SEPSIS ASSOCIATED ENCEPHALOPATHY IN EXTREMELY LOW GESTATIONAL AGE NEONATES

BY

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LIST OF ABBREVIATIONS

aEEG	amplitude-integrated electroencephalography
CNS	central nervous system
EEG	electroencephalography
ELBW	extremely low birth weight
ELGAN	extremely low gestational age neonate
GEE	generalized estimating equations
HFV	high frequency ventilation
IFN	interferon
IL	interleukin
IVH	intraventricular hemorrhage
LPS	lipopolysaccharide
MRI	magnetic resonance imaging
NEC	necrotizing enterocolitis
NICU	neonatal intensive care unit
PMA	postmenstrual age
PVL	periventricular leukoencephalopathy
SAE	sepsis associated encephalopathy
TNF	tumor necrosis factor
WMD	white matter damage

Abstract

Sepsis in premature infants is associated with long term adverse neurodevelopmental outcome. There have been no previous studies to assess the acute changes in brain function during sepsis that may lead to long term adverse neurologic outcomes. The goal of this study was to identify acute changes in brain function during sepsis in premature infants through the use of amplitude-integrated electroencephalography (aEEG). This study was a prospective observational study of 108 premature infants born at less than 28 weeks gestation. aEEG recordings were performed once monthly until 36 weeks postmenstrual age or discharge as well as an additional aEEG recording during their first episode of sepsis. All recordings were assessed for the presence of burst suppression and a maturition score was assigned. Burst suppression occurred more frequently during acute sepsis (OR 2.4, p=0.01), but the rate of aEEG maturation was not different between infants with and without sepsis. We conclude that sepsis is associated with acute encephalopathy, but does not alter the rate of brain wave maturation.

CHAPTER 1: BACKGROUND

Introduction

Advanced technology and neonatal intensive care have greatly improved the survival of extremely premature infants (1). However, these infants are at increased risk for morbidities such as respiratory distress syndrome, chronic lung disease, sepsis and adverse neurologic outcomes (1). Recent studies of extremely low gestational age neonates (infants born at less than 28 weeks gestation, ELGANs) continue to show a high prevalence of neurodevelopmental impairment among these infants, despite trends in improvement in outcome over time. Washburn et al found survival of ELGANs to have improved from 67% to 71% during the 1990s. The rate of neurodevelopmental impairment also improved during this time period from 20% to 14%, but it remains a significant and frequently observed morbidity for these infants (2).

Cranial ultrasonography is the tool used most frequently during the neonatal period to predict subsequent neurologic outcome of premature infants. Presence of intraventricular hemorrhage, ventricular dilatation, cystic lesions and cerebral atrophy are all important predictors of adverse neurologic outcomes. While abnormal cranial ultrasounds may predict neurologic impairment, a normal cranial ultrasound does not ensure the absence of neurologic impairment. Almost one quarter of ELGANs with no cranial ultrasound abnormality have delayed mental development at two years of age (3). If head ultrasounds fail to identify many infants with high risk for abnormal neurologic development, then factors other than brain damage detectable with cranial ultrasound could influence the risk of adverse neurologic outcome in premature infants.

Infection and Neurodevelopmental Outcome

Despite advances in antiseptic measures such as hand gels, hand washing protocols and increased attention to proper central line care, sepsis remains a major cause of morbidity and mortality in premature infants (4). Studies from the National Institute of Child Health and Human Development, Neonatal Research Network suggest that up to 25% of infants born less than 1500 grams have at least one positive blood culture during their initial hospitalization (5). Recent studies suggest as many as 65% of extremely low-birth-weight (ELBW) survivors have at least one infection during their hospitalization after birth (6).

Neonatal infections among ELBW infants are associated with poor neurodevelopmental outcomes. Stoll et al reported that infection in ELBW infants was associated with a significant increase in several adverse neurologic outcomes. In this study, a cohort of 6093 ELBW infants were followed to18 months adjusted age. Those infants who were diagnosed with an infection during their initial hospital stay were at greater risk of cerebral palsy, mental retardation and vision impairment (7). They were also more likely to have growth failure ($<10^{th}$ percentile) at follow-up. In addition, even those infants with clinical infection alone (i.e. those without positive blood cultures) had an increased risk for adverse neurologic and growth outcomes. Patients with necrotizing enterocolitis (NEC) were also included because of the association between NEC and sepsis, and both of these illnesses are associated with inflammation. Patients with NEC and sepsis had the greatest risk of adverse neurologic and growth outcomes.

The high incidence of infection among premature infants and its association with adverse neurodevelopmental outcome prompt further exploration of the possible causal relationship between sepsis and neurodevelopmental impairment and the pathways involved.

A Conceptual Model

Because sepsis is common in premature infants and is associated with neurologic impairment, it is important to elucidate the mechanism by which sepsis is associated with adverse outcome in an effort to improve outcomes. At least three possible mechanisms can be proposed: 1) The association could be explained by confounding variables, such as the degree of prematurity (i.e., the gestational age) which are associated with both sepsis as well as adverse neurological outcome; 2) Sepsis could also lead to morbidities, such as chronic lung disease, which in turn lead to adverse outcome; 3) Sepsis could directly damage the brain through an inflammatory pathway. Animal studies support the role of an inflammation as the pathway linking sepsis and brain damage and implicate cytokines as having a damaging effect on white matter (8). While such studies cannot be directly reproduced in humans, indirect measures of inflammation and brain injury have been associated in humans. If the second and third mechanisms proposed above are operative, then prevention of sepsis and attenuation of brain inflammation could lead to the prevention of neurologic impairment. An understanding of the mechanism by which sepsis is associated with neurologic impairment is an important aspect in the search for treatments to improve outcomes of premature infants.

