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INTERMITTENT HYPOXEMIA IN PRETERM INFANTS

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Elie G. Abu Jawdeh, Student

Dr. Peter Giannone, Major Professor

Dr. Hannah Knudsen, Director of Graduate Studies

INTERMITTENT HYPOXEMIA IN PRETERM INFANTS

DISSERTATION

A dissertation submitted in partial fulfillment of the requirements for the degree of Doctor of Philosophy in the College of Medicine at the University of Kentucky

By

Elie G Abu Jawdeh

Lexington, Kentucky

Co-Directors: Dr. Peter Giannone, Professor of Pediatrics

and Dr. Yang Jiang, Associate Professor of Behavioral Sciences

Lexington, Kentucky

ABSTRACT OF DISSERTATION

INTERMITTENT HYPOXEMIA IN PRETERM INFANTS

Intermittent hypoxemia (IH) is defined as episodic drops in oxygen saturation (SpO_2). Virtually all preterm infants have IH events. Extremely preterm infants have hundreds of IH events per day. The extent of IH is not apparent clinically as accurately documenting cardiorespiratory events for day-to-day patient care management is challenging. High resolution pulse oximeters with 2 second averaging time are currently the ideal methods to measure IH. We have developed novel methods and processes to accurately and efficiently calculate an IH profile that reflects to spectrum of the problem.

The natural progression of IH is dynamic. There is low incidence of IH in the few 2 weeks of life, followed by a progressive increase until peak IH at 4-5 week after which IH plateaus. Multiple factors place preterm infants at high risk for increased IH. These factors include respiratory immaturity, lung disease, and anemia. We also show that preterm infants prenatally exposed to opioids or inflammation (due to maternal chorioamnionitis) have increased IH measures compared to unexposed infants. Interestingly, the increased IH in the exposed groups persists beyond the immediate postnatal period.

Brief episodes of oxygen desaturations may seem clinically insignificant; however, these events may have a cumulative effect on neonatal outcomes. There is mounting evidence from both animal models and clinical studies suggesting that IH is associated with injury and poor outcomes such as impaired growth, retinopathy of prematurity and neurodevelopmental impairment. In addition data from neonatal animal models and adults with obstructive sleep apnea suggest that IH is pro inflammatory itself. We demonstrate in this document for the first time in preterm infants that IH is associated with increased serum inflammatory marker, C-reactive protein.

Finally, a valuable experience throughout this process is working with a talented and dedicated multidisciplinary team. We are a solid example of the value of team science during this new era of clinical and translational research. Our respiratory control research program is one of handful programs nationwide able to perform such high-fidelity studies related to cardiorespiratory events in preterm infants. We will continue to tackle complex questions involving health of infants.

KEYWORDS: Intermittent Hypoxemia, Preterm Infants, Prenatal Opioid Exposure, Chorioamnionitis, Inflammation

Elie G. Abu Jawdeh, M.D.

6/26/2018

Date

INTERMITTENT HYPOXEMIA IN PRETERM INFANTS

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To my parents Giryes and Jeanne D’arc

To my brother Bassam and his family Manal, George and Michael

To my brother Dany

To Farah

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CHAPTER 1: INTRODUCTION AND CLINICAL RELEVANCE

This chapter was published as a review article at the American Academy of Pediatrics *NeoReviews.* The following is a summary of the review with permission from the publisher. The full review is not open access and can be found at the citation below. One section related to prenatal exposure was added to this chapter that was not included in the original publication.

Citation: ***Abu Jawdeh EG****. Intermittent Hypoxemia in Preterm Infants: Etiology and Clinical Relevance. NeoReviews. 2017 November 01; 18(11):e637-e646.*

I. Introduction

Intermittent hypoxemia (IH), generally defined as brief, episodic drops in hemoglobin oxygen saturation (SpO_2). Intermittent hypoxemia is a common disorder in preterm infants with rising evidence linking IH to neonatal morbidities and long term impairment. The definition and thresholds below which IH is clinically relevant are debatable (1-4).

