRIBUTORS STATEMENT

Dr Bithi Roy, Prof Iona Novak and Prof Nadia Badawi conceptualized and designed the study and designed the data collection instruments.

Dr Bithi Roy collected data, carried out the initial analyses, drafted the initial manuscript, and revised the manuscript.

Prof Iona Novak critically analyzed and reviewed and revised the manuscript for important intellectual content.

Prof Nadia Badawi assisted in data collection, and critically reviewed and revised the manuscript for important intellectual content.

Clinical Prof Karen Walker and Dr Catherine Morgan assisted in data collection, and critically reviewed and revised the manuscript for important intellectual content.

Annabel Webb carried out the initial analyses, and critically analyzed and revised the manuscript.

Dr Carlos Nunez collected and curated the data and critically reviewed and revised the manuscript.

Prof Guy Eslick critically analyzed, critically reviewed, and revised the manuscript.

Adjunct Prof Alison L Kent, Prof Rod W Hunt and Prof Mark T Mackay critically reviewed and revised the manuscript for important intellectual content.

All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

Abstract

Objectives: The study objective was to calculate birth prevalence of perinatal stroke and examine the risk factors in term infants (>37 weeks of gestation). This gap exists in literature because some risk factors for perinatal stroke are present in healthy infants, making it difficult to determine at-risk infants.

Study Design: A prospective population-based perinatal stroke study was conducted, and data were compared to the Australian general population data obtained from the Australian Institute of Health and Welfare's (AIHW) Australia's Mothers and Babies report. Maternal and neonatal risk factor data were analyzed using chi-squared and Fisher's exact tests and multivariable logistic regression analysis.

Results: Between 2017 and 2019, 60 perinatal stroke cases were reported between 37 and 42 weeks of gestation with birth weights of 2114-4470 g, including 58% male infants. Estimated stroke prevalence was 9.6/100,000 live births/year including 5.8 for neonatal arterial ischemic stroke and 2.9 for neonatal hemorrhagic stroke. Eighty seven percent had multiple risk factors. Significant risk factors were cesarean section ($p=0.04$), 5-minute Apgar score <7 ($p<0.01$) and neonatal resuscitation ($p<0.01$) when compared with general population data.

Conclusions: This is the first study to compare perinatal stroke risk factors with Australian population data. Cesarean section and fetal distress were associated with perinatal stroke. These data now require validation in a case-control study.

Perinatal stroke includes acute symptomatic perinatal stroke presenting in the perinatal period from 20 weeks of gestation to 28 days postnatal age and presumed perinatal stroke presenting after 28 days of age.(1-3) The focus of this paper is on acute symptomatic perinatal stroke. The subtypes of acute symptomatic perinatal stroke are neonatal arterial ischemic stroke (NAIS), neonatal cerebral sinovenous thrombosis (CSVT) and neonatal hemorrhagic stroke (NHS).(4)

The incidence of perinatal stroke has been variably reported in the literature due to heterogeneity in data sources, reporting biases in non-population clinical samples and inconsistencies in the terminology of stroke subtypes.(5) Diagnostic criteria discrepancies also influence reported incidence rates. The incidence of perinatal stroke is 25-40/100,000 live births, compared to the incidence of childhood stroke 2-3/100,000 children in the United States.(6, 7) The global incidence of perinatal arterial ischemic stroke ranges from 1/2800 to 1/5000 live births.(1, 8, 9) Prior studies in Australia have quoted the incidence of arterial ischemic stroke as 1.8/100,000 children per year and CSVT as 0.34/100,000 children per year, though the true incidence of perinatal stroke remains unknown.(10, 11)

Risk factors for perinatal stroke, underlying etiologies and pathophysiology depends on stroke subtype.(12) Most epidemiological studies are based on retrospective data or small heterogenous samples aggregated into group means, which limits identification of precise risk factors.(7, 13, 14) Moreover, some of these risk factors are present in the general population, leading to uncertainties in etiology and prevention strategies.

The aim of this study was to determine the prevalence and identify risk factors for perinatal stroke in full term infants compared to the general Australian population.

METHODS

Study Design

This was a 2-year study conducted between 1 July 2017 and 30 June 2019. Acute symptomatic perinatal stroke cases were reported prospectively by specialist physicians including neonatologists, pediatricians, or neurologists to the Australian Paediatric Surveillance Unit (APSU). The APSU is a national resource that facilitates active surveillance of rare childhood diseases.(15) It is closely affiliated with the University of Sydney's Faculty of Medicine and Health Sciences and the Sydney Children's Hospital Network. A data questionnaire designed by the investigators (BR, NB and IN) was submitted by reporting physicians with deidentified data to the APSU. Data included postal zip code, risk factors for stroke, clinical presentations, investigations, management, and outcomes of stroke. Diagnosis of stroke was based on the reporting neonatologist, pediatrician or neurologist and independently confirmed by brain MRI reports by three authors (BR, CM and NB). MRI images were not sought to lower respondent burden and reduce the likelihood of missing data. Results were reported according to the revised STROBE statement.(16)
For consistency of reporting and analysis, risk factors for stroke were categorized into maternal, pregnancy, intrapartum and neonatal groups, as follows based on previous studies.(17-20)

a) Maternal risk factors included: ethnicity, age, consanguinity, genetic or hematological abnormalities, miscarriage, still birth and neonatal death history, and history of stroke in other children.
b) Pregnancy risk factors included: gravida, parity, multiple births, type of conception, gestational diabetes, hypertension, and history of illicit drugs, smoking and alcohol.

c) Intrapartum risk factors included: infections, group B streptococcus (GBS) colonization, prolonged rupture of membrane, meconium-stained liquor, placental abnormalities, mode of delivery and history of difficult delivery.

d) Neonatal risk factors included: sex, gestational age, birth weight, head circumference, Apgar scores, resuscitation, cord blood gas, history of receiving vitamin K, sepsis, meningitis, congenital heart disease (CHD), hypoglycemia, central vascular catheterization and hematological or coagulation abnormalities.

Participants

Inclusion criteria were:

1) Term infants (37-42 weeks of gestation) with perinatal stroke in Australia
2) Perinatal stroke presenting from birth to 28 days of age
3) Infants born between 2017 and 2019.

Exclusion criteria were:

1) Perinatal stroke presenting after 28 days of age
2) Strokes secondary to head injury
3) Preterm infants (<37 weeks of gestation)
4) CSVT alone.

Stroke Types

Inconsistent classification of stroke in young children in the literature is one of the major hurdles in aggregating and interpreting data. Some of the classifications are based on: a) age when stroke occurs (e.g. fetal stroke, perinatal stroke, neonatal stroke, and pediatric or childhood stroke); b) type of stroke (e.g. ischemic or hemorrhagic), and c) nature of blockage of blood vessel causing stroke (e.g. arterial ischemic stroke, CSVT, hemorrhagic stroke or periventricular venous infarction).

Dunbar and Kirton's classification of acute symptomatic perinatal stroke, including arterial or venous and ischemic or hemorrhagic subtypes, was used in this study.(4)

Arterial stroke included:

1. Neonatal arterial ischemic stroke (NAIS) – defined as focal ischemic infarction in one or more arterial territories, occurring more commonly in term infants with acute clinical presentation of stroke in the first 28 days of age.

Venous stroke included:

1. Neonatal hemorrhagic stroke (NHS) – a focal bleed within the brain parenchyma in term infants in the first 28 days of age.
2. Cerebral sinovenous thrombosis (CSVT) – includes presence of thrombus in one or more cerebral veins or dural sinuses plus parenchymal venous infarction in cerebral venous territory, occurring in term infants presenting with seizures in the first days of life.

Statistical Analysis

Birth prevalence rates of acute symptomatic perinatal stroke in Australia were calculated by dividing the number of cases by the number of live births, annually, which was determined from the Australian Bureau of Statistics population dataset.(21) The term 'incidence' was used for whole population data and 'prevalence' was used for non-population data. Frequencies and percentages were calculated for maternal and neonatal risk factors across stroke subtype groups. When missing data occurred (reported in *Table 4 (online)*), the denominator for each variable was adjusted as appropriate. Differences in maternal and neonatal risk factors between stroke subtypes were tested using chi-squared tests, or Fisher's exact tests where group sizes were $n < 5$. Odds ratios and associated 95% confidence intervals for the associations between risk factors and stroke subtypes were also calculated

where possible. Multivariable logistic regression models were conducted for the stroke subtype outcomes to identify independent risk factors.

Total population level data was obtained from the Australian Institute of Health and Welfare's (AIHW) Australia's Mothers and Babies report, which is based on 2019 data from the National Perinatal Data Collection, the National Maternal Mortality Data Collection, and the National Perinatal Mortality Data Collection.(22) The prevalence of maternal and neonatal risk factors in the stroke population was compared with their overall population prevalence using chi-squared tests and z-tests of proportions. For all statistical analyses, significance was set at $p < 0.05$.

Statement on Ethics

This study was approved by the Sydney Children's Hospital Network Human Research Ethics Committee (Ethics approval no: 2019/ETH06281).

RESULTS

Patient Population

Sixty acute symptomatic perinatal strokes met all three inclusion criteria (Table 1).

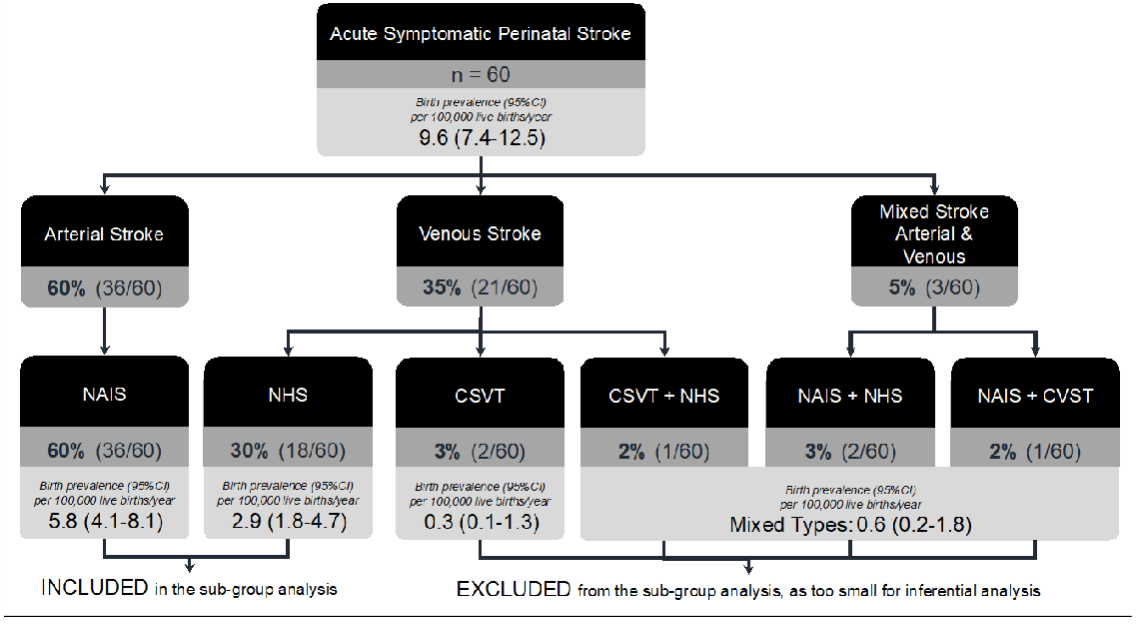
Table 1: Patient characteristics

	Acute symptomatic Perinatal Stroke (<28 days of age)
Total	60
Gestational age (weeks) range (mean)	37 - 42 (39)
Birth weight (grams) range (mean)	2114 - 4470 (3294)
Male:Female	35:25

n – number.

The acute symptomatic perinatal stroke subtypes and birth prevalence are shown in Figure 1.

Figure 1:Types and Birth Prevalence of acute symptomatic Perinatal Stroke (n=60)



LEGEND: CI – confidence interval, CSVT – cerebral sinovenous thrombosis, NAIS – neonatal arterial ischemic stroke, NHS – neonatal hemorrhagic stroke.

Risk Factors for Stroke

Risk factors were identified in 95% (57/60) of acute symptomatic perinatal strokes, and 87% (52/60) had multiple risk factors. In the univariate analysis (Table 2), mode of delivery and meconium-stained liquor were identified as having significant associations with stroke subtype. Specifically, birth via cesarean section was significantly more prevalent among NAIS compared to NHS, with 61.7% of NAIS infants born via cesarean section compared to 29.4% of NHS (OR for NAIS vs. NHS 3.77; 95% CI 1.00, 17.04). Meconium-stained liquor was more common in NAIS than in NHS; 34.3% versus 5.6% (OR for NAIS vs. NHS 8.59; 95% CI 1.07, 400.21). Group B streptococcal colonization and central vascular catheterization were different between the perinatal stroke subtypes, with 17.1% of NAIS having a GBS infection compared to none in the NHS, and 20.0% of NAIS having central vascular catheterization compared to 46.2% in the NHS, although these differences did not reach statistical significance. Both NAIS and NHS had more males than females, but the ratio of males to females was not statistically significant (OR 1.25; 95% CI 0.34, 4.56).

We used multivariable logistic regression models for stroke subtypes, with categorial variables included in the model when they were sufficiently populated ($n \geq 1$ in the smallest level) and univariate analysis yielded a p-value lower than 0.20. Only 3 variables (mode of delivery, meconium-stained liquor and central vascular catheterization) met these criteria. When included together in the model, all 3 variables showed significant association with stroke subtype. Meconium-stained liquor was associated with higher odds of NAIS (adjusted OR for NAIS vs NHS 52.41; 95% CI 2.04, 1348.50). In particular, central vascular catheterization (3 infants with umbilical vessel catheter, 2 with femoral catheter and 1 with peripherally inserted central catheter) was associated with a higher likelihood of NHS, though, none of the infants with central catheters had received anticoagulant treatment. Small sample size with the catheters precluded drawing firm conclusions (adjusted OR for NAIS vs

NHS 0.05; 95% CI 0.00, 0.58). Normal vaginal delivery was similarly associated with higher odds of NHS (adjusted OR for NAIS vs NHS 0.15; 95% CI 0.02, 0.93), while cesarean section was associated with higher odds of NAIS (adjusted OR for NAIS vs NHS 6.91; 95% CI 1.07, 44.54).

Table 2: Risk factors for acute symptomatic perinatal stroke subtypes (n>2)

Risk factors				
	NAIS (n=36)	NHS (n=18)	OR (NAIS vs NHS)	p value
Maternal risk factors				
Racial background:				
Pacific Islander	1 (2.9%)	0 (0.0%)	NA	1.00
Indigenous Australian	0 (0.0%)	1 (6.7%)	NA	0.30
Caucasian	26 (76.5%)	10 (66.7%)	1.61 (0.33, 7.32)	0.50
Asian	4 (11.8%)	3 (20.0%)	0.54 (0.08, 4.25)	0.70
Others	3 (8.8%)	1 (6.7%)	1.35 (0.10, 76.11)	1.00
Age:				
<20 years	0 (0.0%)	0 (0.0%)	NA	
20-39 years	30 (96.8%)	13 (92.9%)	2.26 (0.03, 186.77)	0.50
≥40 years	1 (3.2%)	1 (7.1%)		
Consanguinity	0 (0.0%)	1 (5.9%)	NA	0.38
Hematological/ Genetic abnormalities:				
Factor V Leiden	1 (25.0%)	0 (0.0%)	NA	1.00
Protein C & S	1 (25.0%)	0 (0.0%)	NA	1.00
Homocysteine	0 (0.0%)	0 (0.0%)	NA	1.00
Prothrombin	0 (0.0%)	0 (0.0%)	NA	1.00
AT III	1 (25.0%)	0 (0.0%)	NA	1.00
Fibrinogen	1 (25.0%)	0 (0.0%)	NA	1.00
Normal	3 (75.0%)	2 (66.7%)	NA	
History of miscarriage or still birth or neonatal deaths	3 (8.6%)	0 (0.0%)	NA	0.54
History of stroke in other children	0 (0.0%)	0 (0.0%)	NA	1.00
Pregnancy risk factors				
Gravida:				
Primigravida	18 (56.2%)	10 (58.9%)	0.90 (0.23, 3.44)	1.00
Multigravida (>2)	14 (43.8%)	7 (41.1%)		

Parity: One ≥ Two	 21 (65.5%) 11 (34.5%)	 12 (70.6%) 5 (29.4%)	 0.80 (0.17, 3.29)	 1.00
Multiple births: Twins/triplets	1 (2.8%)	0 (0.0%)	NA	1.00
Type of conception: Natural In-vitro fertilization	 32 (97.0%) 1 (30.0%)	 15 (100.0%) 0 (0.0%)	 NA	 1.00
Pregnancy complications: Gestational diabetes mellitus Pregnancy hypertension	 5 (14.3%) 2 (5.7%)	 1 (5.9%) 1 (5.9%)	 2.70 (0.27, 137.10) 1.00 (0.05, 62.40)	 0.65 1.00
Medications for hypertension, diabetes, thyroid disease	7 (20.0%)	1 (5.9%)	3.91 (0.44, 171.20)	0.25
History of smoking	2 (5.9%)	1 (5.9%)	1.00 (0.05, 62.60)	1.00
History of alcohol	1 (2.9%)	0 (0.0%)	NA	1.00
History of illicit drugs	1 (2.9%)	0 (0.0%)	NA	1.00
Intrapartum risk factors				
Infections: Group B streptococcus colonization HSV infection Parvovirus infection	 6 (17.1%) 0 (0.0%) 0 (0.0%)	 0 (0.0%) 1 (5.9%) 1 (5.9%)	 NA NA NA	 0.16 0.33 0.33
Prolonged rupture of membranes (>24 hours)	1 (2.9%)	0 (0.0%)	NA	1.00
Meconium-stained liquor	12 (34.3%)	1 (5.6%)	**8.59 (1.07, 400.21)**	**0.04**
Chorioamnionitis/Abnormal histopathology	2 (5.0%)	0 (0.0%)	NA	1.00
Mode of delivery: Normal vaginal delivery Instrumental (vacuum, forceps) Cesarean section (elective and emergency) Breech	 7 (20.6%) 6 (17.7%) 21 (61.7%) 0 (0.0%)	 8 (47.1%) 4 (23.5%) 5 (29.4%) 0 (0.0%)	 0.30 (0.07, 1.24) 0.70 (0.13, 3.98) **3.77 (1.00, 17.04)** NA	 0.10 0.71 **0.04** 1.00
History of difficult delivery	4 (12.5%)	3 (18.8%)	0.63 (0.09, 4.90)	0.67

Type of difficult delivery:				
Shoulder dystocia	0 (0.0%)	0 (0.0%)	NA	1.00
Multiple vacuum attempts and/or failed vacuum	0 (0.0%)	1 (50.0%)	NA	1.00
Neonatal risk factors				
Sex:				
Male	22 (61.1%)	10 (55.6%)	1.25 (0.34, 4.56)	0.77
Female	14 (38.9%)	8 (44.4%)		
Birth weight:				
<2500 g	4 (11.4%)	1 (6.7%)	1.78 (0.16, 95.18)	1.00
Small for gestational age (<10th percentile)	4 (12.1%)	2 (13.3%)	0.90 (0.11, 11.12)	1.00
Large for gestational age (>90th percentile)	3 (9.1%)	1 (6.7%)	1.39 (0.10, 78.64)	1.00
Head circumference:				
<10th percentile	3 (11.1%)	0 (0.0%)	NA	0.56
≥90th percentile	6 (22.2%)	1 (11.1%)	2.24 (0.21, 118.02)	0.65
5-minute Apgar score <7	5 (14.7%)	4 (26.7%)	0.48 (0.09, 2.90)	0.43
10-minute Apgar score <7	1 (7.7%)	2 (18.2%)	0.39 (0.01, 8.62)	0.58
Received vitamin K	33 (100.0%)	17 (100.0%)	NA	1.00
Resuscitation at birth	17 (50.0%)	8 (47.1%)	1.12 (0.30, 4.25)	1.00
Abnormal cord blood gas	11 (37.9%)	4 (28.6%)	1.78 (0.11, 22.86)	0.61
Infection (sepsis / meningitis)	7 (31.8%)	2 (15.4%)	1.91 (0.31, 20.98)	0.70
Congenital heart disease	9 (45.0%)	2 (33.3%)	1.61 (0.18, 21.67)	1.00
Hypoglycemia	3 (13.6%)	1 (7.7%)	1.53 (0.11, 85.80)	1.00
Central vascular catheterization	6 (20.0%)	6 (46.2%)	0.30 (0.06, 1.51)	0.14
Hematological abnormalities:				
Factor V Leiden	1 (25.0%)	1 (33.3%)	0.37 (0.01, 34.82)	0.51
Protein C & S	3 (75.0%)	0 (0.0%)	NA	0.52
Homocysteine	1 (25.0%)	0 (0.0%)	NA	1.00
Prothrombin	0 (0.0%)	0 (0.0%)	NA	1.00
AT III	1 (25.0%)	0 (0.0%)	NA	1.00

Fibrinogen	2 (50.0%)	0 (0.0%)	NA	1.00

LEGEND: APPIS – arterial presumed perinatal ischemic stroke, AT III– antithrombin III, CHD – congenital heart disease, CS – cesarean section, CSVT – cerebral sinovenous thrombosis, GDM – gestational diabetes mellitus, HSV – herpes simplex virus, NAIS – neonatal arterial ischemic stroke, NHS – neonatal hemorrhagic, NVD – normal vaginal delivery, p – probability, PPHS – presumed perinatal hemorrhagic stroke

The risk factors for perinatal stroke and its subtypes were also compared with those that are present in the general population, from the AIHW total population data (Table 3). After controlling for risks in the general population, significant risk factors for perinatal stroke included: previous history of miscarriage or still birth or neonatal death ($p<0.01$); cesarean section ($p=0.04$); low Apgar score ($p<0.01$); neonatal resuscitation ($p<0.01$), and nulliparity ($p<0.1$).

Table 3: Comparison of risk factors of perinatal stroke with AIHW total population data

Variable	AIHW Data %	Perinatal Stroke Data n (%)	p-value	NAIS (n=36)	p-value	NHS (n=18)	p-value
Maternal age:							
<20 years	5758 (1.9%)	0 (0%)	0.64	0 (0.0%)	0.91	0 (0.0%)	1.00
20-39 years	283,658	48 (96.0%)	0.69	30 (96.8%)	0.72	13 (92.9%)	1.00
40+ years	(93.6%)	2 (4.0%)	1.00	1 (3.2%)	1.00	1 (7.1%)	1.00
	13,637 (4.5%)						
Smoking	30,912 (10.2%)	4 (7.3%)	0.62	2 (5.9%)	0.58	1 (5.9%)	0.91
Alcohol	15,152 (5.0%)	1 (1.8%)	0.44	1 (2.9%)	0.87	0 (0.0%)	0.77
Delivery:							
NVD	193,954 (64%)	16 (28.6%)	**<0.01**	7 (20.6%)	**<0.01**	8 (47.1%)	0.23
CS	109,099 (36%)	28 (50.0%)	**0.04**	21 (61.8%)	**<0.01**	5 (29.4%)	0.75
Forceps	30,608 (10.1%)	5 (8.9%)	0.94	3 (8.8%)	1.00	1 (5.9%)	0.86
Vacuum	39,700 (13.1%)	7 (12.5%)	1.00	3 (8.8%)	0.63	3 (17.7%)	0.84
Breech	12,425 (4.1%)	0 (0.0%)	0.23	0 (0.0%)	0.44	0 (0.0%)	0.81
GDM	33,336 (11.0%)	7 (12.3%)	0.92	5 (14.3%)	0.73	1 (5.9%)	0.77
PIH	6,061 (2.0%)	3 (5.3%)	0.20	2 (5.7%)	0.33	1 (5.9%)	0.78
LBW	20,002 (6.6%)	6 (10.9%)	0.31	4 (11.4%)	0.42	1 (6.7%)	1.00
SGA	29,487 (9.4%)	7 (13.2%)	0.47	4 (12.1%)	0.81	2 (13.3%)	0.33
5-minute Apgar <7	6,364 (2.0%)	9 (16.7%)	**<0.01**	5 (14.7%)	**<0.01**	4 (26.7%)	**<0.01**
Resuscitation	57,580 (19%)	25 (45.5%)	**<0.01**	17 (50.0%)	**<0.01**	8 (47.1%)	**<0.01**

Parity							
One	130,307 (43%)	35 (64.8%)	**<0.01**	21 (65.5%)	**0.02**	12 (70.6%)	**0.04**
≥ Two	172, 732 (57%)	19 (35.2%)		11 (34.5%)		5 (29.4%)	

LEGEND: AIHW – Australian Institute of Health and Welfare, CS – cesarean section, GDM – gestational diabetes mellitus, LBW – low birth weight, NVD – normal vaginal delivery, p – probability, PIH – pregnancy induced hypertension, SGA – small for gestational age.