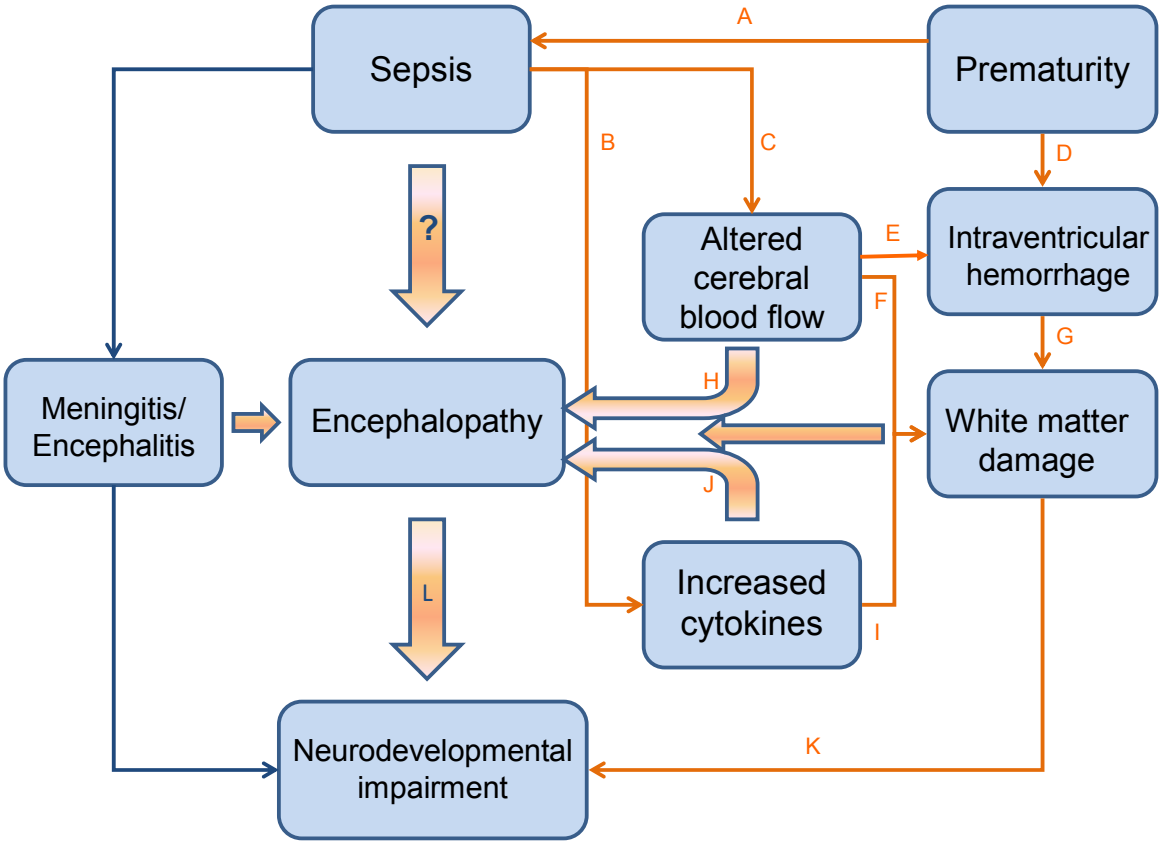


Figure 1: Conceptual Model depicting multiple possible pathways linking sepsis and neurodevelopmental impairment. The arrow marked by a question mark is the association of interest. There are multiple pathways through which sepsis may lead to an acute encephalopathy prior to the ability to identify neurodevelopmental impairment. Evidence for each pathway is identified by a letter with citations below.

A. Prematurity increases risk for sepsis (9).

In a report from the National Institute of Child Health and Human Development Neonatal Research Network, the rate of infection among hospitalized very low birth weight infants was inversely related to gestational age at birth.

B. Sepsis leads to increase in systemic cytokines (10).

Harris et al measured plasma and tracheal aspirate (TA) levels of interleukin-8 (IL-8), epithelial neutrophil activating peptide (ENA-78), IL-10, and IL-18 in 84 neonates with suspected sepsis or NEC. Thirty-one infants had bacterial sepsis, 19 had NEC, and 34 infants with negative results on cultures had sepsis syndrome. Plasma and TA cytokine levels were elevated in critically ill infants

with bacterial sepsis or NEC compared with those in infants with sepsis syndrome.

C. Sepsis is associated with alterations in cerebral blood flow (11).

Hypotension is often a presenting sign of sepsis among premature infants. In a study using continuous bedside recordings of mean arterial pressure (from an indwelling arterial catheter) and cerebral perfusion (using the near-infrared spectroscopy Hb difference signal) to detect cerebral pressure-passivity in the first 5 days after birth in infants with birth weight <1500 g, an association was found between prematurity, hypotension and cerebral blood flow.

D. Premature infants are at higher risk for intraventricular hemorrhage (IVH) (12).

Rates of IVH vary among medical centers and regions. In order to further understand this phenomenon, the rates of IVH in were determined in 5712 infants of 24-30 weeks gestation born from 1995 to 1997 in Australia and New Zealand. Significant antenatal and perinatal variables for major IVH in 1995 and 1996 were identified by univariate and multivariate analysis. Multiple antenatal and perinatal characteristics had significant association with major IVH including gestational age at birth.

E. Alterations in cerebral blood flow may result in intraventricular hemorrhage (13).

Predicting infants at highest risk for IVH is essential for prevention of this morbidity. In a study of 24 premature infants, cerebral blood flow was measured during the first 24 hours of life using near infrared spectroscopy. The infants then had cerebral ultrasound examination performed to define the maximum extent of IVH. Cerebral blood flow was significantly lower in the infants with IVH than those without hemorrhage despite there being no difference in carbon dioxide tension and a higher mean arterial blood pressure. On subgroup analysis, those infants with severe IVH had the lowest cerebral blood flow.

F. Alterations in cerebral blood flow may directly cause white matter damage (14).

Based upon findings in newborn dogs, Young and others found selective hypoperfusion of cerebral white matter with preservation of blood flow to cerebral gray matter during severe hypotension. They proposed this as a mechanism by which sepsis and its associated hypotension leads to periventricular leukomalacia.

G. Intraventricular hemorrhage can lead to white matter damage (15).

In a study of 1605 infants weighing 500 to 1500 g at birth, Kuban and others performed serial cranial ultrasonography to assess IVH and white matter damage (WMD). They found that IVH and ventriculomegaly were strong predictors of WMD occurrence.

H. Alterations in cerebral blood flow may cause acute encephalopathy (16).

Greisen and Pryds measured cerebral blood flow during the first 48 h of life in 20 infants, born after 27 to 33 weeks of gestation, who were under mechanical ventilation and being monitored by amplitude integrated electroencephalography (EEG). Among these infants, cerebral blood flow was related to the level of EEG activity, discontinuous EEG activity being associated with low cerebral blood flow.

I. Increased systemic cytokines may result in white matter damage (17).

In a study examining the relationship between cerebral damage, intrauterine antigen exposure and inflammation in 50 infants who were born at 23-29 weeks' gestation, higher concentrations of cytokines (TNF-alpha, and interleukins, 1beta, 6, and 10) in umbilical blood predicted cerebral lesions detected by magnetic resonance imaging very soon after delivery.

J. Elevated cytokines are associated with acute encephalopathy (18).

Ichiyama and others compared the concentrations of interferon-gamma (IFN-gamma), TNF-alpha, interleukin-2 (IL-2), IL-4, IL-6, IL-10, and soluble TNF receptor 1 (sTNFR1) in serum and cerebrospinal fluid (CSF) in 15 infants with acute encephalopathy to 12 with febrile seizures associated with HHV-6 infection. The serum IL-6, IL-10, sTNFR1, CSF IL-6, and sTNFR1 levels of infants with encephalopathy who had neurological sequelae were significantly higher than those with febrile seizures. In acute encephalopathy, serum IL-6, sTNFR1, and CSF IL-6 levels in infants with neurological sequelae were significantly higher than those without neurological sequelae.

K. White matter damage is an important predictor of neurodevelopmental impairment (3).

In a study of 1017 children born at less than 28 weeks' gestation, O'Shea and others evaluated associations between ultrasound-defined lesions of the brain and developmental delays at 24 months' corrected age. They found that focal white matter damage, as characterized by echolucent/hypoechoic lesion, and diffuse damage, as suggested by late ventriculomegaly, are associated with delayed mental and psychomotor development

L. Encephalopathy can predict adverse neurologic outcome (19).

Hellstrom-Westas and others report on their experience with amplitude-integrated EEG in preterm infants. They state that EEG is a sensitive method for detection of brain injury in preterm infants. Although the acute and chronic EEG changes are mainly non-specific regarding type of damage, they correlate with later neurological and cognitive function.

Animal Models

Several animal models support the association between sepsis and neurologic damage. Multiple pathways are implicated including cytokine-mediated white matter damage as well as direct endotoxin effects.