II. Natural Progression

Intermittent hypoxemia is inversely related to gestational age (GA) (5, 6). Small for gestational age (SGA) are particularly at risk to having increased IH compared to infants appropriate for gestational age (AGA). In addition, IH natural progression varies by postnatal age (1, 2). There is a low frequency of IH during the first week after birth, followed by a progressive increase by weeks 2-3, with a peak around 4-5 weeks then plateau/decrease during weeks 6-10. The factors that influence the rise in IH are poorly defined (7, 8).

III. Factors that Influence Intermittent Hypoxemia

The conventional definition of apnea of prematurity (AOP) may not be applicable to the causality of IH in the current extremely premature NICU population with lung immaturity and lung disease, because as IH can often occur following very brief respiratory pauses, periodic breathing or ineffective ventilation (9-11).

The "Perfect Storm"

The impaired respiratory control along with lung disease/immaturity create a "perfect storm", leading to an increased IH frequency(12). Factors that contribute to increased respiratory pauses and resultant IH in preterm infants include: upregulated inhibitory neurotransmitters, decreased central chemo-sensitivity (7, 10, 13), paradoxical ventilatory depression in response to hypoxia (10, 13), hyper-excitable carotid bodies (14), immature laryngeal chemo-reflex (15) and low baseline functional residual capacity (FRC) (7, 10, 16).

Prenatal Exposure

Prenatal environmental exposures such as opioids, tobacco, and other drugs may have a sustained effect on apnea, lung disease and subsequently IH. Prenatal opioid exposure alters the response to carbon dioxide and depresses central respiratory control centers (17-21). Opioids are known to suppress breathing and respiratory effort especially in neonates (22). Opioid exposed infants often show intrauterine growth retardation and meconium staining, two hallmarks of fetal hypoxia. Similar to the literature from sudden infant death, prenatal opioid use may increase cardiorespiratory events in preterm infants. Prenatal opioids, especially street heroin, cause chronic intrauterine hypoxia leading to brainstem gliosis damaging the central respiratory centers; hence likely more apnea events (19). In addition, infants with intrauterine exposure to drugs of abuse have "down-regulation" of placental neurotransmitter receptors (23). Abnormalities or depletion of receptor sites, especially if the same process occurs in the fetal brain, could impair function of the normal neonatal respiratory control network leading to frequent or prolonged apnea and subsequent IH. Furthermore, prenatal exposure to other illicit drugs such as cocaine perturbs, albeit subtly, the maturation of respiratory control, resulting in disruption of postnatal respiration (24). Prenatal tobacco use is common; around 22% of mothers smoke while pregnant in the USA (25). Prenatal nicotine exposure increased apnea in neonatal mice (26). In addition, studies evaluating pulmonary mechanics in infants of smoking mothers indicated prenatal exposure affects pulmonary function by altering expiratory flow profiles, reducing respiratory compliance and increasing airway resistance (25, 27). Furthermore, prenatal tobacco alters chemoreceptor sensitivity and blunts response to hypoxia in infants (25, 28). Given the rising epidemic of drug abuse in the USA, a larger cohort aimed at understanding these relationships, especially opioids, is imperative and may have a direct impact on management of preterm infants.

Role of Inflammation

Inflammation increases apnea events and worsens lung disease; subsequently increasing IH (29-31). However, because IH is pro-inflammatory, the relationship between inflammation and IH may be bidirectional (7, 14, 32-35).

Anemia

Preterm infants with anemia are at increased risk for IH. As the hematocrit level decreases, the probability of apnea, bradycardia and IH events increases (1, 16, 36).

Target Oxygen Saturation

The target oxygen saturation influences the frequency of IH (8, 37). A lower SpO2 target is associated with greater incidence of IH events compared with higher SpO2 target (16).

IV. Monitoring

Intermittent hypoxemia is very common in preterm infants with hundreds of events per day and accurately documenting those events by bedside providers is challenging without continuous automated recordings (1, 38, 39).

V. Consequences

There is rising evidence linking IH to neonatal morbidities and long term impairment. These brief episodes of oxygen desaturations have been implicated in the following. Data from animal models: neurocognitive handicap, impaired myelination, decreased neuronal integrity, long-term neuro-functional deficits, increased inflammation and oxidative stress, impaired growth and sleep disordered breathing/apnea (32, 40-42). Data from human studies: Retinopathy of prematurity (ROP), Neurodevelopmental Impairment (NDI) (cognitive, motor and language delay) and death (3, 5, 43-46).

