EFFECTS OF DECREASING MATERNAL SEPARATION OF UNDER 6-MONTH OLD INFANTS DIRECTLY BEFORE AND AFTER SURGERY ON ALLOSTATIC LOAD AND OUTCOMES - A RANDOMISED CONTROL TRAIL

By

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DECLARATION

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ACCRYNOMS

ACTH	Adrenocorticotropic Hormone
AVP	Arginine Vasopressin Hormone
ANS	Autonomic Nervous System
ECG	Electrocardiogram
HRV	Heart Rate Variability
HF	High frequency
HPA	Hypothalamic-pituitary-adrenal Axis
IBI	Interbeat interval
ICG	Impedance Cardiograph
LC	Locus Ceruleus
LF	Low-frequency
NE	Noradrenergic
NIPS	Neonatal Infant Pain Scale
NN	Normal to Normal intervals between adjacent QRS complexes resulting from sinus node depolarizationsNeuropeptide Y
NPY	
PVN	Paraventricular Nuclei
RCT	Randomised Controlled Trial
RCWMCH	Red Cross War Memorial Children's Hospital
RMSSD	The square root of the mean of the sum of the squares of differences between adjacent NN intervals
SAM	Sympathetic Adreno-medullary system
SDNN	Standard deviation of all NN intervals
UCT	University of Cape Town
VLF	Very low-frequency
VU-AMS	Vrije Universiteit- ambulatory monitoring system

ABSTRACT

Background

The current understanding is that infants below six months old do not show separation anxiety (Bretherton, 1985).

The objective of the study was to measure the evidence of stress using heart rate variability and impedance cardiograph as indicators of autonomic nervous system activation in order to determine whether decreasing maternal separation of under 6-month-old infants directly before and after surgery decreases their stress experience.

The hypothesis was a mother's presence makes a difference to the autonomic response to stress in infants under six months old undergoing elective hernia surgery.

Methodology:

Heart Rate Variability (HRV), impedance cardiograph (ICG), pain using neonatal pain scale and taking note of the observations of what was happening around the infant were used as a measure of stress in infants undergoing hernia repair surgery. There were 8 cases, 4 in the control group (standard of care) and 4 in the intervention group (intention to treat).

Randomisation was done using sealed envelopes. Infants were randomised into two arms, the control and intervention group.

Results

The observations showed that the babies who were not separated were crying more but they settled quicker than the ones that were separated who cried less but settled much later. HRV showed that there was increased vagal tone in the intervention. ICG indicated that there were increased sympathetic signals of different sympathetic measures in all separated infants. The pain profile was not significant.

Conclusion

The above results suggest that although infants below six months may not outwardly express it, they are most likely experiencing separation anxiety. For this reason, infants should not be separated from their mothers. It is recommended that this study may be continued to get statistical significance.

KEYWORDS: Separation of infants from mothers, stress, heart rate variability, impedance cardiograph, allostatic load, allostatic state.

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GLOSSARY OF TERMS

Heart rate variability (HRV)

Is the oscillation in the interval between consecutive heart beats as well as the oscillations between consecutive instantaneous heart rates (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996)

Respiratory Sinus Arrhythmia (RSA)

These are the temporal changes in heart rate that occur synchronously with respiration. [Heart rate increases (R–R interval shortens) during inspiration and decreases (R–R interval prolongs) during expiration (Billman, 2011)

Impedance Cardiograph

Is characterised by waveforms derived from the change in thorax impedance caused by left ventricular ejection of blood into the descending aorta during the systolic phase of the cardiac cycle.

Pre-Ejection Period (PEP)

Is defined as the interval from the onset of left ventricular depolarization, reflected by the Q-wave onset in the ECG, to the opening of the aortic valves, reflected by the B-point in the ICG signal (Parry and McFetridge-Durdle, 2006)

Stroke volume (SV)

Is the average amount of blood ejected during the cardiac cycle (Parry and McFetridge-Durdle, 2006)

Vrije Universiteit Ambulatory Monitoring System (VU-AMS)

Is an ambulatory monitoring device designed to record ECG and ICG recordings from six spot electrodes (Parry and McFetridge-Durdle, 2006)

Electrocardiogram (ECG)

Is a graph-style representation of electrical activity within the heart as detected by electrodes attached to the surface of the skin and recorded by a monitor

QRS Complex (QRS)

The QRS complex reflects the rapid depolarization of the right and left ventricles

Inter beat Interval (RR interval or NN interval)

The interval between R wave and the next R wave. It is also called Normal to Normal interval. It is the interval between adjacent QRS complexes resulting from sinus node depolarizations (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996)

Heart Rate (HR)

This is the measure of cardiac activity (contractions) expressed as beats per minute

High Frequency (HF)

This is the variance of all NN intervals in high frequency range. Frequency range is 0·15–0·4 Hz. It is calculated from short term recordings of 2 to 5 minutes. The units used are ms^2 (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996)

Low Frequency (LF)

Is the variance of all NN intervals in low frequency range. Frequency range is 0·04–0·15 Hz. The units used are ms^2 (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996)

Very Low Frequency (VLF)

Is the variance of all NN intervals in very low frequency range. Frequency range is 0·003–0·04 Hz. The units used are ms^2 (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996)

SDNN

Standard Deviation of all normal R–R intervals over one minute on an ECG (Billman, 2011)

RMSSD

Root mean square of successive differences. It is used to assess the vagal tone(Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996)

Hypothalamic-pituitary-adrenal Axis (HPA)

It is the neuroendocrine system that is activated in stress leading to the final secretion of glucocorticoids

Central Nervous System (CNS)

It is the system made up of the brain, spinal cord and peripheral nerves

Left Ventricular Ejection Time (LVET)

It is the period of time over which blood is ejected from the left ventricle (Parry and McFetridge-Durdle, 2006)

CHAPTER 1: INTRODUCTION

1.1 Brief layout of the chapters

Chapter one

This chapter introduces the study. It places it in a global and South African context. It introduces the regional and historical perspective of parents and families of patients at the Hospital. It draws together the need for the study, scientific rationale, research question, research aims and objectives and the significance of the study.

Chapter two

Presents the literature reviewed, description of separation of infants from their mothers as a stressor, definition of allostatic state and load, regulation of stress, effects of stress on the body, brain and the developing brain and the measurement of stress using HRV.

Chapter three

Presents the study design and includes; the study setting; study population and sample size; sampling approach and recruitment. The inclusion and exclusion criteria are also described. Ethical considerations surrounding this study are addressed. Data collection, management and analysis are included in this chapter.

Chapter four

This chapter presents the results of the study

Chapter five

The chapter presents the discussion of the results, limitations and practice implications of the study.

Chapter six

This chapter presents the recommendation and conclusion of the study.

1.2 Introduction

This chapter places it in a global and South African context; it introduces the regional and historical perspective of parents and families of patients at the Red Cross war memorial Children's Hospital. It draws together the need for the study, scientific rationale, research question, research aims and objectives and the significance of the study.

Infants continue to be separated from their mothers in many hospital settings. Sometimes, the separation is inevitable due to life circumstances which include; mother's death, mother returning to work or to school, maternal illness or separation of the infant from the mother as a result of a court order. There are a few studies that have investigated the effect of separating infants from their mothers in various settings but none have looked at their separation during the perioperative period (Dettling, Gunnar, & Dinzella, 1999; Bergman, Linley & Fawcus, 2004)).

Maternal separation during childhood leads to increased secretion of stress hormones like adrenaline and glucocorticoids. The infant's brain is vulnerable to the effects of the stress hormones from as early as the immediate postnatal period (Lupien, McEwen, Gunnar & Heim, 2009). Prolonged exposure of the developing brain to the stress hormones leads to adverse effects (McEwen, 1998). These adverse effects lead to the allostatic state changing into allostatic load.

Allostatic load can lead to damage to the developing brain with resultant stress related diseases in adulthood (McEwen, 2003). Allostatic load and its effects will be described in greater detail in the review of the literature.

1.3 Background of Study

Infants respond in different ways when separated from their mothers. As early as 1952 (as cited in Bowlby, 1973), Bowlby observed different stages of a child's response to separation. These include protest, despair and detachment. Protest begins immediately after the infant is separated from the mother, and lasts for hours or even weeks (Field, 1996). The infant in this phase is often distressed, crying and anxiously anticipating his or her mother's return. In the despair phase, the infant shows hopelessness and becomes withdrawn from daily activities (Bowlby, 1973). The last phase is the detachment phase, where the infant shows some interest in the surroundings and interacts with another caregiver who is not the mother.

In his study of juveniles, Bowlby (1944) found that juveniles convicted of theft had been separated from their mothers at an early age for prolonged periods and they did not show any emotions of affect. Bowlby (1969) and Feldman and his colleagues (2002) showed that infants separated from their mothers developed negative emotions and poorer psychological and physiological resilience in both the short and long term.

Studies have also shown that separation of infants from their mothers is linked to an increase in the stress hormones in the circulation (Dettling, Gunnar & Donzella, 1999). Glucorticoids and catecholamines are the primary stress response hormones in the body activated due to stressful situations (McEwen, 2000). Studies by Cacioppo (1994) and Luecken & Lemery (2004) also found that the separation of an infant from the mother over a long period of time may lead to physiological changes which are the result of changes in the epigenetic settings on the DNA of the gene that produces a protein that controls behavior.

Stress is described by McEwen (2000:108) as "ways in which the body copes with psychosocial, environmental, and physical challenge". Stress that is experienced over a prolonged period results in toxic stress. Stress has been found to lead to adverse effects on the child's brain, cardiovascular, immune, and metabolic regulatory systems (Garner et al., 2012).

1.4 Historical perspective and practice norms at Red Cross War Memorial Children's Hospital

At Red Cross War Memorial Children's Hospital , it is not mandatory or expected that mothers stay with infants, except in the short stay wards. Infants are often left with no family and completely in the care of the nursing staff. While some parents stay with the infants in the hospital, which is encouraged, others are unable to stay due to various reasons. The parents that leave visit the infants whenever they can. Occasionally, they call in to find out about the infants' progress. The neonates are usually nursed in incubators while infants are nursed in cots. The feeding and care of these infants is mostly done by the nursing staff as part of their daily care and routine.

A senior clinician nurse at the hospital recounts that initially in the 1980s, mothers were not allowed to stay with the infants in the hospital. This was due to a number of reasons, including what she calls 'professional paternalism' (Bateman, 2012). Healthcare professionals believed that having the mothers present by the bedside upset the infants. The families were instead allotted visiting hours. These were between 15h00 to16h00 for the day and 19h00 to 19h30 for the night. This made it extremely difficult for mothers who worked during the day to visit. The only feasible time for them

to visit was at 19h00 for 30 minutes. During the apartheid era, curfews meant that not everyone was allowed into town. Some mothers gave up on visiting their infants in hospital because of transport issues and the limited time spent with their infants. The laws, added to the hospital norms and practices made it impossible for the mothers to be with their infants.

In the late 1980s, the senior clinician nurse and a social worker in the respiratory department at the Hospital began to advocate for a tracheostomy home care programme where mothers were trained to look after their infants at home. This was implemented with success. Later, in the early 1990s, this team motivated for the mothers to stay in the hospital. The current policies at Red Cross War Memorial Children's Hospital now encourage mothers to stay at the bedside with their infants.

1.5 Statement of the Problem

At times, unavoidable circumstances lead to the separation of infants from their mothers. The challenge is that prolonged periods of separation result in stress. Stress is described by McEwen (2000:108) as "ways in which the body copes with psychosocial, environmental, and physical challenge". Stress that is experienced over a prolonged period results in toxic stress. Stress has been found to lead to adverse effects on the child's brain, cardiovascular, immune, and metabolic regulatory systems (Garner et al., 2012). These adverse effects lead to the allostatic state changing into allostatic load.

Research has been done to measure stress regulation or balance in neonates and preterm infants when separated from their mothers. It demonstrated that infants who were not separated from their mothers' immediately after birth and were put in skin-to-skin contact had a more stable cardiovascular system when compared to those who were separated and nursed in incubators (Bergman, Linley & Fawcus, 2004). Another study was performed on neonates to determine whether there was a difference between sleeping alone or with the mother. It found significant stress indicators in those neonates who slept alone, with increase in high frequency heart rate variation power during quiet sleep when compared to those that maintained skin-to-skin contact with their mothers (Morgan, Horn & Bergman, 2011). While studies like these have demonstrated measurable separation stress in newborns, there was no evidence in the literature of studies done in infants between one and six months old.

At the Children's Hospital, the current standard of practice for infants undergoing surgery is to separate those younger than six months of age from their mothers/guardians as they go to theatre. The evidence for this practice is based on observational studies that suggested that separation anxiety only occurred in infants after 6 months of age (Bretherton, 1985).

1.6 Research Question

Does decreasing maternal separation of under 6-month-old infants directly before and after surgery decrease their stress experience?

1.7 Research Aim

To measure evidence of stress using heart rate variability and impedance cardiograph as indicators of autonomic nervous system activation in order to determine whether decreasing maternal separation of under 6-month-old infants directly before and after surgery decreases their stress experience.

1.8 Objectives

- To determine if separating infants from their mothers before and immediately after undergoing hernia surgery leads to increased allostatic state as measured by increased heart rate variability and impedance cardiograph.
- To determine if shortening the separation period around a surgical intervention decreases stress in infant allostatic state and physiology.
- To compare differences in pain levels between infants who are separated from their mothers before undergoing hernia surgery and those who are not.

1.9 Hypothesis

A mother's presence makes a difference to the autonomic response to stress in infants under six months old undergoing elective hernia surgery.

10 Significance of the Study

The study will examine the current knowledge on infant separation from the mother, with focus on the importance of mother-child interaction. Information generated on the effects of separation of

infants from their parents for these brief periods of stress has the potential to alter practice norms in children's hospitals and to increase our understanding of infant stress in infants under six months old.

11. Conclusion

The background to this study within its global, South African and regional contexts have been presented. The problem has been described, aim, hypothesis, research question, research objectives and significance of the study stated.

CHAPTER 2: LITERATURE REVIEW

2.1 Introduction

This chapter resents the literature reviewed, description of separation of infants from their mothers as a stressor, definition of stress, allostatic state and load, regulation of stress, adaption to stress, effects of stress on the body, brain and the developing brain, the measurement of stress using HRV and pain.

A literature search was conducted from 1990 to 2013 using the following databases: CINAHL; MEDLINE; PubMed Central; EBSCOHost; Science Direct; Cochrane Collaboration; Google Scholar; Academic Search Premier; PsycINFO and relevant textbooks. The keywords that were used include: Separation of infants from mothers, stress, heart rate variability, impedance cardiograph, allostatic load, allostatic state and pain. Original and seminal research of John Bowly published in the 1940s was included.

2.2 Definition of stress

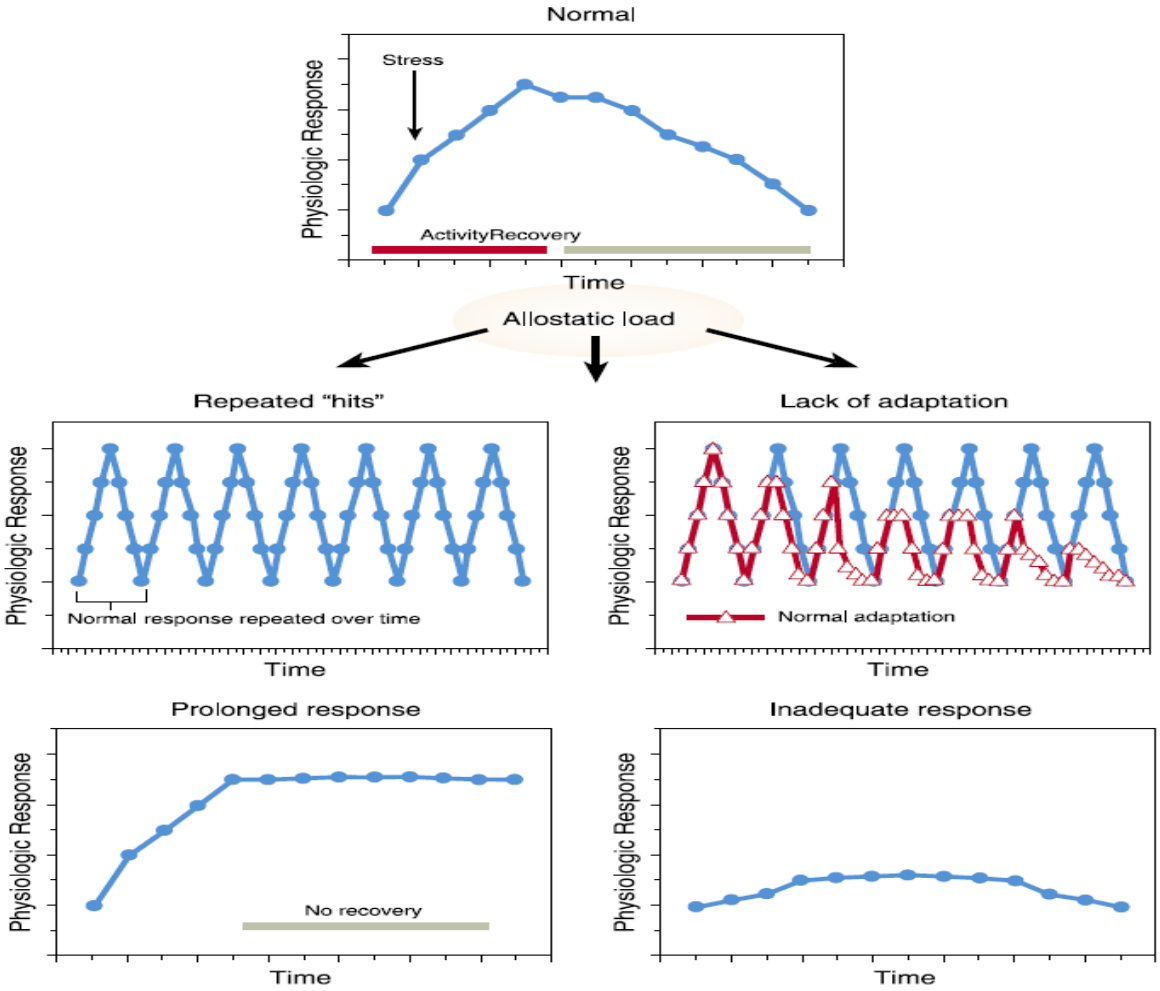
In life, the body is exposed to many acute stressors. Life exists by maintaining a complex adaptive dynamic equilibrium to these intrinsic or extrinsic adverse forces or stressors (Chrousos, 1998; McEwen, 2000a).The stressors may be related to the environment in the home, workplace, or neighbourhood which in turn affects the individual (McEwen BS, 1998).

Due to the ambiguity in the definition of term "stress", Sterling & Eyer (1988) came up with the term "allostasis" to define the body homeostasis (Sterling & Eyer, 1988). The term "allostatic load" was then used to define the state of chronically increased allostasis that can lead to pathology (McEwen BS, 1998; McEwen & Wingfield, 2003).

Body systems like the autonomic nervous system, neuroendocrine system and immune system mediate an individual's adaptation to the adversities of daily life. The active process by which the body responds to daily events and maintains homeostasis is referred to as "allostasis" (Sterling & Eyer, 1988; McEwen, 1998; McEwen, (2000a,b,c); McEwen, 2003). The mediators released from these systems like glucocorticoids, catecholamines and cytokines act on receptors in various organs and tissues to produce the desired adaptive effect in the short term. This is referred to as the "allostatic state". Prolonged effect on the target organs which may lead to receptor desensitization and tissue damage is what is termed as "allostatic load" (McEwen, 1998; McEwen, 2000a, b, c; McEwen & Wingfield, 2003; McEwen 2003; Korte et al, 2005).

In adults, allostatic load could refer to the cumulative wear and tear on body systems caused by too much stress and/or inefficient management of the systems that promote adaptation through allostasis (McEwen, 2006). Allostatic load can therefore be due too much stress, failure to turn off the stress response when it is no longer needed, and inability to turn on an adequate response in the first place or not habituating to the recurrence of the same stressor and thus dampening the allostatic response (McEwen, 2007).

Figure 1: Graphic representation of types of allostatic load



Top panel: illustrates the normal allostatic response, in which a response is initiated by a stressor, sustained for an appropriate interval, and then turned off.
The remaining panels illustrate four conditions that lead to allostatic load:
top left, repeated "hits" from multiple stressors; *top right*, lack of adaptation; *bottom left*, prolonged response due to delayed shut down; *bottom right*, inadequate response that leads to compensatory hyperactivity of other mediators (e.g., inadequate secretion of glucocorticoid, resulting in increased levels of cytokines that are normally counterregulated by glucocorticoids).
[Adapted from McEwen (2007)]

In young children, the stress response can be categorized into three distinct types; positive, tolerable and toxic stress response. The classification is based on the potential to cause lasting physiologic disruptions because of the intensity and duration of the response and the presence or absence of a caring and responsive adult (National Scientific Council on the Developing Child, 2005; Shonkoff & Garner, 2011).

When a child experiences a physiologic state that is brief, with a magnitude that is mild to moderate, in the presence of a caring and responsive adult who helps the child cope with the stressor, this is referred to as a positive stress response. The adult provides a protective effect which facilitates the stress responses to return to their baseline (Shonkoff, Boyce, & McEwen, 200; Shonkoff & Garner,

2011). This may lead to a short-lived increase in heart rate or a mild change in the body's hormone stress levels (National Scientific Council on the Developing Child, 2005).

In the tolerable stress response, the child experiences excessive activation of the stress response system that can lead to physiologic harm and long term consequences for health and ability to learn. The presence of a supportive, caring and protective adult that helps the child to cope and maintain control. This makes the stress response tolerable. It reduces the stress response and facilitates a return to baseline status (National Scientific Council on the Developing Child, 2005; Shonkoff & Garner, 2011).

A toxic stress response results from a strong, frequent, or prolonged activation of the body's stress response systems in the absence of the buffering protection of a supportive, caring and protective adult relationship (National Scientific Council on the Developing Child, 2005; Shonkoff, 2011). Stable and supportive relationships are required to teach and guide infants on how to deal with stress (Middlebrooks & Audage, 2008).

2.3 Regulation of stress in the body

Once the body is stressed, a number of physiologic and behavioural responses are set into motion to try and re-establish homeostasis. Chrousos refers to this phenomenon as the "stress system". It is located in the periphery and the central nervous system (Chrousos and Gold, 1992). Stress hormones are produced that convert the physical or emotional stress into chemical signals that are sent throughout the body as well as to the brain(The National Scientific Council on the Developing Child, 2005).The behavioural and physical changes when activated improve the individual's chances of survival as well as adaptation to the environment (Chrousos, 1998).

2.3.1 Central components of the stress system

The central components of the stress system are located in the hypothalamus and the brainstem and include the parvocellular corticotropin-releasing hormone (CRH) and arginine-vasopressin (AVP) neurons of the paraventricular nuclei (PVN) of the hypothalamus and the CRH neurons of the paragigantocellular and parabranchial nuclei of the medulla, as well as the locus ceruleus (LC) and other mostly noradrenergic (NE) cell groups of the medulla and pons (Chrousos, 1998).

CRH, a 41 amino acid peptide, is the principal hypothalamic regulator of the pituitary–adrenal axis (Chrousos, 1998; McEwen, 2002). CRH receptors are found in the limbic system, basal forebrain, locus ceruleus, noradrenergic cells of the medulla and the pons. This sets into motion a coordinated array of behavioural and peripheral responses, which include characteristic stress behaviours and activation of the pituitary–adrenal axis and the sympathetic nervous system. The secretion of CRH is stimulated by neuropeptide Y (NPY) (Chrousos, 1998).

Substance P (SP) inhibits the paraventricular nuclei (PVN) CRH neuron, while it activates the locus ceruleus (LC) and noradrenergic (NE) cell groups of the medulla and pons (LC/NE system) (Chrousos, 1998). Substance P is elevated centrally, when there is peripheral activation of somatic afferent fibers and may, thus, have relevance to changes in the stress system activity in chronic inflammatory or painful states (Chrousos, 1998).

2.3.2 Peripheral components of the stress system

The peripheral limbs of the stress system are the hypothalamic–pituitary–adrenal (HPA) axis and the autonomic nervous system (the efferent sympathetic/adrenomedullary system [SAM] and components of the parasympathetic system) (Chrousos, 1998; Luecken & Lemery, 2004; The National Scientific Council on the Developing Child, 2005).

2.4 The Hypothalamic-Pituitary-Adrenal Axis

The CRH is released from the hypothalamus into the hypophyseal portal system. It acts on the anterior pituitary gland, leading to secretion of Adrenocorticotropic hormone (ACTH) (Tsigos and Chrousos, 1995). Under non-stressful situations, CRH and Arginine Vasopressin hormone (AVP) are secreted in the portal system in a circadian and highly concordant pulsatile manner with higher amplitudes during the morning. This results in concomitant increases in ACTH and cortisol secretion. This is to enable the body to carry out the stressful activities during the day. In the evening, the secretion levels are lowered to enable the body to rest (Chrousos, 1995; McEwen et al, 1993). This diurnal variation can be interrupted by stress, feeding schedules, and changes in exposure to light and alteration in the level of activity (Chrousos, 1998). In situations of acute stress, there is an increase in the amplitude and frequency of secretion of CRH and AVP (Chrousos, 1995). The pituitary derived ACTH acts on the adrenal cortex leading to the secretion of glucocorticoids, adrenal

androgen and aldosterone from the zonae fasculata, reticularis and glomerulosa respectively (Chrousos, 1998).

Glucocorticoids are involved in actions responsible for generating energy in the form of glucose. They are also involved in production of inflammatory cytokines and the shift in balance of the immune response from cellular towards humoral immunity (Sapolsky et al, 2000; McEwen, 2002).

Proliferation of the stress response is prevented by actions of the adrenal gland steroids. They suppress the production of inflammatory cytokines, minimize the activation of the brain's noradrenergic system and reduce the effects of insulin (McEwen, 2002). Glucocorticoid feedback both delays the onset of the ACTH response to stress and inhibits the magnitude of the increase in ACTH during stress (Keller-Wood, M. & Dallman, 1984). Glucocorticoids control the basal HPA activity and terminate the stress response by acting on the hippocampus and frontal cortex, the hypothalamus and the pituitary gland (Tsigos & Chrousos, 1995).

There are two types of glucocorticoid receptors in the central nervous system. The glucocorticoid Type I receptors respond to low levels of glucocorticoids. The Type II receptors respond to basal and stress levels of glucocorticoids (Chrousos, 1998).

2.5 The Autonomic Nervous System

The autonomic nervous system is the body's initial response to stress. It is divided into the sympathetic and parasympathetic systems. They control a number of systems which include; the cardiovascular, respiratory, gastrointestinal, renal, and endocrine (Chrousos & Gold, 1992). The parasympathetic system can assist the sympathetic functions by withdrawing its activity or it can antagonize them by increasing its activity (Chrousos, 1998).