Observation of white matter damage precipitated by gram-negative bacteremia without intracranial infection prompted laboratory studies of the effect of lipopolysaccharide (LPS) on the developing brain (8). LPS, or endotoxin, is a structural component of the cell wall of gram-negative bacteria. It is a potent inducer of the synthesis of proinflammatory cytokines and other mediators of systemic inflammation, which can, in turn, lead to shock and death. In neonatal kittens, peritoneal exposure to LPS results in a telencephalic leukoencephalopathy characterized by astrogliosis and necrosis (20). Further studies in pigs with E. coli sepsis showed severe brain damage despite the absence of hypoxia and ischemia (21). Histologic examination revealed perivascular edema and macrophage-like cells which resulted in spongiform degeneration in the white matter. Similar findings have been observed in preterm fetal sheep after systemic exposure to LPS (22). White matter necrosis has also been produced in neonatal dogs by both intraperitoneal as well as subcutaneous injection of LPS (14;23)

More recently, tumor necrosis factor - alpha (TNF-α) has been implicated in cerebral signaling during systemic inflammation. A mouse model was used to identify the pathway for communication between the periphery and the brain during inflammatory processes. In mice with hepatic inflammation, there was a significant infiltration of activated monocytes into the brain. TNF-α activated microglia expressed cerebral monocyte chemoattractant protein-1 leading to monocyte infiltration of the central nervous system (CNS). This pathway is a novel immune system-to-CNS communication occurring in the setting of peripheral organ inflammation (24).

Human Studies

While animal models are helpful in understanding possible pathways between sepsis, systemic inflammation and neurologic damage, human fetuses may differ in their response to sepsis. Similar studies in humans are not ethical; however, measures of inflammatory mediators and non-invasive assessments of brain structure or function might provide collaborating evidence.

The finding by Stoll et al that sepsis and NEC combined were associated with worse outcomes than sepsis alone might be the result of a dose dependent relationship between cytokines, inflammation and brain damage. Cytokine levels and patterns vary with disease in infants. Those with bacterial sepsis or NEC have higher plasma IL-8 and IL-10 levels than infants with suspected sepsis alone (10). Infants with both sepsis and NEC have higher plasma IL-6 levels than infants with sepsis alone (25). These findings suggest that the systemic inflammatory response produced by sepsis and NEC varies in magnitude by disease and could explain the differences in outcome experienced by infants who develop NEC and sepsis. This dose response relationship supports a causal link between sepsis, cytokine elevation and adverse neurologic outcome.

Further studies in infants assessing the relationship between cytokines and brain damage suggest that intrauterine exposure to cytokines results in brain lesions on magnetic resonance imaging (MRI). Infants with higher concentrations of cytokines such as TNF- α and IL-1, IL6 and IL-10 in their umbilical cord blood are more likely to exhibit abnormalities on cerebral MRI. These lesions include intraventricular hemorrhage as well as periventricular lesions. This evidence implies that cytokines are important mediators in the pathway between neonatal inflammation and cerebral damage leading to neurologic impairment (17). Current studies by the National Institute for Child Health and Human Development Neonatal Research Network and the Extremely Low

Gestational Age Newborn Study Investigators are in progress to identify specific cytokines predictive of abnormal neurodevelopment among premature infants.

Postnatal sepsis has also been directly linked to noncystic periventricular white matter damage on MRI. Shah et al evaluated 192 infants born at less than 30 weeks' gestation. They found that 35% of these infants had at least one episode of sepsis, the majority of which were caused by coagulase-negative staphylococci, 6% of the cases also had NEC and another 2% had NEC alone. White matter abnormalities consistent with noncystic periventricular leukomalacia (PVL) were present in 80% of those infants with sepsis and/or NEC (26). At age 2 years, these infants with sepsis and/or NEC had lower BSID-II Mental Development Index and Psychomotor Development Index scores even after adjustment for other neonatal characteristics associated with adverse neurologic outcomes.

While the evidence for an association between sepsis and NEC and neurologic outcomes continues to accumulate, to date there have been no studies of the acute effects on the premature infant brain at the time of sepsis.

Sepsis Associated Encephalopathy

Some adults with sepsis develop diffuse cerebral dysfunction, a condition referred to as sepsis-associated encephalopathy (SAE) (27;28). Up to 70% of patients with sepsis develop SAE (29). SAE precedes dysfunction of other organ systems. It is distinguished from meningitis and encephalitis in that it is not related to a direct infection of the central nervous system. In adults, SAE presents with altered mental status. Although reversible in some cases, SAE worsens the overall prognosis (30). The diagnosis can be made by electroencephalography (EEG) which has been found to be more sensitive than clinical exam for this disorder (31). Specific characteristics of the EEG allow grading of the severity of cerebral dysfunction and prediction of outcome.

These include predominant delta, triphasic waves and burst suppression. Specifically, burst suppression is seen in the more severe cases and is associated with a worse prognosis (32). In a study of EEG and clinical features of septic encephalopathy in adults, Young et al found that EEG was a sensitive index of brain function in septic encephalopathy and that it was a valuable tool in evaluating and monitoring intensive care patients with sepsis (33).

Proposed mechanisms for SAE include toxins from microorganisms, cytokines or inflammatory mediators (34). Alterations in cerebral blood flow have also been implicated, but no specific patterns have been identified. The only known intervention for SAE is treatment of the underlying infection, however 50% of cases of adult sepsis are culture-negative, making therapy targeted at a specific organism impossible (35).

Diagnosis of Encephalopathy

Acute neurologic changes during sepsis have not been described in premature infants as they have in adults. While EEG has proven to be helpful in the diagnosis of encephalopathy related to infection in adults (36), there is currently no diagnostic tool to detect alterations in brain function during episodes of sepsis in premature infants, nor have such alterations been adequately described. The EEG, however, has been used to evaluate the effects of acidosis, a common finding in sepsis, on cerebral function (37). Eaton et al measured EEG activity in 14 ventilated infants with gestational ages ranging from 24-32 weeks during 32 episodes of significant acidosis. All episodes of acidosis were associated with deterioration of the EEG tracing as evidenced by increased discontinuity and loss in the variability of EEG discontinuity. In the majority of cases, as acidosis improved, the EEG returned to normal. However, in 11 episodes, the EEG activity improved, but did not return to its previous level of activity. Acute changes in

EEG activity have also been observed during changes in cerebral blood flow and administration of surfactant via endotracheal tube in premature infants (38;39).

Amplitude-Integrated Electroencephalography

While EEG may be useful in diagnosis and prognosis related to cerebral function, it is cumbersome and expensive. It also relies on the availability of qualified personnel to both administer and interpret the tracing. These features limit its feasibility for use in evaluation of neonates with sepsis. A more cost-effective and less time and labor-intensive method of analyzing cerebral function of the neonate may be accomplished by amplitude-integrated electroencephalography (aEEG). Advantages include ease of use, compact and portable equipment and relatively uncomplicated interpretation. Toet et al compared EEG with aEEG in neonates with hypoxic ischemic encephalopathy or suspected seizure activity. They found that aEEG was a reliable tool for monitoring both background patterns, especially normal and severely abnormal, and ictal activity (40). It has also been shown, when assessing neonatal encephalopathy in term infants, to have excellent interobserver agreement (41).