VI. Conclusion

Although IH is very common in preterm infants the extent of the problem is often underestimated by clinical providers. Multiple factors in preterm infants increase their risk for significant IH. Intermittent hypoxemia is clinically relevant with rising evidence from both animal models and preterm infants linking IH to poor outcomes.

CHAPTER 2: METHOD DEVELOPMENT AND VALIDATION

I. Introduction

Intermittent hypoxemia is a common problem in preterm infants due to their immature respiratory control (apnea of prematurity) and lung immaturity/disease (BPD). All preterm infants are at risk for IH. Extreme preterm infants have highest risk for IH, due to their extremely immature respiratory control and lung immaturity/disease. When oxygen saturation (SpO2) is continuously recorded, extreme preterm infants have on average 150 to 200 severe IH events per day during which their SpO_2 drops below 80% (1). Intermittent hypoxemia (IH) is defined as episodic drops in blood oxygen saturation. The specific definition of oxygen saturation (SpO2) drop varies by research group, however most consider SpO2 drop to less than 80% as significant (1-3, 36). Others consider a SpO2 of less than 90% as the starting point (4). Calculating and establishing an IH profile that reflects the spectrum of IH in terms of frequency, severity and duration is imperative.

Accurately documenting cardiorespiratory events for day-to-day patient care management is challenging, as the extent of IH is not apparent clinically. Pulse oximeters are the current standard of care for monitoring oxygenation in the Neonatal Intensive Care Unit (NICU). Bedside providers under-recognize the number of events compared to objective automated recordings. In one study, compared to polysomnography, nursing staff recorded less than 30% and 40% of IH and bradycardia events, respectively. The shorter the event, the less likely that it was recognized by nursing staff (36). For example, bedside providers documented 35% and 29% of IH events that lasted greater than 20 and 10 seconds, respectively (38). Pulse oximeters are the current standard of care for monitoring oxygenation in the NICU. Hence, continuous physiologic recording is required for accurate detection of IH.

In this section we describe the development of methods for SpO2 recording, filtering, analyses, selection of outcome measures and validation of our novel programs.

II. Data Acquisition

Oxygen saturation data were prospectively collected from preterm infants admitted to our level 4 NICU starting November 2014. We used Masimo Radical 7 (Masimo, Irvine, CA) pulse oximeters for continuous data acquisition. Masimo pulse oximeters are widely used in NICUs worldwide due to their proprietary Signal Extraction Technology (SET®) that measures through motion and low perfusion; both important considerations in preterm infants (37, 47-52). All our research pulse oximeters were updated to the latest software prior to study initiation.

Pulse oximeters were equipped with serial data recorders (Acumen Instruments Corp) for continuous data collection (4). The Acumen recorders were connected to the RS232 port located on the Masimo pulse oximeter docking station. Data was collected with 1Hz frequency (every second) and saved on compact flash memory cards connected to the serial data recorders. The compact flash memory cards saved data continuously and were manually downloaded by research personnel to our encrypted servers provided by the University of Kentucky. We programed the Acumen recorders to save the data in daily files (midnight to midnight). The daily files were easier to transfer due to smaller size. In addition, the daily files provided a visual check of data loss if any and troubleshooting if necessary. The Acumen serial data recorder provided a time stamp (date and time, including seconds) for every second of data download. Time stamping is important while linking our IH data to other outcome measures. We downloaded data from serial data recorders weekly. Initially we had difficulty with memory cards not being reliable leading to data loss. However, that problem was transient and resolved with a different brand of memory cards.

We also trialed a different serial data recorder (SeriaGhost Logger) that was placed in series with the Acumen recorders. The SerialGhost recorders were reliable and stored data accurately. The SerialGhost saved all data in one file that at times was tens of gigabytes in size before post processing. The SerialGhost utilized the timestamp from the pulse oximeters versus the Acumen which had its own time stamp (in addition to that of the pulse oximeters). The SerialGhost had the capacity for timestamping however in our experience it was not reliable and was not linked to every second of data download. The SerialGhost was downloaded once at the end of the study period as downloading weekly was not feasible in the absence of a memory card. We tested a SerialGhost with Wi-Fi capabilities. The goal was to download directly to our encrypted serves. This was more challenging than expected given both 1) hospital network restrictions and 2) network changes upon moving infants from one room to another. Currently, we only use the Acumen serial data recorders.