Sympathetic innervation of peripheral organs is derived from the efferent, primarily cholinergic, preganglionic fibres, whose cell bodies lie in the intermediolateral column of the spinal cord. These nerves then synapse in the bilateral chain of sympathetic ganglia (Chrousos, 1998). The mostly adrenergic, postganglionic sympathetic neurons innervate the smooth muscle of the blood vessels, heart, skeletal muscles, kidney, gut, fat, and many other organs (Chrousos, 1998). Through the adrenal medulla, the sympathetic system provides all of the circulating epinephrine and some of the norepinephrine (Chrousos, 1998). The epinephrine and norepinephrine generated mobilizes energy stores and alters blood flow, thereby allowing the body to effectively deal with a range of stresses (Sapolsky et al, 2000).

2.6 Adaptation to stress

Individual adaptation to stress can be behavioural or physical. In adults, behavioural adaptation includes increased arousal, alertness and vigilance, improved cognition, and focused attention, as well as euphoria or dysphoria, depending on the stressor and the memory of the organism (Chrousos, 1998). It also includes enhanced analgesia and elevations in core temperature, along with concurrent inhibition of vegetative functions, such as appetite, feeding, and reproductive function (Chrousos, 1998).

Physical adaptation changes take place principally to promote an adaptive redirection of energy. Thus, oxygen and nutrients are shunted to the CNS and the stressed body site(s), where they are needed the most (Chrousos, 1998). Increases in cardiovascular tone (heart rate, cardiac ejection fraction, and arterial blood pressure), respiratory rate, and intermediate metabolism (gluconeogenesis, lipolysis) all work in concert to promote availability of vital substrates (Chrousos, 1998). Digestion, growth, reproduction, and the immune system are inhibited during stressful periods (Chrousos, 1998).

2.7 Effects of stress on the body

There is a lot of variability between individuals in their adaptive responses to stress. The adaptive responses may be inadequate to re-establish homeostasis or excessive and prolonged (Chrousos, 1998). In either case, a healthy steady state is not attained and pathology may ensue (Chrousos, 1998). It is important that a person's response to stress is in neither of the two extremes (inadequate or excessive) to enable him/her to develop appropriately, perform tasks adequately and have positive social interactions (Charmandari et al, 2003).

Multiple factors determine an individual's stress response. The stress response could be a result of an inherited genetic makeup of the individual. It has been estimated that about half to two-thirds of reliable variance in measured personality traits is due to genetic influences (Chrousos, 1998; Barr et al, 2004). The genetic polymorphisms and/or clinically significant alterations in the expression of genes involved in the regulation of the stress system, their receptors and regulators, are expected to account for the observed variability in the function of the stress system (Chrousos, 1998; Barr et al, 2004). They have been referred to as "vulnerability genes". Early stressors to these genes lead to their dysfunction resulting in problems in stress hormone regulation and behavioural difficulties (Chrousos, 1998; Barr et al, 2004).

The individual variability in the stress responses could also be caused by environmental factors. The environmental factors may be adverse experiences earlier on in life. The increments in the stress

hormones can be temporary, prolonged or excessive (McEwen, 1998; McEwen &Seeman, 1999). Temporary increases in the stress hormone levels are protective to the individual and are necessary for survival but excessive amounts or prolonged exposure to these hormones can be harmful (McEwen, 1998; McEwen & Seeman, 1999). Exaggerated or prolonged exposure to cortisol is associated with accelerated aging and increased risk of cognitive impairments, cardiovascular disease, infectious diseases, and other illnesses (McEwen, 1998).

The SAM component of the stress response results in increased cardiovascular arousal, secretion of norepinephrine and epinephrine into the blood stream, and the halting of non-essential parasympathetic functions. Prolonged cardiovascular arousal has been associated with the development of hypertension and organ damage (Luecken & Lemery, 2004).

The stress response is ideally supposed to be sustained for a limited duration. In this way, its effects of immunosuppression, increased catabolism, prevention of growth and reproduction are temporarily beneficial. Once the effects are sustained over a prolonged period of time, they become a pathological syndrome (Chrousos, 1998). When the high levels of glucocorticoids are sustained for a considerable period of time, the catabolic actions will lead to muscle wasting and loss of bone mass. There will also be suppression of immune function, development of insulin resistance and Type II diabetes (McEwen, 2002)

2.8 Effects of stress on the developing brain

The brain is responsible for learning, memory and the expression of emotions. The brain also controls stress hormone secretion and the autonomic nervous system through a number of pathways which in turn regulate the cardiovascular function, food intake and metabolism, and the immune system (Chrousos, 1998; McEwen, 2002).On the other hand, the brain is a target of stress. It therefore possesses its own allostatic mechanisms in adapting to challenges (McEwen, 2002).

The amygdala mediates fear and anxiety and activates the physiologic stress response. Its stimulation activates the sympathetic system and causes neurons in the hypothalamus to release CRH (Shonkoff & Garner, 2011). The amygdala also has a major role in long term memories associated with traumatic events (Cahill et al, 1996). Adrenaline and glucocorticoids help in the formation of these memories (Cahill et al, 1994; Roozendaal & McGaugh, 1997). Significant stress in early childhood leads to hypertrophy of the amygdala with a resultant chronically activated physiologic stress response. This potentially leads to increased fear and anxiety (Tottenham et al, 2010).

The hippocampus is important for the memory of context, everyday events and general information about the time and place of events (Eichenbaum, H. & Otto, 1992). In association with the amygdala, memories of the time and place of events with strong emotional bias are also stored here (LeDoux, 1995). The hippocampus regulates the stress response by preventing the shutdown of the HPA response (Jacobson & Sapolsky, 1991). High levels of cortisol inhibit neurogenesis in the hippocampus leading to atrophy and reduced connection to the rest of the brain (McEwen et al, 1995; Brunson et al, 2002; Conrad et al, 2009; Shonkoff & Garner, 2011). Toxic stress limits the ability of the hippocampus to promote contextual learning, making it more difficult to discriminate conditions for which there may be danger versus safety later in life and also affects short term memory (Kirschbaum et al, 1996; Shonkoff & Garner, 2011).

The prefrontal cortex (PFC) plays an important role in the development of executive functions like decision making, behavioural self, mood and impulse control and working memory (Shonkoff & Garner, 2011). It regulates the sympathetic and parasympathetic effects and also participates in turning off the cortisol response (Shonkoff & Garner, 2011). It suppresses amygdala activity, allowing for more adaptive responses to potentially threatening or stressful experiences. Stress and elevated cortisol results in changes in the connectivity within the PFC, which may limit its ability to inhibit amygdala activity (Shonkoff & Garner, 2011).Experiences of severe stress early in life is associated with subsequent problems in the development of linguistic, cognitive, and social-emotional skills (Korte et al., 2005).

The changes in the developing brain architecture can have potentially permanent effects on a range of important functions, such as regulating stress physiology, learning new skills, and developing the capacity to make healthy adaptations to future adversity (McEwen & Gianaros, 2011).

In studies done in rats, allostatic load has been shown to increase the brain aging process (Kerr et al., 1991). Animal studies have shown that repeated stress causes brain regions involved in memory and emotions, such as hippocampus, amygdala, and prefrontal cortex, to undergo structural remodeling which leads to impairment of memory, increase in anxiety and aggression (McEwen, 2006).

Stress has been found to lead to adverse effects on the child's brain, cardiovascular, immune, and metabolic regulatory systems and on the child's development (Garner et al., 2012). The most affected areas in the brain are the amygdala, hippocampus, prefrontal cortex and orbito-frontal cortex (McEwen & Gianaros, 2011). The National Scientific Council on the Developing Child (2005) explains that prolonged exposure of infants to stress mediators results in impairment of the

connections of the brain circuits and, in the extreme of cases, results in the development of a smaller brain (The National Scientific Council on the Developing Child, 2005). The foetal, infant and early childhood brain is continuously undergoing reorganization in the neural pathways and synapses. This makes it particularly sensitive and vulnerable to the chemical influences of the stress hormones which may lead to disruption in the structural formation of the developing brain (Chrousos, 1998; The National Scientific Council on the Developing Child, 2005; Middlebrooks & Audage, 2008).

Adverse events in early postnatal life may affect future reactivity to stress experiences by altering the developing neural circuits controlling the neuroendocrine responses. These events in effect control certain characteristics and behaviours in the future life of the individual (Szyf 2009; Roth et al, 2009; Shonkoff & Garner, 2011). Significant stress in the lives of young children is a potential risk factor for the development of health-threatening behaviours and a catalyst for physiologic responses that lays the groundwork for chronic, stress-related diseases and impairment in learning and behaviour later in life (Felitti, 2009; Shonkoff & Garner, 2011).

The potential of developing such devastating effects to the individual later in life as a result of the severe stress experienced in early life underlines the importance of screening for significant stressors in the early years of life. Interventions that strengthen the capacities of families and communities to protect young children from the disruptive effects of toxic stress are likely to promote healthier brain development and enhanced physical and mental wellbeing (Shonkoff & Garner, 2011). However, it is incorrect to conclude that all infants that experience stressful events early in life suffer stress related disorders later in life.

2.9 Separation as a stressor

At times, unavoidable circumstances lead to the separation of infants from their mothers. The challenge is that prolonged periods of separation deprive the infant of a useful buffer during a stressful experience (Nachmias et al, 1996; National Scientific Council on the Developing Child, 2005; Shonkoff & Garner, 2011).

It has been shown that the parasympathetic system is needed in order to have optimal development. However, separation of infants from their mothers results in a sympathetically dominated programme (along with neuroendocrine and somatic responses) where homeorhesis is achieved but with the objective of survival rather than development (Bergman, Linley & Fawcus, 2004). In their study, Morgan and his colleagues found the autonomic activity to increase by 176% during maternal-neonatal separation when compared to skin-to-skin contact (Morgan, Horn & Bergman, 2011). In

their conclusion, they thought maternal separation was a stressor that neonates were unable to cope with.

It is widely acknowledged that surgery is a stressful experience to the children and their families. Because the presence of family members minimizes the anxiety experienced by the children, the Royal College of Surgeons promotes the accompaniment of the children by their family members to the anaesthetic room and from the recovery area (Royal College of Surgeons, 2007). However, there is very little documented evidence in support of who accompanies the child to and from theatre.

Kangaroo mother care has its major component as skin-to-skin contact between the mother and her newborn infant. Other components are frequent and exclusive or almost exclusive breast feeding and early discharge from hospital (Conde-Agudelo & Díaz-Rossello, 2014). In their systemic review, Conde-Agudelo & Díaz-Rossello, 2014, found that kangaroo mother care in low birth weight babies reduced morbidity and mortality increased weight, head circumference, and length gain, breastfeeding, mother satisfaction with method of infant care, some measures of maternal-infant attachment, and home environment. However, there were no differences in neurodevelopmental and neurosensory outcomes at one year of corrected age. In another review, Moore et al., 2012, showed that healthy newborn infants on skin-to-skin contact cried less and interacted more with their mothers, breastfed better and had better physiological adaptation.

2.10 Heart rate variability (HRV)

Heart rate variability (HRV) is the beat-to-beat variation in heart rate or the duration of the R–R interval on the Electrocardiogram [ECG] (Billman, 2011). The temporal changes in the heart rate are synchronous with respiration. Increased heart rate is associated with inspiration and hence, a shortened R-R interval. Conversely, heart rate decreases during expiration and there is associated prolongation of the R-R interval. This is referred to as respiratory sinus arrhythmia (RSA) (Billman, 2011).

The extrinsic control of the heart is by the autonomic nervous system. The sympathetic influence increases the heart rate while the parasympathetic provides tonic inhibitory control thereby reducing the heart rate (Thayer et al., 2012).

The parasympathetic influence on heart rate (HR) is mediated via release of acetylcholine by the vagus nerve while the sympathetic influence on heart is mediated by release of epinephrine and norepinephrine (Jalife and Michaels, 1991). Sympathetic stimulation, occurring in response to stress,

exercise and heart disease, causes an increase in HR by increasing the firing rate of pacemaker cells in the heart's sino-atrial node (Jalife & Michaels, 1991). Under resting conditions, vagal tone prevails and variations in heart period are largely dependent on vagal modulation (Thayer et al., 2012).

The relative sympathetic increases cause the inter-beat interval to become shorter and relative parasympathetic increases cause the inter-beat interval to become longer (Thayer et al., 2012). Sympathetic activity is associated with a low frequency of about 0.15 Hz (Saul, 1990), while parasympathetic activity is associated with a higher frequency range (0.15–0.4 Hz) of modulation frequencies of the HR. This difference in frequency ranges allows HRV analysis to separate sympathetic and parasympathetic contributions (Acharya, Paul Joseph, K., Kannathal, N., et al, 2006). Frequencies below 0.15 Hz represent a mixture of sympathetic and parasympathetic autonomic influences (Thayer et al., 2012). As a result of overlap of the parasympathetic and sympathetic activity, the cardiac function is characterised by irregular time intervals between consecutive heart beats (Thayer et al., 2012). This variability is a result of rhythmic oscillation of the regulatory components of cardiac activity that function to maintain cardiovascular homeostasis within a defined range and to orchestrate responses to challenges (Von Borell et al., 2007).

The sympathetic effects on the heart are slow, on a time scale of seconds, whereas the parasympathetic effects are fast, on a time scale of milliseconds. Therefore the parasympathetic influences are the only ones capable of producing rapid changes in the beat to beat timing of the heart (Thayer et al., 2012). The issue of the definite contributions of the sympathetic and parasympathetic divisions of the ANS to the HRV remains contentious (Parati et al, 2006).

A number of measures have been used to evaluate the variation in the heart rate to provide indices for cardiac autonomic regulation. The two primary methods for analyzing the HRV are the time domain and frequency domain methods (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).

The time domain methods are the simplest to perform. They involve either geometric or statistical measures. Intervals between successive normal QRS complexes (RR or NN intervals) as determined from a continuous ECG record are used for the calculations (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996). In the geometric analysis, a series of RR intervals are converted into a geometric pattern such as a sample density distribution of RR intervals or a sample density distribution of differences between adjacent RR intervals and a formula is used to judge the variability based on the geometric and /or graphic

properties of the resulting pattern (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).

In the statistical analysis, direct measurements of RR intervals from an ECG or those derived from differences between RR intervals are used to calculate variables. The simplest variable to calculate is the standard deviation of the NN interval (SDNN), i.e. the square root of variance (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996). Since variance is mathematically equal to total power of spectral analysis, SDNN reflects all the cyclic components responsible for variability in the period of recording (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).

The statistical analysis provides more detailed information of a continuous sequence of normal beats for the time period of interest when compared to the geometric analysis (Billman, 2011).

Frequency domain methods are derived from power spectral density (PSD) analysis. PSD provides the basic information of how the total variance of a continuous series of beats distributes as a function of frequency (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996; Berntson et al., 1997). PSD can be calculated using either parametric or non-parametric methods. Both methods provide comparable results (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).

Non-parametric methods employ the Fast Fourier Transform (FFT) algorithm which is simple and has a high processing speed. Parametric methods on the other hand depend on Auto-regressive (AR) modelling (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996; Denver et al, 2007). They have smoother spectral components which can be distinguished independently of preselected frequency bands, have easy post-processing of the spectrum with an automatic calculation of low and high frequency (LF and HF) power components and easy identification of the central frequency of each component and they have an accurate estimation of PSD even on a small number of samples on which the signal is supposed to maintain stationarity (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).

During short duration recordings, (2–5min) three main peaks are often identified: very low frequency (VLF) <0.04 Hz, low frequency (LF), 0.04–0.15Hz, and high frequency (HF) 0.15–0.4Hz (Task

Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996). During exercise and in infants, HF is shifted to a higher frequency range (0.24–1.04Hz) (Berntson et al., 1997). Ultra low frequency (ULF) (0.003–0.04 Hz), is obtained during longer recording periods (24 hours) (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).The absolute power at a given frequency is reported as ms^2, but LF and HF power are often measured in normalized units (nu) obtained by dividing the frequency band of interest by total power minus VLF. RSA, RMSSD and the high frequency (HF) component of the power spectrum are closely related and are all primarily parasympathetically mediated (Thayer et al, 2012).

There are non-linear methods that have been used to evaluate HRV which are derived from the Chaos Theory (Billman, 2011). They evolve around the premise that small changes in initial conditions lead to large changes with reiteration. To state it mathematically, a single value *y* can be associated with more than one value of *x* (Denton et al., 1990). In principle, these methods have been shown to be powerful tools for characterization of various complex systems, however; no major breakthrough has yet been achieved by their application to bio-medical data including HRV analysis (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).

2.10.1 Limitations of measuring HRV

Part of the HRV occurs as a consequence of mechanical events like stretching of the atria that result from both changes in cardiac filling and changes in intrathoracic pressure that occur during respiration (Taylor et al., 2001). Independent of cardiac autonomic regulation, respiratory parameters can alter heart rate and RR interval variability (Brown et al., 1993). HRV only provides an indirect assessment of cardiac autonomic activity and does not provide a direct measurement of either cardiac parasympathetic or sympathetic nerve activity (Billman, 2011).

2.10.2 HRV as a measure of stress:

HRV is a marker for flexible dynamic regulation of autonomic activity (Thayer et al, 2012). Since the autonomic nervous system modulates the heart rate on a beat-to-beat timescale, HRV is therefore a measure of parasympathetic-sympathetic balance with higher HRV reflecting higher parasympathetic activity (Thayer et al, 2012). Higher HRV denotes a relatively greater parasympathetic nervous system (PSNS): sympathetic nervous system (SNS) activity ratio (Porges, 2007). It has been found to correlate with physiological and emotional security and interpersonal openness. Conversely, a relatively lesser PSNS: SNS activity ratio, has been correlated with

physiological and emotional insecurity and the fight or flight response (Porges, 2007). Individuals with greater control of their emotions have been shown to have a higher resting HRV (Appelhans & Luecken, 2006).

In adults, it has been shown that phasic increases in HRV in response to situations that require emotional control show effective emotional regulation (Ingjaldsson et al., 2003).There is a connection between the brain and the heart. Efferent outflow from the brain affects the heart and afferent outflow from the heart affects the brain (Thayer et al, 2012). HRV is structurally and functionally linked to the amygdala and the prefrontal cortex (Thayer & Lane, 2000) and may index the degree to which the brain's threat-detection systems produce allostatic load or show an exaggerated stress response (Thayer et al, 2012). The importance of HRV in determining the allostatic load is dependent on its ability to reflect the functioning of the areas in the brain involved in the response to stress.

2.11 Impendence Cardiograph (ICG)

Impedance cardiograph (ICG) is a non-invasive technique of measuring technique of measuring blood flow in the thoracic cavity (Lasater & Von Rueden, 2003). Current is transmitted from one set of electrodes to another and it travels along the path of least resistance which is the blood in the aorta. The ICG then measures the baseline impedance, pulsatile impedance with time and the ECG which are then used to calculate the various measures of cardiac function which include cardiac output, stroke volume (SV), pre-ejection periods (PEP) and left ventricular ejection times (LVET) (Parry & McFetridge-Durdle, 2006) .

Shoemaker and his colleagues found Impedance cardiograph to be a valid, reliable and valid technique for measuring parameters of cardiovascular function (Shoemaker et al.,1996, 1998, 2001).

Table 1:Measures of Cardiac Function Generated by Impedance Cardiograph and their Definitions

Hemodynamic Variable	Parameter	Definition
Thoracic fluid status	Z_0 = thoracic impedance	Baseline fluid status in chest
Left ventricular function	CO = cardiac output	Amount of blood ejected from the left ventricle in 1 minute
	CI = cardiac index	Cardiac output divided by body surface area
Preload	SV = stroke volume	Amount of blood ejected with each beat
Afterload	SVR = systemic vascular resistance	Amount of resistance that the heart must pump against
Contractility	d*Z*/d*t* = impedance changes over time	Reflects the force of ventricular contraction
	PEP = pre-ejection period	Time from ventricular depolarization to ventricular ejection
	LVET = left ventricular ejection time	Period of time over which blood is ejected from the left ventricle

Adapted from (Parry and McFetridge-Durdle, 2006)

2.12 Pain

Pain as defined by the the Committee on Taxonomy of the International Association for the Study of Pain is *"An unpleasant sensory and emotional experience associated with actual or potential tissue damage or described in terms of such damage"*. (Merskey, 1994).

Pain impulses from the nociceptors are transmitted along sensory nerves to the spinothalamic tracts through thalamocortical pathway to the primary somatosensory cortex where pain is perceived (Lee et al, 2005). Pain perception in neonates does not engage the same structures as in human adults (Lowery et al., 2007). In neonates, it involves multi-layered networks of nociceptors, nerve fibres, neurons, and glia distributed in multiple spinal and supraspinal areas, forming diverse feed-back and feed forward loops (Woolf & Salter, 2000). The thalamus plays a central role in pain processing by regulating the spinal– brainstem–spinal loops that mediate descending facilitation or inhibition

(Craig, 2003). There is activation of the HPA axis which is linked to the emotional pain response (Lowery et al., 2007).

2.13 Conclusion

This chapter focused on the definition of stress, its effects and how the body adapts to stressful events. Effects of stress on the developing brain were looked at along with how the different areas of the brain are affected by stress. Separation and surgery were focussed on as stressors and the presence of mother was noted to be a significant buffer to stressful events.

Heart Rate Variability (HRV) and Impedance Cardiograph (ICG) were defined along with the variables that can be derived from them. The relationship between stress and HRV was explored and the limitations of using HRV as a measure of stress.

CHAPTER 3: METHODOLOGY

3.1 Introduction

This chapter presents the study design, setting, population, sample size, sampling approach, recruitment, the inclusion and exclusion criteria. Ethical considerations surrounding this study are addressed. Pilot study, data collection, management and analysis are also included in this chapter.

3.2 Study design

A prospective randomized controlled clinical trial was done to investigate the effects of maternal separation on infants younger than 6 months of age undergoing surgery for hernia repair. This was a double blinded study for both the researcher and the mother before randomisation. Neither the mother nor the researcher knew which group they would be allocated to. The study had a control and experimental group.

3.3 Study setting

The study was conducted at Red Cross War Memorial Children's Hospital. This is a tertiary hospital situated in Rondebosch in the Southern suburbs of Cape Town. It is a public hospital, serving as a referral center for tertiary and quaternary specialist paediatric care. It is run by the Western Cape Government.

3.4 Study population

The target population of the study was infants who were admitted for day surgery for inguinal hernia repair. They were between the ages of one to six months. Both male and female infants were recruited from various socio-demographic backgrounds. There was no discrimination on cultural, religious, economic or ethnic backgrounds. All infants who attended the surgical clinic or arrived directly to the surgical ward having been referred for bi- or unilateral hernias were screened using the inclusion and exclusion criteria.

3.5.1 Sample size

The sample size of 30 participants was calculated using the G power for moderate effect. G Power is a software packages for performing sample size and power calculations. It covers a wide range of study designs and is useful if using effect sizes rather than absolute values (McCrum-Gardner, 2010).

3.5.2 Randomisation

Randomisation was done utilizing a computerized random integer generator from www.random.org. These numbers were printed on 3x5 index cards and kept in order in opaque sealed envelopes. These were allocated to the mother and infant dyad in sequence and indicated whether the dyads are randomized to standard of practice or the intervention group. Neither the researcher nor the mother knew which group they would be assigned to and consent was therefore negotiated prior to randomisation. Blinding of the investigator was not possible as the mother accompanied only the intervention group and not the control group. Data strips (ECG/ICG) were anonymised and therefore this was blinded.

3.6 Inclusion criteria

- Infants who were under six months old undergoing inguinal hernia repair surgery.
- Infants who were admitted on the ward with their mothers or their familiar primary caregiver.
- Mothers of infants who consented to participate in the study.

3.7 Exclusion criteria

- Infants who had more than one clinical condition/ reason for admission.
- Infants whose mother were absent during admission and day of surgery
- Mothers who were not willing to participate in the study

3.8 Recruitment

Recruitment took place in the surgical clinic at Red Cross War Memorial Children's Hospital when the infants came for their appointments prior to the elective surgery. Recruiting infants in the study was done in the outpatient clinic and the ward. For infants who were born at term, recruitment took place in the outpatient clinic. Those who were born prematurely with their corrected age below 40 weeks were seen via the emergency unit. They were triaged orange and then sent directly to the surgical ward. Once there, they were assessed by a doctor who either admitted them for surgery or gave them an appointment day to return for the surgery.

The infants who were recruited in the study were under six months old and were undergoing inguinal hernia repair. The mother was informed about the study and given a chance to decide whether to participate or not. The researcher then managed the informed consent process after the mother had agreed to participate in the study.

3.9 Procedure

There were two groups of mother-infant dyads; the intervention group and the control group.

Intervention group

In this, the mother carried the infant accompanied by a porter and a nurse from the ward to theatre. The mother and the infant were received by one of the theatre staff in the waiting room. Both the mother and the infant together with the theatre staff went to the operation room. The mother remained in the operation room with the infant until the infant was anaesthetized. After this, the mother was accompanied out of the operating room but she remained in the theater suite in the waiting room. The infant was then intubated and the surgery proceeded. After the operation the infant was extubated and taken to the recovery room. Immediately the infant reached the recovery room, the mother was invited in the recovery room.

The initial research plan was to call the mother back into the operating room before the infant was fully awake. This, we thought, could have been crucial to the findings in the study as it might have given an indication on the stress status of the infant when the mother was present. This did not work out in the practice setting as most infants woke up immediately after extubation.

The mothers in the intervention group were re-united with the infants in the recovery room. Some mothers had a chance to reunite and breastfeed their infants in the recovery room, while others infants that were fully awake and stable in the operating room did not stay in the recovery room long enough. They were quickly taken back to the ward.

Control group

This group received the current standard of care for infants under six months old. The mother did not accompany the infant to theatre. She stayed on the ward and waited there until the infant was brought back after surgery. The infant was accompanied by a nurse, a porter and the researcher from the ward to theatre. The infant was received by one of the theatre staff in the waiting room as per the current practice. The infant was taken to the operation room. The infant was then intubated and the surgery proceeded. After the operation the infant was extubated and taken to the recovery room. After the infant was fully awake, the theatre sister called the ward staff to come and accompany the infant

back to the ward. The infant was accompanied by the ward staff, porter and researcher back to the ward where he or she was re-joined with the mother. This is the standard practice.

In the control group, if the infant was obviously in significant distress and crying inconsolably, the researcher noted this in the observational notes and consoled the baby by inserting the dummy in the mouth if the infant had one or by lifting up the baby. It would be unethical to let the infants cry inconsolably.

In both groups, the VU-ambulatory monitoring system was attached prior to the infant going to theatre. The monitor remained in situ during the operation, recovery and 2 hours after the infant was transferred back to the ward. The researcher accompanied the infant to continue the observations and monitoring throughout the whole process.

