Near-Infrared Spectroscopy (NIRS) to measure nociception following noxious stimulation in critically ill infants

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December, 2011

A thesis submitted to McGill University in partial fulfilment of the requirements of the degree of

Doctor of Philosophy (Nursing)

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ABSTRACT

Background

An admission to the intensive care unit causes major physical and psychological stress for children. Pain contributes significantly to this distressing experience. To optimize pain relief, a sound pain assessment tool is essential. Although this need is recognized, only a few pain assessment instruments have been thoroughly tested with this population using a rigorous scientific approach, and none have been shown to be superior to the other. Recent studies using near-infrared spectroscopy (NIRS) in term and premature infants indicate that nociceptive stimuli cause haemodynamic changes in specific cortical regions. This suggests a new avenue for assessing and quantifying pain processing in critically ill infants that could be more sensitive and specific to the nociceptive response.

Aims

In this series of studies we examined regional cerebral and systemic haemodynamic changes, as well as behavioural reactions in critically ill infants with congenital heart defect (CHD) during chest drain removal following open heart surgery. Specifically, we examined changes within subjects, as well as individual factors (age, sex, medication) affecting the change and associations between cerebral haemodynamic changes, systemic physiological changes, and Faces Legs Activity Cry Consolability (FLACC) pain scores.

Subjects

Critically ill infants less than 12 months of age admitted to a cardiac intensive care unit after cardiac surgery for CHB comprised the sample.

Outcome measures

Changes in cerebrovascular haemoglobin concentrations (NIRS), as well as heart rate (ECG), systemic arterial oxygen saturation (pulse oximetry), and mean arterial blood pressure (arterial line) were recorded during three distinct epochs (Baseline, Tactile stimulation, and Chest-drain removal). Behavioural manifestations were also captured through video and were subsequently rated for pain with the FLACC scale.

Design

Descriptive correlational design.

Results

We studied 32 infants with CHD and obtained FLACC pain scores in 20 of these infants. Cerebral deoxygenated haemoglobin concentrations significantly increased across the epochs (p<.01). Physiological systemic responses were not found to be associated with the cerebral haemodynamic parameters. Mean FLACC pain scores significantly increased across the epochs (p < .001) with a mean score of 7/10 during the noxious procedure, despite administration of an analgesic agent (morphine). Sex of patients was found to be a determining factor in the cerebral haemodynamic responses and pain FLACC scores. Pharmacological treatments, age and weight of patients were significantly associated with cerebral and systemic haemodynamic responses, as well as the FLACC pain scores. The administration of a sedating agent (midazolam) had a significant dampening effect on the pain behaviours as assessed by the FLACC scale.

Conclusions

Using a multidimensional pain measurement approach, we demonstrated that significant cerebral, physiological and behavioural activity was present in response to a noxious

procedure in critically ill infants despite the administration of analgesic treatment. Although pain behaviours were significantly dampened by the sedating agent, the cerebral response was still evident. Thus, assessment of cerebral haemodynamics in the context of pain seems to be an important addition when a sedating agent is administered. Our data suggest that NIRS is a potentially useful technique for assessing pain evoked cerebral activation in critically ill infants.

ABRÉGÉ

Introduction

Une admission à l'unité des soins intensifs est une source de stress physique et psychologique chez l'enfant. La douleur contribue grandement à cette expérience affligeante. Pour optimiser le soulagement de la douleur, un bon outil de mesure est essentiel. Malgré que ce besoin soit reconnu, très peu d'instruments ont subi des tests scientifiques rigoureux auprès de cette population et aucun outil ne s'est démarqué des autres. Des études récentes utilisant la spectroscopie par infrarouge (SPIR) chez les nouveau-nés à terme et prématurés ont indiqué que des stimuli nociceptifs causent des changements hémodynamiques dans des régions cérébrales spécifiques. Cette approche semble prometteuse auprès des jeunes enfants gravement malades.

Objectifs

Nous avons examiné les changements hémodynamiques cérébraux et systémiques, ainsi que les réactions comportementales reliés au retrait d'un drain thoracique chez de jeunes enfants ayant subi une chirurgie à cœur ouvert pour une cardiopathie congénitale. Spécifiquement, nous avons exploré et comparé les changements de chaque enfant, ainsi que les facteurs individuels (âge, sexe, médication) affectant ces changements. De plus, les associations entre les changements hémodynamiques cérébraux et physiologiques, ainsi que les scores de douleur selon l'échelle *Faces Legs Activity Cry Consolability* (FLACC) furent étudiées.

Échantillon

L'échantillon comprenait de jeunes enfants gravement malades âgés de moins de 12 mois admis à l'unité des soins intensifs cardiaques après une chirurgie cardiaque pour correction d'une cardiopathie congénitale.

Mesure des paramètres

Les changements de concentrations en oxygène de l'hémoglobine (SPIR), ainsi que le rythme cardiaque (ECG), la saturation artérielle en oxygène (oxymétrie pulsatile), et la pression artérielle moyenne (ligne artérielle) furent recueillis pendant trois périodes distinctes (mesures initiales, stimulation tactile et retrait drain thoracique). Les manifestations comportementales furent obtenues par vidéo et évaluées, subséquemment, pour la douleur à l'aide de l'échelle FLACC.

Devis

Devis descriptif corrélationnel.

Résults

Nous avons étudié 32 enfants avec cardiopathie congénitale et avons obtenu des scores de douleur FLACC auprès de 20 de ces enfants. La concentration cérébrale de désoxygénation de l'hémoglobine a significativement augmenté entre les trois périodes (p<.01). Les réponses physiologiques systémiques ne furent pas associées aux paramètres hémodynamiques cérébraux. Les scores de douleur FLACC moyens ont significativement augmentés entre les périodes (p < .001), dont la moyenne était de 7/10 en réponse à la procédure douloureuse, malgré l'administration d'un agent analgésique (morphine). Le sexe des participants fut un facteur déterminant de la réponse hémodynamique cérébrale ainsi que pour les scores de douleur FLACC. L'administration de traitements

pharmacologiques, l'âge et le poids des enfants furent associés de façon significative aux changements hémodynamiques cérébraux et systémiques, ainsi que ceux des scores de douleur FLACC. L'administration d'un agent sédatif (midazolam) a eu un effet atténuant significatif sur les comportements de douleur tels que mesurés par l'échelle FLACC.

Conclusions

Nous avons démontré, à l'aide d'une approche multidimensionnelle, que des manifestations cérébrales, physiologiques et comportementales significatives étaient présentes en réponse à une procédure nociceptive chez le jeune enfant gravement malade, et ce, malgré l'administration d'un traitement analgésique. Les comportements communiquant une douleur furent significativement atténués par l'administration d'un agent sédatif. Par conséquent, l'évaluation de l'activité hémodynamique cérébrale lors de situation douloureuse s'avère être un important ajout lorsque des médicaments sédatifs sont administrés. Il semble que la SPIR soit une technique potentielle pour évaluer l'activation cérébrale évoquée par une stimulation nociceptive chez le jeune enfant gravement malade.

ACKNOWLEDGEMENTS

My doctoral studies have been remarkable and it is a pleasure to thank those who made this possible. First and foremost, I offer my sincerest gratitude to my supervisor, Dr Celeste Johnston, who has been an exceptional mentor, support, and provided me with amazing learning opportunities. Your infinite knowledge, experience, dedication, and passion for research have contributed greatly to the progression and completion of my doctoral journey. I would like to show my appreciation to my co-advisor, Dr Janet Rennick. Your thoroughness and rigor, as well as valuable guidance have assisted me throughout my studies. You are a remarkable role model for all present and future nurse scientists.

This thesis would not have been possible without the support and assistance of Dr Catherine Limperopoulos. Your inestimable knowledge and experience in infant brain imaging made the completion of my doctoral project possible. Indeed, conducting my study in Boston within the Fetal/Neonatal Brain Development and Injury Group, under the supervision of Dr Adré du Plessis, was in great part due to Dr Limperopoulos. Moreover, it was an honor for me to have worked with Dr du Plessis, a remarkable and passionate clinician, researcher, thinker and professor. You took me under your wing with open arms and for this, I will be forever indebted. In Boston, I was blessed and privileged to have worked with an amazing group of clinicians, researchers and academicians. Through lab meetings, discussions and exchanges with these colleagues, I pushed my thinking beyond my comfort zone and have learned valuable lessons. Many thanks are due to Dr Mustafa Suleymanci, a generous and thoughtful colleague and

friend, as well as other wonderful colleagues such as, Heather, Daniel, Faisal, and Professor Verghese. Finally, a brilliant research scientist at the Massachusetts Institute of Technology, Dr Thomas Heldt, has made available his support in a number of ways. Your patience, dedication, valuable advice and guidance have significantly contributed to my doctoral studies. You have been an exceptional mentor and friend; my gratitude is beyond words.

I would like to thank colleagues and professors at the doctoral program for their support; expressly, a brilliant fellow student and unique friend, Marsha Campbell-Yeo. I could not have made it without your attentive insight, understanding, advice and support. Special thanks must also go to two former PhD students and friends, Sylvie Lambert and Ananda Fernandes. You have been precious role models. I must offer my appreciation to Dr Céline Gélinas, professor and researcher at the School of Nursing. Your interest in my work and valuable insight have contributed to the successful progression of my studies. Acknowledgments are owed to my colleagues at the Montreal Children's Hospital who have always followed the evolution of my projects with much attention. Many thanks go in particular to Annik Otis, Christina Rosmus, Julie Brouillard, Dr Chantal Frigon, and Dr Etienne Goujard for their considerate support.

I am very grateful for the funding that supported my doctoral studies. The Groupe de Recherche Interuniversitaire en Interventions en Sciences Infirmières du Québec (GRIISIQ), the Fonds de recherche en santé du Québec (FRSQ), the Ministère de l'Éducation et des Loisirs (MELS), the Pain in Child Health (CIHR strategic training

initiative in health research), the McGill University Faculty of Medicine, and the Montreal Children's Hospital Foundation.

Many friends have been of invaluable support during my studies. To Olivier for always believing in me; to Katherine, Daphné, Marie-Elizabeth, José, Raoul, Geneviève, David, Jennifer, Hilde, Mathieu, the "architects", and Henri, you kept me confident and sane. I am so grateful for having you in my life. Lastly, I owe my deepest gratitude to my parents, sister and brother-in-law who have stood by me throughout this journey. To my parents, Chantale and Eric, your endless encouragements, interest in my work, and persistent trust in my competence have given me the strength to move forward and never give up on my dreams. For this and much more, I dedicate my doctorate to you.

PREFACE

THESIS FORMAT

According to the Faculty of Graduate and Postdoctoral Studies guidelines, a candidate may present a thesis as a collection of manuscripts illustrating various components of their research program. The candidate, with the approval of her committee, chose this alternative to the traditional thesis style. This dissertation includes three original manuscripts found throughout the dissertation.

The first chapter of the thesis provides an introduction to the research problem leading to the main objective statement. The second chapter presents a comprehensive review of the relevant literature that includes Manuscript #1 entitled: Cerebral Near-Infrared Spectroscopy As a Measure of Nociceptive Evoked Activity in Critically Ill Infants. This chapter closes with the presentation of the study's research objectives, questions and hypotheses.

Chapter three describes the research method utilized to meet the study objectives. The fourth chapter presents the findings of this dissertation study which are depicted in manuscripts #2 and #3. Manuscript #2 focuses on the main objective of the study and is entitled: Near-infrared spectroscopy (NIRS) to assess nociception following noxious stimulation in critically ill infants. Manuscript #3 focuses on the secondary objective of the dissertation study and is entitled: A multidimensional approach to pain assessment in critically ill infants during a painful procedure.

Chapter five provides a summary of the research findings along with a description of the study strengths and limitations. The thesis closes with the dissertation's potential contributions, future research directions and a final conclusion.

CONTRIBUTION OF AUTHORS

This dissertation is the original work of the candidate. Specifically, the candidate was engaged in the conceptualization and design of the thesis study, participant recruitment, data collection, data analysis, interpretation of findings and manuscript preparation. Committee members have provided conceptual and methodological support throughout this scholarly work. Particularly, Dr Johnston, the candidate's advisor, is recognized for her significant support throughout the entire process of the thesis. As the student's co-advisor, Dr Rennick has also contributed intellectually, as well as Dr Limperopoulos. Drs du Plessis, Heldt and Limperopoulos provided intellectual and material support during data collection and analysis in Boston. All five committee members contributed to the critical review of the manuscripts included in the dissertation.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

LIST OF APPENDICES

CHAPTER 1: PROBLEM STATEMENT

An admission to the paediatric intensive care unit (PICU) is associated with major physical, psychological and emotional stress for children and their families (Carnevale & Razack, 2002; Rennick et al., 2004). Pain contributes significantly to this distressing experience. When not well managed, pain can impact many different spheres of recovery because it puts the body in stress response mode, and the child expends valuable energy to cope with it. Pain is a signal that must be treated as equally important as any significant symptom of a critical illness. Nonetheless, the complex nature of pain can impede its specific definition, identification, and measurement.

Despite national and international efforts, guidelines, standards of practice, position statements, and many important discoveries in the field of pain management within the last three decades, some issues remain problematic. The assessment and evaluation of pain in vulnerable populations are key concerns (Dunwoody, Krenzischek, Pasero, Rathmell, & Polomano, 2008). These challenges are increased in the non communicative paediatric critically ill population. Despite the availability of over forty pain assessment tools for infants and children, no single instrument has demonstrated superiority over the others for use across painful conditions or situations. Thus no specific measure has been set as the "gold standard" for pain assessment in children (Anand, 2007; Hummel & van Dijk, 2006). Van Dijk and colleagues (van Dijk et al., 2000) have reported an increased need for reliable and valid pain assessment instruments that can be incorporated into the daily critical care routine. Within the critically ill population, a small number of pain assessment tools have been thoroughly tested using a rigorous scientific approach (Puntillo, Stannard, Miaskowski, Kehrle, & Gleeson, 2002).

In a review paper on the current status and challenges of pain assessment in infants, Hummel and van Dijk (2006) concluded that the basic element of pain management continues to be behavioural pain assessment. Instruments assessing behavioural and/or physiological markers of pain in children that have been tested in the PICU includes, the COMFORT "behavior" scale (van Dijk et al., 2000; Ambuel, Hamlett, Marx, & Blumer, 1992); the Face Leg Activity Cry Consolability scale (FLACC) (Merkel, Voepel-Lewis, Shayevitz, & Malviya, 1997; Manworren & Hynan, 2003); and the Multidimensional Assessment Pain Scale (MAPS) (Abu-Saad, Bulsara, Rees, & McDonald, 2006; Ramelet, Abu-Saad, Rees, & McDonald, 2004; Ramelet, Rees, McDonald, Bulsara, & Abu-Saad, 2007a). However, further validation studies are necessary for these instruments to be used across painful situations and populations.

Unstable and non specific physiological indicators such as heart rate, arterial oxygen saturation, and blood pressure are often relied upon by health professionals to evaluate pain in critically ill patients. These could be viewed as more "objective" than behavioural indicators which require the use of judgement. However, relying upon physiological markers can lead to misinterpretation of pain intensity since they have been shown to decrease the internal consistency of many multidimensional pain assessment instruments (Carnevale & Razack, 2002; Ista, van Dijk, Tibboel, & de Hoog, 2005; van Dijk et al., 2001; van Dijk et al., 2000). Health care professionals' abilities to assess and interpret the level of pain experienced by patients vary widely. Some may prefer to rely on physiological indicators; some may use available pain assessment tools; and others may not know how to properly evaluate pain, thereby missing important signs. Providing

clinicians with valid and reliable methods for assessing pain may amend these inconsistencies in practice and allow standardization.

Controversies in pain assessment might well be surmounted in the near future, as techniques that provide a window into the human brain allowing more direct measurement of pain perception are developed (Anand, 2007). Promising results have been reported on the use of non-invasive electroencephalography and neuroimaging techniques to measure somatosensory and frontal cortex activation (Ranger, Johnston, & Anand, 2007). Recent studies in premature and term infants indicated that painful stimuli cause circulatory and metabolic changes in specific cortical and subcortical regions of the brain (Bartocci, Bergqvist, Lagercrantz, & Anand, 2006). A non invasive technique called near-infrared spectroscopy (NIRS) can detect subtle changes in the brain's haemoglobin oxygenation and de-oxygenation status, which reflect haemodynamic activity. Recently, it has been demonstrated using NIRS that cortical activation occurs in response to painful stimuli in newborns (Bartocci et al., 2006; Slater et al., 2006; Slater, Cantarella, Franck, Meek, & Fitzgerald, 2008). Thus it would appear that there is a new avenue of assessment for populations such as premature and term infants or critically ill children that may be more sensitive and specific to pain response.

Bedside non-invasive neurodiagnostic monitoring with NIRS is showing promising results in its usefulness to detect cortical activation related to painful events (Ranger et al., 2007). Further studies are necessary to determine the utility, specificity, sensitivity, feasibility and clinical significance of novel assessment instruments during differing painful situations. These investigations would open up possibilities of establishing the psychometric properties of the available pain assessment instruments in

non-communicative, critically ill children. Not only would non-invasive neurodiagnostic techniques, such as with NIRS, verify pain perception, they may allow the identification of the most accurate or sensitive observed pain indicators in specific situations. To date, one study has demonstrated moderate correlations among behavioural pain indicators and NIRS cortical measurements in a population of newborn infants (Slater et al., 2008). Thus, the examination of correlations between higher level (cortex) pain processing, physiological signs, and behaviours would permit further validation of observational pain tools (Slater et al., 2008).

The main objective of this study is to verify the ability of NIRS to detect regional cerebral haemodynamic changes in critically ill infants during a noxious procedure. If a significant haemodynamic change is found to be present in response to noxious stimulation, further investigations will be conducted to determine whether significant correlations among cortical, physiological, and behavioural indicators (based on a behavioural pain assessment tool score) exist. If conducted, the latter analyses will allow psychometric testing of a selected paediatric behavioural pain assessment instrument in non-communicative critically ill infants using the NIRS data to establish construct convergence validation.

CHAPTER 2: LITERATURE REVIEW

The literature review covers four main topics. First, an overview of pain concepts and related themes; second, current pain assessment issues; third, near-infrared spectroscopy technology (Manuscript #1); last, instrument evaluation and psychometric testing methods will be reviewed (general methods and those related to pain measurement).

To fully comprehend the various elements involved in the pain phenomenon, as well as the related issues in its recognition and assessment within the critically ill infant population, we must first explore how pain is theoretically described. The next section will describe the advancements in the conceptualization of pain which provide the theoretical background linking the different elements involved in the stated problem and serves as a framework to support the objectives of the study.

CONCEPTUALIZATION OF PAIN

Theories and conceptual frameworks

Theoretical concepts of pain have evolved since Descartes' simple and mechanistic theory that viewed pain as a physiologic phenomenon separate from reason. Attention was placed mainly on the activity at the periphery leaving no place for the psychological aspects of the experience (i.e. meaning, attention, past experience) (Derbyshire & Osborn, 2007; Melzack, 1991; Melzack & Wall, 1965). Based on numerous clinical observations, Descartes' views were shown to be wrong leading the way to new pain theories. In 1965, Melzack and Wall published the seminal Gate Control Theory (Melzack, 1991; Melzack & Wall, 1965) which gained attention in the scientific

community and set the stage for new directions in pain research (Melzack, 1991). Melzack and Wall (1965) defined pain as a multifaceted concept that includes sensory, affective, and evaluative components. Accordingly, the *sensory-discriminative* dimension includes the characteristics linked to the temporal pattern, location and intensity of pain. The *motivational-affective* element relates to the aversive nature of the pain experience and its evoked emotions. C*ognitive-evaluative* aspects of pain are associated with how individuals interpret or evaluate pain, and the meaning assigned to the pain experience.

According to Melzack (1991), "the most important contribution of the Gate Control Theory to biological and medical science is its emphasis on central nervous system (CNS) mechanisms" (p.14). Thus, the brain is the main processor of inputs: it receives the messages, and relays them to the appropriate areas for them to be interpreted and transformed (Melzack, 1999). Melzack proposed that future research should focus on understanding brain function and how it contributes to the pain experience, as opposed to focusing on the periphery. Clinical observations and analysis of phantom limb pain led him to propose a new approach to pain, a *new conceptual nervous system* (Melzack, 1990; Melzack, 1991). In this latter conceptual model of pain, the body's various sets of neuronal connections are represented as *loops* between the thalamus and cortex as well as between the cortex and limbic system, which Melzack named the "neuromatrix". Spatial division and synaptic connections of the neuromatrix are, at the beginning, established genetically, and later shaped by sensory inputs (Melzack, 1990; Melzack, 1991; Melzack, 2001). In the neuromatrix, nerve impulses come and go in particular cyclical patterns, labeled as the "neurosignature". The neurosignature is the structure of all the synaptic links that make up the neuromatrix. Hence, what Melzack proposes is that the body is

made of a genetically designed *matrix* which generates a network of *nerve-impulse patterns* creating the sensations produced by the various sensory activities that we experience (Melzack, 1999).

As various types of sensory inputs can be relayed within the neuromatrix, the neurosignature is subdivided into specialized circuits, the "neuromodules". It is within these structures that the diverse qualities of pain have been innately produced. As such, Melzack points out that it is not required to have had prior pain experience for higher order process and awareness of this sensation. Continual neurosignatures are transmitted to different areas of the brain where they are processed and transformed into conscious messages. These signals may also activate responses such as muscle activation to produce complex motor actions. The pain process is influenced by an array of elements ranging from the neurosignature, to influences from the inner body and the brain. The neuromatrix theory integrates the multidimensional aspects– sensory, affective, and cognitive- of the pain experience and behaviours (Melzack, 1990; Melzack, 1991; Melzack, 1999; Melzack, 2001). Within this frame of reference, behavioural displays of pain are seen as neuronal outputs, that is, motor responses to a noxious stimulation, which may or may not take place depending on the state of the individual. A closer look into brain activity could provide a window into how pain inputs are first received and processed within specific cortical regions offering a direct, and perhaps, more precise indicator of pain.

The advancement in the conceptualization of pain provides theoretical evidence of cortical involvement and facilitates our interpretation of how different brain areas participate in the pain process. These latest views have set the stage for research efforts to

provide empirical support. As such, advances in non-invasive neuroimaging methodologies have revolutionized our understanding of pain processing at the cortical and sub-cortical levels. The specific role of the cerebral cortex has been extensively studied, focusing on its critical role in pain perception and allowing comparison between healthy participants and chronic pain sufferers. Cumulative evidence has permitted researchers to identify two major neuronal projection paths for pain transmission that start in the nociceptive regions of the spinal cord dorsal horn (lamina I and V) (Hodge & Apkarian, 1990). As such, these two main circuits or neuromodules, as Melzack's labeled them, transmit messages to and from specific areas of the brain by two distinct systems: (1) the lateral pain system which transmits information about the intensity, location and duration of noxious stimuli to the primary and secondary somatosensory cortices (SI and SII); and (2) the medial pain system which sends messages regarding affective and motivational dimensions of noxious stimuli to the limbic cortices (insula and anterior cingulate cortex (ACC)) (Apkarian, Bushnell, Treede, & Zubieta, 2005; Derbyshire & Osborn, 2007; Hodge & Apkarian, 1990; Treede, Kenshalo, Gracely, & Jones, 1999) (Figure 1).

In other words, when a noxious stimulus is felt by a body part, a signal is sent to the brain through two major systems, one which transmits the sensory qualities (intensity, location and duration) of the signal, and the other system which relays the affective components of the same signal. Both systems have their gateway point in the dorsal horn of the spinal cord and pass through the thalamus but from there, they are relayed to different brain areas (i.e. SI and SII for the sensory qualities; insula and ACC for the affective motivational qualities) corresponding to different processing modalities. Other

areas of the brain involved in pain processing, that is, related to evaluative cognitive elements of pain, have been identified (i.e. prefrontal cortex, amygdala, hypothalamus, hippocampus, and thalamus) but a complete analysis of the literature is beyond the scope of this review. Moreover, we must mention that recent findings and advances in cerebral pain processing have lead some researchers to reexamine this reductive and simplistic representation (Apkarian, 2008). Findings within this field of research have been systematically summarized by Apkarian *et al.* (2005; Apkarian, 2008), Treede *et al.* (1999), and Derbyshire *et al.* (2007).

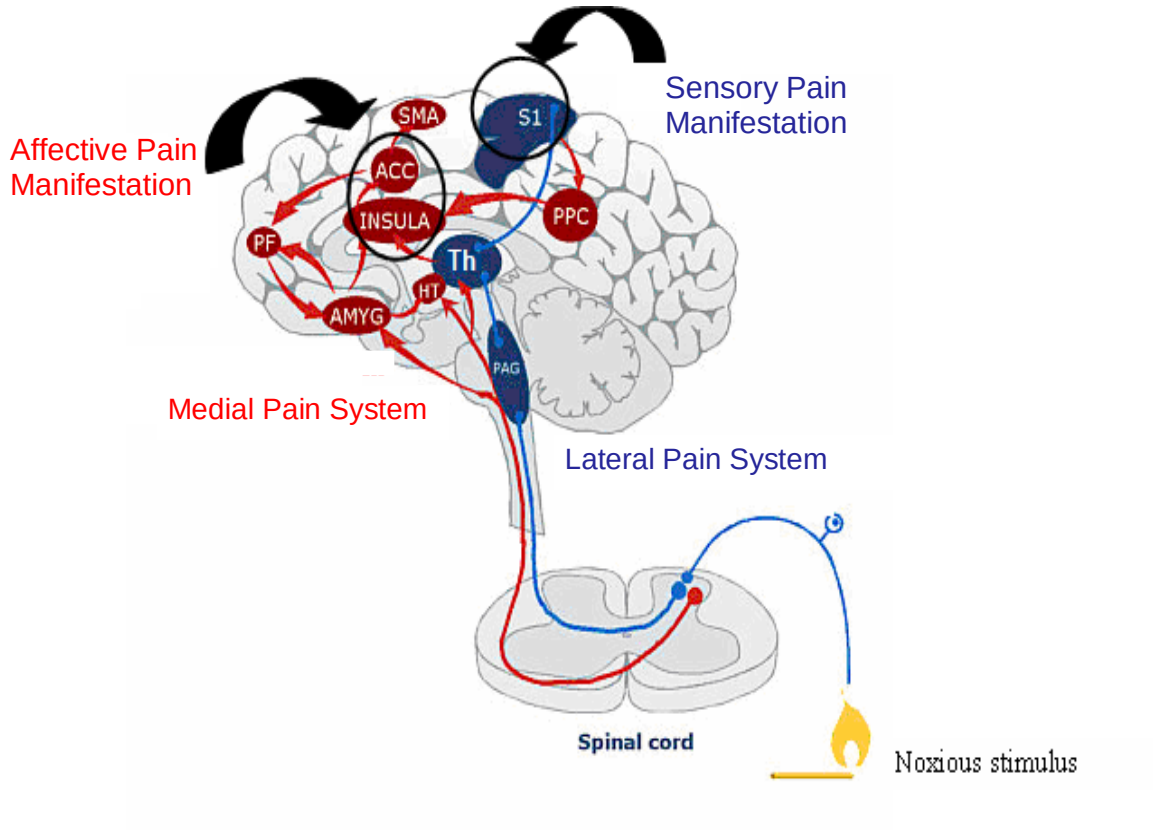

Figure 1 Pain relay systems.

Once a noxious stimulation takes place, inputs are relayed through two systems (1) the Lateral pain system sends the sensorial components qualities of the inputs to primary somatosensory cortex (SI); (2) the Medial pain system sends affective motivational components of the inputs to the insula and anterior cingulate cortex (ACC). Abbreviations: Th, thalamus; AMYG, amygdala; PF, prefrontal cortex; HT, hypothalamus; SMA, supplementary motor area; PAG, periaqueductal gray; PPC, posterior parietal cortex. Figure adapted with permission from http://spinacare.wordpress.com/category/pain-and-the-brain/

This brief overview of pain conceptualization demonstrates just how complex the experience is, and points to the numerous research possibilities emerging within this field. Increasing our understanding of the mechanisms involved is essential for developing better clinical pain management strategies. Many aspects of this multidimensional phenomenon can be studied, such as peripheral nociception, central

processing, cortical modulation, cognitive-emotional and psychological qualities, among others. As described above, certain regions of the brain involved in pain processing have been studied by various techniques that have increased our insight into particular components of this experience, such as the sensory elements. Through the use of neuroimaging, certain regions of the brain could be less easily reached than others because of their anatomical disposition (i.e. insula, ACC) and would require imaging techniques with better spatial resolution such as Magnetic Resonance Imaging (MRI) and Computed Tomography (CT) to be observed. Conversely, the primary somatosensory region, involved in the process of sensory components, can be more readily accessed because it is located at the surface of the cortex. Accordingly, activity in this brain area can be assessed with noninvasive techniques, such as electroencephalography (EEG) and near-infrared spectroscopy (NIRS). This latter optical technique will be further described in Manuscript 1, Chapter 2.

Cortical activation and pain processing

Complex emotional and motivational processing at higher cortical levels is necessary for an individual to experience pain. More primal physiological reactions such as limb withdrawal and increase in heart rate are the result of lower level responses of the nervous system, i.e. brainstem, spinal cord, hypothalamus (Slater, Fitzgerald, & Meek, 2007). Accordingly, behaviours such as facial actions and cry are valued as good proxy measures of the pain experience in those who cannot communicate verbally (Anand & Craig, 1996). Observers use these cues to quantify the internal sensation of pain as a replacement of self-report. As previously explained, it is recognized that the cerebral cortex is involved in various dimensions of pain perception such as the sensory and

affective components and that these are being analyzed in parallel by different divisions of the nociceptive system. The debate now focuses on determining the specific functions served within each cortical region. The evidence thus far supports the primary and secondary somatosensory cortices as providing a sensory discriminative function when activated by noxious stimuli (Apkarian et al., 2005; Treede et al., 1999). However, many questions remain regarding brain activity, both sensory and motor, during noxious stimuli. Observable behaviours, manifestations and reactions to pain could be related only to lower levels of spinal activity, or they may be true reflections of pain perception and awareness that involve cortical processes. While the precise level of involvement of the nervous system in relation to the behavioral manifestations of pain remains unclear, in clinical practice, behaviours remain clinicians' entrée to infants' emotions (Steed et al., 2011).

Sucrose is well known to have analgesic properties in neonates (Stevens, Yamada, & Ohlsson, 2010). A recent study published in the Lancet (Slater et al., 2010a) touched upon an important point regarding the debate between infant behavioural displays of pain and cortical evoked responses during noxious events. After receiving a treatment such as sucrose, somatosensory neuronal activity in neonates was still evident (through EEG assessment) despite dampened behavioural displays. These authors concluded that sucrose does not provide pain relief; however, based on this evidence alone, are these authors entitled to form that conclusion? Pain is a multidimensional phenomenon involving several cortical and subcortical regions and, therefore, results could indicate that behavioural displays are more related to the emotional component of pain, which would be reflected in activation of other areas of the cerebral cortex (e.g. ACC, insula,

and amygdala), which they did not assess (Lasky & van, 2010). Thus, if a treatment (especially non-pharmacological) affects the emotional aspects of the experience more than the nociceptive, can one conclude that it is not effective as a pain-relieving method? Sucrose may not significantly decrease the nociceptive activation compared to placebo but it has been shown in many studies to decrease the observed behavioural displays related to the pain response (Stevens et al., 2010). This debate could take many avenues since it could tell us that behavioural cues are not reliable indicators of nociception in infants and that the only accurate measure is through recordings of brain activity. This would serve as excellent argument to pursue in research within the field of neurodiagnostic techniques (i.e. fNIRS, EEG, etc.). However, it important to keep in mind that when these methods are used to assess pain, they do not provide a complete picture of this complex phenomenon. The term nociception would be more appropriate when reporting somatosensory cortex activation.

Although a painful stimulus usually provokes both noticeable behavioural and/or physiological cues and cortical signaling, a noxious sensation may induce only a cortical response without being accompanied by observational cues (Slater et al., 2008; Slater et al., 2007). Advances in haemodynamic and electrical brain imaging are permitting researchers to look more specifically at nociception processing in those who cannot verbalize, such as infants and the critically ill. Recently, Slater and colleagues (2008) found that pain scores rated with a multidimensional pain assessment tool, the Premature Infant Pain Profile (PIPP) (Stevens, Johnston, Petryshen, & Taddio, 1996), were significantly correlated with the haemodynamic changes in the somatosensory cortex during heel lance in infants. Facial expressions (component of the PIPP scale) was the

item that correlated most strongly with haemodynamic responses (i.e. total haemoglobin concentration measured with NIRS), compared to changes in heart rate and arterial oxygen saturation. However, these results must be interpreted with caution because clear cortical response was recorded in some infants without a corresponding change in facial expression (Slater et al., 2008).

Hence, observable cues may not always reflect cortical activation at the somatosensory level and may not accurately mirror the pain process. Johnston *et al.* (1999), showed that newborns who had undergone a recent painful stimulus were less likely to demonstrate behavioural and physiologic indicators of pain compared to those who had not had a recent noxious procedure. Nonetheless, facial expressions continue to be the strongest behavioural cue related to infants pain experiences, even in the most vulnerable and sick (Craig, Whitfield, Grunau, Linton, & Hadjistavropoulos, 1993; Stevens et al., 2003). Until we have an instrument that can accurately and persistently identify the pain process, health care workers should continue to rely on behavioural indicators, such as facial expressions, to infer the pain experience of their non communicative patients.

Definitions of pain

The International Association for the Study of Pain (IASP) (1979) defines pain as an "unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage"... "Pain is always subjective. Each individual learns the application of the word through experience related to injury in early life" (p. 250). To emphasize the subjectivity involved in the pain experience, McCaffery and Pasero (1999) proposed an alternative definition: "Pain is whatever the experiencing

person says it is, existing whenever the person says it does" (p.17). In the event of a noxious stimulation in a population of non-communicative patients, nociception may represent a more appropriate term to describe the pain phenomenon. Nociception corresponds to the cortical processing of information sent by the activation of a sensory chain reaction between neurones when tissue damage occurs (Turk & Melzack, 2001); in other words, it corresponds to the "neural process of encoding noxious stimuli" (Merskey & Bogduk, 1994). It has been suggested by Anand (1998) that it be used as a replacement to describe the pain experience when self report cannot be obtained.

Although the IASP definition has permitted important advances in the study of pain, it is not flawless. Difficulties in operationalizing the definition were discussed in an editorial by Anand and Craig (1996). Several weaknesses were exposed but ongoing progress in pain research should allow for a revised version of the definition. Furthermore, they proposed that the required self-report inherent within the IASP definition of pain could have led to subsequent failure to acknowledge and properly treat pain in infants and young children (Anand & Craig, 1996). They suggested that "behavioural alterations caused by pain are the infantile forms of self-report and should not be discounted as 'surrogate' measures of pain" (p.5).

Health care professionals are obliged to learn how to identify infants' behavioural cues and pay special attention to their unique way of communicating pain, in order to fulfill their mandate of providing pain relief and comfort. The difficult task of identifying and decoding the infantile form of self-report is complicated by the fact that the nature of these behavioural indicators will vary within each developmental stage of the infant's life (Anand & Craig, 1996; Johnston & Stevens, 1996). The fragile and immature states these

infants are in may also lessen their capability of perceiving and organizing a more regulated response at a higher level.

ISSUES IN PAIN ASSESSMENT IN THE CRITICALLY ILL INFANT

Critically ill infants and children hospitalized in intensive care units are exposed to many distressing experiences, and pain contributes greatly to this (Carnevale, 1997; Rennick et al., 2004). There are several potential sources of pain for children in the PICU, in addition to illness and surgery. Exposure to potentially painful invasive procedures can seriously impact how children internalise their encounter with hospitalization and illness (Rennick, Johnston, Dougherty, Platt, & Ritchie, 2002). Rennick and colleagues (2002) found that exposure to invasive procedures during PICU hospitalisation was a significant predictor of negative psychological responses in children up to six months post-discharge. This study reported an average of 89 invasive procedures per child (n = 60) during their PICU stay, compared to approximately 20 invasive procedures per child (n = 60) on medical and surgical wards. This report emphasizes the importance of monitoring pain exposure of children hospitalized in the PICU.

Over the last two decades, various studies examined different behavioural and physiological indicators observed in the presence of acute and chronic pain situations. Although a considerable body of knowledge now exists, the search for an observational tool that will evaluate pain with sound validity, reliability, specificity and sensitivity remains (Buttner & Finke, 2000). More recently, researchers have directed their efforts towards improving the psychometric properties of existing pain assessment instruments within various populations, types of pain and contextual factors (Stevens et al., 2007). Clinicians still do not know if the behaviours they observe are specific to pain or if they

are manifestations of other experiences such as agitation, distress, fear, stress and sadness (Buttner & Finke, 2000; Ramelet et al., 2004). This issue is even more problematic when dealing with critically ill infants.