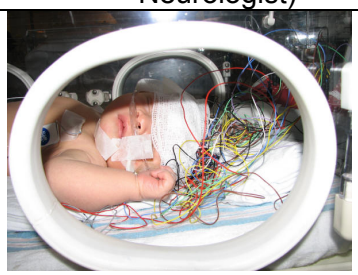
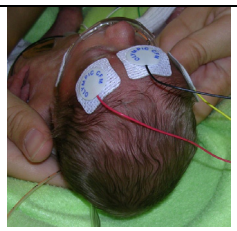
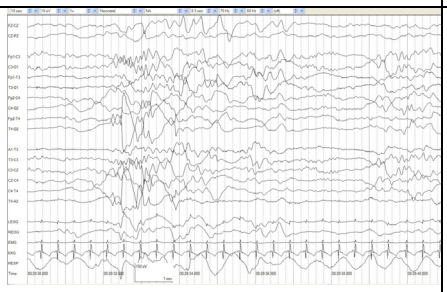
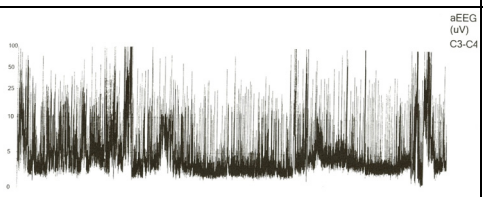
Characteristic	EEG	aEEG
Equipment required	20 leads, monitor, computer, stand, video camera	3 leads, integrated screen and computer atop rolling stand
Training required	Tech: course on lead placement and basic interpretation Interpreter: Subspecialty fellowship in Neurology, could further specialize in EEG	Research assistant: Basic instruction on 3 lead placement Interpreter: 1 day course or literature review with reference to atlas of aEEG
Output	20+ pages of 10 channel recordings	1-2 pages of condensed 1 channel recording
Time for preparation	20-30 minutes	5-10 minutes
Time for interpretation	20-30 minutes (delayed based on availability of Pediatric Neurologist)	Less than 2 minutes
Complexity		
Output		

Table 1: Comparison of EEG and aEEG showing benefits of aEEG as used routinely to monitor brain wave activity in premature infants

In addition to use in patients with HIE and seizures, aEEG has shown promise in evaluating brain function and maturation in premature infants (42-44). Olischar et al have published reference values for aEEG activity in premature infants less than 30 weeks gestation (45). A more complete scoring system developed by Burdjalov et al. has been used to assess brain maturity in premature infants born at <29 weeks gestation. In this system, four components are assessed: continuity, presence of cyclic changes in electrical activity, degree of voltage amplitude depression, and bandwidth. Each component is scored individually within the range of zero to five, depending on the component. For example, continuity has a maximum score of two while the range for cycling is zero to five. All component scores are then summed to yield a total score ranging from 0 to 13. Scores have been shown to increase linearly with increasing gestational age from 24 to 37 weeks ($R=0.84$, $p<0.001$) with an average increase of one point per week (46).

Brain maturation is a more gradual process while sepsis is associated with more acute clinical changes that usually resolve with time. Recent studies have shown characteristic changes in aEEG corresponding to acute changes in clinical status or management. For example, antiepileptic drugs can cause burst suppression and hypoglycemia has also been shown to alter aEEG tracings (47;48).

These findings suggest that aEEG may be a reliable, efficient and accurate tool for monitoring premature infants and detecting alterations related to neonatal encephalopathy. Use of this tool could enable detection of acute changes in brain waves associated with neonatal sepsis which could lead to adverse neurologic outcome among these infants. It could also be useful in observing changes in maturation patterns among premature infants that would help to predict those infants at highest risk for neurologic impairment.

Limitations of the Current Literature

Despite multicenter studies defining an association between neonatal infection and neurologic impairment as well as adult studies specifically describing acute brain effects of sepsis, no study has attempted to address the acute effects of sepsis on the premature infant brain. A few studies have attempted to examine acute effects of metabolic disturbances such as acidosis and hypoglycemia on neonatal brain function, but only through the use of full array EEG. Routine use of EEG in extremely premature neonates is not feasible because of concerns of size, skin integrity and interruptions in infant care. The relatively new tool, aEEG has proven successful in detecting changes in brain function in term infants and brain maturity in preterm infants, but it has not been used in acute sepsis in any infants to characterize disturbances of normal brain function or for prediction of neurologic outcome after neonatal sepsis.

Summary and Significance

Concern regarding the neurologic effects of sepsis prompts exploration into the characteristics and extent of the acute alterations in cerebral function during neonatal infections. Survival rates are increasing for premature infants, but the rate of adverse neurologic outcomes among survivors remains high. The aEEG has proven helpful in evaluation of hypoxic ischemic encephalopathy and seizure activity and is emerging as a method for evaluation of brain maturation (49-51). As sepsis has been shown to have characteristic EEG findings in adults, and since EEG is comparable to the aEEG in evaluation of cerebral function in neonates, the aEEG may be useful in evaluation of neurologic status in neonates with acute infection. The purpose of this study will be to identify aEEG abnormalities that occur in preterm infants in the acute phase of sepsis.

Goals of This Study

Specific Aim 1: To compare the proportion of aEEG recordings with acute abnormalities during sepsis to that of aEEG recordings collected during a time when the infant does not have sepsis.

Hypothesis: The proportion of aEEG abnormalities will be higher in the group with sepsis than the group without sepsis.

Specific Aim 2: To compare the rate of brain wave maturation, as determined by serial aEEG maturation scores, of infants with an episode of sepsis during their initial hospital stay [birth to discharge from the neonatal intensive care unit (NICU)] to the rate of maturation of infants without sepsis during their hospital stay in the NICU.

Hypothesis: Infants with sepsis will have a slower rate of brain wave maturation than those infants who do not develop sepsis during their initial hospital stay.

CHAPTER 2

SEPSIS ASSOCIATED ENCEPHALOPATHY IN EXTREMELY LOW GESTATIONAL AGE NEONATES

Jennifer B. Helderman, Cherrie D. Welch, T. Michael O'Shea

The following manuscript will be submitted to Pediatrics in August 2009. Stylistic variations reflect the requirements of the journal.

INTRODUCTION

Extremely premature infants are at a high risk for infections, which might explain, in part, their high risk for adverse neurodevelopmental outcomes (52). This is true even for infants with clinical infection, but negative blood cultures (52;53). The acute effects of infection that predispose infants to neurologic impairment are not well characterized.

Up to 70% of adults with sepsis develop diffuse cerebral dysfunction, referred to as sepsis-associated encephalopathy (SAE) (54;55). Unlike meningitis and encephalitis it is not related to a direct infection of the central nervous system. SAE is associated with worse prognosis (55). It is not known whether SAE occurs in preterm infants with sepsis.

The objective of this study was to assess the frequency of sepsis-associated encephalopathy in a cohort of extremely premature infants through the use of amplitude-integrated electroencephalography (aEEG). Our hypothesis was that sepsis would be associated with acute changes in brain function detectable as abnormal aEEG tracings and would result in a delay in brain wave maturation.

METHODS

Study Population

Infants born between 24 and 27 6/7 weeks of gestation and at Forsyth Medical Center in Winston-Salem, North Carolina were eligible for recruitment. Exclusion criteria included major congenital anomalies, congenital viral infection or seizures prior to one week of life. Parents of infants were approached for participation in the study by one week of life. Between November 2005 and March 2008, 251 infants were screened and 110 infants were enrolled.

aEEG Recordings

Amplitude-integrated EEG recordings were performed at 28, 32 and 36 weeks' postmenstrual age (PMA). For those infants who became infected, an additional aEEG recording was performed within 72 hours of initiation of treatment. To obtain the aEEG recordings on the first 12 participants, we used the Component Neuromonitoring System (CNS) Model N-300, by Moberg Research, Inc (Ambler, PA). This is a portable EEG monitor that includes an EEG amplifier, compact-sized computer, flat panel touch screen display, and an isolation transformer. The aEEG is displayed as a trended numeric on a semi-log scale with trends of computed numeric values.

A pair of standard gold-disk EEG electrodes was attached to the scalp parietal areas bilaterally using an EEG-electrode paste after first using Nuprep gel to prepare the area. A reference electrode was placed over the mid-frontal area of the scalp and a ground electrode was placed on an earlobe. Each electrode was secured with a small square of gauze and a thin strip of hypoallergenic skin tape. Continuous impedance measurements were done throughout the recording. Any impedance detected over 10 kOhms was corrected by the technician. The EEG signal was processed by amplification, which is a special filtration algorithm used to attenuate signals below 2 Hz and above 16 Hz, amplify, time compress and rectify the signal. The compressed aEEG signal was recorded using a semilogarithmic scale and run at a speed of 6 cm/hour. Continuous recordings were performed for a 3-8 hour period. From each recording, the most uninterrupted period of at least 3-4 hours duration was used for analysis.