A research monitoring unit is connected to the patient after informed consent is obtained. Initially we docked our research units to the clinical stations and utilized the same pulse oximeter for both for clinical and research purposes. An alarm delay was set to avoid alarm fatigue. However, the serial data recorders were sometimes left behind when moving patients among rooms. Early during our study period we changed this practice and currently we utilize an additional research pulse oximeter that moves with the patient. The research pulse oximeter alarm settings are silenced to avoid further noise and alarm fatigue. Patients are connected to the additional research pulse oximeter upon enrollment and monitored for first 2 months of life or 36 weeks corrected age, whichever came last.

Averaging Time

Pulse oximeters are the current standard of care for monitoring oxygenation in the NICU. However, the monitor settings, such as the averaging time, affects the number of IH events recorded (39). Pulse oximeters average SpO_2 values over several heartbeats. Pulse oximeters set to longer averaging times underestimate IH events of short duration and overestimate events of longer duration. This is likely as a result of several short events merged together as one prolonged event **(Figure 2.1)**. Clinical pulse oximeters are set to longer averaging time to decrease alarm fatigue for bedside providers (53). The default averaging times in clinical pulse oximeters range between 8 to 10 seconds but can be as long as 16 seconds. An option for centers who wish to use shorter averaging time is setting a longer alarm delay time (10 to 15 seconds) to reduce alarm fatigue (53). For research purposes, similar to other groups who study IH, we utilized high-resolution pulse oximeters with 2-second averaging time for continuous SpO_2 monitoring (1, 2). We confirmed and tracked the pulse oximeter settings weekly during data download.

III. Data Filtering and Processing

In collaboration with biomedical engineering (Dr. Abhijit Patwardhan laboratory) we developed novel programs to filter and process SpO2 data to analyze IH. Both algorithms were developed using Matlab (Matlab, Natick, MA).

The IH data filtering program excluded artifacts based on both the EXC code provided by Masimo monitors and missing variables in the output. The filtering program imported the raw data in text (.txt) format and exported clean data in text (.txt) format as well. The exported data files were automatically organized daily by the algorithm. The daily file names included the patient identification number and the date of the recorded data. The filtering algorithm has the capacity to filter multiple patients at the same time.

The second program is called Intermittent Hypoxemia Automated Analyses Algorithm (IH-AAA). The IH-AAA process the filtered data files to analyze the IH profile (below). The algorithm imported the clean daily text files (1 Hz frequency) and exported analyzed IH outcome measures in excel files averaged over different durations and intervals (weekly, daily, hourly). This program has the capacity to analyze multiple patients at the same time. The algorithm exports multiple excel files for every patient to reflect the spectrum of IH of different durations (e.g. 4-180 seconds, >180 seconds, etc.) and intervals (weekly, daily, hourly). Each excel file is labeled with patients identification number, date of the recorded data and interval. The IH-AAA also has the capacity to filter raw data in text files.

IV. Intermittent Hypoxemia Profile

The clinical relevance of IH is a relatively new observation (2) with no accurately defined threshold below which IH leads to morbidities and impairment; the exact definition of IH is controversial (54). Therefore, we developed a program that accounts for IH at multiple thresholds and calculate an IH profile. The IH profile reflects the *continuum* of the IH problem making it possible to demonstrate at what level IH causes injury.

In this section we describe the IH profile. For the purpose of demonstration we used a sample patient. We selected the second patient enrolled in our cohort (IH0002). The first patient had an early death and does not have a complete data set.

Frequency

The number of IH events is calculated for every interval (weekly, daily, hourly). The frequency of IH is a primary outcome measure that has been utilized by us and other groups and linked to neonatal morbidities and mortality (1, 2, 4, 55, 56). We define severe IH events as a SpO2 drop to less than 80% (IH-

SpO2<80). Moderate and mild IH are defined as a drop in SpO2 to less than 85% (IH-SpO2<85) and 90% (IH-SpO2<90), respectively. An additional outcome measure is calculated based on Rhein et al. where mild IH is calculated based on a change from baseline by more than 4% and to SpO2<90 (IH-SpO2<90 (>4% Drop))(4). We have the capacity to change our thresholds for IH frequency. Our program outputs frequency of IH at different intervals (weeks, days, hours) as represented in **Figure 2.2**. An upper threshold is often set for IH to differentiate intermittent from sustained hypoxemia. We also document sustained hypoxemia measures.