Most of the available paediatric pain assessment instruments are multidimensional, incorporating both behavioural (facial action, body movement, cry) and physiological (heart rate, respiratory rate, blood pressure, arterial oxygen saturation) indicators. Minimal association between these two classes of indicators has been reported with an average correlation of 0.3 (Barr, 1998; Johnston, Stevens, Yang, & Horton, 1995; Stevens, Johnston, & Horton, 1993). Hence, Foster (2001) recommended that physiological indicators should be viewed as alarming signals that something is occurring, and that further investigations, such as a behavioural pain assessment, should proceed. Behavioural measures, especially facial actions, are more likely to respond selectively to pain (Craig et al., 1993), while physiological indicators change in response to painful stimuli, but also for numerous reasons not specific to pain (Johnston et al., 1999; Stevens et al., 1993; Sweet & McGrath, 1998). This makes interpretation difficult. For example, in a study of sucrose and simulated rocking, facial actions showed large differences between the conditions and favored sucrose, whereas heart rate showed smaller differences and in the opposite direction (Johnston, Stremler, Stevens, & Horton, 1997).

Facial expressions have been reported as being the most stable, sensitive and reliable cue of pain in infants (Craig et al., 1993; Johnston & Strada, 1986; Stevens et al., 2007). Stevens and others (2007) examined the factor structure of 19 pain indicators, both physiological and behavioural, in 149 infants undergoing an acute painful event. Facial

actions accounted for a greater proportion of the variance (close to 40%) with arterial oxygen saturation, heart rate, cry and heart rate variability accounting for lesser, but important, contributions of 8% to 26% additional explained variance.

Available observational pain instruments in critically ill children

Many observational pain measures for neonates, infants and young children are available. A recent review paper (Ramelet et al., 2004) found 28 pain instruments for use in children aged 0 to 3 years, eleven of which were specifically designed for preterm and term neonates. This section will not cover all of these pain measures but will focus on the most widely used behavioural pain scales, the COMFORT and the Faces Legs Activity Cry Consolability (FLACC) scales, and one more recently developed scale, the Multidimensional Assessment Pain Scale (MAPS).

COMFORT scale

A well studied and often cited "pain" assessment tool for use in the PICU is the COMFORT scale (Ambuel et al., 1992). This multidimensional measure was initially developed and tested to assess children's distress, not pain, during hospitalization in an intensive care unit. Although Ambuel *et al.* (1992) described pain as a frequent and associated phenomenon seen in the PICU, they stated that distress could be present without pain. Their conceptualization of distress differentiates this experience from pain, which contributes significantly to the operationalization of the concept to be measured. As such, the COMFORT scale was initially constructed and validated to assess distress and not necessarily pain intensity of children in the PICU.

From the time of its creation this scale has been tested and used by many researchers and clinicians to measure pain and/or sedation in acutely ill children hospitalized in the PICU (Carnevale & Razack, 2002; Ista et al., 2005; van Dijk et al., 2001; van Dijk et al., 2000; van Dijk, Peters, van Deventer, & Tibboel, 2005). Although the differences between the initial purpose of the tool and its applications are often acknowledged by researchers, the problems that may arise have never been fully dealt with. Even though the authors of the COMFORT scale went through all the necessary steps to address validation, changes in its application that have followed have not been critically examined. Given that the COMFORT scale is one of the most widely used tools to measure distress, and/or discomfort, and/or sedation, and/or pain in the paediatric intensive care setting and that there are clearly conceptual and operational inconsistencies with the use of this instrument, it is important to examine the validity of the measure as it relates to applications other than measuring distress. For example, Ramelet and colleagues (2004), questioned the validity and clinical utility of the tool by demonstrating its inability to discriminate sedation from pain. These limitations make score interpretation and clinical decision making difficult.

Validity and reliability issues related to certain components of the COMFORT scale have also been described. An item analysis of this scale in one PICU revealed limited internal consistency of the heart rate and blood pressure items (Pearson's correlations with total score HR r =0.460, BP r = 0.347; p = 0.000). Given these results, a modified version of the scale including only the more reliable behavioural items was suggested (Carnevale & Razack, 2002). A COMFORT "behaviour" scale has undergone further psychometric testing, revealing better reliability and validity coefficients (Ista et

al., 2005; van Dijk et al., 2001; van Dijk et al., 2000; van Dijk et al., 2005). Recently, the COMFORT-B scale was moderately correlated with the FLACC scale in a sample of Chinese children following cardiac surgery supporting its validity (Bai, Hsu, Tang, & van Dijk, In Press).

Face Legs Activity Cry Consolability (FLACC) scale

A unidimensional behavioural pain assessment instrument to measure pain in young children in the post-operative period was developed and tested by Merkel and colleagues (1997). The five item Face Legs Activity Cry and Consolability (FLACC) scale showed evidence of good interrater reliability (Kappa 0.52-0.82), as well as good validation scores (r^2 = .80; p <.001) with the Objective Pain Scale (Norden et al., 1991). From these preliminary results, the instrument was further evaluated with post-surgical children's self-report on the FACES pain scale (Wong & Baker, 1988) (r^2 = .58; p <.001) (Willis, Merkel, Voepel-Lewis, & Malviya, 2003) and was also shown to effectively assess pain in critically ill young children (Merkel, Voepel-Lewis, & Malviya, 2002).

The FLACC scale has the advantage of being easy to use, requiring minimal training and little time to administer. Nonetheless, some authors believe that the unidimensional aspect of this instrument limits its use because it does not measure the multiple qualities of pain. In fact, some authors have concluded that given that fact that no single measure has been shown to be "the" best indicator of pain, a multidimensional approach may be best (Sweet & McGrath, 1998).

The unidimensionality of the FLACC scale inspired the development of a new pain measure for use in critically ill young children, the Multidimensional Assessment Pain Scale (MAPS) (Ramelet et al., 2007a). Although the MAPS is still undergoing

preliminary psychometric testing, it is showing promising results. Similar to the FLACC, it is a 5 item scale with a scoring system of 0 to 10; its distinguishing feature is that it includes a vital sign indicator (heart rate and/or blood pressure). The MAPS was initially validated on 43 PICU patients aged 0 to 31 months and showed good interrater reliability, with correlations varying between *k*: 0.68-0.84. In keeping with other multidimensional pain measures, the MAPS appears to be more stable when the physiological item reflecting vital signs is removed: the overall Cronbach alpha increased from 0.68 to 0.86. Correlation with the FLACC scale and the MAPS has been demonstrated in two studies (Ramelet et al., 2007a; Ramelet, Rees, Mcdonald, Bulsara, & Abu-Saad, 2007b).

This brief overview of three observational pain assessment instruments highlights a number of unresolved issues within this field of research. Since pain is a complex and unique phenomenon, it is not surprising that one "best" observational measure equaling the gold standard of self report has not been found. New avenues in brain imaging could provide an opportunity to better understand this subjective experience. The following section describes a relatively new approach in brain functional research, which refers to a method that monitors neuronal and/or vascular response to brain evoked activation following stimulation (Leff et al., 2011).

NEAR-INFRARED SPECTROSCOPY

In this segment, the review of the literature concerning near-infrared spectroscopy (NIRS) is presented in manuscript #1. This article offers a summary of the literature relevant to the use of cerebral near-infrared spectroscopy as a measure of cerebrovascular activation in response to nociceptive stimuli in infants. Following the presentation of

manuscript #1, the literature review continues where instrument evaluation and psychometric testing methods are explained.

Manuscript #1: Cerebral Near-Infrared Spectroscopy As a Measure of Nociceptive Evoked Activity in Critically Ill Infants

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Published in Pain Research and Management (2011), 16(5), 331-336.

Preface

Manuscript #1 of this dissertation presents a summary of the literature relevant to the use of cerebral near-infrared spectroscopy as a measure of cerebrovascular activation in response to nociceptive stimuli in infants. Advantages and limitations of this neurodiagnostic technique are discussed. The aim is to provide a better understanding of this technique for researchers considering the use of near-infrared spectroscopy (NIRS) for the study of pain in infants.

CEREBRAL NEAR-INFRARED SPECTROSCOPY AS A MEASURE OF NOCICEPTIVE EVOKED ACTIVITY IN CRITICALLY ILL INFANTS

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SHORT TITLE: NEAR INFRARED SPECTROSCOPY TO MEASURE PAIN IN INFANTS

Financial Support

Conflict of Interest and Financial Disclosure – None to Declare

Ranger is supported by the Quebec Health Research Fund (*FRSQ)*, the Quebec Interuniversity Nursing Intervention Research Group (GRIISIQ), the Quebec Pain Research Network, and the Ministère de l'Éducation, Loisirs et Sports (MELS). Johnston and Rennick are also members of GRIISIQ and of the McGill University Health Centre Research Institute, which is funded by FRSQ.

SUMMARY

This article presents a summary of the literature relevant to the use of cerebral near-infrared spectroscopy as a measure of cerebrovascular activation in response to nociceptive stimuli in infants. We discuss both the advantages and limitations of this neurodiagnostic technique. Our aim is to provide a better understanding of this technique for researchers considering the use of near-infrared spectroscopy (NIRS) for the study of pain in infants.

ABSTRACT

Signs of pain may be subtle or absent in the critically ill infant. The complex nature of pain may further obscure its identification and measurement. As the use of monitoring and neuroimaging techniques has become more common in pain research, an understanding of these specialized technologies is important. Near-infrared spectroscopy (NIRS) is a non-invasive technique for monitoring tissue hemodynamics and oxygenation. There are indications that NIRS is capable of detecting the cerebral hemodynamic changes associated with sensory stimuli, including pain, in infants. These developments suggest that NIRS may play an important role in research focusing on pain perception in critically ill infants. This paper briefly describes the cortical responses to noxious stimuli which parallel cerebral hemodynamic responses to various stimuli. This will be followed by an overview of NIRS technology, including a summary of the literature on functional studies that have used NIRS in infants.

Current NIRS techniques have well-recognized limitations which must be considered carefully during the measurement and interpretation of the signals. Nonetheless, until

more advanced NIRS techniques emerge, the current devices have strengths that should be exploited.

Key words: Near-infrared spectroscopy (NIRS), pain, infants, nociception, neurodiagostic technique

Pain is a subjective symptom that, under normal circumstances, triggers a predictable set of objective signs, such as facial grimacing and increased heart rate. Since pain is a warning signal of imminent or ongoing tissue injury its behavioural manifestations signal to caregivers the need for urgent attention (Anand & Craig, 1996). However, in the critically ill infant, the typical signs of pain may be subtle or absent. Their identification and measurement based on observations of behavioural or non-specific physiological signals remain limited. The difficult task of identifying and decoding the infantile form of self-report is complicated by the fact that the nature of the behavioural signals may vary across the developmental stages of infancy (Anand & Craig, 1996; Johnston & Stevens, 1996). Furthermore, in critically ill infants their fragile and immature state may lessen their capability to organize and exhibit perceived pain as a recognizable response.

Consequently clinical researchers have explored the use of associated signals to identify pain. As the use of monitoring and neuroimaging techniques becomes more common in pain research, an understanding of their strengths and limitations is important for professionals considering their application for the study and management of critically ill infants.

To evaluate pain in critically ill infants, health care professionals often rely upon unstable and non-specific physiological indicators such as heart rate, arterial oxygen saturation, and blood pressure. These parameters could be viewed as more "objective" or quantifiable than other more qualitative behavioural indicators. However, relying on physiological markers can lead to misinterpretation of pain intensity since they have been shown to decrease the internal consistency of many multidimensional pain assessment

instruments and are not specific to the pain response (Carnevale & Razack, 2002; Ista et al., 2005; Ramelet et al., 2007a; van Dijk et al., 2001; van Dijk et al., 2000).

Inaccuracies in pain assessment might well be surmounted in the near future, as techniques that provide a window into human cerebral responses allow more direct measurement of pain perception (Anand, 2007). Recent studies in premature and term infants indicate that painful stimuli cause circulatory and metabolic changes in specific cortical and subcortical regions of the brain (Bartocci et al., 2006; Slater et al., 2006; Slater et al., 2008). Near-infrared spectroscopy (NIRS) is a non-invasive technique that can be used to detect subtle changes in the brain concentration of oxygenated and deoxygenated hemoglobin which are inferred to reflect changes in cerebral metabolism and perfusion. Recently, it has been demonstrated with NIRS that cerebral hemodynamic changes (presumably due to cortical activation) occur in response to stressful and/or painful stimuli in both term and preterm newborn infants (Bartocci et al., 2006; Limperopoulos et al., 2008; Slater et al., 2006; Slater et al., 2008).

Promising results have been reported for the use of non-invasive electroencephalography (EEG) (Slater et al., 2010a; Slater et al., 2010b; Slater et al., 2010c) and neuroimaging techniques to measure sensory input processing (Klem, Luders, Jasper, & Elger, 1999), such as in studies on somatosensory and frontal cortex activation (i.e. functional magnetic resonance imaging (fMRI) and positron-emission tomography (PET)) (Apkarian et al., 2005; Treede et al., 1999). Nevertheless, NIRS has the additional feature, as compared to MRI and PET devices, of being directly portable to the bedside which allows for continuous signal recording capable of capturing responses to intermittent stimuli. It would appear that there are new and exciting avenues for the

assessment of nociception that may be more sensitive and perhaps more specific to pain response for populations such as premature and term infants or critically ill children.

Further studies are necessary to determine the feasibility, utility, specificity, sensitivity, and perhaps most importantly, the clinical significance of these novel physiological assessment instruments in different painful conditions. Among other opportunities, this would provide possibilities for establishing the psychometric properties of currently available pain assessment instruments not requiring specialized equipment in critically ill infants. Not only would non-invasive monitoring techniques, such as NIRS, detect pain perception and the respective cortical regions involved in this experience, they may also allow the identification of the most accurate or sensitive observational pain indicators in specific situations (Slater et al., 2008; Slater et al., 2009). To date, only one published study has examined the relationship between NIRS with both physiological and behavioural indicators (i.e. three facial expressions) from a pain assessment instrument in a population of newborn infants; moderate associations were demonstrated among these variables (Slater et al., 2008).

In the present paper, we will briefly describe the cortical responses to noxious stimuli, followed by an overview of NIRS technology, and, finally, a literature review of the functional activation studies using NIRS in critically ill infants, with a particular emphasis on pain activation studies. The objective is to provide the reader with a better understanding of the technology, its applications in pain research, and potential use in the clinical settings.

CORTICAL PAIN RESPONSES

The brain is the principal processor of internal and external sensory experiences, including pain. It receives messages, and relays them to the appropriate areas for interpretation and subsequently transforms them into appropriate responses (Melzack, 1999). Many aspects of the pain experience can be studied, including peripheral nociception, central processing, cortical modulation, cognitive-emotional and psychological qualities. Our understanding of the pain experience has increased through the identification of specific cerebral regions involved in pain processing. It is well established that specific regions implicated in pain processing are located deeper in the brain than others (i.e. insula, anterior cingulate cortex) and in order to be examined advanced neuroimaging techniques with high spatial resolution such as MRI are required (Apkarian et al., 2005; Treede et al., 1999). Conversely, the primary somatosensory cortex, involved in the processing of sensory input, is located more superficially and activity in this cortical region is more accessible to examination by noninvasive optical techniques, such as NIRS.

Cortical activation and pain processing

Emotional and motivational processing at higher cortical levels is necessary for an individual to fully experience pain (Hofbauer, Rainville, Duncan, & Bushnell, 2001). More primal physiological reactions such as limb withdrawal and increase in heart rate are the result of subcortical level responses of the nervous system that is spinal cord, brainstem, hypothalamus, and thalamus (Slater, Fitzgerald, & Meek, 2007). Behaviours such as facial actions and cry are valued as good proxy measures of the pain experience in those who cannot communicate verbally (Anand & Craig, 1996). Although, these

reactions to painful events appear as a vital part of pain (Craig, Versloot, Goubert, Vervoort, & Crombez, 2010), it is unclear whether these cues are true reflections of pain perception and awareness involving cortical processes or whether they are reflexive (Slater et al., 2008). Observers use these cues to quantify the internal sensation of pain as a proxy measure and some even report them as being more honest expressions compared to self-report (Craig et al., 2010). As previously explained, it is recognized that the cerebral cortex is involved in various dimensions of pain perception such as the sensory and affective components and these are being analyzed in parallel by different areas of the nociceptive system. The question now focuses on determining the specific pain-related functions served within each cortical region and how they interact (Hofbauer et al., 2001). The role of the primary and secondary somatosensory cortices in providing a sensory discriminative functional role when activated by noxious stimuli is well established (Apkarian et al., 2005; Treede et al., 1999). Nevertheless many questions about brain activity responses to noxious stimuli remain unanswered. For example, it is not clear why some infants, in certain circumstances, do not display behavioural reactions to noxious procedures (Slater et al., 2009).

The presence of specific topographic areas of cerebral cortex dedicated to pain perception has been extensively studied (Apkarian et al., 2005; Derbyshire & Osborn, 2007). Cumulative evidence has permitted researchers to identify two major neuronal projection pathways for pain transmission from the periphery that start in the nociceptive regions of the spinal cord dorsal horn (laminae I and V). From there, impulses are transmitted to specific areas of the brain by two distinct systems, that is, (1) the lateral thalamus which transmits information about the intensity, location and duration of

noxious stimuli to the primary and secondary somatosensory cortices (SI and SII), and (2) the medial thalamic system which sends messages regarding affective and motivational dimensions of noxious stimuli to the limbic cortices (insula and anterior cingulate cortex (ACC)) (Apkarian et al., 2005; Derbyshire & Osborn, 2007; Treede et al., 1999).

Other areas of the brain involved in the evaluative cognitive elements of pain have been identified (i.e. prefrontal cortex, amygdala, hypothalamus, hippocampus, and thalamus), but a comprehensive analysis of all the anatomical areas of the brain that are associated with pain is beyond the scope of this review. Findings within this field of research have been previously summarized (Apkarian et al., 2005; Derbyshire & Osborn, 2007; Tracey & Mantyh, 2007; Treede et al., 1999).

It has been appreciated since the late 1890s in brain physiology that neuronal activity is closely linked to blood flow and metabolism (Magistretti, 2006; Magistretti & Pellerin, 1999). This founding principle has set the stage for many important discoveries and has provided the basis for functional brain imaging techniques (Magistretti, 2006; Mazziotta, Toga, & Frackowiak, 2000). Accordingly, during specific mental or behavioural tasks, changes in level of hemoglobin oxygenation in specific brain regions are used as indicators of localized brain activity which transiently change the oxy-deoxyhemoglobin levels within these regions and can be measured using functional MRI (fMRI) (Magistretti, 2006). Contrary to results seen in adult fMRI studies, young children (under 5 years of age) show an increase in oxygen consumption that outpaces the increase in blood flow after stimulation to specific regions of the brain (Marcar, Strassle, Loenneker, Schwarz, & Martin, 2004; Meek et al., 1998). This is the case for both oxygenated and deoxygenated hemoglobin increases. This phenomenon has been

tentatively explained by anatomical differences in cerebral neuronal networking in immature regions of the brain. For example, compared to older children (older than 6 years old) and adults, there is a higher number of synaptic connections in the immature visual system (Marcar et al., 2004). Thus, cerebral neurovascular coupling could differ in young children when compared to more mature neuronal networks found in adults (Sava et al., 2009).

During baseline brain metabolism, equilibrium between oxygen utilization and blood flow is present. Consequently, the cerebral oxygen extraction fraction (OEF), which can be depicted as "the percentage of the oxygen delivered to the brain that is utilized by the brain" (Gusnard, Raichle, & Raichle, 2001), is very stable and can be used as a reliable indicator to define baseline brain metabolism. In the awake resting state, brain energy requirements are quite high accounting for about 20% of the oxygen and glucose utilization of the whole body and 10% of the cardiac output (Magistretti, 1999). During brain "activation" there is a modest mobilization of energy (i.e. about 10% increase from baseline activity) characterized by increases in blood flow, glucose utilization, and oxygen delivery. However, the increase in oxygen utilization is slightly less than the increase in oxygen delivery, resulting in a relative *decrease* in OEF because the supply transiently exceeds the demand, reflected by an increase in the concentration of oxygenated hemoglobin ($[HbO_2]$) and decrease in the concentration of deoxygenated hemoglobin ([HbH]). In comparison, a "deactivation" relates to a transient increase in OEF and represents a decrease in neural activity when compared to baseline metabolism but not all decreases are deactivations (Magistretti, 2006; Gusnard et al., 2001).

These neurovascular and neurometabolic coupling mechanisms support the use of techniques that measure regional cerebral hemodynamic responses in order to increase our understanding of pain processing at the cortical level. The following section provides a description of near-infrared spectroscopy (NIRS), a non-invasive bedside technique for monitoring cerebral perfusion and oxygenation. An overview of functional NIRS research papers within the infant population is also featured with a particular focus on pain and NIRS studies.

NEAR INFRARED SPECTROSCOPY

Near-infrared spectroscopy is an optical technique based on the principle that light in the near infrared range (from 700 to 1000 nm) is able to pass through skin, soft tissue, and bone with relative ease, and to penetrate brain tissue to a depth of up to 8 cm (Bartocci, 2006; Wolfberg & du Plessis, 2006). The depth of penetration of the NIR light is dependent on the thickness and density of the tissue. Accordingly, when illuminating the somatosensory cortex area of the premature infant, the light may enter much deeper with signals penetrating the primary somatosensory cortex and parts of the secondary somatosensory cortex, insula, cingulate cortex, thalamus, and amygdale (Bartocci, 2006). In adults, because of the thicker skull, light does not infiltrate more than 5 cm from the surface. The light is mainly absorbed by two chromophores: hemoglobin and cytochrome aa_3. A chromophore is a substance that absorbs light at a given wave length (e.g. NIR light spectrum varies between around 650 and 1000 nm) and those found in living tissues are oxygenated hemoglobin (HbO_2), deoxygenated hemoglobin (HbH), and cytochrome oxidase (Cyt_{ox}) (Soul & du Plessis, 1999). Each of these chromophores has its peak NIR light absorption at a specific distinct wavelength.

The calculation of the chromophore's concentration and absorbance of NIR light in the tissue is possible by a modified Beer-Lambert law (Duncan et al., 1996). This law permits the calculation of the attenuation of a light source that passes through a given substance. When light penetrates a medium that is not homogeneous, such as living tissue, it does not simply travel from the source to the receiver but part of its propagation is scattered and lost. Thus, the distance from the light source to the receiving end is affected by a differential pathlength factor (DPF). The DPF has been calculated for various biological tissues but has also been shown to vary between participants which, may partly explain the complexity in standardizing NIRS variables across participants (Duncan et al., 1996; Wolfberg & du Plessis, 2006).

The hemodynamic signal obtained with the NIRS technique is based on the absorption of NIR light by hemoglobin, which in turn depends on the oxygenation state of hemoglobin circulating through the tissues. Thus, NIRS measures the relative *change* in the tissue concentration of intravascular oxygenated hemoglobin and deoxygenated hemoglobin (Bartocci, 2006; Duncan et al., 1996; Soul & du Plessis, 1999; Wolfberg & du Plessis, 2006).

Cerebral Hemodynamics and NIRS

Research using NIRS to study variations in cerebral oxygenation and hemodynamics of human neonates originated in the work by Brazy and colleagues in 1985 (Brazy, Lewis, Mitnick, & Jobsis, 1985). Although there have been significant advances in this field in the last decade, understanding of how blood flow, metabolism, and neuronal activity interact to affect the NIRS signals remains incomplete (Bartocci,

2006). Establishing validity of the NIRS measures has also proven difficult because few alternative technologies exist to serve as a gold standard (Wolfberg & du Plessis, 2006).

The study of hemodynamic changes to assess the functional activation in the brain is based on the assumption that a given stimulus will induce a neuronal response which in turn triggers local vasodilation, with an increase in cerebral blood volume (CBV) and cerebral blood flow (CBF) (Soul & du Plessis, 1999). Using NIRS we are able to infer changes in cerebral blood flow by measuring changes in the hemoglobin difference (HbD), which is obtained by the difference between the concentration changes in oxygenated hemoglobin and deoxygenated hemoglobin. As for changes in CBV, it largely reflects the changes in total concentration of hemoglobin or "total hemoglobin" (HbT); summation of oxygenated and deoxygenated hemoglobin (Tsuji, duPlessis, Taylor, Crocker, & Volpe, 1998; Wyatt, Delpy, Cope, Wray, & Reynolds, 1986). Under these circumstances, an increase in the relative concentration of the [HbO_2]:[HbT] ratio suggests that CBF has increased in excess of oxygen consumption, which is what is expected to happen during a noxious stimulus. This principle illustrates how neural inputs may provoke hemodynamic changes in the cortical area where the "message" is processed. On the other hand, a decrease in the relative [HbO_2] and [HbT] when compared to a baseline measure can be interpreted as a regional cortical *deactivation* (Bartocci, 2006; Benaron et al., 2000; Obrig et al., 1997; Obrig & Villringer, 1997; Obrig et al., 2000) (Table 1). Such observations have been described in a study of hemodynamic changes in the olfactory cortex following « strong odors » stimulation (i.e. disinfectant or tape remover substances) in premature infants (Bartocci et al., 2001).

Clinical application and functional NIRS studies

Near-infrared spectroscopy has been providing quantitative data on the oxygenation status of living biological tissue since the pioneering work of Jöbsis (1978) in the late 1970s. During the last two decades studies have been conducted to assess the feasibility of this technique for monitoring variations in cerebral oxygenation of patients at risk for brain damage (Bartocci, 2006; Soul & du Plessis, 1999). Studies with NIRS can be categorized in two: (1) measurement of brain activity through assessment of dynamic relative changes in regional cerebral blood flow in real time (continuous-wave-type instrument); (2) imaging of brain activity as a function of time (time-resolved instrument) (Hoshi, 2003). For further references on these NIRS technique, refer to Hoshi (Hoshi, 2003).

Many advantages of this optical technique have been described. Specifically it is a safe, noninvasive, bedside technique for exploring pathophysiological mechanisms underlying brain injury in sick infants (Wolf & Greisen, 2009). It has enormous potential as a tool for measuring cerebral hemodynamic responses to a variety of stimuli including changes in blood pressure, oxygenation, carbon dioxide, and neuronal activation. The excellent temporal resolution of NIRS makes it a potentially valuable tool for assessing various pathologies and their management. It can be adapted to many experimental and clinical situations, and combined with other electrophysiological and neuroimaging techniques (Bartocci, 2006; Soul & du Plessis, 1999; Wyatt et al., 1986). Currently it is a clinical and research tool providing anesthesiologists, neurologists, neurosurgeons, physiologists, cardiac surgeons, neonatologists, and nurses with important insights into the hemodynamic and oxygenation activity of the brain in adults and children (Soul & du Plessis, 1999). NIRS has been used in studies of brain neurophysiological development

and reactivity in the preterm and term infant (Bassan et al., 2006; Limperopoulos et al., 2008; Roche-Labarbe, Wallois, Ponchel, Kongolo, & Grebe, 2007; Soul & du Plessis, 1999; van Alfen-van der Velden et al., 2009; Wolfberg & du Plessis, 2006), as well as in intra- and postoperative cardiac infant studies (du Plessis et al., 1995; Fallon et al., 1993; Roberts et al., 1998). Regarding this last area, NIRS technology has assisted researchers in determining the complex changes in cerebral hemodynamics that persist during the early postoperative period. As such, these ongoing cerebral hemodynamic disturbances may impact upon the cerebral hemodynamic-activation coupling (Soul & du Plessis, 1999), thereby confounding the interpretation of cortical activation by noxious stimuli. Furthermore, certain treatments provided to critically ill infants may have significant effects on cerebral circulation, such as surfactant administration, mechanical ventilation, blood transfusion, surgery, hypothermia, analgesic/sedative, caffeine and indomethacin therapies (Bartocci, 2006; Lemmers, Toet, & van, 2008; van Alfen-van der Velden et al., 2009; Wolfberg & du Plessis, 2006; Yanowitz, Potter, Bowen, Baker, & Roberts, 2006). The effects of these agents on NIRS measurements need to be considered carefully during studies in this complex clinical scenario.

In the last decade, there has been an increase in brain functional activation studies in the newborn evaluating cortical activation to certain stimuli using NIRS technology, such as olfactory (Bartocci et al., 2001; Bartocci et al., 2000), visual (Meek et al., 1998; Carlsson, Lagercrantz, Olson, Printz, & Bartocci, 2008), auditory (Bartocci, 2006), tactile and pain stimulation (Bartocci et al., 2006; Slater et al., 2006; Slater et al., 2008). Brain functional activation studies have opened the door to a whole new area of research. These studies have already provided valuable insights into the functional topography of the

different components of the sensory system and this is likely to remain an important approach for future research. Bartocci and colleagues (Bartocci et al., 2006), for example, demonstrated that a noxious (i.e. venipuncture) stimulus on the hand induced a significant oxygenated hemoglobin increase ($[HbO_2]$) in the contralateral somatosensory cortex of preterm infants. Since there was no accompanying change in the occipital cortex this argued against a global sympathetically mediated response as might be occurring after sudden changes in blood pressure in infants with tenuous cerebral pressure autoregulation, or after a general state change response to pain (Roche-Labarbe et al., 2007).

Currently multichannel NIRS devices that cover multiple regions of the scalp are not readily available but their application in experimental contexts is increasing in adult and pediatric functional NIRS (fNIRS) research (Becerra et al., 2008; Hoshi, 2003; Hoshi & Chen, 2002). Recently, researchers using diffuse optical tomography (DOT) (multichannel NIRS device) were able to demonstrate a specific pain signal response in the somatosensory cortex of healthy adults after a noxious thermal stimulation (Becerra et al., 2008). As these techniques continue to advance, obstacles in their application in younger less healthy populations is likely to decrease. Multichannel NIRS devices bring increasing spatial resolution to the already excellent temporal resolution of NIRS, allowing better functional mapping and providing more insight into brain function (Hoshi, 2003). For a comprehensive review of the progress and state of NIRS instrumentation and their clinical applications in preterm and term neonates, we invite readers to refer to Wolf and Greisen (2009).

NIRS and Pain research in critically ill infants

The literature examining pain and brain functional activation studies in critically ill infants is relatively recent. To date, only three studies reporting on pain and NIRS have been published (Bartocci et al., 2006; Slater et al., 2006; Slater et al., 2008), all of which concern preterm and term neonates.

Slater *et al.* (2006) were the first to report somatosensory cortical activation following heel lance in a group of 18 newborns aged 25 to 45 weeks postmenstrual age (each newborn studied 1 to 5 times). They demonstrated a clear distinction between spinal and cortical processing, where strong reflex reactions (i.e. spinal) did not automatically correspond to more perceived pain (i.e. cortical), as demonstrated with non significant changes in the cerebral total hemoglobin concentration. Notably, these cortical activations were shown to be discriminative to pain since Von Frey hair stimulation (i.e. filaments used to measure threshold for touch evoked sensation) of the plantar surface of the foot caused a flexion withdrawal response but did *not* lead to a cortical activation. Thus, spinal reactions could serve as protective reflexes. Increases in total hemoglobin concentration in the contralateral somatosensory cortex was found following heel lance in infants as young as 25 weeks (mean Δ 7.74 μmol/L [HbT]; SE 1.10) and were independent of global hemodynamic changes. Finally, the increases in total hemoglobin concentration depended on the gestational age and awake/sleep states of the infants, with less robust contralateral cortical responses in younger neonates than older ones, and in neonates who were asleep versus awake.

Bartocci *et al.* (2006) demonstrated specific somatosensory cortical activation to venipuncture in 40 preterm neonates born between 28-36 weeks of gestation and a postnatal age of 25 to 46 hours. One group of 29 neonates was monitored bilaterally and

symmetrically overlying the somatosensory cortex. Tactile and painful stimuli were applied on both sides of the hand (half of the group receiving stimulation on the left hand and the rest received it on the right). In a second group of 11 infants the contralateral somatosensory and occipital areas were recorded allowing measurement of specificity of cerebral regions. Increases in oxygenated hemoglobin concentrations ($[HbO_2]$) in somatosensory areas in both groups occurred after skin disinfection (tactile stimulus) and an even stronger response occurred after venipuncture (noxious stimulus); in the latter group, no changes were detected in the occipital cortex. Thus, differing areas of blood flow indicated a specific response to painful stimulation in the brain. Interestingly, additional findings showed that somatosensory responses to painful stimulation measured with NIRS were more prominent in the cortex of the left hemisphere, of higher intensity in male infants, directly associated with postnatal age, and inversely correlated with gestational age.

Differences in the time sequence of examined changes in SaO_2 and $[HbO_2]$, led Bartocci *et al.* (2006) to conclude that changes in vital signs did not explain the $[HbO_2]$ augmentations noted over the somatosensory cortex in their study population. Accordingly, SaO_2 dropped following venipuncture (lasting 20-30 seconds), while the cerebral $[HbO_2]$ significantly increased and remained so for 2 to 3 minutes, well after the changes in vital signs had subsided. Furthermore, even though crying influences cerebral hemodynamics, this behavioural response started to influence cortical circulation after approximately five minutes; whereas the functional activation in the somatosensory areas appeared in the *"silent" phase* of this response, between the venipuncture and the observable indicator.

The ongoing search for a gold standard in pain assessment in non-verbal populations such as critically ill infants has provided direction for future research. A recent study examining the connection between behavioural pain indicators and cortical activation appears to be a promising approach. Slater *et al.* (2008) measured correlations between scores of the Premature Infant Pain Profile (PIPP) pain assessment tool (Stevens et al., 1993) and somatosensory cortical activation to heel lance in 12 infants aged 25 to 43 weeks (33 test occasions). Moderate correlations between the total PIPP score and level of cerebrovascular response were found (regression coefficient = 0.72, CI [0.32, 1.11] (p=0.001); r = 0.57). Among the different indicators in the PIPP, facial expressions were most highly correlated with changes in total hemoglobin concentrations (regression coefficient = 1.26, CI [0.84, 1.67] (p<0.0001); r = 0.74). Importantly, however, in some infants, cortical "pain" responses occurred without displays of change in facial expression, stressing the importance of relying on other means of pain assessment in populations with limited observable behavioural displays.

The use of NIRS to study pain perception in humans is relatively recent, and many avenues remain unexplored. Generalizability of findings from the three studies conducted to date is limited (Bartocci et al., 2006; Slater et al., 2006; Slater et al., 2008). Sample sizes were small, and possible confounding factors were not accounted for. With the exception of two premature infants receiving morphine during one study (Slater et al., 2006), there have been scant descriptions of study samples regarding diagnosis, severity of illness, co-morbidity, and medication administration. Many of these factors could affect cerebral hemodynamics which, in turn, could have altered the results. In future studies, these factors must be carefully accounted for to enhance the general applicability

of these findings. In addition, very little data were provided in the three studies conducted to-date regarding systemic physiological responses, which are important factors to consider when measuring cerebral hemodynamic responses to noxious stimuli. Only two studies (Bartocci et al., 2006; Slater et al., 2008) provided data regarding heart rate and peripheral arterial blood oxygen saturation in comparison to cerebral oxygenated and total hemoglobin concentrations changes. Most importantly, none of the studies provide blood pressure data, which is known to be related to cerebral blood flow and blood volume changes during nociceptive events in sick infants (Limperopoulos et al., 2008; Soul et al., 2007). Advances in this field will depend upon the careful measurement and analysis of other simultaneous physiologic changes that may confound the interpretation. In addition to factors that have been found to be associated with cerebral hemodynamic changes (i.e. gestational age, sex, sleep/wake state), important confounders such as sound and lighting in the NICU, as well as infants' severity of illness remain unexplored but should be accounted for in future studies.

Limitations of the NIRS technique

Near-infrared spectroscopy technology is not without limitations, some of which may be more difficult to overcome than others. The amount of NIR light reaching the detector optode is influenced not only by the concentration of the absorbing chromophores (e.g., hemoglobin) but also by the amount of photon scattering. With more scattering the light has a longer path through the tissues and, therefore, has a greater chance of being absorbed. Currently a major drawback of most available devices is the unknown path length of the NIR light within tissues. For this reason it is still necessary to apply a constant 'differential pathlength factor' to the measurements and to assume that

this path length does not change significantly during the course of a study. This remains a tenuous assumption in many clinical situations. Tables of different path length factors in brain tissues for differing participants are used, but are known to vary significantly with age (Duncan et al., 1996; Wolfberg & du Plessis, 2006). As such, measured data should be regarded as absolute *change* in hemoglobin concentration rather than absolute values (Bartocci, 2006). Another major drawback of NIRS technology is related to the difficulty in accurately identifying the exact region that is sampled by the NIR light (Hoshi, 2003). However, conducting multichannel fNIRS trials, as mentioned previously, allows for a more accurate mapping of cortical areas and improved discrimination (Becerra et al., 2008).