For the subsequent 98 participants, the Olympic Cerebral Function Monitor 6000, Olympic Medical (Seattle, WA) was used to obtain aEEG recordings. This change in monitoring system was necessary to continue following the participants when they were transferred from Forsyth Medical Center to Brenner Children's Hospital. Only one Moberg monitoring system was available and a second machine was needed for

Brenner Children's Hospital. To ensure consistent monitoring within participants, two Olympic monitoring systems were obtained, one for each hospital, and all subsequent participants were monitored with Olympic monitoring systems.

When using the Olympic Cerebral Function Monitor, the skin was prepared as described previously and bifrontal leads were used. With this method, three hydrogel electrodes were placed, one in the frontal midline region as a reference and one 3.75 centimeters from the midline on each side, as close to the coronal plane as possible based on the amount of hair on the infant's scalp.

aEEG Interpretation

The scoring system developed by Burdjalov et al. was used to assign the degree of maturation. In this system, four components are assessed: record continuity, presence of cyclic changes in electrical activity, degree of voltage amplitude depression, and bandwidth. Each component was scored and then all were summed to yield a total score ranging from 0 to 13. Scores have been shown to increase linearly with increasing gestational age from 24 to 37 weeks ($R=0.84$, $p<0.001$) (56). All tracings were scored individually by two evaluators who were blinded to the patient's identification, gestational age and infection status. From each recording, the most uninterrupted period of at least 3-4 hours duration was used for analysis. Tracings with score discrepancies greater than 1 point were adjudicated. A previous study found an inter-coder reliability of 82% and 100% for one and two points of difference between scores, respectively, using this system (57).

The tracings were also assessed for the presence or absence of burst suppression. Burst suppression is pattern characterized by overall decrease in numbers of bursts per hour resulting in a less dense appearing tracing, usually associated with low voltage and increase in bandwidth of bursts.

Definitions of Infection

Sepsis was defined as clinical signs and symptoms of infection and a positive blood culture at >72 hours of age requiring 5 or more days of antibiotic therapy. Culture negative infection was defined as new onset signs and symptoms of infection with a negative blood culture after 72 hours of life and intention to treat for 5 or more days. Necrotizing enterocolitis (NEC) was defined as treatment with antibiotics for NEC for seven or more days and diagnosed and classified according to the system of Bell et al (58). Recordings were not performed unless the infant was at least 14 days of age at the time of infection. This restriction allowed us to compare aEEG recordings from infants of similar gestational age within 2 weeks. If aEEG recordings were performed on infants at 25 weeks adjusted age, there would have been no recordings of infants without infection at a similar gestational age with which to compare. Also, the infection could possibly have been present at birth and would not meet the definition of a hospital-acquired infection.

Management of Infection

All infants were managed according to the discretion of the attending neonatologist based on the clinical and laboratory evidence for infection. Routine practice was to obtain a blood culture in infants with clinical signs and/or symptoms of infection, then to initiate intravenous antibiotics to cover both gram positive and gram negative organisms. In some cases, urine and cerebrospinal fluid cultures were also obtained. After 48-72 hours, the decision to continue antibiotics was based on either a positive culture or strong suspicion of infection despite negative cultures.

Those infants suspected of having NEC on the basis of abnormal abdominal radiograph(s), gastric aspirates or an abnormal abdominal exam were managed according to the discretion of the attending neonatologist. Usual care included obtaining a blood culture and starting antibiotics to cover gram negative and anaerobic organisms.

In addition, enteral feedings were held, a gastric tube was placed to suction and routine labs and abdominal radiographs were obtained. Antibiotics were continued for 7-14 days based on severity of illness and resolution of clinical signs.

Data Analysis

Data were analyzed using SAS (SAS Institute, Cary, NC) version 9.1.3. Group attributes were described as proportions and means/standard deviations. The incidence of burst suppression and the rate of maturation of aEEG scores were compared for infants with and without sepsis. Slopes of maturation of aEEG scores were computed for all infants with at least two non-septic aEEG recordings using linear regression. The slopes of maturation were then compared for infants with and without sepsis during their hospital stay using t-tests. T-tests were also used to compare the slope of maturation of infants with burst suppression during sepsis to infants without evidence of burst suppression during sepsis.

A mixed model approach was also used to compare the rate of aEEG maturation between the groups with and without sepsis. In this model we used sepsis and postmenstrual age at the time of the aEEG recording as fixed effects. The intraindividual correlation was taken into account by using an autoregressive covariance structure. We also introduced an interaction term between sepsis and postmenstrual age to examine the difference in slope between the two groups.

To compare the incidence of aEEG recordings with burst suppression for septic infants and non-septic infants we used generalized estimating equations (GEE), where intraindividual correlation was taken into account by using an autoregressive covariance structure.

Sample size considerations

Based on preliminary aEEG maturity scoring in this cohort to establish inter-rater reliability, we estimated that the standard deviation of rate of aEEG maturity is 0.5 points per week. Assuming a frequency of sepsis of 0.5, and at significant level of alpha = 0.05, our sample of 110 infants provided 88% power to detect a 0.3 point difference in the rate of maturation of aEEG scores in infants with and without sepsis.

RESULTS

A total of 251 infants were screened for participation in the study. Of these infants, 88 parents declined participation, 6 had language barriers to consent, 19 expired prior to consent, 1 was anticipated to expire, 6 were transferred to other hospitals, 5 had congenital abnormalities, 6 had legal guardianship issues, 7 were not approached and one had congenital HSV and one had candidal meningitis. Consent was obtained for 110 infants. Two infants withdrew from the study, one prior to any aEEG recordings and another after 2 aEEGs had been obtained, both because of parental request to withdraw. Post menstrual age at birth ranged from 24 to 27 and 6/7 weeks with a mean of 26 weeks and standard deviation of 1.09 weeks. The only significant difference in baseline variables between the group with sepsis and the group without sepsis was PMA at birth (Table 1). Of the 108 infants who completed the study, 67 (62%) developed sepsis during their initial hospital stay in the intensive care nursery.

Table 1: Clinical Characteristics of Infants with Sepsis and Without Sepsis

	Sepsis (n=67)	No Sepsis (n=41)	p-value
Gestational age, mean (SD)	25.76 (1.07)	26.32 (1.06)	0.009
Birth weight in g, mean (SD)	791 (178)	822 (179)	0.38
Male, %	59.7	46.3	0.18
Antenatal steroids, %	83.6	82.9	0.93
C-section, %	65.7	68.3	0.78
Ethnicity			0.98
Caucasian, %	44.8	65.9	
African-American, %	40.3	19.5	
Hispanic, %	13.4	14.6	
Other, %	1.5	0	
5 min APGAR score <6, %	22.4	19.5	0.82
IUGR, n	4	4	0.47

Twenty-four infants died during their hospital stay. Mortality was significantly lower in the group without sepsis. Three out of 41 infants (7%) died in the group without sepsis and 21 out of 67 (31%) infants died in the group with sepsis (p=0.007). Seventy one infants completed all three aEEG recordings for maturity. Forty two (63%) infants who developed sepsis completed all aEEG recordings and 29 (71%) infants without sepsis completed all aEEG recordings. Of those infants who developed sepsis, 48 (72%) had an aEEG recording performed during sepsis.