DISCUSSION

Prevalence of Perinatal Stroke

The estimated birth prevalence of acute symptomatic perinatal stroke in this population-based study was 9.6/100,000 live births per year, compared with 0.54-18.60/100,000 live births reported in studies from USA and 15.87/100,000 live births reported in a Canadian population-based study and in a perinatal stroke registry.(23-25) Our estimates were based on prospective reports from the entire country, and controlled for by birth prevalence in each state. Standardized stroke classification reporting was conducted by specialist physicians.(26, 27)

In our study the birth prevalence of NAIS was 5.8/100,000 live births per year compared with 10.2/100,000 live births in Canada, 7.0/100,000 in Germany, 1.3/100,000 in Denmark with a global pooled incidence of 24.6/100,000 live births.(28, 29) The birth prevalence of NHS in our study was 2.9/100,000 live births per year, which was significantly lower than a retrospective study that quoted 6.2/100,000 and 10.5-15.9/100,000 annually from a combined retrospective prospective study.(30, 31). The two countries with the highest stroke prevalence in the literature have registers, suggesting possible under-reporting in our method and other countries.

Epidemiology

Risk factors associated with stroke subtypes

Our study found no difference in the risk factors between ischemic (NAIS) and hemorrhagic strokes (NHS), except for higher numbers of cesarean section ($p=0.04$) and meconium-stained liquor ($p=0.04$) in NAIS compared to NHS (Table 2). There were 11 infants with CHD: 9 had NAIS and 2 had NHS. No equivalent studies were found in the published literature for comparison. Previous studies have reported maternal infection in the perinatal

and postnatal period as important risk factors, but these were not significant in our sample.(32-34)

Maternal illicit drug use is associated with stroke. In our study, one mother had a history of buprenorphine and her baby had NAIS.(35, 36) The other risk factors in this case were low birth weight, small for gestational age, emergency cesarean section and Apgar score <7, requiring oxygen for resuscitation.

We confirm a male predominance (male:female ratio 1.4:1.0), similar to a high male incidence (male:female ratio 1.5:1.0) quoted in the literature.(29, 37) The cause for male dominance was not clear, though studies have indicated an association between elevated endogenous testosterone and risk of cerebral thromboembolism.(38) Previous meta-analyses have suggested male predominance was not associated with a greater vulnerability of males to adverse neonatal outcome.(20)

In our study cohort we had no genetic findings and no reports of sickle cell anemia, a known risk factor.(34, 39) It was not clear whether this was due to a unique Australian phenomenon, ethnicity of the participants, insufficient data entry, or an artefact of the sample size.

Risk factors compared to population data

Some risk factors were significantly higher in the stroke cases compared to the babies from the control AIHW group (Table 3), for example history of miscarriage, still birth and neonatal death, and cesarean section, consistent with previous studies.(40) However, risks such as miscarriage, still birth and neonatal death were aggregated in the AIHW data, and could not be analyzed individually. This was problematic because still birth and neonatal death are presumed individual risk factors for stroke. Elective and emergency cesarean sections were reported collectively in AIHW population data, whereas in our study 7% had elective cesarean section and 40% had emergency cesarean section. Emergency cesarean

sections are likely to be ascribed to fetal compromise, which is presumed to be a risk factor for stroke.

The 5-minute Apgar score <7 and neonatal resuscitation were significant risk factors. Similar findings have been reported in previous studies.(41) Though emergency cesarean section, low 5 minute Apgar score, meconium stained liquor and neonatal resuscitation were coded as risk factors, these variables in combination could be signs and symptoms of a perinatal stroke in progress. Hence infants with these signs and symptoms should have a brain MRI to detect stroke as early as possible.

In this study there was no association between neonatal infection, CHD and hypoglycemia because of the lack of information on these in the AIHW data set. A well powered case-control study is warranted to further examine the relationship between these risks.

The strength of our study was the prospective comparison with Australian population data, and the main limitation was the inability to obtain MRI images. Therefore, the diagnosis of perinatal stroke was based on the physician's report and the MRI brain report, which may have clinical interpretation differences. There could also be under-reporting due to different ethics requirements in different Australian jurisdictions and the burden of notifications from busy clinicians leading to a small sample size. Under-reporting could potentially have been overcome by a longer study or if allied health experts like physiotherapists were allowed to report cases to the APSU, as currently only doctors can notify.

Not all individual risk factors could be weighed against the AIHW data because of the aggregate nature of this data. Finally, a well-powered case-control study is recommended to further examine individual risk factors.

In conclusion, this Australian population-based study reported the birth prevalence of acute symptomatic perinatal stroke, including for the subtypes. This was the first study to compare

the risk factors for perinatal stroke with the Australian population data (AIHW) and elucidate the significant risk factors which may be present in healthy infants, such as low Apgar scores and cesarean section.

Further research is needed to investigate placental pathology and the role of genetic factors in perinatal stroke. This may then allow even more precise identification of risk factors, leading to preventative measures, earlier diagnosis, and individualized neuroplasticity treatment. Therefore, full population ascertainment reporting to a national pediatric stroke register is critical to substantiate the risk factors and true incidence.

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REFERENCES

1. Chabrier S, Husson B, Dinomais M, Landrieu P, Nguyen The Tich S. New insights (and new interrogations) in perinatal arterial ischemic stroke. Thrombosis Research. 2011;127 13-22.
2. Ilves P, Laugesaar R, Loorits D, Kolk A, Tomberg T, Loo S, et al. Presumed Perinatal Stroke: Risk Factors, Clinical and Radiological Findings. J Child Neurol. 2016;31 621-8.
3. Ramaswamy V, Miller SP, Barkovich AJ, Partridge JC, Ferriero DM. Perinatal stroke in term infants with neonatal encephalopathy. 2004;1 2088-91.
4. Dunbar M, Kirton A. Perinatal stroke: mechanisms, management, and outcomes of early cerebrovascular brain injury. Lancet Child Adolesc Health. 2018;2 666-76.
5. Benders MJNL, Groenendaal F, Uiterwaal CSPM, de Vries LS. Perinatal Arterial Stroke in the Preterm Infant. Seminars in Perinatology. 2008;32 344-9.
6. Fullerton HJ, Wu YW, Sidney S, Johnston SC. Risk of recurrent childhood arterial ischemic stroke in a population-based cohort: the importance of cerebrovascular imaging. Pediatrics. 2007;119 495-501.
7. Laugesaar R, Kolk A, Tomberg T, Metsvaht T, Lintrop M, Varendi H, et al. Acutely and retrospectively diagnosed perinatal stroke: a population-based study. Stroke. 2007;38 2234-40.
8. Nelson KB, Lynch JK. Stroke in newborn infants. Lancet neurol. 2004;3 150-8.
9. Heljic S, Mornjakovic A, Merhemic Z, Maksic H, Hadzipasic E. [Stroke in newborn infants]. Med Arh. 2007;61 62-4.
10. Barnes C, Newall F, Furmedge J, Mackay M, Monagle P. Arterial ischaemic stroke in children. Journal of Paediatrics & Child Health. 2004;40 384-7.
11. Barnes C, Newall F, Furmedge J, Mackay M, Monagle P. Cerebral sinus venous thrombosis in children. Journal of Paediatrics & Child Health. 2004;40 53-5.
12. Gerstl L, Bonfert MV, Heinen F, Olivieri M, Schroeder AS, Tacke M, et al. Childhood Arterial Ischaemic Stroke: Clinical Presentation, Risk Factors and Management. Hamostaseologie. 2020;40 165-73.
13. Lee J, Croen LA, Backstrand KH, Yoshida CK, Henning LH, Lindan C, et al. Maternal and infant characteristics associated with perinatal arterial stroke in the infant. Jama. 2005;293 723-9.
14. Benders MJ, Groenendaal F, Uiterwaal CS, Nikkels PG, Bruinse HW, Nievelstein RA, et al. Maternal and infant characteristics associated with perinatal arterial stroke in the preterm infant. Stroke. 2007;38 1759-65.
15. Teutsch SM, Nunez CA, Morris A, Eslick GD, Khandaker G, Berkhout A, et al. Australian Paediatric Surveillance Unit (APSU) Annual Surveillance Report 2020. Commun Dis Intell (2018). 2021;45 28.
16. von Elm E, Altman DG, Egger M, Pocock SJ, Gotzsche PC, Vandenbroucke JP, et al. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. Bull World Health Organ. 2007;85 867-72.
17. Chabrier S, Saliba E, Nguyen The Tich S, Charollais A, Varlet MN, Tardy B, et al. Obstetrical and neonatal characteristics vary with birthweight in a cohort of 100 term newborns with symptomatic arterial ischemic stroke. European Journal of Paediatric Neurology. 2010;14 206-13.
18. Harteman JC, Groenendaal F, Kwee A, Welsing PM, Benders MJ, de Vries LS. Risk factors for perinatal arterial ischaemic stroke in full-term infants: a case-control study. Archives of Disease in Childhood Fetal & Neonatal Edition. 2012;97 F411-6.

19. Darmency-Stamboul V, Chantegret C, Ferdynus C, Mejean N, Durand C, Sagot P, et al. Antenatal factors associated with perinatal arterial ischemic stroke. Stroke. 2012;43 2307-12.
20. Li C, Miao JK, Xu Y, Hua YY, Ma Q, Zhou LL, et al. Prenatal, perinatal and neonatal risk factors for perinatal arterial ischaemic stroke: a systematic review and meta-analysis. Eur J Neurol. 2017;24 1006-15.
21. Australian Bureau of Statistics. Births, Australia 2019 [Available from: https://www.abs.gov.au/statistics/people/population/births-australia/2018.
22. Australian Institute of Health and Welfare (AIHW). Australia's mothers and babes. Available from: https://www.aihw.gov.au/reports/mothers-babies/australias-mothers-babies/contents/about (Accessed 28 Junel 2022)
23. Surmava AM, Maclagan LC, Khan F, Kapral MK, Hall RE, deVeber G. Incidence and Current Treatment Gaps in Pediatric Stroke and TIA: An Ontario-Wide Population-Based Study. Neuroepidemiology. 2019;52 119-27.
24. Agrawal N, Johnston SC, Wu YW, Sidney S, Fullerton HJ. Imaging data reveal a higher pediatric stroke incidence than prior US estimates. Stroke. 2009;40 3415-21.
25. Dunbar M, Mineyko A, Hill M, Hodge J, Floer A, Kirton A. Population Based Birth Prevalence of Disease-Specific Perinatal Stroke. Pediatrics. 2020;146 11.
26. Pohlman KA, Carroll L, Tsuyuki RT, Hartling L, Vohra S. Comparison of active versus passive surveillance adverse event reporting in a paediatric ambulatory chiropractic care setting: a cluster randomised controlled trial. BMJ open qual. 2020;9 11.
27. Ferriero DM, Fullerton HJ, Bernard TJ, Billinghurst L, Daniels SR, DeBaun MR, et al. Management of Stroke in Neonates and Children: A Scientific Statement From the American Heart Association/American Stroke Association. Stroke. 2019;50 e51-e96.
28. Oleske DM, Cheng X, Jeong A, Arndt TJ. Pediatric Acute Ischemic Stroke by Age-Group: A Systematic Review and Meta-Analysis of Published Studies and Hospitalization Records. Neuroepidemiology. 2021;55 331-41.
29. deVeber GA, Kirton A, Booth FA, Yager JY, Wirrell EC, Wood E, et al. Epidemiology and Outcomes of Arterial Ischemic Stroke in Children: The Canadian Pediatric Ischemic Stroke Registry. Pediatr Neurol. 2017;69 58-70.
30. Armstrong-Wells J, Johnston SC, Wu YW, Sidney S, Fullerton HJ. Prevalence and predictors of perinatal hemorrhagic stroke: results from the kaiser pediatric stroke study. Pediatrics. 2009;123 823-8.
31. Cole L, Dewey D, Letourneau N, Kaplan BJ, Chaput K, Gallagher C, et al. Clinical Characteristics, Risk Factors, and Outcomes Associated With Neonatal Hemorrhagic Stroke: A Population-Based Case-Control Study. JAMA Pediatrics. 2017;171 230-8.
32. Sporns PB, Fullerton HJ, Lee S, Kim H, Lo WD, Mackay MT, et al. Childhood stroke. Nat Rev Dis Prim. 2022;8 12.
33. Vitagliano M, Dunbar M, Dyck Holzinger S, Letourneau N, Dewey D, Oskoui M, et al. Perinatal arterial ischemic stroke and periventricular venous infarction in infants with unilateral cerebral palsy. Developmental Medicine & Child Neurology. 2022;64 56-62.
34. Jankovic M, Petrovic B, Novakovic I, Brankovic S, Radosavljevic N, Nikolic D. The Genetic Basis of Strokes in Pediatric Populations and Insight into New Therapeutic Options. International Journal of Molecular Sciences. 2022;23 29.
35. Armon C. Which drugs increase the risk of stroke during pregnancy? 2019 [Available from: https://www.medscape.com/answers/1149405-164921/which-drugs-increase-the-risk-of-stroke-during-pregnancy.
36. Mullen M. Stroke associated with drug abuse: Medlink Neurology; 2021 [Available from: https://www.medlink.com/articles/stroke-associated-with-drug-abuse.

37. Golomb MR, Fullerton HJ, Nowak-Gottl U, Deveber G, International Pediatric Stroke Study G. Male predominance in childhood ischemic stroke: findings from the international pediatric stroke study. Stroke. 2009;40 52-7.
38. Normann S, de Veber G, Fobker M, Langer C, Kenet G, Bernard TJ, et al. Role of endogenous testosterone concentration in pediatric stroke. 2009;1 754-8.
39. Rodrigues DO, Ribeiro LC, Sudario LC, Teixeira MT, Martins ML, Pittella AM, et al. Genetic determinants and stroke in children with sickle cell disease. J Pediatr (Rio J). 2016;92 602-8.
40. Munoz D, Hidalgo MJ, Balut F, Troncoso M, Lara S, Barrios A, et al. Risk Factors for Perinatal Arterial Ischemic Stroke: A Case-Control Study. Cell med. 2018;10 2155179018785341.
41. Sorg AL, von Kries R, Klemme M, Gerstl L, Weinberger R, Beyerlein A, et al. Risk factors for perinatal arterial ischaemic stroke: a large case-control study. Developmental Medicine & Child Neurology. 2020;62 513-20.

Chapter 6: CASE-CONTROL STUDY FOR RISK FACTORS OF PERINATAL STROKE

6.1 Introduction

The impact of perinatal stroke continues for the life of the child and its aetiology remains unclear, thus hampering prevention research. This comparative study elucidated the risk factors for perinatal stroke in term infants. Our study tested the theory of multifactorial aetiology.

Multivariable logistic regression analysis indicated exposure to smoking during pregnancy (OR: 1.48, 95% CI: 1.09, 1.99), 1-minute Apgar score <7 (OR: 1.54, 95% CI: 1.15, 2.08), 10-minute Apgar score <7 (OR: 1.26, 95% CI: 1.02, 1.54) and hypoglycaemia (OR: 1.49, 95% CI: 1.07, 2.06) are independent risk factors for perinatal stroke. Emergency caesarean section, resuscitation at birth and abnormal cord blood gas are additional risk factors.

6.2 Published Manuscript

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ORIGINAL ARTICLE

Risk factors for perinatal stroke in term infants: A case–control study in Australia

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Aim: The aetiology of perinatal stroke is poorly understood. This study aimed to prospectively confirm the risk factors and identify any previously unknown variables.

Methods: A prospective case–control study was conducted in Australia. Univariate odds ratios (ORs), associated 95% confidence intervals (CIs) and multivariable logistic regression models fitted with backwards stepwise variable selection were used.

Results: Sixty perinatal stroke cases reported between 2017 and 2019 included 95% (57/60) with multiple risk factors. Univariate analysis identified emergency caesarean section rather than NVD ($P < 0.01$), low Apgar score (<7) at 1, 5 and 10 min of age ($P < 0.01$), resuscitation at birth ($P < 0.01$), abnormal cord blood gas ($P < 0.01$), neonatal infection/sepsis ($P < 0.01$), congenital heart disease ($P < 0.01$) and hypoglycaemia ($P < 0.01$) as significant risk factors. Multivariate analysis found smoking during pregnancy (OR: 1.48; 95% CI: 1.09–1.99), 1-min Apgar score < 7 (OR: 1.54; 95% CI: 1.15–2.08), 10-min Apgar score < 7 (OR: 1.26; 95% CI: 1.02–1.54) and hypoglycaemia (OR: 1.49; 95% CI: 1.07–2.06).

Conclusions: Perinatal stroke is associated with multiple risk factors. Exposure to smoking, 10-min Apgar score < 7, neonatal infection and hypoglycaemia were independent risk factors. Emergency caesarean section, resuscitation at birth and abnormal cord blood gas were additional risk factors.

Key words: aetiology; neonatology; neurology; perinatal stroke; risk factors.

What is already known on this topic

1 Perinatal stroke is an important cause of neurological disability in infants.
2 The diagnosis is often missed because of multifactorial risk factors and non-neurological presentation.

What this paper adds

1 Exposure to smoking during pregnancy, 10-min Apgar score < 7, neonatal infection and hypoglycaemia were identified as independent risk factors.

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Conflict of interest: None declared.

Author contributions: Dr Bithi Roy, Prof Iona Novak and Prof Nadia Badawi conceptualised and designed the study and designed the data collection instrument. Dr Bithi Roy collected data, carried out the initial analyses, drafted the initial manuscript and revised the manuscript. Prof Iona Novak critically analysed, reviewed and revised the manuscript for important intellectual content. Prof Nadia Badawi assisted in data collection, critically reviewed and revised the manuscript for important intellectual content. Clinical Prof Karen Walker and Dr Catherine Morgan assisted in data collection, and critically reviewed and revised the manuscript for important intellectual content. Annabel Webb carried out the initial analyses, and critically analysed and revised the manuscript. All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

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Perinatal stroke is a known cause of neurodevelopmental disability in infants. The reported annual incidence of perinatal stroke is about 1 in 2500–4000 live births.[1] Although previous studies have identified several risk factors, aetiology is still poorly under- stood, making it difficult to identify the population at highest risk. Pregnancy elevates the risk for maternal stroke, particularly in the presence of hypertensive disease of pregnancy.[2–4] This increased vulnerability in the mother activates clotting and poses an increased risk of arterial ischemic stroke in the neonate. Stud- ies have included nulliparity, male sex, maternal fever, prema- ture rupture of membranes, meconium-stained liquor, neonatal sepsis and meningitis as significant risk factors for perinatal arte- rial ischemic stroke.[5–7] A meta-analysis of mostly retrospective studies, not based on individual patients, suggested chorioamnionitis and fetal-placental insufficiency are also closely associated with perinatal arterial ischemic stroke.[8]

Some stroke risk factors are also present in healthy infants, compounding the difficulty of identifying infants at risk. This

study aimed to prospectively confirm stroke risk factors in a case–control study and identify any previously unknown variables.

Methods

Study design

This was an Australian case–control study of term infants (>37 weeks' gestational age) with perinatal stroke. The comparison group included three healthy controls for every infant with stroke, matched for gestational age and date of birth (≤7 days).

Case definition

Babies included those who had acute perinatal stroke before 28 days post-natal age, diagnosed by brain magnetic resonance imaging (MRI) and confirmed by the reporting physician.

Case data collection

An active surveillance system based on pre-specified criteria was used for case identification. Perinatal strokes secondary to non-accidental head injury were excluded. Expert physicians, including neonatologists, paediatricians or neurologists from tertiary hospitals, were asked to report babies with perinatal stroke using a deidentified data sheet to the Australian Paediatric Surveillance Unit (APSU) on a weekly basis from 1 July 2017 to 30 June 2019. The APSU is a national resource that facilitates active surveillance of rare childhood diseases.[9] It is closely affiliated with the University of Sydney's Faculty of Medicine and Health Sciences and the Sydney Children's Hospital Network. Data collected included risk factors found in babies with perinatal stroke and control babies, perinatal stroke types, clinical presentation and outcome. Case reports were independently validated, and the perinatal stroke types (ischemic, haemorrhagic and cerebral sinovenous thrombosis (CSVT)) were confirmed from MRI brain reports by three independent raters (NB, CM and BR). Original MRI images were not sought, to lower respondent burden and reduce the likelihood of missing data.

Control definition

Control babies were born at ≥37 weeks of gestation and were not admitted to the neonatal unit.

Control data collection

Control babies were born at a tertiary neonatal unit in a major public hospital in Sydney, Australia. This single hospital is known to treat patients from diverse socio-economic and geographic backgrounds and is widely considered representative of Australian families. Three presumed healthy controls per baby with stroke, in total 180, were randomly selected and matched for gestational age and actual age of the baby with stroke (≤7 days old). The only exclusion criteria over and above no neonatal unit admission were infants with congenital malformations.

Stroke types

Dunbar and Kirton's classification of acute symptomatic perinatal stroke, including arterial or venous and ischemic or haemorrhagic subtypes, were used in this study.[10] Arterial stroke included neonatal arterial ischemic stroke (NAIS) and venous stroke included neonatal haemorrhagic stroke (NHS) and CSVT.

Statistical analysis

All statistical analysis was carried out using R.[11] Frequencies and percentages were calculated for prenatal, perinatal and neonatal risk factors for the perinatal stroke cases, and for the controls, as well as for the NAIS, NHS and CSVT stroke subtypes. Rates of missing data for each risk factor of interest were calculated for babies with stroke. Univariate odds ratios (ORs) and associated

Table 1 Patient population

	Perinatal stroke cases	Controls
Total (*n*)	60	180
Gestational age (weeks), range (mean)	37–42 (39)	37–41 (39)
Birthweight (g), range (mean)	2114–4470 (3294)	1743–4760 (3324)
Male:Female	35:25	98:82

n, frequency counts.

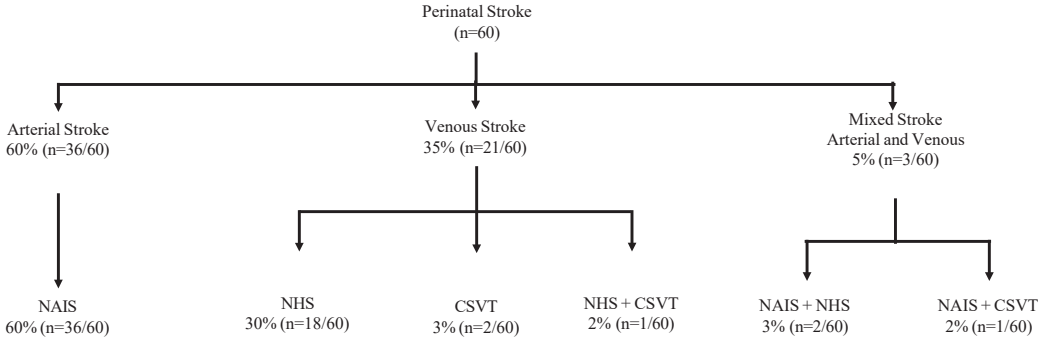

Fig. 1 Types of perinatal stroke (*n* = 60). CSVT, cerebral sinovenous thrombosis; NAIS, neonatal arterial ischemic stroke; NHS, neonatal hemorrhagic stroke.