3.10 Measurement instrumentation

3.10.1Vrije Universiteit Ambulatory Monitoring System (VU-AMS)

The VU-AMS measures and records data recorded using an ambulatory ECG/ICG (Electro cardiograph/ Impedance cardiograph). Heart rate variability data for calculating HRV was recorded using an ambulatory Electro cardiograph data acquisition system. Heart rate variability has been validated as a measure of stress in infants (Morgan, Horn &Bergman, 2011).
The VU-AMS is customised to work with seven disposable ECG electrodes. Before the electrodes were attached, the infant's skin was cleaned with an alcohol swab at the seven positions where the electrodes where going to be attached. The metal stud was attached at the center of the electrode, and then the sticky plastic brim was attached to the skin firmly spread out by the contact gel. The VU-AMS monitoring system electrodes have different colours and functions in measuring the autonomic nervous system.
Below is what the VU-AMS electrodes were measuring on the infants ANS and their colours. (Adopted from VU-AMS Manual version 1.1, 2013).
The white, green and black electrodes measure the ECG

1 White electrode was placed slightly below the right collar bone 4 cm to the right of the sternum

2 Green electrode was placed on the right side, between the lower two ribs

3 The black electrode was placed at the apex of the heart on the left lateral margin of the chest approximately at the level of the processus xiphodius.

The two blue electrodes measure the ICG. They consist of two wires of which one is short and the other one is long. The short wire is labelled number 4 and the long wire is number 5.

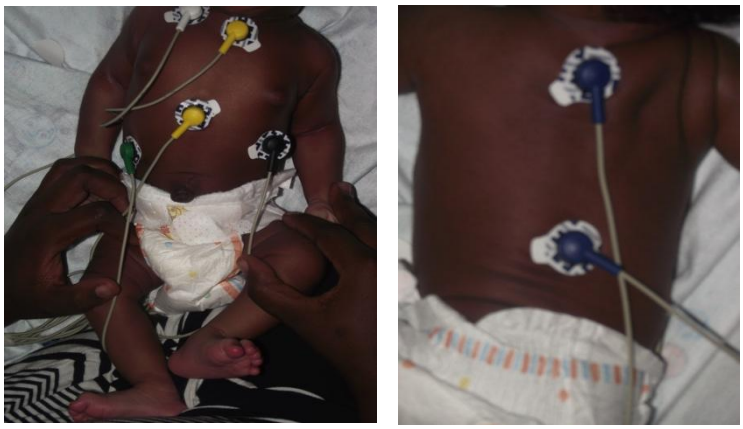
4 The blue short wire was placed at the back, on the spine, at least 3 cm (1") above electrode 6

5 The blue long wire was placed at the back, on the spine, at least 3 cm (1") below electrode 7

6 The short yellow wire was placed at the suprasternal notch above the top of the sternum

7 The long yellow wire was placed at the processus xiphodius at the bottom of the sternum

Figure 2: Position of VU−AMS electrodes on an infant



3.10.2Observational tools

An observational tool was designed and used by the researcher to document what happened according to the flow or the events from the time the infant was admitted until after surgery. The observational tool consisted of biographical data and research timeline events.

Biographical data

On admission, the following was documented; the date when the research will take place, research number, informed consent given, date of birth, weeks at birth, current age in months, weight at birth, current weight, sex, date of admission, antenatal history, postnatal history, onset of problem.

On the day of the surgery, the following was documented; time when nil per mouth commenced, type of feed that was last given and the person who gave it, any medication given on the ward and theatre and the person who gave it.

Research timeline events

On the day of surgery the following times were documented. The time the researcher arrived to meet patient, the time the patient got connected to the VU-AMS, the start of the recording, the time the mother put the infant down on trolley in the control group or carried the infant in the intervention group, the time the patient left the ward, the time the patient arrived in theatre, the time the mother put the infant down in the intervention group, the time anaesthesia was started, time of intubation, time surgery started and when it ended, time of extubation, time mother arrived in recovery room to meet infant (for those in the intervention group), time mother and infant left theatre and when they arrived back to the ward, the time the infant met the mother (for those in the control group), time of attachment of monitors on the ward and 30, 60,90 and 120 minutes later after surgery. The time of commencement of feeds and time when recording was stopped were also noted. See appendix IV

Pain tool

The Neonatal Infant Pain Scale (NIPS) was used as an additional observational tool to assess infant state, comfort and pain. (Lawrence et al., 1993).It was validated by Hudson-Barr et al., 2002 and found to be an appropriate pain-tool for infant of this age group. The researcher scored the infants and the mother was also asked if the infant was in pain. There was correlation between the researcher and the mother's score. See appendix IV.

Table 2: Illustration of the procedure

Control Group – (Standard of care)			***Observation period and location***	**Experiment Group**		
Infant	Mother	Researcher's role observing and monitoring the infant:	**Monitoring period**	Infant	Mother	Researcher's role observing and monitoring the infant:
VU-AMS monitor attached on the body	Present	Monitor - Heart rate Respiration Temperature, pain score Observe and track	On the ward prior to theatre	VU-AMS monitor attached on the body	Present	Monitor - Heart rate Respiration Temperature, pain score, Observe and track all activities

		all activities				
VU-AMS monitor attached on the body	Absent	Observe and track all activities	Trolley transfer to theatre	VU-AMS monitor attached on the body	Present	Observe and track all activities
VU-AMS monitor attached on the body	Absent	Observe and track all activities	Theatre reception	VU-AMS monitor attached on the body	Present	Observe and track all activities
VU-AMS monitor attached on the body	Absent	Observe and track all activities	Operating theatre	VU-AMS monitor attached on the body	Present	Observe and track all activities
VU-AMS monitor attached on the body	Absent	Observe and track all activities	During the procedure	VU-AMS monitor attached on the body	Absent	Observe and track all activities
VU-AMS monitor attached on the body	Absent	Observe and track all activities	Recovery room	VU-AMS monitor attached on the body	Present	Observe and track all activities
VU-AMS monitor attached on the body	Absent	Observe and track all activities	Trolley transfer to the ward	VU-AMS monitor attached on the body	Present	Observe and track all activities
VU-AMS monitor attached on the body	Present	Heart rate Respiration Temperature Pain score Observe and track all activities	Ward	VU-AMS monitor attached on the body	Present	Heart rate Respiration Temperature pain score Observe and track all activities
VU-AMS monitor attached on the body	Present	Heart rate Respiration Temperature Observe and track all activities	2 hour post-surgery on the ward	VU-AMS monitor attached on the body	Present	Heart rate Respiration Temperature Observe and track all activities

3.11 Reliability and validity

Reliability refers to the consistency and dependability of a research instrument to measure a variable, including stability, equivalence and internal consistency (Brink, 2002).Validity refers to the ability of an instrument to measure the variable that it is intended to measure (Brink, 2002). Studies by Acharya et al., 2006, Von Borell et al, 2007 have proven heart rate variability to be a valid and reliable marker for the effects of the autonomic nervous system on the heart rate.

The Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology (1996) standardised the frequency bands for analysing HRV. The High Frequency band is related to breathing rate and is a measure of parasympathetic activity while low frequency is regarded as a mixture of sympathetic and parasympathetic activities. These bands are valid markers of the HRV.

3.12 Ethical Considerations

Ethical considerations were an important part of the study because the research involved infants who were patients, are minors and comprise a particularly vulnerable population.

Ethical considerations included having to ensure that the study caused no harm and did not place participants at risk in any way. It also had to comply with the latest Helsinki Convention (World Medical Association Declaration of Helsinki Ethical Principles for Medical Research Involving Human Subjects, 2013). The study also involved these infants' mothers. In order to ensure that the study was ethically compliant, approval was sought from the Research Ethics Committee of the University of Cape Town (UCT) as well as Red Cross War Memorial Children's Hospital. Below are the ethical considerations.

3.12.1 Autonomy

The nature, risks and benefits of the study were explained to the legal guardian and was given a chance to ask questions. This principle recognises the rights of individuals to self-determination and to make informed decisions (Gillon, 1994).The researcher respected each legal guardian as a person capable of making an informed decision regarding participation in the research study.

3.12.2 Informed consent

The principal researcher recruited the infants into the study. Verbal consent was initially sought from recruited participants .The infants' mothers and legal guardians were fully informed about the nature of the study and what would be required (Appendix ii). Thereafter they were given an opportunity to make a decision about participation in the study. It was always reassured that their infant's care was not going to be compromised in any way if they decided not to participate in the study. They were also informed that they could withdraw from the study at any time without being required to give a reason and with no repercussions in terms of the kind of care their babies would

receive. The legal guardians were also provided the opportunity to ask any questions or raise any concerns. These were responded to by the primary researcher.
One copy of the informed consent was provided to the parents, one copy was placed in the infant's medical record and the original was retained in the study file. See appendix iii

3.12.3 Beneficence and Non-Maleficence

Beneficence emphasizes that the researcher to do good. Non-maleficence refers to the obligation of the researcher not to cause harm (Brink 2002).In this study the researcher attempted to maximize benefits for the individual participants while minimizing risk of harm to the participants by gathering data via a set of non-invasive skin electrodes. There were two blue electrodes at the back. Padded pieces of gauze were placed at the back to prevent friction between the operation bed and the electrodes. They were also carefully placed above the naval area to prevent any interference with the surgical procedure. The continuous presence of the researcher ensured safety while also providing comfort to infants in the control group.

3.12.4 Confidentiality

The right to privacy of the participants was ensured through maintaining anonymity and confidentiality. Anonymity is understood as concealing the identities of participants in all documents resulting from the research, while confidentiality entails ensuring that unauthorized people (as per agreement with participants) have no access to the information provided by the participants (King & Horrocks, 2010). Anonymity was maintained by using a coding system instead of actual names for all the participants. Confidentiality was achieved by restricting access to raw data and materials to only the researcher. Both of these principles are retained throughout this report and in subsequent presentation of data and study outcomes.

3.12.5 Justice

The principle of justice includes the right to fair selection and treatment and the right to privacy and confidentiality (Brink, 2002).Inclusion and exclusion criteria ensured that all infants were equally considered and all that met the inclusion criteria were accepted into the study. The randomisation process was carefully done and maintained. The legal guardians participating were given the opportunity to open the randomised sealed envelope to ensure transparency and fairness to the participants.

3.13 Implication of the Research

A detailed report on the research findings will be provided to the departments of nursing, surgery and anaesthesia at Red Cross War Memorial Children's Hospital. The results of the study will serve to motivate for practice changes and policies in hospitals for children if the hypothesis is confirmed. The results will be presented at a number of clinical settings and academic meetings as deemed relevant.

3.14 Pilot study

A pilot study was carried out on a small population to identify potential practical problems and test the research instrument in the research procedure (de Vos, 2005).

The pilot study was carried out on two patients, one in the control group and the other in the intervention group.

The observational tool which was designed to track changes was modified according to the activities that were noted to happen around the infant. The researcher realised that it was difficult to record regular vital signs between the times when the infant left the ward to the time she/he returned to the ward after surgery. This is because she did not have direct access to the infant and she had to keep from interfering with the surgical procedure.

There were initial challenges using the VU-AMS ambulatory recording system machine. The electrodes were often dislodged when the infant was moved.

The researcher also discovered that she had to be hyper vigilant at all times to ensure that the VU-AMS monitor was tracing and at the same time monitor and record what was happening around the infant. The pilot study helped to prepare for this complexity, to identify and control for errors such as those due to the instrument. These were reduced as much as possible. This helped to make the data accurate and meaningful (de Vos, 2005).

Data from one dyad was good and it was decided to use it in the analysis along with the study cases. The second set of data was discarded because of a recording error. It was later discovered that the machine had accidentally stopped during the time the infant was being transferred.

3.15 Data collection

Ethical approval had been obtained from the Human Research Ethics Committee of the Faculty of Health Sciences at the University of Cape Town and from the Hospital Ethics Committee. As soon as this was granted, the nursing managers and staff in the surgical clinic and the ward where these

patients were seen and admitted were contacted in person. The researcher discussed with them the study protocol and reassured them that she would do her best to prevent any interference in the normal ward routine so that the research would have minimal effect on the care of the infants. The researcher also spoke to the head of the anaesthesia team who was also in agreement with the method of data collection.

Data was collected using the VU-AMS ambulatory ECG/ICG. It has seven electrodes which were attached on the infant's body. (http://www.vu-ams.nl/vu-ams/) The machine has software that needs to be downloaded on the computer before it is able to do any recording.

The machine was attached in the morning before the infant went for surgery. This stayed on the infant throughout surgery. It was removed after two hours of recording after the patient had returned to the ward after the operation.

3.16 Data storage management

The VU-AMS ambulatory monitoring system has a memory device where all the data is recorded during the monitoring process. A card reader was used to transfer data onto the researcher's computer and saved by the AMS Electro Cardiograph data acquisition software. Offline computation of RMSSD (root mean square of successive differences), high frequency, low frequency and very low frequency was done. Standard deviation of normal R-R intervals (SDNN, standard deviation of all heart periods over one minute) was computed. According to previous research, this data was found to correlate well with overall autonomic tone and can be used to check the integrity of the data (Morgan, 2011). Data was then transferred to the statician via an external hard drive where it was analysed.

3.17 Statistical Analysis

Due to the small sample size, non-parametric values were used to calculate the sample size using the two-sample Wilcoxon rank-sum (Mann-Whitney) test. They were plotted as p50 or medians and put into a STATA computer programme which produced the p values that were reported in the results. Graphs were plotted for easy presentation and the statistics were reflected in the median because of the non-parametric values.

3.18 Drawing of polygraphs

For each case, the observations were plotted on the polygraphs for visualization and to guide interpretation and subsequent statistical analysis. This included graphically representing the different periods of epochs described in figure 1 with observed significant events. Key outcomes were plotted in separate graphs with graphic time alignment.

3.19 Conclusion

This chapter discussed the methodology and the design used to collect the data. The instruments of collecting and analyzing data were also analysed. The ethical issues were discussed and every step of the way the mother was explained what was happening around the child.

CHAPTER 4: RESULTS

This chapter describes the results of this study and is organised to ensure that the results are aligned with the intended study design as described in the previous chapter.

4.1 Trial profile

40 infants were assessed for eligibility and 30 recruited into the study. The full trial profile is illustrated below in diagram 1.

4.1.1 A total of 40 infants were assessed for eligibility to participate in the study

4.1.2 10 of these infants were not eligible to participate in the study. Of these, 4 infants were excluded because they were accompanied by caregivers who were not their legal guardians and so legal consent could not be obtained. 2 infants did not meet the inclusion criteria because they had co-existing conditions and 4 legal guardians declined to participate. They were not required to provide a reason for their decision.

4.1.3. There were 30 infants who were eligible and they consented to participate in the study. 15 were randomized to the control group (standard of practice) and 15 to the intervention group.

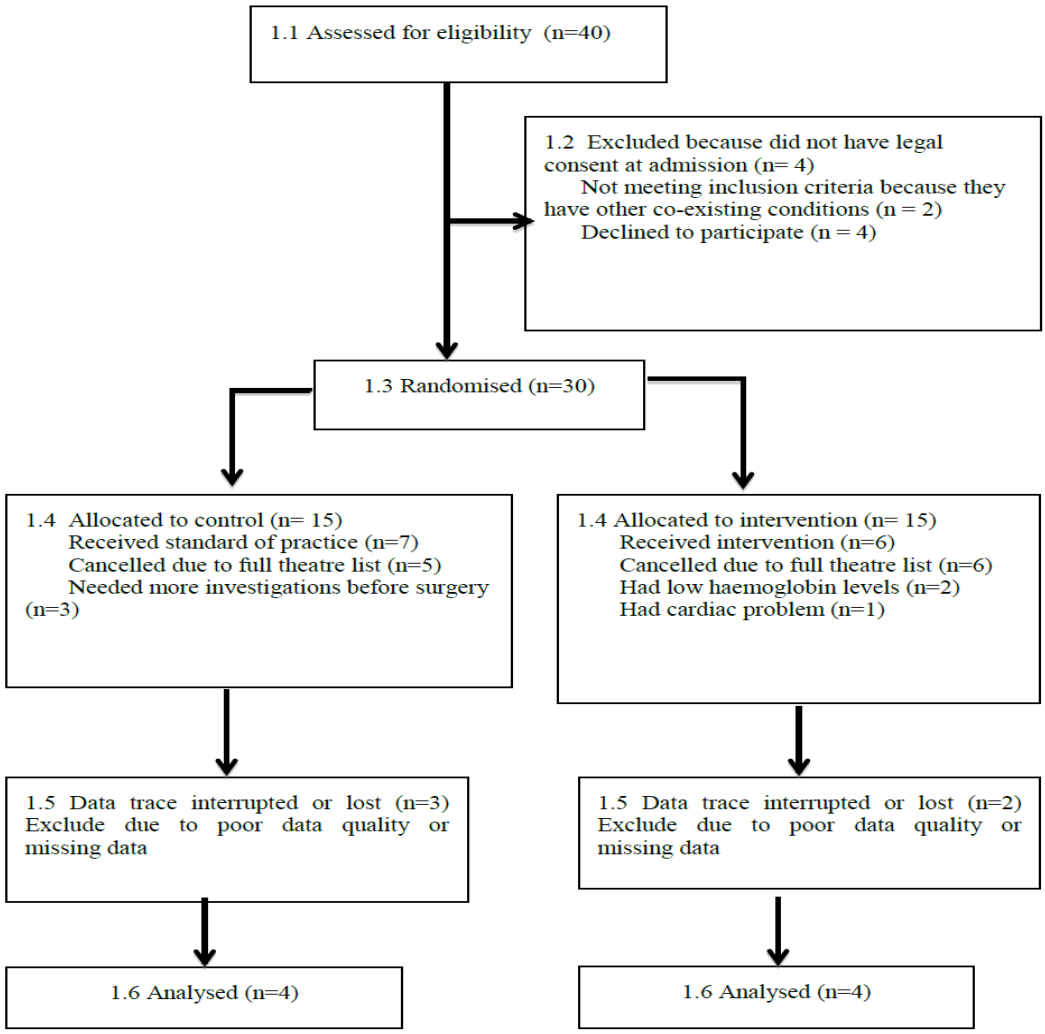
4.1.4 In the control group, 7 received the standard of practice. 5 infants were cancelled citing a full theatre list from the doctors and theatre staff despite the fact that they had been kept nil per mouth in preparation for the surgery. 3 were cancelled because the doctors made a decision that they had to do more investigations before they could proceed with the surgery.

In the intervention group, there were 15 infants. 6 infants received the intervention. 6 infants were cancelled due to a full theatre list despite being kept nil per mouth in preparation for the surgery. 2 patients went to the operation theatre, but surgery was not started due to low haemoglobin levels. Another infant had the surgery cancelled after going into the operating theatre because the cardiac surgeons had not assessed him prior to the surgery.

4.1.5 In the control group, 3 infants were not analysed due to poor data quality.
In the intervention group, 2 were not analysed due to missing and poor quality data.

4.1.6 The total number of patients who had their data analysed was 8 out of the consented 30 infants. In the control group, 4 infants' data and in the intervention group, 4 infants' data was analysed.

Figure 3 TRIAL PROFILE



Format adopted from CONSORT 2010 statement (Schulz, Altman & Moher, 2010).

4.17 Baseline characteristics of the study subjects: Biographical and Clinical data for the 8 infants

Data from 8 infants was analysed; 4 in the control group and 4 in the intervention group. 7 of these infants were male and one was female. The mean weight was 3618g and the range was between 2800g and 5500g. The average age was 10 weeks with the range between 5weeks and 17 weeks. All the infants were scheduled for and had an inguinal hernia repair.

Table 3: Biographical and Clinical data for the 8 infants

		Mass		Age				
		Birth(g)	Current	Birth Gestation	Corrected age	Current		

Case	Infant's sex		(g)	(weeks)	(weeks)	(weeks)	Diagnosis	Group
1	M	2900	3200	35	46	11	Inguinal Hernia	Intervention
2	F	1200	2800	28	41	13	Inguinal Hernia	Intervention
3	M	3500	5500	42	55	13	Inguinal Hernia	Intervention
5	M	3000	3320	40	48	8	Inguinal Hernia	Control
6	M	970g	2900	26	43	17	Inguinal Hernia	Control
7	M	3200	4000	38	46	8	Inguinal Hernia	Intervention
8	M	2000	2700	35	40	5	Inguinal Hernia	Control
9	M	3200	4520	40	46	6	Inguinal Hernia	Control
Mean	-	2496	3618	35.5	45.6	10	-	-
SD	-	976.20	985.06	5.81	4.69	4.09	-	-

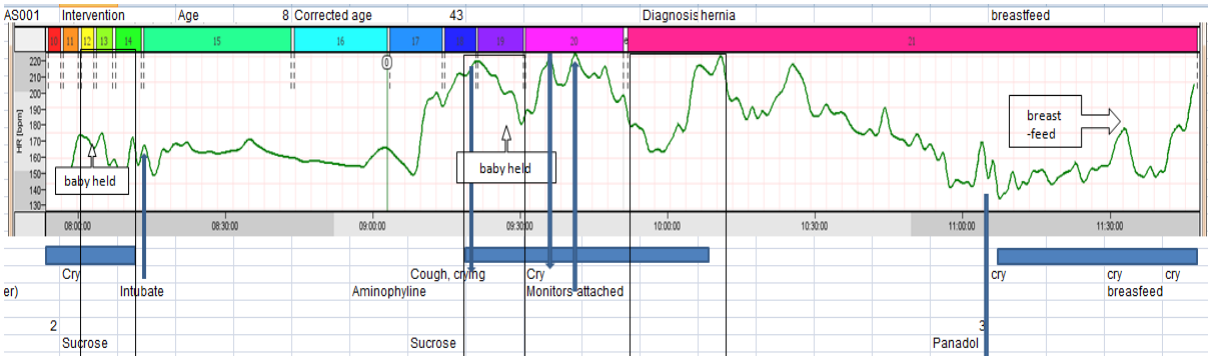
4.2 Case reports of individual infant data sets

Each infant was fully monitored using the VU-AMS monitoring equipment. These data were aligned with the detailed notes made on events throughout the observation period while outcome variables were being recorded. These events were noted according to predefined epoch periods and tracked in assigned colours. The outcome variables for each case report are drawn as polygraphs with the epoch and the events with the heart rate extracted from the VU-AMS software.

Each selected outcome variable is provided as follows:

4.2.1 Top window

Figure 3:Example of polygraph of data set.



The top window is from the VU-AMS software and it is a jpg screen shot. The observational data is seen in the lower half as an annotation of observed events. Important to note is the horizontal blue bar when the mother is present. There are key events that took place and these were also tracked on in the lower window. The annotation of observed events of importance included administered medications, sucrose for pain relief and feeding.

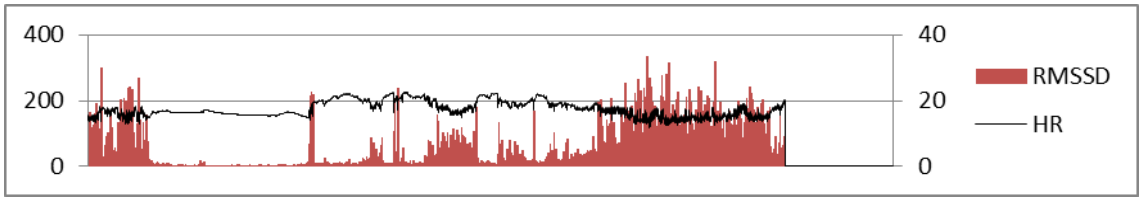
The top window has different colours which represent different events from the time when the infants were on the ward, through to theatre, recovery room until they were brought back to the ward.

Table 4:Different colours and what they represent in the top window

Colours	Events
10	Waiting for trolley on the ward
11	Waiting on the trolley on the ward
12	Moving to theatre
13	In the waiting room
14	In theatre until being anaesthetized
15	Being anaesthetized
16	Being operated
17	Waking up in theatre
18	Recovery room
19	Moving to ward
20	Settling
21	Resting

4.2.2 Root mean square of successive differences and Heart rate

Figure 4:Root mean square of successive differences and Heart rate



RMSSD is the measure of how much effort is being exerted by the cardiac muscle. This was correlated with the heart rate which is the number of heartbeats per unit of time, expressed as beats per minute.

4.2.3 Standardised Low frequency (LF),High frequency (HF) and very low frequency (VLF)

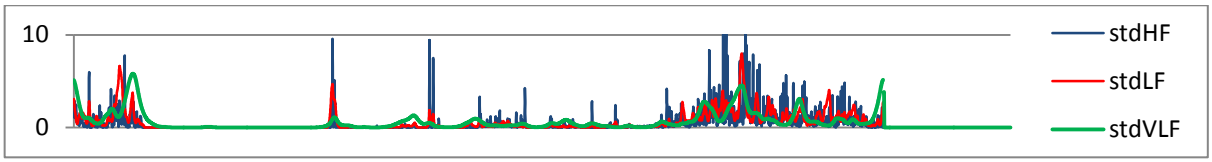


Figure 5:Standardised Low frequency,High frequency and very low frequency

The standardized frequency is regarded as a combination of sympathetic and parasympathetic tone. HF is related to the breathing rate and is well validated as a measure of parasympathetic (social vagal tone).

4.2.4 Heather index, Low frequency (LF) andHigh frequency (HF) ratio

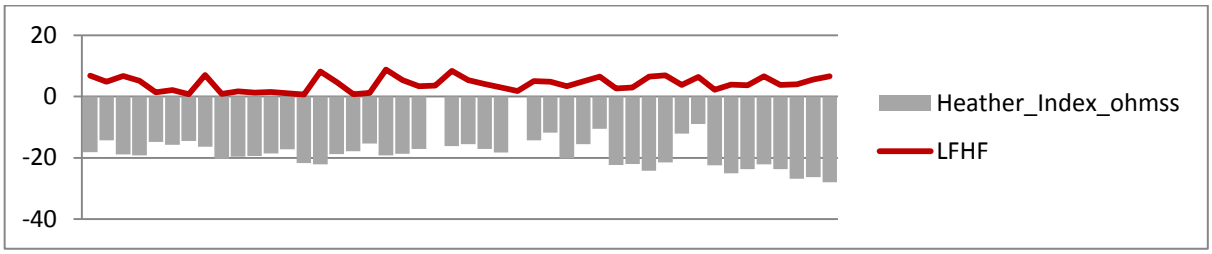


Figure 6: Heather index, Low frequency and High frequency ratio

Heather index is an alternative contractility measure in ohm/sec^2 and has been shown to be especially sensitive to changes in cardiac contractility.

Low frequency (LF) and High frequency (HF) are frequency bands.

LF is regarded as a mixture of sympathetic and parasympathetic tone.

HF is related to the breathing rate and is well validated as a measure of parasympathetic (social vagal) tone.