Near-infrared spectroscopy technology is sensitive to various factors that may confound results. Conditions related to critical illness that may result in metabolic somatosensory changes could confound pain related activation measurement using NIRS. Environmental stimuli need to be taken also into account when doing functional NIRS as these can bias the results. Patient movement can cause artefacts and disruptions in data collection. Nonetheless, this technology has the major advantage of allowing for continuous bedside monitoring of cerebral activity and hemodynamics in a noninvasive manner, which is particularly valuable for the critically ill infant. Finally, despite the fact that NIRS has some reliability issues limiting its widespread use for close clinical monitoring of cerebral hemodynamics, it can provide significant insight into the multifaceted physiological and pathological responses to stimuli (Becerra et al., 2008; Lagercrantz, 2009; Wolfberg & du Plessis, 2006). Until more advanced NIRS technologies become available, the strengths of the current devices should be exploited

and the limitations carefully considered when interpreting data generated by this unique neurodiagnostic technique.

CONCLUSION AND RECOMMENDATIONS FOR FUTURE RESEARCH

Although understanding of the multidimensional experience of pain has advanced over the last century, avenues remain unexplored, particularly in vulnerable populations such as the non-communicative patients. Near-infrared spectroscopy has potential as a technique for assessing pain evoked cerebral activation in critically ill infants. Given the complexity of NIRS technology, the paucity of research supporting its use in pain measurement in critically ill infants, and the need for tight control of many confounding factors as well as artefacts, more studies are clearly needed. At this stage it may be best to consider this neurodiagnostic technique solely as a research tool that will improve our understanding of pain perception, increase the psychometric features of currently available pain assessment instruments, and perhaps assess the efficacy of pharmacological and non-pharmacological treatments.

Determining what constitutes a clinically significant change in the measured parameters (i.e. [HbO_2], [HbH], and [HbT]) when compared with normal fluctuations that occur in the brain tissue by sampling both healthy and critically ill infants of differing developmental ages is also needed. Finally, as a variety of devices using NIRS technology are currently available for research (e.g. NIRO devices, Hamamatsu Photonics, Japan) and clinical use (e.g. INVOS® Systems, Somanetics, Troy, MI), setting standards for specific use in pain measurement could facilitate the generalization of findings.

A major question that is raised by clinicians caring for non-communicative patients is whether this technique will move beyond research to become a bedside monitoring technique for pain assessment. Moreover, can this technology help us monitor the cerebral hemodynamic changes due to prolonged pain versus acute procedural pain, and can it make this distinction. When looking at the research findings and current available devices, this may seem improbable. Nevertheless, this approach should not be abandoned since its usefulness as a portable means to functional brain mapping is evolving well and setbacks are being resolved (Becerra et al., 2008; Hoshi, 2003). If NIRS technique is validated over time as an accurate measure of pain, other issues will arise, as with all proxy measures of pain, such as determining successful analgesic response. All pain indicators for non-communicating persons require series of validation studies with replication and NIRS would be no exception.

Table 1 Summary of hemodynamic measures derived by NIRS data (Soul et al., 2007; Soul, Taylor, Wypij, duPlessis, & Volpe, 2000; Tsuji et al., 1998; Tsuji et al., 2000)

Name	**Formula**
Total Hemoglobin (Δ[HbT])	$\Delta[HbO_2] + \Delta[HbH]$
Hemoglobin difference (Δ[HbD])	$\Delta[HbO_2] - \Delta[HbH]$
Cerebral Blood Flow (ΔCBF)	$\Delta[HbO_2] - \Delta[HbH] = \Delta[HbD] \cong \Delta CBF$
Cerebral Blood Volume (ΔCBV)	$\Delta[HbO_2] + \Delta[HbH] = \Delta[HbT] \cong \Delta CBV$
Principle	If $\uparrow \frac{[HbO_2]}{[HbT]} \Rightarrow$ CBF > O_2 consumption

Data measured by the NIRS technique represent relative changes in the oxygenation state of the hemoglobin (Δ).

As it has been elucidated in manuscript #1, the use of NIRS technology to study pain processing is rather novel. Validation testing is thus necessary. To better comprehend instrument evaluation and psychometric testing methods, this last section reviews the general concepts and those related to pain measurement.

PSYCHOMETRIC TESTING METHODS

Measurement is necessary in the study of pain. It permits health care professionals to quantify and qualify the subjective experience unique to each patient. It is necessary to create a proper and complete pain management plan which includes prescribing appropriate treatment and verifying its efficacy. Moreover it enables researchers and clinicians to "examine the nature, origins and correlates of pain in children" (p.1) (Finley & McGrath, 1998). The attainment of sound psychometric qualities is an ongoing process in measurement. Every time a tool is used, tested in different settings or population or context, it is increasing its psychometric records.

The two main properties verified in classic psychometrics are reliability and validity. Reliability measures the amount of variability in scores reflecting true disparity between participants; proportion of true score reflected within the measured score (Johnston, 1998; Streiner & Norman, 2008). It demonstrates if the instrument measures the construct in a consistent and accurate manner when it is used under different circumstances, at varying points in time, and with different raters. The internal consistency of a scale refers to how well different items are connecting with the main attribute of a given instrument (Streiner, 1993). Validity corresponds to the degree to which the instrument truly measures the concept of interest. It involves not only peer judgements but empirical support to demonstrate that the tool is measuring what it is

designed to evaluate (Johnston, 1998; Streiner & Norman, 2008). There are many different approaches to reliability and validity testing in the field of psychometrics. Differentiating these essential steps can be confusing as the terminology found in the literature tends to vary among authors. Nonetheless, the objectives of the different approaches are generally the same that is, measuring the true score of the concept in question (Johnston, 1998).

In validity testing of pain instruments, determining the level of correlation between the "new" measure and established tools is essential. This important step in validation is often referred to as *concurrent* and *convergent* validation. These criteria both involve testing the instrument under study against other measures that have varying degrees of conceptual relationship with the tool in question. Where *concurrent* validation involves correlating the new measure with a "gold standard" (criterion) or another tool measuring the same construct at the same moment in time, *convergent* validation involves correlations with measures of varying degrees of theoretical relation (i.e. pain and fear; pain and depression) (Johnston, 1998). As no "gold standard" exists to measure pain in children, especially observational pain evaluation, the approach to criterion validation should be viewed more as an estimate than a precise test (Johnston, 1998).

CONCLUSION

Advancement in pain conceptualization has led the way to prolific empirical research. The use of various imaging techniques has provided researchers with means to support their theoretical propositions. Melzack's (1990; 1991) *new conceptual nervous system*, the neuromatrix, provides a sound framework for studies looking into higher

order pain processing. Blending Melzack's view with empirical research findings in neuroimaging further strengthens both avenues. In view of this, it has become evident that monitoring brain activity and more precisely, the area which reacts to the level of the physical stimulus (i.e. the somatosensory cortex) could offer a means of detecting the presence of pain when verbal expression is not possible.

Measuring pain in critically ill children is not a simple task, especially when self-report cannot be obtained. Inferring the pain level that someone else is experiencing is complex. Although a wide variety of pain measures exist for assessing pain in non communicative infants and children, no one measure has been selected as the "best measure". Various issues surrounding the evaluation of pain in critically ill children have been described in this chapter and serve as a starting point for this study.

Haemodynamic and electrical brain studies regarding pain processing are innovative and show potential for exploring these issues. More precisely, a noninvasive technology, near-infrared spectroscopy, is a method that may provide the bridge between behavioural observational indicators and cortical pain processing, thus serving as an alternative, and perhaps more specific instrument in the measure of nociception in critically ill infants.

STUDY OBJECTIVES, QUESTIONS AND HYPOTHESES

The overall objective of this study is to describe the regional cerebral haemodynamic changes in critically ill infants following noxious stimulation activation as measured by near-infrared spectroscopy (NIRS). If significant changes are measured, further investigations will be pursued to explore the relationship between the NIRS measurements, physiological indicators, and behavioural pain scores as measured by a validated pain assessment instrument (FLACC scale). Data will provide new and important information regarding the psychometric properties of the measure. Guiding the study are the following research questions and hypotheses.

RESEARCH QUESTIONS

1. Is a noxious stimulation associated with regional cerebral haemodynamic changes of critically ill infants as measured with NIRS?
2. Do regional cerebral haemodynamic (as measured with NIRS), physiological (vital signs) and behavioural (as measured with the behavioural pain assessment tool- FLACC scale) reactions significantly correlate in response to a noxious stimulation in critically ill infants?

RESEARCH HYPOTHESES

H_1: An acute noxious stimulation will be associated with significant regional cerebral haemodynamic changes in critically ill infants as measured with the NIRS technique.

H_2: Regional cerebral haemodynamic changes in critically ill infants (measured with NIRS) correlate moderately with physiological indicators (vital signs) and behavioural pain assessment scores (measured with the FLACC scale).

CHAPTER 3: METHOD

The following chapter presents the steps of the proposed study, and is composed of a description of the design, setting, sample, variables and instruments, as well as sample size requirements, procedures, and finally, the statistical analyses. Given that pain is a multidimensional phenomenon and that this study proposes to measure a specific component and neurological signal of the pain process, the term nociception will be used as a reflection of the regional cerebral activation.

Minor adjustments to the study protocol were made throughout data collection because of: (1) the heterogeneous population taking part in this study; (2) the presence of confounding factors that were difficult to control for during NIRS measurement which may have affected the interpretability of the results; and (3) the novelty of this indicator of nociception with the critically ill infant population. However, no changes to the study objectives, research questions, and hypotheses were made.

INVESTIGATOR TRAINING

Preceding the commencement of the research project, a one month training at the Children's Hospital Boston (Harvard Medical School) took place. This introduction allowed the principal investigator to become familiar with the equipment, setting, and data analysis process, as well as to identify possible issues that could preclude the proper advancement of the study.

Design

A prospective descriptive correlational design was proposed to study regional cerebral haemodynamic changes in critically ill infants following noxious stimulation

measured with NIRS. Given that measuring such changes in the brain, in this population, was largely unexplored, a descriptive design was the appropriate method to answer the main research question (Brink & Wood, 1998).

Previous research (described in manuscript 1) provided reasons to believe that relationships between cortical (measured with NIRS), behavioural (measured with a pain assessment instrument) and physiological changes may be present. As such, if a significant haemodynamic change is found to be present in response to noxious stimulation, further investigations will be conducted to determine whether significant correlations exist among cortical, physiological, and behavioural indicators.

Setting

Initially, the study was to take place in a Montreal university-affiliated children's hospital but this was found to be impossible given the lack of equipment (i.e. NIRS device- NIRO 300 and interface monitor). Consequently, the study took place at a Boston university-affiliated children's hospital where a similar research study was in progress and where the initial training took place. Given that a previous study examining the immediate cerebral oxygenation changes associated with congenital heart defect (CHD) in critically ill infants following open heart surgery using near-infrared spectroscopy was ongoing at this center, the present research was considered as an addendum to this project (i.e. same population and setting). This change in the initial protocol enabled the availability of the most accurate device setup and assured the support by a well established multidisciplinary team of specialized clinicians and researchers (i.e. Drs. A.J. du Plessis and T. Heldt).

The study took place in a 32-bed Cardiac Intensive Care Unit (CICU) at Children's Hospital Boston devoted solely to providing critical care services to children with heart conditions. This unit admits on average 1400 patients per year. Surgeons perform over 1200 cardiac surgeries per year at this hospital, with the majority of patients admitted to the CICU. This unit consist of 24 individual closed rooms, and 3 open rooms with two to three bed spaces each. As a result, the surrounding environment is quieter than a comparable open ICU setting. Noise level due to alarms and health care staff activity is kept to a minimum. During data collection the surrounding environment of each participant was kept stable and calm to prevent other stimuli from affecting the child's cerebral activity. This may be difficult to achieve in an intensive care unit where many alarms, health care professionals and other stimuli cannot be controlled. Setting up the equipment in a standardized manner and controlling the infant's environment as much as possible without intervening with the normal course of patient care during the procedure proved important, but challenging.

Sample

Infants less than 12 months of age requiring a surgical procedure for repair of a congenital heart defect (e.g. transposition of the great arteries (TGA), ventricular septal defect (VSD), coarctation of the aorta, truncus arteriosus, tetralogy of Fallot (TOF), and other right outflow tract obstructive lesions) were the selected population for this study. Infants were excluded if they were: (1) diagnosed with a spinal or peripheral nervous system illness; (2) supported by extracorporeal membrane oxygenation (ECMO) or high frequency/nitric oxyde ventilation; and (3) had severe anemia (haemoglobin less than 70 g/dl) at the time of data collection.

Description of noxious stimulus

Since most infants in the CICU have central intravenous (IV) access for fluid administration and arterial line access for continuous blood pressure measurement/blood procurement, noxious procedures such as heel lance or IV insertion rarely take place, and are unpredictable. However, postoperative cardiac patients typically have chest drains in place that are removed when chest drainage is minimal and the infant is relatively stable. Chest drain removal may or may not be carried out with prior analgesia administration and is usually planned in the daily care of patients. At the study center, the chest drains are removed by certified CICU registered nurses. Given that this procedure is highly predictable within the postoperative course and that it is a short, acute, tissue damaging painful event, it was selected as the noxious stimulus to be studied herein. This procedure has been previously studied in critically ill adults and was described as being moderately painful by patients (average pain self-report of 5 on a 0-10 numerical rating scale) (Puntillo et al., 2001). According to CICU nurses' observations of behavioural pain indicators, chest drain removal seems to be quite painful, even when patients are pre-medicated (personal informal communications).

Sample size

As previously stated, sample size calculations were based on results from previous studies. The estimation of the sample size was based on a power analysis capable of detecting a 5 micromol/litre (SD 4.5) difference in NIRS indicators between pre/post noxious stimulation; moderate effect size of 0.5. With a desired alpha of 0.05 and a desired beta of 0.10 to provide 90% power, the estimated sample size was 26

participants. To account for possible missing data and drop outs, we aimed to recruit 30 infants.

Measures

The study variables were measured using numerous methods. Cortical activity was measured with a NIRS monitor; physiological variables (i.e heart rate, mean arterial blood pressure, and arterial oxygen saturation) with a vital sign bedside monitor; and pain scores were obtained using a clinically validated pain assessment instrument according to filmed behavioural displays. The following section describes each measure.

Regional cerebral haemodynamic activity ([HbO_2] and [HbH])

The main variables related to regional cerebral haemodynamic activity following a noxious stimulation as measured with NIRS: [HbO_2] and [HbH]. The principal outcomes were the mean changes in the *absolute* concentration of oxygenated haemoglobin ([HbO_2]) and deoxygenated haemoglobin ([HbH]) during three different events, but we also examined changes in their calculated derivatives ([HbT] and [HbD]). Changes in [HbO_2] and [HbH] are measured in μmol/L; [HbO_2] and [HbH] values have been found to vary typically between -10 to 10 μmol/L (Bartocci, 2006).

NIRS monitor

To measure the regional cerebral haemodynamic changes a non-invasive tissue oxygenation monitor that uses NIRS, the NIRO 300 (Hamamastu Photonics, Japan), was used. The NIRS technology employed in this study does not measure the absolute cerebral concentrations in oxygenated haemoglobin and deoxygenated haemoglobin, but rather absolute change in concentration from an arbitrary reference value (Bartocci, 2006;

Soul & du Plessis, 1999; Wolfberg & du Plessis, 2006). We labelled the oxygenated haemoglobin concentration as [HbO_2] and deoxygenated haemoglobin concentration as [HbH]. The NIR light is produced by laser diodes and carried to and from a spectrometer via a bundle of glass fibres; these form a channel. The NIRO 300 is equipped with 2 channels formed by two optodes: emitting optode and receiving optode. It is through the emitting optode that the light is transmitted into the tissue over which it is placed; the receiving optode detects what is left of the light, that is, the residual light that was not absorbed by the tissue, and sends it back to the spectrometer. The emitter can generate NIR light approximately every 6 kHz and each pulse lasts 100 nanoseconds. The device is capable of sampling data at 1/6, 0.5, 1, 2, 5, 10, 30, or 60 seconds. In this study, data sampling was set at the fastest rate (i.e. 6 Hz- 6 samples per second).

Two channels (i.e two couples of optodes) can be used at the same time to measure different areas of the body. For this study, bilateral measurement of the cortical somatosensory areas was conducted. In theory, the closer the emitter optode is to the receiver, the less deeply the NIR light penetrates the illuminated tissue. Thus, when conducting measures to precisely explore cortical responses, it is recommended that optodes be placed 4 to 5 cm apart, and kept constant for the duration of the measure (Bartocci, 2006; Wolfberg & du Plessis, 2006). The positioning of these two optodes on the skin of the infants is safe, painless and does not cause any tissue injury. Optodes were placed in a flexible rubber holder that prevented interference from external light sources and kept the distance between the emitter and receiver optodes constant at 4 cm. This apparatus is non adhesive and was kept in place with a stretchy self-adhesive wrap (dispensed after each use). According to Bartocci and colleagues (2006), as well as other

groups conducting functional NIRS studies, placement of the optodes on the head of the participant is done according to the *international 10-20 EEG system* (Figure 2) (Bartocci, 2006).

Pain measurement

Pain was measured by two independent clinical nurse specialists (CNSs) in paediatric pain through observation of video footage (recorded during the data collection) with a clinically validated pain assessment instrument: the Face Legs Activity Cry Consolability scale. The CNSs were blinded to the purpose of the study, as well as the NIRS and physiological data. A description of the instrument and its psychometric properties follows.

FLACC (Face Legs Activity Cry Consolability) Scale

The FLACC is a unidimensional behavioural instrument developed initially to assess pain in children aged 2 months to 7 years in the post anaesthesia care unit (PACU) (Merkel et al., 2002; Merkel et al., 1997). It has been further validated in over 250 children younger than 7 years old admitted to PACU, PICU, surgical and trauma units, oncology and haematology units, and in cognitively impaired populations (Merkel et al., 2002; Voepel-Lewis, Zanotti, Dammeyer, & Merkel, 2010; Voepel-Lewis, Merkel, Tait, Trzcinka, & Malviya, 2002; Willis, Merkel, Voepel-Lewis, & Malviya, 2003). The scale is easy and quick to use. The five categories are scored from 0 to 2 resulting in a total score ranging from 0 to 10; recommended time for observation is 1 to 5 minutes or longer and interpretation of the behavioural score is provided by the authors (Merkel et al., 2002) (Appendix A).

Initial psychometric data showed acceptable interrater reliability: high correlations between two observers (in 30 children, 3 observations measured at 5-minute intervals) for the FLACC total score ($r(87)= 0.94$, $p<0.001$), and kappa values above 0.50 for each category ($k= 0.52$-0.82). There were significant positive correlations between the FLACC scores with Objective Pain Scale (OPS) scores (Norden et al., 1991) ($n=30$, $r = 0.81$, $p<0.001$). Finally, predictive validation was tested through response to analgesia. Significant differences between pre and post analgesia scores for three different time intervals (10, 30, and 60 minutes post analgesia) were reported ($n=29$, $p<0.001$ for each time interval) (Merkel et al., 1997).

Physiological measures

Vital signs, such as heart rate (HR), mean arterial blood pressure (MAP), and arterial oxygen saturation (SaO_2) were collected simultaneously with regional cerebral haemodynamic activity measurements. They were captured from the CICU bedside monitor system already in place and usually connected to the patients as standard of care (Philips Intellivue MP70, Andover, MA). The analogue signals were passed through a band-pass filter and a notch filter at 60 Hz. The band-pass filter was used for anti-aliasing, and the notch filter to exclude potential 60 Hz artifact common in the ICU setting. During measurement recording, physiological data were simultaneously transferred to a portable user interface monitor (Component Neuromonitoring System (CNS) device, Day One Medical, LLC and Moberg Research Inc, Ambler PA, USA). This is a computer-based system that continuously records, displays, stores, and analyses physiological data from multiple monitoring sources in real-time, allowing data

synchronization, display, and analysis. The NIRO 300 monitor and WebCam were also connected to the CNS device.

Control variables

Important demographic characteristics and medical data were collected as they could influence the study results. As such, these possible confounding factors were taken into account and entered in the analyses. A chart review of the study participants was conducted to collect demographic data: age, sex, weight, diagnosis, date of surgery, blood test results, co-morbidity, and time from surgery at noxious procedure. The following medical and nursing data were also collected from the charts for the 24 hour period prior to the data collection: medication received (i.e. analgesic, sedative, inotrope drugs), noxious procedures, pain scores, mean vital signs (HR, MAP, SaO_2), and ventilation status. An illness severity index for the last 24 hours preceding data collection was calculated using the Pediatric Risk of Mortality (PRISM III) (Pollack, Patel, & Ruttimann, 1996) (Appendix B).

Data collection procedure

Prior to commencement of recruitment, a meeting with the nurse manager of the CICU and other key advance practice nurses to discuss the research project took place. Following this meeting, an informal presentation about the study to the unit's nursing staff was provided; and a reminder note was placed in the communication book and emailed to all staff nurses. Following the Children's Hospital Boston Institutional Scientific Review and Ethical Board (REB) approvals (April 2009) (Appendix C), the principal investigator (PI) received the weekly cardiac surgery list and screened for

eligible patients. If potential patients were identified, the PI visited the pre-operative cardiac clinic to ask for permission to approach parents of those patients during their pre-operative visit (i.e. usually held the day prior to the surgery). As part of the screening for eligibility process, patients' charts were occasionally examined prior to talking to families, again to ensure the infant met the eligibility criteria. If authorization was granted, parents of infants meeting the inclusion criteria were approached by the PI; study goals, procedure, and potential risks were explained and consent forms were handed to them for further consultation. Questions were answered and parents were given the time they felt necessary to make their decision. When the decision had been made in favour of participation, the consent form was signed and photocopied. A copy was placed in the participant's chart, another one was given to the parents, and the original consent was kept by the PI.

During the post-operative recovery period, each enrolled patient's nurse was informed of the patient's participation in the study. A sign with study name and PI's pager number was posted at the bedside of the participant. The participating patient was followed closely during this period to assure that no chest drain removals were missed. When the painful intervention was scheduled to take place, parents and CICU staff involved with the participant were advised about the data collection procedure. The principal investigator set up the equipment (CNS monitor, NIRO 300 and WebCam) at least 30 minutes prior to the intervention to ensure the participant was calm, stable and not stimulated by the equipment when recordings were to begin. Data collection was conducted at the bedside of the patient and the staff nurse performing the procedure was asked to carry out the removal of the chest drain as usual. No interruption on the part of

the study personnel took place. Once the equipment was set up, prior to the noxious procedure, preliminary data were gathered to allow calibration and adjustments for artefacts. The two pairs of the NIRO 300 optodes were placed symmetrically on each side of the head in the bendable rubber holder and held in place with a disposable self-adherent wrap- Coban^{tm} (3M^{TM} St-Paul, MN). When possible, the emitter optode was positioned approximately 2cm below and slightly posterior to the C_3/C_4 position (according to the international EEG 10-20 system (Bartocci, 2006; Bartocci et al., 2006); as a result the receiver optode was positioned 4cm from the emitter optode allowing illumination of the primary somatosensory cortex and possibly the underlying structures (Figure 2). When this optode configuration was not possible due to hair, which causes interference with the laser signal, or other obstacles (i.e. IV indwelling catheter), optodes were placed more in the frontal or temporal brain regions. Exact placement of optodes for each participant was carefully recorded in a log book. As previously stated, the NIRO 300 monitor was set for data sampling at 6Hz (6 samples per second). Simultaneously, vital signs (HR, SaO_2, MAP) were continuously monitored and recorded. Behavioural displays during data collection were filmed on a WebCam (Orbit-Logitech); the camera was placed to allow filming of the whole body of each participant while ensuring that the facial expression was clearly visible. Pain assessment ratings were not conducted during this time.

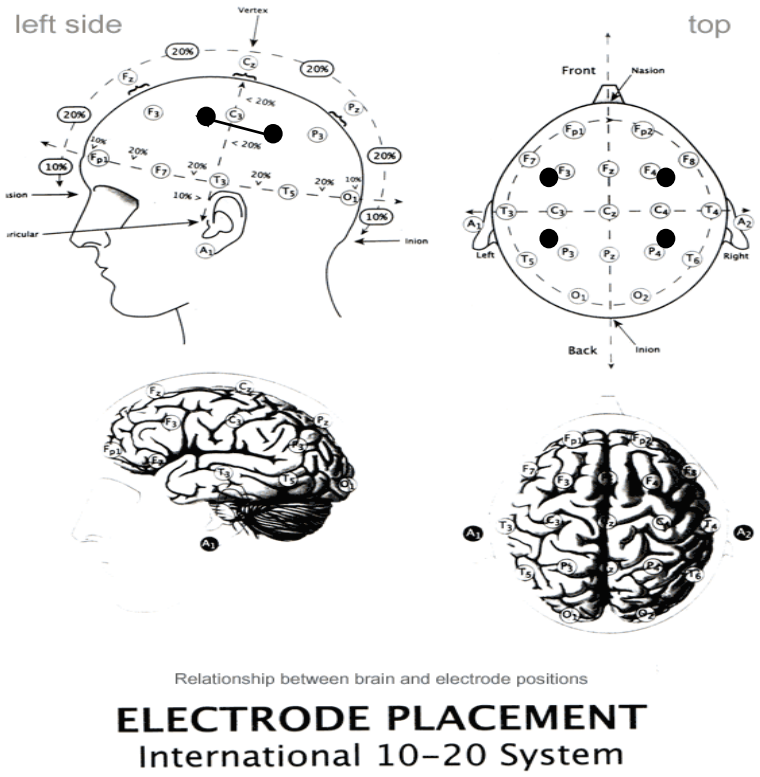

Figure 2 International 10-20 EEG system.

Used as referenced for optodes placement for NIRS functional studies; in this study the pair of optodes will be placed symmetrically near T4 and C4 (right side) and T3 and C3 (left side) to capture bilateral readings of the primary somatosensory cortex area. Black circles indicate optode placement.

All data recordings began 10 to 60 minutes prior to the drain removal and carried on for the duration of the procedure. Data extraction and analysis of the measures (NIRS, behavioural and physiological) were divided into three equal time intervals (Table 3); these were marked during recordings (i.e. excel event login sheet). Once the data measurement was completed, the principal investigator stopped all recordings and equipment was removed. Data collection (chart review) came to an end once all related information was gathered.

Video recorded behavioural displays were rated according to the recommended procedure for clinical use of the FLACC scale by a clinical nurse specialist (CNS) in paediatric pain blinded to the purpose of the study. The footages were played back in real time on a personal computer; each 30-second segment previously identified by the PI was coded for pain (Baseline, Tactile stimulation, and Noxious stimulation; always coded in this order). Thus, the coder generated three pain scores corresponding to the study time intervals, and this for each participant. A second blinded CNS in paediatric pain coded half of the sample to establish interrater reliability. This same procedure was repeated by the same coder six weeks after the first ratings were done to test intrarater reliability.

Table 2. Description of data extraction during testing period surrounding painful procedure unfoldment

Time interval	**Name**	**Duration (second)**
T_0	Baseline	30
T_1	Tactile stimulation	30
T_2	Noxious stimulation	30

Statistical analysis

Data were analyzed using descriptive, correlational, and multiple regression statistics. NIRS and physiological raw data recordings were archived to DVDs and converted into Matlab files (Matlab Student version 7.0) after the recording session had been completed. A custom-made Matlab script was used to process the recordings and analyze the data. Statistical analyses were completed using the Statistical Package for the

Social Sciences (SPSS) 15.0. Descriptive statistics were calculated for each variable and for participants' demographic/medical data.

NIRS data can be reduced and analyzed in various ways. Concentrations in oxygenated haemoglobin, deoxygenated haemoglobin, total haemoglobin, and haemoglobin difference obtained for the three time intervals can be examined as mean values for the three 30-second period, for each participant or across the sample. We were also interested in determining mean changes between these epochs (Table 4).

Table 3. Description of NIRS data reduction for analyses

	Participant*
T_0 - baseline	$\mu[HbO_2]$, μ[HbH], μ[HbT], μ[HbD]
T_1 - tactile	$\mu[HbO_2]$, μ[HbH], μ[HbT], μ[HbD]
T_2 - noxious	$\mu[HbO_2]$, μ[HbH], μ[HbT], μ[HbD]
ΔT_1-T_0	$\Delta[HbO_2]$, Δ[HbH]
ΔT_2-T_0	$\Delta[HbO_2]$, Δ[HbH]
ΔT_2-T_1	$\Delta[HbO_2]$, Δ[HbH]

*Take note that for NIRS data each signal is computed for each hemisphere (i.e. right and left).

Hypothesis 1

To verify if an acute noxious stimulation resulted in significant regional cerebral haemodynamic changes in the total sample, repeated measure ANOVA were conducted to compare NIRS data. More precisely, the mean $[HbO_2]$, [HbH], [HbT], and [HbD] at T_0 for each side (right and left hemisphere) were compared to their counterparts at T_1, and T_2. Additionally, repeated measure ANOVAs with sex as between-subjects factor to compare the differences between male and female infants were performed.

Secondary statistical procedures were conducted to further examine NIRS data. Pearson correlation coefficients (r) between the NIRS data, demographic and clinical data were calculated. Univariate linear regression analyses were executed to verify

associations between each change in NIRS signal values ($\Delta[HbO_2]$ and $\Delta[HbH]$), analgesic medication doses, age, weight, pediatric risk of mortality score (PRISM III), and time from surgery at chest drain removal. Sub-group secondary analyses were performed to evaluate cerebral haemodynamic responses depending on optode placement (somatosensory area versus frontotemporal or temporoparietal); repeated measure ANOVAs with optode placement as between-subjects factor.

Hypothesis 2

Pearson correlation coefficients (*r*) between the changes in NIRS data, physiological measures, and pain scores were calculated. When associations were present, univariate linear regression analyses were executed to further examine between each changes in NIRS signal values ($\Delta[HbO_2]$ and $\Delta[HbH]$), systemic physiological responses (ΔHR, ΔMAP, ΔSaO_2), and pain scores from the behavioural pain scale (and each component of the instrument).

Psychometric testing of pain scale

The pain scores obtained through ratings of the video recordings taken during the painful procedure were used to verify certain psychometric properties of the FLACC scale. Among the two main properties verified in classic psychometrics, the following analyses were conducted to test five important criteria:

Rater Reliability:

- Internal consistency of the FLACC scale using Cronbach's alpha (α) aiming for results between 0.70 and 0.90 (Nunnally, 1978; Streiner & Norman, 2008).

- Interrater using Inter-Class Coefficient (ICC) and aiming for agreement between the two raters to be above 0.80 (minimum 0.60) (Streiner, 1993).
- Intrarater (Intra-Class Coefficient) correlation between two scores by the same rater, but conducted at two different times (initial time and 10 weeks later); ICC should be above 0.80 (Streiner, 1993).

Validity:

- Construct (convergent) validation where NIRS data were tested against FLACC scale scores through bivariate correlations (Pearson correlation coefficients) (Johnston, 1998).
- Responsiveness or sensitivity to change to evaluate the capacity of the NIRS and the FLACC behavioural pain scale to provide different results between noxious versus non noxious (baseline and tactile) stimuli using repeated measure ANOVAs (Streiner & Norman, 2008). As such, measures obtained during baseline and tactile stimulation periods were compared to the ones obtained during the noxious stimulation time.

ETHICAL CONSIDERATIONS

The study protocol first received scientific and ethical approvals from both McGill University Health Center and the Montreal Children's Hospital Research Ethics Board (REB) (Appendix E). Given that the research took place at the Children's Hospital Boston (CHB), institutional review board approval was also obtained at this center (Appendix C). Within the preoperative cardiac clinic setting of the CHB, informed consent was obtained from the participant's parent(s) or legal guardian. The research objectives were explained, each step of the study detailed, and the benefits, risks,

participants' rights and confidentiality matters reviewed. In any paediatric research, special considerations must be taken into account. As stated by the Research Ethics Office of the Research Institute of the McGill University Health Center: " In Quebec all health research conducted with children is subject to special provision under the law due to the vulnerable standing of these minor persons who have not attained the age of majority (18 years old for the purpose of research). Only REBs that have been designated by the Minister of Health may review and approve research involving legally incompetent persons, a category that includes minor children. The Pediatric REB maintains the appropriate ministerial designation in compliance with Article 21 of the Quebec Civil Code." (Research Institute of the McGill University Health Center, 2008; Éditeur officiel du Québec, 2008).

The study posed minimal risk to participants. The use of additional equipment (NIRS monitor) may have been uncomfortable for the infant during data collection. However, the positioning of the optodes on the child's head was completely painless and did not provoke any tissue damage. The average power output from the emitting optode was around 1 mW and the irradiation intensity to the participant from this source is kept at the Class 1 level according to the International Electrotechnical Commission 60825-1 standard (Bartocci, 2006; Hamamastu Photonics, 2003; International Electrotechnical Commission, 2007). Class 1 refers to lasers that have an output power that is kept below the point at which eye injury can occur. Although a WebCam was used, this did not cause discomfort to the participant, and the footage obtained was to be used only for the purposes of this study. Care providers could not be identified through this footage and so this should not pose a threat to them. As previously stated, during data recordings, the

principal investigator did not interfere in any way with the procedure taking place, thus disruption or alteration of "normal" care was kept to a minimum.

Certain strategies were implemented to minimize risks related to confidentiality issues. Participants' names were not recorded except on consent forms and study code list. A random and unique code was assigned to each infant at the point of enrolment. The link between the infant and this study code was kept in a password protected computer file accessible only by the researchers involved with the study. All hard copy data were stored in a locked file in the Neurology Department, Children's Hospital Boston, which will be kept for 25 years. The PI and co-supervisors are the only ones who will have access to the data. The results of this study have been and will be published and presented at professional meetings. No information has been or will be disclosed that could reveal the infant's identity. Films have only been used for the purpose of this study and have been viewed only by members of the research team. It may be possible to identify the participants as their faces were captured. The films have not and will not be used for publications, presentations or any other purpose.

Infants' parents were made aware of their right to withdraw at any point in the course of the study. Three different consent forms received approval from their respective review boards: two for the Montreal Children's Hospital (English and French- Appendix F) and one form for the Children's Hospital Boston (Appendix D), although the two former forms were never used to consent parents. As stated previously, the present research was considered an addendum to a study that took place at the Children's Hospital Boston (*Development of a technique for continuous measurement of cerebral venous oxygen saturation and cerebral oxygen extraction*). Thus, the consent form relates

principally to the primary project to which this research was added. Information concerning the rights of study participants was provided in all consent forms. This study had no direct benefit for participants and no compensation was provided.

CHAPTER 4: RESULTS

The findings of this dissertation study are presented in manuscripts #2 and #3. Manuscript #2 focuses on the main objective of the study and is entitled: Near-infrared spectroscopy (NIRS) to assess nociception following noxious stimulation in critically ill infants. This article describes the haemodynamic changes in the somatosensory cortical region following noxious stimulation activation in critically ill infants as measured by near-infrared spectroscopy (NIRS); the relationship between NIRS measurements and systemic physiological parameters are also presented. Manuscript #3 focuses on the secondary objective of the dissertation study and is entitled: A multidimensional approach to pain assessment in critically ill infants during a painful procedure. This article reports on the associations between cerebral haemodynamic changes, systemic physiological changes, and behavioural pain scores of critically ill infants during a routine painful procedure.

Manuscript #2: Near-Infrared Spectroscopy (NIRS) to Assess Nociceptive Following Noxious Stimulation in Critically Ill Infants

Authors: Manon Ranger, N, PhD candidate; Celeste C. Johnston, N, DEd, FCAHS; Janet E. Rennick, N, PhD; Catherine Limperopoulos, PhD; Thomas, Heldt, PhD; & Adre J. du Plessis, MBChB, MPH.