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Table 2 Univariable risk factor analysis for perinatal stroke cases versus controls

Risk factors	Perinatal stroke cases *n* = 60 *N* (%)	Controls *n* = 180 *N* (%)	OR (95% CI)	*P* value
Prenatal risk factors				
Indigenous status				
Aboriginal or Torres Strait Islander	1 (2%)	9 (5%)	0.36 (0.01–2.70)	0.46
Non-Indigenous	53 (98%)	171 (95%)		
Maternal age				
<20 years	0 (0%)	0 (0%)	NA	1.00
20–34 years	41 (82%)	112 (62%)	(ref)	–
≥35 years	7 (14%)	68 (38%)	0.29 (0.10–0.68)	<0.01
History of miscarriage or stillbirth or neonatal deaths	4 (7%)	70 (39%)	0.12 (0.03–0.35)	<0.01
History of stroke in other children	0 (0%)	0 (0%)	NA	1.00
Multigravida (≥2)	24 (44%)	118 (66%)	0.42 (0.22–0.82)	<0.01
Parity ≥ 2	19 (35%)	85 (47%)	0.61 (0.30–1.19)	0.16
Multiple births – twins/triplets	1 (2%)	2 (1%)	1.51 (0.03–29.41)	1.00
Pregnancy complications:				
Gestational diabetes	7 (12%)	23 (13%)	0.96 (0.33–2.48)	1.00
Pregnancy hypertension	2 (4%)	5 (3%)	1.27 (0.12–8.03)	0.68
Antepartum haemorrhage	2 (4%)	3 (2%)	2.14 (0.17–19.15)	0.60
Breech	0 (0%)	8 (4%)	NA	0.21
History of smoking	4 (7%)	9 (5%)	1.49 (0.32–5.61)	0.51
History of alcohol	1 (2%)	5 (3%)	0.59 (0.01–6.35)	1.00
History of illicit drugs	1 (2%)	1 (1%)	3.36 (0.04–265.97)	0.41
Perinatal risk factors				
GBS colonisation	8 (14%)	23 (17%)	1.11 (0.40–2.79)	0.83
Prolonged rupture of membranes (>24 h)	1 (2%)	5 (3%)	0.70 (0.02–6.42)	1.00
Mode of delivery				
Normal vaginal delivery	16 (32%)	81 (45%)	(ref)	(ref)
Instrumental (vacuum, forceps)	12 (24%)	32 (18%)	1.89 (0.73–4.81)	0.17
Elective caesarean section	4 (8%)	36 (20%)	0.57 (0.13–1.92)	0.43
Emergency caesarean section	24 (48%)	31 (17%)	3.88 (1.72–8.98)	<0.01
Breech	0 (0%)	0 (0%)	NA	NA
Type of difficult delivery				
Shoulder dystocia	0 (0%)	2 (1%)	NA	1.00
Multiple vacuum attempts and/or failed vacuum	1 (2%)	0 (0%)	NA	0.22
Neonatal risk factors				
Sex:				
Male	35 (58%)	98 (54%)	1.17 (0.62–2.22)	0.65
Female	25 (42%)	82 (46%)		
Birthweight < 2500 g	6 (11%)	8 (4%)	2.62 (0.71–9.08)	0.10
Small for gestational age (<10th percentile)	13 (25%)	38 (21%)	1.21 (0.54–2.61)	0.58
Large for gestational age (>90th percentile)	5 (9%)	10 (6%)	1.77 (0.45–6.01)	0.54
1-min Apgar score < 7	21 (39%)	12 (7%)	8.79 (3.72–21.70)	<0.01
5-min Apgar score < 7	9 (17%)	2 (1%)	17.51 (3.46–171.90)	<0.01
10-min Apgar score < 7	3 (5%)	1 (1%)	22.66 (1.74–1221.74)	<0.01
Received vitamin K	55 (100%)	180 (100%)	NA	1.00
Resuscitation at birth	25 (46%)	22 (12%)	5.93 (2.82–12.66)	<0.01
Abnormal cord blood gas	16 (34%)	13 (7%)	6.55 (2.66–16.47)	<0.01
Infection/sepsis	14 (23.3%)	0 (0%)	NA	<0.01
Congenital heart disease	11 (42%)	0 (0%)	NA	<0.01
Hypoglycaemia	5 (8.3%)	0 (0%)	NA	<0.01

CI, confidence interval; n, frequency counts; OR, odds ratio.

95% confidence intervals (CIs) were computed to investigate the association between risk factors and perinatal stroke, including the stroke subtypes. Multivariable logistic regression models fitted with backwards stepwise variable selection were also used to identify independent factors associated with the odds of stroke. Adjusted ORs and associated 95% CIs were computed from these

models. A significance level of 5% was used for all statistical tests.

Statement on ethics

This study was approved by the Sydney Children's Hospital Network Human Research Ethics Committee (Ethics approval no: 2019/ETH06281).

Results

Sixty babies with perinatal stroke were reported in the selected period, including NAIS, NHS and CSVT (Fig. 1) and 180 controls (Table 1). Results were reported according to the revised STROBE statement.[12]

Perinatal stroke subtypes are shown in Figure 1.

Risk factors for stroke

Prenatal, perinatal and neonatal risk factors for perinatal stroke and its subtypes were compared with the controls (Table 2). Risk factors were identified in 95% (57/60) of the perinatal stroke cases.

Univariate analysis

All stroke types

In the univariate analysis of babies with perinatal stroke versus controls, risk factors that were significantly associated with increased odds of stroke (all subtypes) were: birth via emergency caesarean section rather than NVD ($P < 0.01$); low Apgar score (<7) at 1, 5 and 10 min of age ($P < 0.01$); resuscitation at birth ($P < 0.01$); abnormal cord blood gas ($P < 0.01$); neonatal infection/sepsis ($P < 0.01$); congenital heart disease ($P < 0.01$) and hypoglycaemia ($P < 0.01$) (Table 2). Conversely, factors associated with lowered odds of stroke were maternal age above 35 years ($P = 0.02$), history of miscarriage, stillbirth or neonatal death ($P < 0.01$) and multigravida ($P < 0.01$).

Stroke subtypes

Similarly, when comparing the odds of NAIS versus controls, risk factors significantly associated with increased risk of stroke included birth via emergency caesarean (compared to NVD), low Apgar score (<7) at 1 and 5 min of age, resuscitation at birth and abnormal cord blood gas. History of miscarriage, stillbirth or neonatal death, multigravida and maternal age of 35 or above decreased the odds of NAIS. For NHS versus controls, only low Apgar score (<7) at 1, 5 and 10 min of age, resuscitation at birth and abnormal cord blood gas were identified as significant risk factors, with all these factors associated with higher odds of NHS. Due to small group sizes, ORs and CIs were not computed to compare CSVT and controls.

Multivariate analysis

To identify risk factors independently associated with perinatal stroke, we performed multivariable logistic regression. The multivariable model was constructed using all covariates considered in the univariate analysis, apart from those with a rate of missing data above 20%. The covariates included in the full multivariable model are listed in Table 3. Backwards stepwise variable selection was performed to identify significant independent risk factors of stroke.

Table 3 Multivariable logistic regression models for the odds of stroke

	Full model OR (95% CI)	Reduced model OR (95% CI)
Covariates		
Aboriginal or Torres Strait Islander	0.98 (0.68–1.42)	–
Maternal age > 35 years	0.98 (0.85–1.13)	–
History of miscarriage, stillbirth or neonatal death	0.89 (0.73–1.08)	0.87 (0.78–0.98)
Parity ≥ 2	0.93 (0.76–1.14)	–
Multiple birth	0.96 (0.61–1.51)	–
Gestational diabetes	1.09 (0.94–1.26)	–
Pregnancy hypertension	0.99 (0.69–1.41)	–
Antepartum haemorrhage	0.82 (0.59–1.15)	–
History of smoking	1.48 (1.09–1.99)	1.39 (1.06–1.81)
History of alcohol	1.05 (0.78–1.40)	–
Group B Streptococcus infection	1.10 (0.94–1.28)	–
Prolonged rupture of membranes	0.92 (0.59–1.42)	–
Mode of delivery: Normal vaginal delivery	(ref)	–
Mode of delivery: Instrumental (vacuum, forceps)	1.11 (0.93–1.32)	–
Mode of delivery: Elective Caesarean section	1.06 (0.89–1.27)	–
Mode of delivery: Emergency Caesarean section	0.96 (0.82–1.13)	–
Male	0.96 (0.85–1.08)	–
Birthweight < 2500 g	1.04 (0.81–1.33)	–
1-min Apgar score < 7	1.54 (1.15–2.08)	1.61 (1.38–1.89)
5-min Apgar score < 7	1.08 (0.81–1.44)	–
10-min Apgar score < 7	1.26 (1.02–1.54)	1.27 (1.05–1.53)
Resuscitation at birth	1.09 (0.87–1.36)	–
Sepsis/infection	1.35 (0.99–1.82)	1.39 (1.07–1.80)
Hypoglycaemia	2.26 (1.56–3.28)	2.07 (1.50–2.86)

CI, confidence interval; OR, odds ratio.

In the full multivariable logistic regression model (before variable selection), risk factors identified as significantly associated with stroke were: a maternal history of smoking (OR: 1.48; 95% CI: 1.09–1.99); 1-min Apgar score < 7 (OR: 1.54; 95% CI: 1.15–2.08); 10-min Apgar score < 7 (OR: 1.26; 95% CI: 1.02–1.54), and hypoglycaemia (OR: 1.49; 95% CI: 1.07–2.06). After variable selection was performed, all these variables remained significant. Miscarriage, stillbirth and neonatal death were identified as statistically significant but favouring controls. History of miscarriage, stillbirth or neonatal death was associated with a 13% (95% CI: 2–22%) decrease in the odds of stroke in the reduced model.

Table 4 Risk factors for perinatal stroke types versus controls

Risk factors	NAIS (*n* = 36)	OR† (95% CI)	NHS (*n* = 18)	OR† (95% CI)	Control (*n* = 180)
Prenatal risk factors					
Indigenous status					
Aboriginal or Torres Strait Islander	0 (0%)	NA	1 (7%)	1.35 (0.03–11.1)	9 (5%)
Non-Indigenous	34 (100%)		14 (93%)		171 (95%)
Maternal age					
<20 years	0 (0%)	NA	0 (0%)	NA	0 (0%)
20–34 years	27 (87%)	(ref)	11 (79%)	(ref)	112 (62%)
≥35 years	4 (13%)	0.25 (0.06–0.75)	3 (21%)	0.45 (0.08–1.79)	68 (38%)
History of miscarriage or stillbirth or neonatal deaths	3 (9%)	0.15 (0.03–0.50)	0 (0%)	NA	70 (39%)
Multigravida (≥2)	14 (44%)	0.41 (0.18–0.94)	7 (41%)	0.37 (0.11–1.14)	118 (66%)
Parity ≥ 2	11 (34%)	0.59 (0.24–1.36)	5 (29%)	0.47 (0.12–1.50)	85 (47%)
Multiple births – twins/triplets	1 (3%)	2.52 (0.04–49.8)	0 (0%)	NA	2 (1%)
Pregnancy complications					
Gestational diabetes	5 (14%)	1.10 (0.30–3.28)	1 (6%)	0.40 (0.01–2.83)	23 (8%)
Pregnancy hypertension	2 (6%)	2.05 (0.19–13.2)	1 (6%)	2.05 (0.04–19.9)	5 (3%)
Antepartum haemorrhage	1 (3%)	1.68 (0.03–21.6)	1 (6%)	3.44 (0.06–45.6)	3 (2%)
Breech	0 (0%)	NA	0 (0%)	NA	8 (4%)
History of smoking	2 (6%)	1.19 (0.12–6.13)	1 (6%)	1.12 (0.02–8.97)	9 (5%)
History of alcohol	1 (3%)	1.06 (0.02–9.93)	0 (0%)	NA	5 (3%)
History of illicit drugs	1 (3%)	5.34 (0.07–426)	0 (0%)	NA	1 (1%)
Perinatal risk factors					
Group B Streptococcus colonisation	6 (18%)	1.06 (0.32–3.03)	0 (0%)	0.00 (0.00–1.51)	23 (17%)
Prolonged rupture of membranes (>24 h)	1 (3%)	1.06 (0.02–9.93)	0 (0%)	NA	5 (3%)
Mode of delivery					
Normal vaginal delivery	7 (22%)	(ref)	8 (47%)	(ref)	81 (45%)
Instrumental (vacuum, forceps)	6 (19%)	2.16 (0.55–8.15)	4 (24%)	1.26 (0.26–5.12)	32 (18%)
Elective Caesarean section	5 (16%)	1.60 (0.37–6.32)	0 (0%)	0.00 (0.00–1.41)	36 (20%)
Emergency Caesarean section	16 (50%)	5.89 (2.05–18.6)	5 (29%)	1.62 (0.39–6.16)	31 (17%)
Breech	0 (0%)	NA	0 (0%)	NA	–
Type of difficult delivery					
Shoulder dystocia	0 (0%)	NA	0 (0%)	NA	2 (1%)
Multiple vacuum attempts and/or failed vacuum	0 (0%)	NA	1 (6%)	NA	0 (0%)
Neonatal risk factors					
Sex					
Male	22 (61%)	1.31 (0.60–2.97)	10 (56%)	1.05 (0.35–3.20)	98 (54%)
Female	14 (39%)		8 (44%)		82 (46%)
Birthweight < 2500 g	4 (11%)	2.76 (0.57–11.1)	1 (7%)	1.53 (0.03–12.9)	8 (4%)
Small for gestational age (<10th percentile)	9 (27%)	1.40 (0.53–3.45)	3 (20%)	0.94 (0.16–3.71)	38 (21%)
Large for gestational age (>90th percentile)	3 (9%)	1.70 (0.28–7.11)	1 (7%)	1.12 (0.03–9.74)	10 (6%)
1-min Apgar score < 7	15 (44%)	10.8 (4.10–29.7)	6 (40%)	9.13 (2.28–34.9)	12 (7%)
5-min Apgar score < 7	5 (31%)	15.0 (2.32–165)	4 (27%)	30.8 (3.94–372)	2 (1%)
10-min Apgar score < 7	1 (4%)	7.98 (0.10–638)	2 (18%)	37.2 (1.79–2311)	1 (1%)
Received vitamin K	33 (100%)	NA	17 (100%)	NA	180 (100%)
Resuscitation at birth	17 (50%)	7.09 (2.94–17.3)	8 (47%)	6.29 (1.90–20.6)	22 (12%)
Abnormal cord blood gas	11 (79%)	44.8 (10.3–280)	4 (67%)	24.6 (3.20–294)	13 (7%)
Infection/sepsis	7 (19%)	NA	2 (11%)	NA	0 (0%)
Congenital heart disease	9 (45%)	NA	2 (33%)	NA	0 (0%)
Hypoglycaemia	3 (8%)	NA	1 (6%)	NA	0 (0%)

† All ORs are stroke subtype versus control group. CI, confidence interval; n, frequency counts; NAIS, neonatal arterial ischemic stroke; NHS, neonatal hemorrhagic stroke; OR, odds ratio.

Conversely, in the reduced model: maternal history of smoking was associated with a 39% (95% CI: 6–81%) increase in the odds of stroke; a 1-min Apgar score < 7 was associated with a 61% (95% CI: 38–81%) increase in the odds of stroke; a 10-min Apgar score < 7 was associated with a 27% (95% CI: 5–53%) increase in the odds of stroke; neonatal infection/sepsis was associated with a 39% (95% CI: 7–80%) increase in the odds of stroke; and hypoglycaemia was associated with a 107% (95% CI:

50–186%) increase in the odds of stroke. Notably, history of smoking was not found to be a statistically significant predictor of stroke in the univariate analysis but became significant after adjusting for the other maternal and neonatal risk factors included in the logistic regression.

Discussion

The pathophysiology of perinatal stroke remains ambiguous among most babies. A search for specific risk factors justified a prospective case–control study. Most studies to date have been retrospective, preventing an accurate assessment of each of the multiple risk factors. Our study identified exposure to smoking, 10-min Apgar score < 7, neonatal infection and hypoglycaemia as independent risk factors.

Prenatal risk factors

Similar to other studies, perinatal stroke occurred more frequently in nulliparous mothers aged 20–34 years than in mothers aged 35 years or older (82% vs. 14%).[5] In our study, multivari- ate analysis showed exposure to smoking to be an important risk factor. Except for one study, in which it was reported as an insignificant factor, no other studies investigated smoking exposure.[6] As there was only one baby with an Indigenous background, no conclusions could be drawn regarding ethnicity.[13]

Hypertensive disease of pregnancy,[14,15] gestational diabetes,[7] antepartum haemorrhage, miscarriage, stillbirth, neonatal death, maternal alcohol and illicit drug use were not risk factors in our study. Studies have found inconsistent associations between hypertensive disease of pregnancy, gestational diabetes, and arterial ischemic stroke.[5–7,16,17]

Placental pathology was available in only two of our cases and both were abnormal. In general, studies have shown a low utilisation of placental testing, likely due to placental disposal shortly after birth.[18,19]

Perinatal risk factors

In our study, emergency caesarean section and resuscitation at birth were significant risk factors in the univariate analysis and, on adjusting with other variables, fetal distress and 1-min and 10-min Apgar scores <7 were independent risk factors. An Apgar score of <7 likely reflects adverse events during delivery and suggests an important role for fetal distress, emergency caesarean section and subsequent hypoxia-ischemia in the pathogenic pathway of stroke.

Instrumental delivery may indicate trauma to the head and cause haemorrhagic stroke. In our study (Table 2), instrumental delivery was higher in the perinatal stroke cases (24%) than in the controls (18%, OR: 1.89; 95% CI: 0.73–4.81). Also, when compared within the perinatal stroke types (Table 4), instrumen- tal delivery was higher in the NHS than in the NAIS (24% vs. 19%), supporting the hypothesis, although these were not statistically significant.

As in other studies, prolonged rupture of membranes was not significant in our study.[20]

Neonatal risk factors

In our study, the male:female ratio was higher in perinatal stroke cases than in controls (1.4:1.2), though this was not statistically significant. However, this is in keeping with other neurodevelopmental conditions among male babies.[21,22] There were no uniform results regarding sex and perinatal stroke in previous studies.[17,23,24]

Neonatal infection was an independent risk factor in our study, though GBS colonisation and prolonged rupture of membranes were not significant.[17] Gestational diabetes was similar in both babies with perinatal stroke and controls in our data, but neonatal hypoglycaemia was an independent risk factor. Other studies have also confirmed that hypoglycaemia was an independent risk factor for perinatal arterial ischemic stroke.[5,25] Unlike in some of the published studies, low birthweight, and small and large for gestational age were not risk factors in our study.[8,26] Congenital heart disease is an important risk factor for perinatal arterial ischemic stroke.[27] It was a significant risk factor in our study in the univariate analysis, but it could not be included in the multivariate analysis because of the missing data and a potential risk of bias.

Limitations

Although this study has strengths in its prospective nature, its main limitation was the lack of MRI images of the brain. The diagnosis of stroke was based on MRI reports and reporting phy- sicians. However, in Australia, paediatric MRIs are performed in tertiary care hospitals, so MRI reporting is done by senior radiolo- gists and specialists.

A final limitation of the current study is that small group sizes for some risk factors and some stroke subtypes may have limited the statistical power to detect significant ORs. The CIs for some ORs were extremely wide, which may limit interpretability.

Conclusion

This comparative study confirmed exposure to smoking, 10-min Apgar score < 7, neonatal infection, and hypoglycaemia as independent risk factors for perinatal stroke. Emergency caesarean section, resuscitation at birth and abnormal cord blood gas were additional risk factors. Our study also supports the theory of multifactorial aetiology, with a combination of prenatal, perinatal, and neonatal risk factors being involved in the pathogenesis of perinatal stroke.

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References

1 deVeber GA, Kirton A, Booth FA *et al*. Epidemiology and outcomes of arterial ischemic stroke in children: The Canadian Pediatric Ischemic Stroke Registry. *Pediatr. Neurol.* 2017; 69: 58–70.
2 Ban L, Abdul Sultan A, Stephansson O *et al*. The incidence of first stroke in and around pregnancy: A population-based cohort study from Sweden. *Eur. Stroke J.* 2017; 2: 250–6.
3 Katsafanas C, Bushnell C. Pregnancy and stroke risk in women. *Neurobiol. Dis.* 2022; 169: 105735.
4 Curry CJ, Bhullar S, Holmes J, Delozier CD, Roeder ER, Hutchison HT. Risk factors for perinatal arterial stroke: A study of 60 mother-child pairs. *Pediatr. Neurol.* 2007; 37: 99–107.
5 Harteman JC, Groenendaal F, Kwee A, Welsing PMJ, Benders MJNL, De Vries LS. Risk factors for perinatal arterial ischaemic stroke in full-term infants: A case-control study. *Arch. Dis. Child. Fetal Neonatal Ed.* 2012; 97: F411–F6.
6 Chabrier S, Saliba E, Nguyen The Tich S *et al*. Obstetrical and neonatal characteristics vary with birthweight in a cohort of 100 term newborns with symptomatic arterial ischemic stroke. *Eur. J. Paediatr. Neurol.* 2010; 14: 206–13.
7 Darmency-Stamboul V, Chantegret C, Ferdynus C *et al*. Antenatal factors associated with perinatal arterial ischemic stroke. *Stroke* 2012; 43: 2307–12.
8 Li C, Miao JK, Xu Y *et al*. Prenatal, perinatal and neonatal risk factors for perinatal arterial ischaemic stroke: A systematic review and meta-analysis. *Eur. J. Neurol.* 2017; 24: 1006–15.
9 Teutsch SM, Nunez CA, Morris A *et al*. Australian Paediatric Surveillance Unit (APSU) annual surveillance report 2020. *Commun. Dis. Intell.* 2018; 2021: 28.
10 Dunbar M, Kirton A. Perinatal stroke: Mechanisms, management, and outcomes of early cerebrovascular brain injury. *Lancet Child Adolesc. Health* 2018; 2: 666–76.
11 R Core Team. *R: A Language and Environment for Statistical Computing*, R Foundation for Statistical Computing. Vienna, Austria; 2022 Available from: https://www.R-project.org/
12 von Elm E, Altman DG, Egger M *et al*. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: Guidelines for reporting observational studies. *Bull. World Health Organ.* 2007; 85: 867–72.
13 Edmonds LK, Kandasamy Y, Lamont A, O'Connor S. Perinatal arterial ischemic stroke in northern Queensland. *Am. J. Perinatol.* 2012; 29: 483–8.
14 Lee J, Croen LA, Lindan C *et al*. Predictors of outcome in perinatal arterial stroke: A population-based study. *Ann. Neurol.* 2005; 58: 303–8.
15 Wu YW, Lynch JK, Nelson KB. Perinatal arterial stroke: Understanding mechanisms and outcomes. *Semin. Neurol.* 2005; 25: 424–34.
16 Lee J, Croen LA, Backstrand KH *et al*. Maternal and infant characteristics associated with perinatal arterial stroke in the infant. *JAMA* 2005; 293: 723–9.
17 Martinez-Biarge M, Cheong JL, Diez-Sebastian J, Mercuri E, Dubowitz LM, Cowan FM. Risk factors for neonatal arterial ischemic stroke: The importance of the intrapartum period. *J. Pediatr.* 2016; 173: 62–8.e1.
18 Roy B, Arbuckle S, Walker K *et al*. The role of the placenta in perinatal stroke: A systematic review. *J. Child Neurol.* 2020; 35: 773–83.
19 Roy B, Gonzalez J, Kwok A *et al*. Routine placental histopathological examination: Provides answers in neonatal management. *J. Pediatr.* 2022; 8: 10–3.
20 Munoz D, Hidalgo MJ, Balut F *et al*. Risk factors for perinatal arterial ischemic stroke: A case-control study. *Cell Med.* 2018; 10: 2155179018785341.
21 Dan B. Sex differences in neurodevelopmental disorders. *Dev. Med. Child Neurol.* 2021; 63: 492.
22 May T, Adesina I, McGillivray J, Rinehart NJ. Sex differences in neurodevelopmental disorders. *Curr. Opin. Neurol.* 2019; 32: 622–6.
23 Golomb MR, Fullerton HJ, Nowak-Gottl U, Deveber G, International Pediatric Stroke Study Group. Male predominance in childhood ischemic stroke: Findings from the international pediatric stroke study. *Stroke* 2009; 40: 52–7.
24 Golomb MR, Dick PT, MacGregor DL, Curtis R, Sofronas M, deVeber GA. Neonatal arterial ischemic stroke and cerebral sinovenous thrombosis are more commonly diagnosed in boys. *J. Child Neurol.* 2004; 19: 493–7.
25 Benders MJ, Groenendaal F, Uiterwaal CS *et al*. Maternal and infant characteristics associated with perinatal arterial stroke in the preterm infant. *Stroke* 2007; 38: 1759–65.
26 Sorg AL, von Kries R, Klemme M *et al*. Risk factors for perinatal arterial ischaemic stroke: A large case-control study. *Dev. Med. Child Neurol.* 2020; 62: 513–20.
27 Gerstl L, Bonfert MV, Heinen F *et al*. Childhood arterial ischaemic stroke: Clinical presentation, risk factors and management. *Hamostaseologie* 2020; 40: 165–73.