Similar to IH, hyperoxemic events are calculated. Documenting hyperoxemia is important given both the associated morbidities and to assess fluctuations in oxygenation. We currently have the hyperoxemia severity set at 2 thresholds with SpO2 more than 95% (IH-SpO2>95) and 97% (IH-SpO2>97). Sample patient for hyperoxemic events frequency is presented in **Figure 2.3**.

Percent time

The percent time in hypoxemia is another primary measure. The benefit of this outcome measure is that it represents cumulative IH events of short and long duration. The same 3 SpO2 thresholds for percent time in hypoxemia are selected here for severe (%time-SpO2<80), moderate (%time-SpO2<85) and mild hypoxemia (%time-SpO2<90 and %time-SpO2<90 (>4% drop)) (**Figure 2.4**). This measure of percent time spent with SpO2 below threshold was chosen per Poets et al. (3). Percent time is calculated at multiple intervals (weeks, days, hours). Similarly, hyperoxemia is analyzed demonstrating percent time spent with SpO2 more than 95% and 97%, (**Figure 2.5**). Percent time outcome measure is not affected by averaging time and is clinically relevant in all NICUs (39).

Mean, Nadir and Peak

The mean, nadir and peak SpO2 measures provide an additional perspective for IH. Mean is calculated for every interval (weeks, days, hours) and provides a baseline for IH during that interval (**Figure 2.6**). Both an average nadir for all events and lowest nadir are calculated. The nadir provides insight regarding the severity of IH (**Figure 2.7**). Similarly the average peak and highest signal are calculated for every interval (weeks, days, hours), (**Figure 2.8**).

Duration

The duration of IH is addressed in multiple forms. First, the average duration of IH events below every threshold is calculated for the three intervals (weeks, days, hours) (**Figure 2.9**). Similarly the average duration is calculated for hyperoxemia (**Figure 2.10**). However, the duration of IH may vary widely and the average duration may not be representative as it is influenced by outliers. Hence, we developed our algorithm to output multiple files of different duration cutoffs (for example multiples of 60 seconds). E.g. 1-59seconds, 60-119 seconds, 120-179 seconds, 180-239 seconds, 240-299, >300 seconds. By dividing the duration cutoffs, the average duration is influenced less by outliers. The duration cutoffs can be easily adjusted to any duration (for example multiples of 30 seconds, etc.) In addition, we use the 4-180 second cutoff for the primary measure as previously described by Abu Jawdeh et al. and Rhein et al. (1, 4).

Bradycardia

Heart rate decelerations or bradycardia events are part of the apnea of prematurity problem. Our algorithm calculates bradycardia below 2 thresholds of 80 and 100 beats per minute (bpm) (**Figure 2.11**) (11). The relative time of bradycardia to IH is also calculated. However, the role of heart rate deceleration is beyond the scope of this document.

Perfusion Index

Perfusion index (PI) is a noninvasive measure of perfusion thought to reflect the general hemodynamic status of the preterm infant (57, 58). Perfusion index assesses the pulse strength derived from pulse oximetry. Perfusion index is measured by infrared light, and is calculated as the ratio of the pulsatile to non-pulsatile components of the blood flow in tissue. The value of PI has been demonstrated in multiple neonatal morbidities (59, 60), including prediction of patent ductus arteriosus patency shown from this cohort (61). **Figure 2.12** demonstrates PI in a sample patient.

V. Statistical Analyses

Statistical modeling must account for the covariance among repeated measurements from the same subject. A general strategy to do so is to use linear mixed models, also known as linear multilevel models, hierarchical models, or multivariate Gaussian models (62). Correctly accounting for this correlation will ensure standard errors are appropriately estimated, thus yielding correct p-values and thus valid inference. Furthermore, in our experience and elsewhere, the need for an appropriate transformation, such as the square root, is needed for IH-based measures (2).