4.2.5 Stroke volume and minute volume

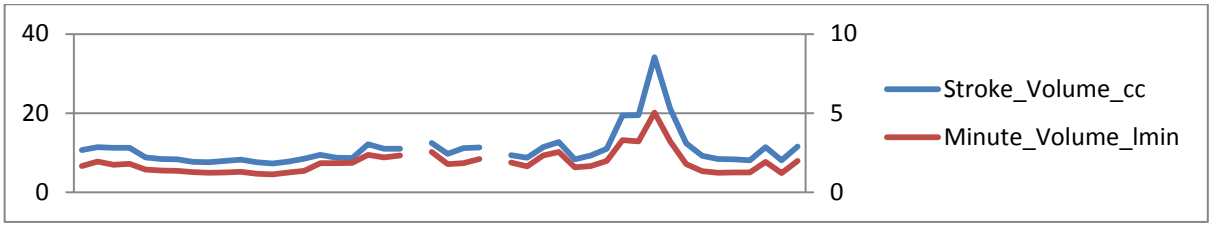


Figure 7: Stroke volume and minute volume

Stroke volume (SV) is the average amount of blood ejected during the cardiac cycle.

Minute volume (MV) is the total amount of blood circulated through the body per minute. The events and behaviours noted in observations were matched to specific outcome variables as results for each case report.

4.2.6 Brief descriptions of each infant data set

Each infant's data trace was aligned with a set of observational data. The setting and sequence of events were well prepared and the procedure considered the most 'routine' and probably simplest of surgical interventions. Each infant's data set included a variety of events, predicted and unpredicted that added complexity to the results.

4.2.6.1 Control group

The control group consisted of data sets AS005; AS006, AS008, AS009. Each is briefly described below.

Data period: 12hrs 17 minutes

Table 5: First Infant: AS005

Infant's sex	Mass		Age			Diagnosis
	Birth(g)	Current (g)	Birth Gestation (weeks)	Corrected age (weeks)	Current (weeks)	
M	3000	3320	40	48	8	Inguinal Hernia

Time	Event	Additional

		comment
03h00	Infant breastfed by mother then nil per mouth commenced. Mother and infant were sleeping	
	VUAMS monitor attached -	
10h57	Infant left the ward with the porter and theatre staff	Infant was nil per mouth for 8 hours
11h04	Infant was intubated and caudal block given	
11h15	Surgery started	
12h25	Surgery completed	
12h28	Infant extubated	Arrived in the recovery room, awake and crying
12h30	Infant taken to recovery room - infant visibly agitated and crying	
12h54	Infant back on the ward and reunited with the mother	
13h00	Infant started breastfeeding	
13h30	Infant crying very loudly, mother struggling to console	
14h00	Sleeping	
14h18	Coughing while sleeping	
14h38	Woke up and started crying	
15h00	Crying. Mother comforting infant	
15h16	Crying	
15h17	Stopped recording	

Summary: The RMSSD and the LFHF increased when the infant was being transferred to the ward which is a sign of stress. Despite the fact that the infant was crying at this time, the heart rate was not high. Heart rate alone may not be a good measure of stress.

Case no 6 CONTROL GROUP

Table 6:Second Infant: AS006

Infant's sex	Mass		Age			Diagnosis
	Birth(g)	Current (g)	Birth Gestation (weeks)	Corrected age (weeks)	Current (weeks)	
M	970g	2900	26	43	17	Inguinal Hernia

Time	Event	Comment
06h00	Infant breastfed by mother and nil per mouth started	Mother was very agitated pacing up and down in the corridor while carrying the infant
09h00	Infant irritable and crying. A nurse or doctor dummy with sucrose was inserted in the mouth	
11h00	Infant and the mother very agitated. Infant was crying	
11h30	Infant fell asleep and woke up just after 10 minutes. Started crying again. Theatre staff were contacted and they said they will collect infant within 30 minutes time	Mother was very agitated pacing up and down in the corridor while carrying the infant
12h00	The VU-AMS machine was connected earlier after theatre confirmed to say that they were fetching the patient	
12h05	Theatre staff arrive with the trolley to fetch the patient	
12h08	Patient left the ward with the theatre staff	
12h11	Patient arrives the waiting room Cries, dummy in the mouth, researcher lift infant due to over crying, dummy in the mouth with sucrose	Infant was made to wait in the waiting room for 35 minutes
12h48	Leave the waiting room and arrives in theatre	
12h59	Infant is intubated	Nil by mouth for 7 hours – no fluids in that time
13h22	Caudal block given	
13h33	Surgery starts	Heather index visibly higher indicating significant stress While mother left the bed side and infant's heart rate increased though he was sleeping
13h51	Surgery stopped	
13h54	Extubated, coughing and crying	

13h59	Infant is in recovery room, crying dummy was put in the mouth with sucrose.	
14h41	Infant transferred to the ward and meets mother	
14h52	Started breastfeeding	
15h00	Infant slept	
15h20	Mother left the bed side and came back at 16h00	
16h45	Stopped recording	

Summary: The Heather index was high at one point during the time when the infant was being anaesthetised and the minute volume rose during the time the infant was waking up. These are sympathetic measures that indicate stress. The RMSSD and standard HF were raised during the time of resting when the mother was away even though the heart rate was low and the infant

Case no 8 CONTROL GROUP

Data period: Table 4.3

Table 7:Third Infant: AS008

Infant's sex	Mass		Age			Diagnosis
	Birth(g)	Current (g)	Birth Gestation (weeks)	Corrected age (weeks)	Current (weeks)	
M	2000	2700	35	40	5	Inguinal Hernia

Time	Event	Comments
02h00	Nil per mouth commenced	
07h54	Infant left the ward	
07h57	Arrives in waiting room while crying	Nil per mouth for 6 hours
08h00	Is given a dummy with sucrose and is still crying	
08h15	In theatre and is intubated	
08h29	Caudal block given	
08h47	Surgery starts	
09h19	Surgery ends	
09h24	Is extubated	Infant had a high VLF and high SV/MV when waking up.
09h30	Transferred to recovery room is awake and crying	

09h33-09h44	Is crying continuously, dummy with sucrose is inserted in the mouth	
10h00	Transferred to the ward still crying	
10h05	Returns on the ward and is still crying despite of give the dummy with sucrose. Mother was not present on the ward	
10h25	Dummy with sucrose given by ward staff	
10h30	Infant crying	
11h00	Mother walks in and comforts the infant	
11h10	Mother carries infant who is still crying	
11h30	Infant keeps quiet while being held by mother. She is singing a lullaby	
12h00	Stopped recording	

Summary: The very low frequency was high during anaesthesia and the operation. Stroke and minute volume were high in theatre when the infant was waking up. These are sympathetic measures and are an indication of stress.

Case no 9 CONTROL GROUP

Data period: Table 4.4

Table 8:Forth Infant: AS009

Infant's sex	Mass		Age			Diagnosis
	Birth(g)	Current (g)	Birth Gestation (weeks)	Corrected age (weeks)	Current (weeks)	
M	3200	4520	40	46	6	Inguinal Hernia

Time	Event	Comment
02h00	Infant breastfed by mother and nil per mouth started	
08h20	Infant quiet, dummy in the mouth with sucrose	
08h57	Mother is agitated and crying because infant is crying	
09h12	Doctor puts up IV line on the infant	

09h21	IV fluids commenced	NPO for 13 hours Iv commenced 7 hours after last feed. Had a high heart rate during operation compared with HR before? Clarify
09h45	Infant is sleeping	
13h36	Infants leaves ward to go theatre, crying	
13h41	Infant in the waiting room and crying	
13h48	Infant is in theatre, crying	
13h59	Infant is intubated	
14h12	Caudal block given	
14h22	Surgery starts	
14h45	Surgery stops	
14h53	Is extubated	
14h56	Transferred to recovery room	
15h21	On the ward and reunites with mother, crying	
15h25	Infant crying, dummy in the mouth with sucrose	
15h30	Starts breastfeeding	
15h45	Infant is crying	
16h00	Mother picks infant and carries her	
16h30	infant is crying	
16h55	Mother comforts the infant who is still crying	
17h20	Infant sleeping	
17h25	Stopped recording	

Summary: This infant was starved for 13 hours and a lower heart rate but when he was separated from the mother, even during anaesthesia and surgery, the heart rate was raised. Infant had a normalised very low frequency during surgery which is a stress indicator.

4.2.6.2. Intervention group

The intervention group consisted of data sets AS001; AS002; AS003; AS005. Each is briefly described below

Case no 1 INTERVENTION GROUP

Data period: Table 4.5

Table 9:First Infant: AS001

Infant's sex	Mass		Age			Diagnosis
	Birth(g)	Current (g)	Birth Gestation (weeks)	Corrected age (weeks)	Current (weeks)	
M	2900	3200	35	46	11	Inguinal Hernia

Time	Event	Comment
04h00	Infant breastfed by mother and nil per mouth started	
08h00	Mother leaves the ward carrying the infant to theatre.	
08h07	Mother and infant arrive in the operating room	
08h10	Staff takes out dummy from the infant, he cries	
08h11	Mother puts dummy back in the mouth	
08h14	Mother leaves operation room after infant is anaesthetised	
08h17	Intubation	Nil per mouth for 4 hours
08h28	Caudal block given	
08h43	Surgery starts	
09h00	Surgery stops	
09h08	Infant is extubated, moving hand and crying	
09h14	Infant is in recovery room	
09h19	Mother arrives in the recovery room	

09h20	Mother and infant leave the recovery room to go to the ward	
09h23	Mother puts dummy in the infant's mouth	
09h30	Arrive on the ward crying	Could not breastfeed immediately. Did not stay for long in the recovery room
09h35	Infant crying	
09h45	Started feeding on half dextrose water	
10h00	Infant is quiet but not asleep	
10h12	Mother leaves bedside and still waiting for nurses to tell her when to feed the baby.	
10h40	Infant moving arms and legs	
10h57	Mother comes back	Mother leaving bedside caused baby's heart rate to go up response from the infant
11h00	Infant woke up	
11h07	Started crying	
11h30	Started breastfeeding	
11h45	Stopped recording	

Summary: In this intervention case, the baby was quiet and calm when the mother was present but her absence caused a very low frequency effect and later was replaced by high frequency response which eventually came down when she returned but it took a long time. The separation caused an autonomic response which looked like stress.

Case no2 INTERVENTION GROUP

Data period: Table 4.6

Table 10:Second Infant: AS002

Infant's sex	Mass		Age			Diagnosis
	Birth(g)	Current (g)	Birth Gestation (weeks)	Corrected age (weeks)	Current (weeks)	
F	1200	2800	28	41	13	Inguinal Hernia

Time	Event	Comment
04h00	Infant breastfed by mother and nil per mouth started	
07h46	Mother lifts infant off the bed	
07h54	Mother arrives in theatre	
07h58	Mother sits down, infant is anaesthetised while mother holding infant	
07h59	Mother leaves theatre	
08h06	Intubation	for 4 hours
08h08	Caudal block is given	
08h39	Surgery starts	Infant had a high stress response during surgery.
09h15	Surgery stops	
09h16	Extubation, infant coughing	
9h25	In recovery room, crying	
09h32	Mother enters recovery room and she starts breastfeeding	
09h37	Infant crying, refuses to take the breast	
09h44	Infant and mother leave the recovery room back on the ward	
09h47	Staff pricks infant's foot to take off blood for haemoglobin testing and infant screams.	
10h08	Infant cries continuously, mother holds the infant	
10h13	Mother starts to breast feed	
10h17	Infant is sleeping while breastfeeding	
10h47	Mother leave the bedside	

10h51	Infant wakes up crying	
10h55	Infant is crying and dummy is inserted in the mouth by mother	
11h02	Mother walks out and infant is sleeping	
11h32	Mother comes back at the bed side	
11h42	Infants wakes up crying and mother breast feeds the baby	
11h50	Stopped recording	

Summary: This infant had high normalised HF during surgery and low normalised low frequency. This is a mixed

Case no3 INTERVENTION GROUP

Data period: Table 4.7

Table 11: Third Infant: AS003

Infant's sex	Mass		Age			Diagnosis
	Birth(g)	Current (g)	Birth Gestation (weeks)	Corrected age (weeks)	Current (weeks)	
M	3500	5500	42	55	13	Inguinal Hernia

Time	Event	Comment
02h00	Infant breastfed by mother and nil per mouth started. Then was given half dextrose water at 11h00	
12h08	Mother let the ward carrying the infant	
12h15	Mother arrives in theatre	
12h23	Mother holds infant while is being anesthetised and she leaves theatre	
12h32	Intubation commences	
12h40	Caudal block is given	
13h00	Surgery starts	Nil per mouth for 10 hours
13h48	Surgery stops	
13h53	Extubation, coughing	
13h56	In recovery room	
14h03	Mother walks in the recovery room infant crying, dummy in the mouth	
14h05	Mother leaves recovery room in to waiting room to wait for ward staff to fetch her	
14h13	Infant is crying, mother picks up the infant from the trolley in the ward	Mother was told not to breastfeed immediately by a nurse as infant would vomit
14h20	Infant crying nurse, dummy inserted in the mouth	
14h28	Mother given half dextrose water to give infant by	

	a nurse	Infant had increased normalised high frequency
14h32	Infant quiet and sleeping	
15h40	Infant wakes up crying	
15h42	Mother gives more half dextrose water	
15h43	Refused dextrose water	
16h00	Sleeping	
16h18	Stopped recording	

Summary The normalised HF was high during anaesthesia and surgery and during the periods when the infant was sleeping during resting time. These are sympathetic measure indicating stress

Case no 7 INTERVENTION GROUP

Data period: Table 4.10

Table 12: Forth Infant: AS007

Infant's sex	Mass		Age			Diagnosis
	Birth(g)	Current (g)	Birth Gestation (weeks)	Corrected age (weeks)	Current (weeks)	
M	3200	4000	38	46	8	Inguinal Hernia

Time	Event	Comment
04h00	Infant breastfed by mother and nil per mouth started	
08h43	Mother carries infant off the bed and leaves for theatre	
08h49	Mother and infant are made to wait in the waiting room	
08h51	Infant is crying and the dummy is inserted in the mouth	
08h57	Mother is in operating room and carrying the infant	
09h00	Infant is anesthetised while on mother's lap	Nil per mouth for 5 hours
09h05	Mother leaves theatre	
09h08	Infant has IV inserted	
09h22	Surgery starts	
09h42	Surgery stops	Displayed stress responses during surgery.
09h45	Extubation, infant is moving hands and coughing	
09h49	In recovery room	
09h57	Mother walks in the recovery room	

10h05	Mother breastfeeds infant in recovery room	
10h25	Mother leaves recovery room while carrying infant	
10h27	Arrives on the ward and infant starts to cry	
10h32	Infant crying, mother breastfeeds	
10h48	Infant crying, mother breast feeds	
10h50	Mother leaves the bed side	
11h06	Infant asleep	
11h48	Infant vomiting	
12h07	Infant crying, dummy inserted in the mouth	
12h27	Mother walks in then walks out again	
12h29	Infant is sleeping	
12h34	Mother walks in	
12h46	Stopped recording	

Summary: The normalised VLF was high during surgery. When the infant was back on the ward, the mother left the ward and within that time the infant had episodes of increased heart rate, RMSSD and increased heather index but the infant was asleep.

4.3 Collated results of the heart rate variability and impedance cardiograph

Data were collated and then labelled into identifiable periods which were statically analysed.

Below are the labels that were made identifying carefully the periods that would be compared between the control and the intervention group for analysis. These periods were the same for each group.

Table 13: Periods for analysis

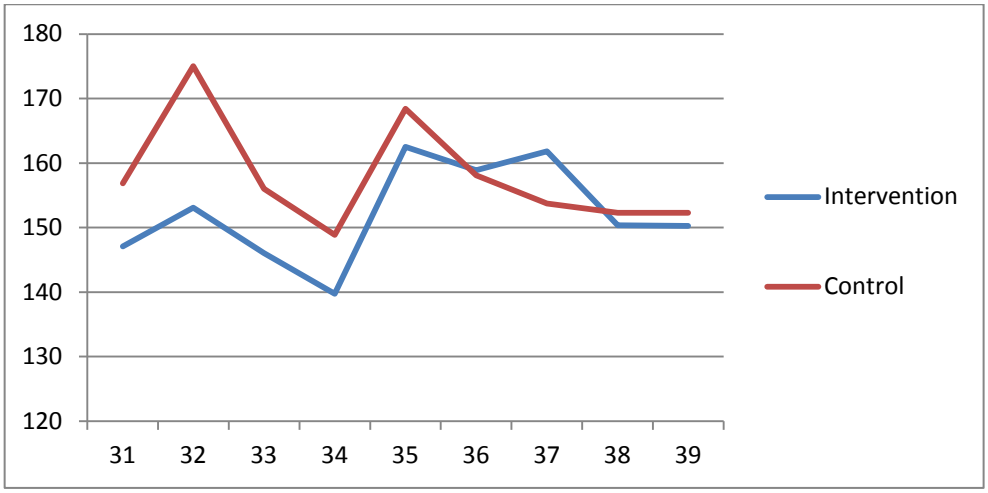
31	preparatory
32	preoperative
33	Anaesthesia
34	Surgery
35	Recovery
36	postoperative
37	Wards
38	Settling early 20 minutes
39	Settling later

Heart Rate Variability.

4.3.1 Heart rate (HR)

At point 32 when the babies were separated, the heart rate was higher in the control, (control vs intervention: the p50 is 166 vs 153, p 0.03) which is significant.

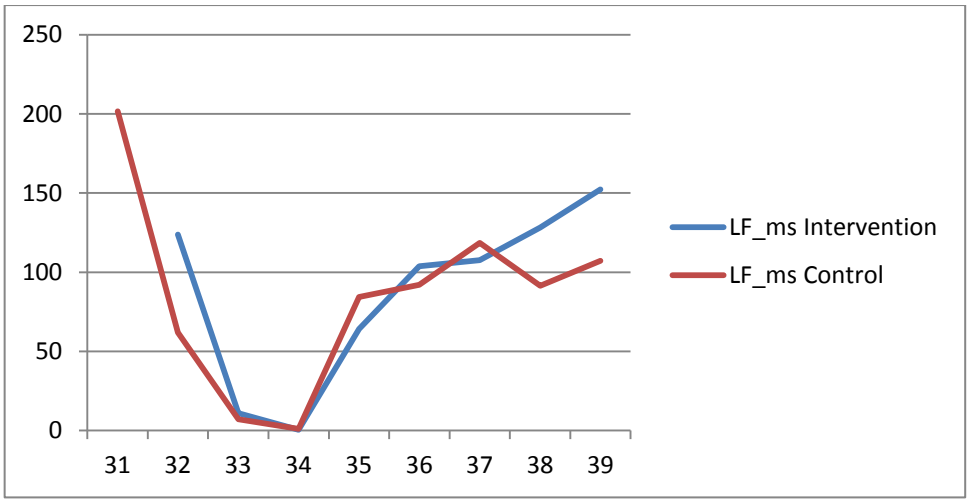
Figure 8: Heart rate



4.3.2 Low frequency

At point 32 when the babies were separated, the low frequency was significantly lower in the control, (control vs intervention: the p50 is 63 vs 111, p 0.034) which is significant. A higher low frequency is interpreted as a healthy and mature autonomic response.

Figure 9:Low frequency

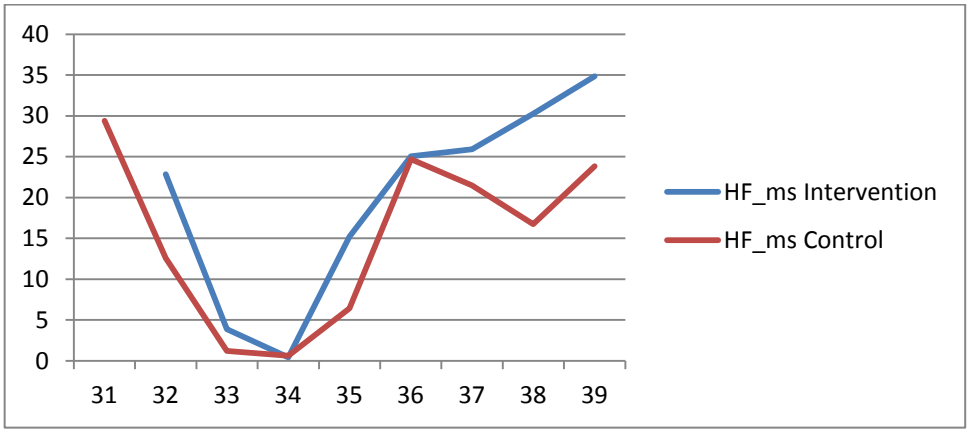


4.3.3 High frequency

The higher HF is interpreted as a healthy autonomic health response.

At point 32 when the babies are separated, the high frequency is significantly lower in the control group, (control vs intervention: the p50 is 14 vs 22.7, p 0.07) though it is not statistically significant. No other points are significant.

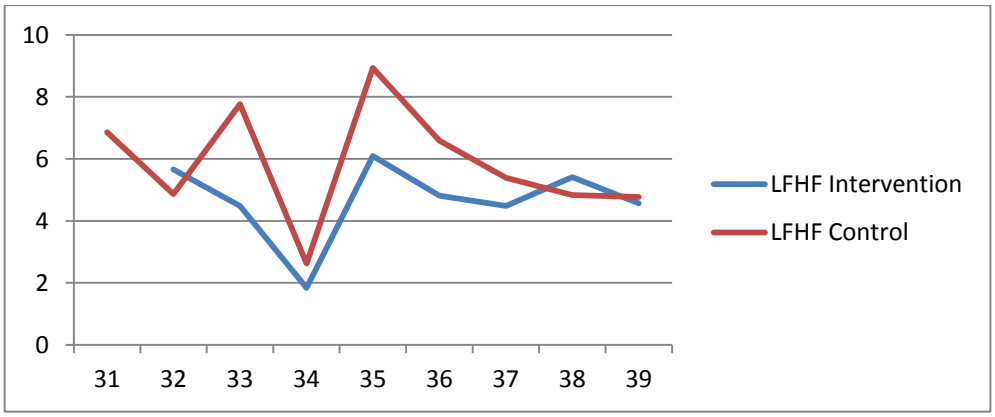
Figure 10:High frequency



4.3.4 Low frequency high frequency (LFHF)

The LFHF is the proportion of LF to HF and therefore serves as a proxy for sympathetic activity. The more sympathetic it is, the higher LF is compared to the HF and therefore, the higher the numbers. The control is more sympathetic especially during anaesthesia. At point 33 when the babies are separated, LFHF is higher in the intervention and lower in the control, (control vs intervention: the p50 is 9 vs 4, p 0.2) which is not statistically significant.

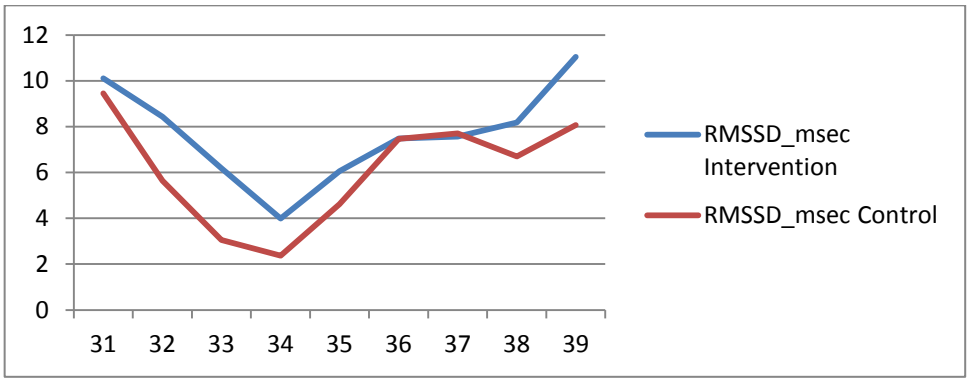
Figure 11:Low frequency high frequency



4.3.5Root mean square of successive heartbeat interval differences (RMSSD)

At point 33 when the babies are separated, the RMSSD is lower in the control, (control vs intervention: the p50 is 2.7 vs 5.4, p 0.14) which is not statistically significant.

Figure 12:Root mean square of successive heartbeat interval differences

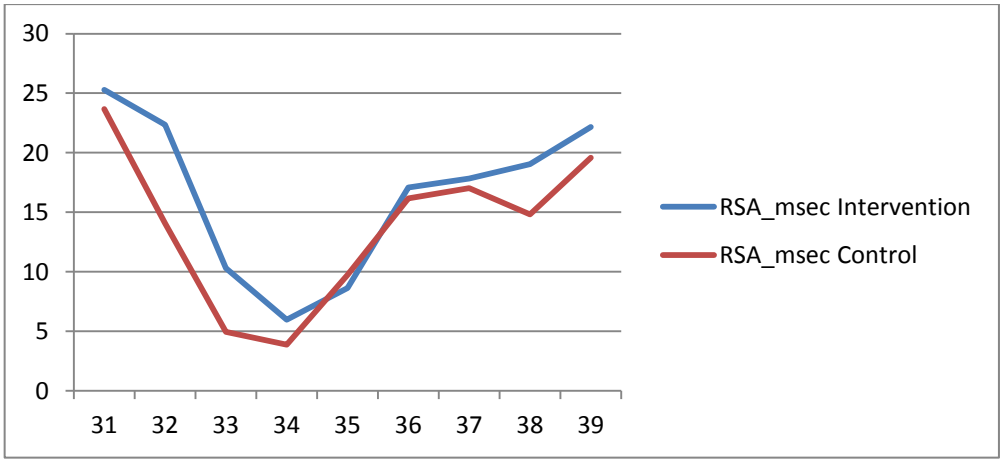


4.3.6 Respiratory Sinus Arrhythmia (RSA)

The RSA is a vagal tone. The intervention is stronger and so, there is a better vagal tone.

At point 32 when the babies are separated, the RSA is lower in the control, (control vs intervention: the p5012 vs 21, p 0.034) which is statistically significant. At point 33, it is not statistically significant but it is close at 0.08 and then becomes less significant. It is not influenced during the operation except for some patterns of higher vagal tone in the intervention group.