Submitted, revised, and re-submitted for publication to the European Journal of Pain (Appendix G)

Preface

Manuscript #2 of this dissertation is the first of two on the results of the study. This first article describes the regional cerebral haemodynamic changes in critically ill infants following noxious stimulation activation as measured by near-infrared spectroscopy (NIRS), which is the overall objective of the study. Precisely, it reports the recorded changes during three distinct epochs (Baseline, Tactile stimulation, and Chest-drain removal) in cerebrovascular haemoglobin concentrations (by NIRS) as well as heart rate (by ECG), systemic arterial oxygen saturation (by pulse oxymetry), and mean arterial blood pressure (by arterial line). As the noxious stimulation resulted in significant regional cerebral haemodynamic changes, further analyses were possible to evaluate the associations between cerebral and systemic haemodynamic changes. This partially addresses the second objective of the study dissertation.

Near-Infrared Spectroscopy (NIRS) to Assess Nociception Following Noxious Stimulation in Critically Ill Infants

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ABSTRACT

To optimize pain relief, a sound pain assessment tool is essential. Although this need is recognized, only a few pain assessment instruments have been thoroughly tested with a rigorous scientific approach for critically ill infants, and none have been shown to be superior to the other. Studies using near-infrared spectroscopy (NIRS) in neonates indicated that nociceptive stimuli are associated with haemodynamic changes in specific cortical regions. This approach may provide a more sensitive and specific technique for assessing and quantifying pain processing in critically ill infants. The objective of this study was to use continuous NIRS to describe cerebrovascular and systemic haemodynamic changes in critically ill infants during chest-drain removal following open-heart surgery for congenital heart defects (CHD). In addition to NIRS, we employed physiological monitoring of heart rate, systemic blood oxygenation, and mean arterial blood pressure. The relationship between NIRS measurements and systemic physiological parameters was examined along with the influence of specific clinical factors. In a sample of 32 infants (<12 months) cerebral deoxygenated haemoglobin concentrations significantly differed across the epochs ($p<.01$). Cerebral blood volume and blood flow, as assessed by NIRS, differed also significantly ($p<.05$). Physiological systemic responses were not found to be associated with the cerebral haemodynamic parameters. Pharmacological treatments significantly affected cerebral and systemic haemodynamic responses. In critically ill infants noxious stimuli may be associated with cerebral haemodynamic changes, independent of systemic haemodynamic changes, despite of pain medication. These findings suggest that NIRS is a potential technique for assessing pain evoked cerebral activation.

1. INTRODUCTION

Despite wide-ranging efforts over the last three decades to increase knowledge and improve practice in the field of pain management, the assessment and evaluation of pain in vulnerable populations remain key concerns (Dunwoody et al., 2008). This is particularly true in the non-communicative paediatric critically ill population. Despite the availability of over forty pain assessment tools for infants and children, no single instrument has demonstrated superiority over the others for use across painful conditions or stimuli (Anand, 2007; Duhn & Medves, 2004; Ranger et al., 2007). Although behavioural cues remain the primary means by which to evaluate pain in non-verbal populations (Hummel & van Dijk, 2006), health professionals working with critically ill children are often forced to rely on non-specific physiological indicators such as heart rate, arterial oxygen saturation, and blood pressure to assess pain. Relying upon physiological markers can lead to misinterpretation of pain intensity since they have been shown to decrease the validity of many multidimensional pain assessment instruments (Carnevale & Razack, 2002; Ista et al., 2005; Ramelet et al., 2007a; van Dijk et al., 2001; van Dijk et al., 2000).

Given the known limits of pain measures based on behavioral and physiological observations and the growing interest in the mechanisms involved in cortical functions in the brain, research on pain has focused more recently on brain activation in response to noxious stimuli through various measurement techniques such as near-infrared spectroscopy (NIRS) (Bartocci et al., 2006; Slater et al., 2006; Slater et al., 2008) and electroencephalography (EEG) (Fabrizi et al., 2011; Slater et al., 2010a; Slater et al., 2010b; Slater et al., 2010c). Recent studies in premature and term infants indicate that

painful stimuli are associated with haemodynamic changes in specific cortical regions of the brain (Bartocci et al., 2006; Slater et al., 2006; Slater et al., 2008; Slater et al., 2010c). Interestingly, Limperopoulos *et al*. (2008) demonstrated that even routine care-giving in critically ill preterm infants is associated with major cerebral and circulatory fluctuations. Although these studies have increased our understanding of brain response to various events in the preterm infant, the majority of the research on stress and pain has focused on preterm and term neonates, overlooking other critically ill paediatric populations.

In this study we examined cerebrovascular and systemic haemodynamic changes in critically ill infants with congenital heart defect (CHD) following open heart surgery during a specific routine noxious event. We assessed the infants' cerebrovascular and cardiovascular changes using continuous NIRS, as well as usual physiological parameters including heart rate, systemic blood oxygenation, and mean arterial blood pressure. We hypothesized that an acute noxious stimulation (i.e. chest drain removal) would result in significant haemodynamic changes (detectable by NIRS) in the somatosensory cortex of critically ill infants. Further analyses were conducted to examine within and between-subjects differences, as well as associations between cerebral haemodynamic changes, systemic physiological changes, and specific clinical variables.

2. METHODS

2.1. Study population

Forty infants less than 12 months of age who required a surgical procedure for repair of a CHD, who necessitated a chest drain post-operatively, were prospectively recruited between April 2009 and May 2010. Infants diagnosed with a spinal or

peripheral nervous system illness were excluded from the study. The research protocol was approved by the institutional review board at Children's Hospital Boston, USA. We obtained parental written informed consent in all cases, and 89% of those approached to participate agreed to do so.

2.2. Instrumentation

The infants' cerebral haemodynamic and associated physiological signals were continuously monitored and recorded for approximately 30 minutes before and after the chest drain removal.

A non-invasive NIRS-based tissue oxygenation monitor (NIRO 300, Hamamastu Photonics, Japan) was used to measure changes in cerebral tissue oxygenation. This NIRS device has two channels, each consisting of an emitting and a receiving optode. When possible, the emitter optode was positioned approximately 2 cm beneath and slightly posterior to the C_3/C_4 position (according to the international EEG 10-20 system) (Bartocci, 2006; Bartocci et al., 2006). As a result, the receiver optode was positioned 4 cm above the emitter optode allowing interrogation of the primary somatosensory cortex and possibly the underlying structures. When this optode configuration was not possible due to hair coverage or other obstacles (i.e. intravenous indwelling catheter), optodes were placed in the frontoparietal or temporoparietal regions. Optodes were placed in a flexible rubber holder that prevented interference from external light sources and kept the distance between the emitter and receiver optodes constant at 4 cm. This configuration was held in place on the participant's head with a disposable self-adherent wrap- Coban^{tm} (3M^{TM} St-Paul, MN).

The cerebral oxygenation signal obtained with the NIRS technique is based on the absorption of near-infrared light by haemoglobin, which in turn depends on the oxygenation state of haemoglobin circulating through the tissues. The NIRS technology employed in this study does not measure the absolute cerebral concentrations in oxygenated haemoglobin ([HbO_2]) and deoxygenated haemoglobin ([HbH]), but rather absolute change in concentration from an arbitrary reference value (Bartocci, 2006; Soul & du Plessis, 1999; Wolfberg & du Plessis, 2006). From these measures we derive the haemoglobin difference ([HbD] = [HbO_2] – [HbH]) and total haemoglobin concentration (HbT = [HbO_2] + [HbH]) (Tsuji et al., 1998; Wyatt et al., 1986). Under certain physiological conditions, [HbD] and [HbT] are related to changes in cerebral blood flow and cerebral blood volume, respectively (Soul et al., 2007; Soul et al., 2000; Tsuji et al., 1998; Tsuji et al., 2000). All four cerebral parameters ([HbO_2], [HbH], [HbD], and [HbT]) were included in the analyses (see Section 2.5). The NIRS device allows for data sampling at 6 Hz.

Systemic arterial oxygen saturation (SaO_2), the electrocardiogram (ECG), and arterial blood pressure data were recorded continuously from the patient's bedside monitor (Philips Intellivue MP70, Andover, MA). An indwelling arterial line placed either in the femoral or radial arteries provided continuous monitoring of the arterial blood pressure waveform, from which mean arterial blood pressure (MAP) was computed. In a sub-sample of infants, we simultaneously captured behavioural responses and these results are reported elsewhere (Ranger et al., under review).

2.3. Data collection

The analogue signals were passed through an anti-aliasing filter and a notch filter at 60 Hz to exclude potential 60 Hz power-line interference common in the ICU setting. During measurement recording, cerebral and systemic physiological data were simultaneously streamed to a portable monitor (Component Neuromonitoring System, Day One Medical, LLC and Moberg Research Inc., Ambler PA, USA), which is a computer-based system that continuously records, displays, stores, and analyzes in real time physiological data from multiple monitoring sources, allowing data synchronization, display, and analysis.

Material set-up and data acquisition were completed by the same study investigator (MR) who was continuously present during the data-acquisition process to document in the patient's study file the precise beginning, end, and nature of all events (e.g. study events, administration of medication, handling by care provider or parent, etc.) that occurred throughout the study.

2.4. Chest-drain removal procedure

Data collection was carried out in the cardiac intensive care unit (CICU) at the patient's bedside, and the nurses who performed the chest-drain removal were asked to carry out the procedure as usual. Once the equipment was set up, preliminary data were gathered prior to the noxious procedure to allow calibration of the NIRS device and adjustments for artefacts. Each procedure was performed according to a standardized care plan. The chest-drain removal procedure comprised a sequence of steps and a variety of sensory stimuli, including administration of analgesics and/or sedatives (e.g. morphine and/or midazolam) for the majority of cases, removal of the dressing, untying of purse-string sutures around the exit site, removal of the drain at the height of expiration, and

pulling tightly on the purse string to allow closure of the exit site, which was immediately followed by application of a pressure bandage. The duration of each step, and, therefore, the entire procedure varied somewhat from subject to subject. All data recordings began 10 to 60 minutes prior to drain removal and carried on beyond the duration of the procedure.

Data analysis was divided into three 30-second intervals (epochs); these were precisely marked during recordings by the study investigator on an Excel event log sheet. Once the measurements were completed, all recordings were stopped and the equipment was removed.

2.4.1. Data selection

Three 30-second epochs representing quiet baseline (T_0), tactile stimulation (T_1) (removal of dressing), and removal of chest drain (T_2), were examined. The beginning of the baseline (T_0) period, when the infant was calm and not disturbed, was captured approximately 25 minutes prior to drain removal. About 5 minutes prior to the removal of the chest drain, a period of tactile stimulation (T_1) which was defined by removal of the dressing and preparation of the procedure took place. The epoch corresponding to the noxious stimulus response (T_2) was signaled by the unit nurse as soon as the tube began to be pulled on peak expiration by the patient. Although infants were kept quiet and as still as possible by the staff nurses during the T_1 and T_2 events, participants were moving their head and limbs.

2.5. Data pre-processing and statistical analysis

For ease of data handling, the physiological signals were down-sampled to 20 Hz prior to further analysis. From the raw data, we computed average [HbO_2], [HbH],

[HbD], [HbT], HR, MAP, and SaO_2 values for each 30-second epoch. For ease of naming, we will refer to each variable by its name without mentioning "mean", even though the reported variables correspond to the computed mean of each signal over the 30-second epochs. We also computed average changes for each signal ($\Delta[HbO_2]$, Δ[HbH], Δ[HbD], Δ[HbT], ΔHR, ΔMAP, and ΔSaO_2) between the different event epochs: Tactile vs Baseline (T_1–T_0), Noxious vs Baseline (T_2-T_0), and Noxious vs Tactile (T_2-T_1).

A custom Matlab script was used to process the raw recordings and analyze the data (Matlab Student version 7.0, The Mathworks, Natick, MA). To determine whether chest-drain removal procedures induced statistically significant cerebral and systemic haemodynamic changes compared to the other epochs, a repeated measure ANOVA was conducted to compare $[HbO_2]$, [HbH], [HbT], [HbD], HR, MAP, and SaO_2 responses between the epochs. Sex was added as a between-subjects factor. Univariate linear regression analyses were used to verify associations between each change in NIRS signal values ($\Delta[HbO_2]$ and Δ[HbH]), systemic physiological responses (ΔHR, ΔMAP, ΔSaO_2), analgesic medication doses, age, weight, pediatric risk of mortality score (PRISM III), and elapsed time since surgery of chest-drain removal. Sub-group secondary analyses were performed to evaluate cerebral haemodynamic responses depending on optode placement. Statistical analyses were performed using the Statistical Package for Social Sciences (SPPS) version 15.0 (IBM, Somers, NY); *p*-values of less than 0.05 were considered significant, and Bonferroni correction was used to adjust for multiple comparisons. Mauchly's test was used to verify the assumption of sphericity, and if

violated, degrees of freedom were corrected using Greenhouse-Geisser estimates of sphericity.

3. RESULTS

3.1. Population

Families of 45 infants were approached in the pre-operative cardiac clinic to participate in the study. Forty infants were enrolled, and 32 (18 males; 56%) participated within the immediate post-operative period. Eight recruited participants could not be studied because chest drain removal was missed (n = 6) or the patient was transferred to another unit prior to procedure (n = 2). Not all 32 recorded data sets could be used in the analysis due to equipment issues, loss of signal, or excessive noise in the recordings. Twenty-seven NIRS data sets for left hemisphere and 28 for right hemisphere were analyzed, 29 for HR, and 23 for SaO_2. Twenty infants still had an indwelling arterial catheter allowing for continuous MAP monitoring. Infants' characteristics are presented in Table 1.

All patients underwent cardiopulmonary bypass during corrective surgeries for congenital heart defects. Eight children had a co-morbid condition, with the most common being Trisomy 21 in six patients (4 males; 2 females), one had a connective tissue disorder and one had a syndrome associated with digital abnormality not well characterized otherwise. At the time of chest-drain removal, 15 out of the 32 participants required respiratory support from a conventional ventilator (n = 13) or CPAP (n = 2). A review of the participants' chart for the last 24 hours prior to removal of chest-drain showed that the mean maximum reported pain score on the FLACC instrument (Merkel

et al., 1997) was 4.6 (scale from 0 to 10). Cortical somatosensory placement of optodes was not achieved in 9 participants because of hair or access issues; in these cases, optodes were placed in the frontotemporal and temporoparietal regions. Analysis of cerebral haemodynamic data was carried out on the entire sample, as well as with a sub-group that had somatosensory cortex optode placement (n = 23).

3.2. Cerebral NIRS changes

In response to chest drain removal, significant bilateral increases in regional cerebral [HbH] were found in critically ill infants (left hemisphere (F(2, 52) = 5.51, p = .007); right hemisphere (F(2, 54) = 7.28, p = .002) (Figure 1).

Total haemoglobin [HbT] in the right hemisphere differed significantly between the 30-second epochs (F(1.64, 44.26) = 4.62, p = .02) (Supplementary Figure S1). Left hemisphere haemoglobin difference [HbD] differed significantly between the 30-second epochs (F(2, 52) = 3.98, p = .03) (Supplementary Figure S2).

No significant changes were found in bilateral cerebral regional oxygenated haemoglobin [HbO_2], left hemisphere [HbT], and right hemisphere [HbD]. Table 2 shows the results of our analyses (Supplementary Table S1 shows median (IQR) of NIRS data across 30-second epochs).

3.2.1. Sex differences

The sex of the patient was a determining factor in the response direction of the bilateral cerebral regional [HbO_2] but not in [HbH]. In response to chest drain removal we observed bilateral increases in [HbO_2] from baseline in female infants (n = 13): mean change 0.97 μmol/l (SE 1.76) left hemisphere and 4.04 μmol/l (SE 2.19) right hemisphere. Males showed bilateral decreases in [HbO_2]: mean change -2.36 μmol/l (SE

2.34) left hemisphere (n = 14), and -0.88 μmol/l (SE 0.9) right hemisphere (n = 15) (Supplementary Figures S3 and S4). Additionally, females showed a significantly higher change across the epochs in the left hemisphere [HbO_2] ($F(2, 25) = 4.49$, $p = .04$) when compared to male infants.

3.3. Systemic physiological changes

In response to chest drain removal, we found significant increases in systemic MAP ($F(1.47, 27.99) = 15.05$, $p < .001$) and heart rate (HR) ($F(1.31, 36.64) = 9.47$, $p = .002$) in our sample of critically ill infants. We did not find significant differences in the SaO_2. Results are shown in Table 2.

3.4. Changes in NIRS and physiological signals across 30-second epochs

Univariate analyses showed that only changes (Δ) in left hemisphere Δ[HbH] were inversely associated with changes in ΔSaO_2. No other NIRS signal change across the three 30-second epochs was found to be significantly correlated with physiological signals. Table 3 shows those relationships that achieved statistical significance.

3.4.1. Effects of medications

The dose of analgesic medication (morphine in mcg/kg/dose) administered was inversely associated with changes in the left hemisphere $\Delta[HbO_2]$. Thus, administering morphine explained 26% ($R^2 = .26$) of the variance in the response to noxious stimulation and 19% ($R^2 = .19$) to tactile stimulation in the left hemisphere [HbO_2]. When we performed the same analysis for male and female infants separately, we found that the administered doses of morphine were associated with changes in the left hemisphere $\Delta[HbO_2]$ in females only; explaining 40% of the variance ($R^2 = .40$) (Table 3).

Administration of morphine was not found to correlate significantly with any of the changes in the other parameters.

The dose of sedative medication (midazolam in mcg/kg/dose) administered was inversely associated with changes in HR and SaO_2, explaining 20% (R^2 = .2) of the variance in HR and 25% (R^2 = .25) (Table 3). Administration of midazolam was not associated with cerebral haemodynamic changes and other physiological signal changes.

3.4.2. Effects of demographic and clinical data

Age of infants was found to be significantly associated with changes in the right hemisphere $\Delta[HbO_2]$, while it had an inverse association with changes in left hemisphere Δ[HbH]. In all of these instances age explained approximately 20% of the variance in the responses (R^2 between 0.20-0.21). Age was significantly correlated with ΔMAP and the explained variance was 39% (R^2 = 0.39). Infant's age was not associated with any other cerebral and physiological systemic haemodynamic change (Table 3). Infants with the comorbid condition Trisomy 21 or who were supported by a ventilator at the time of the study did not differ significantly in their cerebral and physiological haemodynamic response when compared to the rest of the sample. PRISM III score and time from surgery to chest-drain removal were not associated with any of the cerebral and physiological haemodynamic changes.

3.5. Somatosensory cortex optode placement sub-group analyses

We proceeded with secondary analysis in the sub-group of infants (n = 22) for whom optode placement was on the somatosensory cortex. Bilateral [HbH] differed significantly between the different 30-second epochs (left hemisphere p = .003; right hemisphere p = .035). This sample did not significantly differ from the total sample of

infants in regards to age, weight, PRISM III score, and doses of analgesic/sedative medication.

In this sub-group of infants, the dose of morphine administered produced a similar effect on cerebral haemodynamic indicators as in the total sample (Supplementary Table S2). We found that administering morphine explained 22% (R^2 (T_2-T_0) = .22) of the variance in the response to tactile and noxious stimulation in left hemisphere [HbO_2]. For comparison purposes, the remaining infants that had their cerebral NIRS recordings in the frontoparietal or temporoparietal regions showed a stronger inverse association to morphine administration and explained 59% (R^2 = .59) of the variance in the response to noxious stimulation in left hemisphere [HbO_2] (Supplementary Table S2).

4. DISCUSSION

We demonstrate varying cerebrovascular and systemic haemodynamic changes in response to chest-drain removal in critically ill infants with CHD following open heart surgery. To our knowledge, this is the first study that describes continuous cerebral haemodynamic responses coupled with systemic cardiovascular activity (continuous systemic MAP and SaO_2) during routine chest-drain removal. Surprisingly, it was the deoxygenated haemoglobin concentration ([HbH]) that significantly increased in response to tactile and, even more so, noxious stimulation when compared to baseline. While males demonstrated bilateral decreases in oxygenated haemoglobin concentrations ([HbO_2]) in response to the procedure, females showed bilateral increases. Interestingly, MAP changes were not associated with any of the cerebral haemodynamic changes we recorded. Additionally, we did not find any statistically significant changes in SaO_2.

Regional cerebral activation typically results in regional increases in both [HbO_2] and [HbT] with a decrease in [HbH] (Hoshi, 2003). However, contrasting results have been reported, such as no change or increases in [HbO_2] with increases in both [HbH] and [HbT] (Kato, Kamei, Takashima, & Ozaki, 1993, Kleinschmidt et al., 1996; Hoshi & Tamura, 1993). Previous studies that have reported cerebral haemodynamic responses to stressful and noxious stimuli involved preterm and term born neonates (Bartocci et al., 2006; Limperopoulos et al., 2008; Slater et al., 2006; Slater et al., 2008). None of these studies reported changes in the [HbH] or incorporate pharmacological treatment for pain into their analyses. In our total sample, we found paradoxical results to those reported by Bartocci *et al.* (2006). They described significant bilateral increases in mean [HbO_2] recorded from the somatosensory areas after venipuncture. Slater *et al*. (2006) reported results more similar to ours, namely significant increases in [HbT] when compared to baseline in the contralateral somatosensory cortex of stable preterm infants following heel lance. Bilateral prefrontal decreases in [HbO_2], independent of gender, have been reported in school-age children in response to unpleasant emotions (Hoshi & Chen, 2002).

In our total sample we were unable to demonstrate significant changes in the [HbO_2] responses, possibly because only females showed the *expected* (i.e. previously reported (Bartocci et al., 2006; Limperopoulos et al., 2008; Slater et al., 2006)) [HbO_2] increases, while males showed decreases. Additionally, contrary to Bartocci *et al.*'s (2006) findings, and direction of change set aside, we found significantly larger [HbO_2] changes in females. However, experimental findings have consistently demonstrated that women experience pain more intensely than men do (Fillingim King, Ribeiro-Dasilva,

Rahim-Williams, & Riley, 2009; Goffaux et al., 2011; Riley, Robinson, Wise, Myers, & Fillingim, 1998). A number of physiological, structural, hormonal, psychological, and environmental factors influence how the human brain responds to noxious stimulation. To address whether and how these influences explain our observed gender differences in brain activation would require a much larger sample size than was available in our study (Derbyshire, 2008). Furthermore, very few experimental and clinical studies in human adult have described gender differences in the analgesic effect of mu-opioid agonists when compared to animal (i.e. rodent) studies; of those in humans, results have been equivocal (Bijur et al., 2008; Comer et al., 2010). A recent systematic review described mixed results, showing a stronger effect of morphine in adult woman in experimental pain studies but this effect was not present in clinical trials (Niesters et al., 2010). To our knowledge, no published studies have reported on sex differences of analgesic effect in the paediatric population.

Another factor that may explain our results is the administration of analgesic medication which explained 22% of the variance in the changes in [HbO_2] during chest-drain removal (T_2-T_0) in the sub-group, and 59% of the variance in infants whose optode placement was not on the somatosensory region, but in the frontal or temporal areas. The inverse association between administered dose of morphine and the change in [HbO_2] suggests that analgesic aids in blunting the cerebrovascular response to the nociceptive stimulus. As such, morphine appears to decrease pain-induced neurovascular *activation* (i.e. regional increase in blood flow and [HbO_2] with decreases in [HbH], reflecting increase in neuronal activity), thus dampening the cortical activity in response to noxious stimulation and confirming its inhibitory effect (Apkarian et al., 2005; Casey et al.,

2000). Interestingly, this effect was more profound in the infants that had their optodes placed in the frontoparietal or temporoparietal regions, areas that are closer to regions that are known to be rich in μ-opioid receptors (Apkarian et al., 2005; Schlaepfer et al., 1998). Analgesics such as morphine were shown to decrease the functional magnetic resonance imaging (fMRI) signal (i.e. blood level oxygen dependent (BOLD) signal caused by a decrease in [HbH]) in the prefrontal cortex (Becerra et al., 2006). Furthermore, administration of sedatives during imaging has been shown to reverse the direction of the BOLD signal in adults (Hirth et al., 1996); thus demonstrating an increase in deoxygenated haemoglobin with a decrease in oxygenated haemoglobin.

Cyclical oscillatory changes every 10 to 20 seconds in cerebral [HbO2], [HbH], and (HbT) have been reported during crying episodes in premature neonates; a pattern concordant with recurring obstruction of cerebral venous return (Brazy et al., 1985). Given that 63% (19/30) of our sample cried in response to the noxious procedure, this may have impacted cerebral haemodynamics of these critically ill infants. This may also explain increases of [HbH] we found in the total sample, and even the dual increases in [HbO2] and [HbH] observed in the majority of females. However, some authors report that crying after a painful procedure affects brain oxygenation (through changes in intrathoracic pressure affecting venous return) starting only after approximately 5 minutes (Bartocci et al., 2006). Nevertheless, it is possible that pain induced by the procedure could have caused such a *Valsalva*-type reaction with increased intrathoracic pressure and reduced cerebral venous outflow, thus resulting in an increase in cerebral venous volume and accounting for the observed bilateral rise in [HbH]. However, one

would also expect a concomitant bilateral rise in [HbT] and [HbO2], which we did not observe in more than half of the infants.

Although it has been reported that blood pressure is related to cerebral blood flow and blood volume changes during nociceptive events in sick infants (Limperopoulos et al., 2008; Soul et al., 2007), we found no significant associations between MAP and cerebral haemodynamic measured changes. While both cerebral NIRS parameters and MAP showed significant changes in response to the painful procedure, the measured cerebral changes were independent of the systemic haemodynamic response.

While there have been significant advances in the last decade in research using NIRS to study variations in cerebral oxygenation and haemodynamics of humans, understanding how blood flow, metabolism, and neuronal activity interact to affect the NIRS signals remains incomplete (Bartocci et al., 2006; Oyama, Kondo, Komatsu, & Sugiura, 2009). Additionally, most of the focus in brain-function research has been on [HbO_2]. In general, changes in [HbH] are not reported in the functional NIRS literature, which contributes to the difficulty of interpreting our results. Furthermore, generalizations are difficult because inconsistencies between NIRS and fMRI studies have been reported (Hoshi, 2003; Yamamoto & Kato, 2002). Nonetheless, variations in [HbH] have been described as reflecting neuronal activity, and as being a better indicator of cellular oxygen consumption than [HbO_2] (Oyama et al., 2009). Thus, based on our results, it appears that in response to chest-drain removal neuronal activation caused an increase in cerebral oxygen consumption that surpassed the increase in oxygen delivery, as reflected in a significant increase in [HbH] while [HbO_2] remained relatively constant.

Functional MRI studies in infants and young children (i.e. under 5-year old) suggest that haemodynamic coupling may be different in that age group as compared to adults (Marcar et al., 2004; Sava et al., 2009), possibly even exhibiting a reversal of the adult response pattern (Meek et al., 1998). Contrary to results seen in adult fMRI studies, young children (under 5 years of age) show an increase in oxygen consumption that outpaces the increase in blood flow after stimulation to specific regions of the brain (Marcar et al., 2004; Meek et al., 1998). This is the case when both oxy- and deoxygenated haemoglobin increase, which was the case in our female sample. This phenomenon has been tentatively explained by anatomical differences in cerebral neuronal networking in immature regions of the brain. For example, compared to older children (older than 6 years old) and adults, there is a higher number of synaptic connections in the immature visual system (Marcar et al., 2004). Thus, cerebral neurovascular coupling could differ in young children when compared to more mature neuronal networks found in adults (Sava et al., 2009). An immature neuronal network characterized by a higher number of synaptic connections could explain the significant increase in [HbH] that we found in our sample.

Lateralization in the cerebral haemodynamic response could not be verified in the present study as stimulation in the majority (90%) of cases was in the midline chest area (i.e. where chest drains were located) or bilaterally (i.e. one drain on right and one on the left side of chest). Nonetheless, our results indicate that the noxious stimulation seems to provoke differing hemispheric patterns of change. It is possible that these varying hemispheric recorded changes in response to noxious stimulus may be due to interference with the cortical motor area, which is located just adjacent to the somatosensory area.

During the procedure, patients were not fully restrained. Consequently, limbs as well as the head frequently moved during stimulation, likely causing changes in cerebral haemodynamics in this brain area as well. This calls for further investigations in which motor activity would be statistically controlled. It also seems that the left hemisphere is more associated with other parameters such as when compared to the right. Morphine administered doses significantly dampened the response in [HbO_2] only in the left hemisphere which perhaps explains the modest variations we have found. However, the few studies on this topic in the literature do not suggest an explanation (Bartocci et al., 2006; Limperopoulos et al., 2008; Slater et al., 2006) and none of these studies reported an effect of analgesic or sedation medication. Moreover, we must remember that it is possible that these associations could be correlated with each other. Furthermore, because of our sample size, we were limited in the level of analysis (i.e. only univariate analyses were conducted). Thus, this effect could not be verified and remains a major limitation of the current study.

Several limitations of our study are apparent. First, our sample size limited the extent of our analyses and did not permit us to perform multivariate statistical tests, which would have allowed us to assess interactions among variables and possibly include further important clinical indicators. A larger sample size is needed to further assess and evaluate confounding factors (such as administration of various medications, pain intensity/score, chest drain type, etc.). Second, optode placement and consistent interrogation of the same brain regions is a major concern with NIRS. Functional NIRS recordings, using multichannel NIRS devices that cover multiple regions of the scalp, are not readily available, though their use in adult and older children is increasing (Becerra et

al., 2008; Hoshi, 2003; Hoshi & Chen, 2002). Controlling for movement artefact and possible confounding environmental factors in this clinical research setting is very difficult. However, since the purpose of this study was to explore, through the use of NIRS technology, cerebral haemodynamic changes to a frequently performed noxious procedure in critically ill patients, we wanted to mimic as closely as possible the clinical care routine. Thus, we chose not to interfere with standard care and did not apply more strict control over possible confounding factors.

CONCLUSIONS

This study demonstrates that alterations in cerebral haemodynamics in response to a noxious stimulus in critically ill infants may occur independently of a systemic response, and despite pain medications. One of the surprising findings of this study is the disparate direction in oxygenated haemoglobin response between male and female infants. Based on our results, it appears that NIRS has potential as a technique for assessing pain-evoked cerebral activation in critically ill infants. Given the complexity of NIRS technology, the paucity of research supporting its use in pain assessment and quantification in critically ill infants, and the need for tight control of many confounding factors as well as artefacts, more studies are clearly needed with larger sample sizes. Particularly important will be studies that combine the strengths of NIRS with those of other complimentary techniques such as EEG, electromyography, and fMRI to increase our understanding of neurovascular coupling in the immature brain.

ACKNOWLEDGEMENTS

This work was supported in part by the United States National Institutes of Health through grants R01EB001659, K24NS057568, and R21HD056009 by the National Institute of Biomedical Imaging and Bioengineering, the National Institute of Neurological Disorders and Stroke, and the National Institute of Child Health and Human Development, respectively. Further support was provided by the Lifebridge Fund. M. Ranger was supported by doctoral and international fellowships from the Quebec Health Research Fund and the Quebec Interuniversity Nursing Intervention Research Group. We are very thankful to the nurses of the pre-operative clinic and cardiac intensive care unit at Children's Hospital Boston and families of patients, whose generous help and participation made this work possible. We also thank our colleagues Dr. Mustafa Sulemanji and Ms. Heather O'Leary for significant contributions and guidance in the performance of this study.

Table 1. Demographic and clinical characterization of the critically ill infants (*n* = 32)

Sex distribution	Male 56% (18); Female 44% (14)
Mean age (months)	3.6 ± 1.6; range 1 to 7
Mean weight (kg)	5.5 ± 1.4; range 2.8-10
Mean PRISM III scores	9 ± 4; range 3-18
Diagnosis: TOF (+ pulmonary stenosis) VSD & VSD/PFO ASD/VSD (+PDA) CAVC (+PDA) Combination of 2 or more Other	*n* = 9 *n* = 8 *n* = 3 *n* = 2 *n* = 5 *n* = 5
Mean morphine dose (*n* = 31)	60 mcg/kg/dose; range 25-200
Mean midazolam dose (*n* = 26)	60 mcg/kg/dose; range 25-100
Type of inotrope infusions (*n* of infants receiving infusions 6)[a]	Nitroprusside (*n*=3); dopamine (*n*=1); nitroglyceride (*n*=1); procainamide (*n*=1); milirone (*n*=1)
Mean time from surgery at noxious procedure	33.3 hours; range 16-92 (median 23.5)
Location of chest drain	Midline (*n*= 22) Right (*n*=9)[b] Left (*n*=5)[b]
Type of chest drain[c]	Chest tube (*n* = 9); Blake drain (*n* = 25)
Mean maximum FLACC pain score in last 24h (*n* = 30)	4.6 (± 2.03); range 0-8

PRISM III: Pediatric RISk of Mortality; TOF: Tetralogy of Fallot; VSD: Ventricular septal defect; ASD: Atrial septal defect; PFO: Patent foramen ovale; PDA: Patent ductus arteriosis; CAVC: Complete atrioventricular canal; Combination: CAVC/TOF, TOF/ASD, TOF/PFO; Other: Aortic root aneurysm, hypoplastic left heart syndrome, Transitional AV canal; pain score was taken from patients' charts.

[a] One infant was receiving two infusions of inotropes (nitroprusside and nitroglyceride);

[b] Four infants had bilateral chest drains- right and left- removed at the same time;

[c] Two infants had both a chest tube and Blake drain that were removed at the same time.

FLACC: Face Legs Activity Cry Consolability pain scale (Merkel et al., 1997) . Range of scores 0 to 10.

Table 2. Within-subject pairwise comparisons of signals between epochs (mean differences).

	$T_1 - T_0$ **Tactile vs Baseline**	$T_2 - T_0$ **Noxious vs Baseline**	$T_2 - T_1$ **Noxious vs Tactile**
[HbO_2] (μmol/L)	L: $p = 1$ R: $p = 1$	L: $p = 1$ R: $p = 0.76$	L: $p = 1$ R: $p = 0.41$
[HbH] (μmol/L) ($n_L = 27$; $n_R = 28$)	L: $p = 1$ R: $p = 0.51$	L: 2.87[a] CI 0.02, 5.54 R: 3.52[b] CI 0.86, 6.18	L: 2.21[a] CI 0.37, 4.05 R: 2.97[a] CI 0.06, 5.88
[HbT] (μmol/L) ($n_L = 27$; $n_R = 28$)	L: $p = 1$ R: $p = 1$	L: $p = 0.65$ R: 4.93[a] CI 0.28, 9.58	L: $p = 0.11$ R: $p = 0.1$
[HbD] (μmol/L) ($n_L = 27$; $n_R = 28$)	L: $p = 0.37$ R: $p = 0.89$	L: -3.63[a] CI -7.17, -0.08 R: $p = 0.36$	L: $p = 0.51$ R: $p = 1$
MAP (mmHg) ($n = 20$)	5.49[a] CI 0.38, 10.58	12.52[b] CI 4.93, 20.12	7.04[b] CI 2.09, 12
HR (beats/min) ($n = 29$)	$p = 0.06$	9.53[b] CI 2.59, 16.47	3.63[a] CI 0.55, 6.71
SaO_2 (%) ($n = 23$)	$p = 0.80$	$p = 0.11$	$p = 0.88$

[HbO_2]: oxygenated haemoglobin concentration; [HbH]: deoxygenated haemoglobin concentration; [HbT]: total haemoglobin concentration; [HbD]: haemoglobin difference concentration; MAP: mean arterial blood pressure; HR: heart rate; SaO_2: arterial blood oxygen saturation; [a] $p < 0.05$; [b] $p < 0.01$.
T_0: baseline; T_1: tactile stimulus; T_2: noxious stimulation (chest drain removal); L: left; R: right.

Table 3. Relationship (*p* value) between events, cerebral and circulatory measures and medication administration (univariate analysis)

Parameter	$\Delta[HbO_2]$	$\Delta[HbH]$	ΔMAP	ΔHR	ΔSaO_2
T_1-T_0					
ΔMAP					
ΔHR					
ΔSaO_2		-0.29 (.008)[a]			
Morphine	-75.65 (.023)[a]				
Midazolam					-63.6 (.015)
Age	1.77 (.017)[b]				
T_2-T_0					
ΔMAP					
ΔHR					
ΔSaO_2					
Morphine	-88.97 (.007)[a]				
Midazolam					
Age		-1.53 (.017)[a]			
T_2-T_1					
ΔMAP					
ΔHR					
ΔSaO_2					
Morphine					
Midazolam				-94.09 (.012)	
Age	2.26 (.012)[b]		3.11 (.002)		

[a] Left hemisphere; [b] Right hemisphere.

$[HbO_2]$: oxygenated haemoglobin concentration; [HbH]: deoxygenated haemoglobin concentration; MAP: mean arterial blood pressure; HR: heart rate; SaO_2: arterial blood oxygen saturation.

T_0: baseline; T_1: tactile stimulus; T_2: noxious stimulation (chest-drain removal).