In order to attain valid inference, the model for the mean structure of the given outcome over time, as well as the model for the covariance among outcomes from the same subject (not of interest to the research question, but a necessity for inference), must be correctly specified (62). Otherwise, inference may be biased. A simple solution to this issue is to look at outcomes aggregated over weekly periods; e.g., weekly IH totals. In such a case, a statistical model can treat time as categorical to ensure a correctly specified mean structure with respect to time. Furthermore, a working unstructured covariance can be used with such a mean structure to ensure appropriate standard error estimation. Finally, this flexible modeling of the mean and covariance structures allows inference to be valid if any missing data are missing completely at random (MCAR) or missing at random (MAR) (63).

Outcome data is sometimes not captured for periods of time for example due to patient leaving the NICU for procedures or imaging. Therefore, as outcomes are usually aggregated over time, e.g. the total IH count for a given weekly period, such outcomes should be weighted by the amount of time they were observed. For instance, if interest is in weekly IH count, but IH data were only obtained for exactly half of the week, then that subject's total IH count would need to be doubled for use in the statistical analysis such that it represents the desired weekly total. A weight of one half would then need to be assigned to this outcome value in the analysis. If this weighting procedure is not done, estimated means and standard errors may be biased.

VI. Validation

In order to validate the novel program, we performed an assessment comparing IH measures calculated by the algorithm to those of independent observers. The observers were masked (blind) to the algorithm analyses. We obtained SpO2 data from 20 preterm infants less than 30 weeks GA randomly selected from our cohort. A total of 60 hours were analyzed. Each subject contributed 3 hours of SpO2 data; 1 hour from each postnatal age epoch (1 week, 1 months and end of study period) as defined by Abu Jawdeh et al. (1). We included IH events 4-180 second duration per Abu Jawdeh et al. (1). The validation presented focuses on two primary measures, IH-SpO2<80 and %time-SpO2<80. Other thresholds of less than 85% and 90% were examined with similar results.

The observers were masked to both other observers and algorithm counts. Three observers manually counted the first measure of IH-SpO2<80 from the raw data. The second measure of %time-SpO2<80 was analyzed by a singled masked observer utilizing Microsoft Excel (Excel Version 2010). Pearson correlations among observers and algorithm were performed using GraphPad (Prism 7). There was excellent correlation among observers as presented in **Figure 2.13**. For IH-SpO2<80, there was excellent correlation between mean observer count and algorithm count as presented in **Figure 2.14**. For %time-

SpO2<80, there was excellent correlation between observer and algorithm as presented in **Figure 2.15**.

VII. Discussion

There is rising evidence linking IH to both short and long term morbidities in preterm infants and hence, accurate recording of these events is paramount in determining their impact. In this chapter we described methods development to collect and process SpO2 data to measure IH. We defined our IH outcome measures with emphasis on IH profile. Finally, we presented the process for validating IH-AAA for accurate and reliable measurement of IH. We have developed an automated, convenient, and time efficient strategy to record such events, with exceptional accuracy when compared to human measurements.

The clinical significance of IH in preterm infants is a relatively new observation (2, 3, 54). In the past, brief IH events occur hundreds of times per day seemed clinically insignificant. However in the last 5-10 years, the interest in accurately documenting these IH events increased given the recent evidence linking IH to neonatal morbidities. Accurately documenting IH should involve a continuous physiologic recording with an automated system as bedside providers under-recognize the number of events (36, 38). An important factor in continuous monitoring is averaging time of pulse oximeters. As longer averaging times will underestimate IH events of short duration and overestimate events of longer duration (39, 64). This is likely as a result of several short events merged together as one prolonged event (39, 64).

An additional challenge relates to IH is variation in definitions. This variation is likely related to the unknown thresholds below which IH leads to injury. Most centers however consider a SpO2 drop to less than 80% as clinically relevant. We developed an IH profile that represents the continuum of IH. The IH profile allows us to better define at what threshold (e.g. severity, duration, etc.) IH matters clinically.

In conclusion, over the last 5 years we developed efficient and validated methods to accurately assess IH. We are one of few centers in the nation able to perform high fidelity studies related to IH in preterm infants.