Figure 13:Respiratory Sinus Arrhythmia



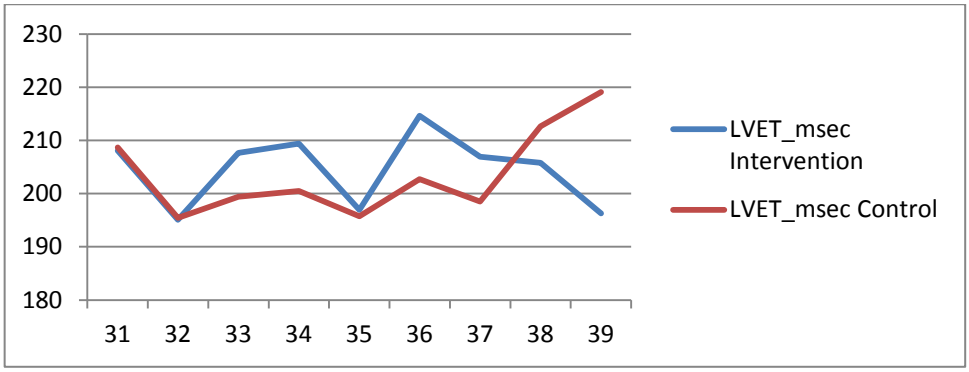
Impendence cardiograph

4.3.7 Left Ventricular Ejection Time (LVET)

LVET refers to sympathetic control of how long the heart beats. The controls are producing less at point 32 to 37 but more afterwards.

At point 39 when the babies are separated, the LVET is higher in the control, (control vs intervention: the p50 is 223 vs 193, p 0.14) but it is not statistically significant. This is a delayed stress response in the control group.

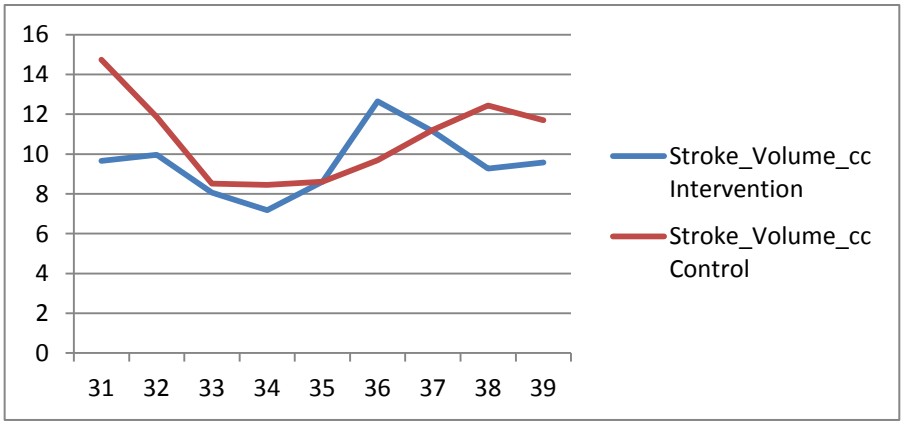
Figure 14:Left Ventricular Ejection Time



4.3.8 Stroke volume (SV)

At point 31 when the babies are separated, there is a sympathetic beat time (Control vs intervention: the p50 is 14.7 vs 8.8, p 0.17) but it is not statistically significant.

Figure 15: Stroke volume

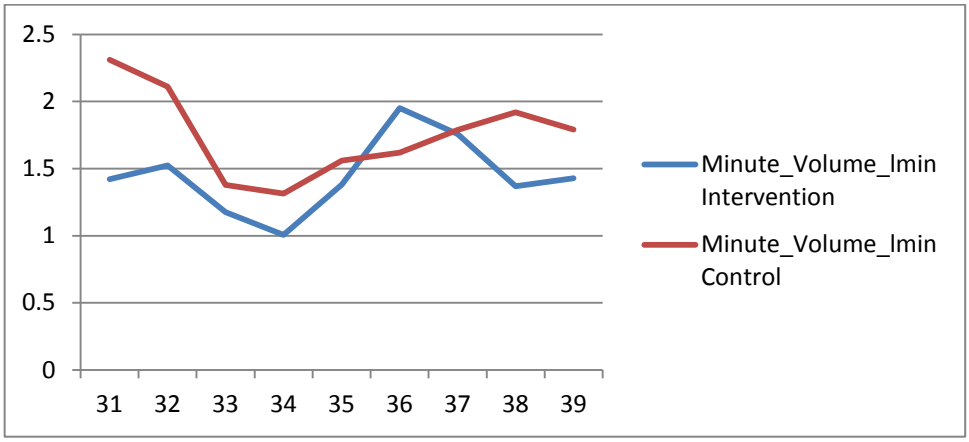


4.3.9 Minute Volume

The minute volume is based on stroke volume but factors in the heart rate.

At point 31 when the babies are separated, the minute volume is higher in the control, (control vs intervention: the p50 is 2.3vs 1.3, p.18) and there is a big difference between 2.3 and 1.3 but it is not statistically significant. The patterns remain the same. In the last part of the response, they rise and fall.

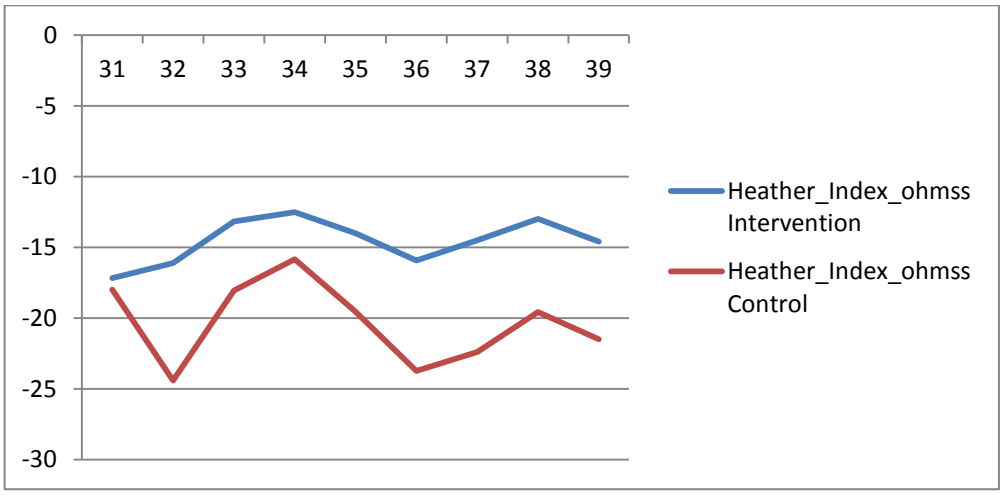
Figure 16: Minute Volume



4.3.10 Heather index

At point 32 when the babies are separated, the heather index is lower in the control, (control vs intervention: the p50 is -2 vs -17, p 0.07) but it is not statistically significant. The more minus the heather index is, the more stress there is.

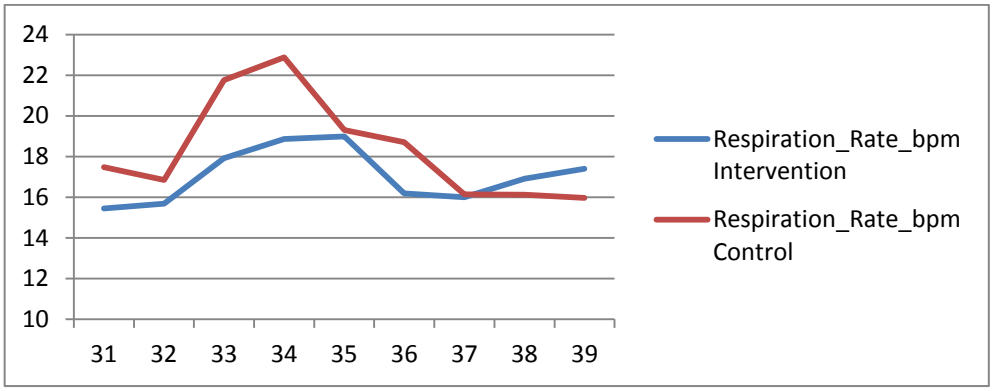
Figure 17: Heather index



4.3.11 Respiration rate

At point 36 when the babies are separated, the respiratory rate is higher in the control, (control vs intervention: the p50 is 23 vs 16, p 0.02). This is statistically significant. It is higher in the control group and perhaps they are experiencing more anxiety at this point.

Figure 18: Respiration rate



4.3.12 Pain profile

The neonatal pain score was planned to be used as an additional indicator of infant pain and discomfort. This proved to be very difficult to use with one researcher because of the observations and additional monitoring that needed to be done. It was more complex than anticipated. In the pilot cases, the scale was used. It was not removed from the schedule as it was thought that regular scores could be recorded. However, this was not possible. If pain was recorded at the same regular intervals it might have yielded different results which I am not certain what they may have been. The scores that were tracked are indicated below.

Table 14: Time and score

Case	Time and score											
	07h00	08h00	09h00	10h00	11h00	12h00	13h00	14h00	15h00	16h00	17h00	18h00

AS001						3		1				
AS002	1		4		0							
AS003						2		3				
AS005				2		3		1				
AS006								2	0			
AS007		1		3		1						
AS008		2		3								
AS009				4	1				3			

The variability of time intervals meant that it was not easy to meaningfully analyse the data. There is no trend or significance to be reported in this data.

5. DISCUSSION

This study set out to investigate the effects of maternal separation of infants younger than 6 months of age and undergoing hernia repair surgery. A prospective, single blinded, randomized control clinical trial design has ensured a set of detailed data. Unfortunately, as explained below, only 8 infants were recruited and therefore the results are not statistically significant. There are trends that can be flagged in a variety of aspects around infant stress in clinical settings which warrant further study and consideration in view of the current clinical practices.

The objective of the study was to show if decreasing maternal separation of under 6-month old infants directly before and after surgery decreased their allostatic load and improved outcomes. The support for this hypothesis is detailed below.

5.1 Trial profile

There were 40 infants assessed for eligibility and 30 recruited into this study. All of these infants were admitted to hospital through the full admissions procedure and prepared for theatre. 17 of these infants did not receive their surgery as scheduled. There were 5 data sets that could not be used for

technical reasons and this left 8 data sets that were usable. These were included in the full data analysis. All thirty infants' mothers or legal guardians were consented and fully prepared. While data from these infants may provide interesting insights about infants requiring surgery in this setting, some of the observations and data are not included in this discussion because the researcher was not intending to study them.

5.2 Polygraphs and analysed periods of cases reports

The observation tool allowed detailed description of events and helped to recognize interesting trends and behaviour of infants. These included admission procedures, practices around preoperative fasting, complexities of very small for age infants, those that are breastfed or have formula milk and communication challenges between different service areas and teams in this setting. These findings are interesting and may provide rich baseline data for clinical practice development and communication norms and practices. They have been shown in the results for each individual case study. As in the profile data, this data was additional findings and not directly related to the mother's presence and allostatic load in the infant which was my study objective. These circumstances were not the objective of my study. They are reported but will not be discussed.

The time period that was analysed yielded different results for the heart rate variability and the impedance cardiograph. The different outcomes are discussed below for both the control and intervention groups.

5.2.1 Low frequency (LF)

The low frequency (LF) was the same from the time infants left the ward until surgery for both the control and the intervention groups. During recovery, it started going higher and by the time infants were settling on the ward, the intervention group had higher low frequency (LF) than the control group. A higher low frequency is interpreted as a healthy and mature autonomic response (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).

5.2.2 High frequency (HF)

Similar to the low frequency, the high frequency was the same for both groups from the time the infants were on the ward until surgery. During recovery, the HF was high in both groups but later on dropped in the control group during the post-operative period while going even higher in the intervention group. HF is a parasympathetic measure and a high HF is interpreted as a healthy and

mature autonomic response (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).

5.2.3 Low frequency high frequency (LFHF)

The LFHF was higher in the control group most especially during anaesthesia and recovery. The LFHF is the proportion of LF to HF and is therefore a proxy for sympathetic activity. The more sympathetic it is, the higher LF is compared to the HF and therefore the higher these numbers. The control was more sympathetic than the intervention group and was therefore more stressed (Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology, 1996).

5.2.4 Respiratory Sinus Arrhythmia (RSA)

The control group had a lower RSA compared to the intervention group. This was observed right from the ward through to recovery. The RSA is a vagal tone. The intervention group had a higher RSA and therefore a better vagal tone.

5.2.5 Left Ventricular Ejection Time (LVET)

LVET indicates the length of sympathetic control on the heart beats. The control group had lower values than the intervention group. The intervention group was probably experiencing greater activation of the stress response during the periods of separation but this quickly came down when the infants were re-united with their mothers. Charney, 2004 refers to it as "resilience" which is the ability to bring down a stress response. There is a delayed stress response from the control group.

5.2.6 Stroke volume (SV)

The higher the SV, the more sympathetic it is. The control group had a higher SV compared to the intervention group through the period from the ward until recovery room. The SV was higher in the intervention group from the post-operative period until the infants came back to the ward. With such results from SV, an observation was made that infants in the intervention group cried much more when they were re-united with their mothers. This could have influenced the rise in the SV because later, it came down while the SV in the control group remained high.

5.2.7 Heather index

The minus in the heather index means that is more sympathetic. The heather index was more minus in the control group than the intervention group throughout the whole process from the ward to theatre and back to the ward.

5.2.8 Observation and analysis

There was a time when an infant received a heel prick. On the ICG, the LVET and minute volume went up. The heel prick event showed an acute stress response while maternal separation showed a delayed stress response. Babies may not express separation anxiety but it is measurable by ICG.

The intervention infants cried more when they met with their mothers, who matched the acute response but they later came down. The control group did not cry that much but they demonstrated a high stress response. Infants may not express their stress response but it can be measured.

5.3 Heart Rate Variability

The HRV results showed that there was an increased vagal tone, higher LF and HF in the infants when the mothers present. This is a reflection of maturation but in these infants, it may be a reflection of improvement in the autonomic control.
The HRV of the controls revealed lower values of the vagal tone, a reflection of poor autonomic functions.

5.4 Impedance Cardiograph

The ICG showed a pattern of responses consistent with increased sympathetic response. The intervention ones are showed an acute response and then calmed down. The controls did not show any acute response but a delayed response was revealed by the measurements. Studies done in monkeys by Feng et al., 2011 showed that maternal separation produced lasting changes in cortisol secretion and behaviour. The function of cortisol is vital to the wellbeing of an individual but if it remains persistently high, it starts to cause harm (Charney, 2004). Bringing down the cortisol levels is therefore very important. A mother's presence is vital in bringing down the sympathetic response as evidenced on ICG. Her absence causes the sympathetic response to stay up for much longer. Mothers should therefore be present with their infants.

5.5 Pain profile

It was the complexity of the situation, the different times and lack of standardization that made the data unusable.

5.6 Strengths of this study

- Detailed and complex data sets were collected and they have provided insight into a range of practice complexities that may not have been well described before. A robust description of biological parameters and indicators of infant stress that have not been measured in the context of clinical interventions before.
- Acquisition, mastery and use of complex yet non-invasive data gathering equipment and skill refined to utilize this equipment over a periods of several hours in the real world context of various clinical environments – surgical ward → transfer of an infant on a large trolley → transfer onto an operating bed → during surgery → back to trolley → recovery room→ back on a trolley → settled in ward.
- As a nurse, I was able to follow these infants through this process and collect these recordings and collate them with observations.
- A standardised group of infants was identified. They all had the same diagnosis and even in the statistics, they all had the same average age. Hence, they were comparable.
- For all the cases in the intervention group, it was real intention to treat.

Weaknesses of the study

- While 30 infants were recruited and consented, the sample size of eight was too small to prove statistical difference.
- It was a challenging environment. I was not able to be present at all times when the baby was being moved and at the same time take notes for the observations.
- The cases got cancelled despite the fact that they had been prepared.
- A second researcher would have been useful to gather data from the mother and also to gather the pain comfort data with the NIPS. While the control group was subjected to the practice norm, it became evident that this was not very clear; hence a number of unforeseen variables like feeding norms, pre and post. Theatre affected the results and added to the single variable of the mothers' absence or presence.
- In the absence of a predictable theatre list, all participants were not equally prepared and the monitoring started at various times rather than uniformly (at least 2 hours before theatre). The control period of the observations before randomisation became too short.

6. RECOMMENDATIONS

There are trends that clearly deserve further study and stronger statistical analysis but in the meantime, the data does support the hypothesis made that infants below six months should have their mothers with them in the operating theatre and recovery room when the infants wake up.

This also creates an opportunity for follow-up research and practice development work.

The machine creates the potential to further study this particular topic that has been addressed and to also look at other situations in the hospital because it captures a lot of variables that can be analysed.

A reflection from my observations is that infants in the intervention group were fed in the recovery room while those in the control did not feed immediately, even after they returned to the ward. They were given half dextrose water. This could be an aspect of study in the future.

CONCLUSION

My hypothesis was that mother's presence makes a difference to the autonomic response to stress in infants under six months old undergoing elective hernia surgery. The results as discussed above showed that heart rate variability, impedance cardiograph, observations and behaviours support the hypothesis that there is a difference. By using HRV as a measure of sympathetic influence and ICG as a sympathetic response, it was shown that infants who were separated from their mothers were stressed even though they did not express separation anxiety. My recommendation is that mothers should accompany infants into the operating room. Because these results are not statistically significant, more research needs to be done before policy changes can be implemented.

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APPENDICES

Appendix I: Informed consent letter

Information Section:

Dear Parent,

My name is Lydia Ssenyonga, a registered nurse with a Postgraduate Diploma in Child Nursing. I am currently a Masters' student, at the University of Cape Town. I am conducting a study that is looking at how babies experience stress in babies under six months of age. I am focusing on babies who will be undergoing a hernia repair operation. The goal is to understand if babies under 6 months of age that are separated from their mothers experience more stress than they can handle. The information gained from this study will help in improving practice and policies in paediatric hospitals.

You are invited to participate in the study with your baby because you baby meets the study requirements. There are other mothers and babies who will also participate. You will be allocated to one of two groups. In the one group, the baby will be treated in the normal way which is that the baby is settled in by the mother and then is taken to the operating theatre by a nurse and porter. When the baby wakes up after the operation, he/she is brought back to the mother in the ward.

In the second group, the mother will be asked to accompany the baby to theatre with a nurse and a porter. The mother will stay with her baby until he/she is asleep in theatre. She will then be asked to wait outside while the operation is performed. After the operation, she will be asked to come in just before her baby wakes up. The mother, nurse and porter will accompany the baby back to the ward. The group to which you will belong will be determined by the number in the envelope that will be randomly picked at the beginning of the study.

Your baby's stress will be measured by a heart monitoring machine. This machine has electrodes with stickers which will be placed on your baby's chest from the time the child goes for the operation until when he/she returns to the ward. The stickers are not painful and will be secured with tape, so your baby will be able to sleep on his/her back without being disturbed. On completion of the data collection, the buttons will be gently removed without causing any pain to the child.

The information that is collected from you and your baby will be kept in a way that does not identify you or your baby. The final results from the study will be shared with the University of Cape Town and Red Cross War Memorial Children's Hospital but you and your baby's name will not appear on any of the results. .

During the research you are free to contact myself, the University of Cape Town or my supervisor should you have any questions or concerns regarding your rights. Below are the contacts. You are free to withdraw from the study at any time without giving a reason and it will have no bearing to the treatment of your child.

Thank you for considering participating in this study which will benefit the care of babies in future.

Primary researcher
Lydia Ssenyonga
University of Cape Town
Division of Nursing and Midwifery
Cell. 0730366336
Email: lydiasenyonga@yahoo.com

Supervisor
ProfessorMinette Coetzee
University of Cape Town
Child Nurse Practice Development initiative
School of Child and Adolescent Health
Phone: 021-6855392
Email:Minette.coetzee@uct.ac.za

University of Cape Town
Faculty of Health Sciences
Human Research Ethics Committee (HREC)
Tel. 021 406 6338

Appendix II: Consent form

Measuring stress of infants under 6 months old undergoing hernia repair

I _____ have read (or had read to me by _____) the Information Sheet. I understand what is required of me and my child and I have had all my questions answered. I do not feel that I am forced to take part in this study and I am doing so of my own free will. I know that I can withdraw at any time if I so wish and that it will have no bad consequences for me or my child.

Signed:

_____ _____

Participant Date and place

_____ _____

Researcher Date and place

_____ _____

Witness Date and place

Appendix III: Observational data tool

Date ……../……. / 20…….	Informed consent Yes/ No. circle	Research number.....
Date of birth	Current age in months	Weeks at birth
Weight (kg)	sex	Date of admission
Antenatal history	Postnatal history	Immunisation (up-to-date) Yes No circle
On set of problem	Time of NPO	
Type of feed.....	Time given.....	Given by.....

Medication administered

On the ward	Time	Given by
In theatre	Time	Given by

Role Players

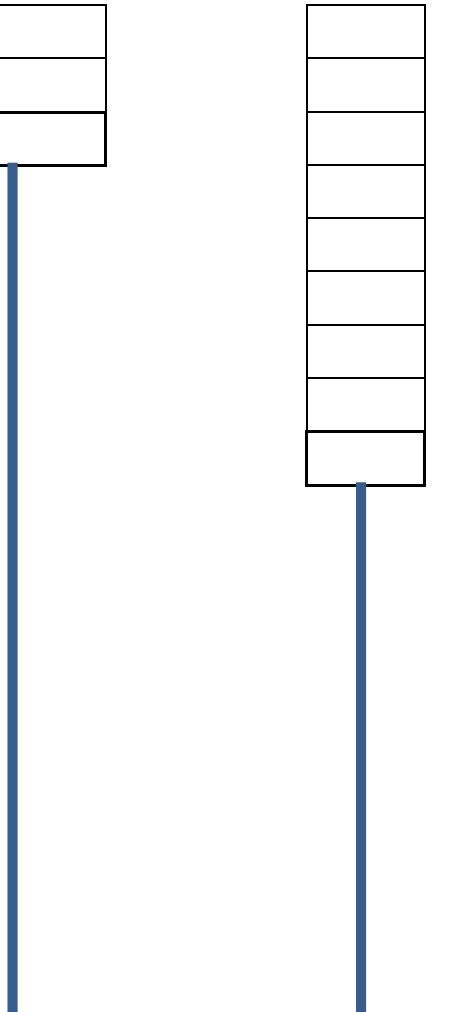
Symbol	**Category**
PN	Professional Nurse

PS	Principal Researcher
SUR	Surgeon
PAR	Parent
INF	Infant
AN	Anaesthetist
⬇	Time of mother – infant separation

Research number.....

RESEARCH TIMELINE ...

	Event overview	**Time for label button**	**CONTROL**	**INTERVENTION**
	Arrived to meet patient		Mother	Mother
	Patient connected		infant	infant
			separation	separation
1	Start recording			
2	Mother puts baby down			
3	Patient leaves ward			
4	patient into theatre			
5	Mother puts baby down			
6	Anaesthesia starts			
7	Intubation			
8	Surgery starts			



9 Surgery stops

10 Extubation

11 Mother arrives recovery room

12 Mother leaves theatre

13 Arrive aback on the ward

14 Attachment of monitors

15 30 minutes later

16 60 minutes later

17 90 minutes later

19 120 minutes later

20 Recording stops

21 Time feeds commenced

Research number Date

		State	

OUT COMES

During VU-ams	**What happened**	**What to do next time**
Day Report		
Night Report		
Analgesia	**Ward**	**Theatre**
Mother's experience **(What was it like for you?)**		
Reflections		

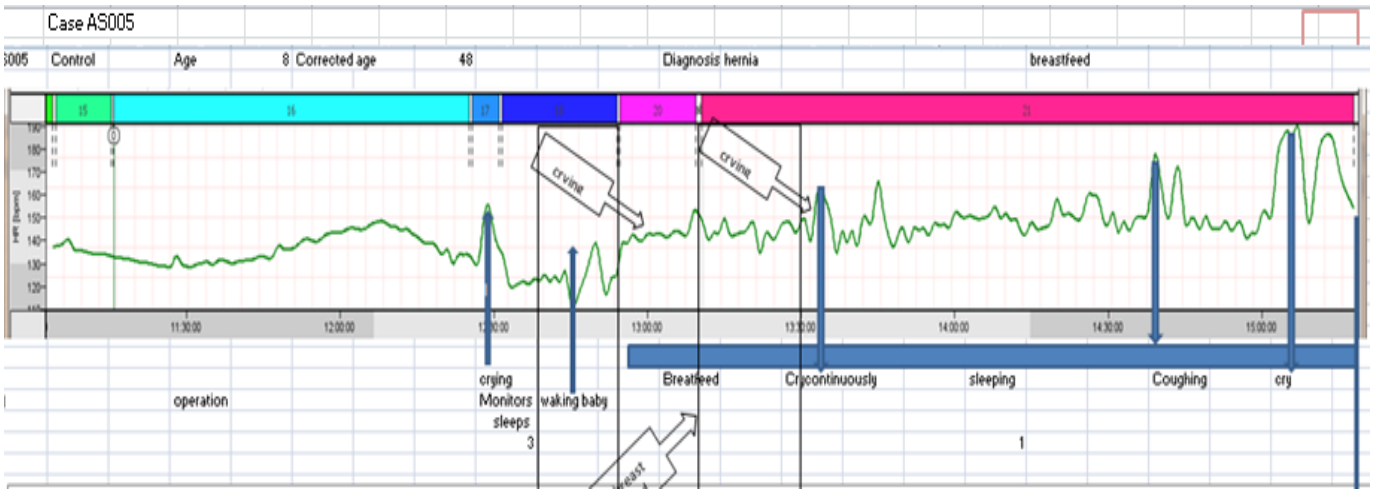
Appendix IV: Neonatal Infant Pain Scale (NIPS)

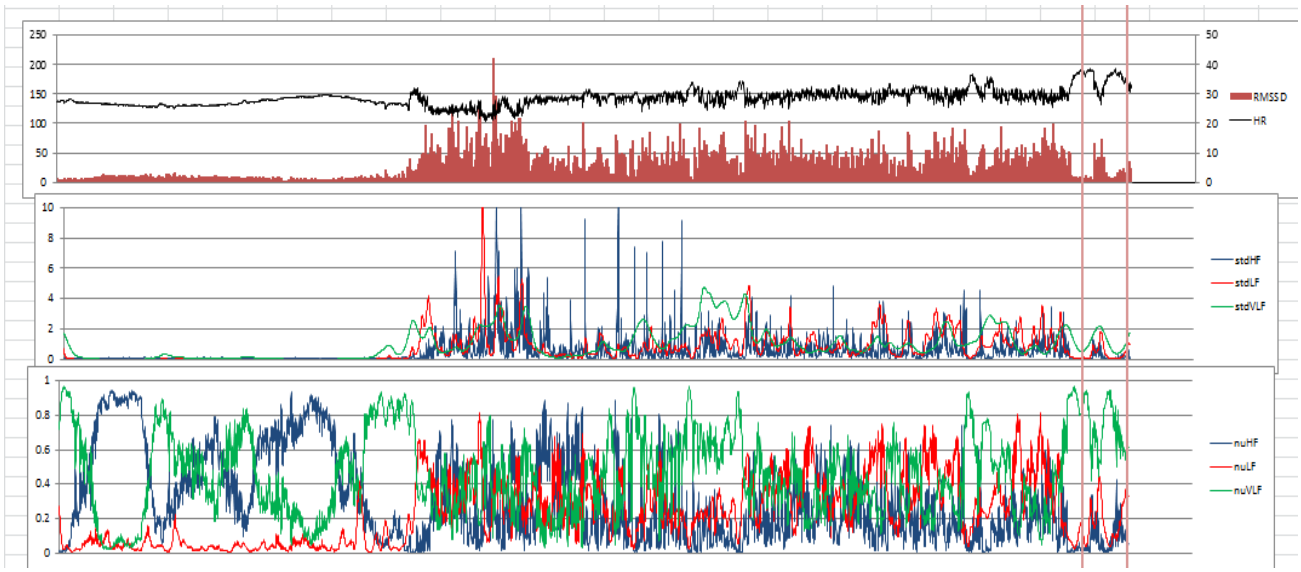
		Score
Facial Expression		
0 – Relaxed 1 – Grimace	Restful face, neutral expression Tight facial muscles; furrowed brow, chin, jaw, (negative facial expression – nose, mouth and brow)	
Cry		
0 - No Cry 1 – Whimper 2 - Vigorous Cry	Quiet, not crying Mild moaning, intermittent Loud scream; rising, shrill, continuous (Note: Silent cry may be scored if baby is intubated as evidenced by obvious mouth and facial movement)	
Breathing Patterns		
0 – Relaxed 1 - Change in Breathing	Usual pattern for this infant In drawing, irregular, faster than usual; gagging; breath holding	
Arms		
0 –Restrained 1- Flexed/Extended	No muscular rigidity; occasional random movements of arms Tense, straight legs; rigid and/or rapid extension, flexion	
Legs		
0- Relaxed/Restrained 1- Flexed/Extended	No muscular rigidity; occasional random leg movement Tense, straight legs; rigid and/or rapid extension, flexion	
State of Arousal		
0- Sleeping/Awake 1 - Fussy	Quiet, peaceful sleeping or alert random leg movement Alert, restless, and thrashing	

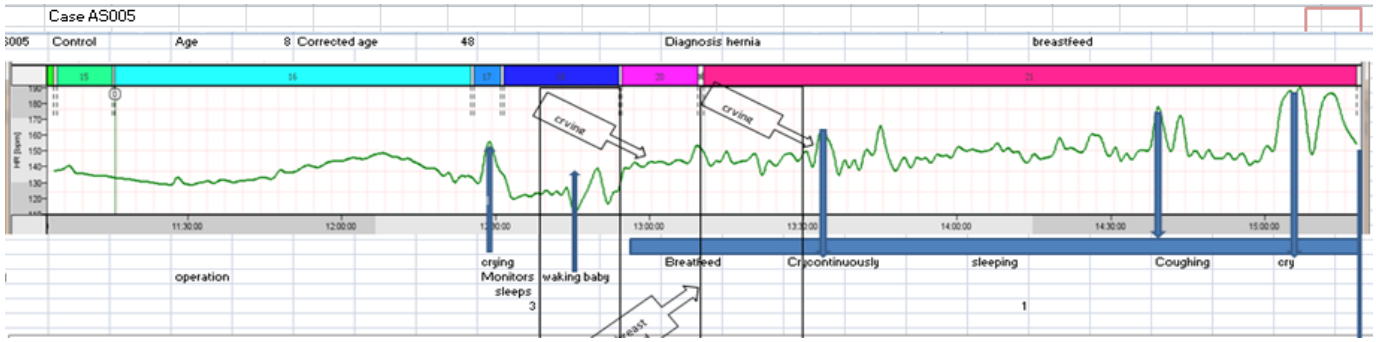
Lawrence, Alcock, McGrath, Kay et al (1993).