TableS1. Median and Interquartile ranges for cerebral NIRS across 30-sec epochs

	Median[a]	**IQR[a]**	**Median[b]**	**IQR[b]**
$[HbO_2]\ T_0$	-0.141	[-1.841, 2.168]	0.429	[-1.402, 1.754]
$[HbO_2]\ T_1$	-1.145	[-4.150, 3.041]	-0.676	[-3.472, 2.771]
$[HbO_2]\ T_2$	-0.906	[-4.79, 2.479]	-0.507	[-3.182, 4.661]
$[HbH]\ T_0$	0.417	[-1.165, 1.359]	-0.035	[-1.803, 1.087]
$[HbH]\ T_1$	0.447	[-1.545, 3.568]	0.318	[-1.784, 2.924]
$[HbH]\ T_2$	2.892	[-0.498, 7.144]	2.684	[-0.693, 7.066]
$[HbT]\ T_0$	0.135	[-2.308, 3.615]	0.145	[-1.548, 2.106]
$[HbT]\ T_1$	-0.72	[-3.7, 4.7]	-0.875	[-3.536, 4.205]
$[HbT]\ T_2$	2.854	[-3.309, 1.009]	2.299	[-1.115, 5.859]
$[HbD]\ T_0$	0.162	[-1.887, 1.481]	0.542	[-1.436, 2.73]
$[HbD]\ T_1$	-0.125	[-5.238, 2.563]	0.611	[-4.412, 2.511]
$[HbD]\ T_2$	-3.726	[-8.748, 0.672]	-1.415	[-5,927, 2.178]

IQR: Interquartile range; [a] Left hemisphere, [b] Right hemisphere.
$[HbO_2]$: oxygenated haemoglobin concentration; [HbH]: deoxygenated haemoglobin concentration; [HbT]: total haemoglobin concentration; [HbD]: difference in haemoglobin concentration; all shown in µmo/l.
T_0: baseline, T_1: tactile stimulation, T_2: noxious stimulation.

TableS2. Somatosensory cortex optode placement sub-group regression analysis (n = 22) compared to "other" cortex optode placement sub-group regression analysis (n = 5) with morphine administration.

Optode	**Parameter**	***B***	**95% CI**	***r***	***p*-value**
Somatosensory	$\Delta[HbO_2]$ Left[a]	-66.81	[-131.38, -2.25]	-0.47	.043
Somatosensory	$\Delta[HbO_2]$ Left[b]	-79.69	[-156.92, -2.46]	-0.47	.044
"Other"	$\Delta[HbO_2]$ Left[b]	-150.77	[-276.4, -25.15]	-0.77	.026

$\Delta[HbO_2]$: change in oxygenated haemoglobin concentration; left hemisphere
"Other" optode placement: frontoparietal or temporoparietal cortical region
[a] For Tactile stimulation - Baseline
[b] For Chest-drain removal - Baseline

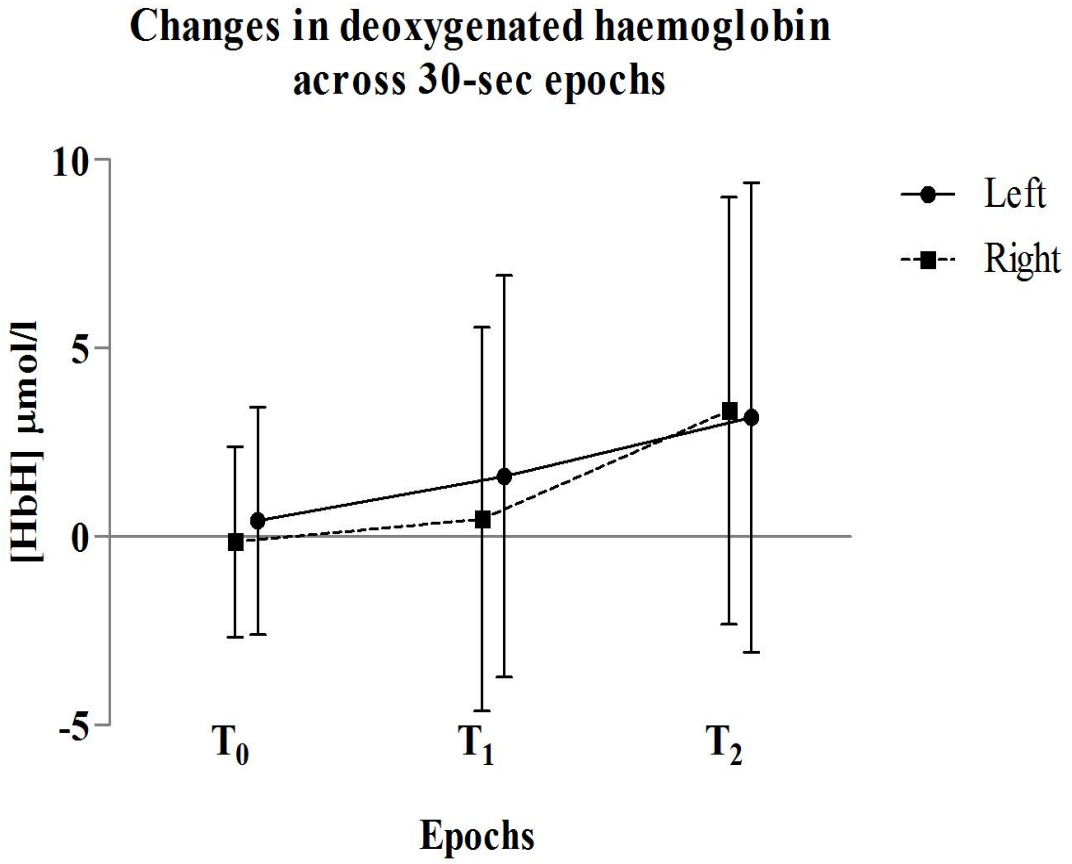

Figure 1. Changes in left (n = 27) and right (n = 28) hemispheres [HbH] values in all subjects. The figure shows significant [HbH] increases from baseline (T_0) to noxious stimulation (T_2) (left $p < 0.05$; right $p < 0.01$). Significant increases from tactile (T_1) to noxious on left hemisphere ($p < 0.05$). Vertical bars denote the 95% confidence intervals and the middle points denote the mean values.
[HbH] : deoxygentated haemoglobin concentration.

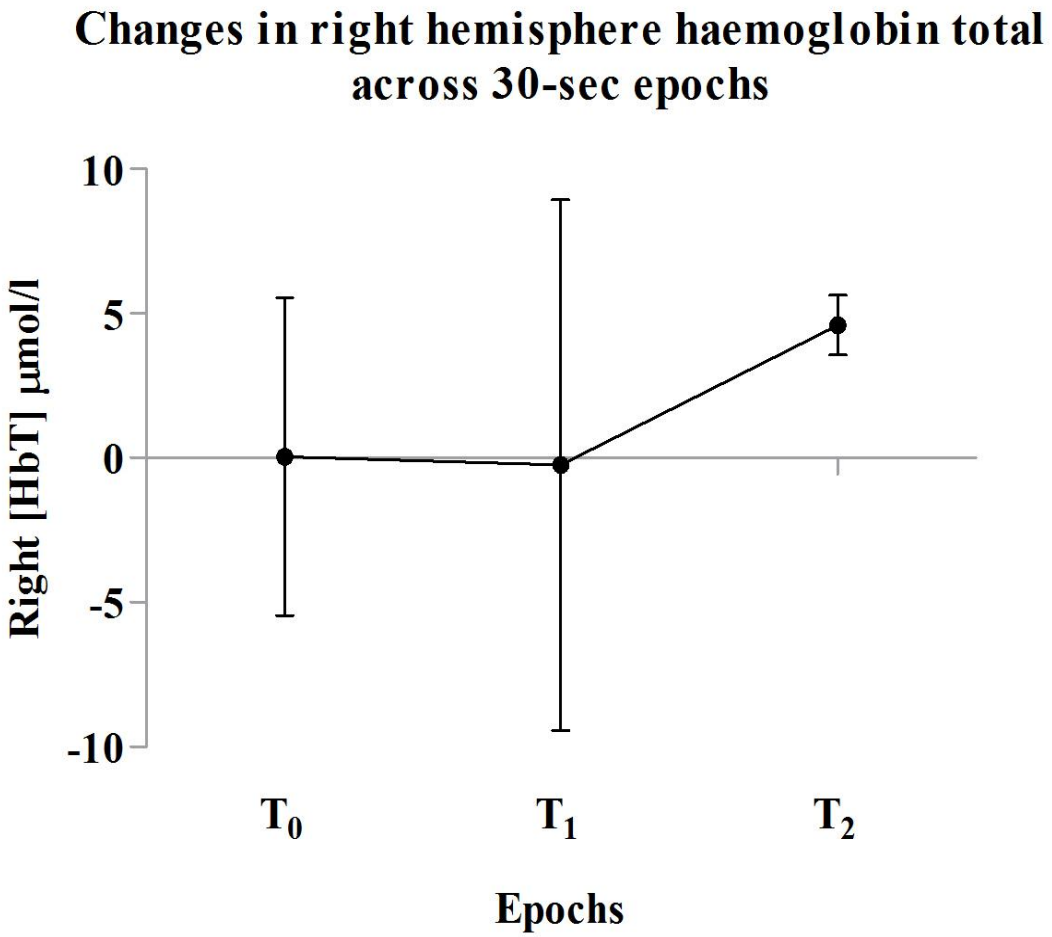

Figure S1. Changes in right hemiphere [HbT] values in all subjects (n = 28). The figure shows significant [HbT] increases from baseline (T_0) to noxious stimulation (T_2) ($p < 0.05$). Vertical bars denote the 95% confidence intervals and the middle points denote the mean values.

[HbT] : total haemoglobin concentration; T_1: tactile stimulation.

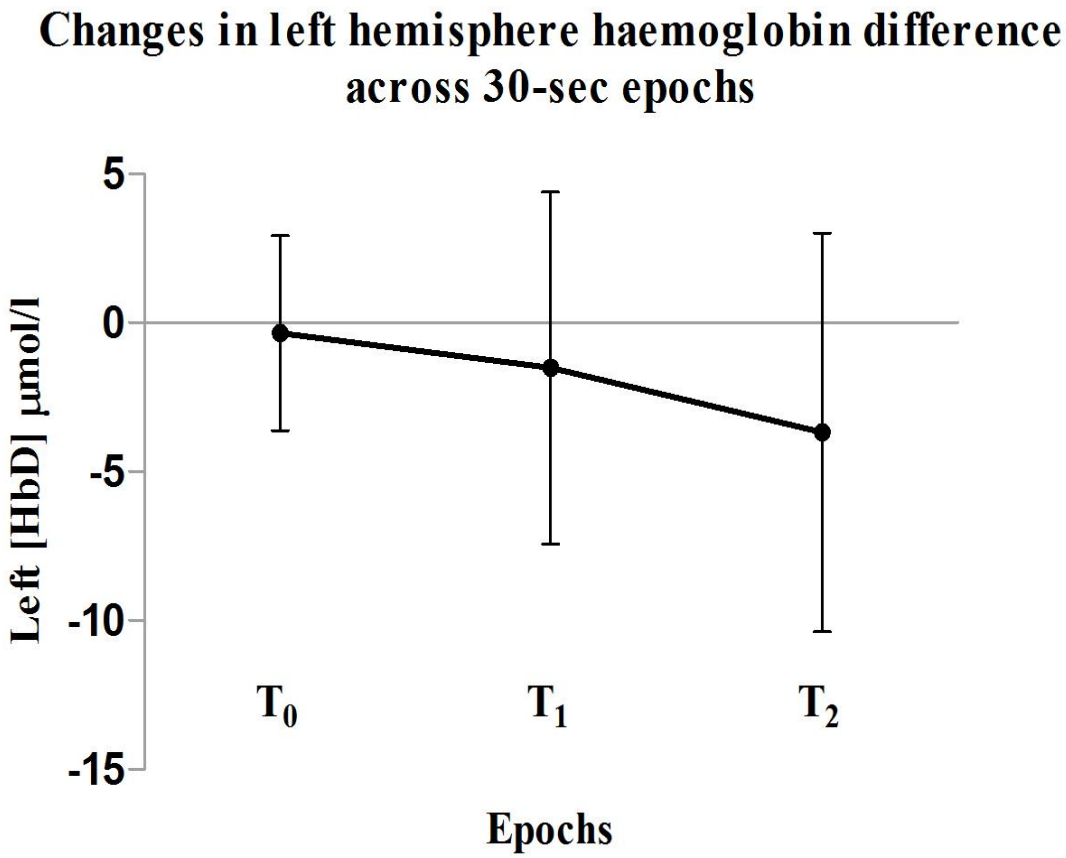

Figure S2. Changes in left hemiphere [HbD] values in all subjects (n = 27). The figure shows significant [HbD] decreases from baseline (T_0) to noxious stimulation (T_2) ($p < 0.05$). Vertical bars denote the 95% confidence intervals and the middle points denote the mean values.
[HbD]: haemoglobin difference concentration; T_1: tactile stimulation.

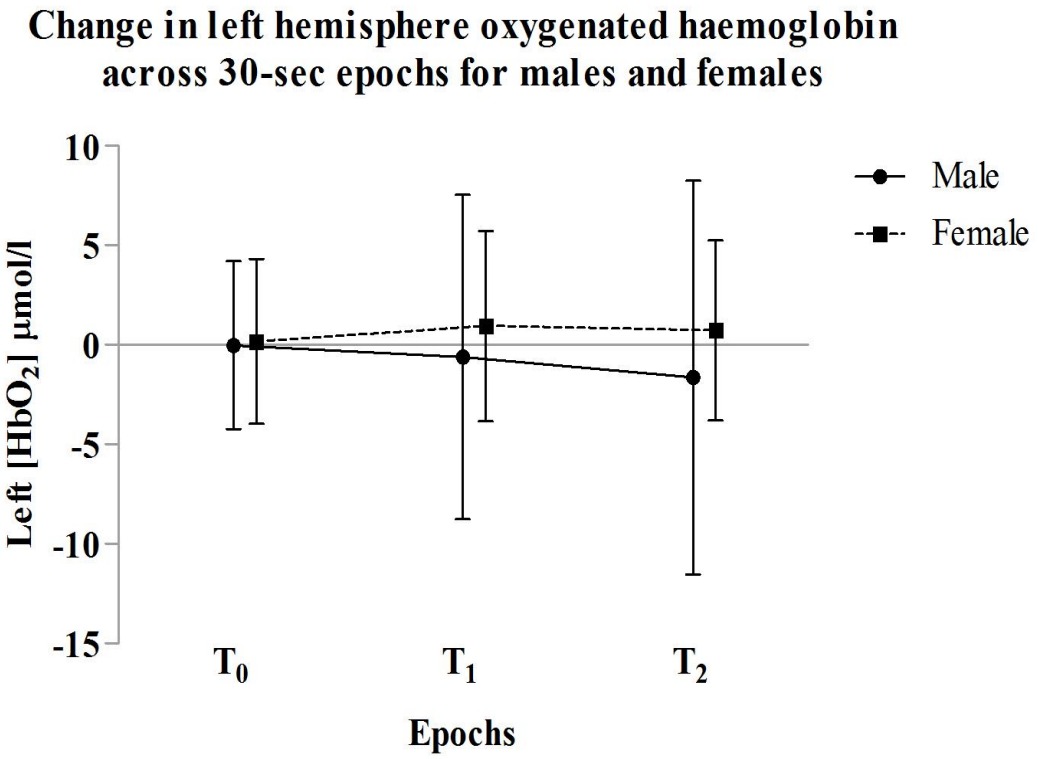

Figure S3. Changes in left hemisphere [HbO_2] in males (n = 14) and females (n = 13) infants. Females showed a significantly higher change across the epochs in the left hemisphere [HbO_2] (p = .04). Vertical bars denote the 95% confidence intervals and the middle points denote the mean values.
[HbO_2]: oxygenated haemoglobin concentration; T_0: baseline, T_1: tactile stimulation, T_2: noxious stimulation.

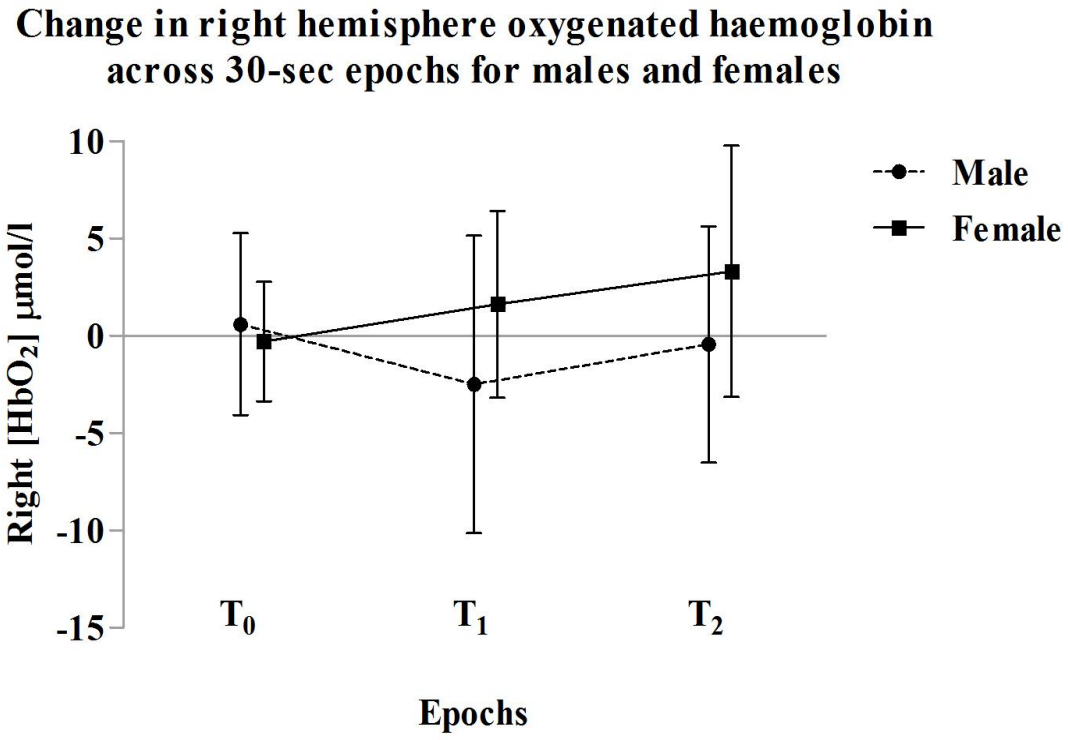

Figure S4. Changes in right hemisphere [HbO_2] in males (n = 15) and females (n = 13) infants. Vertical bars denote the 95% confidence intervals and the middle points denote the mean values.

[HbO_2]: oxygenated haemoglobin concentration; T_0: baseline, T_1: tactile stimulation, T_2: noxious stimulation.

Manuscript #3: A multidimensional approach to pain assessment in critically ill infants during a painful procedure

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Submitted for publication to the Clinical Journal of Pain (Appendix H)

Preface

Manuscript #3 of this dissertation is the second of two papers on the results of the study. This last article explores associations between regional cerebral haemodynamic changes, behavioural pain scores, systemic physiological changes, and specific clinical variables in critically ill infants during chest-drain removal. Thus, it aims to address the second objective of the study dissertation. Although all infants comprised in the total sample (in manuscript #2) were filmed during data collection, we were able to capture good quality video recordings of the procedure to enable behavioural pain coding in about 65% of the total sample. These participants consist of the study sample comprised in manuscript #3.

A multidimensional approach to pain assessment in critically ill infants during a painful procedure

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ABSTRACT

Objectives: Inferring the pain level of a critically ill infant is complex. The ability to accurately extract the appropriate pain cues from observations is often jeopardized when heavy sedation and muscular blocking agents are administered. Near-infrared spectroscopy (NIRS) is a noninvasive method that may provide the bridge between behavioral observational indicators and cortical pain processing. We aimed to describe regional cerebral and systemic hemodynamic changes, as well as behavioral reactions in critically ill infants with congenital heart defects (CHD) during chest drain removal following cardiac surgery.

Methods: Our sample included 20 critically ill infants with CHD less than 12 months of age admitted to the cardiac intensive care unit following surgery.

Results: Cerebral deoxygenated hemoglobin concentrations significantly differed across the epochs (i.e. Baseline, Tactile stimulus, Noxious stimulus) (p = .01). Physiological systemic responses and FLACC pain scores differed significantly across the events (p < .01). The three outcome measures were not found to be associated with each other. Mean FLACC pain scores during the painful procedure was 7/10 despite administration of morphine. Midazolam administration accounted for 36% of the variance in pain scores.

Discussion: We demonstrated with a multidimensional pain assessment approach that significant cerebral, physiological and behavioral activity was present in response to a noxious procedure in critically ill infants despite the administration of analgesic treatment. Considering that the sedating agent significantly dampened pain behaviors,

assessment of cerebral hemodynamic in the context of pain seems to be an important addition.

1. Introduction

To appropriately respond and treat pain in the non-communicative child, one must be able to accurately extract the appropriate pain cues from observations. Although a wide variety of pain measures exist for assessing pain in non-communicative infants and children, no one measure has been uniformly accepted. Although a considerable body of knowledge now exists, an observational tool that evaluates pain with sound validity, reliability, specificity, and sensitivity remains elusive (Buttner & Finke, 2000). More recently, researchers have directed their efforts towards improving the psychometric properties of existing pain assessment instruments within various populations, types of pain and contextual factors (Stevens et al., 2007). Clinicians still do not know if the behaviors they observe are specific to pain or if they are manifestations of other experiences such as agitation, distress, fear, stress, or sadness (Buttner & Finke, 2000; Ramelet et al., 2004). This issue is even more problematic when dealing with critically ill infants, in which the expressive capacity is often jeopardized by the administration of heavy sedation and muscular blocking agents.

Most of the available paediatric pain assessment instruments are multidimensional, incorporating both behavioral (facial action, body movement, cry) and physiological (heart rate, respiratory rate, blood pressure, arterial oxygen saturation) indicators. Dissociation between these two classes of indicators has been reported with an average correlation of 0.3 (Barr, 1998; Johnston et al., 1995; Stevens et al., 1993). Behavioral measures, especially facial actions, are more likely to respond selectively to pain (Craig et al., 1993), while physiological indicators change in response to painful

stimuli, but also for numerous reasons not specific to pain (Johnston et al., 1999; Stevens et al., 1993; Sweet & McGrath, 1998).

Hemodynamic and electrical brain studies into pain processing show potential for exploring these issues (Bartocci et al., 2006; Sava et al., 2010; Slater et al., 2006; Slater et al., 2008; Slater et al., 2010a; Slater et al., 2010b; Slater et al., 2010c). One such approach employs continuous near-infrared spectroscopy (NIRS), which may provide the bridge between behavioral observational indicators and cortical pain processing, thus serving as an alternative and perhaps more specific instrument in the measure of pain in critically ill infants. In fact, recent studies in premature and term infants indicated that painful stimuli cause hemodynamic changes in specific cortical regions of the brain (Bartocci et al., 2006; Slater et al., 2006; Slater et al., 2008).

In this study we report on regional cerebral and systemic hemodynamic changes, as well as behavioral reactions in critically ill infants with congenital heart disease (CHD) during a routine painful procedure. We assessed the cerebrovascular and cardiovascular responses to postoperative chest-drain removal by means of continuous NIRS as well as usual monitoring of heart rate, systemic blood oxygenation, and mean arterial blood pressure. Behavioral reactions were also captured through video recordings and rated for pain retrospectively with the Face Legs Activity Cry Consolability (FLACC) pain scale (Merkel et al., 1997). Specifically, we examined differences within subjects across phases of the procedure, factors associated with the response (sex, age, weight, and medication), as well as associations between cerebral hemodynamic changes, systemic physiological changes, FLACC pain scores and specific clinical variables.

2. Materials and Methods

2.1. Study population

Forty infants less than 12 months who required a surgical procedure for repair of a CHD and who required a chest drain post-operatively were prospectively recruited between April 2009 and May 2010. We excluded infants who were diagnosed with a spinal or peripheral nervous system illness. The research protocol was approved by the Institutional Review Board at Children's Hospital Boston, USA. We obtained parental written informed consent in all cases, and 89% of the approached parents gave their consent.

2.2. Instrumentation

The infants' cerebral hemodynamic and associated physiological signals, as well as video recordings were continuously monitored and recorded for approximately 30 minutes surrounding the chest drain removal.

A non-invasive NIRS-based tissue oxygenation monitor (NIRO 300, Hamamastu Photonics, Japan) was used to measure changes in cerebral tissue oxygenation. This NIRS device has two channels, each consisting of an emitting and a receiving optode. When possible, the emitter optode was positioned approximately 2 cm beneath and slightly posterior to the C_3/C_4 position (according to the international EEG 10-20 system (Bartocci, 2006; Bartocci et al., 2006)); the receiver optode was positioned 4 cm apart from the emitter optode allowing illumination of the primary somatosensory cortex and possibly the underlying structures. When this optode configuration was not possible due to hair coverage, which causes interference with the laser signal, or other obstacles (i.e. intravenous indwelling catheter), optodes were placed in the frontoparietal or

temporoparietal regions. Optodes were placed in a flexible rubber holder that prevented interference from external light sources and kept the distance between the emitter and receiver optodes constant at 4 cm.

The cerebral oxygenation signal obtained with the NIRS technique is based on the absorption of near-infrared light by hemoglobin, which in turn depends on the oxygenation state of hemoglobin circulating through the tissues. The NIRS technology employed in this study does not measure the absolute cerebral concentrations in oxygenated hemoglobin ($[HbO_2]$) and deoxygenated hemoglobin ([HbH]), but rather *absolute change* in concentration from an unknown reference value (Bartocci, 2006; Soul & du Plessis, 1999; Wolfberg & du Plessis, 2006). The NIRS device allows for data sampling at 6 Hz.

The systemic arterial oxygen saturation (SaO_2), the electrocardiogram (ECG), and arterial blood pressure were recorded continuously from the patient's bedside monitor (Philips Intellivue MP70, Andover, MA). An indwelling arterial line placed either in the femoral or radial arteries provided continuous monitoring of the arterial blood pressure waveform, from which mean arterial blood pressure (MAP) was computed on a beat-by-beat basis.

2.2.1. Face Leg Activity Cry Consolability (FLACC) scale

The FLACC scale is a unidimensional behavioral pain assessment instrument to measure pain in young children in the post-operative period (Merkel et al., 1997). It includes five items (Face, Legs, Activity, Cry, and Consolability) and has good inter-rater reliability (Kappa 0.52-0.82), as well as good content and convergent validity

(Merkel et al., 1997). The FLACC has been shown to be reliable in critically ill young children (Merkel et al., 2002; Voepel-Lewis et al., 2010).

2.3. Data collection

The analogue signals were passed through a band-pass filter and a notch filter at 60 Hz. The band-pass filter was used for anti-aliasing, and the notch filter to exclude potential 60 Hz artifact common in the intensive care unit (ICU) setting. During measurement recording, cerebral, systemic physiological data, and video were simultaneously transferred to a portable user interface monitor (Component Neuromonitoring System, Day One Medical, LLC, Ambler PA, USA). This device is a computer-based system that continuously records, displays, stores, and analyzes in real-time physiological data from multiple monitoring sources in real-time, allowing data synchronization, display, and analysis.

Equipment set-up and data acquisition were completed by the same study investigator (MR), except on one occasion. The investigator was continuously present during the data-acquisition process to document in the patient's study file the precise beginning and end of the chest-drain removal procedure and steps leading to the actual noxious stimulation, as well as the nature of all other events (e.g. study events, administration of medication, handling by care provider or parent, etc.) that occurred throughout the study.

2.4. Chest-drain removal procedure

Data collection was carried out in the cardiac intensive care unit (CICU) at the bedside of the patient, and the nurses performing the chest-drain removal were asked to carry out the procedure as usual. No interruption on the part of the study investigator

took place. Once the equipment was set up, baseline data were gathered prior to the noxious procedure to allow calibration of NIRS device and adjustments for artefacts. Each procedure was performed according to a standardized care plan. The chest-drain removal procedure comprised a sequence of steps and a variety of sensory stimuli, including: (1) administration of analgesics and/or sedatives (e.g. morphine and/or midazolam) in the majority of the cases, (2) removal of dressing, (3) untying of purse-string sutures around the exit site, (4) removal of the drain at the height of expiration, and (5) pulling tightly on the purse string to allow closure of the exit site, immediately followed by application of a pressure bandage. The duration of each step, and therefore the duration of the entire procedure, varied from subject to subject (ranging from 2 to 20 minutes).

All data recordings began 10 to 60 minutes prior to the drain removal and carried on for the duration of the procedure. Data analysis was divided into three 30-second intervals (epochs); these were precisely marked during recordings by the study investigator on an Excel event log sheet. Once the procedure was completed, all recordings were stopped and the equipment was removed.

2.5. Data selection

Three 30-second epochs that represent quiet baseline (T_0), tactile stimulation (T_1) (removal of dressing), and removal of chest drain (T_2), respectively were documented. The beginning of the baseline (T_0) period during which the infant was calm and not disturbed was captured at approximately 25 minutes prior to the drain removal. This period was followed by a tactile stimulation (T_1) due to removal of dressing and preparation of the procedure preceding the chest-drain removal. The epoch corresponding

to the noxious stimulus response (T_2) was signaled by the unit nurse as soon as the tube began to be pulled on peak expiration by the patient. A recovery period was captured in only half of the participants and thus, was not included in the analysis.

2.5.1. Video

Filmed behavioral displays were rated according to the recommended procedure for clinical use of the FLACC scale by a clinical nurse specialist (CNS) in paediatric pain experienced with the use of the instrument and blinded to the purpose of the study. The footages were played back in real time on a personal computer; each of the 30-second segments previously identified by study investigator (MR) was coded for pain scores by the CNS. Thus, the blinded coder generated three pain FLACC scores related to the study epochs, and this for each participant (Cronbach alpha of 0.8). This same procedure was repeated ten weeks after the first ratings were done to test intrarater reliability of 0.9. A second blinded CNS in paediatric pain coded half of the sample to establish interrater reliability of 0.86 (0.71-0.96) (ICC).

2.6. Data pre-processing and statistical analysis

For ease of data handling, the physiological signals were subsequently down-sampled to 20 Hz prior to further analysis. From the raw data we computed average [HbO_2], [HbH], HR, MAP, and SaO_2 values for each 30-second epoch. For ease of naming, each variable will be referred to by its name without mentioning "mean". We also computed average changes for each signal (Δ[HbO_2], Δ[HbH], ΔHR, ΔMAP, and ΔSaO_2) between the different event epochs: Tactile vs. Baseline (T_1–T_0), Noxious vs. Baseline (T_2-T_0), and Noxious vs. Tactile (T_2-T_1).

A custom Matlab script was used to process the raw recordings and analyze the data (Matlab Student version 7.0, The Mathworks, Natick, MA). To determine whether chest-drain removal procedures induced statistically significant cerebral and systemic hemodynamic changes compared to the other epochs, a repeated measure ANOVA was conducted to compare [HbO_2], [HbH], HR, MAP, and SaO_2 responses, as well as FLACC pain scores between the epochs. Sex was added as a between-subject factor. Univariate linear regression analyses were used to verify associations between changes in NIRS signal values (Δ[HbO_2] and Δ[HbH]), systemic physiological responses (ΔHR, ΔMAP, ΔSaO_2), FLACC pain score changes, analgesic medication doses, age, weight, pediatric risk of mortality score (PRISM III) (Pollack et al., 1996), and elapsed time since surgery of chest- drain removal.

Statistical analyses were performed using the Statistical Package for Social Sciences (SPSS) version 15.0 (IBM, Somers, NY); *p*-values of less than 0.05 were considered significant, and Bonferroni correction was used to adjust for multiple comparison. Mauchly's test was used to verify the assumption of sphericity, and if violated, degrees of freedom were corrected using Greenhouse-Geisser estimates of sphericity.

3. Results

3.1. Population

The families of 45 infants were approached in the pre-operative cardiac clinic to participate in the study. Forty infants were enrolled, and 32 (18 males; 14 females) participated in the immediate post-operative period. Eight recruited participants were not recorded because chest drain removal was missed (n = 6) or the patient was transferred to

another unit prior to procedure (n = 2). Although all 32 infants were filmed, we were able to capture good quality video recordings of the procedure to enable behavioral pain coding in only 20 infants (12 males; 8 females). These participants comprised the sample for the present study, though not all 20 recorded data sets could be used in the analysis due to equipment issues, loss of signal, or excessive noise in the recordings. Twenty NIRS data sets for left hemisphere and 19 for right hemisphere were analyzed, 20 for HR, and 18 for SaO_2. Fourteen infants had an indwelling arterial catheter allowing for continuous MAP monitoring. Infants' characteristics are presented in Table 1.

The 20 infants underwent cardiopulmonary bypass for various CHD. Seven children had a co-morbid condition with the most common being Trisomy 21 in five patients (3 females; 2 males), one had a connective tissue disorder and one had a syndrome associated with digital abnormality not well characterized otherwise. When their chest drain was removed 7 out of the 20 participants required support from a conventional ventilator (n = 6) or CPAP (n = 1). Cortical somatosensory placement of optodes was not achieved in six participants because of hair or access issues. In these 6 cases, optodes were placed in the frontotemporal and temporoparietal regions.

3.2. Cerebral NIRS changes

In response to chest drain removal, significant increases in regional cerebral deoxygenated hemoglobin concentrations [HbH] were found. Specifically, the right hemisphere [HbH] differed significantly between the different 30-second epochs $F(2, 36) = 5.07$ ($p = .01$). Within-subject pairwise comparisons showed significant differences between Baseline (T_0) and Noxious (T_2) [HbH] ($p = .04$) in the right hemisphere (Figure 1). We found no significant changes in the left hemisphere [HbH] or bilateral cerebral

regional oxygenated hemoglobin [HbO_2]. Optode placement or chest tube location were not determining factors in cerebral hemodynamic responses.

3.3. Systemic physiological changes

In response to chest-drain removal, we found significant increases in systemic mean arterial blood pressure (MAP). MAP differed significantly between the different 30-second epochs $F(1.38, 16.52) = 19.18$ ($p < .001$). Within-subject pairwise comparisons showed significant differences between the three 30-second epochs: T_2 and T_0 MAP (15.75 CI [6.62, 24.89]; $p = .001$); T_2 and T_1 MAP ($p = .005$); T_0 and T_1 MAP ($p = .012$).

Heart rate (HR) differed significantly between the different 30-second epochs $F(1.28, 24.27) = 6.87$ ($p = .01$). Within-subject pairwise comparisons showed significant differences between T_2 and T_0 HR (11.35 CI [1.27, 21.42]; $p = .02$). No other significant differences were found; systemic arterial blood oxygenation (SaO_2) level was not found to differ significantly across the 30-second epochs.

3.4. FLACC pain scores

Mean FLACC pain scores at the three epochs were 0.25 (SD 0.12); CI [0.01, .51] at baseline, at tactile 3.25 (SD 0.56); CI [2.08, 4.23], and at noxious 6.7 (0.66); CI [5.32, 8.08]. Overall, FLACC pain scores differed significantly between the different 30-second epochs $F(1, 19) = 102.64$ ($p < .001$) (Table 2; Figure 2). Female infants had significantly higher FLACC scores than males $F(1, 18) = 9.65$ ($p = .006$) across all events. In addition to the standard pharmacological treatment, two participants received sucrose prior to chest-drain removal and both had 9/10 FLACC pain scores in response to the noxious procedure.

3.5. Changes in NIRS, physiological signals and pain scores across 30-second epochs

We performed univariate analyses to explore the association among study parameters related to changes (Δ) observed during the different 30-second epochs, as well as exploring the relationship between these and clinical factors. Table 3 shows those relationships that achieved statistical significance on univariate analysis. Only left hemisphere $\Delta[HbO_2]$ was associated with ΔHR, explaining 32% of the change. No other NIRS signal change across the three 30-second epochs was found to be significantly correlated with physiological signals. Total FLACC pain scores and its five indicators were not significantly associated with the NIRS and physiological signal changes across the different epochs.

In response to chest-drain removal, infants with the comorbid condition Trisomy 21 did not differ significantly in their cerebral and physiological hemodynamic response or FLACC pain scores when compared to the rest of the sample. Infants' age and weight significantly correlated with some of the measured variables. Results from the univariate analyses are found in Table 3.

3.5.1. Effects of medication

The administration of analgesic medication (morphine) was significantly associated with less changes in $[HbO_2]$ (β -137.78, p = .005) and [HbH] (β -82.54, p = .002). Morphine doses also decreased the response in HR (β -90.24, p = .022). The administration of analgesic was not significantly associated with FLACC pain scores changes.