Chapter 7: A REPORT ON PERINATAL STROKE IN AUSTRALIA

7.1 Introduction

The final study was a population-based prospective longitudinal follow-up study (from Studies 4 and 5) of perinatal stroke in children up to 2 years of age. This age range was prioritized because the first two years are critical in ameliorating deficits through appropriate and timely interventions.

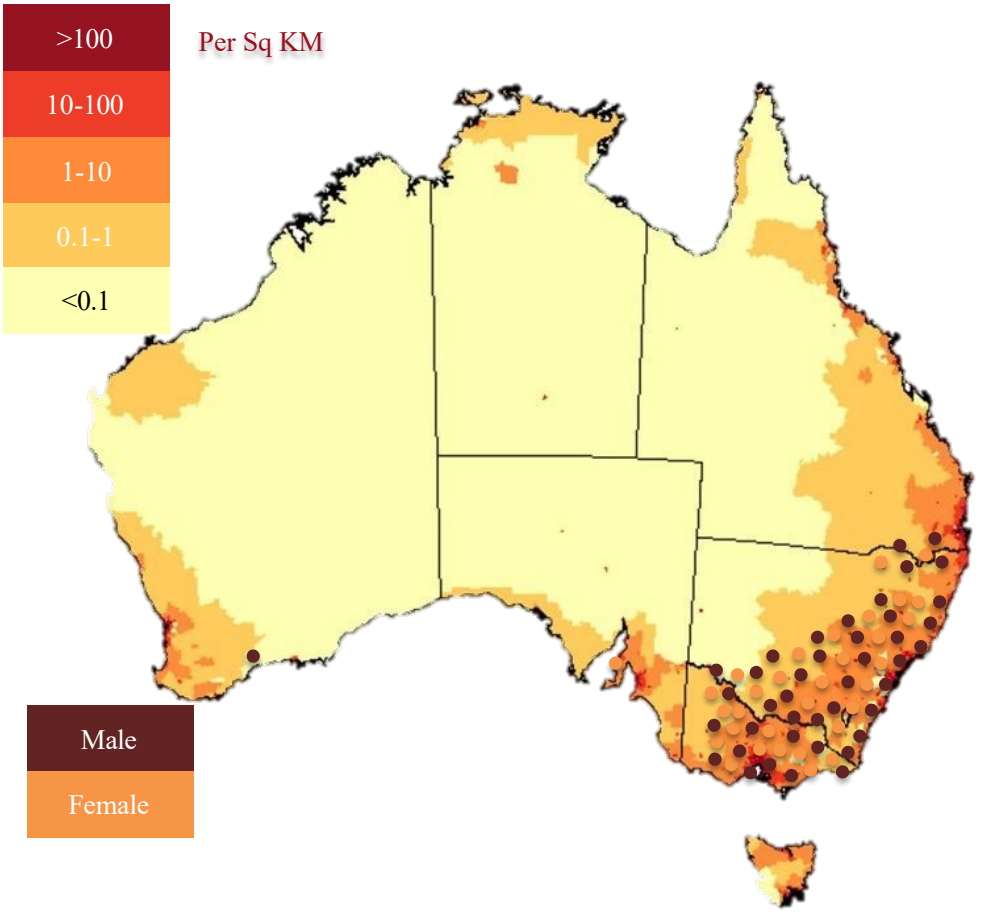
There were 87 children with perinatal stroke reported in Australia and enrolled in this study (Figure 3). Clinical characteristics and outcomes of perinatal stroke were analysed. Seventy two percent of perinatal stroke occurred in the first week of life. The study highlighted the burden of perinatal stroke with 48% disability rate. The disability rate increased to 72% with presumed perinatal stroke. The incidence of cerebral palsy (CP) was 29% with acute symptomatic perinatal stroke and 72% with presumed perinatal stroke.

The first week of a child's life was most critical in terms of lifelong disability. Universal screening programme for CP will help to reduce the incidence for CP. Collaborative studies are needed to ensure timely diagnosis despite non-neurological presentations and allow early implementation of multidisciplinary therapy service.

7.2 Manuscript Under Review

Roy B, Webb A, Walker K, Morgan C, Badawi N, Novak I. A report on perinatal stroke in Australian children: population-based longitudinal study.

Figure 3: Perinatal stroke cases from Australia (in the population density map)



Clinical Characteristics and Outcomes of Perinatal Stroke in Australia: Population-based Longitudinal Study

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Article Summary
Unlike adults, perinatal stroke may not present with neurological symptoms. Non-neurological symptoms may delay diagnosis and may delay intervention.

What's Known on This Subject
Perinatal stroke in children occurs most frequently in the first month of life. Cerebral palsy is an important consequence of perinatal stroke.

What This Study Adds
The Non-neurological presentations of perinatal stroke can lead to a cascade of adverse outcomes in children due to delayed diagnosis and delayed initiation of interventional therapy.

CONTRIBUTORS STATEMENT

Dr Bithi Roy, Prof Iona Novak and Prof Nadia Badawi conceptualized and designed the study and designed the data collection instrument.

Dr Bithi Roy collected data, carried out the data curation, initial analyses, drafted the initial manuscript, and revised the manuscript.

Prof Iona Novak critically analysed and reviewed and revised the manuscript for important intellectual content.

Prof Nadia Badawi, Clinical Prof Karen Walker and Dr Catherine Morgan assisted in data collection, data curation and critically reviewed and revised the manuscript for important intellectual content.

Annabel Webb carried out the initial analyses, and critically analysed and revised the manuscript.

All authors approved the final manuscript as submitted and agree to be accountable for all aspects of the work.

Abstract

Background: The incidence and severity of cerebral palsy (CP) is decreasing, but the incidence of hemiplegic CP remains the same because the perinatal stroke rate is unchanged. Perinatal stroke is one of the main causes of hemiplegic CP. This study aimed to analyse the clinical characteristics and outcomes of perinatal stroke in a cohort of Australian children for early detection of CP.

Methods: A population-based prospective longitudinal study on perinatal stroke up to 2 years of age, was conducted from 2017 to 2019.

Results: Eighty-seven children with perinatal stroke were reported. Seventy-four percent (51/69) of acute symptomatic perinatal strokes presented in the first 3 days of age and 78% (14/18) of presumed perinatal strokes presented within 6 months of age. Sixty-two percent had an arterial stroke, 29% had a venous stroke and 5% had a combined arterial and venous stroke.

Unexpectedly, 35% (24/69) with acute symptomatic perinatal stroke had only respiratory symptoms and 50% (9/18) with presumed perinatal stroke were asymptomatic. The incidence of CP was 29 % (20/69) with acute symptomatic perinatal stroke and 72% (13/18) with presumed perinatal stroke.

Conclusions: The first week of a child's life is the most critical period in terms of lifelong disability. Universal screening programme for CP will help to reduce the incidence for CP.

INTRODUCTION

Although the incidence of cerebral palsy (CP) has decreased, the incidence of hemiplegic CP has not changed as perinatal stroke rates have not changed.(1) Because of life expectancy and involvement of all family members, the burden of post-stroke care in children is believed to be much greater than that of post-stroke care in adults.(2) National strategies for perinatal stroke prevention, early detection, and treatment to reduce the severity of CP are top priorities for health systems and families caring for CP.

Perinatal stroke is the most common cause of hemiplegic CP.(3) It is defined as a clinical syndrome due to focal cerebral interruption of blood supply between 20 weeks of gestation and 28 days of postnatal age.(4) There are two types of perinatal stroke depending on the age of diagnosis: a) acute symptomatic perinatal stroke – which are diagnosed within the perinatal period (<28 days of age), and b) presumed perinatal stroke – which are diagnosed after 28 days of age when they present with motor abnormality or delayed milestones.(5) A composite classification of perinatal stroke includes the type of blood vessel affected and age at diagnosis.

The aetiology is multifactorial and some of the risk factors are present in healthy children. Some of the known risk factors includes infection, foetal distress, hypoglycaemia, congenital heart disease, thrombophilia, and prothrombotic disorders. Despite the critical role of placenta, which is a highly vascular organ between the mother and the foetus, the aetiological role is not completely clear because of lack of placental histopathology reports.(6) The other causes include head and neck trauma, genetic abnormalities, and inborn errors of metabolism. A lower socioeconomic status has also been associated with higher risk of stroke.(7)

Seizures are a common symptom of acute symptomatic perinatal stroke.(8) However, the nonspecific presenting symptoms such as poor feeding, respiratory distress, apnoea, cyanosis, or lethargy can lead to delayed diagnosis or misdiagnosis.(9, 10) Presumed perinatal stroke

usually presents with motor asymmetry, abnormal muscle tone, delayed milestones, and/or epilepsy.(11) Most of these presentations are in the first two years of life when the developmental milestones are emerging.

The gold standard for early diagnosis of perinatal stroke is based on magnetic resonance imaging (MRI) of the brain.(12, 13) For diagnosis later in infancy, the workup involves a combination of meticulous clinical history taking and a series of motor and neurological assessments such as Prechtl's qualitative assessment of General Movements (GMA), Hammersmith Infant Neurological Examination (HINE), and Hand Assessment in Infants (HAI), which are known to be the best early predictors for cerebral palsy at 3-4 months of age.(14)

In this prospective longitudinal study, we aimed to describe the clinical characteristics and neurodevelopmental outcomes in a cohort of children under 2 years of age with perinatal stroke in Australia. The 2 year follow-up point was prioritized because the first two years are critical in ameliorating deficits through appropriate and timely interventions and a conventional time point for confirmation of disability.

An insight into early diagnosis and early intervention strategies are important for ameliorating the deficits through timely and evidence-based interventions such as physiotherapy, occupational therapy, speech pathology, exercise physiology and assistive technology and equipment assistance to improve daily living skills that harnesses neuroplasticity.

METHODS

Study Design

This was a two-year population-based longitudinal prospective follow-up study conducted at the University of Sydney, Australia between July 1, 2017 and June 30, 2019. Data were prospectively collected via physician (neonatologist, paediatrician, or neurologist) report of

perinatal stroke cases to the Australian Paediatric Surveillance Unit (APSU). APSU is a national body affiliated with The University of Sydney, Australia and is responsible for population surveillance of rare childhood diseases.

Physicians completed a questionnaire designed by the investigators (BR, IN, NB) based on key variables identified from published systematic reviews and large case-control studies.(8, 15, 16) Data included postal zip code, risk factors of stroke, clinical presentation, investigations, management, and outcome of stroke. Results were reported according to the revised STROBE statement.(17)

Participants

Inclusion criteria were –

1) Children less than two years of age with perinatal stroke occurring between 2017 and 2019 residing anywhere in Australia.
2) Perinatal stroke diagnosed clinically and/or by Magnetic Resonance Imaging (MRI) of the brain.

Exclusion criteria was –

1) Strokes secondary to head injury.

Analysis

Study data were collected and managed using REDcap® electronic data capture tools. Characteristics for mother and children, clinical presentations, and outcomes were reported using descriptive statistics as frequency counts (N) and percentages (%). The frequency of missing data was reported for each variable of interest. Differences in long-term outcomes between stroke subtypes were investigated using Fisher's exact tests. A p-value of 0.05 was considered statistically significant.

Statement on Ethics

This study was approved by the Sydney Children's Hospital Network Human Research Ethics Committee (Ethics approval no: 2019/ETH06281).

RESULTS

Patient demographics

Characteristics of 87 children with perinatal stroke (Table 1) reported prospectively from New South Wales (53%; 46/87), Victoria (33%; 29/87), Queensland (12%; 10/87) and one each from Western Australia and South Australia (Figure 1) including the aetiological subtypes and birth prevalence (Figure 2).

Table 1: Perinatal stroke characteristics (n=87)

Characteristics of mother	n (%) [Total=87 cases]
Maternal age	
20-35 years	57 (66%)
>35 years	11 (13%)
Missing	19 (22%)
Racial background	
Caucasian	54 (62%)
Indigenous	1 (1%)
Pacific islander	2 (2%)
Asian	10 (12%)
Other	12 (14%)
Missing	8 (9%)
Parity	
Nullipara	44 (54%)
Multiparity (>1)	35 (40%)
Missing	8 (9%)
Miscarriage	7 (8%)
Stillbirth	0
Neonatal death	0
Missing	8 (9%)
Plurality	
Singletons	84 (97%)
Twins and triplets	3 (3%)
Missing	0
Type of conception	
Natural	74 (85%)
In vitro fertilisation	2 (2%)
Other	2 (2%)

Missing	9 (10%)
Pregnancy complications	
Fetal distress	20 (23%)
Group B Streptococcal colonisation	9 (10%)
Gestational diabetes mellitus	10 (11%)
Prolonged rupture of membranes (>24 hours)	3 (3%)
Herpes Simplex infection	1 (1%)
Parvovirus infection	1 (1%)
Missing	3 (3%)
Abnormal antenatal Ultrasound	14 (16%)
Missing	7 (8%)
Meconium-stained liquor	15 (17%)
Missing	5 (6%)
Placental pathology available	4 (5%)
Chorioamnionitis	3 (3%)
Missing	83 (95%)
Maternal medication (metformin, antidepressant, prednisolone, aspirin, thyroxine, insulin, hydralazine, fluconazole)	20 (23%)
Missing	6 (7%)
Smoking	5 (6%)
Missing	11 (13%)
Alcohol	2 (2%)
Missing	14 (16%)
Illicit drugs (methadone, oxycodone, benzodiazepam, quetiapine, buprenorphine, barbiturates)	3 (3%)
Missing	10 (12%)
Family history of stroke	0
Missing	4 (5%)
Mode of delivery	
Normal vaginal delivery	28 (32%)
Instrumental delivery	14 (16%)
Emergency caesarean section	31 (36%)
Elective caesarean section	8 (9%)
Missing	5 (6%)
Difficult delivery	1 (1%)
Missing	10 (12%)
Hematological / Genetic tests attended	10 (12%)
Abnormal studies	4/10 (40%)
Characteristics of children	
Gestational age	
Preterm (<37 weeks)	14 (16%)
Term (37-42 weeks)	65 (75%)
Post term	2 (2%)
Missing	6 (7%)
Males	48 (55%)

Females Missing	39 (45%) 0
Apgar scores 1-minute Apgar score <7 5-minute Apgar score <7 10-minute Apgar score <7 Missing	 26 (30%) 11 (13%) 4 (5%) 10 (12%)
Abnormal cord blood gas Missing	19 (22%) 22 (25%)
Resuscitation at birth Missing	35 (40%) 6 (7%)
Birth weight Normal birth weight Low birth weight (<2500 grams) Small for gestational age (<10th percentile) Large for date (>90th percentile) Missing	 60 (69%) 18 (21%) 16 (18%) 9 (10%) 8 (9%)
Head circumference <10th or >90th percentile Missing	 15 (17%) 33 (38%)
IM Vitamin K Oral vitamin K	87 (100%) 0
Central vascular catheterisation Missing	10 (12%) 14 (16%)
Congenital heart disease Missing	15 (17%) 47 (54%)
Infection Positive blood culture Positive blood and cerebrospinal fluid culture Missing	22 (25%) 8 (9%) 1 (1%) 7 (8%)
Hypoglycaemia (Blood sugar level <2.6 mmol/L)	5 (6%)
Hematological / Genetic tests attended Abnormal studies	23 (26%) 11/23 (48%)

*n – frequency counts.

Note regarding overlapping variables: some babies had more than one characteristic such as with pregnancy complications or birth weight (low birth weight and small for gestational age), which means the count data does not always equal 87.

Figure 1: Perinatal stroke from Australia (shown in the population density map)

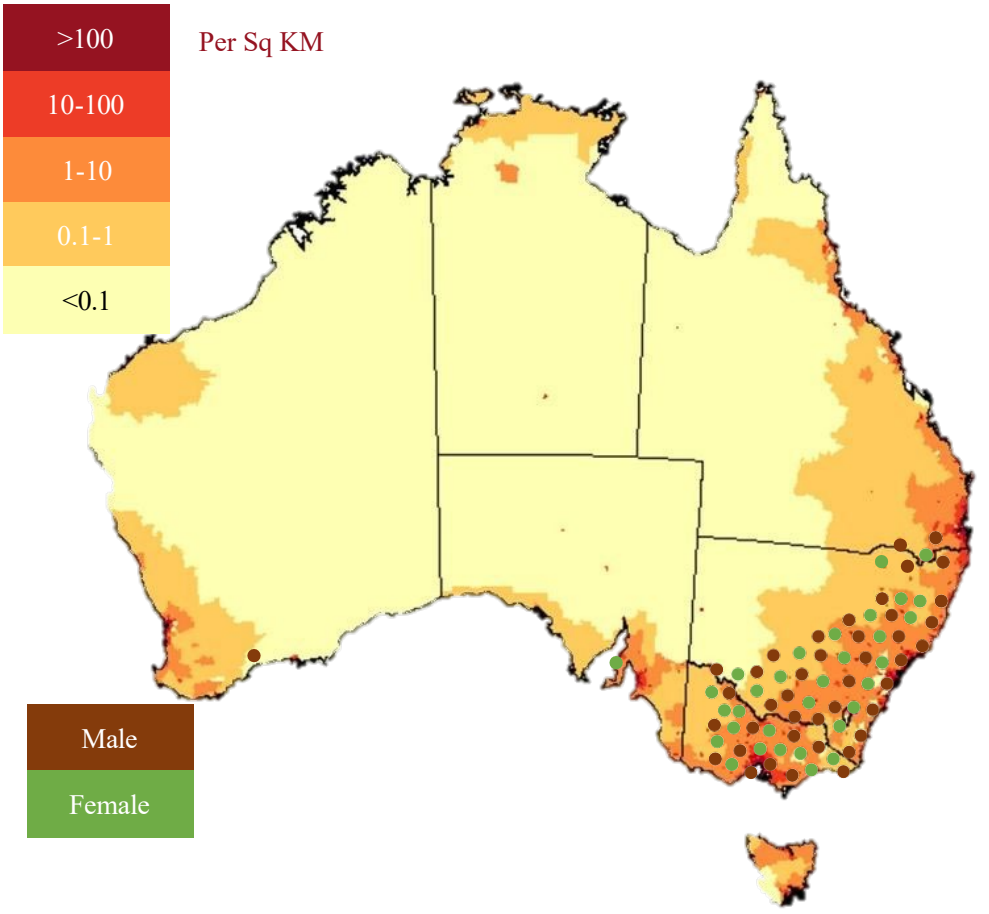
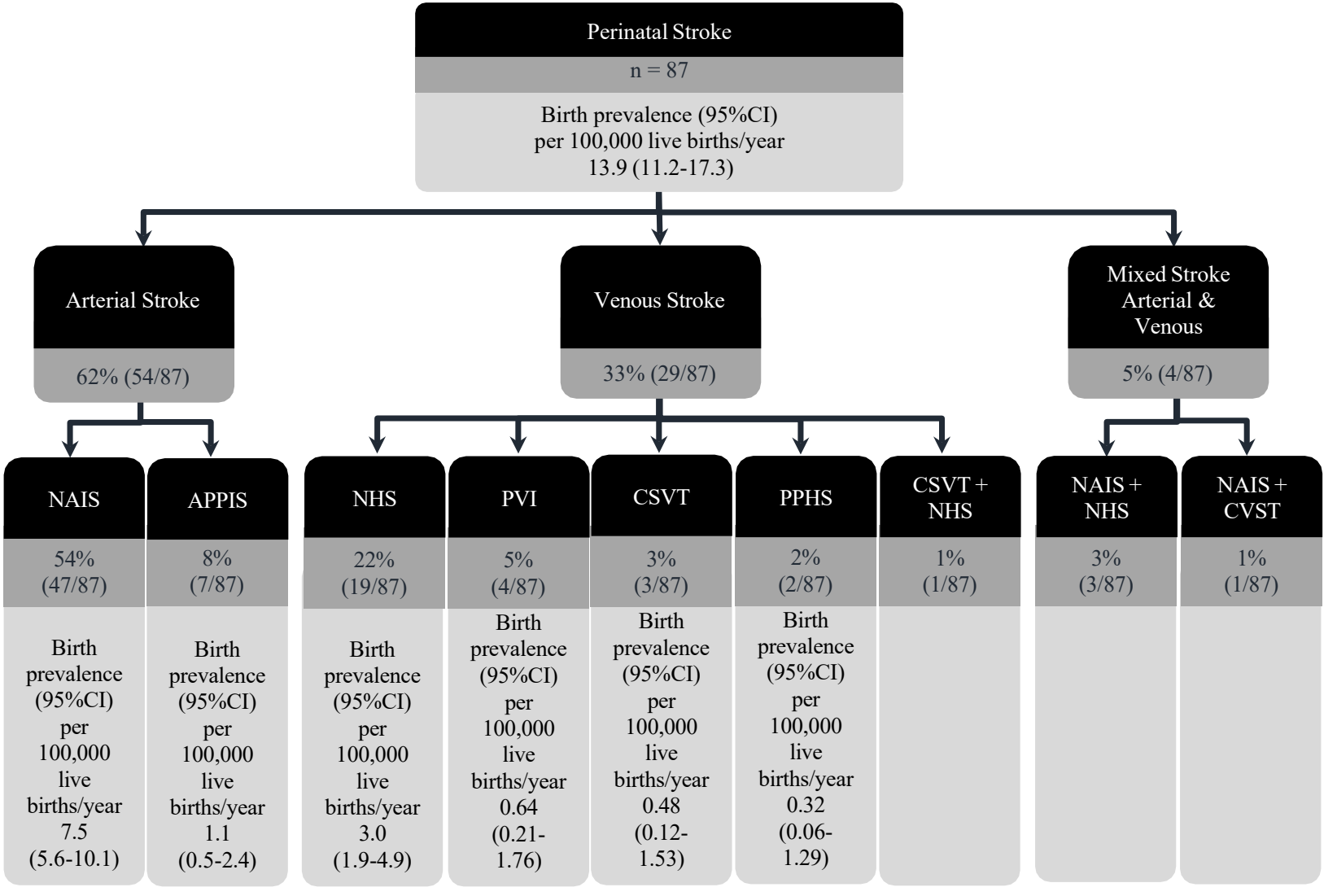


Figure 2: Perinatal stroke subtypes (n=87) and Birth prevalence



*APPIS – arterial presumed perinatal stroke, CSVT – cerebral sinovenous thrombosis, PPHS – presumed perinatal haemorrhagic stroke, PVI – periventricular venous infarction, NAIS – neonatal arterial ischaemic stroke, NHS – neonatal haemorrhagic stroke

Clinical presentations

Clinical presentations of perinatal stroke were analysed separately for acute symptomatic (n=69) and presumed perinatal strokes (n=18) (Table 2). Seventy four percent (51/69) of acute symptomatic perinatal stroke presented in the first 3 days of life, 17% (12/69) between 4 and 7 of age, and 9% (6/69) between 7 and 28 days of age. Seventy eight percent (14/18) of presumed perinatal stroke presented within 6 months of age and 22% (4/18) between 6 and 24 months of age.