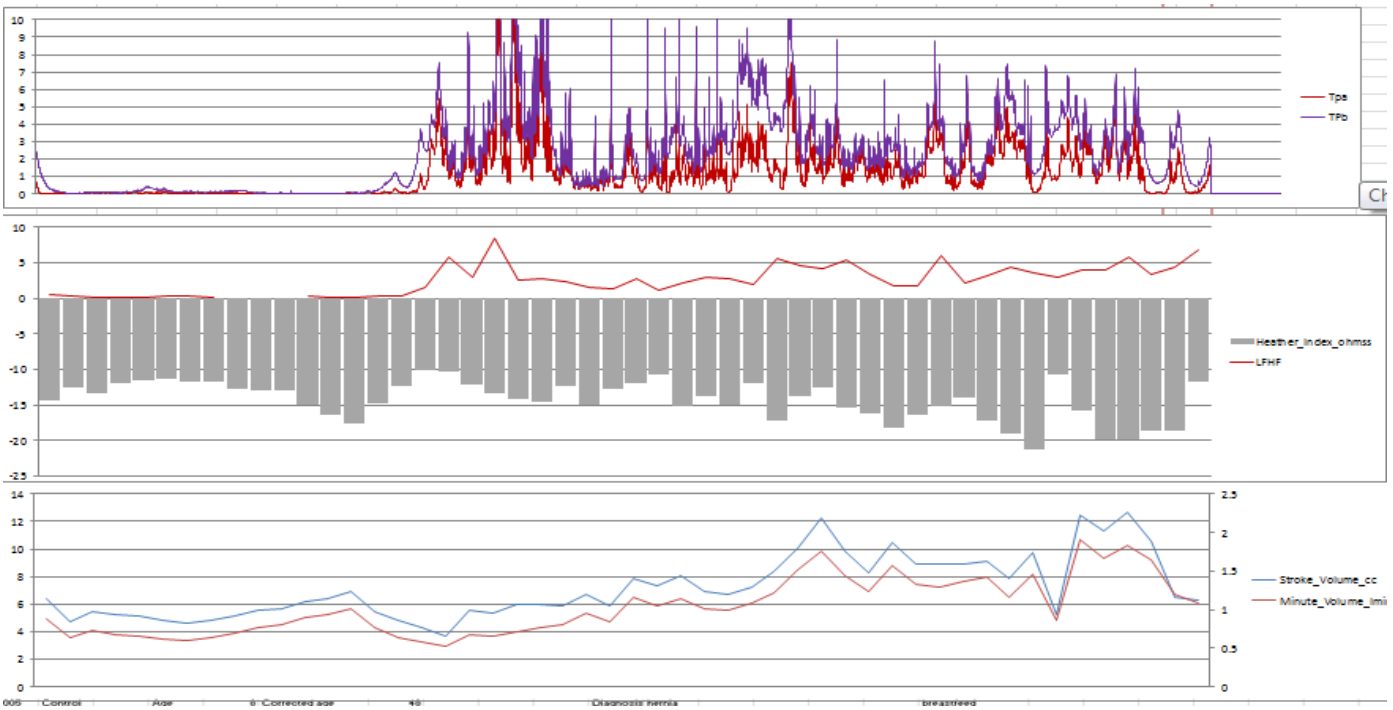
Appendix V: Control Group- First Infant

FIRST INFANT: AS005 Case no 5 Data period: 12hrs 17 minutes CONTROL GROUP





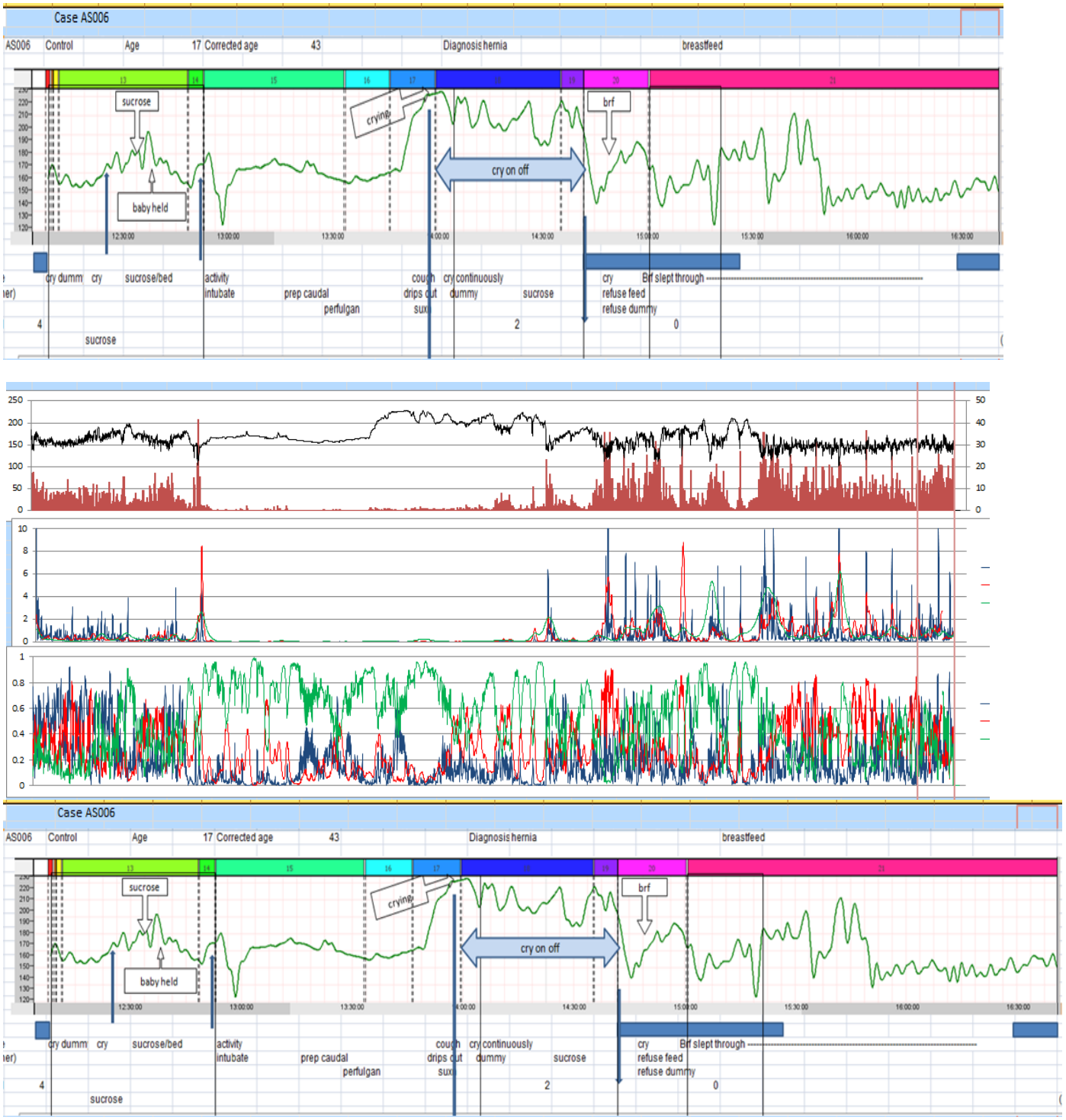


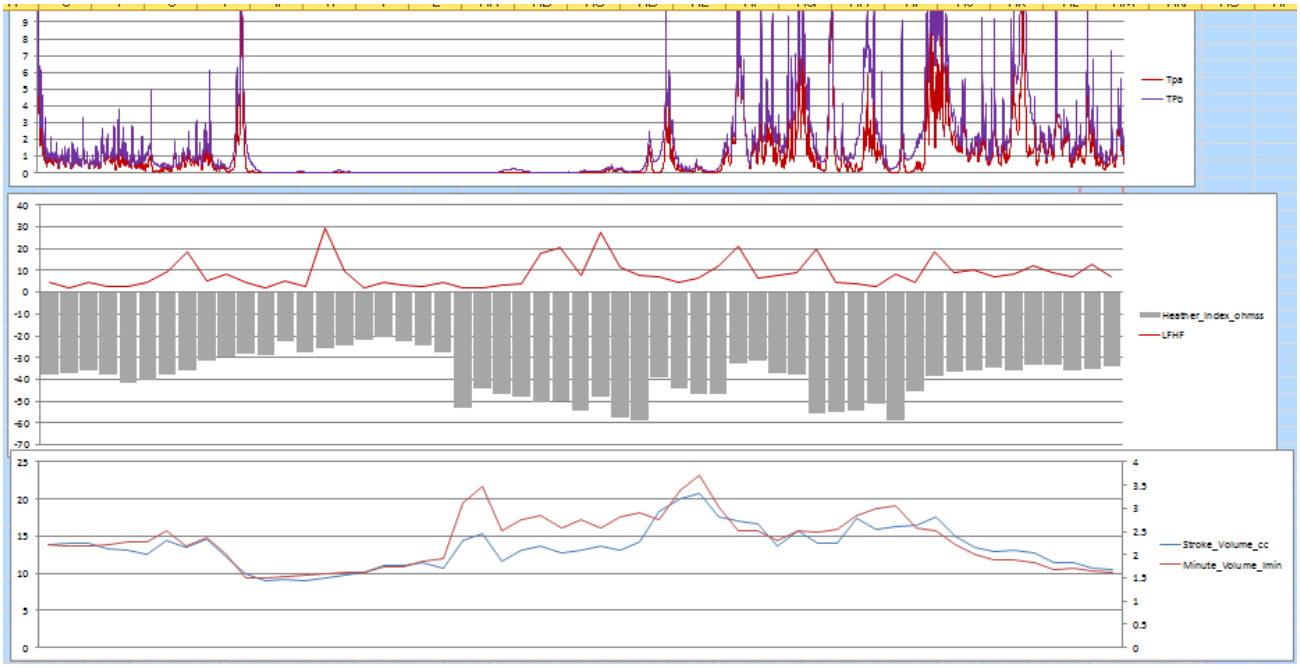


Summary: The RMSSD and the LFHF increase when the infant is being transferred to the ward which is a sign of stress. Despite of the fact that the infant is crying at this time, the heart rate was not high.

Appendix VI: Control Group- Second Infant

SECOND INFANT: AS006 Case no 6 Data period: 1 CONTROL GROUP

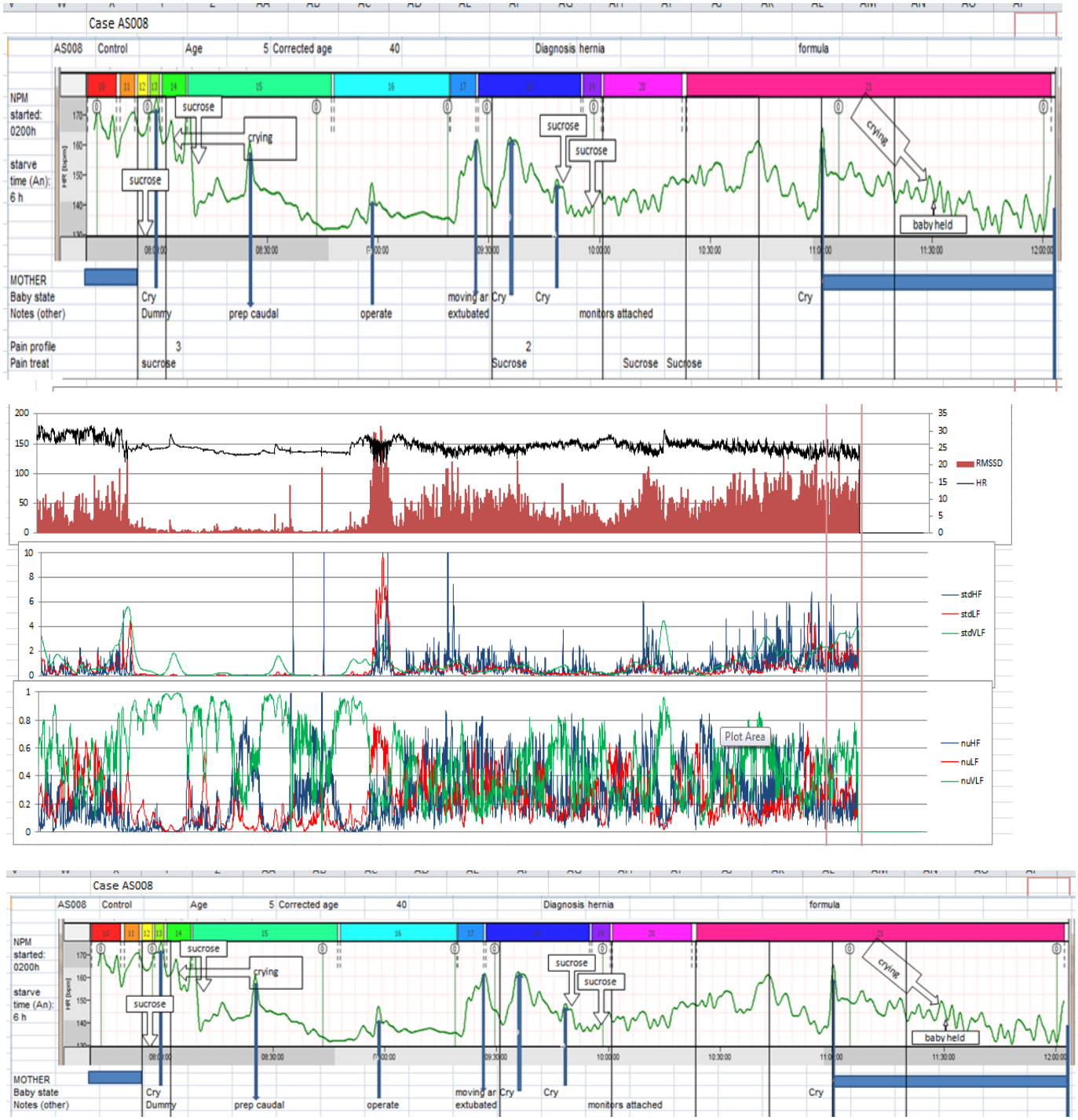




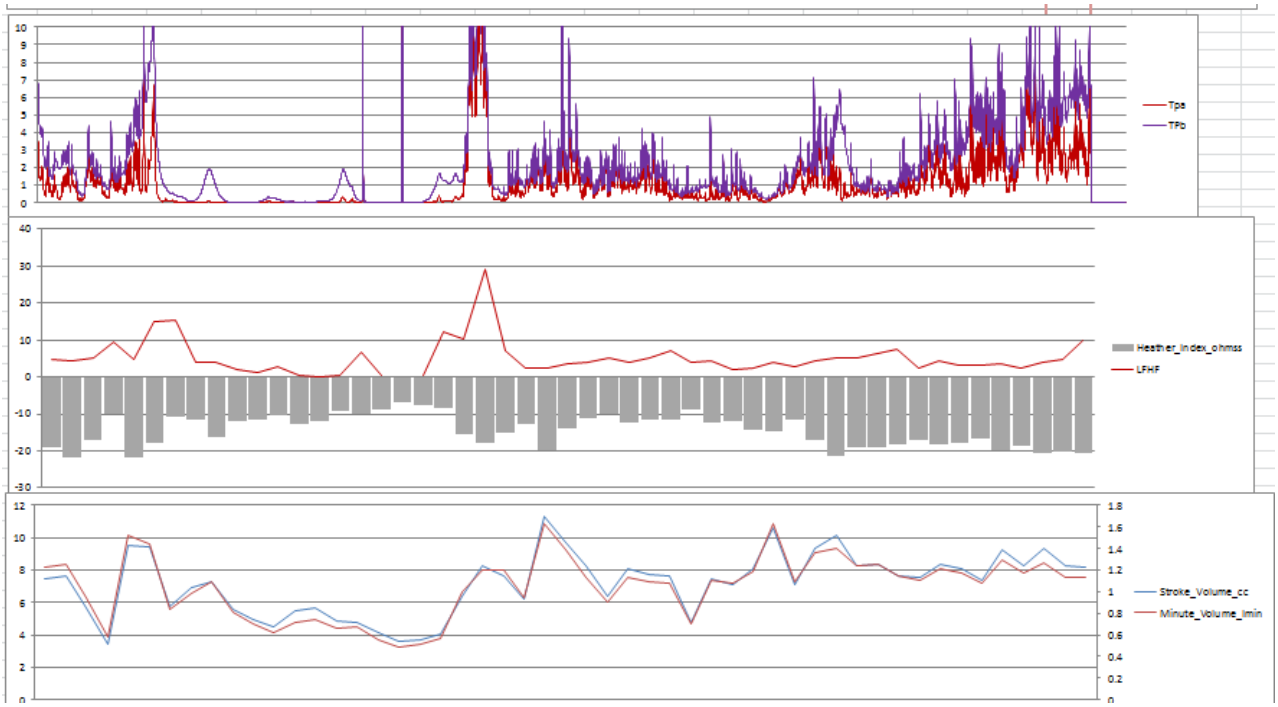
Summary: The heather index is high at one point during the time when the infant is anaesthetised and the minute volume raised during the time when the infant is waking up. These are sympathetic measures and which indicate stress. The RMSSD and standard HF are raised during the time of resting when the mother is away despite of the heart rate being low and the infant is sleeping.

Appendix VII: Control Group- Third Infant

THIRD INFANT: AS008 Case no 8 CONTROL GROUP



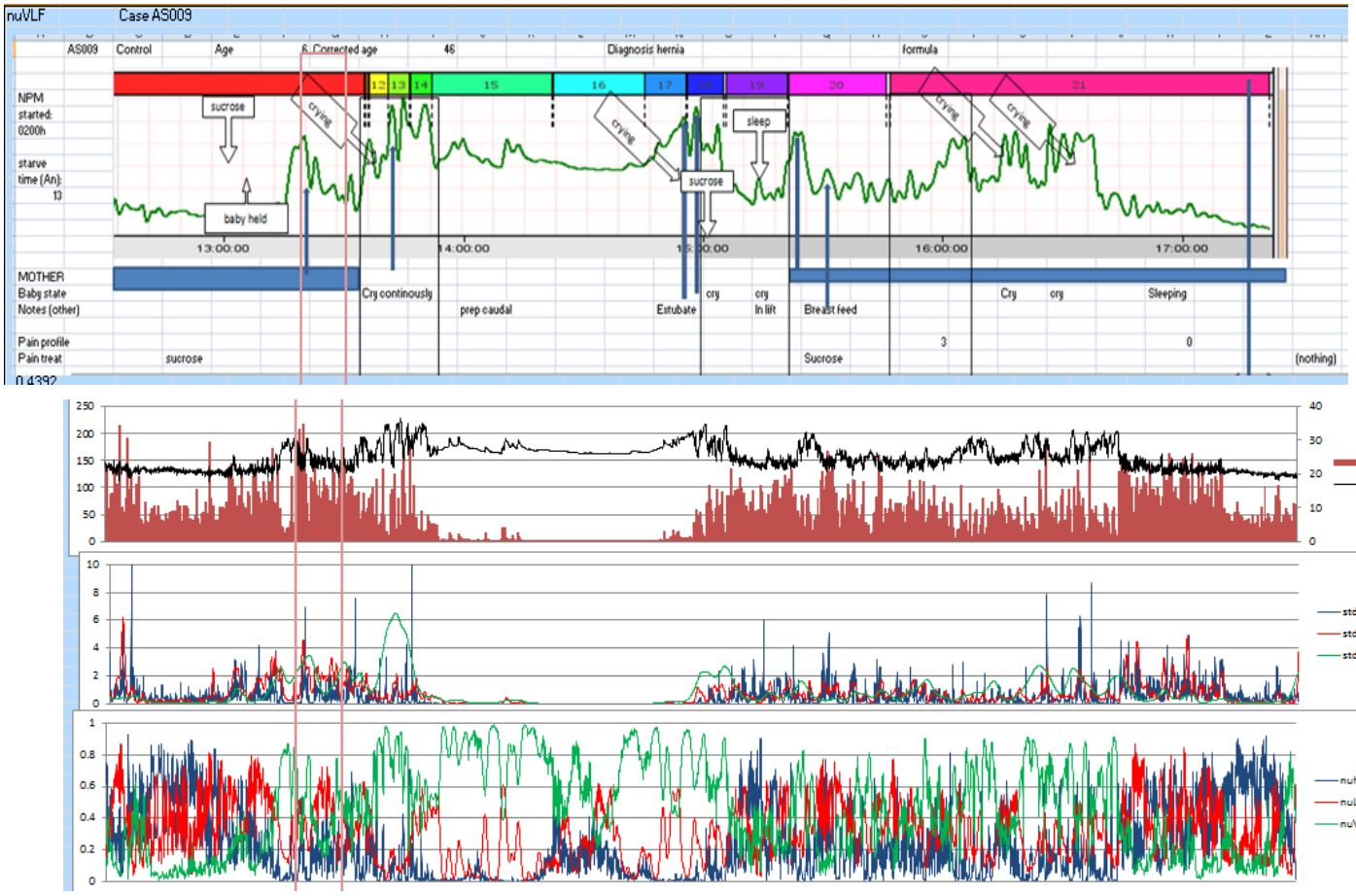
Case AS008
AS008 Control Age 5 Corrected age 40 Diagnosis hernia formula
NPM started: 0200h
starve time (An): 6 h
sucrose
crying
baby held
MOTHER
Baby state
Notes (other)
Cry
Dummy
prep caudal
operate
moving ar
extubated
Cry
Cry
monitors attached
Cry