Sedation medication administration (midazolam) was associated with significantly less changes in left hemisphere $[HbO_2]$ and bilateral [HbH] (β -114.87, p =

.017; β -63.7, p = .015; β -137.49, p = .008). Midazolam doses also significantly decrease the responses of the HR and SaO_2 (β -146.63, p = .005; β -71.27, p = .012). Administration of a sedative was significantly correlated with less FLACC pain scores changes between T_0 and T_2 (-65.76; p = .005) and 19% of the changes between T_0 and T_1 (-40.53; p = .05). These data are presented in Table 3.

3.5.2. Individual changes between T_2-T_0

For comparison purposes, we divided the sample in two groups: (1) the moderate to severe pain group (FLACC pain scores ≥ 5) and (2) the no pain to mild pain group (FLACC pain scores ≤ 4). For the first group, mean FLACC scores were 8/10, mean cerebral hemodynamic (i.e. [HbO_2] and [HbH]) varying from -0.6 to 4.7 μmol/l with large standard deviations, and smaller mean doses of morphine/midazolam. The second group received higher mean doses of these same drugs with lower mean FLACC scores 2/10, and mean cerebral hemodynamic changes between -0.33 to 3.3 μmol/l. These results can be found in the supplemental Tables S1 and S2 which show the results of the descriptive analyses of individual cerebral hemodynamic and FLACC score changes from baseline to chest-drain removal.

3.6. Sex disparity

Sex of the patient was found to be a determining factor in the cerebral and behavioral responses (supplemental Figures S1 and S2). Females responded more strongly than males across the events in the right hemisphere [HbO_2] $F(1, 17) = 4.3$ (p = .05). As stated previously, female infants had significantly higher pain scores across the events as well (Figure 2). We found that the change in FLACC scores of males from T_0

to T_2 were significantly dampened the midazolam administered doses (β = -70.43, CI [-134.43, -6.43], p = .03; $r(11)$ = -0.61). This was not the case in female infants.

4. Discussion

We demonstrated varying cerebrovascular and systemic hemodynamic changes, as well as pain behavioral manifestations in response to chest-drain removal in critically ill infants with CHD following open heart surgery. We demonstrated the gender differences: females showed significantly stronger cerebral hemodynamic response and higher pain scores across the events when compared to males. Finally, we failed to find significant associations between these three multidimensional "pain" parameters.

Previous studies that have reported on cerebral hemodynamic responses to noxious stimuli have focused on preterm and term born neonates (Bartocci et al., 2006; Slater et al., 2006; Slater et al., 2008). These studies showed significant bilateral mean increases in [HbO_2] (14) or in the maximum change in the contralateral total hemoglobin concentrations ([HbT] = [HbO_2] + [HbH]) (Slater et al., 2006; Slater et al., 2008) when compared to baseline. None of these studies reported on the measured changes in the [HbH] or incorporated pharmacological treatment for pain into their analyses, therefore making comparison with our findings difficult. Regional cerebral activation typically results in regional increases in both [HbO_2] and [HbT] with a decrease in [HbH] (Hoshi, 2003). However, contrasting results have been reported, including no change or increases in [HbH] with increases in both [HbO_2] and [HbT] (Hoshi & Tamura, 1993; Kato et al., 1998; Kleinshmidt et al., 1996).

To date, there is only one study reporting on associations between pain scores and cerebral hemodynamic responses in infants. In twelve preterm and term neonates (measured on 33 occasions), Slater *et al*. (2008) found moderate correlations (r = .57; p = .001) between scores of a multidimensional pain scale (Premature Infant Pain Profile-PIPP (Stevens et al., 1996)) and maximum change in the contralateral [HbT] in response to heel lance. Moreover, they showed that the cortical response had a stronger association with the behavioral components of the PIPP (i.e. facial expression) compared with the physiological components (i.e. HR and SaO_2 changes from baseline) (r = .4; p = .04). Although some correlations between the behavioral FLACC pain scores and cerebral hemodynamic responses were present in our study, they did not attain statistical significance. However, this is most certainly due to lack of power since in order to attain a statistically significant correlation between any of these signals, a minimal association of 0.55 was needed. Similar to our finding, Gelinas *et al*. (2011) did not find significant associations between behavioral pain scores, adult patient's self-report, and regional cerebral oxygenation (rSO_2) during nociceptive procedures, although taken individually, these parameters did show significant changes from their baseline measure.

Recently Frank *et al*. (2011), recommended that the COMFORT (Ambuel et al., 1992) pain scale be used as the main pain assessment instrument for recognizing post-operative and procedural pain in the critically ill neonate when compared to three other tools. The COMFORT scale was best at identifying analgesic effectiveness. This composite pain assessment instrument may be more suitable for critical care contexts where significant clinical factors (e.g. inotropes, ventilator support, etc) can impact pain

measures (Frank et al., 2011). The FLACC scale, not designed specifically for this population, is not sensitive enough to these and other factors.

Experimental findings have consistently demonstrated that women experience pain more intensely than men do (Fillingim et al., 2009; Goffaux et al., 2011; Riley et al., 1998). Nonetheless, the lower pain scores found in male infants in our study population could be explained by the significant impact (i.e. r = -.61) that midazolam had on their behavioral manifestations when compared to females; thus dampening males' capacity to express their pain experience. The exact mechanism underlying this gender difference and the role of midazolam calls for further investigations with a larger sample to insure reproducibility of these findings. As many physiological, structural, hormonal, psychological, and environmental factors influence how the human brain responds to noxious stimulation, whether and how these explain the gender differences in brain activation that we found would require a much larger sample size than was available in our study (Derbyshire, 2008).

Although we expected some level of pain in response to chest-drain removal, we did not anticipate the mean FLACC pain scores to be severe (7/10) since nearly all infants (n = 19) received an analgesic. Additionally, the sedating agent's (midazolam) ability to blunt/dampen the behavioral response to pain should be taken into account when conducting pain evaluations solely on the basis of behavioral manifestations. Although cerebral hemodynamics responses were decreased by pharmacological treatments, changes in [HbO_2] and [HbH] irrespective of hemisphere were still present in the infants with lower FLACC pain scores (e.g. FLACC 1/10 with Δ[HbO_2] Left, Right = 6.32, 1.12 and Δ[HbH] Left, Right = 7.29, 3.79 μmol/l). Thus, adding cerebral

measurements to a behavioral assessment may complement the pain evaluation when there is a risk of dampened behaviors, e.g., highly sedated or paralyzed patients. These data underscore the need for further investigations of pain responses in non-communicative patients which combine simultaneous cerebral and behavioral assessment.

A recent study by Slater *et al.* (2010) demonstrated that neonates were still showing somatosensory neuronal activity after receiving sucrose, a treatment known to be effective for reducing procedural pain response (Stevens et al., 2010), even though their behavioral pain scores were significantly lowered. The effectiveness of this widely used non-pharmacological pain treatment was questioned, but the validity of the behavioral pain measure was not questioned. This discrepancy stresses the importance of assessing the two dimensions of pain (Apkarian, 2008; International Association for the Study of Pain, 1979; Melzack & Wall, 1965): the *sensory* dimension as reflected by somatosensory hemodynamic activity and the *emotional* dimension as reflected by behavioral manifestations. Each dimension may be more differentially sensitive to non-pharmacological and pharmacological treatments. This again emphasizes the necessity of using a multidimensional approach to completely capture the pain experience, especially in non-verbal critically ill populations.

Several limitations of our study are apparent. First or foremost, our sample size limited the extent of our analyses and conceivably our ability to obtain statistically significant associations between cerebral hemodynamic changes and behavioral pain scores. Additionally, we could not perform multivariate statistical tests which would have allowed us to describe more precisely interactions between our variables and possibly

important clinical indicators. Second, optode placement and consistent interrogation of the same brain regions is a major concern with NIRS. Currently multichannel NIRS devices that cover multiple regions of the scalp are not readily available but their use in adult and older children functional NIRS research is increasing in popularity (Becerra et al., 2008; Hoshi, 2003; Hoshi & Chen, 2002).

Controlling for movement artefact and possible confounding environmental factors in this clinical research, although very difficult, represents an additional limitation to the study. However, since the purpose of this research was to explore, through the use of NIRS technology, cerebral hemodynamic changes to a frequently performed noxious procedure in critically ill patients, we wanted to capture changes in a routine clinical setting. For this reason, we elected not to interfere with standard care and apply a more strict control over possible confounding factors.

Using a multidimensional pain assessment approach, we demonstrated that significant cerebral, physiological and behavioral activity was present in response to a noxious procedure in critically ill infants despite the administration of analgesic treatment. The fact that the average FLACC scores were high and that the sedative agent, rather than analgesic treatment, reduced the pain ratings significantly may urge health-care professionals to question current pain management protocols during chest-drain removal in critically ill infant. Despite low pain scores in some infants, this group still showed relatively high cerebral hemodynamic activity in response to chest-drain removal. Hence, there is additional information from brain activity in these circumstances. Dampened behaviors by a sedating agent during a procedure known to be painful may jeopardize the discriminate validity of certain behavioral cues. This is

especially relevant when heavy sedation and/or blocking agents are administered. Further research using an approach that combines cerebral and behavioral evaluation is warranted.

ACKNOWLEDGEMENTS

This work was supported in part by the United States National Institutes of Health through grants R01EB001659, K24NS057568, and R21HD056009 by the National Institute of Biomedical Imaging and Bioengineering, the National Institute of Neurological Disorders and Stroke, and the National Institute of Child Health and Human Development, respectively. Further support was provided by the Lifebridge Fund. M. Ranger was supported by doctoral and international Fellowships from the Quebec Health Research Fund and the Quebec Interuniversity Nursing Intervention Research Group. We are very thankful to the nurses of the pre-operative clinic and CICU at Children's Hospital Boston and families of patients, whose generous help and participation made this work possible. We also thank our colleagues Dr. Mustafa Sulemanji and Ms. Heather O'Leary for significant contributions and guidance in the performance of this study.

Table 1. Demographic and clinical characteristics of our cohort (N= 20).

Mean age (months)	3.7 ±1.4; range 1.5-7
Mean weight (Kg)	5.5 ±1.6; range 2.8-10
Mean PRISM III scores	8.5 ± 3.9; range 3-18
Diagnosis: TOF (+ pulmonary stenosis) VSD & VSD/PFO ASD/VSD CAVC Combination of 2 or more Other	*n* = 7 *n* = 5 *n* = 3 *n* = 1 *n* = 2 *n* = 2
Mean morphine dose mg/kg/dose	0.05; range 0.0-0.2 (median 0.05)
Mean midazolam dose mg/kg/dose	0.04; range 0.0-0.1 (median 0.05)
Mean time from surgery at noxious procedure	32.9 hours; range 16-96 (median 24.5)
Location of chest drain	Midline (*n*= 16) Right (*n*= 4)
Type of chest drain	Chest tube (*n* = 2); Blake drain (*n* = 18)
Mean maximum pain score in last 24h (*n* = 18)	4.5/10; range 0-7

PRISM III: Pediatric RISk of Mortality; TOF: Tetralogy of Fallot; VSD: Ventricular septal defect; ASD: Atrial septal defect; PFO: Patent foramen ovale; PDA: Patent ductus arteriosis; CAVC: Complete Atrioventricular Canal; Combination: VSD/ASD/PDA, CAVC/TOF; Other: Aortic root aneurysm, hypoplastic left heart syndrome, Transitional AV canal; pain score was taken from patients' charts.

Table 2. FLACC pain scores regression analysis (*n* = 20)

Epochs	**Mean difference**	**95% CI**	***p*-value**
T_1-T_0[1]	3.00	[1.49, 4.51]	< .001
T_2-T_0[2]	6.45	[4.65, 8.25]	< .001
T_2-T_1[3]	3.45	[1.65, 5.25]	< .001

[1]For Tactile stimulation - Baseline
[2] For Chest-drain removal - Baseline
[3] For Chest-drain removal - Tactile stimulation

Table 3. Relationship (*p* value) between cerebral and physiological measures, and clinical factors (univariate analysis) during chest-drain removal.

Parameter	$\Delta[HbO_2]$	Δ[HbH]	ΔMAP	ΔHR	ΔSaO_2
T_1-T_0					
ΔMAP					
ΔHR	0.19 (.035)[1]				
ΔSaO_2					
Morphine	-137.78 (.005)[1]				
Midazolam	-114.87 (.017)[1]				-71.27 (.012)
Age					
Weight				4.6 (.032)	
T_2-T_0					
ΔMAP					
ΔHR					
ΔSaO_2					
Morphine					
Midazolam					
Age		-1.94 (.044)[1]			
Weight			3.93 (.029)		-1.46 (.002)
T_2-T_1					
ΔMAP					
ΔHR					
ΔSaO_2					
Morphine		-82.54 (.002)[1]		-90.24 (.022)	
Midazolam	105.97 (.029)[1]	-63.7 (.015)[1]		-146.63 (.005)	
		-137.49 (.008)[2]			
Age			2.9 (.044)		
Weight					-1.11 (.017)

1: Left hemisphere; 2: Right hemisphere.

$[HbO_2]$: oxygenated hemoglobin concentration; [HbH]: deoxygenated hemoglobin concentration; MAP: mean arterial blood pressure; HR: heart rate; SaO_2: arterial blood oxygen saturation.

T_0: baseline; T_1: tactile stimulus; T_2: noxious stimulation (chest-drain removal).

Table S1. Description of cerebral hemodynamic response change between Baseline and Chest-drain removal for infants with FLACC pain score above 5.

Sex	FLACC	L$\Delta[HbO_2]$	R$\Delta[HbO_2]$	LΔ[HbH]	RΔ[HbH]	Morphine	Midazolam
M	7	-2.94	-1.86	-1.05	.30	.05	.05
F	7	10.30	2.09	3.39	2.09	.00	.04
F*	7	-.51	.08	3.61	1.86	.05	.05
F	7	-12.16	16.38	-14.18	-.17	.10	.05
M	8	-5.03	-5.41	4.15	6.37	.03	.03
F*	8	7.98	2.86	.09	-.76	.05	.00
F	8	2.06	18.75	.66	12.81	.05	.00
F*	8	-.54	-4.38	12.25	5.30	.02	.02
M	8	-7.30	5.08	-3.37	3.35	.10	.05
M	8	-4.61	5.01	8.41	23.16	.05	.00
M*	8	3.85	.02	2.05	.91	.03	.03
M	9	-4.64	-3.84	-2.29	5.61	.05	.05
F	10	-2.70	-4.88	10.39	6.38	.05	.05
F	10	7.46	-1.26	6.50	-1.41	.05	.03
Mean(SD)	8(1)	-.63(6.35)	2.05(7.43)	2.19(6.62)	4.7(6.53)	.05(.03)	.03(.02)
Median	8	-1.62	.05	2.72	2.72	0.05	0.04

*: infants with Trisomy 21;
Face Leg Activity Cry Consolability (FLACC) pain scores at chest-drain removal – baseline;
$\Delta[HbO_2]$: oxygenated hemoglobin concentration difference between chest-drain removal and baseline in micromole/l; Δ[HbH]: deoxygenated hemoglobin concentration difference between chest-drain removal and baseline in micromole/l; L: left hemisphere, R: right hemisphere; Morphine: dose given in mg/kg/dose; Midazolam: dose given in mg/kg/dose; SD: standard deviation.

Table S2. Description of cerebral hemodynamic response change between Baseline and Chest-drain removal for infants with FLACC pain score less than 4.

Sex	FLACC	L$\Delta[HbO_2]$	R$\Delta[HbO_2]$	LΔ[HbH]	RΔ[HbH]	Morphine	Midazolam
M	0	.15	-1.74	.67	.05	.05	.05
M	1	-2.92	1.54	1.95	-.13	.05	.05
M	1	6.32	1.12	7.29	3.79	.10	.10
M	2	.37	-4.65	3.25	-.95	.09	.09
M*	4	-1.58	2.09	3.52	2.78	.07	.07
Mean(SD)	1.6(1.5)	.47(3.54)	-.33(2.83)	3.34(2.49)	1.11(2.05)	.07(.02)	.07(.02)
Median	1	.15	1.12	3.25	.05	.07	.07

*: infants with Trisomy 21;
Face Leg Activity Cry Consolability (FLACC) pain scores at chest-drain removal – baseline;
$\Delta[HbO_2]$: oxygenated hemoglobin concentration difference between chest-drain removal and baseline in micromole/l; Δ[HbH]: deoxygenated hemoglobin concentration difference between chest-drain removal and baseline in micromole/l; L: left hemisphere, R: right hemisphere; Morphine: dose given in mg/kg/dose; Midazolam: dose given in mg/kg/dose; SD: standard deviation.

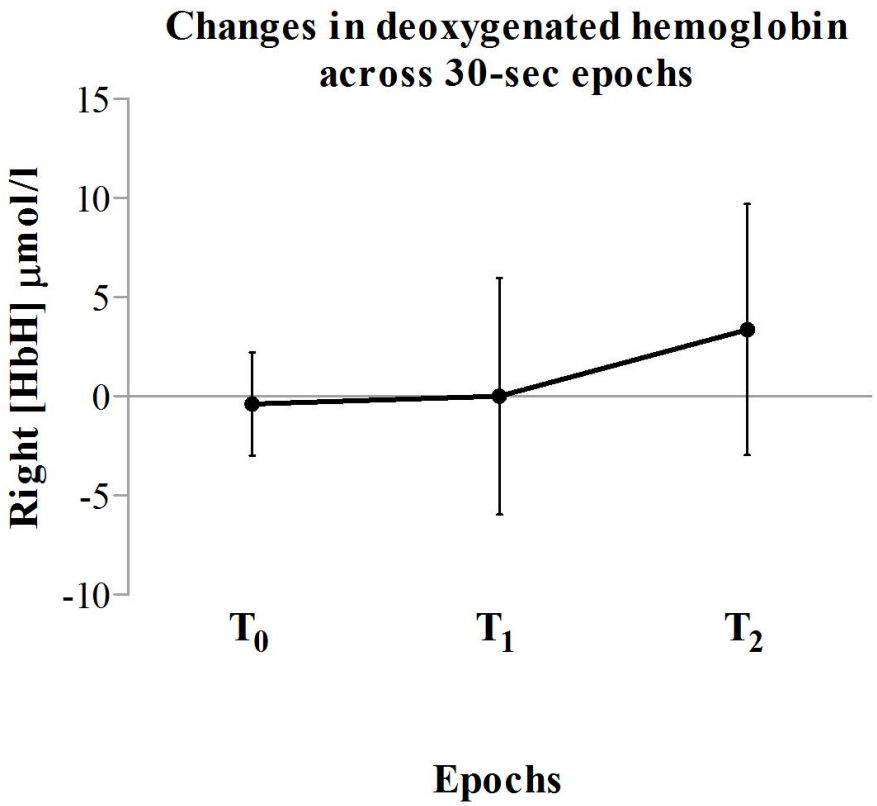

Figure 1. Changes in right hemisphere [HbH] values in all subjects (n = 19). The figure shows significant [HbH] increases from baseline (T_0) to noxious stimulation (T_2) $*p < 0.05$. Vertical bars denote the 95% confidence intervals and the middle points denote the mean values.
T_0: baseline; T_1: tactile stimulation; T_2: noxious stimulation.

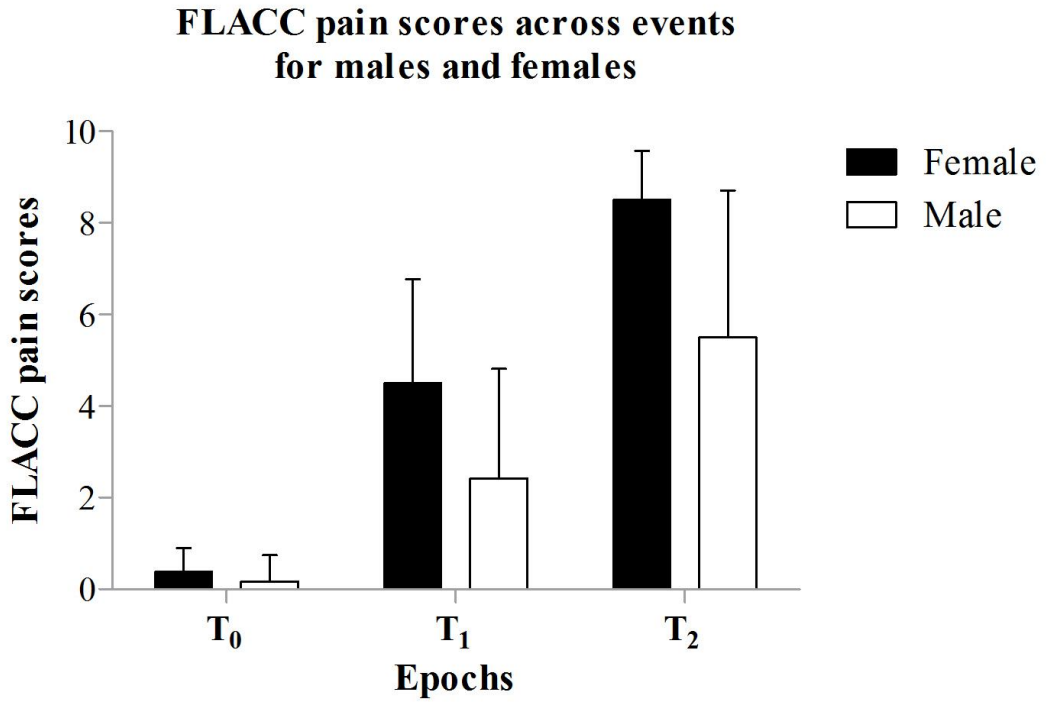

Figure 2. Mean FLACC pain scores for baseline (T_0), tactile (T_1), and noxious (T_2) events between males (n = 12) and females (n = 8). The figure shows significant differences in overall FLACC scores across the three events ($p < 0.001$) and between males and females ($p < 0.01$).

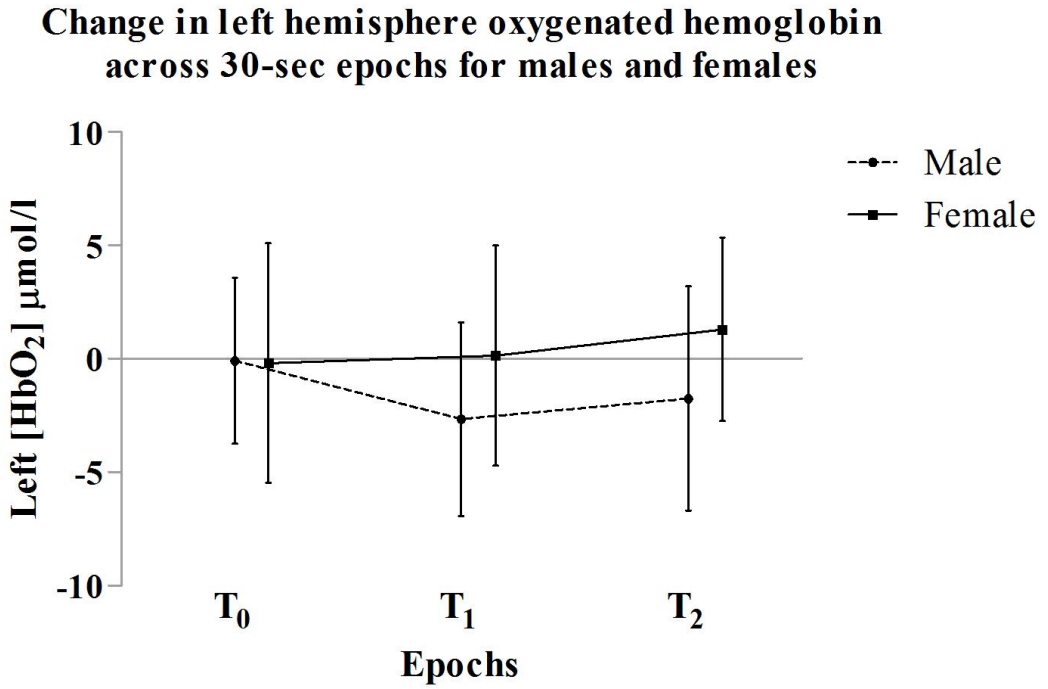

Figure S1. Mean changes in left hemisphere [HbO_2] across events in males (n = 12) and females (n = 8). Vertical bars denote the 95% confidence intervals and the middle points denote the mean values. T_0: baseline; T_1: tactile stimulation; T_2: noxious stimulation.

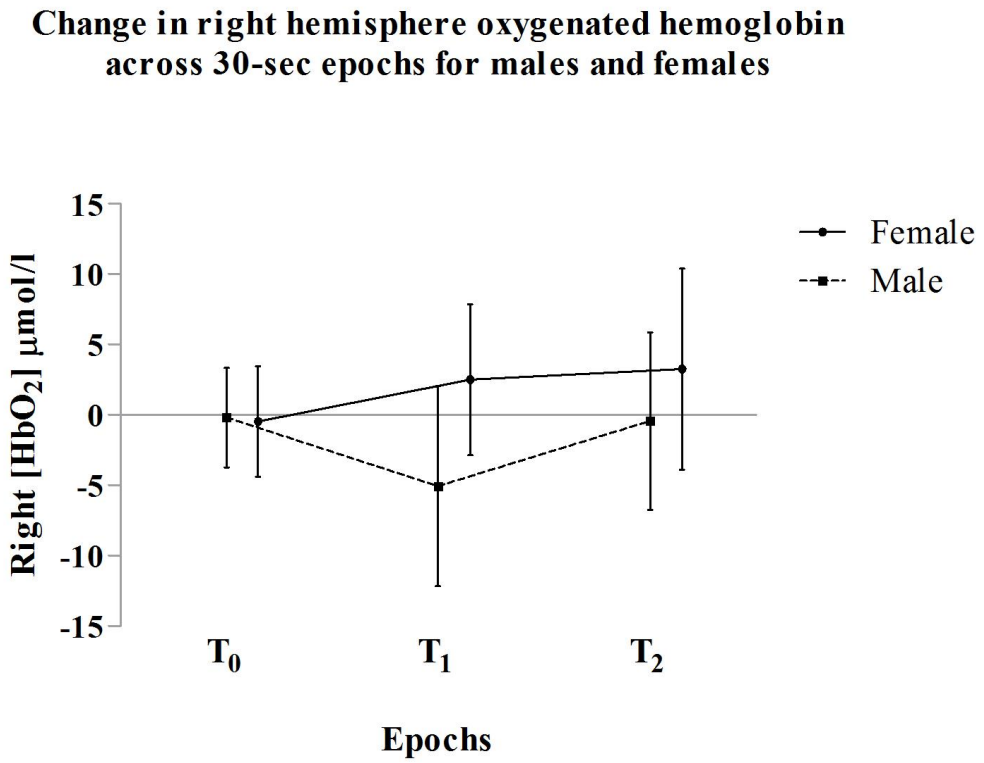

Figure S2. Mean changes in right hemisphere [HbO_2] across events in males (n = 11) and females (n = 8). Vertical bars denote the 95% confidence intervals and the middle points denote the mean values. T_0: baseline; T_1: tactile stimulation; T_2: noxious stimulation.

CHAPTER 5: FINAL CONCLUSION AND SUMMARY

The overall objective of this dissertation was to describe the regional cerebral haemodynamic changes, as measured by NIRS, in critically ill infants in response to a noxious stimulation. In view of the fact that significant changes were measured, further analyses were pursued to explore the relationship between the cerebral haemodynamic and systemic physiological responses, as well as behavioural pain scores (FLACC scale). Three manuscripts resulted from this work. The first manuscript reviewed the literature regarding the use of cerebral near-infrared spectroscopy technique as a measure of cerebrovascular activation in response to nociceptive stimuli in infants. Manuscript #2 focused on the main objective of the study and described the haemodynamic changes in the somatosensory cortical region following noxious stimulation activation in critically ill infants as measured by NIRS. The relationship between NIRS measurements and systemic physiological parameters was also presented in this article. The third manuscript centered on the secondary objective of the study and reported on the associations between cerebral haemodynamic changes, systemic physiological responses, and behavioural pain scores in a sub-sample of 20 infants in which adequate video footage was available for pain coding.

The following section describes the main research findings congruent with our research hypotheses, bearing in mind the limitations of this work.

Hypothesis 1

We hypothesized that an acute noxious stimulation would be associated with significant regional cerebral haemodynamic changes in critically ill infants which could be measured using NIRS. The main research findings supported this hypothesis. As such,

we have shown that it was possible through the use of NIRS to monitor significant cerebral haemodynamic changes in response to chest-drain removal in critically ill infants following open-heart surgery for CHD. To our surprise, it was the deoxygenated haemoglobin concentration ([HbH]), and not the oxygenated haemoglobin concentration ([HbO_2]), that significantly increased in response to tactile and, even more so, to noxious stimulation when compared to baseline ($p<.01$). It was difficult to compare these findings (i.e. changes in [HbH]) with results from previous studies since this parameter is generally not reported (most authors have reported changes in [HbO_2] and/or [HbT]). Moreover, regional cerebral activation typically results in regional increases in both oxygenated haemoglobin concentrations [HbO_2] and [HbT] with a decrease in [HbH] (Hoshi, 2003). However, contrasting results have been reported, such as no change or increases in [HbO_2] with increases in both [HbH] and [HbT] (Hoshi & Tamura, 1993; Kato et al., 1993; Kleinschmidt et al., 1996). Nonetheless, [HbT] increased significantly across the epochs ($p<.05$) in our sample, and females showed bilateral increases in [HbO_2], which were significantly larger than in male infants, who showed bilateral decreases in [HbO_2] in response to the procedure.

When we added clinical factors into our analyses, we found that administered pharmacological treatments and age of infants had significant effects on cerebral haemodynamic responses. More precisely, we demonstrated that the administered dose of morphine significantly dampened the change in right hemisphere [HbO_2] ($p<.05$), explaining 26% ($R^2 = .26$) of the variance in its response to noxious stimulation. The age of infants was also shown to affect both cerebral haemodynamic indicators, explaining approximately 45% of the variance in the respective responses. The inverse association

between administered dose of morphine and the change in [HbO_2] suggests that this analgesic aids in blunting the cerebrovascular response to the nociceptive stimulus. As such, morphine appears to decrease pain-induced neurovascular *activation* (i.e. regional increase in blood flow and [HbO_2] with decreases in [HbH], resulting from a possible increase in neuronal activity), thus dampening the cortical activity in response to noxious stimulation and confirming its inhibitory effect (Apkarian et al., 2005; Schlaepfer et al., 1998). These data suggest that this non-invasive portable cerebral haemodynamic monitoring system appears to be able to help discriminate between a painful and non-painful stimulation, and, in addition, is sensitive to the administration of analgesic medication.

As stated previously, changes in [HbH] are generally not reported in the functional NIRS literature. This lack of reference values or previous results contributes to the difficulty in interpreting our results. Recently, Leff et al. (2011) recommended that researchers report on all haemodynamic parameters (HbO_2, HbH, HbT) and not just one *best* indicator to improve specificity. Although [HbO_2] is often the parameter of choice, variations in [HbH] have been described as being more closely linked to neuronal activity, and possibly better indicative of cellular oxygen consumption than [HbO_2] (Oyama, Kondo, Komatsu, & Sugiura, 2009).

Furthermore, based on our results, it appears that in response to chest-drain removal, neuronal activation was associated with an increase in cerebral oxygen consumption that surpassed a possible increase in oxygen delivery, as reflected in a significant increase in [HbH] while [HbO_2] remained relatively constant. Finally, while there have been significant advances in the last decade in research using NIRS to study

variations in cerebral oxygenation and haemodynamics of humans, understanding how blood flow, metabolism, and neuronal activity interact to affect the NIRS signals remains incomplete (Bartocci, 2006; Oyama et al., 2009) and warrants further study.

Hypothesis 2

We hypothesized that regional cerebral haemodynamic changes in critically ill infants (measured with NIRS) would correlate moderately with physiological indicators (vital signs) and behavioural pain assessment scores (measured with the FLACC scale). This second hypothesis was not supported. While we found that both physiological indicators (i.e. MAP and HR) and behavioural pain scores as measured with the FLACC scale significantly increased across the epochs, these changes were not associated with cerebral haemodynamic changes. Thus, construct validation between cerebral haemodynamic responses as measured by NIRS, and pain behavioural scores as measured with the FLACC scale remains questionable.

Based on previous reports supporting associations between changes in cerebral and systemic haemodynamic (i.e MAP), changes during nociceptive events in sick infants (Limperopoulos et al., 2008; Soul et al., 2007), we were expecting to find correlations between these parameters in our population. While both cerebral NIRS measures ([HbH]) and vital signs showed significant changes in response to the painful procedure, the measured cerebral changes were surprisingly uncorrelated of the systemic haemodynamic response.

In response to chest-drain removal, average FLACC pain scores were high (mean scores 7/10) despite the administration of standard doses of pharmacological treatment. This may urge health-care professionals to question current pain management protocols

during chest-drain removal in critically ill infants. To our surprise, we found that it was the sedating agent that significantly dampened (by 36%) the behavioural pain scores. This was more evident in males, who had significantly lower FLACC scores when compared to females. However, significant cerebral haemodynamic changes still occurred, thus there is additional information from brain activity that can be measured with NIRS when a sedative is administered. Hence, dampened behavioural responses by a sedating agent during a procedure known to be painful may jeopardize the discriminative value of these cues (i.e. pain behavioural indictors).

Our results, however, may reflect certain limits of the behavioural FLACC scale. According to Frank and colleagues' (2011) findings, the COMFORT scale could perhaps be a more appropriate choice for recognizing post-operative and procedural pain in a critically ill infant population. In this study, the COMFORT scale was best at identifying analgesic effectiveness and discriminating between procedure pain intensity levels. As such, the COMFORT composite pain assessment instrument may be more suitable for critical care contexts where significant clinical factors (e.g. inotropes, ventilator support, severity of illness, diagnosis, etc) can impact pain measures. This scale includes an indicator related to level of sedation (i.e. includes one indicator measuring alertness), which the FLACC does not, and as such, can account for medications typically used in critical care settings.

In circumstances where morphine was administered, we demonstrated with a multidimensional pain measurement approach that significant cerebral, physiological and behavioural activity were present in response to a noxious procedure in critically ill infants. Based on these findings, it seems that NIRS has potential as a technique for

assessing pain evoked cerebral activation in critically ill infants. However, given the paucity of research supporting its use in pain measurement in this population, the clinical use of NIRS is currently not recommended. At this time, NIRS remains a promising research tool that has potential to improve our understanding of pain perception. In moving forward, it will be important to determine what constitutes a clinically significant change in the measured parameters (i.e. [HbO_2] and [HbH]) when compared with normal fluctuations that occur in the brain tissue, by sampling both healthy and critically ill infants of differing developmental ages. In addition, conducting research with appropriate sample sizes, evaluating response repeatability and reliability, as well providing results on time to peak/time to nadir and back to baseline are just a few examples of recommended measures to improve NIRS research (Leff et al., 2011).

CONTRIBUTIONS

Our preliminary findings provide important insights on higher order pain processing and should inspire future studies. In our sample of critically ill infants we were able to capture activation of the cortical nociceptive system with NIRS. We demonstrated that although pharmacological treatment was provided, pain was still present as reflected by localized changes in cerebral blood flow and oxygen consumption that surpassed its supply.

To our knowledge, this is the first study to describe continuous cerebral haemodynamic responses coupled with systemic cardiovascular activity and behavioural manifestations during routine chest-drain removal in critically ill infants. In describing both oxygenated and deoxygenated haemoglobin concentration changes in response to a noxious event, this study has provided a more complete representation of cerebral

activation than has been reported in the past. Moreover, controlling for the effects that pharmacological treatments have on the measured indicators has not been previously reported and is an additional strength of this thesis.

The use of a multimodal approach to assess pain in non-communicative critically ill infants revealed certain limitations in solely using behavioural cues to evaluate pain in certain clinical contexts, such as when sedating treatments are administered. Although cerebral haemodynamics, systemic physiological and behavioural pain indicators were not found to be associated with each other, they all showed a significant change from their baseline measures in response to pain. Thus, each means of assessment or *output modality* showed their own strengths and limitations, as well as illustrating their complementarity.

LIMITATIONS

Limitations of this research included issues related to sample size (lack of power for some analyses), data collection, the complexity of the setting and the sample population, the control of confounding variables, as well as the NIRS technique itself. These limitations were presented in manuscript #1, #2 and #3, and are summarized below.