Table 2: Clinical presentations

Clinical presentations	Acute symptomatic perinatal stroke (n=69)	Presumed perinatal stroke (n=18)	*p value*
Seizures - Focal seizure on right side - Focal seizure on left side - Multifocal (generalised)	65% (45/69) 20% (9/45) 31% (14/45) 49% (22/45)	0	**<0.001**
Lethargy / poor feeding / respiratory symptoms	77% (53/69)	44% (8/18)	**0.034**
Only respiratory symptoms	35% (24/69)	0	**0.002**
Asymptomatic	0	50% (9/18)	**<0.001**
Missing	0	1	

*n – frequency counts.

Investigations

All children were diagnosed with brain MRI, and 57% (50/87) had an initial cranial ultrasound (cUS). On brain MRI, 62% of the stroke lesions were diagnosed as arterial and 33% as venous stroke. Electroencephalogram (EEG) was performed in 47% (41/87). Echocardiography was performed in 46% (40/87), of which 38% (15/40) had cardiac abnormalities, which included atrial septal defect (excluding patent foramen ovale),

ventricular septal defect, a combination of atrial and ventricular septal defect, and patent ductus arteriosus (2 cases each) and patent ductus arteriosus with tricuspid regurgitation and pulmonary hypertension, transposition of great arteries and tetralogy of Fallot (1 case each). Hematological and genetic studies were reported in 12% (10/87) of mothers, of which 40% (4/10) were abnormal and in 26% (23/87) of the children, of which 48% (11/23) were abnormal. The abnormalities were factor V Leiden, Protein C and S, homocysteine, antithrombin III and fibrinogen anomalies.

Acute Treatment

Forty one percent (36/87) received respiratory support and 7% (6/87) required therapeutic cooling. Eight percent (7/87) required anticoagulant therapy, such as unfractionated heparin infusion (2/87), low molecular weight heparin (2/87), oral aspirin (2/87), and cryoprecipitate (1/87). Three children required ventriculo-peritoneal shunt placement.

Outcomes

The perinatal stroke outcome was analysed separately for acute symptomatic and presumed perinatal strokes (Table 3). Perinatal stroke outcomes were reported in different age groups, ranging from 3 months to 24 months, so there were different motor sequels such as abnormal General Movements assessments, abnormal motor tone (hypertonia and hypotonia) and hemiplegia, depending on the developmental course of the disorder.

Table 3: Outcome of Perinatal stroke

	Acute symptomatic perinatal stroke (n=69)	Presumed perinatal stroke (n=18)***	p value
Normal	48% (33/69)	17% (3/18)	0.023
Death	1% (1/69)	0	0.999
Neurodeficits	42% (29/69)	72% (13/18)	0.064

- Early CP (generalised hyper or hypotonia)	6% (4/69)	0	0.329
- Hemiplegic cerebral palsy Right hemiplegia Left hemiplegia	20% (14/69) 57% (8/14) 43% (6/14)	72% (13/18) 69% (9/13)** 31% (4/13)	<0.001 0.695
- Diplegic cerebral palsy	3% (2/69)	0	0.999
- Abnormal General Movements Absent fidgety Poor repertoire	7% (5/69) 60% (3/5) 40% (2/5)	6% (1/18) 100% (1/1) 0% (0/1)	0.999
- Hearing loss	3% (2/69)	6% (1/18)	0.498
- Language delay/aphasia	1% (1/69)	6% (1/18)	0.366
- Delayed milestones	1% (1/69)	0	0.999
- Seizure disorder/epilepsy	0	28% (5/18)	<0.001
Missing	6	2	

*n – frequency counts.
** – one child with right side hemiplegia was associated with hearing loss, another with aphasia, and another with abnormal General Movements.
*** There were overlapping neurodeficit outcomes for presumed perinatal stroke, meaning that the count data is greater than 18 for this group.

DISCUSSION

Between 1997 and 2014, the overall rate of CP decreased by 30% in high-income countries, whereas the rate of hemiplegic CP showed no such decrease.(1) There are no studies outlining the cost burden of cerebral palsy in Australia, but the average estimated cost is AUD 43, 431 per person per year.(18) This study analysed a range of clinical presentations to help the clinicians (neonatologists and paediatricians) and related health professionals (physiotherapists, occupational therapists, and speech pathologists) to recognise the signs and symptoms of perinatal stroke. A cost benefit analysis of CP indicates that the best practices for early detection of CP may include universal screening for CP.(19) This will allow timely detection of the onset of the disability, lower the severity of neurodeficit and lessen the burden of cost on the health care system and families of children with CP.

In this national cohort of 87 children with perinatal stroke (Figure 2), birth prevalence of perinatal stroke and its subtypes were comparable to the global pooled data.(20)

Perinatal stroke characteristic (Table 1) showed that most were singletons (97%) following a spontaneous pregnancy (85%), from nulliparous (54%), Caucasian (62%) mothers, born by emergency caesarean section (36%) with 1, 5 or 10-minute Apgar score of <7. Some of these characteristics are also found in healthy children, but similar associations have been cited in a meta-analysis study, suggesting careful consideration should be given in suspected children.(21) There were more males (55%) in our study, which was supported by a case-control study (62%), though, the cause for such correlation was not clear.(22) Placental pathology was reported in only 4 cases, 3 of which were intrauterine infections, similar to the rarity of placental pathology in the literature.(23, 24) However, chorioamnionitis has been described as an independent risk factor for arterial ischaemic stroke.(21)

Seizures are one of the most common presentations of perinatal stroke (Table 2), though, non-conventional presentations have been described by other authors.(5, 24, 25) In our study, 65% (45/69) with an acute symptomatic perinatal stroke presented with seizures and unexpectedly 35% (24/69) had only respiratory symptoms, such as tachypnoea, apnoea, cyanosis, and/or respiratory distress. We can safely conclude that those presenting with purely respiratory symptoms are at risk of presumed perinatal stroke due to late diagnosis. Fifty percent (9/18) of presumed perinatal strokes were asymptomatic, and 44% (8/18) had nonspecific signs and symptoms in the perinatal period. It is not clear why some infants are asymptomatic or have non-neurological symptoms. Another likely reason for the occurrence of presumed perinatal stroke is the difficulty in recognising stroke in unwell newborns.

Perinatal stroke has some of the most debilitating lifetime lasting disabilities, namely cerebral palsy. Adverse outcome (Table 3) was present in 42% (29/69) of acute symptomatic perinatal stroke and 78% of presumed perinatal stroke. Additionally, the incidence of CP was higher with presumed perinatal stroke (72% versus 29%). While motor deficits are the hallmark of hemiparetic CP, children may also experience cognitive and impairments plus epilepsy. In

our presumed perinatal stroke cohort, hemiplegia was also associated with hearing loss and aphasia in two children, respectively. Seventy-two percent (63/87) of all perinatal strokes occurred within the first week of life, demonstrating the importance of this age.

Prechtl's General Movement Assessment (GMA) was underreported in our study; in only 7% (6/87) of all perinatal strokes. Although GMA has a 95% sensitivity to predict CP from a score of "absent fidgety", this confirmation was lacking in this study because further follow-up data were not available.(26) The under utilitilisation of the GMA may be due to its lower predictability of hemiplegia, because not all assessors identify hemi-fidgety as absent fidgety, and not all babies with hemiplegia have absent fidgety. Therefore, a combination of tests is recommended to identify hemiplegic CP.(14, 27)

The strength of this study is that it is the first prospective study of perinatal stroke in Australia and lays the groundwork for future large global studies. A separate outcome analysis of two types of perinatal stroke, namely acute symptomatic and presumed perinatal stroke, identified the need for early detection of perinatal stroke.

A limitation of this study was the lack of ability to review original MRI images of the brain. The diagnosis of stroke was based on MRI reports and the reporting physicians. However, in Australia, paediatric MRIs are performed in tertiary care hospitals, thus MRI reporting is done by senior radiologists which ensures accurate reporting. The second limitation was the lower number of reported cases from some states in Australia, partly due to the small population and ethical constraints of reporting within the jurisdictions.

CONCLUSION

The first week of a child's life is the most critical period in terms of lifelong disability, as 72% of perinatal strokes occurred during this period. This study highlighted the burden of perinatal stroke with an overall disability rate of 48% (42/87) and a disability rate of 72%

(13/18) with presumed perinatal stroke. This brings the universal screening programme for CP into focus. Prospective collaborative studies are needed to improve timely detection and ensure prompt early intervention therapy to reduce the severity of disability.

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REFERENCES:

1. Novak I, Morgan C, Fahey M, Finch-Edmondson M, Galea C, Hines A, et al. State of the Evidence Traffic Lights 2019: Systematic Review of Interventions for Preventing and Treating Children with Cerebral Palsy. Current Neurology & Neuroscience Reports. 2020;20(2):3.
2. Rajsic S, Gothe H, Borba HH, Sroczynski G, Vujicic J, Toell T, et al. Economic burden of stroke: a systematic review on post-stroke care. Eur J Health Econ. 2019;20(1):107-34.
3. Wu YW, Lindan CE, Henning LH, Yoshida CK, Fullerton HJ, Ferriero DM, et al. Neuroimaging Abnormalities in Infants With Congenital Hemiparesis. Pediatr Neurol. 2006;35(3):191-6.
4. Knowledgebase A. Perinatal period: Australian Institute of Health and Welfare 2015–2023; 2005 [Available from: https://meteor.aihw.gov.au/content/327314.
5. Dunbar M, Kirton A. Perinatal stroke: mechanisms, management, and outcomes of early cerebrovascular brain injury. Lancet Child Adolesc Health. 2018;2(9):666-76.
6. Cardona F, Linduska N, Thom K, Schmook M, Prayer D, Seidl R. Perinatal arterial ischemic infarction associated with placental pathology. Neuropediatrics Conference: 12th International Congress of the European Society of Magnetic Resonance in Neuropediatrics Vienna Austria Conference Publication:. 2013;44(SUPPL. 1).
7. Chiang KL, Cheng CY. Epidemiology, risk factors and characteristics of pediatric stroke: a nationwide population-based study. Qjm. 2018;111(7):445-54.
8. Kirton A, Armstrong-Wells J, Chang T, Deveber G, Rivkin MJ, Hernandez M, et al. Symptomatic neonatal arterial ischemic stroke: the International Pediatric Stroke Study. Pediatrics. 2011;128(6):e1402-10.
9. Srinivasan J, Miller SP, Phan TG, Mackay MT. Delayed recognition of initial stroke in children: need for increased awareness. Pediatrics. 2009;124(2):e227-34.

10. Srivastava R, Kirton A. Perinatal Stroke: A Practical Approach to Diagnosis and Management. Neoreviews. 2021;22(3):e163-e76.
11. Hori I, Tsuji T, Miyake M, Ueda K, Kataoka E, Suzuki M, et al. Delayed recognition of childhood arterial ischemic stroke. Pediatrics International. 2019;61(9):895-903.
12. Lee S, Mirsky DM, Beslow LA, Amlie-Lefond C, Danehy AR, Lehman L, et al. Pathways for Neuroimaging of Neonatal Stroke. Pediatr Neurol. 2017;69:37-48.
13. Mackay MT, Monagle P, Babl FE. Improving diagnosis of childhood arterial ischaemic stroke. Expert rev. 2017;17(12):1157-65.
14. Morgan C, Fahey M, Roy B, Novak I. Diagnosing cerebral palsy in full-term infants. Journal of Paediatrics & Child Health. 2018;54(10):1159-64.
15. Li C, Miao JK, Xu Y, Hua YY, Ma Q, Zhou LL, et al. Prenatal, perinatal and neonatal risk factors for perinatal arterial ischaemic stroke: a systematic review and meta-analysis. Eur J Neurol. 2017;24(8):1006-15.
16. deVeber GA, Kirton A, Booth FA, Yager JY, Wirrell EC, Wood E, et al. Epidemiology and Outcomes of Arterial Ischemic Stroke in Children: The Canadian Pediatric Ischemic Stroke Registry. Pediatr Neurol. 2017;69:58-70.
17. von Elm E, Altman DG, Egger M, Pocock SJ, Gotzsche PC, Vandenbroucke JP, et al. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. Bull World Health Organ. 2007;85(11):867-72.
18. Alliance CP. Facts about cerebral palsy: Cerebral Palsy Alliance; 2018 [Available from: https://cerebralpalsy.org.au/our-research/about-cerebral-palsy/what-is-cerebral-palsy/facts-about-cerebral-palsy/.
19. te Velde A, Morgan C, Novak I, Tantsis E, Badawi N. Early Diagnosis and Classification of Cerebral Palsy: An Historical Perspective and Barriers to an Early Diagnosis. Journal of Clinical Medicine. 2019;8(10):03.
20. Oleske DM, Cheng X, Jeong A, Arndt TJ. Pediatric Acute Ischemic Stroke by Age-Group: A Systematic Review and Meta-Analysis of Published Studies and Hospitalization Records. Neuroepidemiology. 2021;55(5):331-41.
21. Sorg AL, von Kries R, Klemme M, Gerstl L, Weinberger R, Beyerlein A, et al. Risk factors for perinatal arterial ischaemic stroke: a large case-control study. Developmental Medicine & Child Neurology. 2020;62(4):513-20.
22. Cole L, Dewey D, Letourneau N, Kaplan BJ, Chaput K, Gallagher C, et al. Clinical Characteristics, Risk Factors, and Outcomes Associated With Neonatal Hemorrhagic Stroke: A Population-Based Case-Control Study. JAMA Pediatrics. 2017;171(3):230-8.
23. Roy B, Arbuckle S, Walker K, Morgan C, Galea C, Badawi N, et al. The Role of the Placenta in Perinatal Stroke: A Systematic Review. J Child Neurol. 2020;35(11):773-83.
24. Elgendy MM, Puthuraya S, LoPiccolo C, Liu W, Aly H, Karnati S. Neonatal stroke: Clinical characteristics and neurodevelopmental outcomes. Pediatrics & Neonatology. 2022;63(1):41-7.
25. Ilves P, Laugesaar R, Loorits D, Kolk A, Tomberg T, Loo S, et al. Presumed Perinatal Stroke: Risk Factors, Clinical and Radiological Findings. J Child Neurol. 2016;31(5):621-8.
26. Novak I, Morgan C, Adde L, Blackman J, Boyd RN, Brunstrom-Hernandez J, et al. Early, Accurate Diagnosis and Early Intervention in Cerebral Palsy: Advances in Diagnosis and Treatment. JAMA Pediatrics. 2017;171(9):897-907.
27. Mackay MT, Slavova N, Pastore-Wapp M, Grunt S, Stojanovski B, Donath S, et al. Pediatric ASPECTS predicts outcomes following acute symptomatic neonatal arterial stroke. Neurology. 2020;94(12):e1259-e70.

Chapter 8: SUMMARY and CONCLUSION

8.1 Introduction

Perinatal stroke remains one of the most common causes of hemiplegic cerebral palsy. Even in developed countries, it is an important cause of neurodevelopmental delay and disability in infants. Yet, it remains under-researched with knowledge gaps in its aetiology, clinical presentation, and diagnosis, which precludes identifying and testing novel prevention and intervention strategies that might further lower the falling rate of cerebral palsy (CP).

This thesis makes an innovative and important contribution to the field of perinatal stroke. The studies have focused on perinatal stroke rate in Australia, identifying risk factors, recognising the clinical presentations, and reporting the outcomes.

8.2 Key Findings

The thesis set out to address the six questions outlined in Chapter 1. Six studies using different methodologies were designed, conducted, analysed, and discussed. Thirteen major findings answered the six questions put forward. Four findings elucidated risks of perinatal stroke in literature, and nine findings described perinatal stroke in the Australian population.

Key Findings Elucidating Risk Factors

1. Placental abnormality is an under-recognised cause of perinatal stroke in the published literature. A lack of uniformity exists in describing placental pathological abnormalities.
2. Placental examination is underutilised in neonatal practice.
3. Aetiology of perinatal stroke in preterm infants is unclear and is further

confounded when researchers aggregate preterm and term-born data.

4. Clinical presentation of perinatal stroke in preterm infants can be non-neurological symptoms and does not include seizures.

Key Findings in an Australian Cohort

1. This was the first population-based study to elucidate the birth prevalence of perinatal stroke in Australia, which was: 13.9 per 100,000 live births per annum (p.a.) (95% CI 11.2, 17.3); 9.6/100,000 p.a. (95% CI 7.4, 12.5) for acute perinatal stroke; 5.8/100,000 p.a. (95% CI 4.1, 8.1) for NAIS; 2.9/100,000 p.a. (95% CI 1.8, 4.7) for NHS, and 0.3/100,000 p.a. (95% CI 0.1, 1.3) for CSVT.
2. Perinatal stroke is associated with multiple risk factors, some of which are also present in healthy babies.
3. Most babies in the perinatal stroke cohort were singletons born to nulliparous mothers.
4. Primiparity ($p<0.01$), caesarean section ($p=0.04$), 5-minute Apgar score <7 ($p<0.01$), and resuscitation at birth ($p<0.01$) were significant risk factors for perinatal stroke, over and above the risks in healthy babies in the Australian Institute of Health and Welfare dataset.
5. Multivariate analysis in a case-control study found exposure to smoking during pregnancy (OR 1.48; 95% CI 1.09, 1.99), 1-minute Apgar score <7 (OR 1.54; 95% CI 1.15, 2.08), 10-minute Apgar score <7 (OR 1.26; 95% CI 1.02, 1.54), and hypoglycaemia (OR 1.49; 95% CI: 1.07, 2.06) as significant risk factors for perinatal stroke.
6. Most infants with perinatal stroke presented early. Seventy eight percent (68/87) presented in the neonatal period, 10% (9/87) between 1–3 months of age, 5% (4/87) between 3–6 months of age and 2% (2/87) each between 6–12 months and

18–24 months of age.

7. Many of the clinical presentations were non-neurological symptoms; 35% presented with respiratory symptoms (tachypnoea, apnoea, cyanosis, and respiratory distress) and 20% with lethargy or poor feeding.
8. Predictors of disability such as absent fidgety General Movements were present in 6% of babies with perinatal stroke.
9. Ninety nine percent survived the perinatal stroke; however, 40% had neurological deficits such as hemiplegic cerebral palsy.

8.3 Implications for Practice, Policy, and Research

Perinatal stroke remains a relatively less-researched entity compared to stroke in adults, which has established clinical express pathways facilitating rapid diagnosis and initiation of therapy to restore speech, comprehension, movement, and vision. Major gaps in the perinatal stroke literature include aetiology (multifactorial and present in healthy infants) and non-neurological clinical presentations (Figure 1), which impede the discovery of comprehensive preventive strategies, and delay diagnosis, worsening adverse neurological outcomes.

Based on the key findings from the studies of this thesis, practice recommendations are proposed for clinicians and healthcare services, policy recommendations are proposed for governments and funders, as well as recommendations for areas of future research.

Recommendations for Practice

1. Maternity hospitals should follow NSW Health guidelines for placental histopathological examination and establish local policy for placental storage duration for all newborn babies.(71, 72)

All placentas should have a macroscopic examination at birth by the

attending clinician (obstetricians, neonatologists, and paediatricians) or neonatal nurse or midwife. Since most perinatal strokes occur within the first 7 days of life, all placentas should be stored – refrigerated in individually labelled plastic bags or plastic containers for at least 48 to 72 hours, ideally for one week to match the diagnostic window. Placental histopathological examination should be performed in infants presenting with seizures or non-specific symptoms, which may yield information regarding the cause of perinatal stroke, such as a thrombus originating from the placenta or chorioamnionitis. This information will form the basis for prevention strategies such as prophylactic aspirin for mothers, placental ultrasound doppler studies, or close monitoring for infection in future pregnancies.

2. Brain MRI examination should be carried out for infants with suspected perinatal stroke.

   It is important to differentiate some of the risk factors for perinatal stroke such as emergency caesarean section, 5- or 10-minute Apgar score of <7, meconium-stained liquor and neonatal resuscitation as signs of perinatal stroke in progress. A combination of meticulous clinical history, clinical surveillance, and neuroimaging (preferably brain MRI with diffusion) allows rapid accurate diagnosis, identification of nature of brain injury to predict CP, and diagnostic-specific time-critical implementation of multi-disciplinary therapy services to avoid lost opportunities during the critical window for neuroplasticity.(73)

Recommendations for Policy

1. Establish an Australian Perinatal Stroke Register.

Clinical practice guideline and Australian state-based clinical consensus guideline exist for the management and rehabilitation of childhood stroke and are available at the state level.(74, 75) However, an Australia-wide Perinatal Stroke Registry with an integrated perinatal stroke improvement national audit program is warranted. Audits could be used to monitor and report the changing rates of perinatal stroke in Australia and aid comparison to global statistics, to gain further understanding of the causative mechanisms (Appendix A). A Register team could also be co-led by consumer stakeholders such as Little Stroke Warriors Australia, which is a group of families and survivors of paediatric stroke providing support to families through the journey of stroke recovery. Together consumers and knowledge translation experts could collaborate with those generating evidence and research end-users to improve child outcomes. Knowledge brokers could also establish knowledge translation training programs to create placental stroke awareness programs for consumers, hospitals and clinicians.

A centralised global registry with pooled data from an Australia-wide registry and the International Pediatric Stroke Study (IPSS) would increase sample sizes for advancing discoveries in aetiology, clinical presentations and outcomes of perinatal stroke.(76) The key findings (8.2) from studies 4, 5 and 6 could be used within these registries to create new knowledge.

Recommendations for Research

1. Collaborative prospective studies are needed to further investigate the risk factors for perinatal stroke.

   The published literature did not previously emphasise the aetiological role of the placenta (Study 1) and the prospective perinatal stroke research (studies 4

and 5) substantially under-collected and under-reported perinatal strokes with the placental pathology report (only n=4/87). An Australian Perinatal Stroke Registry should promote routine placental histopathological examination via evidence-based knowledge translation strategies. This would advance the discovery of the role of the placenta in the pathogenesis of perinatal stroke, given the known association between placental thromboinflammatory changes and perinatal stroke (Study 1). Based on Studies 1 and 2, power calculations to detect aetiological correlation of placental abnormalities in perinatal stroke with 80% power and 5% significance level, a case control study with a 1:3 ratio of cases to matched controls would require n = 29 perinatal stroke cases and n = 87 controls (Appendix I).

It is also important to identify perinatal stroke-related genetic/haematological abnormalities, especially after known causes of perinatal stroke are excluded or in the presence of a family history of perinatal or childhood stroke. Research proposals to identify genetic and/or haematological abnormalities would require a wide range of possible sample sizes. Therefore, a multicentre international collaborative study is required to reach the target sample size (Appendix I).

This research found that preterm infants are at risk of intraventricular haemorrhage, but the pathogenesis of perinatal stroke was unclear (Study 3). Furthermore, the clinical presentation of perinatal stroke in preterm infants may be masked by incomplete maturation of other human body systems presenting with respiratory distress, apnoea, cyanosis, lethargy, poor sucking reflex, or sepsis. A case-control study is needed to differentiate the precise symptoms of perinatal stroke to enable timely diagnosis and timely initiation of multidisciplinary intervention.

Finally, inconsistent classification of perinatal stroke in the literature was one of the major hurdles in aggregating and interpreting data. Therefore, Dunbar's classification on perinatal stroke subtypes was used for the prospective perinatal stroke research (Studies 4, 5 and 6) to maintain a consistent classification based on the timing of presentation of perinatal stroke.(13) Future studies should also adopt Dunbar's standard perinatal stroke classification so that more definitive conclusions can be drawn.