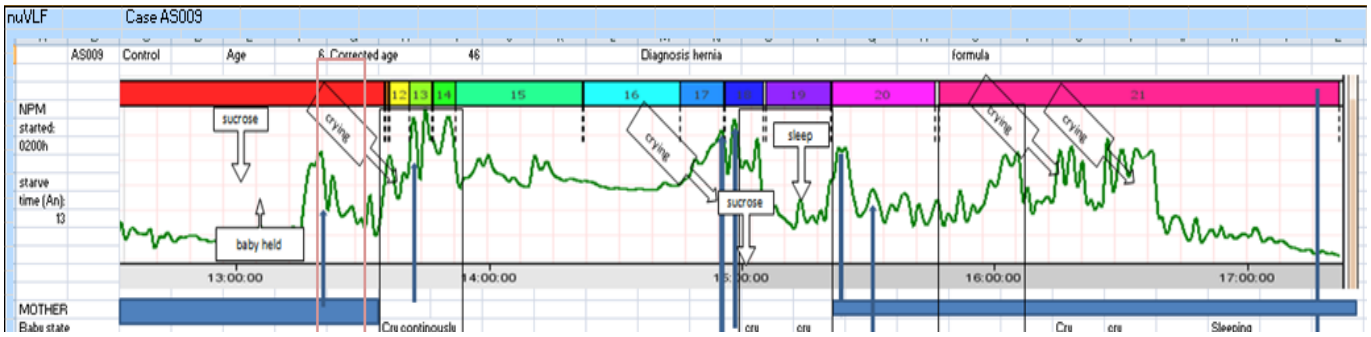


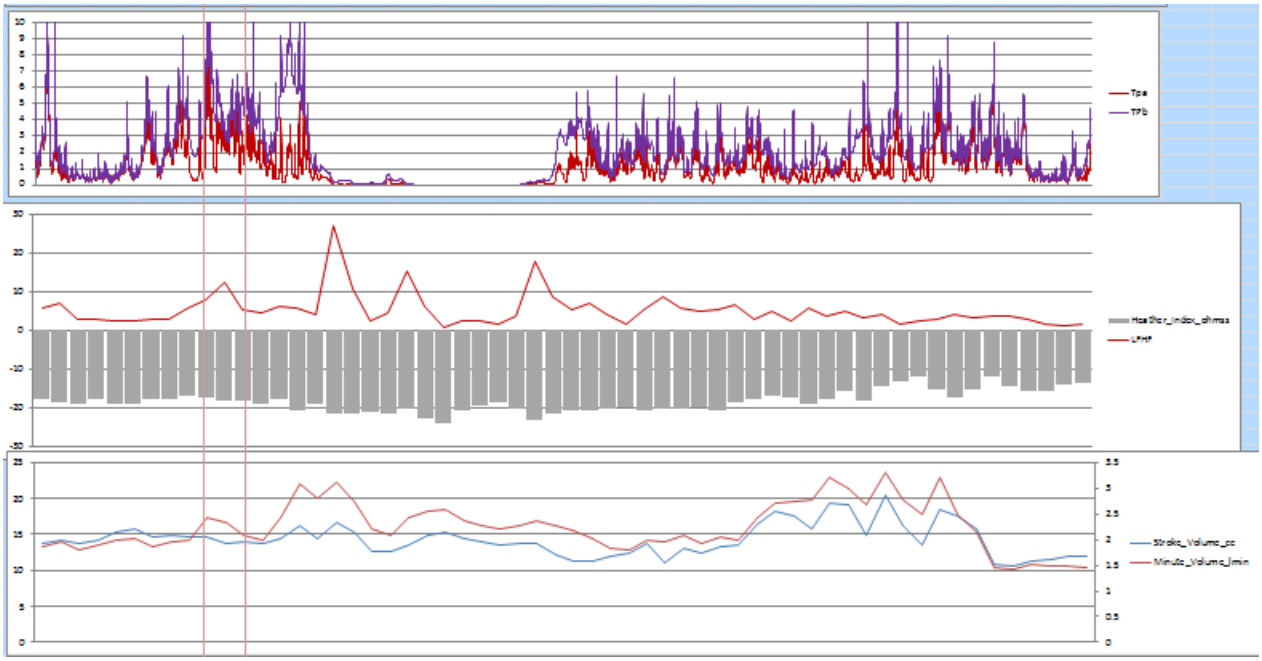
Summary: The VLF was high during anaesthesia and surgery. Stroke and minute volume were high in theatre when the infant was waking up. These are sympathetic measures and indication of stress

Appendix VIII: Control Group- Forth Infant

FORTH INFANT: AS009 Case no 9 CONTROL GROUP



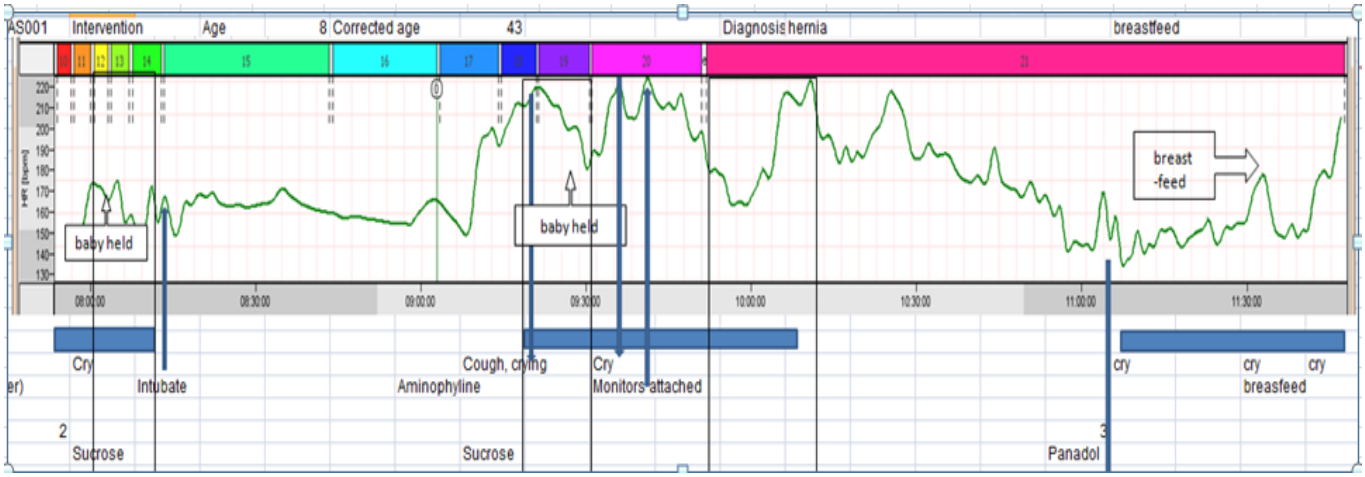


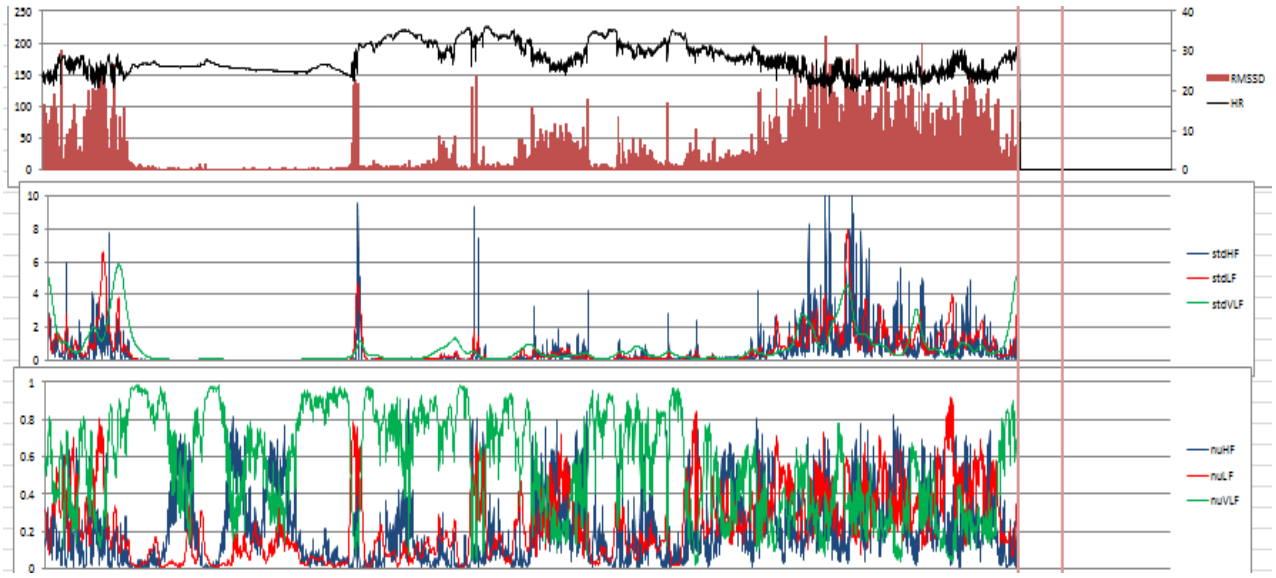


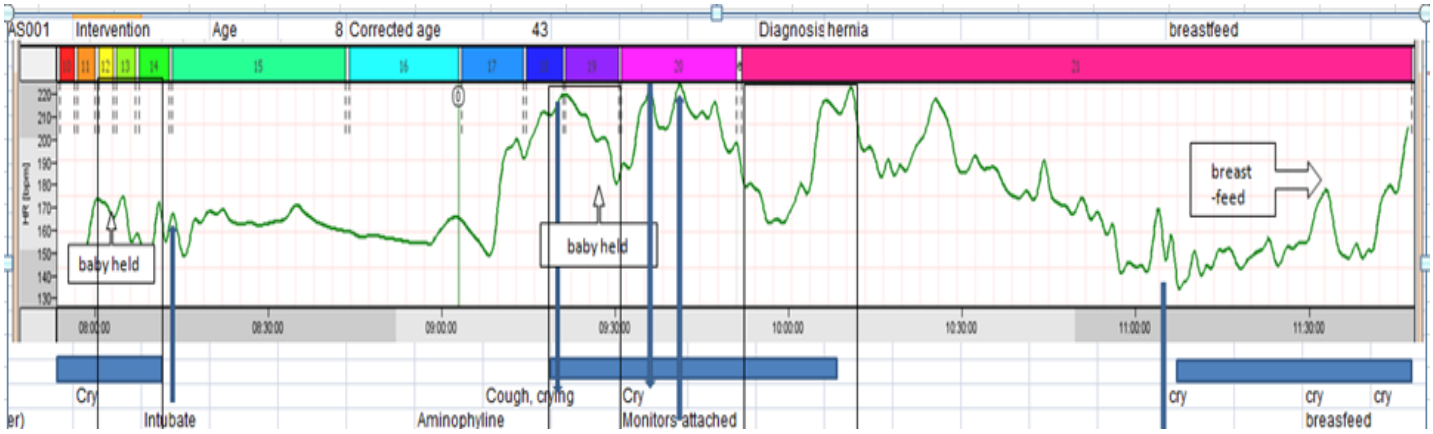
Summary:This infant was starved for 13 hours. Had a lower heart rate but when he was separated from the mother even during anaesthesia and surgery the heart rate was raised. Infant had a normalised very low frequency during surgery which is a stress indicator.

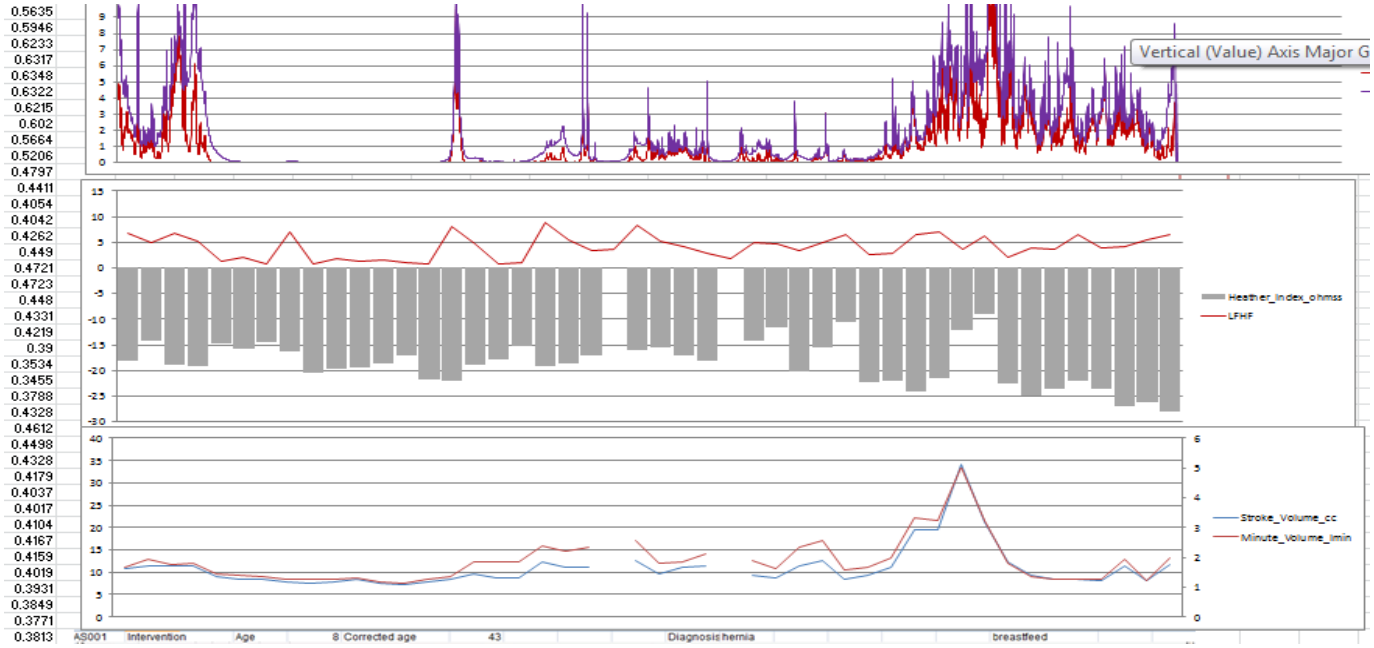
Appendix IX: Intervention Group-First Infant

FIRST INFANT: AS001 Case no 1 INTERVENTION GROUP





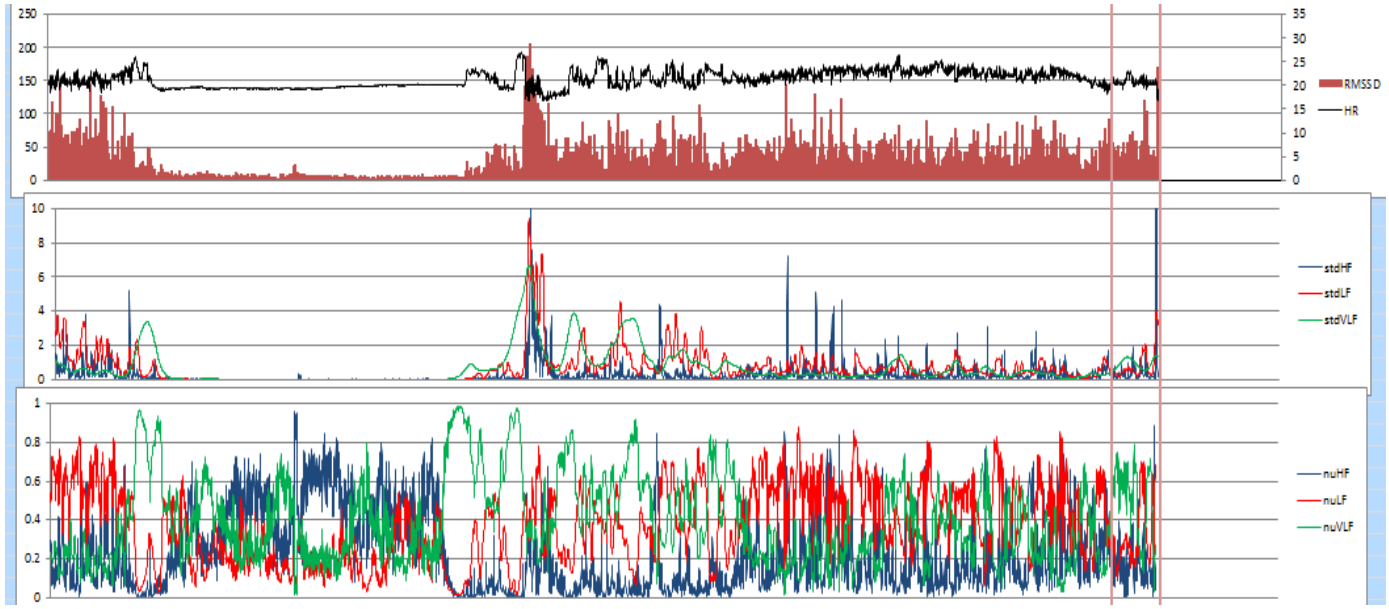


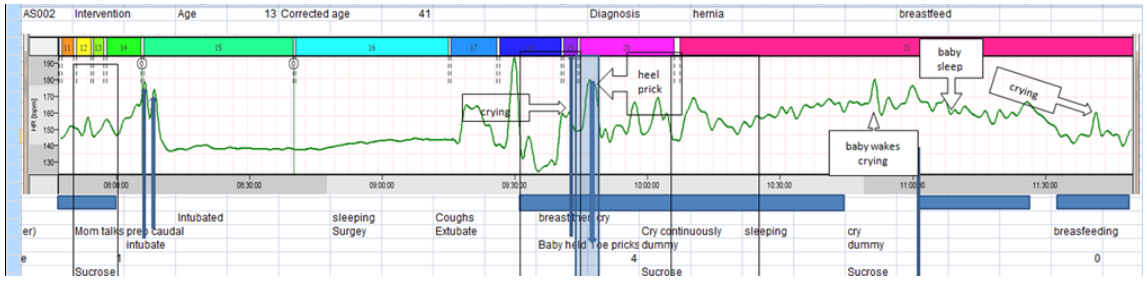


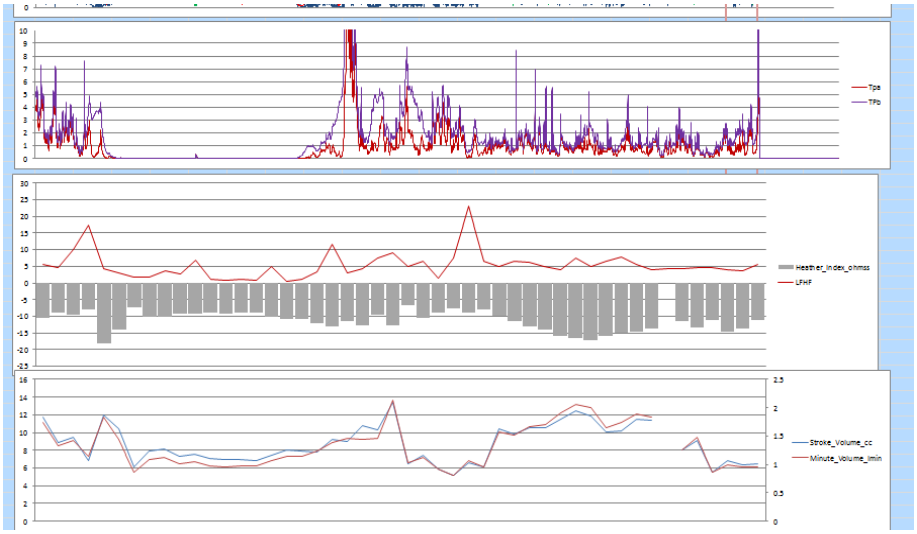
Summary: In this intervention case, the baby was quiet calm when the mother was present but her absence caused a very low frequency effect and later was replaced by high stress response which eventually comes down when she returned but it took a long time. The separation caused an autonomic response which looked like stress.

Appendix X: Intervention Group-Second Infant

SECOND INFANT: AS002 Case no2 INTERVENTION GROUP



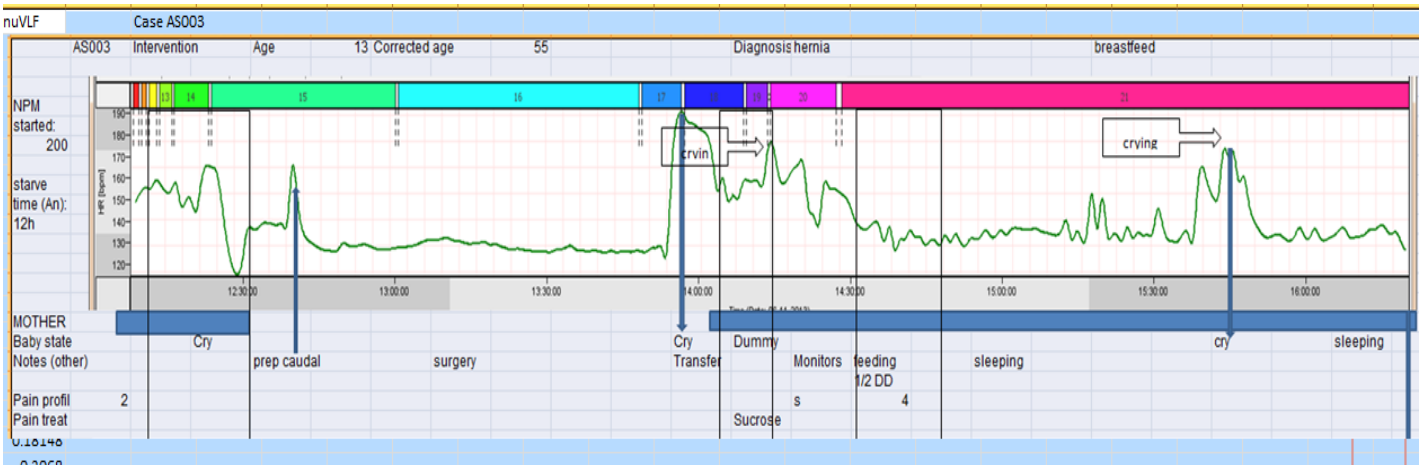


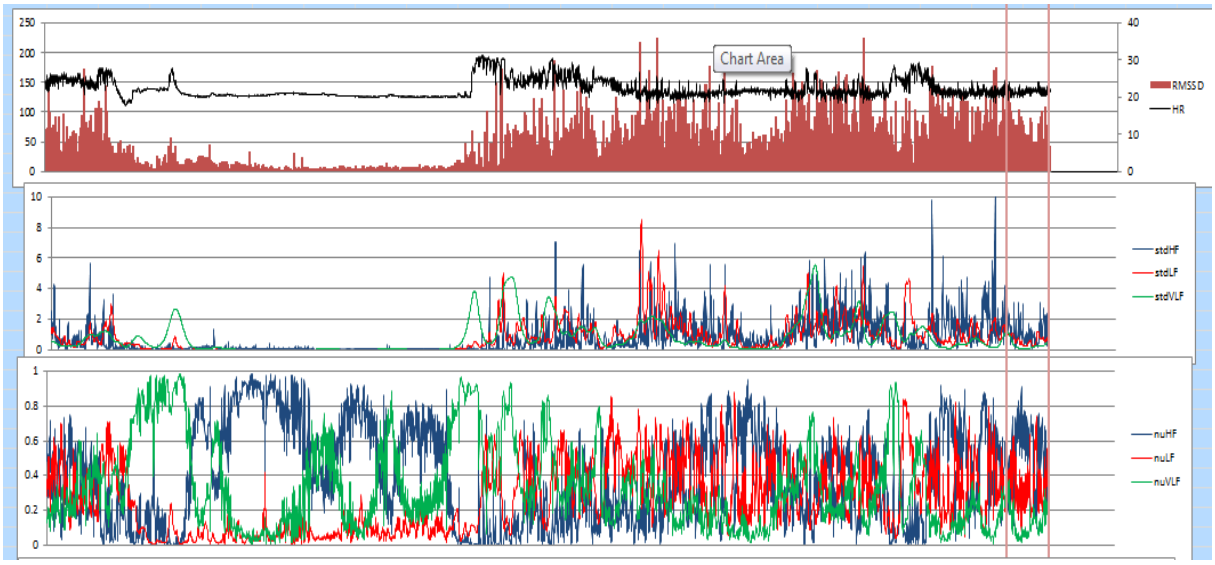


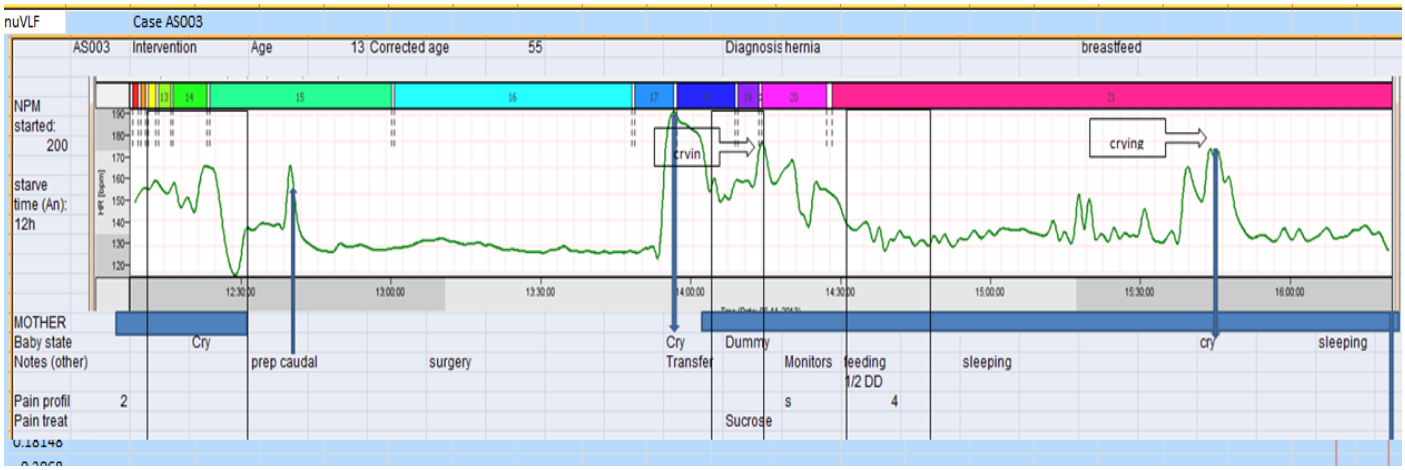
Summary:This infant had high normalised HF during surgery and low normalised low frequency

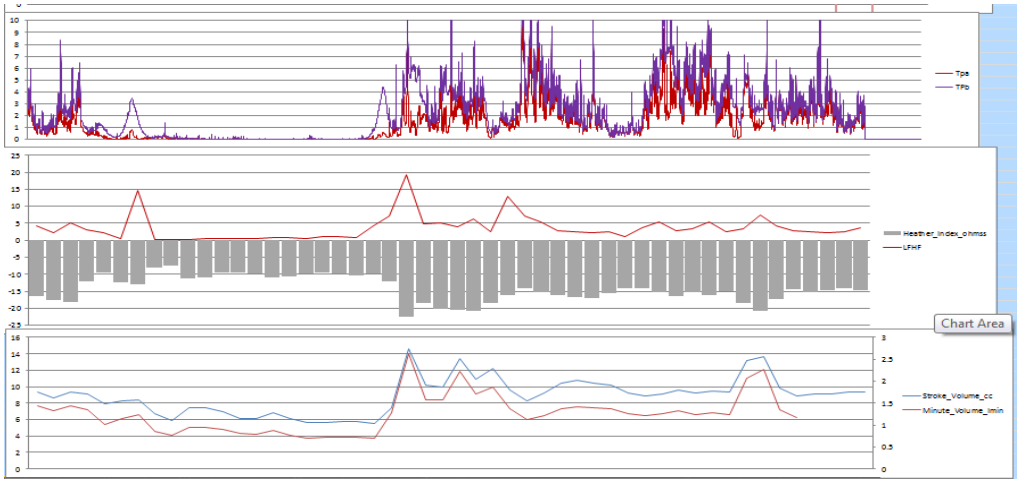
Appendix XI: Intervention Group-Third Infant