Our sample size limited the extent of our analyses and conceivably our ability to attain statistically significant associations between cerebral haemodynamic changes and behavioural pain scores. Additionally, we could not perform multivariate statistical tests, which would have allowed us to examine more precisely interactions between our variables and possibly important clinical indicators. A larger sample size is certainly needed to further assess and evaluate potentially confounding factors, such as

administration of various medications, co-morbidity and diagnosis, chest-drain type and placement, to mention just a few.

The selection of 30second epochs, which was based on previous reports on infant pain assessment, specifically the Premature Infant Pain Profiles (PIPP) used in numerous studies of neonates and infants (Stevens, Johnston, Taddio, Gibbins, & Yamada, 2010), may not have been the most appropriate choice. However, chest-drain removal being a very brief event lasting less than 5 seconds, a longer epoch duration of 60second, such as employed by Bartocci *et al.* (2006), may not have reflected well the cerebral response we were aiming to capture. Additionally, our data analysis approach (i.e. taking averages of haemodynamic indicators) may have washed-out important information. Conducting temporal dynamic trends of the observed cortical haemodynamic responses of this brief event may have been a better option, allowing better insight on the unfolding of the neurovascular coupling process.

Optode placement and consistent interrogation of the same brain regions is a major concern with NIRS. Ideally, the accuracy of the interrogated cerebral region by the NIRS device should be verified by simultaneous MR imaging. In clinical research, this is extremely difficult to achieve. Thus, we were limited in our ability to ascertain that we assessed the somatosensory cortial region. In the near future, the use of multichannel NIRS devices that cover multiple regions of the scalp (Becerra et al., 2008; Hoshi, 2003; Hoshi & Chen, 2002) and whole head coverage investigations (Franceschini, Joseph, Huppert, Diamond, & Boas, 2006), may help resolve this issue. Finally, controlling for movement artefact and possible confounding environmental factors in this clinical study, although challenging, represents an additional limitation to the study. The motor center

being adjacent to the somatosensory region, it is not possible at this time to ascertain that the motor area was not accessed during our data collection. This must be kept in mind when interpreting our results.

Conducting clinical research has its share of issues but it is through rigorous and well planned studies that we can surmount these obstacles and gain significant insight into the complex interactions between various stimuli and cortical activity in critically ill infants (Wolfberg & du Plessis, 2006). Several confounding factors may have affected our study results due to the complex nature of the PICU population that participated in the study (e.g., diagnosis, severity of illness, haemodynamic and respiratory instability, medication administration, etc). Additionally, important limitations regarding the validity and reliability of NIRS must be taken into account in attempting to generalize our findings. With all pain assessment tools, validation is an everlasting process. All pain indicators in infants require series of validation studies with replication, and NIRS would be no exception. Nevertheless, these limitations should not prevent researchers from pursing ongoing studies using NIRS. Studies are surely needed to advance our understanding of the complex interaction between these different *output modalities* (behavioural manifestations, cerebral haemodynamic, and physiological activity) of the pain experience in critically ill infants.

IMPLICATIONS

Practice

A significant finding of this study was that the FLACC scale, a commonly used behavioural pain measure in the PICU, had limitations in the context of sedation; whereas cerebral haemodynamic changes, measured with NIRS, indicated nociception. The

sedating agent midazolam dampened FLACC pain scores while the analgesic morphine did not. Thus, the administration of midazolam seemed to lessen the infants' capacity to express their pain experience in response to the chest-drain removal. It is now apparent that adding cerebral measurements to a behavioural assessment may complement the pain evaluation when there is a risk of dampened behaviours, such as when dealing with highly sedated or paralyzed patients.

Although we expected some behavioural manifestations in response to chest-drain removal, the average pain scores in our sample revealed severe pain (7/10), which could have been even higher if the sedating agent had not been administered in 81% of infants. Actually, the three infants that did not receive midazolam prior to the procedure had FLACC pain scores in response to chest-drain removal of 8/10. These results may urge health care professionals to question current pain management protocols during chest-drain removal in critically ill infants. Our findings will hopefully lead to a better pain management care plan for this vulnerable population.

A recently published Position Statement for pain assessment in the patient unable to self-report (Herr, Coyne, McCaffery, Manworren, & Merkel, 2011) stressed the significant role nurses play in ensuring the assessment and treatment of pain in vulnerable populations such as, infants and pre-verbal toddlers, as well as the critically ill patient. Certain limitations of common approaches to assessment such as behavioral displays and physiological indicators were pointed out. Pain behaviors were viewed as not specific reflections of pain intensity, and in some cases indicative of other source of distress, such as physiological or emotional distress. It was pointed out that no single objective assessment strategy, such as interpretations of behaviors, pathology or estimates of pain

by observers, is sufficient by itself. This position statement corroborates with our view that a multidimensional pain assessment approach is necessary to allow a more complete view of the pain experience and may overcome the limitations of each single means of assessment. Nurses' clinical judgment is also crucial and we believe that our results demonstrate that pain assessment is not a straightforward task that is done without thinking; it is essential that nurses keep this in mind when conducting a pain assessment in a at risk population.

Future research

We believe that the findings from this dissertation may contribute theoretically and empirically. They set the stage for future studies and, possibly add to the evidence for consideration of NIRS technology as a complementary objective non-invasive approach to pain assessment infants. Results from this thesis assisted in advancing the reliability and validity properties of NIRS as a measure of cerebral haemodynamic changes, contributing to its usefulness as a bedside research instrument. We have increased our understanding of cerebral and systemic haemodynamic responses of critically ill infants to routine chest-drain removal. This was achieved, for the first time, through continuous measurement of cerebral haemodynamic responses coupled with systemic cardiovascular activity (i.e. continuous systemic arterial blood pressure, heart rate, and arterial oxygen saturation). Examining the effects that various demographic (such as age and weight) and clinical factors (such as pharmacological treatments) have on the cerebral haemodynamic response during this commonly performed noxious procedure is an additional important contribution of this study.

We sought to increase our understanding of higher level pain processing in critically ill infants, and although our preliminary findings contribute new information related to this field, it may be premature to draw definitive conclusions. Thus, the need for further investigations of pain responses in infants which combine simultaneous cerebral and behavioural assessment was highlighted within this dissertation and is certainly needed.

CONCLUSION

The evidence revealed in this dissertation has elucidated the complex nature of pain demonstrated by the large degree of between-subject-variability in the measured cerebral response to the noxious event. Nonetheless, the main objective of this thesis has been realized as shown by the ability of NIRS to detect significant regional cerebral haemodynamic changes in critically ill infants during a noxious procedure. Our study findings enhance the *psychometric properties* of this novel means of pain assessment. This non-invasive portable technique seems especially useful in situations in which the reliability of behavioural pain assessment instruments may be affected by sedation or paralytic agents. Replication of these findings is essential to allow more robust conclusions related to NIRS clinical utility as a complementary pain assessment instrument in critically ill infants.

REFERENCES

Ambuel, B., Hamlett, K. W., Marx, C. M., & Blumer, J. L. (1992). Assessing distress in pediatric intensive care environments: the COMFORT scale. *Journal of Pediatric Psychology, 17(1),* 95-109.

Anand, K. J. S. (1998). Neurophysiological and Neurobiological Correlates of supraspinal pain processing: Measurement Techniques. In A.Finley & P. McGrath (Eds.), *Measurement of pain in infants and children* (pp. 21-46). Seattle: IASP Press.

Anand, K. J. S. (2007). Pain Assessment in Preterm Neonates. *Pediatrics, 119,* 605-607.

Anand, K. J. S. & Craig, K. D. (1996). New perspectives on the definition of pain. *Pain, 67,* 3-6.

Apkarian, A. V. (2008). Pain perception in relation to emotional learning. *Current Opinion in Neurobiology, 18,* 464-468.

Apkarian, A. V., Bushnell, M. C., Treede, R. D., & Zubieta, J. K. (2005). Human brain mechanisms of pain perception and regulation in health and disease. *European Journal of Pain, 9,* 463-484.

Bai, J., Hsu, L., Tang, Y., & van Dijk, M. Validation of the COMFORT Behavior Scale and the FLACC Scale for Pain Assessment in Chinese Children after Cardiac Surgery. *Pain Management Nursing, In Press, Corrected Proof*.

Barr, R. G. (1998). Reflections on measuring pain in infants: dissociation in responsive systems and "honest signalling". *Arch.Dis.Child.Fetal Neonatal Ed., 79,* F152-F156.

Bartocci, M. (2006). *Brain Functional Near Infrared Spectroscopy in Human Infants.* PhD Karolinska Institutet. http://diss.kib.ki.se/2006/91-7357-034-6

Bartocci, M., Bergqvist, L. L., Lagercrantz, H., & Anand, K. J. (2006). Pain activates cortical areas in the preterm newborn brain. *Pain, 122(1-2),* 109-17.

Bartocci, M., Winberg, J., Papendiek, G., Mustica, T., Serra, G., & Lagercrantz, H. (2001). Cerebral Hemodynamic Response to Unpleasant Odors in the Preterm Newborn Measured by Near-Infrared Spectroscopy. *Pediatric Research, 50,* 324-330.

Bartocci, M., Winberg, J., Ruggiero, C., Bergqvist, L. L., Serra, G., & Lagercrantz, H. (2000). Activation of olfactory cortex in newborn infants after odor stimulation: a functional near-infrared spectroscopy study. *Pediatric Research.48(1),* 18-23.

Bassan, H., Benson, C. B., Limperopoulos, C., Feldman, H. A., Ringer, S. A., Veracruz, E. et al. (2006). Ultrasonographic Features and Severity Scoring of Periventricular Hemorrhagic Infarction in Relation to Risk Factors and Outcome. *Pediatrics, 117,* 2111-2118.

Becerra, L., Harris, W., Joseph, D., Huppert, T., Boas, D. A., & Borsook, D. (2008). Diffuse optical tomography of pain and tactile stimulation: Activation in cortical sensory and emotional systems. *NeuroImage, 41,* 252-259.

Becerra, L., Morris, S., Bazes, S., Gostic, R., Sherman, S., Gostic, J. et al. (2006). Trigeminal neuropathic pain alters responses in CNS circuits to mechanical (brush) and thermal (cold and heat) stimuli. *J Neurosci, 26,* 10646-10657.

Benaron, D. A., Hintz, S. R., Villringer, A., Boas, D., Kleinschmidt, A., Frahm, J. et al. (2000). Noninvasive functional imaging of human brain using light. *Journal of Cerebral Blood Flow & Metabolism., 20,* 469-477.

Bijur,P.E.P.M., Esses,D.M., Birnbaum,A.M., Chang,A.K.M., Schechter,C.M., and Gallagher,E.J.M. (2008). Response to Morphine in Male and Female Patients: Analgesia and Adverse Events. *Clinical Journal of Pain, 24,* 192-198.Brazy, J. E., Lewis, D. V., Mitnick, M. H., & Jobsis, V. (1985). Noninvasive monitoring of cerebral oxygenation in preterm infants: preliminary observations. *Pediatrics,75(2),* 217-25.

Brink, P. & Wood, M. (1998). *Advanced design in nursing research*. (2nd ed.) Thousand Oaks, California: Sage publications.

Buttner, W. & Finke, W. (2000). Analysis of behavioural and physiological parameters for the assessment of postoperative analgesic demand in newborns, infants and young children: a comprehensive report on seven consecutive studies. *Pediatric Anesthesia, 10,* 303-318.

Carlsson, J., Lagercrantz, H., Olson, L., Printz, G., & Bartocci, M. (2008). Activation of the right fronto-temporal cortex during maternal facial recognition in young infants. *Acta Paediatr., 97,* 1221-1225.

Carnevale, F. A. (1997). The experience of critically ill children: narratives of unmaking. *Intensive & Critical Care Nursing, 13(1),* 49-52.

Carnevale, F. A. & Razack, S. (2002). An item analysis of the COMFORT scale in a pediatric intensive care unit. *Pediatr Crit Care Med, 3,* 177-180.

Casey, K. L., Svensson, P., Morrow, T. J., Raz, J., Jone, C., & Minoshima, S. (2000). Selective Opiate Modulation of Nociceptive Processing in the Human Brain. *Journal of Neurophysiology, 84,* 525-533.

Comer,S., Cooper, Z., Kowalczyk, W., Sullivan, M., Evans, S., Bisaga, A., and Vosburg, S. (2010). Evaluation of potential sex differences in the subjective and analgesic effects of morphine in normal, healthy volunteers. *Psychopharmacology, 208,* 45-55.

Craig, K. D., Versloot, J., Goubert, L., Vervoort, T., & Crombez, G. (2010). Perceiving pain in others: automatic and controlled mechanisms. *J.Pain, 11,* 101-108.

Craig, K. D., Whitfield, M. F., Grunau, R. V. E., Linton, J., & Hadjistavropoulos, H. D. (1993). Pain in the preterm neonate: behavioural and physiological indices. *Pain, 52,* 287-299.

Derbyshire, S. W. G. (2008). Gender, Pain, and the Brain. *Pain: Clinical Updates, 16,* 1-4.

Derbyshire, S. W. G. & Osborn, J. (2007). Modeling Pain Circuits: How Imaging May Modify Perception. *Neuroimaging Clinics of North America, 17,* 485-493.

du Plessis, A. J., Newburger, J., Jonas, R. A., Hickey, P., Naruse, H., Tsuji, M. et al. (1995). Cerebral oxygen supply and utilization during infant cardiac surgery. *Ann Neurol., 37,* 488-497.

Duhn, L. J. & Medves, J. M. (2004). A systematic integrative review of infant pain assessment tools. *Advances in Neonatal Care, 4(3),* 126-40.

Duncan, A., Meek, J. H., Clemence, M., Elwell, C. E., Fallon, P., Tyszczuk, L. et al. (1996). Measurement of cranial optical path length as a function of age using phase resolved near infrared spectroscopy. *Pediatric Research, 39(5),* 889-94.

Dunwoody, C. J., Krenzischek, D. A., Pasero, C., Rathmell, J. P., & Polomano, R. C. (2008). Assessment, Physiological Monitoring, and Consequences of Inadequately Treated Acute Pain. *Pain Management Nursing, 9,* 11-21.

Fallon, P., Roberts, I., Kirkham, F. J., Elliott, M. J., Lloyd-Thomas, A., Maynard, R. et al. (1993). Cerebral hemodynamics during cardiopulmonary bypass in children using near-infrared spectroscopy. *Ann Thorac.Surg., 56,* 1473-1477.

Fabrizi,L., Slater,R., Worley,A., Meek,J., Boyd,S., Olhede,S., and Fitzgerald,M. (2011). A Shift in Sensory Processing that Enables the Developing Human Brain to Discriminate Touch from Pain. *Curr. Biol., 21(18),* 1552-1558.

Fillingim,R.B., King,C.D., Ribeiro-Dasilva,M.C., Rahim-Williams,B., and Riley,J.L., III (2009). Sex, gender, and pain: a review of recent clinical and experimental findings. *J. Pain, 10,* 447-485.

Finley, G. A. & McGrath, P. (1998). Introduction: The Roles of Measurement in Pain Management and Research. In G.A.Finley & P. McGrath (Eds.), *Measurement on Pain in Infants and Children* (pp. 1-4). Seattle: IASP Press.

Foster, R. L. (2001). Nursing judgment: the key to pain assessment in critically ill children. *Journal of the Society of Pediatric Nurses, 6(2),* 90-3, 96.

Franceschini, M. A., Joseph, D. K., Huppert, T. J., Diamond, S. G., & Boas, D. A. (2006). Diffuse optical imaging of the whole head. *J.Biomed.Opt., 11,* 054007.

Gelinas, C., Choiniere, M., Ranger, M., Denault, A., Deschamps, A., & Johnston, C. (2011). Toward a new approach for the detection of pain ináadult patients undergoing cardiac surgery: Near-infrared spectroscopy--A pilot study. *Heart & Lung: The Journal of Acute and Critical Care, 39,* 485-493.

Goffaux,P., Michaud,K., Gaudreau,J., Chalaye,P., Rainville,P., and Marchand,S. (2011). Sex differences in perceived pain are affected by an anxious brain. *Pain, 152,* 2065-2073.

Gusnard, D. A., Raichle, M. E., & Raichle, M. E. (2001). Searching for a baseline: functional imaging and the resting human brain. *Nature Reviews Neuroscience, 2(10),* 685-94.

Hamamastu Photonics. (2003). NIRO-200 Near Infrared Oxygenation Monitor.

Herr, K., Coyne, P. J., Key, T., Manworren, R., McCaffery, M., Merkel, S. et al. (2006). Pain Assessment in the Nonverbal Patient: Position Statement with Clinical Practice Recommendations. *Pain Management Nursing, 7,* 44-52.

Herr, K., Coyne, P.J., McCaffery, M., Manworren, R, & Merkel, S. Pain Assessment in the Patient Unable to Self-Report: Position Statement with Clinical Practice Recommendations. *Pain Management Nursing, 12(4),* 230-250.

Hirth, C., Obrig, H., Villringer, K., Thiel, A., Bernarding, J., Muhlnickel, W. et al. (1996). Non-invasive functional mapping of the human motor cortex using near-infrared spectroscopy. *Neuroreport, 7,* 1977-1981.

Hodge, C. J., Jr. & Apkarian, A. V. (1990). The spinothalamic tract. *Crit Rev.Neurobiol., 5,* 363-397.

Hofbauer, R. K., Rainville, P., Duncan, G. H., & Bushnell, M. C. (2001). Cortical representation of the sensory dimension of pain. *J.Neurophysiol., 86,* 402-411.

Hoshi, Y. (2003). Functional near-infrared optical imaging: utility and limitations in human brain mapping. *Psychophysiology, 40,* 511-520.

Hoshi, Y. & Chen, S. J. (2002). Regional cerebral blood flow changes associated with emotions in children. *Pediatr Neurol., 27,* 275-281.

Hoshi, Y. & Tamura, M. (1993). Detection of dynamic changes in cerebral oxygenation coupled to neuronal function during mental work in man. *Neurosci Lett., 150,* 5-8.

Hummel, P. & van Dijk, M. (2006). Pain assessment: Current status and challenges. *Seminars in Fetal and Neonatal Medicine, 11,* 237-245.

International Association for the Study of Pain (1979). Editorial The need of a taxonomy. *Pain, 6,* 247-252.

International Electrotechnical Commission (2007). Safety of laser products - Part 1: Equipment classification and requirements. International Electrotechnical Commission on-line store [On-line]. Available: http://webstore.iec.ch/webstore/webstore.nsf/artnum/037864?opendocument

Ista, E., van Dijk, M., Tibboel, D., & de Hoog, M. (2005). Assessment of sedation levels in pediatric intensive care patients can be improved by using the COMFORT "behavior" scale. *Pediatric Critical Care Medicine, 6(1),* 58-63.

Jobsis, F. F. (1978). Noninvasive, infrared monitoring of cerebral and myocardial oxygen sufficiency and circulatory parameters. *Science.,* 1264-1267.

Johnston, C. C. (1998). Psychometric Issues in the Measurement of Pain. In G.A.Finley & P. McGrath (Eds.), *Measurement of Pain in Infants and Children* (pp. 5-20). Seattle: IASP Press.

Johnston, C. C. & Stevens, B. J. (1996). Experience in a Neonatal Intensive Care Unit Affects Pain Response. *Pediatrics, 98,* 925-930.

Johnston, C. C., Stevens, B. J., Franck, L. S., Jack, A., Stremler, R., & Platt, R. (1999). Factors Explaining Lack of Response to Heel Stick in Preterm Newborns. *Journal of Obstetric, Gynecologic, & Neonatal Nursing, 28,* 587-594.

Johnston, C. C., Stevens, B. J., Yang, F., & Horton, L. (1995). Differential response to pain by very premature neonates. *Pain, 61,* 471-479.

Johnston, C. C. & Strada, M. E. (1986). Acute pain response in infants: a multidimensional description. *Pain, 24,* 373-382.

Johnston, C. C., Stremler, R. L., Stevens, B. J., & Horton, L. J. (1997). Effectiveness of oral sucrose and simulated rocking on pain response in preterm neonates. *Pain, 72(1-2),* 193-9.

Kato, T., Kamei, A., Takashima, S., & Ozaki, T. (1993). Human visual cortical function during photic stimulation monitoring by means of near-infrared spectroscopy. *J Cereb.Blood Flow Metab, 13,* 516-520.

Kleinschmidt, A., Obrig, H., Requardt, M., Merboldt, K. D., Dirnagl, U., Villringer, A. et al. (1996). Simultaneous recording of cerebral blood oxygenation changes during human brain activation by magnetic resonance imaging and near-infrared spectroscopy. *J Cereb.Blood Flow Metab, 16,* 817-826.

Klem, G. H., Luders, H. O., Jasper, H. H., & Elger, C. (1999). The ten-twenty electrode system of the International Federation. The International Federation of Clinical Neurophysiology. *Electroencephalography & Clinical Neurophysiology – Supplement 52,* 3-6.

Lagercrantz, H. (2009). The birth of consciousness. *Early Hum.Dev., 85,* S57-S58.

Lasky, R. E. & van, D. W. (2010). Is sucrose an effective analgesic for newborn babies? *Lancet, 376,* 1201-1203.

Leff, D. R., Orihuela-Espina, F., Elwell, C. E., Athanasiou, T., Delpy, D. T., Darzi, A. W. et al. (2011). Assessment of the cerebral cortex during motor task behaviours in adults: a systematic review of functional near infrared spectroscopy (fNIRS) studies. *NeuroImage, 54,* 2922-2936.

Lemmers, P. M., Toet, M. C., & van, B. F. (2008). Impact of patent ductus arteriosus and subsequent therapy with indomethacin on cerebral oxygenation in preterm infants. *Pediatrics, 121,* 142-147.

Limperopoulos, C., Gauvreau, K. K., O'Leary, H., Moore, M., Bassan, H., Eichenwald, E. C. et al. (2008). Cerebral hemodynamic changes during intensive care of preterm infants. *Pediatrics, 122(5),* e1006-13.

Magistretti, P. J. (1999). Brain energy metabolism. In M.Zigmond, F. Bloom, S. Landis, J. Roberts, & L. Squire (Eds.), *Fundamental in Neuroscience* (pp. 389-413). San Diego: Academic Press.

Magistretti, P. J. (2006). Neuron-glia metabolic coupling and plasticity. *J Exp Biol, 209,* 2304-2311.

Magistretti, P. J. & Pellerin, L. (1999). Cellular mechanisms of brain energy metabolism and their relevance to functional brain imaging. *Philos.Trans.R.Soc.Lond B Biol.Sci., 354,* 1155-1163.

Manworren, R. C. & Hynan, L. S. (2003). Clinical validation of FLACC: preverbal patient pain scale. *Pediatric Nursing, 29(2),* 140-6.

Marcar, V. L., Strassle, A. E., Loenneker, T., Schwarz, U., & Martin, E. (2004). The influence of cortical maturation on the BOLD response: an fMRI study of visual cortex in children. *Pediatr Res, 56,* 967-974.

Mazziotta, J. C., Toga, A. W., & Frackowiak, R. S. J. (2000). *Brain Mapping.* San Diego: Academic Press.

McCaffery, M. & Pasero, C. (1999). *Pain and Clinic Manual*. (2nd ed.) St-Louis: Mosby.

Meek, J. H., Firbank, M., Elwell, C. E., Atkinson, J., Braddick, O., & Wyatt, J. S. (1998). Regional hemodynamic responses to visual stimulation in awake infants. *Pediatric Research, 43(6),* 840-3.

Melzack, R. (1990). Phantom limbs and the concept of a neuromatrix. *Trends in Neurosciences, 13,* 88-92.

Melzack, R. (1991). The Gate Control theory 25 years later: new perspecties on phantom limb pain. In M.Bond, J. Charlton, & C. Woolf (Eds.), *Pain research and*

therapy: Proceedings of the VIth World Congress on Pain (pp. 9-26). Amsterdam: Elsevier Science Publishers.

Melzack, R. (1999). From the gate to the neuromatrix. *Pain, 82,* S121-S126.

Melzack, R. (2001). Pain and the neuromatrix in the brain. *J Dent Educ., 65,* 1378-1382.

Melzack, R. & Wall, P. D. (1965). Pain mechanisms: a new theory. *Science., 150,* 971.

Merkel, S., Voepel-Lewis, T., & Malviya, S. (2002). Pain assessment in infants and young children: the FLACC scale. *American Journal of Nursing, 102(10),* 55-8.

Merkel, S. I., Voepel-Lewis, T., Shayevitz, J. R., & Malviya, S. (1997). The FLACC: a behavioral scale for scoring postoperative pain in young children. *Pediatric Nursing, 23(3),* 293-7.

Merskey, H., & Bogduk, N. (1994). Part III: Pain Terms, A Current List with Definitions and Notes on Usage. In *Classification of Chronic Pain, Second Edition, IASP Task Force on Taxonomy* (pp. 209-214). Seatle: IASP Press. Update 2011, retrieved on December 13, 2011 from: http://www.iasp-pain.org/Content/NavigationMenu/GeneralResourceLinks/PainDefinitions/default.htm

Niesters, M., Dahan, A., Kest, B., Zacny, J., Stijnen, T., Aarts, L., and Sarton, E. (2010). Do sex differences exist in opioid analgesia? A systematic review and meta-analysis of human experimental and clinical studies. *Pain, 151,* 61-68.

Norden, J., Hannallah, R., Getson, P., O'Donnell, R., Kelliher, G., & Walker, N. (1991). Conccurrent validation of an objective pain scale for infants and children. *Anesthesiology, 75*, A934.

Nunnally, J. J. (1978). *Psychometric theory*. (2nd ed.) New York: McGraw-Hill.

Obrig, H., Hirth, C., Junge-Hulsing, J. G., Doge, C., Wenzel, R., Wolf, T. et al. (1997). Length of resting period between stimulation cycles modulates hemodynamic response to a motor stimulus. *Advances in Experimental Medicine & Biology, 411*, 471-80.

Obrig, H. & Villringer, A. (1997). Near-infrared spectroscopy in functional activation studies. Can NIRS demonstrate cortical activation?. *Advances in Experimental Medicine & Biology, 413*, 113-27.

Obrig, H., Wenzel, R., Kohl, M., Horst, S., Wobst, P., Steinbrink, J. et al. (2000). Near-infrared spectroscopy: does it function in functional activation studies of the adult brain? *Int.J.Psychophysiol., 35*, 125-142.

Oyama, K., Kondo, T., Komatsu, H., & Sugiura, T. (2009). Evaluation of NIRS data based on theoretical analysis of oxygen transport to cerebral tissue. *Advances in Experimental Medicine & Biology, 645*, 181-6.

Pollack, M. M., Patel, K. M., & Ruttimann, U. E. (1996). PRISM III: An updated Pediatric Risk of Mortality score. *Critical Care Medicine, 24*, 743-52.

Puntillo, K. A., Stannard, D., Miaskowski, C., Kehrle, K., & Gleeson, S. (2002). Use of a pain assessment and intervention notation (P.A.I.N.) tool in critical care nursing practice: Nurses' evaluations. *Heart & Lung: The Journal of Acute and Critical Care, 31,* 303-314.

Puntillo, K. A., White, C., Morris, A. B., Perdue, S. T., Stanik-Hutt, J., Thompson, C. L. et al. (2001). Patients' perceptions and responses to procedural pain: results from Thunder Project II. *American Journal of Critical Care, 10(4),* 238-51.

Ramelet, A. S., Abu-Saad, H. H., Bulsara, M. K., Rees, N., & McDonald, S. (2006). Capturing postoperative pain responses in critically ill infants aged 0 to 9 months. *Pediatric Critical Care Medicine, 7(1),* 19-26.

Ramelet, A. S., Abu-Saad, H. H., Rees, N., & McDonald, S. (2004). The challenges of pain measurement in critically ill young children: a comprehensive review. *Australian Critical Care, 17(1),* 33-45.

Ramelet, A. S., Rees, N., McDonald, S., Bulsara, M., & Abu-Saad, H. H. (2007a). Development and preliminary psychometric testing of the Multidimensional Assessment of Pain Scale: MAPS. *Paediatric Anaesthesia, 17(4), 333-40.*

Ramelet, A. S., Rees, N. W., McDonald, S., Bulsara, M. K., & Abu-Saad, H. H. (2007b). Clinical validation of the Multidimensional Assessment of Pain Scale. *Pediatric Anesthesia, 17,* 1156-1165.

Ranger, M., Johnston, C. C., & Anand, K. J. (2007). Current controversies regarding pain assessment in neonates. *Semin.Perinatol., 31,* 283-288.

Ranger, M., Johnston, C.C., Limperopoulos. C., Rennick, J.E., & du Plessis, A.J. (2011). Cerebral Near-Infrared Spectroscopy as a Measure of Nociceptive Evoked Activity in Critically Ill Infants. *Pain Research & Management, 16(5),* 331-336.

Ranger, M., Johnston, C.C., Rennick, J.E., Limperopoulos, C., Heldt, T., & du Plessis, A.J. Near-infrared spectroscopy to assess nociception following a noxiouss stimulation in critically ill infants. *European Journal of Pain* (under review).

Rennick, J. E., Johnston, C. C., Dougherty, G., Platt, R., & Ritchie, J. A. (2002). Children's psychological responses after critical illness and exposure to invasive technology. *Journal of Developmental & Behavioral Pediatrics, 23(3),* 133-44.

Rennick, J. E., Morin, I., Kim, D., Johnston, C. C., Dougherty, G., & Platt, R. (2004). Identifying children at high risk for psychological sequelae after pediatric intensive care unit hospitalization. *Pediatric Critical Care Medicine, 5(4*), 358-63.

Riley,J.L.I., Robinson,M.E., Wise,E.A., Myers,C.D., and Fillingim,R.B. (1998). Sex differences in the perception of noxious experimental stimuli: a meta-analysis. *Pain, 74,* 181-187.

Roberts, I. G., Fallon, P., Kirkham, F. J., Kirshbom, P. M., Cooper, C. E., Elliott, M. J. et al. (1998). Measurement of cerebral blood flow during cardiopulmonary bypass with near-infrared spectroscopy. *J Thorac.Cardiovasc.Surg., 115,* 94-102.

Roche-Labarbe, N., Wallois, F., Ponchel, E., Kongolo, G., & Grebe, R. (2007). Coupled oxygenation oscillation measured by NIRS and intermittent cerebral activation on EEG in premature infants. *Neuroimage, 36,* 718-727.

Sava, S., Lebel, A. A., Leslie, D. S., Drosos, A., Berde, C., Becerra, L. et al. (2009a). Challenges of functional imaging research of pain in children. *Mol.Pain, 5,* 30.

Schlaepfer, T. E., Strain, E. C., Greenberg, B. D., Preston, K. L., Lancaster, E., Bigelow, G. E. et al. (1998). Site of Opioid Action in the Human Brain: Mu and Kappa Agonists' Subjective and Cerebral Blood Flow Effects. *Am J Psychiatry, 155,* 470-473.

Slater, R., Cantarella, A., Franck, L., Meek, J., & Fitzgerald, M. (2008). How well do clinical pain assessment tools reflect pain in infants? *PLoS Medicine / Public Library of Science, 5(6),* e129.

Slater, R., Cantarella, A., Gallella, S., Worley, A., Boyd, S., Meek, J. et al. (2006). Cortical Pain Responses in Human Infants. *J.Neurosci., 26,* 3662-3666.

Slater, R., Cantarella, A., Yoxen, J., Patten, D., Potts, H., Meek, J. et al. (2009). Latency to facial expression change following noxious stimulation in infants is dependent on postmenstrual age. *Pain, 146,* 177-182.

Slater, R., Cornelissen, L., Fabrizi, L., Patten, D., Yoxen, J., Worley, A. et al. (2010a). Oral sucrose as an analgesic drug for procedural pain in newborn infants: a randomised controlled trial. *Lancet, 376,* 1225-1232.

Slater, R., Fabrizi, L., Worley, A., Meek, J., Boyd, S., & Fitzgerald, M. (2010b). Premature infants display increased noxious-evoked neuronal activity in the brain compared to healthy age-matched term-born infants. *NeuroImage, 52,* 583-589.

Slater, R., Fitzgerald, M., & Meek, J. (2007). Can cortical responses following noxious stimulation inform us about pain processing in neonates? *Semin.Perinatol., 31,* 298-302.

Slater, R., Worley, A., Fabrizi, L., Roberts, S., Meek, J., Boyd, S. et al. (2010c). Evoked potentials generated by noxious stimulation in the human infant brain. *European Journal of Pain, 14,* 321-326.

Soul, J. S. & du Plessis, A. J. (1999). Near-infrared spectroscopy. *Seminars in Pediatric Neurology, 6,* 101-110.

Soul, J. S., Hammer, P. E., Tsuji, M., Saul, J. P., Bassan, H., Limperopoulos, C. et al. (2007). Fluctuating Pressure-Passivity Is Common in the Cerebral Circulation of Sick Premature Infants. *Pediatric Research, 61,* 467-473.

Soul, J. S., Taylor, G., Wypij, D., duPlessis, AJ., & Volpe, JJ. (2000). Noninvasive Detection of Changes in Cerebral Blood Flow by Near-Infrared Spectroscopy in a Piglet Model of Hydrocephalus. *Pediatric Research, 48,* 445-449.

Steed, D., Port, L., Connell, T. G., Standish, J., Munro, J., Takacs, L. et al. (2011). Oral sucrose for procedural pain in infants. *Lancet, 377,* 25-28.

Stevens, B., Franck, L., Gibbins, S., McGrath, P. J., Dupuis, A., Yamada, J. et al. (2007). Determining the structure of acute pain responses in vulnerable neonates. *Can.J Nurs Res, 39,* 32-47.

Stevens, B. J., Johnston, C. C., & Horton, L. (1993). Multidimensional Pain Assessment in Premature Neonates: A Pilot Study. *Journal of Obstetric, Gynecologic, & Neonatal Nursing, 22,* 531-541.

Stevens, B., Johnston, C., Petryshen, P., & Taddio, A. (1996). Premature Infant Pain Profile: development and initial validation. *Clinical Journal of Pain, 12(1),* 13-22.

Stevens, B., Johnston, C., Taddio, A., Gibbins, S., & Yamada, J. (2010). The premature infant pain profile: evaluation 13 years after development. *The Clinical journal of pain, 26,* 813-30

Stevens, B., McGrath, P., Gibbins, S., Beyene, J., Breau, L., Camfield, C. et al. (2003). Procedural pain in newborns at risk for neurologic impairment. *Pain, 105,* 27-35.

Stevens, B., McGrath, P., Gibbins, S., Beyene, J., Breau, L., Camfield, C. et al. (2007). Determining behavioural and physiological responses to pain in infants at risk for neurological impairment. *Pain, 127,* 94-102.

Stevens, B., Yamada, J., & Ohlsson, A. (2010). Sucrose for analgesia in newborn infants undergoing painful procedures. *Cochrane Database of Systematic Reviews.(1):CD001069, [Update of Cochrane Database Syst Rev. 2004;(3):CD001069; PMID: 15266438]*.

Streiner, D. (1993). A checklist for evaluating the usefulness of rating scales. *Canadian Journal of Psychiatry, 38,* 140-147.

Streiner, D. L. & Norman, G. R. (2008). *Health measurement scales: A practical guide to their development and use*. (4th ed.) New York: Oxford University Press.

Sweet, S. D. & McGrath, P. J. (1998). Physiological Measures of Pain. In G.A.Finley & P. J. McGrath (Eds.), *Measurement of Pain in Infants and Children* (pp. 59-81). Seattle: IASP Press.

Tracey, I. & Mantyh, P. W. (2007). The Cerebral Signature for Pain Perception and Its Modulation. *Neuron, 55,* 377-391.

Treede, R. D., Kenshalo, D. R., Gracely, R. H., & Jones, A. K. P. (1999). The cortical representation of pain. *Pain, 79,* 105-111.

Tsuji, M., duPlessis, A., Taylor, G., Crocker, R., & Volpe, J. J. (1998). Near infrared spectroscopy detects cerebral ischemia during hypotension in piglets. *Pediatric Research, 44(4),* 591-5.

Tsuji, M., Saul, J. P., du, P. A., Eichenwald, E., Sobh, J., Crocker, R. et al. (2000). Cerebral intravascular oxygenation correlates with mean arterial pressure in critically ill premature infants. *Pediatrics, 106(4),* 625-32.