A total of 297 aEEG recordings were performed among the cohort. Each infant had between one and four total aEEG recordings. Twenty eight percent of the aEEG recordings showed evidence of burst suppression. Burst supression was found in 22% of aEEG recordings from infants without sepsis and 57% of recordings from infants with sepsis at the time of the recording (odds ratio = 4.2; 95% confidence limits = 2.4, 7.2;p<0.001). After adjustment for postmenstrual age at the time of the recording using GEE model, sepsis remained an important predictor of burst suppression on aEEG

(odds ratio = 2.4; 95% confidence limits =1.2,4.8; p=0.01). When this analysis was separated by monitoring system used, the odds ratio for the association between sepsis and burst suppression was 27.8 (95% confidence limits = 3.95, 195.5;p=0.0008) for recordings performed with the Moberg monitor and 1.8 (95% confidence limits = 0.91,3.4;p=0.09) for recordings performed with the Olympic monitor.

Using a two sample t test, there was no statistically significant difference in the slopes of aEEG maturation between infants with sepsis and those without (p=0.3). Using linear regression, there was also no effect of gestational age at birth on slope of aEEG maturation. There was also no statistically significant difference in the slope of maturation of infants with burst suppression during sepsis to those without burst suppression during sepsis (0.62 and 0.84 respectively, p=0.2).

In the mixed model approach, there was no significant interaction between sepsis and postmenstrual age at the time of the aEEG recording. Sepsis did not affect aEEG maturation over time. There was a linear increase in the rate of maturation over time with an increase of 0.87 points per week of postmenstrual age (Figure 1).

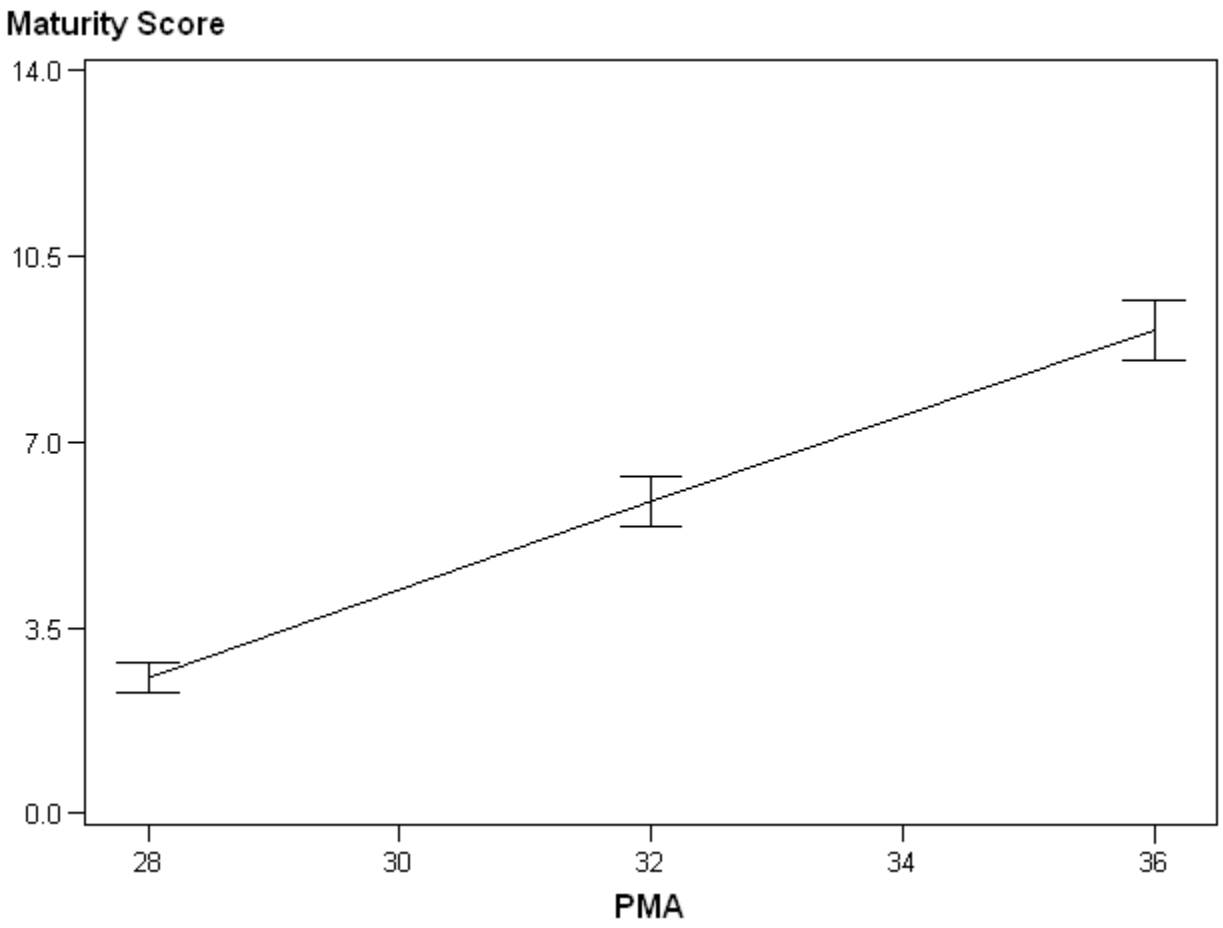


Figure 1: Average aEEG maturation scores at 28, 32 and 36 weeks postmenstrual age.

DISCUSSION

We found that extremely premature infants with sepsis were more likely than non-infected controls to exhibit electroencephalographic burst suppression. This finding suggests that SAE occurs in neonates, as has been described in up to 70% of adults with sepsis(29). In adults SAE has been defined by specific changes on EEG (55). One of these findings, burst suppression, is associated with worse prognosis (59). To our knowledge, ours is the first study that describes this sign of SAE among neonates. We did not anticipate the high prevalence of burst suppression (i.e., 22%) among infants without infection. Our observations suggest that this pattern is related to extreme immaturity, because it was associated with postmenstrual age at the time of the recording. When adjusting for this potential confounder, the association of sepsis and burst suppression persisted.

Research in animal models suggests that inflammatory cytokines might mediate at least some of the neonatal brain damage associated with sepsis. Inflammation of the brain might explain the sepsis-burst suppression association that we observed if, in fact, the association is causal. Increases in systemic cytokines have been associated with white matter injury in both animal models and human studies. In a porcine model, abdominal sepsis elevated cerebrospinal levels of tumor necrosis factor-alpha and interleukin-6 resulting in cerebral edema and death of neurons (21). In very preterm infants higher concentrations of cytokines (TNF-α, and IL-1β, IL-6 and IL-10) in umbilical blood predicted cerebral lesions detected by magnetic resonance imaging very soon after delivery (17;21). In a study of infants born at less than 30 week/s gestation, 35% had at least one episode of sepsis and that white matter abnormalities were present in 80% of those with sepsis and/or NEC (26). Sepsis and NEC also have been associated with developmental impairments such as cerebral palsy, mental retardation and vision impairment (60).

If the pathogenesis of both SAE and cerebral white matter damage involves inflammatory cytokines, then we would expect evidence of white matter injury among infants who exhibited encephalopathy at the time of sepsis. Using aEEG to serially assess brain function, we found no evidence that SAE results in altered brain maturation. Magnetic resonance imaging might be a more sensitive method to detect sequelae of SAE.

While these findings are suggestive that encephalopathy does occur during neonatal sepsis, there are differences in EEG and aEEG. EEG is accepted as the gold standard in bedside measurement of brain function. It has been validated in adults and neonates in predicting outcome (54;61). Despite its complexity, EEG measurement would be a less disputed outcome measurement than aEEG because of the large volume of material published about its clinical and research-oriented use. When used for prediction of abnormal neurologic outcome, EEG has been shown to have a sensitivity and specificity of 100% as compared to magnetic resonance imaging (62). However, premature infants have less surface area for lead placement, often do not tolerate extensive handling require with EEG and have poor skin integrity. Amplitude-integrated EEG has fewer leads and requires minimal time for lead placement. Multiple studies have demonstrated a close association between the aEEG and standard EEG, however exact sensitivity and specificity is only available in direct comparison of aEEG with prediction of neurologic outcome (63).