Recommendations for Diagnosis

1. Best practice clinical guidelines for the management of perinatal stroke are required.

   Variable clinical presentation of perinatal stroke, including non-neurological respiratory presentations (35% in Study 6), is one of the major reasons for the delay in diagnosis and late intervention (Figure 1). However, due to scanner time resource constraints and anaesthesia safety issues in infants >6 weeks of age, MRI may be performed at inappropriate times (i.e. well after the event of perinatal stroke) or not performed at all. Diagnosis of perinatal stroke and disability after stroke is a multilayered clinical decision. Diagnosticians need: a high index of suspicion when a cluster of risk factors present; an open mind about varied clinical presentations; adherence to guidelines and policies outlining comprehensive investigations; consideration of the safety and feasibility of brain MRI; and input from allied health professionals (such as physiotherapist, occupational therapist, and speech pathologist) about emerging motor and cognitive impairments. Together these practices will allow for earlier and more accurate diagnosis and initiation of early interventions to optimise infant outcomes.(77) It is important that an early

diagnosis (i.e. 48-72 hours) of perinatal stroke be followed by prompt referral for early intervention rehabilitation services.

Continuing education and awareness programs about perinatal stroke in preterm and term infants are essential because clinicians play a critical role in realising early diagnosis and intervention. Placental examination, genetic evaluation when indicated, and MRI of the brain should become the routine standard of care.

Recommendation for Research Landscape with No Funding Barriers

1. Comprehensive early management of perinatal stroke may lower the severity of disability after stroke.
    - A comprehensive work-up for perinatal stroke entails investigating the list of risk factors including placental histopathology, cardiac echocardiography, and prothrombin markers (Appendix A), brain MRI, EEG, physiotherapy assessment and ophthalmic examination for all infants prior to discharge from the hospital.
    - A combination of neurological assessments such as Prechtl's qualitative assessment of General Movements (GMA), Hammersmith Infant Neurological Examination (HINE), and Hand Assessment in Infants (HAI) are known to be the best early predictors for cerebral palsy as an outcome after stroke.
    - Follow-up neuroimaging if indicated to monitor the white and grey matter, and basal ganglia growth and development, in case of comorbidities such as impairments to vision, language, and learning.
    - Referral to social worker is recommended to assist with coordination of outpatient follow-up and financial supports, such as the carer's allowance

and National Disability Insurance Scheme (NDIS), to optimise the child's health and wellbeing, and likelihood of achieving long-term independent living, educational outcomes, and employment.

- Multicentre fully funded audit for surveillance of perinatal stroke that contributes to Australian Perinatal Stroke Registry, which will enable answering the prevention and early diagnosis questions.

8.4 Limitations and Strengths

The specific limitations and strengths of each study have been discussed in the publications included in chapters 2–7. Here, the limitations and strengths of the thesis will be discussed.

Limitations

A major limitation of the research program was the under-reporting of perinatal stroke cases in some states, affecting the total sample size and consequently the ability to calculate the incidence of perinatal stroke in the total Australian population. Under-reporting was due to different ethical requirements in different Australian jurisdictions and the burden of notification by time-poor doctors.

The second limitation was the inability to acquire actual MRI images for central analysis from a skilled single rater; instead, analyses were conducted from MRI report data. The decision to not acquire the MRI images was made a priori to ease the reporting burden of clinicians, with volume of cases reported prioritised over the data source. In this research program, the diagnosis of perinatal stroke was based on the physician's report and the MRI brain report, which may have introduced clinical interpretation differences.

Strengths

This research program prospectively generated definitive information on the risk factors for perinatal stroke in an Australian cohort using a population-based prospective sample, compared to healthy infant population samples. Another, major strength of this research program is that it was the first to report the prevalence of perinatal stroke in Australia.

The questionnaires included in the data collection (appendices A and B) could serve as a template for a future perinatal stroke registry in Australia and even for global registry data linkage.

8.5 Significance of Research and Contribution to the Field

The lasting contribution of this research will be threefold.

First, perinatal stroke prevention strategies can now be identified and tested, because risk factors have been elucidated and confirmed. In addition, this study provided evidence-based recommendations for implementing placental histopathological examination into the perinatal stroke work-up, since the placenta can have a causative role via the thromboinflammatory process.

Furthermore, ensuring an early diagnosis via a high index of suspicion in response to a wide range of clinical presentations could initiate early intervention that might lower the severity of disability after stroke.

Some of the tools in this research, such as the data collection questionnaires (Appendices A and B), could serve as templates for identifying pathogenesis and signs and symptoms of perinatal stroke. These data could also be the foundation for an Australian neonatal stroke registry, which is not currently available but is needed.

Second, the Australian perinatal stroke prevalence calculated in this study will serve as a benchmark for global comparisons to gauge the trends in global incidence.

A third outcome is the substantial contribution to new knowledge about the aetiological pathways to perinatal stroke that will enable further identification of significant risk factors, including genetic causes, and identification of novel prevention treatments.

8.6 Conclusion

This thesis asked the who? what? and how? of perinatal stroke. New knowledge was generated to answer these questions through: a series of systematic reviews; a chart audit; and a prospective longitudinal population-based study with both population and case-controls.

Who gets perinatal stroke? Infants born preterm and term experience perinatal stroke, but the strokes are different, not just in the timing but also in their type and clinical presentation. More is known about strokes in term-born infants. There were insufficient data to elucidate the exact aetiology and mechanisms of perinatal stroke in preterm infants. Placental thromboinflammatory abnormalities were identified as a risk factor for perinatal stroke, but the results were inconclusive due to the small sample sizes in published literature. Placental examinations were also under-collected and under-reported in real-world neonatal practice, compounding the sample size problem, further limiting conclusive findings. Recommendations for population-based prospective placental collections were made to overcome sample size barriers.

What are the risks for perinatal stroke in the Australian population? For the first time, this population-based study reported a prevalence of perinatal stroke in Australian children of 13.9/100,000 live births per year. In addition, for the first time, risks for

perinatal stroke were prised apart from risks in healthy babies, paving the way for an earlier diagnosis. The following independent risk factors for perinatal stroke were identified: exposure to smoking during pregnancy; caesarean delivery; 5- and 10-minute Apgar score <7; neonatal infection, and hypoglycaemia. Resuscitation at birth and abnormal cord blood gas were identified as additional risk factors. Recommendations for diagnostic practices and global registries were made to improve the care of infants with stroke and to promote prevention discoveries.

How does perinatal stroke present? Contrary to conventional wisdom, not all perinatal strokes present with seizures, which may explain why some babies are diagnosed late. As many as 35% of infants presented with non-neurological symptoms such as tachypnoea, apnoea, and cyanosis. As much as 39% had lifelong neurological impairment after stroke, including cerebral palsy in 35%. Recommendations for early management were made to improve the infants' quality of life and outcomes.

In adults with stroke, there are well-established rapid management pathways. The same standard of care is possible for infants with a higher index of suspicion for perinatal stroke, adoption of earlier diagnosis practices, collection of the placenta, notification to registries, uptake of early intervention practices and the testing of novel preventative agents.

References

1. Kirton A. Advancing non-invasive neuromodulation clinical trials in children: Lessons from perinatal stroke. European Journal of Paediatric Neurology. 2017;21(1):75-103.
2. Australian Institute of Health and Welfare. Perinatal period identifying and definitional attributes. 2005.
3. Ferriero DM, Fullerton HJ, Bernard TJ, Billinghurst L, Daniels SR, DeBaun MR, et al. Management of Stroke in Neonates and Children: A Scientific Statement From the American Heart Association/American Stroke Association. Stroke. 2019;50(3):e51-e96.
4. Raju TN, Nelson KB, Ferriero D, Lynch JK, Participants N-NPSW. Ischemic perinatal stroke: summary of a workshop sponsored by the National Institute of Child Health and Human Development and the National Institute of Neurological Disorders and Stroke. Pediatrics. 2007;120(3):609-16.
5. Lehman LL, Rivkin MJ. Perinatal Arterial Ischemic Stroke: Presentation, Risk Factors, Evaluation, and Outcome. Pediatr Neurol. 2014;51(6):760-8.
6. Kirton A, Deveber G, Pontigon AM, Macgregor D, Shroff M. Presumed perinatal ischemic stroke: vascular classification predicts outcomes. Annals of Neurology. 2008;63(4):436-43.
7. Lai PT, Reilly JS. Language and affective facial expression in children with perinatal stroke. Brain Lang. 2015;147:85-95.
8. Lynch JK, Nelson KB. Epidemiology of perinatal stroke. Current Opinion in Pediatrics. 2001;13(6):499-505.
9. Nelson KB, Lynch JK. Stroke in newborn infants. Lancet neurol. 2004;3(3):150-8.
10. Nelson KB. Perinatal ischemic stroke. Stroke. 2007;38(2 Suppl):742-5.
11. Oleske DM, Cheng X, Jeong A, Arndt TJ. Pediatric Acute Ischemic Stroke by Age-Group: A Systematic Review and Meta-Analysis of Published Studies and Hospitalization Records. Neuroepidemiology. 2021;55(5):331-41.
12. Rivkin MJ, Bernard TJ, Dowling MM, Amlie-Lefond C. Guidelines for Urgent Management of Stroke in Children. [Review]. 2016;1:8-17.
13. Dunbar M, Kirton A. Perinatal stroke: mechanisms, management, and outcomes of early cerebrovascular brain injury. Lancet Child Adolesc Health. 2018;2(9):666-76.
14. Katsafanas C, Bushnell C. Pregnancy and stroke risk in women. Neurobiol Dis. 2022;169:105735.
15. Martinez-Biarge M, Cheong JL, Diez-Sebastian J, Mercuri E, Dubowitz LM, Cowan FM. Risk Factors for Neonatal Arterial Ischemic Stroke: The Importance of the Intrapartum Period. Journal of Pediatrics. 2016;173:62-8.e1.
16. Li C, Miao JK, Xu Y, Hua YY, Ma Q, Zhou LL, et al. Prenatal, perinatal and neonatal risk factors for perinatal arterial ischaemic stroke: a systematic review and meta-analysis. Eur J Neurol. 2017;24(8):1006-15.
17. Lee J, Croen LA, Backstrand KH, Yoshida CK, Henning LH, Lindan C, et al. Maternal and infant characteristics associated with perinatal arterial stroke in the infant. Jama. 2005;293(6):723-9.
18. Harteman JC, Groenendaal F, Kwee A, Welsing PM, Benders MJ, de Vries LS. Risk factors for perinatal arterial ischaemic stroke in full-term infants: a case-control study. Archives of Disease in Childhood Fetal & Neonatal Edition. 2012;97(6):F411-6.
19. Hunt RW, Inder TE. Perinatal and neonatal ischaemic stroke: a review. Thrombosis Research. 2006;118(1):39-48.
20. Jankovic M, Petrovic B, Novakovic I, Brankovic S, Radosavljevic N, Nikolic D. The Genetic Basis of Strokes in Pediatric Populations and Insight into New Therapeutic Options. International Journal of Molecular Sciences. 2022;23(3):29.

21. Redline RW, Ravishankar S. Fetal vascular malperfusion, an update. APMIS. 2018;126(7):561-9.
22. Bernson-Leung M, Boyd T, Danehy A, Meserve E, Rivkin M. Placental pathology in neonatal stroke: A retrospective case-control study. Neurology Conference: 68th American Academy of Neurology Annual Meeting, AAN. 2016;86(16 SUPPL. 1).
23. Kirton A, Armstrong-Wells J, Chang T, Deveber G, Rivkin MJ, Hernandez M, et al. Symptomatic neonatal arterial ischemic stroke: the International Pediatric Stroke Study. Pediatrics. 2011;128(6):e1402-10.
24. Elgendy MM, Puthuraya S, LoPiccolo C, Liu W, Aly H, Karnati S. Neonatal stroke: Clinical characteristics and neurodevelopmental outcomes. Pediatrics & Neonatology. 2021;28:28.
25. Mallick AA, Ganesan V, Kirkham FJ, Fallon P, Hedderly T, McShane T, et al. Childhood arterial ischaemic stroke incidence, presenting features, and risk factors: a prospective population-based study. Lancet neurol. 2014;13(1):35-43.
26. Van Der Aa NE, Benders MJNL, Nikkels PG, Groenendaal F, De Vries LS. Cortical Sparing in Preterm Ischemic Arterial Stroke. Stroke. 2016;47(3):869-71.
27. Glass HC, Ferriero DM. Neonatal stroke. Fetal and Neonatal Brain Injury, Fourth Edition2009. p. 296-303.
28. Hori I, Tsuji T, Miyake M, Ueda K, Kataoka E, Suzuki M, et al. Delayed recognition of childhood arterial ischemic stroke. Pediatrics International. 2019;61(9):895-903.
29. Fink M, Slavova N, Grunt S, Perret E, Regenyi M, Steinlin M, et al. Posterior Arterial Ischemic Stroke in Childhood. Stroke. 2019;50(9):2329-35.
30. Lin J, Li X, Wu G, Chen X, Weng Y, Wang H, et al. White Matter High Signals Interfere with Noncontrast Computed Tomography in the Early Identification of Cerebral Infarction. Cerebrovasc Dis. 2020;49(2):135-43.
31. Biswas A, Mankad K, Shroff M, Hanagandi P, Krishnan P. Neuroimaging Perspectives of Perinatal Arterial Ischemic Stroke. Pediatr Neurol. 2020;113:56-65.
32. Srivastava R, Rajapakse T, Carlson HL, Keess J, Wei XC, Kirton A. Diffusion Imaging of Cerebral Diaschisis in Neonatal Arterial Ischemic Stroke. Pediatr Neurol. 2019;100:49-54.
33. Olive G, Agut T, Echeverria-Palacio CM, Arca G, Garcia-Alix A. Usefulness of Cranial Ultrasound for Detecting Neonatal Middle Cerebral Artery Stroke. Ultrasound Med Biol. 2019;45(3):885-90.
34. Rajsic S, Gothe H, Borba HH, Sroczynski G, Vujicic J, Toell T, et al. Economic burden of stroke: a systematic review on post-stroke care. Eur J Health Econ. 2019;20(1):107-34.
35. Munoz D, Hidalgo MJ, Balut F, Troncoso M, Lara S, Barrios A, et al. Risk Factors for Perinatal Arterial Ischemic Stroke: A Case-Control Study. Cell med. 2018;10:2155179018785341.
36. Loo S, Ilves P, Mannamaa M, Laugesaar R, Loorits D, Tomberg T, et al. Long-term neurodevelopmental outcome after perinatal arterial ischemic stroke and periventricular venous infarction. European Journal of Paediatric Neurology. 2018;22(6):1006-15.
37. Laugesaar R, Vaher U, Loo S, Kolk A, Mannamaa M, Talvik I, et al. Epilepsy after perinatal stroke with different vascular subtypes. Epilepsia Open. 2018;3(2):193-202.
38. Redline RW. Classification of placental lesions. American Journal of Obstetrics & Gynecology. 2015;213(4 Suppl):S21-8.
39. Chabrier S, Husson B, Dinomais M, Landrieu P, Nguyen The Tich S. New insights (and new interrogations) in perinatal arterial ischemic stroke. Thrombosis Research. 2011;127(1):13-22.
40. Ilves P, Laugesaar R, Loorits D, Kolk A, Tomberg T, Loo S, et al. Presumed Perinatal Stroke: Risk Factors, Clinical and Radiological Findings. J Child Neurol. 2016;31(5):621-8.
41. Ramaswamy V, Miller SP, Barkovich AJ, Partridge JC, Ferriero DM. Perinatal stroke in term infants with neonatal encephalopathy. 2004;1(11):2088-91.

42. Benders MJNL, Groenendaal F, Uiterwaal CSPM, de Vries LS. Perinatal Arterial Stroke in the Preterm Infant. Seminars in Perinatology. 2008;32(5):344-9.
43. Fullerton HJ, Wu YW, Sidney S, Johnston SC. Risk of recurrent childhood arterial ischemic stroke in a population-based cohort: the importance of cerebrovascular imaging. Pediatrics. 2007;119(3):495-501.
44. Laugesaar R, Kolk A, Tomberg T, Metsvaht T, Lintrop M, Varendi H, et al. Acutely and retrospectively diagnosed perinatal stroke: a population-based study. Stroke. 2007;38(8):2234-40.
45. Heljic S, Mornjakovic A, Merhemic Z, Maksic H, Hadzipasic E. [Stroke in newborn infants]. Med Arh. 2007;61(1):62-4.
46. Barnes C, Newall F, Furmedge J, Mackay M, Monagle P. Arterial ischaemic stroke in children. Journal of Paediatrics & Child Health. 2004;40(7):384-7.
47. Barnes C, Newall F, Furmedge J, Mackay M, Monagle P. Cerebral sinus venous thrombosis in children. Journal of Paediatrics & Child Health. 2004;40(1-2):53-5.
48. Gerstl L, Bonfert MV, Heinen F, Olivieri M, Schroeder AS, Tacke M, et al. Childhood Arterial Ischaemic Stroke: Clinical Presentation, Risk Factors and Management. Hamostaseologie. 2020;40(2):165-73.
49. Benders MJ, Groenendaal F, Uiterwaal CS, Nikkels PG, Bruinse HW, Nievelstein RA, et al. Maternal and infant characteristics associated with perinatal arterial stroke in the preterm infant. Stroke. 2007;38(6):1759-65.
50. Teutsch SM, Nunez CA, Morris A, Eslick GD, Khandaker G, Berkhout A, et al. Australian Paediatric Surveillance Unit (APSU) Annual Surveillance Report 2020. Commun Dis Intell (2018). 2021;45:28.
51. von Elm E, Altman DG, Egger M, Pocock SJ, Gotzsche PC, Vandenbroucke JP, et al. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. Bull World Health Organ. 2007;85(11):867-72.
52. Chabrier S, Saliba E, Nguyen The Tich S, Charollais A, Varlet MN, Tardy B, et al. Obstetrical and neonatal characteristics vary with birthweight in a cohort of 100 term newborns with symptomatic arterial ischemic stroke. European Journal of Paediatric Neurology. 2010;14(3):206-13.
53. Darmency-Stamboul V, Chantegret C, Ferdynus C, Mejean N, Durand C, Sagot P, et al. Antenatal factors associated with perinatal arterial ischemic stroke. Stroke. 2012;43(9):2307-12.
54. Australian Bureau of Statistics. Births, Australia 2019 [Available from: https://www.abs.gov.au/statistics/people/population/births-australia/2018.
55. Australian Institute of Health and Welfare (AIHW). Australia's mothers and babes. Available from: https://www.aihw.gov.au/reports/mothers-babies/australias-mothers-babies/contents/about (Accessed 28 Junel 2022)
56. Surmava AM, Maclagan LC, Khan F, Kapral MK, Hall RE, deVeber G. Incidence and Current Treatment Gaps in Pediatric Stroke and TIA: An Ontario-Wide Population-Based Study. Neuroepidemiology. 2019;52(3-4):119-27.
57. Agrawal N, Johnston SC, Wu YW, Sidney S, Fullerton HJ. Imaging data reveal a higher pediatric stroke incidence than prior US estimates. Stroke. 2009;40(11):3415-21.
58. Dunbar M, Mineyko A, Hill M, Hodge J, Floer A, Kirton A. Population Based Birth Prevalence of Disease-Specific Perinatal Stroke. Pediatrics. 2020;146(5):11.
59. Pohlman KA, Carroll L, Tsuyuki RT, Hartling L, Vohra S. Comparison of active versus passive surveillance adverse event reporting in a paediatric ambulatory chiropractic care setting: a cluster randomised controlled trial. BMJ open qual. 2020;9(4):11.
60. deVeber GA, Kirton A, Booth FA, Yager JY, Wirrell EC, Wood E, et al. Epidemiology and Outcomes of Arterial Ischemic Stroke in Children: The Canadian Pediatric Ischemic Stroke Registry. Pediatr Neurol. 2017;69:58-70.

61. Armstrong-Wells J, Johnston SC, Wu YW, Sidney S, Fullerton HJ. Prevalence and predictors of perinatal hemorrhagic stroke: results from the kaiser pediatric stroke study. Pediatrics. 2009;123(3):823-8.
62. Cole L, Dewey D, Letourneau N, Kaplan BJ, Chaput K, Gallagher C, et al. Clinical Characteristics, Risk Factors, and Outcomes Associated With Neonatal Hemorrhagic Stroke: A Population-Based Case-Control Study. JAMA Pediatrics. 2017;171(3):230-8.
63. Sporns PB, Fullerton HJ, Lee S, Kim H, Lo WD, Mackay MT, et al. Childhood stroke. Nat Rev Dis Prim. 2022;8(1):12.
64. Vitagliano M, Dunbar M, Dyck Holzinger S, Letourneau N, Dewey D, Oskoui M, et al. Perinatal arterial ischemic stroke and periventricular venous infarction in infants with unilateral cerebral palsy. Developmental Medicine & Child Neurology. 2022;64(1):56-62.
65. Armon C. Which drugs increase the risk of stroke during pregnancy? 2019 [Available from: https://www.medscape.com/answers/1149405-164921/which-drugs-increase-the-risk-of-stroke-during-pregnancy.
66. Mullen M. Stroke associated with drug abuse: Medlink Neurology; 2021 [Available from: https://www.medlink.com/articles/stroke-associated-with-drug-abuse.
67. Golomb MR, Fullerton HJ, Nowak-Gottl U, Deveber G, International Pediatric Stroke Study G. Male predominance in childhood ischemic stroke: findings from the international pediatric stroke study. Stroke. 2009;40(1):52-7.
68. Normann S, de Veber G, Fobker M, Langer C, Kenet G, Bernard TJ, et al. Role of endogenous testosterone concentration in pediatric stroke. 2009;1(6):754-8.
69. Rodrigues DO, Ribeiro LC, Sudario LC, Teixeira MT, Martins ML, Pittella AM, et al. Genetic determinants and stroke in children with sickle cell disease. J Pediatr (Rio J). 2016;92(6):602-8.
70. Sorg AL, von Kries R, Klemme M, Gerstl L, Weinberger R, Beyerlein A, et al. Risk factors for perinatal arterial ischaemic stroke: a large case-control study. Developmental Medicine & Child Neurology. 2020;62(4):513-20.
71. Secretary NSW Health. Maternity - Indications for Placental Histological Examination 2014 [Available from: https://www1.health.nsw.gov.au/pds/ActivePDSDocuments/GL2014_006.pdf.
72. Yee Khong VF, Jane Dahlstrom, David Ellwood, Lisa Hui, Dell Horey, Lynn Sinclair, Jane Zucculo, Fran Boyle. Clinical Practice Guideline for Care Around Stillbirth and Neonatal Death 2019 [Available from: https://sanda.psanz.com.au/assets/Uploads/Section-4-PerintalPostMortemExamination-V3.2-121219.pdf.
73. Morgan C, Fahey M, Roy B, Novak I. Diagnosing cerebral palsy in full-term infants. Journal of Paediatrics & Child Health. 2018;54(10):1159-64.
74. Greenham M, Knight S, RoddaPh DJ, Scheinberg A, Anderson V, Fahey MC, et al. Australian clinical consensus guideline for the subacute rehabilitation of childhood stroke. Int j. 2021;16(3):311-20.
75. The Royal Children's Hospital Melbourne. Stroke 2019 [Available from: https://www.rch.org.au/clinicalguide/guideline_index/Stroke.
76. Committee IP. International Pediatric Stroke Study (IPSS) 2023 [Available from: https://internationalpediatricstroke.org/ipss-research/.
77. Morgan C, Fetters L, Adde L, Badawi N, Bancale A, Boyd RN, et al. Early Intervention for Children Aged 0 to 2 Years With or at High Risk of Cerebral Palsy: International Clinical Practice Guideline Based on Systematic Reviews. JAMA Pediatrics. 2021;175(8):846-58.