THIRD INFANT: AS003 Case no3 INTERVENTION GROUP





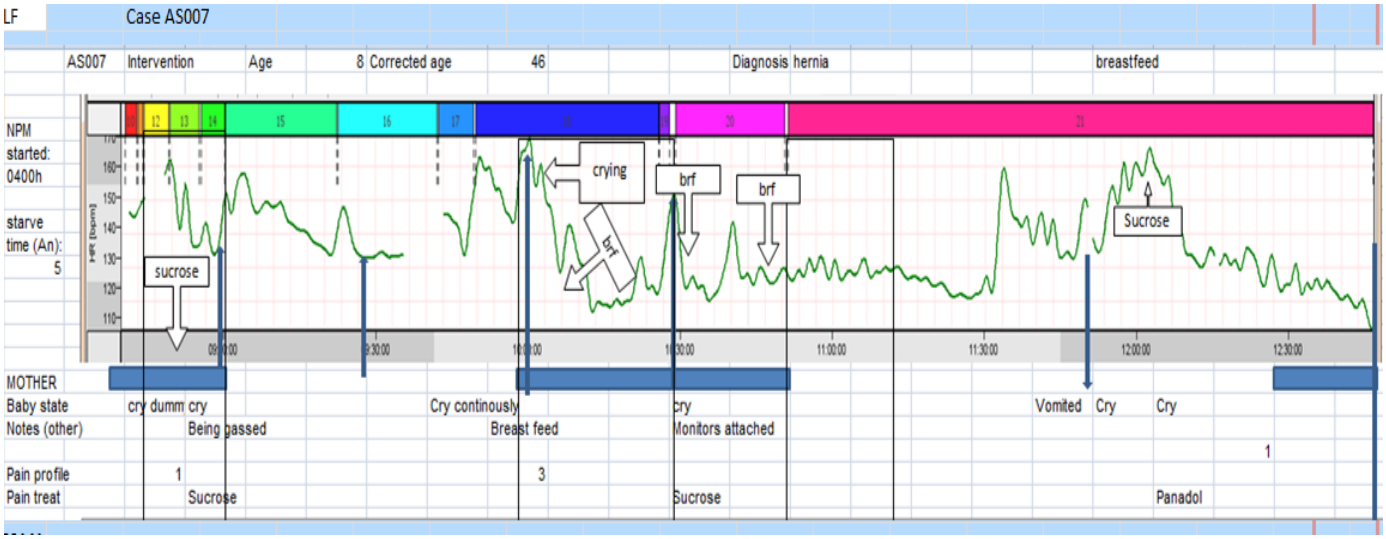


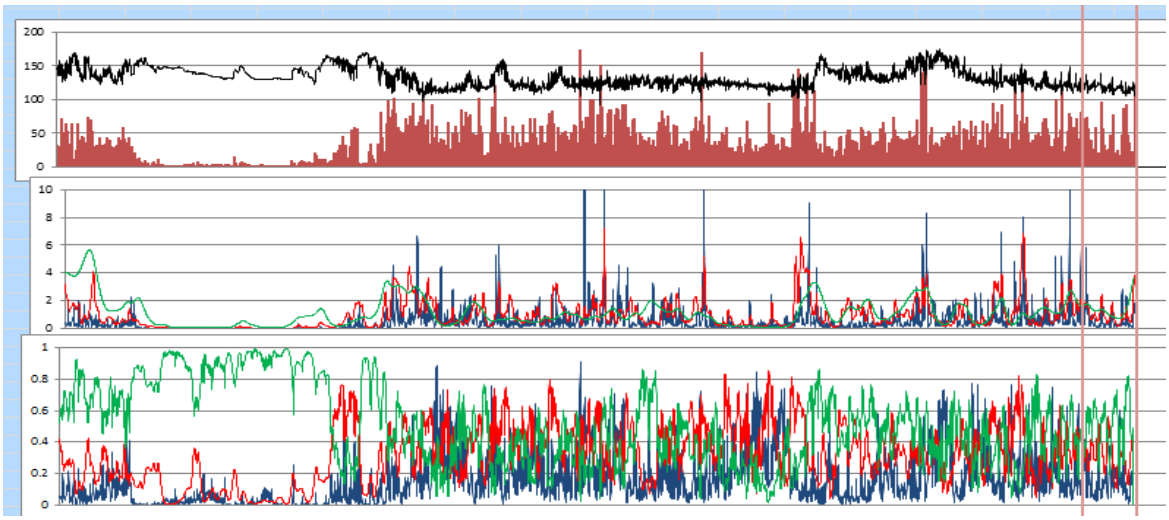


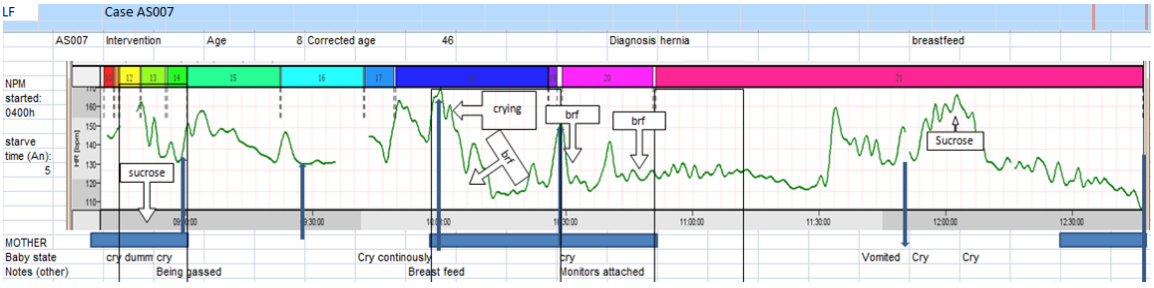
Summary: The normalised HF is high during anaesthesia and surgery and during the periods when the infant was sleeping during resting time. These are sympathetic measure indicating stress

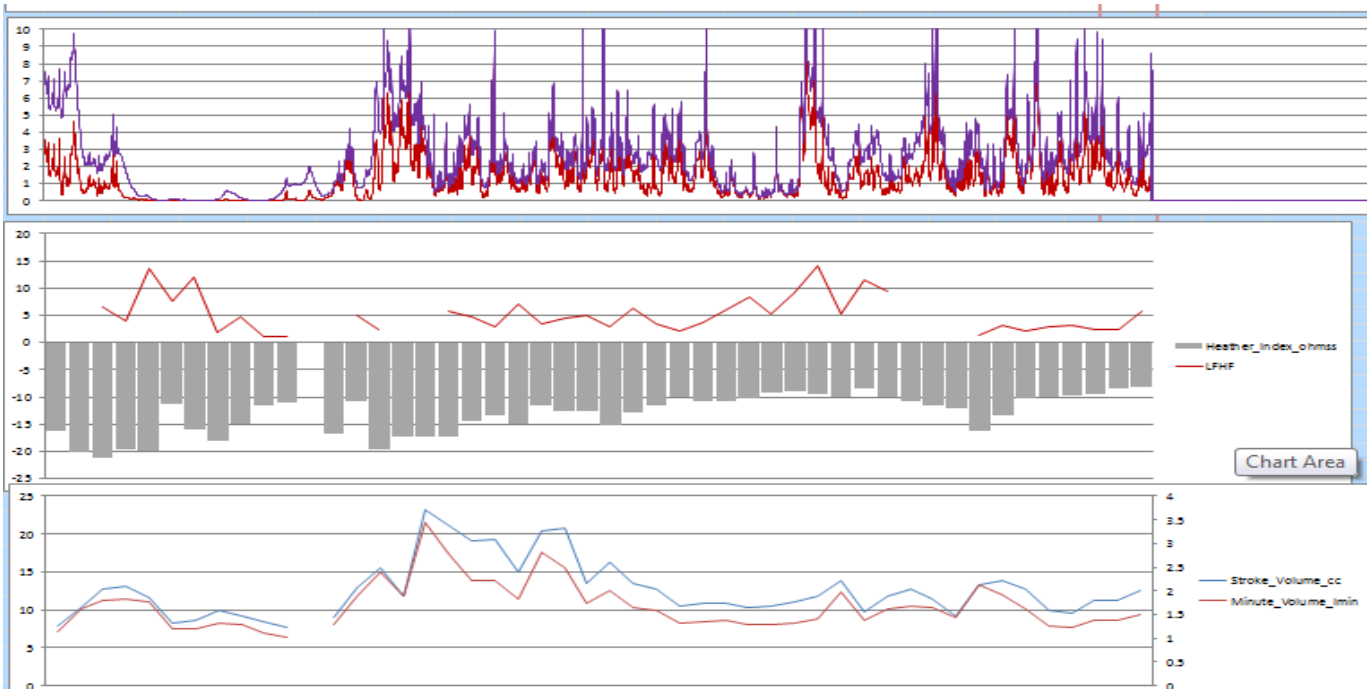
Appendix XII: Intervention Group-Fourth Infant

FOURTH INFANT: AS007Case no 7 INTERVENTION GROUP









Summary: The normalised VLF was high during surgery. When the infant was back on the ward, the mother left the ward and within that time the infant had episodes of increased heart rate, RMSSD and increased heather index but the infant was asleep

Appendix XIII: Collated Data

ID	Label	Correct_age	Randomized	Status_1_2	Average_HR_bpm	RMSSD_msec	LF_ms	HF_ms	LFHF	LVET_msec	Stroke_Volume_cc	Minute_Volume_lmin	Heather_Index_ohmss	RSA_msec	Respiration_Rate_bpm	Analytical_condition	Analytical_conditio
AS001	31	43	Intervention	1	147.7191861	9.231385995	0	0	0	221.8253968	12.83524977	1.896012649	-23.9611625	27.11111111	14.24630735	Preparatory	31
AS001	32	43	Intervention	1	162.1372023	7.231707754	167.2147534	26.88071291	6.220621972	185.7142857	10.41532491	1.688711641	-17.982344	20.84337349	15.72494408	Preoperative	32
AS001	33	43	Intervention	1	163.3267618	3.316433024	1.669008412	0.272665011	6.121094906	182.1428571	7.431684322	1.213792935	-16.6515573	6.005263158	19.16389838	Anaesthesia	33
AS001	34	43	Intervention	1	156.0647159	2.334863511	0.050192343	0.031912558	1.572808502	185.3174603	7.027107473	1.096683531	-18.6273237	3.421383648	20.13701247	Surgery	34
AS001	35	43	Intervention	1	181.6448697	6.003467092	70.61257077	11.06759714	6.38011755	177.3809524	7.367652157	1.338296216	-19.64852251	6.277227723	22.53771159	Recovery	35
AS001	36	43	Intervention	1	207.3524436	2.51913681	3.787207833	0.57657159	6.568495395	193.6507937	10.57138499	2.19200251	-19.04116284	5.423357664	16.13345629	Postoperative	36
AS001	37	43	Intervention	1	207.502174	4.737752471	8.330767418	1.624794847	5.127273412	195.7777778	10.83385644	2.248048764	-16.70539048	6.507874016	16.06804775	Ward	37
AS001	38	43	Intervention	1	189.060342	4.592127618	35.66610341	8.174308644	4.363195099	189.6825397	8.527204655	1.612156228	-17.07009738	11.22175732	17.32557399	Settling_early20	38
AS001	39	43	Intervention	1	170.498159	9.193162823	132.8822058	27.73378394	4.791347842	171.031746	9.072291698	1.546809032	-20.02655665	18.80682927	16.92141737	Settling_later	39
AS002	31	41	Intervention	1													
AS002	32	41	Intervention	1	150.1915481	6.860160184	105.5775985	15.41698999	6.848133039	190.9727626	9.508942375	1.428162776	-9.53811987	18.71186441	16.48802442	Preoperative	32
AS002	33	41	Intervention	1	144.8244849	6.17289466	13.7099526	1.946266506	7.044231897	218.6031746	7.842388215	1.135769834	-9.778169787	10.87280702	16.84475108	Anaesthesia	33
AS002	34	41	Intervention	1	141.3223903	3.427715232	0.965905137	0.243137653	3.972667857	206.3492063	7.213042492	1.019364406	-9.503393866	5.337883959	22.88325913	Surgery	34
AS002	35	41	Intervention	1	155.4917562	5.394635979	39.72887511	9.291070458	4.27602775	199.1904762	8.562509328	1.331399613	-11.44538888	8.67539267	20.70476525	Recovery	35
AS002	36	41	Intervention	1	141.0219815	5.668790555	62.46657212	14.10461202	4.428804709	216.2222222	11.30443616	1.594173987	-11.1418261	13.6779661	16.25461013	Postoperative	36
AS002	37	41	Intervention	1	157.0743928	5.620942542	36.37790371	8.672913834	4.194426973	191.4126984	6.622883699	1.040285435	-8.549301707	13.72850679	15.91065616	Ward	37
AS002	38	41	Intervention	1	154.3868054	4.84377037	44.00550584	4.761607258	9.241733611	188.4920635	7.529503657	1.162456016	-8.855594204	11.67410714	16.04990858	Settling_early20	38
AS002	39	41	Intervention	1	159.8072375	5.893556563	45.86237088	9.134358749	5.020863767	175.3968254	8.587083962	1.372278166	-12.99905651	12.97745209	15.88518936	Settling_later	39
AS003	31	55	Intervention	1	148.532199	13.75487482				185.9922179	8.881418383	1.319176602	-15.38833564	31.5	17.14995217	Preparatory	31
AS003	32	55	Intervention	1	155.1198345	12.11388153	114.3766433	30.33719303	3.770178841	186.540856	8.537335283	1.324310037	-16.65738537	27.77439024	15.52857457	Preoperative	32
AS003	33	55	Intervention	1	133.3321899	9.905757054	8.395561446	7.947829649	1.056333844	211.3151751	7.434508635	0.991259317	-10.23080926	14.23333333	16.93394655	Anaesthesia	33
AS003	34	55	Intervention	1	127.8106186	6.438667761	0.690988117	1.298436181	0.532169487	217.7587549	6.093011456	0.778751563	-9.883035991	9.508009153	16.446548	Surgery	34
AS003	35	55	Intervention	1	164.6509834	5.406779202	64.67545244	5.697844907	11.35086221	208.9494163	6.627440566	1.091214607	-10.51604524	9.341269841	17.59376274	Recovery	35
AS003	36	55	Intervention	1	155.5186504	7.303478877	103.731621	24.42087454	4.247662007	193.385214	9.991220706	1.55382116	-18.94509929	20.86363636	16.29376758	Postoperative	36
AS003	37	55	Intervention	1	155.9697902	7.821175615	139.8694984	29.56441264	4.731008871	200	10.41776802	1.624857093	-19.0487069	22.28378378	15.69274337	Ward	37
AS003	38	55	Intervention	1	132.6506001	13.70878744	220.2740253	56.14686417	3.923175917	213.540856	9.648970049	1.279941667	-15.28143185	30.42918455	16.44463295	Settling_early20	38
AS003	39	55	Intervention	1	138.6474197	16.86205511	160.8217669	52.25055897	3.077895625	212.0972763	9.411316921	1.304854807	-15.77069241	33.01734694	17.12166531	Settling_later	39
AS005	31	48	Control	2													
AS005	32	48	Control	2													
AS005	33	48	Control	2	135.7219838	5.793423344	0.354322987	1.39702196	0.253627357	199.540856	4.510263481	0.612141907	-13.86557649	8.12605042	12.57928523	Anaesthesia	33
AS005	34	48	Control	2	136.3733959	5.101109076	3.589335369	1.803451147	1.990259273	196.9299611	4.07775008	0.556096626	-12.73015437	9.115332429	14.8062443	Surgery	34
AS005	35	48	Control	2	135.0955761	8.15298256	56.87551568	12.61696909	4.507858844	196.8171206	3.421610312	0.462244416	-9.402777011	19.21176471	17.09880349	Recovery	35
AS005	36	48	Control	2	123.3122201	13.373261	187.6128567	49.73661447	3.77212761	216.0972763	5.161924138	0.636528325	-13.62945762	23.23045267	23.25558772	Postoperative	36
AS005	37	48	Control	2	142.1794149	9.76478641	36.08954567	23.33683334	1.546462845	181.2645914	5.183421492	0.736975835	-12.35107064	18.22981366	16.60793342	Ward	37
AS005	38	48	Control	2	144.0972832	7.652080265	53.1089749	19.65734875	2.701736413	223.8210117	7.471376469	1.076605051	-12.91920933	15.51612903	15.96587822	Settling_early20	38
AS005	39	48	Control	2	153.7129723	7.14253574	65.46015323	17.80451559	3.676603999	217.2101167	9.238706709	1.420109069	-16.22572733	17.11774744	15.70688822	Settling_later	39
AS006	31	43	Control	2													
AS006	32	43	Control	2	164.6364488	5.517717667	63.92524342	14.07721842	4.541042237	193.4241245	12.56209486	2.068178686	-33.2412426	12.91386861	17.07992901	Preoperative	32
AS006	33	43	Control	2	166.144118	0.92670625	0.255380798	0.022850821	11.17600078	183.7548638	9.308890837	1.546617458	-23.93651949	1.673202614	32.71812199	Anaesthesia	33
AS006	34	43	Control	2	158.4207208	1.144820739	0.118579309	0.026112195	4.541146712	189.9766537	11.2555476	1.783111964	-21.9722358	2.195876289	28.32828435	Surgery	34
AS006	35	43	Control	2	205.3273688	1.443131951	0.130590154	0.064017958	2.039898772	188.1478599	12.6565196	2.598729868	-34.80428261	2.470779221	24.86794536	Recovery	35
AS006	36	43	Control	2	206.6582529	1.346066448	5.863876681	0.376378904	15.57971665	151.7509728	13.34829669	2.758535673	-50.34325551	3.250554324	16.67366973	Postoperative	36
AS006	37	43	Control	2	171.6229443	7.39593223	230.989565	24.10513019	9.582589399	168.536965	18.73764004	3.215808953	-45.98902082	13.14285714	16.22046566	Ward	37
AS006	38	43	Control	2	155.0096398	6.725428535	230.7960473	22.53183159	10.24311079	182.9805447	15.58370935	2.415625173	-34.75097379	15.32380952	16.5392234	Settling_early20	38
AS006	39	43	Control	2	160.1267718	6.371036157	149.9613868	18.93266954	7.920773479	198.6498054	13.82038555	2.213013724	-37.00154661	16.075	16.04570608	Settling_later	39
AS007	31	46	Intervention	1	144.9988589	7.357051963				216.3735409	7.247305324	1.050851002	-12.14577795	17.2	14.93676522	Preparatory	31
AS007	32	46	Intervention	1	144.9842333	7.49700795	108.0222125	18.69220448	5.77899801	217.0389105	11.40154959	1.653044925	-20.26874534	22.08080808	14.98718393	Preoperative	32
AS007	33	46	Intervention	1	142.6219136	5.362592326	19.95824294	5.386116377	3.705497904	218.536965	9.55626304	1.362932522	-16.05465324	10.10526316	18.7260726	Anaesthesia	33
AS007	34	46	Intervention	1	133.8250666	3.73753813	0.175113537	0.140142028	1.249543337	228.0894942	8.409042047	1.125340612	-12.05882744	5.637795276	15.95945055	Surgery	34
AS007	35	46	Intervention	1	148.2236249	7.403879501	81.58066676	34.83838042	2.341689417	202.3346304	11.86192361	1.758217315	-14.46037623	10.25	15.13684685	Recovery	35
AS007	36	46	Intervention	1	131.6124014	14.44452224	244.5725187	61.11004825	4.002165367	255.2062257	18.70711597	2.462088456	-14.59654046	28.34754098	16.05938542	Postoperative	36
AS007	37	46	Intervention	1	126.6596942	12.08077453	245.5963949	63.72110083	3.854239675	240.540856	16.66537219	2.110830945	-13.69069516	28.79295154	16.29424709	Ward	37
AS007	38	46	Intervention	1	125.377816	9.562070734	212.9655529	51.89317694	4.103922046	231.4280156	11.35453587	1.423606909	-10.72840819	22.85714286	17.79474069	Settling_early20	38
AS007	39	46	Intervention	1	132.191279	12.2222617	269.2453369	50.32606411	5.350017762	226.7042802	11.2572899	1.48811555	-9.63069201	23.81296992	19.64180745	Settling_later	39
AS008	31	40	Control	2													
AS008	32	40	Control	2	166.2060066	3.821989274	36.60145142	7.800563342	4.692154889	201.536965	7.314583004	1.215727631	-19.59316613	11.59459459	16.51787302	Preoperative	32
AS008	33	40	Control	2	149.2716011	3.635950867	26.9636884	3.399704121	7.931186786	224.7587549	5.976932501	0.892186284	-12.66450146	7.188481675	16.67952577	Anaesthesia	33
AS008	34	40	Control	2	136.3122953	2.438908744	0.814018318	0.61903722	1.314974757	222.5291829	4.19068929	0.571242476	-8.766718454	3.031365314	19.92920827	Surgery	34
AS008	35	40	Control	2	148.1025508	5.996210925	270.9307193	11.06366537	24.48833278	237.0389105	6.062491185	0.897870409	-11.91037255	11.53787879	15.85031412	Recovery	35
AS008	36	40	Control	2	146.78547	6.177490461	56.89804111	18.39870948	3.092501742	241.8132296	7.38385427	1.08384252	-11.2997914	14.69607843	16.94595782	Postoperative	36
AS008	37	40	Control	2	142.574174	5.322705693	67.26510262	15.2742171	4.403833085	238.9805447	7.62996282	1.087835647	-11.47924941	14.91954023	16.25467949	Ward	37
AS008	38	40	Control	2	150.0560272	4.324947544	20.77730707	7.270550749	2.857734961	226.6498054	8.638524622	1.296262686	-13.29598225	10.1659919	16.53839345	Settling_early20	38
AS008	39	40	Control	2	143.6869898	6.459320309	101.4484307	24.03028657	4.221690424	230.5953307	8.715525703	1.252307652	-18.65303397	19.11368653	15.43659969	Settling_later	39
AS009	31	46	Control	2	156.8652738	9.451523495	201.610946	29.41058902	6.855046181	208.6498054	14.73232945	2.310990894	-17.9869495	23.65819209	17.47328018	Preparatory	31
AS009	32	46	Control	2	194.2382551	7.575099884	85.02655635	15.87219038	5.356951644	191.3190661	15.70049719	3.049637177	-20.42209612	17.64779874	16.95828412	Preoperative	32
AS009	33	46	Control	2	172.8880694	1.857328316	0.750937007	0.064234214	11.69060783	189.4824903	14.2407755	2.462060182	-21.73339966	2.782051282	25.08366654	Anaesthesia	33
AS009	34	46	Control	2	164.3490725	0.797358791	0.029106071	0.011055867	2.632635728	192.6070039	14.28575511	2.347850603	-19.84918105	1.191099476	28.42630686	Surgery	34
AS009	35	46	Control	2	185.2300343	2.903115945	9.168811741	1.957542905	4.683836926	160.9805447	12.28242289	2.275073613	-22.05570314	5.728323699	19.42216817	Recovery	35
AS009	36	46	Control	2	155.7017054	8.988999013	117.8530691	30.26631592	3.893868993	201.1478599	12.82816443	1.997367078	-19.6888209	23.51470588	17.94931407	Postoperative	36
AS009	37	46	Control	2	158.4829099	8.345180701	139.8064311	23.17499887	6.032640254	205.3696498	13.32434851	2.111681524	-19.81919068	21.76356589	15.45913037	Ward	37
AS009	38	46	Control	2	159.9853934	8.079095061	61.08898844	17.43593309	3.503625996	217.151751	18.07867168	2.8923234	-17.28904724	18.31512605	15.45652035	Settling_early20	38
AS009	39	46	Control	2	151.6152171	12.29892322	111.8159668	34.49473644	3.241537066	229.8754864	15.04012763	2.280312215	-14.09461104	25.98333333	16.66232747	Settling_later	39

Appendix XIV: STATA do file

Commands

```
sheet("Data_long") firstrow

bysort Label  Randomized : tabstatAverage_HR_bpm, stats (count min max p50)

bysort Label  Randomized : tabstatRMSSD_msec, stats (count min max p50)

bysort Label  Randomized : tabstatLF_ms, stats (count min max p50)

bysort Label  Randomized : tabstatHF_ms, stats (count min max p50)

bysort Label  Randomized : tabstat    LFHF, stats (count min max p50)

bysort Label  Randomized : tabstatLVET_msec, stats (count min max p50)

bysort Label  Randomized : tabstatStroke_Volume_cc, stats (count min max p50)

bysort Label  Randomized : tabstatMinute_Volume_lmin, stats (count min max p50)

bysort Label  Randomized : tabstatHeather_Index_ohmss, stats (count min max p50)

bysort Label  Randomized : tabstatRSA_msec, stats (count min max p50)

bysort Label  Randomized : tabstatRespiration_Rate_bpm, stats (count min max p50)

bysort Label  Randomized : tabstatAnalytical_condition_Code, stats (count min max p50)
```

Appendix XV: Human Research Ethics Committee Approval Letter

UNIVERSITY OF CAPE TOWN

Faculty of Health Sciences
Human Research Ethics Committee
Room E52-24 Groote Schuur Hospital Old Main Building
Observatory 7925
Telephone [021] 406 6338 • **Facsimile** [021] 406 6411
e-mail: shuretta.thomas@uct.ac.za
Website: www.health.uct.ac.za/research/humanethics/forms

28 June 2013

HREC REF: 373/2013

Ms L Ssenyonga
c/o A/Prof M Coetzee
Nursing & Midwifery
2nd floor, ICH Building, Room 201
Red Cross War Memorial Children's Hospital

Dear Ms Ssenyonga

PROJECT TITLE: DOES DECREASING MATERNAL SEPARATION OF UNDER 6-MONTH OLD INFANTS DIRECTLY BEFORE AND AFTER SURGERY DECREASE THEIR ALLOSTATIC LOAD AND IMPROVE OUTCOMES? A RANDOMISED CONTROL TRIAL

Thank you for submitting your study to the Faculty of Health Sciences Human Research Ethics Committee for review.

It is a pleasure to inform you that the HREC has **formally approved** the above-mentioned study.

Approval is granted for one year till the 30th June 2014

Please submit a progress form, using the standardised Annual Report Form if the study continues beyond the approval period. Please submit a Standard Closure form if the study is completed within the approval period.
(Forms can be found on our website: www.health.uct.ac.za/research/humanethics/forms)

Please note that the ongoing ethical conduct of the study remains the responsibility of the principal investigator.

Please quote the HREC. REF in all your correspondence.

Yours sincerely

pp TuBurgess

PROFESSOR M BLOCKMAN
CHAIRPERSON, FHS HUMAN ETHICS
Federal Wide Assurance Number: FWA00001637.
Institutional Review Board (IRB) number: IRB00001938
This serves to confirm that the University of Cape Town Human Research Ethics Committee complies to the Ethics Standards for Clinical Research with a new drug in patients, based on the Medical Research Council (MRC-SA), Food and Drug Administration (FDA-USA), International Convention on Harmonisation Good Clinical Practice (ICH GCP) and Declaration of Helsinki guidelines.

s.thomas

HREC Ref no 373/2013 – 28Jun2013
The Human Research Ethics Committee granting this approval is in compliance with the ICH Harmonised Tripartite Guidelines E6: Note for Guidance on Good Clinical Practice (CPMP/ICH/135/95) and FDA Code Federal Regulation Part 50, 56 and 312.