This study used a maturation system developed by Burdjalov et al (57). In his study, the expected increase in maturation was one point per week. In this study, the average score increased 0.87 points per week. This discrepancy is most likely related to differences in the study populations. While the participants in this study were all born at less than 28 weeks' gestation, the infants in the prior study included infants up to 39

weeks' gestation at birth. The inclusion of more mature infants likely resulted in a healthier population.

Potential limitations of this study should be noted. First, all three aEEG studies were obtained from only 66% of the study sample. Explanations for missing values include death of some infants with sepsis and discharge of infants prior to their undergoing a final aEEG at 36 weeks postmenstrual age. It is reasonable to speculate that had they survived, the infants who died with sepsis would have had slower rates of maturation of their aEEG tracings. Additionally, we can speculate that infants discharged before 36 weeks postmenstrual age were less likely to have had sepsis and would have had more accelerated maturation of their aEEG tracings. On this basis we suggest that the bias due to missing values probably is not an explanation for the sepsis-burst suppression association that we observed but could be an explanation for our failure to detection an association of sepsis and slower aEEG maturation. Second, although a previous study has reported inter-coder reliability of 82% and 100% for one and two points of difference between scores, respectively, using this system, judgments about aEEG maturation scoring are somewhat subjective (57). If, as we expect, the resulting misclassification was non-differential with respect to whether the infant had sepsis, the bias would be towards the null. Third, the outcome that we studied, aEEG tracings, might not be reflective of clinically significant neurologic outcomes. For this reason, we are following the study participants to obtain neurodevelopmental assessments at 18 months adjusted age.

Strengths of this study include a sample that was sufficiently large to detect moderate sized associations and the use of aEEG interpretations from independent and blinded readers.

CONCLUSIONS

In premature infants, sepsis is often associated with an acute encephalopathy that can be detected with aEEG. Sepsis-associated encephalopathy does not alter the rate of brain maturation as assessed with aEEG. Further investigation of this disorder could inform the design of strategies to reduce the risk for associated white matter damage and neurodevelopmental impairment and to identify those patients at highest risk for adverse outcome.

CHAPTER 3

Project Summary

After careful review of the literature, there is an obvious deficiency in our knowledge of the acute effects of sepsis on the premature infant brain. While there is evidence to support a relationship between neonatal sepsis and adverse neurologic outcome as well as animal research linking sepsis, inflammatory cytokines and white matter injury, there are no studies specifically addressing the acute effects of infection on cerebral function among premature neonates (8;52).

This study was designed to answer two questions: whether or not neonates experience encephalopathy during sepsis and if sepsis affects brain wave maturation in the short term. Use of generalized estimating equations, a type of generalized linear model specifically tailored for repeated measures, allowed for comparison of the rate of burst suppression between infants with sepsis and those without sepsis on repeated aEEG recordings. Interpretation of 297 aEEG recordings revealed an incidence of burst suppression on non-septic infants of 21% and 57% among septic infants. Using GEE, adjusting for postmenstrual age at the time of the recording, this difference was statistically significant ($p = 0.011$).

The rate of brain wave maturation over time was measured by serial aEEG recordings and maturation scores performed monthly through 36 weeks adjusted gestational age. Using linear regression, the slope of the maturation curve was calculated for each patient. The slopes were then compared between groups using t-test. There was no significant difference in the rate of maturation between the group of infants who developed sepsis during their hospitalization and those that did not.

Additional Analyses

During interpretation of aEEG recordings, we noted that many recordings looked very immature, but had an elevated baseline, which is usually an indication of maturity. Such instances were recorded in the comments section of the reporting forms. Discussion concerning possible explanations led us to consider high frequency ventilation as the cause for this phenomenon. High frequency ventilation (HFV) delivers 360 to 900 breaths per minute resulting in "jiggling" of the chest and entire body of the infant. Previous studies have not described characteristic aEEG findings during HFV.

A study assistant collected data on whether or not an infant was receiving HFV at the time of the aEEG recording. The variable "HFV" was then added into the model containing postmenstrual age and sepsis. Again using GEE, postmenstrual age remained significant ($p<0.001$). Sepsis also remained significant, but the odds ratio (OR) decreased from 2.4 ($p=0.011$) to 2.1 ($p=0.048$). HFV approached significance level with an OR of 2.3 ($p =0.062$). The effect of HFV on the OR of sepsis prompted an analysis for possible interaction. Again using generalized estimating equations, the product of sepsis and HFV was added into the model. The interaction term was not significant ($p=0.32$).

While the association of HFV with the presence of burst suppression does not reach statistical significance at the 0.05 significance level it does alter the OR of sepsis. These observations suggest that HFV might alter the aEEG recording. The concern is whether HFV produces actual changes in cerebral function or if the effect on the aEEG recording is primarily mechanical, resulting from the vibrations of the infant's body. While the effect of high frequency vibrations seems most likely, the possibility that HFV could change brain function, even if transiently, poses questions about risks of HFV on the developing premature infant brain.

One case series suggests that the effects of HFV on the aEEG recording is through artifact alone (64). In this series of three infants, all were simultaneously monitored with both EEG and aEEG. The presence of artifact on aEEG was confirmed by coincident EEG findings. In one infant the baseline was elevated as a result of HFV. This report is the only description in the literature of effects of HFV on aEEG.

The more important question is whether HFV could alter brain function. An observational follow up study of premature infants requiring mechanical ventilation for respiratory distress did not show a difference in neurodevelopmental outcome of infants managed with HFV versus conventional ventilation (65). This study only looked at 21 infants managed with high frequency oscillatory ventilation and compared them with age-matched controls; therefore the power was limited by a small sample size. Further studies may help to address the possible implications of providing HFV for premature infants with respiratory distress.

Lessons Learned

This project has created many learning opportunities. As with HFV, there are other variables to which these infants are exposed daily and may act in conjunction with or independently from our measured variables to affect our results. Such exposures could include postnatal steroids, which have been shown to play a role in neurodevelopment. While antenatal steroids may protect the developing brain from the negative effects of inflammation, postnatal steroids, when used at high doses, have been shown to increase the risk of cerebral palsy (66). The acute effects of postnatal steroids have not been previously described. Collecting information about this exposure could help in interpretation of our results, or provide new information to study.

Probably the most important lesson resulting from this project arose during analysis of the results. It is extremely important to understand the biology behind your

proposed associations as well as the actual distribution and characteristics of the data that you observed. These two pieces are essential in deciding which analyses are appropriate and how to interpret the results. The biologic pathways and possible causal relationships are keys to determining which factors may be predictor variables, confounding variables and outcome variables. An understanding of the relationships between variables is greatly enhanced by spending time developing and using literature to support a conceptual model. This model not only ensures a firm grasp on the relevant body of literature, but also provides insight into other possible associations which should be included in data collection and analysis.

Implications

While no research project is able to fully address all aspects of a relationship between exposure and outcome, every attempt should be made to provide as complete a picture of the hypothesized association as possible. In this case, more extensive data collection could have provided further insight about the effects of everyday neonatal exposures, such as HFV and postnatal steroids, on the aEEG tracings of these infants. Even though HFV has been previously reported in one patient as being artifact-creating, it could actually be producing changes in brain waves which alter the tracing. Without further investigation, this seemingly benign intervention may inadvertently create risk for future neurodevelopmental impairment in this population.