Appendices

Appendix A: APSU_Stroke_Questionnaire_8-05-18

Stroke in Australian Children Under 2 Years of Age Australian Paediatric Surveillance Unit Please contact the APSU (02) 9845 3005; SCHN-APSU@health.nsw.gov.au If you have any questions about this form	*APSU Office Use Only*	
	Study ID #:	
	Month/Year Report:	
Instructions: Please answer each question by ticking the appropriate box or writing your response in the space provided. DK=Don't Know; NA = Not Applicable	*Version 3: 08.05.2018*	

REPORTING CLINICIAN'S DETAILS:

1. APSU Dr Code/Name: ☐☐☐☐/_____ **2.** Date questionnaire completed: __ __/__ __/_____

PATIENT DETAILS (THIS CHILD)

3. First 2 letters of first name: ___ ___ **4.** First 2 letters of surname: ___ ___ **5.** Postcode of family: _____

6. Racial background *(select all that apply)*: ☐ Aboriginal ☐ Caucasian ☐ Pacific Islander ☐ Torres Strait Islander ☐ African ☐ Asian ☐ DK ☐ Other *(specify):* _____ **7.** Date of Birth: __ __ / __ __/ __ ____ (dd/mm/yyyy)

8. Sex: ☐ Male ☐ Female ☐ Indeterminate **9.** Did you make the diagnosis? ☐ Yes (***please go to Q10***) ☐ No – if this patient is primarily cared for by another physician who you believe could provide additional details, please write their name below and return this form to the APSU. If no other reports are received for this child we will contact you for futher information.

Physcian's Name: _____ Clinic/Hospital: _____

DIAGNOSIS OF STROKE THIS CHILD

10. Date of Diagnosis of Stroke: __ /__ /____ (dd/mm/yyyy) **11.** Type of Stroke event (a patient can have multiple types of stroke event): (Select all that apply) ☐ Arterial Ischemic Stroke (AIS) ☐ Periventricular Venous Infarction (PVI) ☐ Cerebral Sinovenous Thrombosis (CSVT) ☐ Haemorrhagic Stroke (NHS) ☐ Other; Specify:_____

12. Other diagnoses? ☐ Generalised Sepsis ☐ Meningitis ☐ Other infection; Specify:_____

☐ Congenital heart disease (specify CHD type):_____

(specify interventions): _____

☐ Other congenital anomalies, specify_____

☐ Other Diagnoses (specify)_____

HISTORY DURING PREVIOUS PREGNANCIES

13. (a) Miscarriage: ☐ Yes ☐ No ☐ DK **If yes,** Specify number_____

(b) Stillbirth: ☐ Yes ☐ No ☐ DK **If yes,** Specify number_____ Specify gestation for each _____

(c) Neonatal death: ☐ Yes ☐ No ☐ DK **If yes,** Specify number_____ Specify gestation for each _____

PREGNANCY HISTORY (THIS PREGNANCY)

14. Maternal age *(completed yrs)*:_____ (yrs) **15.** Consanguinity: ☐ Yes ☐ No ☐ DK **16.** Parity: Gravida____ Parity__ ☐ DK

17. Was conception: ☐ Natural ☐ IVF ☐ Other (*specify):*_____ ☐ DK

18. (a) Complications during this pregnancy: ☐ Yes ☐ No ☐ DK **If Yes,** ☐ pre-eclampsia ☐ IUGR ☐ Placental blood flow abnormality ☐ Other *(specify)*: _____

(b) Were there any abnormal antenatal US reports: ☐ Yes ☐ No ☐ DK **If yes,** Specify: _____

(c) Was there evidence of meconium stained liqor: ☐ Yes ☐ No ☐ DK

(d) Was there evidence of Chorioamnionitis?☐ Yes ☐ No ☐ DK **If yes,** ☐ Clinically suspected ☐ Pathologically proven ☐ Both ☐ DK

Please attach de-identified placental pathology report, if available.

(e) Did the mother have any positive microbial cultures during pregnancy? ☐ Yes ☐ No ☐ DK,

***If yes*,** GBS in High Vaginal Swab: ☐ Yes ☐ No ☐ DK

☐ Urine culture (Specify pathogen(s): _____;

☐Other +ve cultures, Specify pathogen(s): _____

(f) During prenancy did the mother take: ☐ warfarin ☐ phenytoin ☐ barbiturates ☐ Other medications (specify): _____

☐ No ☐ DK

(g) During prenancy did the mother: Smoke ☐ Yes ☐ No ☐ DK; Drink alcohol ☐ Yes ☐ No ☐ DK; Take Illicit drugs ☐ Yes ☐ No ☐ DK **If yes,** specify all: _____

19. Is there a family history of childhood stroke: (i) In parents ☐Yes ☐No ☐DK (ii) siblings ☐Yes ☐No ☐DK (iii) first degree relatives of parents ☐Yes ☐No **If yes,** specify first degree relative: _____

BIRTH INFORMATION AND INTERVENTIONS (THIS CHILD)

20. Gestational age:_____ (completed wks) ☐ DK

21. i)Birth Weight: _____ (g) ☐ DK ii)Birth Length: _____ (cm) ☐ DK iii)Birth Head Circumference: _____ (cm) ☐ DK

22. Vitamin K given: ☐ Yes ☐ No ☐ DK **If yes,** ☐ Oral ☐ IM, **If Oral** were all 3 doses given? ☐ Yes ☐ No ☐ DK

23. Mode of delivery: ☐ Normal Vaginal Delivery ☐ Vacuum ☐ Forceps ☐ Elective Caeseran Section ☐ Emergency Caeseran Section ☐ Vaginal Breech Delivery ☐ DK

24. Was this a difficult delivery: ☐ Yes ☐ No ☐ DK **If yes,** ☐ Shoulder dystocia ☐ Multiple vacuum attempts ☐ Failed vacuum ☐ Other Please specify:_____

25. Plurality: ☐ Singleton ☐ Twin 1 ☐ Twin 2 ☐ Triplet 1 ☐ Triplet 2 ☐ Triplet 3 ☐ Other ☐ DK If twin or triplet, type (select one): ☐ MCMA ☐ MCDA ☐ DCDA ☐ MCTA ☐ TCTA ☐ DK ☐ Other: _____

26. Death of Co-twin / Co-triplet: ☐ Yes ☐ No ☐ DK **If yes,** ☐ Death before birth ☐ after birth Cause of death:_____ ☐DK

27. Apgar Scores: 1min ___ ☐ DK; 5min ___ ☐ DK; 10min ___ ☐ DK

28. Cord blood gas**?** ☐ Yes ☐ No ☐ DK **If yes,** ☐ Arterial Cord / ☐ Venous cord: pH _____ /_____ pCO2 ____ /_____ Base Excess _____ /_____ Lactate ___ /_____

29. Resuscitation required at birth? ☐ Yes ☐ No ☐ DK

If yes, ☐ Suction ☐ Oxygen ☐ IPPV ☐ CPAP ☐ Intubation ☐ Chest compression ☐ Adrenaline ☐ Fluid bolus

30. Did the child need vascular catheterisation? ☐ Yes ☐ No ☐ DK **If yes,** ☐ Umbilical artery ☐ Umbilical vein ☐ Femoral artery ☐ Femoral vein ☐ Cardiac catheterisation ☐ Other specify:_____

CLINICAL PRESENTATION OF STROKE (THIS CHILD)

31. Date of clinical onset of symptoms: _ /_ /_____ (dd/mm/yyyy)

32. Clinical Presentation *(please tick all that apply)*: ☐ Poor feeding ☐ Tachypnoea ☐ Apnoea ☐ Cyanosis ☐ Hypoglycemia ☐ Abnormal tone and reflexes ☐ Lethargy ☐ Abnormal level of consciousness ☐ DK

☐ Seizures: ☐ Yes ☐ No ☐ DK **If yes,** Type of Seizures - ☐ Focal right ☐ Focal left ☐ Multifocal ☐ Generalised ☐ Subtle

☐ Hemiparesis: ☐ Yes ☐ No ☐ DK **if yes,** Specify ☐ Right ☐ Left ☐ Bilateral ☐ None ☐ DK

☐ Other; specify: _____

INVESTIGATIONS FOR STROKE (THIS CHILD)

33. Brain Imaging: **Please attach de-identified Brain imaging (MRI, CT, Head US) and EEG reports, if available.**

(i) Was MRI done? ☐ Yes ☐ No ☐ DK **If yes,** Date __ /__ / _____ (dd/mm/yyyy)

(ii) Was CT done? ☐ Yes ☐ No ☐ DK **If yes,** Date __ /__ /_____ (dd/mm/yyyy)

(iii) Was Ultrasound done? ☐ Yes ☐ No ☐ DK **If yes,** Date __ /__ /_____ (dd/mm/yyyy)

34. (i) EEG report: ☐ Yes ☐ No ☐ DK **If yes**: Date __ /__ /_____ (dd/mm/yyyy)

(ii) Full blood count: ☐ Yes ☐ No ☐ DK **If yes,** any abnormalities? _____

(iii) Routine coagulation screening tests (PT, aPTT): ☐ Yes ☐ No ☐ DK **If yes,** any abnormalities? _____

(iv) Positive culture: ☐ Yes ☐ No ☐ DK **If yes,** ☐ Blood ☐ Urine ☐ CSF Specify: _____

(v) Echocardiography: ☐ Yes ☐ No ☐ DK **If yes,** findings? _____

(vi) Newborn Screening Test : ☐ Yes ☐ No ☐ DK **If yes,** any abnormalities? _____

INVESTIGATIONS FOR STROKE (MOTHER AND CHILD)

35. (i) Were any of the results of the following investigations abnormal? (Please tick all that were abnormal):

Mother	**Child**
☐ Activated Protein C Resistance (APCR)	☐ Activated Protein C Resistance (APCR)
☐ Anti-thrombin III (ATIII)	☐ Anti-thrombin III (ATIII)
☐ Fibrinogen	☐ Fibrinogen
☐ Plasminogen	☐ Plasminogen
☐ Protein S	☐ Protein S
☐ Protein C	☐ Protein C
☐ Factor V Leiden (FVL)	☐ Factor V Leiden (FVL)
☐ Methylenetetrahydrofolate Reductase (MTHFR)	☐ Methylenetetrahydrofolate Reductase (MTHFR)
☐ Prothrombin Gene	☐ Prothrombin Gene
☐ Homocysteine	☐ Homocysteine
☐ Factor VIII	☐ Factor VIII
☐ Factor IX	☐ Factor IX
☐ Factor XI	☐ Factor XI
☐ Lipoprotein (a)	☐ Lipoprotein (a)
☐ **Positive X 1** Anticardiolipin antibody (ACLA IgG)	☐ **Positive X 1** Anticardiolipin antibody (ACLA IgG)
☐ **Positive X 2** Anticardiolipin antibody (ACLA IgG)	☐ **Positive X 2** Anticardiolipin antibody (ACLA IgG)
☐ **Positive X 3** Anticardiolipin antibody (ACLA IgG)	☐ **Positive X 3** Anticardiolipin antibody (ACLA IgG)
☐ **Positive X 1 for** Lupus Anticoagulant	☐ **Positive X 1 for** Lupus Anticoagulant
☐ **Positive X 2 for** Lupus Anticoagulant	☐ **Positive X 2 for** Lupus Anticoagulant
☐ **Positive X 3 for** Lupus Anticoagulant	☐ **Positive X 3 for** Lupus Anticoagulant

(ii) Hypercoagulable disorder: ☐ Yes ☐ No ☐ DK **If yes,** Specify tests:_____

(iii) Known platelet aggregation disorder: ☐ Yes ☐ No ☐ DK **If yes,** Specify: _____

(iv) Any other haematological abnormality: ☐ Yes ☐ No ☐ DK **If Yes,** Specify: _____

TREATMENT/OUTCOME AT DISCHARGE (THIS CHILD)

36. (i) Did the child require respiratory support? ☐ Yes ☐ No ☐ DK **If yes,** ☐ CPAP ☐ Mechanical ventilation ☐ Other _____

(ii) Did the child undergo: Therapeutic cooling: ☐ Yes ☐ No ☐ DK; Surgical intervention:☐ Yes ☐ No ☐ DK; Neuroradiological intervention :☐ Yes ☐ No ☐ DK; ☐ Other; Specify _____

(iii) Did the child receive: Anticoagulation treatment ☐ Yes ☐ No ☐ DK **If yes,** Specify *(tick all that apply)*: ☐ Unfractionated heparin ☐ Low molecular weight heparin ☐ Aspirin ☐ Other; Specify _____

37. Outcome at discharge:

(a) Is the child alive? ☐ Yes ☐ No ☐ DK **If No,** date of death: _____ /_____ /_____ (dd/mm/yyyy)

(b) Did the child have any neurological deficits at discharge? ☐ Yes ☐ No ☐ DK **If yes,** Specify:_____

Thank you for your help with this research project. Please return this questionnair to the APSU via email (SCHN-APSU@health.nsw.gov.au) or fax to 02 9845 3082 even if you don't complete all items. Australian Paediatric Surveillance Unit, Kid's Research Institute, Locked Bag 4001, Westmead NSW 2145.

The APSU is affiliated with the Royal Australasian College of Physicians (Paediatrics and Child Health Division) and Sydney Medical School, The University of Sydney. APSU is funded by the Australian Government Department of Health. This study has been approved by a Human Research Ethics Committee properly constituted under NHMRC guidelines.

Appendix B: APSU_Stroke Followup_Questionnaire_v 1.3

Stroke in Australian Children Under 2 Years of Age – Follow-up **Australian Paediatric Surveillance Unit** Please contact the APSU (02) 9845 3005 or SCHN-APSU@health.nsw.gov.au if you have any questions about this form *Instructions: Please answer each question by ticking the appropriate box or writing your response in the space provided. DK = Don't Know; NA = Not Applicable*	*APSU Office Use Only*	
	Study ID #:	
	Month/Year:	
	Version 1.3_26.08.2019	

This study is a follow-up to the cases of stroke that you have previously reported.
Please include de-identified reports not already supplied with the initial questionnaire.

REPORTING CLINICIAN'S DETAILS:

1. APSU Dr Code/Name: _____ /_____

PATIENT DETAILS (of the child reported in the previous study)

2. First 2 letters of first name: ___ ___ **3.** First 2 letters of surname: ___ ___ **4.** Postcode of family: _____

5. Date of Birth: __ __ / __ __ / __ ____ (dd/mm/yyyy)

INVESTIGATIONS FOR STROKE (THIS CHILD)

6. Placental pathology: Please attach de-identified report, if available.

7. Brain Imaging: Please attach de-identified Brain imaging reports:

(a) MRI

(b) CT

(c) Head US reports

8. EEG: Please attach de-identified EEG report.

INVESTIGATIONS FOR STROKE (MOTHER AND CHILD)

9(a). (i) Any hypercoagulable disorder, platelet aggregation disorder and/or haematological abnormality (mother): ☐ Yes ☐ No ☐ DK

If Yes, specify: _____

9(b). (i) Any hypercoagulable disorder, platelet aggregation disorder and/or haematological abnormality (child): ☐ Yes ☐ No ☐ DK

If Yes, specify: _____

FOLLOW-UP (THIS CHILD)

10. Paediatrician/Neonatologist/Allied health professional/General Movements/HINEs/Growth & Development follow up: Please attach de-identified report(s) - please ensure the date of report is visible.

OUTCOME (THIS CHILD)

11(a). Outcome of the child? ☐ Alive ☐ Deceased ☐ DK

11(b). ***If deceased,*** date of death: _____ /_____ /_____ *(dd/mm/yyyy)*

11(c). Did the alive or deceased child have any neurological deficits at the time of last review? ☐ Yes ☐ No ☐ DK

If yes, specify: _____

_____

_____

_____

Thank you for your help with this research project.

The APSU is affiliated with the Royal Australasian College of Physicians (Paediatrics and Child Health Division)
and Faculty of Medicine and Health, The University of Sydney.
The APSU is funded by the Australian Government Department of Health.
This study has been approved by a Human Research Ethics Committee properly constituted under NHMRC guidelines.

Appendix C: APSU_Stroke_Protocol_July 2017

Australian Paediatric Surveillance Unit
STUDY PROTOCOL

COMMENCING JULY 2017

Stroke in Australian Children under 2-years of age

BACKGROUND

Stroke is an acute vascular event causing focal interruption of blood supply to the brain. This can occur at any age including in a foetus. The incidence of stroke is high in the newborn period[1] and in adulthood. It is the third most common cause of adult mortality in Australia[2,3] though neonates survive stroke better than adults.

We are interested in studying all types of stroke in children up to 2 years of age diagnosed by radiological and or clinical examination irrespective of the timing of the cerebral insult and including all outcomes of stroke including death and disability. We are excluding children with intraventricular haemorrhage (IVH) and stroke following accidental head injury or following physical abuse.

Previous estimates suggest an incidence of perinatal stroke of approximately 0.25/1000 live births annually[6], however, more recent studies estimate the incidence in preterm infants to be as high as 7 per 1000 live births annually[7]. The incidence of pediatric stroke in children aged <18 years is estimated at 1.2 to 13 cases per 100,000 children annually[8,9].The true incidence is likely to be higher in more recent studies with the availability of advanced diffusion-weighted (dw) magnetic resonance (MR) imaging[4,5], especially in the case of hyper acute and acute stroke.

The most common clinical presentation of stroke is focal seizures. Perinatal stroke may be associated with nonspecific symptoms such as poor feeding, cyanosis, unexplained tachypnoea or apnoea[10]. Some children may present later in infancy with signs of motor deficit or cerebral palsy[11]. The diagnosis of stroke is by radiological imaging including brain MRI or CT, by the General Movements assessment[12] or neurological examination.

There is a gap in the epidemiological information regarding perinatal stroke in Australia as currently there is no national stroke registry for children. Some Australian centres have collaborated to contribute to the International Paediatric Stroke Study Group (IPSSG) registry[13].

This study will improve our understanding of childhood stroke and assist in ascertaining its incidence in Australia. Most importantly, through this surveillance study we aim to establish the need for a 'national stroke register for children under 2-years of age' which is currently lacking.

This study will help to identify high-risk groups and to inform development of preventive measures. It will also raise awareness of neonatal stroke among Australian paediatricians and the importance of neurodevelopmental assessment for an early and accurate diagnosis and/or early intervention therapies.

STUDY OBJECTIVES

1. To determine the incidence of stroke in Australian children < 2 years of age
2. To describe the epidemiology of stroke in the same population

3. To describe high risk groups that could be targeted for prevention strategies
4. To raise awareness among clinicians of the need to perform detailed neurodevelopmental assessments and imaging to support early diagnosis and intervention

Please see over for Case Definition

CASE DEFINITION [4,5]

Please report any child < 2 years of age, diagnosed with cerebral stroke by radiological and/or clinical examination, irrespective of timing of insult and including all outcomes of stroke i.e. death and disability.

Exclusion

Please exclude children with intraventricular haemorrhage and stroke following accidental head injury or following physical abuse.

REPORTING INSTRUCTIONS

1. Please report any child newly diagnosed with cerebral stroke that meets the case definition above, whom you have seen within the last month and that you have not previously reported to APSU.

Investigators (*Principal Investigator and contact person): *Dr Bithi Roy (The Mater Hospital, Sydney, The University of Sydney, The University of Notre Dame NSW), Professor Iona Novak (Cerebral Palsy Alliance, The University of Sydney NSW), Professor Nadia Badawi (The Children's Hospital at Westmead, Cerebral Palsy Alliance, The University of Sydney, NSW), A/Professor Karen Walker (The Children's Hospital at Westmead, Cerebral Palsy Alliance, The University of Sydney, NSW), Dr Cathy Morgan (Cerebral Palsy Alliance, The University of Sydney, NSW)

National reference group members: Professor Nadia Badawi (The Children's Hospital at Westmead, Cerebral Palsy Alliance, The University of Sydney, NSW), A/Professor Rodney Hunt (The Royal Children's Hospital, Melbourne, Murdoch Children's Research Institute, University of Melbourne VIC), A/Professor Mark Mackay (The Royal Children's Hospital, Melbourne, Murdoch Children's Research Institute, Florey Institute of Neurosciences and Mental Health, Wimmera Base Hospital, University of Melbourne, Geelong University VIC), Professor Alison Kent (Centenary Hospital for Women and Children, Australian National University Medical School ACT), Dr Lakshmi Nagarajan (Princess Margaret Hospital for Children, School of Paediatrics and Child Health WA), Dr Adriane Sinclair (Lady Cilento Children's Hospital, Children's Health Queensland Hospital and Health Service QLD)

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REFERENCES

1. Mineyko A, Kirton A. The black box of perinatal ischemic stroke pathogenesis. J Child Neurol 2011;26:1154–62.
2. Leading causes of death (AIHW) www.aihw.gov.au/deaths/leading-causes-of-death
3. Facts and figures about stroke – Stroke Foundation – Australia https://strokefoundation.com.au/about-stroke/facts-and-figures-about-stroke
4. Nelson KB, Lynch JK. Stroke in newborn infants. Lancet Neurol 2004;3:150-158
5. Krishnamoorthy KS, Soman TB, Takeoka M, Schaefer PW. Diffusion-weighted imaging in neonatal cerebral infarction: Clinical utility and follow-up. J Child Neurol. 2000;15:592–602
6. Lee J, Croen LA, Backstrand KH, Yoshida CK, Henning LH, Lindan C, Ferriero DM, Fullerton HJ, Barkovich AJ, Wu YW. Maternal and infant characteristics associated with perinatal arterial stroke in the infant. JAMA. 2005; 293: 723–729
7. Benders MJ, Groenendaal F, De Vries LS. Preterm arterial ischemic stroke. Semin Fetal Neonatal Med. 2009;14:272–277
8. Lynch JK, Hirtz DG, DeVeber G, et al. Report of the National Institute of Neurological Disorders and Stroke Workshop on Perinatal and Childhood Stroke. Pediatrics. 2002;109:116–123

9. Lynch JK, Han CJ. Pediatric stroke: what do we know and what do we need to know? Seminars in Neurology. 2005;25(4):410–423
10. Stroke in the newborn – UpToDate (2017) www.uptodate.com/contents/stroke-in-the-newborn
11. Badawi N, Keogh JM (2013) Causal pathways in cerebral palsy. J Paediatr Child Health 49:5–8
12. Morgan, Catherine et al. "Sensitivity And Specificity Of General Movements Assessment For Diagnostic Accuracy Of Detecting Cerebral Palsy Early In An Australian Context". J Paediatr Child Health 52.1 (2015): 54-59.
13. Prospective web based stroke registry - https://app3.ccb.sickkids.ca/cstrokestudy/other/studyInfo.jsp

Appendix D: APSU_Stroke_Protocol_21-12-21

STUDY PROTOCOL
Stroke in Australian Children under 2-years of age

BACKGROUND

Stroke is an acute vascular event causing focal interruption of blood supply to the brain. This can occur at any age including in a foetus. The incidence of stroke is high in the newborn period[1] and in adulthood. It is the third most common cause of adult mortality in Australia[2,3] though neonates survive stroke better than adults.

We are interested in studying all types of strokes in children up to 2 years of age diagnosed by radiological and or clinical examination, irrespective of the timing of the cerebral insult and including all outcomes of stroke such as death and disability. We are excluding children with intraventricular haemorrhage (IVH) and stroke following accidental head injury or following physical abuse.

Previous estimates suggest an incidence of perinatal stroke of approximately 0.25/1000 live births annually[6], however, more recent studies estimate the incidence in preterm infants to be as high as 7 per 1000 live births annually[7]. The incidence of pediatric stroke in children aged <18 years is estimated at 1.2 to 13 cases per 100,000 children annually[8,9]. The true incidence is likely to be higher in more recent studies with the availability of advanced diffusion-weighted (dw) magnetic resonance (MR) imaging[4,5], especially in the case of hyper acute and acute stroke.

The aetiology of stroke is complex and multifactorial. Some of the risk factors also occur in the general population.

There is a gap in the epidemiological information regarding perinatal stroke in Australia as currently there is no national stroke registry for children. Some Australian centres have collaborated to contribute to the International Paediatric Stroke Study Group (IPSSG) registry[13].

RESEARCH PLAN

Study Design

This is an observational case control study.

This study has two stages. The first stage was to enrol stroke cases prospectively between July 2017 and June 2019. Data collection for this stage is completed. In stage one, we enrolled 74 cases of stroke via the Australian Paediatric Surveillance Unit (APSU) methodology, via a paid special one-off data collection. APSU is the national resource, established in 1993 to facilitate active surveillance of uncommon and rare childhood diseases, complications of common diseases or adverse effects of treatment. It is closely affiliated with University of Sydney, Discipline of Child and Adolescent Health and Sydney Children's Hospital Network. We jointly own this stroke dataset with APSU. The preliminary analysis of the stroke data indicates, that, the risk factors in the stroke population were likely to be present in some of the general population.