Turk, D. & Melzack, R. (2001). *Handbook of Pain Assessment*. (2nd ed.) New York: Guilford Press.

van Alfen-van der Velden AA, Claessen, V. P., Hopman, J. C., Klaessens, J. H., Sengers, R. C., & Liem, K. D. (2009). Changes in cerebral oxygenation and hemodynamics during cranial ultrasound in preterm infants. *Brain Dev., 31,* 427-434.

van Dijk, M., de Boer, J. B., Koot, H. M., Duivenvoorden, H. J., Passchier, J., Bouwmeester, N. et al. (2001). The association between physiological and behavioral pain measures in 0- to 3-year-old infants after major surgery. *Journal of Pain & Symptom Management, 22(1),* 600-9.

van Dijk, M., de Boer, J. B., Koot, H. M., Tibboel, D., Passchier, J., & Duivenvoorden, H. J. (2000). The reliability and validity of the COMFORT scale as a postoperative pain instrument in 0 to 3-year-old infants. *Pain, 84(2-3),* 367-77.

van Dijk, M., Peters, J. W., van Deventer, P., & Tibboel, D. (2005). The COMFORT Behavior Scale: a tool for assessing pain and sedation in infants. *American Journal of Nursing, 105(1),* 33-6.

Voepel-Lewis, T., Merkel, S., Tait, A. R., Trzcinka, A., & Malviya, S. (2002). The reliability and validity of the Face, Legs, Activity, Cry, Consolability observational tool as a measure of pain in children with cognitive impairment. *Anesthesia & Analgesia, 95(5),* 1224-9.

Voepel-Lewis, T., Zanotti, J., Dammeyer, J. A., & Merkel, S. (2010). Reliability and Validity of the Face, Legs, Activity, Cry, Consolability Behavioral Tool in Assessing Acute Pain in Critically Ill Patients. *American Journal of Critical Care, 19,* 55-61.

Willis, M. H., Merkel, S. I., Voepel-Lewis, T., & Malviya, S. (2003). FLACC Behavioral Pain Assessment Scale: a comparison with the child's self-report. *Pediatric Nursing, 29(3),* 195-8.

Wolf, M. & Greisen, G. (2009). Advances in near-infrared spectroscopy to study the brain of the preterm and term neonate. *Clin.Perinatol., 36,* 807-34.

Wolfberg, A. J. & du Plessis, A. J. (2006). Near-Infrared Spectroscopy in the Fetus and Neonate. *Clinics in Perinatology, 33,* 707-728.

Wong, D. L. & Baker, C. M. (1988). Pain in children: comparison of assessment scales. *Pediatr.Nurs., 14,* 9-17.

Wyatt, J. S., Delpy, D. T., Cope, M., Wray, S., & Reynolds, E. O. R. (1986). Quantification of Cerebral Oxygenation and Haemodynamics in Sick Newborn Infants bt Near Infrared Spectrophotometry. *The Lancet, 328,* 1063-1066.

Yamamoto, T. & Kato, T. (2002). Paradoxical correlation between signal in functional magnetic rasonance imaging and deoxygenated hemoglobin content in capillaries: a new theoretical explanation. *Physics in Medicine and Biology, 47,* 1121-1141.

Yanowitz, T. D., Potter, D. M., Bowen, A., Baker, R. W., & Roberts, J. M. (2006). Variability in cerebral oxygen delivery is reduced in premature neonates exposed to chorioamnionitis. *Pediatr.Res., 59,* 299-304.

APPENDIX A. FLACC BEHAVIORAL PAIN SCALE

The FLACC Behavioural Pain Assessment Scale			
Categories	**0**	**1**	**2**
Face	No particular expression or smile	Occasional grimace or frown, withdrawn, disinterested	Frequent to constant quivering chin, clenched jaw
Legs	Normal position or relaxed	Uneasy, restless, tense	Kicking, or legs drawn up
Activity	Lying quietly, normal position, moves easily	Squirming, Shifting back & forth, Tense	Arched, rigid or jerking
Cry	No cry (awake or asleep)	Moans or whimpers Occasional complaint	Crying steadily, screams or sobs frequent complaints
Consolability	Content, relaxed	Reassured by touching, hugging, or being talked to, distractible	Difficult to console or comfort

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Merkel, S., Voepel-Lewis,T., Sayevitz, J., & Malviya, S. The FlACC: A behavioral scale for scoring post-op pain in young children. *Pediatric Nursing*, 1997, 23, 293-297.

APPENDIX B.

Pediatric RISk of Mortality Score (PRISM) III (Pollack *et al.*, 1996)

Patient's Initials: |_|_|_| Patient Enrolment Number: |_|_|_|_|_|_|

ADRENAL INSUFFICIENCY IN PEDIATRIC CRITICAL ILLNESS

Page 5: DAILY PRISM III SCORE

2.a. Daily PRISM III - Score the PRISM III for Day 1 ACTH Stimulation Test – Use the worst value within the same calendar day. Circle the appropriate units.

	Actual Value	1	2	3	4	5	6	7	11	Po
Heart Rate beats/min) Neonate* Infant* Child* Adolescent*				215 - 225 215 - 225 185 - 205 145 - 155	> 225 > 225 > 205 > 155					
Systolic BP (mmHg) Neonate* Infant* Child* Adolescent*				40 - 55 45 - 65 55 - 75 65 - 85				< 40 < 55 < 55 < 65		
Temperature (Rectal/PO/Blood/Ax)				< 33°C or > 40.0°C						
Pupillary Reflexes								one fixed, one reactive	both fixed	
Mental Status						stupor/coma (GCS < 8)				
Acidosis			pH 7.0 - 7.28 or total CO_2 5 - 16.9				pH < 7.0 or total CO_2 < 5			
Total CO_2 (mmol/L)					> 34.0					
PH			7.48 - 7.55	> 7.55						
PaO_2(mmHg)				42.0 - 49.9			< 42.0			
$PaCO_2$ (mmHg)		50 - 75		> 75						
Glucose			> 200mg/dL or > 11.0 mmol/L							
Potassium (mmol/L)				> 6.9						
Creatinine Neonate* Infant* Child* Adolescent*			> 0.85 mg/dL or > 75 µmol/L > 0.90 mg/dL or > 80 µmol/L > 0.90 mg/dL or > 80 µmol/L > 1.30 mg/dL or > 115 µmol/L							
BUN Neonate* All other ages				>11.9 mg/dL or > 4.3 mmol/L >14.9 mg/dL or > 5.4 mmol/L						
WBC					< 3000 (cells/mm^3) or < 3.0 (10^9/L)					
PT or PTT Neonate* All other ages				PT > 22.0 or PTT > 85.0 PT > 22.0 or PTT > 57.0						
Platelet Count			100,000 - 200,000 (cells/mm^3) or 100 - 200 (10^9/L)		50,000 - 99,999 (cells/mm^3) or 50 - 90 (10^9/L)	<50,000 cells/mm^3 or < 50 (10^9/L)				
									Total score:	

* Neonate = 0 < 1 month, Infant = ≥ 1 month - 12 months, Child = ≥ 12 months - 144 months (1 - 11 years), Adolescent = ≥ 144 months (≥ 12 years).

APPENDIX C. RESEARCH ETHIC BOARD APPROVAL CHILDREN'S HOSPITAL BOSTON

Children's Hospital Boston

Clinical Investigation Office
333 Longwood Avenue, 4th floor
phone 617-355-7052 | *fax* 617-730-0226

To: Adre Du Plessis, MB, ChB

Date: 03/31/2010

Re: **NOTICE OF EXPEDITED CONTINUING APPROVAL**
APPROVAL DATE: **03/30/2010**
EXPIRATION DATE: **03/29/2011**

PROTOCOL NUMBER 07-04-0151

PROTOCOL TITLE DEVELOPMENT OF A QUANTITATIVE MONITORING TECHNIQUE OF CEREBRAL HEMODYNAMICS AND OXYGEN METABOLISM IN INFANTS WITH CONGENITAL HEART DISEASE

The Committee on Clinical Investigation has approved the continuing renewal above referenced protocol through expedited review procedures.

The approved consent form is available on-line through the CHB Informed Consent Library. To obtain the consent form, please go to http://chbcfapps/research/consents. The ICLibrary should be accessed each time you need a consent form to ensure that the current version of the consent is always used. Do not store the consent forms on your computer or make copies for future use. Note that the activation/expiration date on the consent form can only be changed or modified by the staff of the Clinical Investigation Office. Please also note that subjects cannot be enrolled in a study if the consent form has expired. A copy of the consent should be kept in your files. The subject/family should also be given a signed copy.

The occurrence of unanticipated problems should promptly be reported to this office. Any revisions, amendments, or changes to the protocol require prior Committee approval.

The Committee has asked this office to notify investigators that clinical investigation protocol files are subject to audits at some future time.

Sincerely,

Ashley Pysczynski
For the Committee on Clinical Investigation
cc. Mustafa Suleymanci

APPENDIX D. CONSENT FORM CHILDREN'S HOSPITAL BOSTON

Children's Hospital Boston

RESEARCH CONSENT FORM

Use Plate or Print:

MRN#:

DOB:

Pt Name:

Gender:

Protocol Title: Development of a technique for continuous measurement of cerebral venous oxygen saturation and cerebral oxygen extraction

Why is this research study being conducted; What is its purpose?

The goal of this research study is to develop a technique that will help us to determine whether babies are getting sufficient oxygen to their brain during periods of illness. There are currently no non-invasive techniques for measuring the oxygen used by the brain of sick infants. As you know, your baby was born with a heart problem and either currently requires help from a ventilator to breathe or is expected to require help from a ventilator for a limited time after a planned operation.

Who is conducting this research study, and where is it being conducted?

This is a single site study, being conducted at Children's Hospital, Boston. The research is being sponsored by the Lifebridge Foundation and the Principal Investigator is Adre du Plessis, MD, MPH. Dr. du Plessis is a senior attending neurologist at Children's Hospital. We hope to enroll approximately 100 babies in this study.

How are individuals selected for this research study? How many will participate?

We will study infants less than one year of age, born with a heart problem, who need the help of a bedside ventilator to breathe. We will try to study infants before and after heart surgery during times when they are using a ventilator. We have come to speak to you about this study today because we have learned from the cardiac surgery patient lists that your baby either will have or has had a heart operation and that your baby currently does or will require help with breathing from a ventilator. We have received permission from your baby's cardiologist to speak with you about our study. We would like to ask you to enroll your baby, _____, in this study.

What do I have to do if I am in this research study?

We plan to measure the flow of blood and oxygen to your baby's brain using a technique called near infrared spectroscopy (NIRS). NIRS allows us to measure blood flow and oxygen changes

in the brain by shining a light across the skin and skull into the brain and then measuring the amount of light that returns. In this study we are particularly interested in measuring the flow of blood and oxygen to your baby's brain during changes in blood pressure and ventilator pressure that occur during the routine care of babies with heart problems.
Secondly, we will use a technique that measures brain waves called amplitude integrated EEG. This technique is a simplified version of a technique that is widely used by doctors interested in following a patient's brain function. Amplitude integrated EEG uses two electrodes that are placed on your baby's scalp to tell us what the level of brain activity is at any time. This technique measures electrical activity coming from your baby's brain. No electrical activity goes toward your baby's brain. These measurements will be used to test whether there are changes in blood and oxygen delivery to your infant's brain as the brain activity changes.

Thirdly, we will collect a drop of discarded blood left in the syringe tubing after routine blood tests at three time points: (i) just prior to the start of cardiopulmonary bypass, (ii) at the end of cardiopulmonary bypass, and (iii) around the time of the postoperative bedside studies described above. We will be using these specimens to look for signs of inflammation, in order to see whether this plays a role in the delivery of oxygen to the infant's brain following cardiac surgery. There will be no extra blood collected for our study. The samples will be sent to a research laboratory at Brigham & Women's Hospital and will have no information identifying your baby. The samples will be used only for this research study and may be stored for up to five years.

If you agree to enroll your infant into this study, the following events will occur. During times when your infant is receiving mechanical ventilation, we will place two optodes (NIRS), or small discs, one on either side of your baby's head. These optodes will be held in place by a stretchy dressing. We will also place two tiny discs that measure brain wave activity (EEG), one on each side of your baby's head. These discs will be held in place with a sticky gel. These two procedures are for research purposes only.

These are the only three aspects of the study that will not be part of your baby's routine care. All our other measurements are made from sources that are routinely used by doctors in the management of infants like yours. We will collect measurements from the back of the bedside monitors used in the care of infants with heart disease. Our measurements will not cause any interruption of your baby's monitoring. Specifically, we will measure changes in the ventilator pressure, changes in oxygen levels measured in the hands or feet, changes in blood pressure, and changes in blood gases. If your baby's doctors are measuring the pressures in the lungs through a tube passed through your baby's nose or mouth and into her/his throat, we will also collect this information. All of these measurements will be made for up to six hours at a time, and will be repeated at intervals during the period in which your baby requires management at the intensive care unit. We will also collect certain items of information from your baby's medical record, such as her/his birth history, cardiac diagnosis, and laboratory results. This study requires no additional needle sticks, extra blood drawn, or medications that would not be used for the routine care of your baby in the intensive care unit. Once the breathing tube is removed, the research study will end for your baby. No long-term follow-up visits are required.

What are the risks of this research study? What could go wrong?

There are no known risks associated with either the near infrared spectroscopy or amplitude integrated EEG techniques. We have used the NIRS technique in more than 500 babies without any complications. We will begin our measurements only after we have checked with your baby's doctors and established that your baby is stable enough to be studied. We will carefully watch the skin on your baby's scalp and change the position of the optodes regularly to minimize skin irritation. None of these studies will interfere with your infant's routine care. Although it is possible that your baby may experience some discomfort during the placement of our optodes, this is unlikely since we have extensive experience in minimizing the discomforts to babies in our studies. In addition, infants on ventilators are often given medications to keep them comfortable, making it even less likely that they will experience any significant discomfort from the study procedures.

What are the benefits of this research study?

There is no direct benefit to your baby from participation in this study. The importance of these studies is the potential benefit for future infants in the intensive care unit. The ability to monitor the control of brain blood flow and brain oxygen use in sick infants on a continuous basis would be a major advance in the care of infants with heart disease and those who require heart surgery, as well as other sick babies receiving intensive care.

Are there costs associated with this research study? Will I receive any payments?

There will be no extra costs to you or to your insurance carrier for any studies done solely for the purposes of this research. You or your insurance carrier will be billed for all routine aspects of your baby's care during this period. You will not receive any payments for your baby's participation in this study.

In the event of an injury resulting directly from your participation in this research study, medical treatment will be provided if the injury is reported in a timely manner. Provision of such medical care does not imply any negligence or other wrongdoing on the part of the Hospital or any of the physicians or other personnel involved in the study. Where applicable, the Hospital reserves the right to seek payment from third-party payors for any medical care or services rendered. The Hospital has no program to provide you with any additional compensation as a result of any such injuries.

What will happen with the information obtained as part of this research study? What about

confidentiality?

If you agree to enroll your baby into this research study, we will assign an anonymous code to your baby and will store any identifying information in locked and password protected files. Your baby will not be identifiable to anyone outside this research study. The information gathered from your baby may be reviewed and analyzed by collaborators in this research from

the Department of Electrical Engineering and Computer Science at the Massachusetts Institute of Technology (M.I.T.). If this occurs, the information will be anonymous.

Since the information gathered in this study will not be used for clinical purposes, none of this information will be placed on your baby's medical record. In this manner it will be unlikely that others within the hospital, an insurance company or employer would ever learn of such results. Information collected during the study will be stored in separate research files maintained by the investigator. These research records will not be made available to any individuals who are not part of the research team unless you so request or as required by law. If you/your child withdraw from the research study, information that has already been collected will become part of the research data, however, you/your child will not be identified.

If I do not want to take part in this research study, what are the other choices?

Currently there are no ways for measuring, at the bedside of sick infants, the control of blood flow to the brain or the amount of oxygen used by the brain. Your alternative is to not enroll your infant in this study.

What are my rights as a research participant?

Your participation in this research study is voluntary. Regardless of your decision to enroll your infant or not, your baby's doctors will continue to manage her/him in the best available manner. A decision to not participate in this study, or to withdraw from the study, will not interfere with your baby's current or future care at Children's Hospital.

Why would I be taken off the study early?

Your baby's participation in this study may be terminated if the doctors caring for your baby determine that your baby's clinical condition does not allow for the safe performance of the study. On occasion, if another baby is being studied elsewhere, then your baby's studies may not occur if the equipment is not available.

What information do I need to know about the Health Insurance Portability and Accountability Act (HIPAA)?

During this research, information about your or your child's health will be collected. In general, under federal law, information about patients is private, but there are exceptions and you should know who will have access to this information and might see it. Researchers may be collecting information about you or your child from medical records. They may also learn things from procedures that are part of the research itself such as tests, office visits, questionnaires and interviews.

The following people will be able to see this information:

- Medical and research staff at Children's Hospital, including people listed on your informed consent.

- Medical staff who are directly involved in your care that is related to the research or arise from it.
- People who oversee, advise or conduct research at Children's Hospital, and people who oversee or evaluate research and care, including the Committee on Clinical Investigation, staff working on quality improvement, and other clinicians and administrative staff of Children's Hospital..
- People from agencies and organizations that provide independent accreditation and oversight of research
- Sponsors or others involved in funding the research
- Federal agencies that oversee or review research information.
- Government agencies and sponsors.
- If some law or court requires us to share the information, we would have to follow that law or final ruling

You/your child should be aware that the federal privacy rule does not cover all of these possible uses. This means that once some of the above mentioned users receive your/your child's health information they do not have to follow the same rules. Other laws may or may not protect sharing of private health information. If you have a question about this you may contact the Children's Hospital Privacy Officer at 617-355-5502

There is no set time for destroying this information and no time limit for its use. Researchers continue to analyze data for many years and it is not possible to know when they will be done.

You or your child do not have to sign this form. If the form is not signed, however, you won't be able to participate in the study. Not signing will not affect your care or your child's care at Children's Hospital in any way now or in the future. Also, there will be no penalty or loss of benefits if you choose not to sign and participate.

You or your child also have the right to withdraw from this study at any time. You have the right to end your permission for Children's Hospital to use or share the protected information about you or your child that was collected as part of the research.

Researchers may also continue to use information already collected to protect the integrity of the study. This means that your withdrawal won't make the whole study useless. Once you remove your permission and you or your child is no longer in the study, no more private health information will be collected. If you wish to withdraw you will need to do so in writing. Your investigator will have a form for you to use. If you or your child decide to share private information with anyone not involved in the study, the federal law designed to protect privacy may no longer apply to this information.

Although there are some legal limitations, you or your child have the right to get protected information resulting from this research that relates to your treatment or to payments. This information is available after the study analysis is done. To request the information, please contact the Hospital's Privacy Officer at 617-355-5502. If you have questions, please be sure to ask for answers.

Research at Children's Hospital: Children's Hospital has recently developed a web-based, interactive educational program for parents called "A Parent's Guide to Medical Research." To find out more about research at Children's Hospital, please visit the program at www.researchchildren.org

Children's Hospital is interested in hearing your comments, answering your questions and responding to any concerns regarding clinical research at Children's Hospital. If you would like further information about the type of clinical research performed at the hospital or have suggestions, questions or concerns regarding clinical research you may send an email to cci@childrens.harvard.edu or call 617 355-7052 between the hours of 8:30 and 5:00.

INVESTIGATOR'S AND/OR ASSOCIATE'S STATEMENT:

I have fully explained to all involved parties (participant/parent/guardian as applicable) the nature and purpose of the above-described procedures and the risks involved in its performance. I have provided the subject/family with the Privacy Rule if requested. I have answered and will answer all questions to the best of my ability. I will inform the participant of any changes in the procedures or the risks and benefits if any should occur during or after the course of the study. I have given a copy of the consent/ authorization form to the subject/family.

☒ _____

Date (MM/DD/YEAR) Signature of **Investigator or Associate**

CONSENT/AUTHORIZATION:

I understand that I may use the following contact information to reach the appropriate person/office to address any questions or concerns I may have about this study. I know:

I can call … about …	At …	If I have questions or concerns
Investigator: Adré J. du Plessis MD emergencies complaints	Phone: **617-355-8025** Pager: **617-355-6000 #0877**	▪ General questions about the study ▪ Research-related injuries or ▪ Any research-related concerns or
Office of Clinical Investigations	Phone: **617-355-7052**	▪ Rights of a research subject ▪ Use of protected health information ▪ Compensation in event of research-related injury ▪ Any research-related concerns or complaints ▪ If investigator/study contact cannot be reached ▪ If I want to speak with someone other than the Investigator, Study Contact or research staff

I have been satisfactorily informed of the above-described procedure with its possible risks and benefits. I have been provided with the applicable Privacy Rule provisions under the Health Insurance Portability and Accountability Act. I give permission for my child's participation in this study and for use of the associated protected health information as described above.

I understand that participation in this study is voluntary. If I refuse to permit my child to participate, or choose to withdraw my child from the study at any time, I understand there will be no penalty or loss of benefits to which my child is otherwise entitled, and this decision will not affect present or future care by the doctors or the hospital. I am signing this consent form before my child has participated in any research activities. I have been given a copy of this form.

_____ _____ _____

Date (MM/DD/YEAR) Signature of **Parent or Guardian** Relationship to child

WITNESS SIGNATURE REQUIRED BELOW ONLY IF: (check which one applies)

- ☐ the consent document needs to be read to subject or legal representative **or**
- ☐ communication impairments limit the subject's ability to clearly express consent **or**
- ☐ required by sponsor/CCI.
- ☐ other reason: please specify _____

I confirm that the information in this consent form was accurately explained to, and understood by the subject or legally authorized representative, and that informed consent was given freely.

☒ _____ _____

Date (MM/DD/YEAR) Signature of **Witness**

APPENDIX E. RESEARCH ETHIC BOARD MONTREAL CHILDREN'S HOSPITAL

L'Hôpital de Montréal pour enfants
The Montreal Children's Hospital
Centre universitaire de santé McGill
McGill University Health Centre

August 21, 2009

Dr. C. Johnston
Manon Ranger
McGill School of Nursing
3605 rue University
Montreal, QC
H3A 2A7

Re : ***PED-08-037 Near Infrared Spectrscopy (NIRS) to Measure Nocieption Following Noxious Stimulation in Critically Ill Infants***

Dear Dr. Johnston,

We have received an Application for Continuing Review of the Montreal Children's Hospital Research Ethics Board for the research study referenced above and the report was found to be acceptable for ongoing conduct at the McGill University Health Centre. At the MUHC, sponsored research activities that require US federal assurance are conducted under Federal Wide Assurance (FWA) 00000840.

The re-approval for the study protocol was provided via expedited review of the Chair on August 20, 2009 will be reported to the Research Ethics Board (REB) at its meeting of August 24, 2009, and will be entered accordingly into the minutes. No subjects are being recruited at this site.

All research involving human subjects requires review at a recurring interval and the current study approval is in effect until August 19, 2010. **It is the responsibility of the principal investigator to submit an Application for Continuing Review to the REB prior to the expiration of approval to comply with the regulation for continuing review of "at least once per year".**

However, should the research conclude for any reason prior to the next required review, you are required to submit a Termination Report to the Committee once the data analysis is complete to give an account of the study findings and publication status.

Should any revision to the study, or other unanticipated development occur prior to the next required review, you must advise the REB without delay. Regulation does not permit initiation of a proposed study modification prior to REB approval for the amendment.

Sincerely,

Elizabeth Craven
Coordinator, Research Ethics Board

APPENDIX F. CONSENT FORMS ENGLISH/FRENCH MONTREAL CHILDREN'S HOSPITAL

MONTREAL CHILDREN'S HOSPITAL
PEDIATRIC CRITICAL CARE DEPARTMENT

INFORMED CONSENT

BRAIN ACTIVITY MEASUREMENT RELATED TO PAINFUL PROCEDURES IN THE CRITICALLY ILL INFANT

INVESTIGATORS

- Manon Ranger, RN, MSc — PhD candidate, McGill University, School of Nursing
- Dr Céleste Johnston, RN, Ed — Professor, McGill University, School of Nursing
- Dr Janet Rennick, RN, PhD — Assistant professor, McGill University, School of Nursing
- Dr Catherine Limperopoulos, OT, PhD — Assistant Professor, McGill University Neurology & Neurosurgery
- Dr Davinia Withington, MD — Assistant Professor, Anesthesiology & Critical Care

PURPOSE OF THE STUDY

An admission to the hospital can be quite stressful for children. Pain contributes significantly to this distressing experience, especially if surgical events are to take place. During the post-operative course of your child, certain painful procedures may occur. To provide good pain relief a good pain assessment is essential. This may be quite difficult when children are too young to communicate their pain with words. To know if children have pain, we usually rely on certain behaviors like facial expressions and crying but these may not always be specific to pain. Recent studies with babies showed that painful events cause activity in specific regions of the brain. This activity was measured with a non invasive technology called Near Infrared Spectroscopy (NIRS). These results are quite promising as they could help us the observers, to link brain activity with behaviors used to indicate pain. It may also demonstrate that even if some infants do not demonstrate such vigorous pain behaviors, because they are so sick, they are in fact experiencing pain as measured by brain activity. The purpose of this study is to measure brain activity during chest tube removal, a common procedure that takes place after cardiac surgery.

STUDY PROCEDURE

During the post-operative course of your child, many procedures will take place. Most children after cardiac surgery have a chest tube to collect fluids around the lungs. When the time is right, this tube must be removed. This is done by the cardiovascular surgery team (CVT) and they decide when it is time to remove the chest tube of your child. If you decide to take part in the study, 30 minutes prior to the procedure, the principal investigator will begin to set-up the equipment to gather some data. To measure brain activity during the removal of your child's chest tube, two sets of non adhesive electrodes will be placed on each side of your child's head and kept in place with a cotton hat. These electrodes will be connected to the NIRS machine.

This machine resembles much to a portable vital signs monitor. To assess the behaviors during the procedure, your child will be videotaped using a small digital camera. The medical team will not be visible on the videotape and this footage will serve only for this study. The vital signs, such as heart beat, respiratory rate, blood pressure and blood oxygen level, will be collected from PICU standard monitoring system at the same time as the NIRS monitor. Recordings will take place during the procedure and once it is finished, the equipment will be taken off. This will mark the end of the study. Our purpose is just to gather some numbers and video to make links afterwards, we do not intervene whatsoever with the normal course of the procedure. Information regarding age, weight, health status, and medications administered, which can be found in your child's medical chart will also be collected. These data will be used in some analyses of the study.
Taking part in this study is completely voluntary. You may choose not to be in this study. If at any time you decide not to continue participating in our study you can contact us. This will not affect the care of your child.

RISKS AND INCONVENIENCES
The participation of your child in this study does not entail any risks or inconveniences except that your child might be slightly bothered by the electrodes connected to the NIRS equipment. The light source emitted by the machine is totally safe and painless. It will not cause any tissue damage where it is positioned.

BENEFITS
You should not expect any direct benefits from your child's participation in this study. However, the information from this study may benefit future patients by contributing to improve pain assessment in critically ill children unable to speak.

CONFIDENTIALITY
The information gathered will be treated as confidential as required and permitted by law and will be kept in a locked cabinet at McGill University for 25 years. No information that can identify your child will be used in any reports, publications or presentations, which may result from this study. Films will only be used for the purpose of this study and will be viewed only by members of the research team. The films will not be used for publications, presentations, or teaching.

CONTACT PERSON
If you wish to discuss the information or have questions at any time, please feel free to contact any one of the investigators:

Manon Ranger, PhD candidate	(514) 299-0187 (mobile phone)
Dr Davinia Withington	(514) 412-4400 ext 22367

For additional information regarding your child's rights as a research subject, you may contact the hospitals' Patient Representative, (514) 412-4400 ext. 22223, who is independent of the investigator, and works to protect patients' right.

CONSENT

The study described above has been explained to me, and I voluntarily consent for my child to take part in this study. I have had the opportunity to ask questions and understand that the investigators listed above will answer future questions, should I have any, about the research.

Participant's name: Date

Parent or Guardian's printed name:

Parent or Guardian's signature: Date

Relationship to child:

Name of the person who obtained consent:

Signature of the person who obtained consent: Date

HÔPITAL DE MONTRÉAL POUR ENFANTS
DÉPARTEMENT DES SOINS CRITIQUES

FORMULAIRE DE CONSENTEMENT

MESURE DE L'ACTIVITÉ CÉRÉBRALE PENDANT UNE PROCÉDURE DOULOUREUSE CHEZ L'ENFANT EN SOINS CRITIQUES

INVESTIGATEURS:

• Manon Ranger, Inf., MSc	PhD candidate, Université McGill, École des Sciences Infirmières
• Dr Céleste Johnston, Inf., Ed	Professeur, Université McGill, École des Sciences Infirmières
• Dr Janet Rennick, Inf., PhD	Professeur adjoint, Université McGill, École des Sciences Infirmières
• Dr Catherine Limperopoulos, Erg., PhD	Professeur adjoint, Université McGill, Départements de Neurologie & Neurochirurgie
• Dr Davinia Withington, MD	Professeur adjoint, Anesthésiologie & Soins critiques

BUT DE L'ÉTUDE

Une admission à l'hôpital peut être stressante pour un enfant. La douleur contribue à cette expérience stressante, surtout si une chirurgie a lieu. Après la chirurgie de votre enfant, certaines procédures douloureuses peuvent avoir lieu. Pour bien soulager la douleur une bonne évaluation est essentielle. Ceci peut être assez difficile si les enfants sont trop jeunes pour communiquer avec des mots leur douleur. Pour savoir si les bébés ont mal, nous devons observer les comportements comme des expressions du visage ou les pleurs, mais ceux-ci ne sont pas toujours liés à la douleur. Des études récentes chez les bébés ont démontré que la douleur était associée à de l'activité dans certaines régions du cerveau. Une technologie noninvasive appelée, Spectroscopie par infrarouge (SPIR), peut détecter cette activité dans le cerveau. Ces résultats sont très prometteurs puisqu'ils pourront peut-être nous aider à faire le lien entre ce qui se passe dans le cerveau et les comportements que nous observons lorsque les enfants ont mal. Ceux-ci vont peut-être aussi indiquer que même si nous n'observons pas de comportement, parce que l'enfant est trop malade par exemple, la douleur est présente telle que mesurée dans le cerveau. Le but de cette étude est de mesurer l'activité dans le cerveau pendant que le drain thoracique est retiré, une procédure commune après une chirurgie cardiaque.

LE DÉROULEMENT DE L'ÉTUDE

Pendant la période post-opératoire de votre enfant, certaines procédures auront lieu. Après une chirurgie cardiaque, la plupart des enfants ont un drain thoracique pour accumuler le liquide autour des poumons. Quand le temps sera venu, ce drain devra être retiré. Ceci est fait par les médecins de chirurgie cardiaque et ce sont eux qui décident quand le temps est bon pour votre enfant. Si vous décidez de participer à notre recherche,

30 minutes avant que le drain thoracique sera retiré, l'investigateur principal de l'étude commencera à installer l'équipement afin d'obtenir des données. Pour mesurer l'activité dans le cerveau de votre enfant pendant que son drain thoracique sera retiré, deux électrodes non collantes seront déposées de chaque côté de sa tête et tenues en place par une petite tuque de cotton. Ces électrodes seront rattachées à la machine de SPIR. Cette machine ressemble beaucoup à la machine qui enregistre les signes vitaux. Pour évaluer les comportements de votre enfant pendant la procédure, celui-ci sera filmé avec une petite camera digitale. L'équipe médicale ne sera pas visible sur les vidéos et ceux-ci ne seront utilisés que pour l'étude. Les signes vitaux, comme les battements de coeur, le rythme de la respiration, la pression artérielle et le niveau d'oxygène, seront recueillis par les moniteurs des soins intensifs en même temps que le SPIR. L'enregistrement aura lieu pour la durée de la procédure et une fois qu'elle sera terminée, l'équipement sera retiré. Ceci marquera la fin de l'étude. Le but de l'étude est d'obtenir des chiffres et des images afin de faire des liens par la suite, nous n'intervenons pas sur la façon dont la procédure est habituellement faite.
Des informations relatives à l'âge, le poids, l'état de santé, et la médications administrées, pouvant être trouvées dans le dossier de votre enfant, seront recueillies. Ces données seront utilisées pour certaines analyses reliées à cette recherche.
Prendre part à cette étude est complètement volontaire. Vous pouvez choisir de ne pas participer à cette recherché. Si, à tout moment, vous ne voulez plus participer à cette étude, vous pouvez nous contacter. Ceci n'affectera aucunement les soins que votre enfant reçoit.

RISQUES ET INCONVENIENTS
La participation de votre enfant à cette étude n'engendre aucun risque ou inconvénient à l'exception que celui-ci puisse être un peu dérangé par l'équipement du SPIR. La source lumineuse émise par la machine est totalement sécuritaire et ne cause aucune douleur ou dommage à la peau.

BÉNÉFICES
Il n'y a pas de bénéfices directes relies à la participation de votre enfant à cette étude. Par contre, les informations obtenues par cette étude pourront peut-être aider les futurs patients en améliorant l'évaluation de la douleur chez les enfants qui sont incapables de parler.

CONFIDENTIALITÉ
L'information recueillie sera traitée confidentielle tel que requis et permis par la loi. Celle-ci sera gardée dans une filière barrée à l'université McGill pour une durée de 25 ans. Aucune information qui résultera de cette étude pouvant identifier votre enfant ne sera utilisée pour des fins de reportages, publications ou présentations. Les vidéo ne seront utilisés que pour cette recherche et visionnés que pas les membres de l'équipe de recherche. Les images ne seront pas utilisées pour des publications, des présentations ou de l'enseignement.

PERSONNES RESSOURCES

À tout moment, si vous désirez discuter des informations ou si vous avez des questions, s'il vous plait, contacter les investigateurs mentionnés ici-bas:

Manon Ranger, PhD candidate	(514) 299-0187 (cellulaire)
Dr Davinia Withington	(514) 412-4400 ext 22367

Pour obtenir plus d'informations concernant les droits de votre enfant en tant que participant à une étude, vous pouvez contacter le Représentant de patients de l'hôpital, (514) 412-4400 ext. 22223. Cette personne est indépendante de l'investigateur et travaille afin de protéger les droits des patients.

Signature investigateur Date

DÉCLARATION DU PARTICIPANT

L'étude décrite ci-dessus m'a été expliquée et je consens de façon volontaire à ce que mon enfant participe à cette recherché. J'ai eu l'opportunité de poser mes questions et de comprendre que les investigateurs seront disponibles pour répondre à mes questions portant sur cette étude dans le futur, si cela survient.

Signature du participant Date

Signature du parent/gardien légal Date

Lien avec l'enfant:

Nom de la personne ayant obtenu le consentement:

Signature de la personne ayant obtenu le consentement: Date

APPENDIX G. MANUSCRIPT #2 CONFIRMATION OF JOURNAL SECOND RE-SUBMISSION

May 8, 2011
A manuscript number has been assigned

ees.eurjpain.0.110019.eccdf1dfc@eesmail.elsevier.com on behalf of the European Journal of Pain [ejp@meditos.de]

Dear Dr. Ranger,

Your submission entitled "Near-Infrared Spectroscopy (NIRS) to Assess Nociception Following Noxious Stimulation in Critically Ill Infants" has been been assigned the following manuscript number: EURJPAIN-D-11-00247.

You will be able to check on the progress of your paper by logging on to EES as an author.
The URL is http://ees.elsevier.com/eurjpain/.

Thank you for submitting your work to this journal.

Kind regards,

Dr. Bettina Haake-Weber
Editorial Assistant
European Journal of Pain

November 22, 2011
Second re-submission
Ref.: Ms. No. EURJPAIN-D-11-00247R2
Near-Infrared Spectroscopy (NIRS) to Assess Nociception Following Noxious Stimulation in Critically Ill Infants

Dear Dr. Ranger,

European Journal of Pain has received your revised submission.

You may check the status of your manuscript by logging onto Editorial Manager at (http://eurjpain.edmgr.com/).

Kind regards,

Dr. Bettina Haake-Weber
Editorial Assistant
European Journal of Pain

APPENDIX H. MANUSCRIPT #3 CONFIRMATION OF JOURNAL SUBMISSION

Sep 29, 2011

Dear Mrs Ranger,

Your submission entitled "A multidimensional approach to pain assessment in critically ill infants during a painful procedure" has been assigned the following manuscript number: CJP-D-11-00333.

If you have not already done so, please have all authors sign the copyright transfer form found on the journal's website and send it by email (julieaporter@mindspring.com), fax (303-791-3885), or mail (9312 Fernwood Ct., Littleton, CO 80126) as soon as possible.

You will be able to check on the progress of your paper by logging on to Editorial Manager as an author.

http://cjp.edmgr.com/

Thank you for submitting your work to The Clinical Journal of Pain.

Kind Regards,

Julie Porter,

Managing Editor

The Clinical Journal of Pain