The findings from this project support the need for further exploration into the idea of a neonatal "sepsis-associated encephalopathy." While the brain wave maturation, as assessed by aEEG, was not significantly different between the groups with and without sepsis, there was evidence of an acute effect on cerebral function as evidenced by an increased incidence of burst suppression. Future studies should

address the pathophysiology behind sepsis-associated encephalopathy in neonates as will as implications for future neurodevelopmental outcome of these infants.

Future Directions

Since little is known about acute neurologic changes in premature infants and long term neurodevelopmental outcome, follow up of infants who have had acute changes during their initial hospitalization will improve our understanding of predictors of neonatal outcomes. The primary goal of the proposed follow up study will be to evaluate the association between specific aEEG abnormalities such as burst suppression and neurodevelopmental outcome.

There also exists a deficit in our knowledge concerning postnatal brain wave maturation and its ability to predict later neurodevelopment as measured by Bayley Scales of Infant Development or neurologic exam. A follow up study of these infants will enable us to better define the association between rate of brain wave maturation in preterm neonates and its correlation with long term neurodevelopmental outcome.

Infants admitted to Forsyth Medical Center who are less than 1000 grams at birth are invited to return at 18 months adjusted age to the follow-up clinic at Amos Cottage for developmental assessments using the Bayley Scales of Infant Development as well as an examination by a practitioner trained in neurodevelopmental exams. We plan to use the data obtained from this developmental assessment to evaluate the association between aEEG findings during acute infection, as determined by the present study, and long-term neurodevelopmental outcomes.

The proposed study will be a prospective observational follow up study of infants who were enrolled in this project. The predictors of interest are prior evidence of burst suppression on aEEG tracing, brain maturity at 36 weeks post menstrual age or discharge (adjusted for PMA) as assessed by aEEG maturation score and rate of brain

wave maturation from 28-36 weeks PMA or discharge as assessed by aEEG. The outcomes of interest are neurologic development measured by the Bayley Scales of Infant Development-Second Edition Mental Developmental Index and Psychomotor Developmental Index at 18-22 month adjusted age as well as presence or absence of cerebral palsy.

After completion of the follow up study, we will have identified and described a cohort of extremely low gestational age neonates who have experienced various exposures such as sepsis and have had serial assessments of their neurologic function. This longitudinal data on 108 ELGANs will be a wealth of information that should greatly enhance our understanding of the role of neonatal exposures in the neurodevelopment of premature infants.

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EDUCATION:

1992-1996	North Carolina State University Bachelor of Science in Zoology, *Summa Cum Laude*
1996-2000	East Carolina University School of Medicine Doctorate of Medicine
2007-present	Wake Forest University Graduate School Masters of Science in Health Sciences Research Anticipated graduation August 2009

POSTDOCTORAL TRAINING:

2000-2003	Pediatric Residency Program Wake Forest University Baptist Medical Center
2004-2007	Fellowship in Neonatal-Perinatal Medicine Wake Forest University Baptist Medical Center

PROFESSIONAL LICENSURE:

2001-Present	North Carolina Medical Board License Number 200101063

SPECIALTY CERTIFICATION:

October 2003 American Board of Pediatrics

October 2008 Neonatal-Perinatal Medicine

ACADEMIC APPOINTMENTS:

2003-2004 Clinical Instructor in Pediatrics
Wake Forest University Baptist Medical Center

2007-present Assistant Professor in Pediatrics/Neonatal-Perinatal Medicine
Wake Forest University School of Medicine

EMPLOYMENT:

2003-2004 Clinical Instructor in Pediatrics
Wake Forest University Baptist Medical Center

1997 Family Medicine Externship at Family Medical Care in Greenville

1997 Tutor for Student Affairs, East Carolina University School of Medicine

1995-1996 Office Assistant, Cary Pediatrics Center

1994-1995 Supplemental Chemistry Instructor
North Carolina State University

1994 Manager, The Video Bar in Cary, NC

INSTITUTIONAL SERVICE:

2005-Present Member
Neonatal Pain and Sedation Working Group
Wake Forest University Baptist Medical Center

PROFESSIONAL MEMBERSHIPS AND SERVICE:

2000-Present	American Academy of Pediatrics
1997-2000	Christian Medical and Dental Association
1997-1998	President of Student Chapter of Christian Medical Dental Association
1996-Present	North Carolina Medical Society
1997-1998	Delegate to North Carolina Medical Society
1996-Present	American Medical Association

HONORS AND AWARDS:

2000	William E. Laupus Pediatric Award East Carolina University School of Medicine
2000	American Medical Women's Association Glasgow Memorial Achievement Citation East Carolina University School of Medicine
1999	Alpha Omega Alpha East Carolina University School of Medicine
1999	Merck Manual Award East Carolina University School of Medicine
1998	Bakerman Scholarship East Carolina University School of Medicine
1996	Phi Beta Kappa North Carolina State University
1995	Phi Kappa Phi Honor Society North Carolina State University
1992-1996	Caldwell Scholar North Carolina State University

PROFESSIONAL INTERESTS:

My current area of research interest is sepsis and its effect on brain function and neurodevelopment. I am currently concluding an observational study entitled "Sepsis and Electroencephalographic Changes in Extremely Low Gestational Age Neonates." The objective of this study is to understand the correlation between infection and acute neurologic changes in extremely low gestational age neonates as measured by the amplitude integrated electroencephalogram (aEEG). I am using the data obtained from this study as part of my thesis on Sepsis Associated Encephalopathy in Premature Infants. My future plans include following this cohort of infants at 18 to 22 months corrected age to explore associations between acute changes in brain function and long term neurodevelopment.

INVITED PRESENTATIONS:

April 2009	What Would You Do? Interesting Cases in Neonatal-Perinatal Medicine Perinatal Symposium Forsyth Medical Center, Winston-Salem, NC
May 2008	Electroencephalographic Changes in ELGANs Pediatric Academic Societies' Annual Meeting Poster presentation Honolulu, HI
April 2008	Sepsis and Acute aEEG Abnormalities NC Neonatology Meeting Durham, NC
Feb. 2008	Sepsis and Encephalopathy 32nd Southeastern Conference on Perinatal Research Key Largo, FL
Nov. 2007	Viability and Ethics For Brenner Children's Hospital Transport Team Brenner Children's Hospital and Health Services
July 2006	Ventilator Management in Neonates Presentation for Fellows, Residents and Neonatal Staff Wake Forest University Baptist Medical Center

June 2006	Sepsis and Amplitude-Integrated Electroencephalography in Extremely Low Gestational Age Neonates: Poster Presentation 68th Perinatal & Developmental Medicine Symposium Aspen, CO
Jan. 2006	Sepsis and Electroencephalography in Extremely Low Gestational Age Neonates Perinatal Research Conference Wake Forest University Baptist Medical Center
Dec. 2005	Limits of Viability Obstetrics and Gynecology Grand Rounds Forsyth Medical Center
Nov. 2005	Sepsis and aEEG Neonatology Research Conference Wake Forest University Baptist Medical Center
March 2005	Indirect Hyperbilirubinemia Pediatric Resident Conference Wake Forest University Baptist Medical Center
Feb. 2005	Cyanotic Newborns Presented to Pediatric Residents Wake Forest University Baptist Medical Center
Feb. 2003	Sudden Infant Death Syndrome Pediatric Grand Rounds Wake Forest University Baptist Medical Center

COMMUNITY ACTIVITIES AND SERVICE:

1990-Present	North Carolina Farmers' State Alliance Member and Committee Chair
2004-Present	North Carolina State University Alumni Association Member
2006-2008	Choir member Calvary Baptist Church