In the second stage, we aim to collect control data. Control data will enable inferential statistical analysis of the risk factors for stroke in children especially in infants, compared to control cases.

Aim of the study

Primary aim:

1. To identify the rates of risk factors in the causal pathway of stroke in Australian children under 2-years of age by comparing with age matched controls.

Secondary aims:

1. To identify high-risk groups for stroke and development of preventive measures.
2. To ascertain incidence of stroke in Australia in this age group.

3. To establish the need for a 'national stroke register for children under 2-years of age' which is currently lacking.
4. To raise awareness among clinicians of the need to perform detailed neurodevelopmental assessments and imaging to support early diagnosis and intervention

Participants
Seventy-four stroke cases have been enrolled between July 2017-June 2019.
We now require 150 controls (1 stroke case:2 controls) in the same period as the stroke cases i.e July 2017-June 2019.

Sample size
We have consulted with our biostatistician. We have been advised to recruit 150 control cases to the existing 74 stroke cases i.e 2:1 ratio, to enable adequate power to control for the large number of risk factors for stroke and the cooccurrence of some of these risk factors in the general population.

Inclusion criteria
1. Well babies born at Royal Prince Alfred Hospital between July 2017 and June 2019,
2. Above 35 weeks gestational age,
3. Not admitted to the neonatal unit

Exclusion criteria
1. Babies with congenital malformations

RECRUITMENT METHODS
Identification of participant
This is a retrospective de-identified data from the Royal Prince Alfred Hospital medical records as per the inclusion criteria. Randomly every 10th, 15th or 20th control case will be selected manually between the period July 2017-June 2019.
Note: We have selected RPA because Prof Karen Walker is Neonatal Clinical Nurse Consultant at RPA and is one of the authors of this study.

Consent process
This is a retrospective non-identifiable dataset from the RPA medical records, and therefore consent will not be required.
Note: The RPA investigator, Prof Karen Walker will prepare and convert the re-identifiable control case into non-identifiable cases for the non-RPA investigators.
We will apply for waiver of consent as per SCHN Ethics guideline.

DATA MANAGEMENT PLAN
Data collection
METHOD: Retrospective non-identifiable RPA Hospital medical record review.
DATABASES: RPA medical records in the hospital database.
VARIABLES: All variables are specified in the attached excel sheet.
QUESTIONNAIRES: Not applicable.
DATA COLLECTION METHOD: The RPA control data we are seeking, has already been collected as part of routine care, and is stored in the hospital medical record. To protect privacy, before export of the RPA control dataset to the Principal Investigator, the RPA investigator will identify potential RPA controls meeting the inclusion criteria from medical records and convert these data to non-identifiable data format.
PROCESSES OF IDENTIFIABILITY: RPA investigator, Prof. Walker will prepare the RPA data sheet into a non-identifiable data format using codes, that, only she will be able to reidentify. For example, DOB will be converted to age in days to prevent re-identification. Non-RPA study investigators will have only access to non-identifiable data and have no capability to re-identify RPA participants.

Analysis of the Data Plan
Data analysis will be conducted by biostatistician and epidemiological personnel at the APSU. The rates of risk factors in the RPA non-identifiable controls will be compared to the rates of risk factors in the APSU stroke population using logistic regression.

Reporting of Results
The results will be reported in de-identified group format, in medical conference presentations and peer-reviewed journal publications. Privacy issues are not applicable as it is de-identified data. It will not be possible for the Principal Investigator to provide RPA control participants with a copy of the results, given no contact will be made with participants as this is non-identifiable data. We will, however, provide the results to the staff at RPA.

Data storage
Professor Walker, RPA Investigator will be the only study personnel to access the RPA hospital server, using two passwords - the first to initially access the computer, and the second to access the restricted-access server. RPA data will also be linked to a password-protected electronic laboratory notebook (ELN). This is password protected at two levels (first, to access the computer; second to access the ELN), and only authorized users will have access to the relevant ELN. Prof Walker will convert the RPA control dataset from re-identifiable to non-identifiable data in an Excel format and handover to the Principal Investigator via a secured file transfer format in compliance with the University of Sydney digital security and privacy policies e.g., Cloud Store.

De-identified data will be stored in a secure, web-based REDCap database, that is password protected. The REDCap system is hosted on a server that is behind The University of Sydney's firewall and access is restricted with multi-factor authentication and TLS encrypted connections. Each participant will be given a unique code by the RPA Investigator before the de-identified data is uploaded onto the secure, web-based REDCap database. This process meets The University of Sydney data storage and privacy requirements, where the Principal Investigator is enrolled as higher degree candidate. We are seeking a waiver of consent for non-identifiable data, and these approvals will be obtained before moving the data offsite.

Access to data
Access to data is limited to the study investigator listed on the ethics application. Prof. Walker, RPA Investigator will be the only study personnel to access the RPA control re-identifiable data.

Research data storage time
The data will be stored for 5 years post completion of the study. There are no audio or video recording being collected.

Sharing and re-use of data
Professor Walker, RPA Investigator will be the only study personnel to have access to the RPA control re-identifiable data.

The de-identified RPA control dataset will be shared with only the study personnel for the purpose of answering the aim of the study. RPA control participants will have no awareness of this process, as it is non-identifiable data. We are seeking a consent waiver for this process.
The data will not be re-used.

Disposal of Data
Electronic data will be deleted after the end of the stipulated storage period of 5 years.

STUDY RISKS AND BENEFITS
Risk and Benefits: There is no risk to participants as this is not an interventional study, nor is there any risk to patient privacy as we are seeking to access non-identifiable data.
Findings from this research are in the public interest as identification of preventable factors could potentially impact the burden of stroke in Australian children.

Investigators (*Principal Investigator and contact person): *Dr Bithi Roy (North Shore Private Hospital, NSW, The University of Sydney, The University of Notre Dame NSW), Professor Iona Novak (Cerebral Palsy Alliance, The University of Sydney NSW), Professor Nadia Badawi (The Children's Hospital at Westmead, Cerebral Palsy Alliance, The University of Sydney, NSW), A/Professor Karen Walker (Royal Prince Alfred (RPA) Hospital), Dr Cathy Morgan (Cerebral Palsy Alliance, The University of Sydney, NSW), Annabel Webb (Cerebral Palsy Alliance, The University of Sydney, NSW)

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REFERENCES

1. Mineyko A, Kirton A. The black box of perinatal ischemic stroke pathogenesis. J Child Neurol 2011;26:1154–62.
2. Leading causes of death (AIHW) www.aihw.gov.au/deaths/leading-causes-of-death
3. Facts and figures about stroke – Stroke Foundation – Australia https://strokefoundation.com.au/about-stroke/facts-and-figures-about-stroke
4. Nelson KB, Lynch JK. Stroke in newborn infants. Lancet Neurol 2004;3:150-158
5. Krishnamoorthy KS, Soman TB, Takeoka M, Schaefer PW. Diffusion-weighted imaging in neonatal cerebral infarction: Clinical utility and follow-up. J Child Neurol. 2000;15:592–602
6. Lee J, Croen LA, Backstrand KH, Yoshida CK, Henning LH, Lindan C, Ferriero DM, Fullerton HJ, Barkovich AJ, Wu YW. Maternal and infant characteristics associated with perinatal arterial stroke in the infant. JAMA. 2005; 293: 723–729
7. Benders MJ, Groenendaal F, De Vries LS. Preterm arterial ischemic stroke. Semin Fetal Neonatal Med. 2009;14:272–277
8. Lynch JK, Hirtz DG, DeVeber G, et al. Report of the National Institute of Neurological Disorders and Stroke Workshop on Perinatal and Childhood Stroke. Pediatrics. 2002;109:116–123
9. Lynch JK, Han CJ. Pediatric stroke: what do we know and what do we need to know? Seminars in Neurology. 2005;25(4):410–423
10. Stroke in the newborn – UpToDate (2017) www.uptodate.com/contents/stroke-in-the-newborn
11. Badawi N, Keogh JM (2013) Causal pathways in cerebral palsy. J Paediatr Child Health 49:5–8
12. Morgan, Catherine et al. "Sensitivity And Specificity Of General Movements Assessment For Diagnostic Accuracy Of Detecting Cerebral Palsy Early In An Australian Context". J Paediatr Child Health 52.1 (2015): 54-59.
13. Prospective web based stroke registry - https://app3.ccb.sickkids.ca/cstrokestudy/other/studyInfo.jsp

Appendix E: LNR Approval_19-01-17

The Sydney children's Hospitals Network

care, advocacy, research, education

Contact for this correspondence:
Research Ethics Office
Research Ethics Administration Assistant
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ABN 53 188 579 090

19 January 2017

Dr Bithi Roy
Neonatology
The Mater Hospital

Dear Dr Roy,

HREC Reference: **LNR/16/SCHN/449**

Project title: **Stroke in Australian Children under 2-year age**

Sites: **The Children's Hospital at Westmead**

Thank you for submitting the above project for single ethical and scientific review. This project was considered by the Sydney Children's Hospitals Network Human Research Ethics Committee's Executive Committee ("the Committee") at its meeting **1 December 2016**, and subsequently by the Executive of SCHN HREC on the **16 January 2017.**

This HREC has been accredited by the NSW Department of Health as a lead HREC under the model for single ethical and scientific review, and by the National Health and Medical Research Council as a certified committee in the review of multi-centre clinical research projects.

This HREC is constituted and operates in accordance with the National Health and Medical Research Council's *National Statement on Ethical Conduct in Human Research* and *CPMP/ICH Note for Guidance on Good Clinical Practice.*

I am pleased to advise that the Committee has granted ethical approval of this research project. Your approval is valid for three (3) years, effective the date of this letter.

This application has been assessed in accordance with, and meets the requirements of the National Statement on Ethical Conduct in Human Research (2007).

The documents reviewed and approved by the Committee are:

Document Reviewed	*Version*	*Date*
LNR Application – AU/6/E38A26		29 November 2016
APSU Study Protocol		Received 30 November 2016
Stroke Prevalence Calculations		Received 30 November 2016
Email correspondence from Investigator		Received 30 November 2016
Ethics cover letter		22 November 2016
APSU Stroke Questionnaire	V1	8 November 2016
LNR Study Protocol	V2	Received 11 January 2017
C V - Bithi Roy		15 May 2016
APSU New Study Guidelines Full application		Received 11 January 2017
LNR Response cover letter		10 January 2017

Please note the following conditions of approval:

1. The Coordinating Investigator will immediately report anything which may warrant review of ethical approval of the project in accordance with the SCHN adverse event reporting policy.

2. All proposed changes to the research protocol, including the conduct of the research, changes to site or personnel, or an extension to HREC approval, are to be provided to the HREC or its delegate for review before those changes can take effect.

3. The HREC will be notified, giving reasons, if the project is discontinued at a site before the expected date of completion.

4. The co-ordinating investigator will provide an annual report to the HREC on the anniversary of this approval letter, and a final report on completion of the study.

5. Your approval is valid for three (3) years from the date of the final approval letter. If your project extends beyond that three year period and you are still actively recruiting you will be required to resubmit your application incorporating any amendments within six (6) months of that approval expiry date. If your project is in follow up on, or analysis, please submit and application for amendment to extend the approval period. Ethics approval can be extended for a period of twelve (12) months at a time.

6. In the event of a project **not having commenced** within 12 months of its approval, the approval will lapse and reapplication to the HREC will be required.

Should you have any queries about the HREC's consideration of your project please contact the Research Ethics Administration Assistant on (02) 9845 1253.

You are reminded that this letter constitutes ethical approval only. You must not commence this research project at a site until separate authorisation from the Chief Executive or delegate of that site has been obtained. A copy of this letter must be forwarded to all site investigators for submission to the relevant Research Governance Officer.

The HREC wishes you every success in your research.

Yours faithfully

[redacted]

Dr Peter Cooper
Chair, Sydney Children's Hospitals Network Human Research Ethics Committee
Sydney Children's Hospitals Network Human Research Ethics Committee

Appendix F: LNR_Study Protocol_Stroke Followup_v1.1

Australian Paediatric Surveillance Unit
STUDY PROTOCOL

COMMENCING JULY 2019

Stroke in Australian Children under 2-years of age – Follow-up

BACKGROUND

We have prospectively collected the epidemiological information of stroke in Australian children under 2-years of age via Australian Paediatric Surveillance Unit.

We are now collecting follow up information of the previously reported cases of stroke. This will include follow up reports from paediatrician, neonatologist and/or physiotherapist, General Movements (GMs), Hammersmith Infants Neurological Examination (HINE) and/or Growth and Developmental follow up report.

STUDY OBJECTIVES

1. To follow up the outcome of stroke in Australian children <2 years of age

Please see over for Case Definition

CASE DEFINITION

Please include the follow up reports from paediatrician, neonatologist and/or physiotherapist, General Movements (GMs), Hammersmith Infants Neurological Examination (HINE) and/or Growth and Development follow up reports of the previously reported cases of stroke by yourself.

Investigators (*Principal Investigator and contact person): *Dr Bithi Roy (The Mater Hospital, Sydney, The University of Sydney, The University of Notre Dame NSW), Professor Iona Novak (Cerebral Palsy Alliance, The University of Sydney NSW), Professor Nadia Badawi (The Children's Hospital at Westmead, Cerebral Palsy Alliance, The University of Sydney, NSW), A/Professor Karen Walker (The George Institute for Global Health, Newtown, The University of Sydney, NSW), Dr Cathy Morgan (Cerebral Palsy Alliance, The University of Sydney, NSW)

National reference group members: Professor Nadia Badawi (The Children's Hospital at Westmead, Cerebral Palsy Alliance, The University of Sydney, NSW), Professor Rodney Hunt (The Royal Children's Hospital, Melbourne, Murdoch Children's Research Institute, University of Melbourne VIC), Professor Mark Mackay (The Royal Children's Hospital, Melbourne, Murdoch Children's Research Institute, Florey Institute of Neurosciences and Mental Health, Wimmera Base Hospital, University of Melbourne, Geelong University VIC), Professor Alison Kent (Centenary Hospital for Women and Children, Australian National University Medical School ACT), Dr Lakshmi Nagarajan (Princess Margaret Hospital for Children, School of Paediatrics and Child Health WA), Dr Adriane Sinclair (Lady Cilento Children's Hospital, Children's Health Queensland Hospital and Health Service QLD)

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REFERENCES

Mineyko A, Kirton A. The black box of perinatal ischemic stroke pathogenesis. J Child Neurol 2011;26:1154–62.

Leading causes of death (AIHW) www.aihw.gov.au/deaths/leading-causes-of-death

Facts and figures about stroke – Stroke Foundation – Australia https://strokefoundation.com.au/about-stroke/facts-and-figures-about-stroke

Nelson KB, Lynch JK. Stroke in newborn infants. Lancet Neurol 2004;3:150-158

Krishnamoorthy KS, Soman TB, Takeoka M, Schaefer PW. Diffusion-weighted imaging in neonatal cerebral infarction: Clinical utility and follow-up. J Child Neurol. 2000;15:592–602

Lee J, Croen LA, Backstrand KH, Yoshida CK, Henning LH, Lindan C, Ferriero DM, Fullerton HJ, Barkovich AJ, Wu YW. Maternal and infant characteristics associated with perinatal arterial stroke in the infant. JAMA. 2005; 293: 723–729

Benders MJ, Groenendaal F, De Vries LS. Preterm arterial ischemic stroke. Semin Fetal Neonatal Med. 2009;14:272–277

Lynch JK, Hirtz DG, DeVeber G, et al. Report of the National Institute of Neurological Disorders and Stroke Workshop on Perinatal and Childhood Stroke. Pediatrics. 2002;109:116–123

Lynch JK, Han CJ. Pediatric stroke: what do we know and what do we need to know? Seminars in Neurology. 2005;25(4):410–423

Stroke in the newborn – UpToDate (2017) www.uptodate.com/contents/stroke-in-the-newborn

Badawi N, Keogh JM (2013) Causal pathways in cerebral palsy. J Paediatr Child Health 49:5–8

Morgan, Catherine et al. "Sensitivity And Specificity Of General Movements Assessment For Diagnostic Accuracy Of Detecting Cerebral Palsy Early In An Australian Context". J Paediatr Child Health 52.1 (2015): 54-59.

Prospective web based stroke registry - https://app3.ccb.sickkids.ca/cstrokestudy/other/studyInfo.jsp

Appendix G: LNRSSA Approval_19-04-17

The Sydney children's Hospitals Network
care, advocacy, research, education

Contact for this correspondence:
Research and Development
Name: Jessica Grundy
Phone: (02) 9845 3084
Facsimile: (02) 9845 1317
Email: jessica.grundy@health.nsw.gov.au

Date: 19 April 2017

Dr Bithi Roy
University of Sydney

Site Authorisation Letter

Dear Dr Roy,

HREC reference number: LNR/16/SCHN/449

SSA reference number: LNRSSA/17/SCHN/57

Project title: **Stroke in Children**

Site: **The Children's Hospital at Westmead**

Thank you for submitting an application for authorisation of this project. I am pleased to inform you that authorisation has been granted for this study to take place at the above site.

The following conditions apply to this research project. These are additional to those conditions imposed by the Human Research Ethics Committee that granted ethical approval:

1. Please advise us of the date when the project starts at this site.

[redacted]ed amendments to the research protocol or conduct of the research which may affect [redacted]ical acceptability of the project, and which are submitted to the lead HREC for review, [redacted]ied to the research governance officer.

3. Proposed amendments to the research protocol or conduct of the research which may affect

the ongoing site acceptability of the project are to be submitted to the research governance officer.

Yours sincerely,

Jessica Grundy
Research Governance Officer

Appendix H: LNR Approval_RPA

From: "no_reply@regis.health.nsw.gov.au" <no_reply@regis.health.nsw.gov.au> Subject: 2019/ETH06281: Notification of an amendment to a research study - Addition of a New Site - (74546) - Approved
Date: 9 December 2021 at 3:25:11 pm AEDT
To: "broy8525@uni.sydney.edu.au" <broy8525@uni.sydney.edu.au>
Cc: "broy8525@uni.sydney.edu.au" <broy8525@uni.sydney.edu.au>, "Karen Walker (Sydney LHD)" <karen.walker@health.nsw.gov.au>, SCHN-Ethics <SCHN-Ethics@health.nsw.gov.au>

Date of Decision Notification: **09 Dec 2021**
Low or negligible risk review pathway

Dear Dr Bithi Roy,

Thank you for submitting a Notification of an amendment to a research study - Addition of a New Site with ID (74546 for the following study;
2019/ETH06281: Stroke in Australian Children under 2-year age

The Amendment has been reviewed by the Executive of the **Sydney Children's Hospitals Network Human Research Ethics Committee** at its meeting held on **29/11/2021** who have determined the Amendment has been approved.

Notification of an amendment to a research study - Addition of a New Sitewith form ID 74546

- Royal Prince Alfred Hospital

PI name:

- Associate Professor Karen Walker

PI email:

- karen.walker@health.nsw.gov.au

The following documentation is included in this approval:

- 074546_Notification of an amendment to a research study - Addition of a New Site, 22 November 2021
- 074546_WalkerK_CV_Short, received 22 November 2021

It is noted that the Sydney Children's Hospitals Network Human Research Ethics Committee is constituted in accordance with the National Statement on Human Conduct in Research, 2007 (NHMRC).

This email constitutes ethical and scientific approval only.
This project cannot proceed at any site until separate research governance authorisation has been obtained from the Institution at which the research will take place.

Should you require any further information, please don't hesitate to contact the Ethics team at SCHN-Ethics@health.nsw.gov.au.

Yours Sincerely,
Victoria
Sent on behalf of Associate Professor Sarah Garnett
Chair, Sydney Children's Hospitals Network Human Research Ethics Committee

Victoria Luccitti | **Research Ethics Support Officer** | **Ethics and Governance**
t: (02) 9845 1253 | **e:**victoria.luccitti@health.nsw.gov.au | **w:**https://www.schn.health.nsw.gov.au/research/ethics-governance/ethics
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Appendix I: Stroke_Prevalence calculation

Stroke Prevalence Calculations from the Australian Cerebral Palsy Register

Strokes result in cerebral palsy 30-40% of the time. Children on the Australian Cerebral Palsy Register all have cerebral palsy and therefore represent 30-40% of the strokes that occur.

From 1993-2006 the number of children on the Australian Cerebral Palsy Register with postneonatal cerebral palsy arising from stroke, in the 3 states with population data (WA; SA; VIC 43% of the Australian population) was: n=13 (associated with surgery); n= 9 (associated with cardiac complications); n=38 (spontaneous), making a total of n=60 cases/13 years.

If we estimate the other states had the same prevalence, this would mean: 140 cases in Australia/13 years = n=11 strokes/yr resulting in postneonatal cerebral palsy.

Accounting for the fact that only 30-40% of strokes result in CP, this would conservatively mean a total of n=37 strokes occur per year in Australia.

In addition to post neonatal stokes a small number of strokes occur neonatally. The Australian New Zealand Neonatal Network (ANZNN) report does not report on rates of stroke in the preterm population and therefore we are unable to accurate report on these numbers.

n=37p.a. +/- neonatal cases n=unknown

Stroke Prevalence Calculations Australian Stroke Registry

In 2014 – recorded n=16 strokes in children<13yrs old. Note this a relatively new register and has not yet reached full population ascertainment.

n=16p.a. +/- neonatal cases n=unknown

International Prevalence of Perinatal Stroke

Incidence of perinatal stroke = 1/4000 live births, with a 10% mortality rate (Lynch & Nelson 2001)

Australian Birth Rate in 2014 = 299,700 births
Equates to n=74 perinatal strokes p.a. Then accounting for 90% survival n=67

n=67p.a. perinatal strokes

Appendix J: Sample size calculations for future studies

Placental histopathology

In a systematic review of studies investigating the role of the placenta in perinatal stroke (Study 1), a total of 10 studies investigated placental abnormality in 83 stroke cases and 110 controls. 72/83 (87%) of the stroke cases had placental abnormality and 66/110 (60%) of the control had placental abnormality.

Using these rates, the odds ratio for stroke among those with placental abnormality is approximately 4.33. To detect an effect of this size with power of 80% and significance level of 5%, a case control study with a 1:3 ratio of cases to matched controls would require n = 29 stroke cases and n = 87 controls, for a total sample size of n = 116.

Genetic/hematological abnormalities

In the APSU data (Study 6), 23 of the babies with stroke had genetic/hematological tests performed and 11 (48%) of these were abnormal. Rates of genetic/hematological abnormality in a control population are not currently available in the literature. Here we present some possible sample sizes for future case control studies under some scenarios for the possible rate of genetic/hematological abnormality in the control population. However, we stress that before a large case control study is carried out, a pilot study will be necessary to ascertain the rate of genetic/hematological abnormality more accurately in the control population.

If 48% of babies with stroke have genetic/hematological abnormalities and we consider a scenario where 20% of the control population have genetic/hematological abnormalities, then the odds ratio for stroke among those with genetic/hematological abnormalities would be 3.67. Under this scenario, the required number of stroke cases in a case control study with a 1:3 ratio of cases to matched controls would be n = 28 and the number of required controls would be n = 84, for a total sample size of n = 112, assuming statistical power of 80% and a significance level of 5%.

If 48% of babies with stroke have genetic/hematological abnormalities and we consider a more extreme scenario where 40% of the control population have genetic/hematological abnormalities, then the odds ratio for stroke among those with genetic/hematological abnormalities would be 1.38. Under this scenario, the required number of stroke cases in a case control study with a 1:3 ratio of cases to matched controls would be n = 405 and the number of required controls would be n = 1215, for a total sample size of n = 1640, assuming the same statistical power and significance level as above.

The wide range of possible required sample sizes here underlines the need for a small pilot study (for example of approximately n = 20 controls) to better estimate the rate of genetic/hematological abnormalities in the control population.