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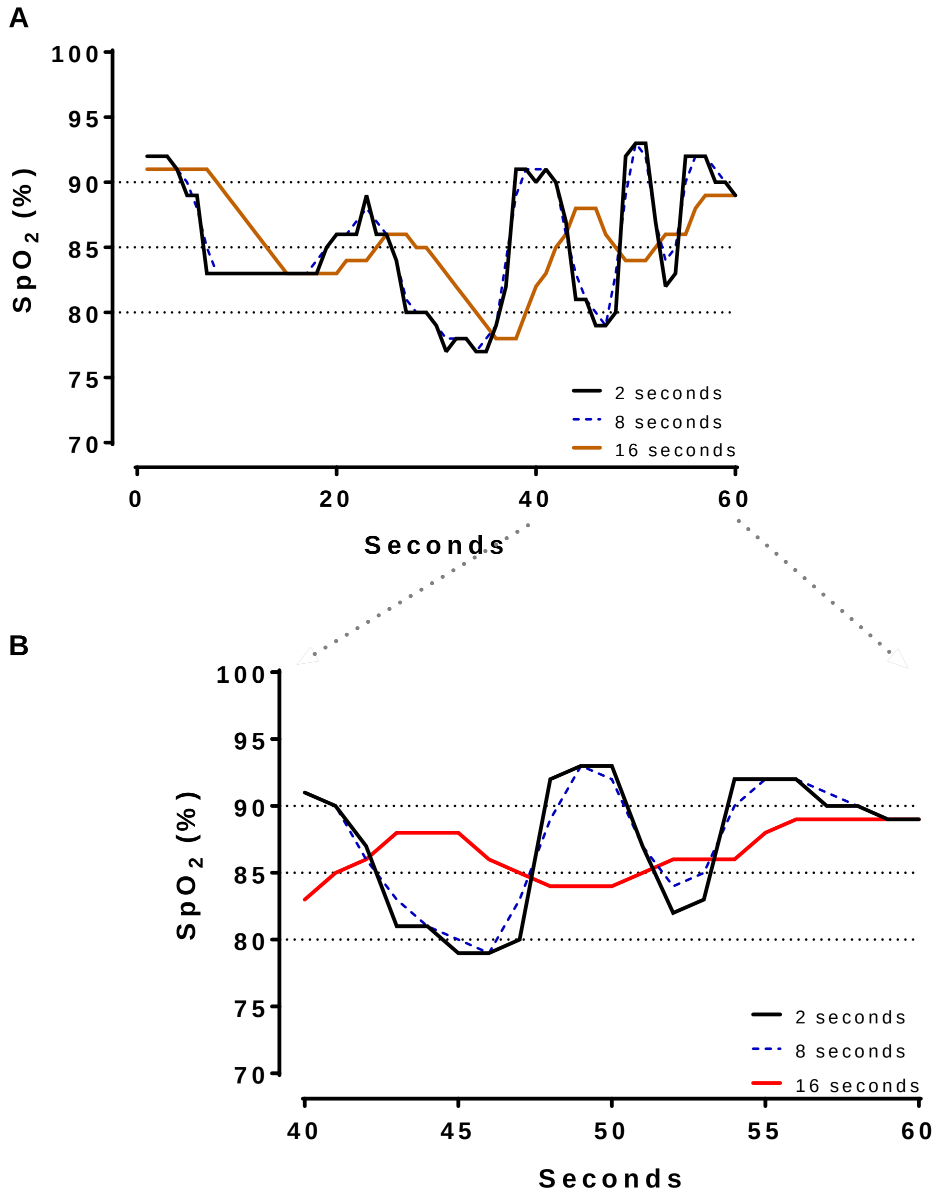

Figure 2. 1: A sample showing the effect of averaging time on the number of IH events.

A) The 2 second averaging time recording shows 3 events IH-SpO2<90, 3 event IH-SpO2<85 and 2 event IH-SpO2<80. In contrast, 16 second averaging conversion shows 1 event IH-SpO2<90, 3 events IH-SpO2<85 and 1 event IH-SpO2<80. **B)** This figure zooms in to seconds 40-60 to show how increased averaging time smooths the waveform by merging multiple short events.

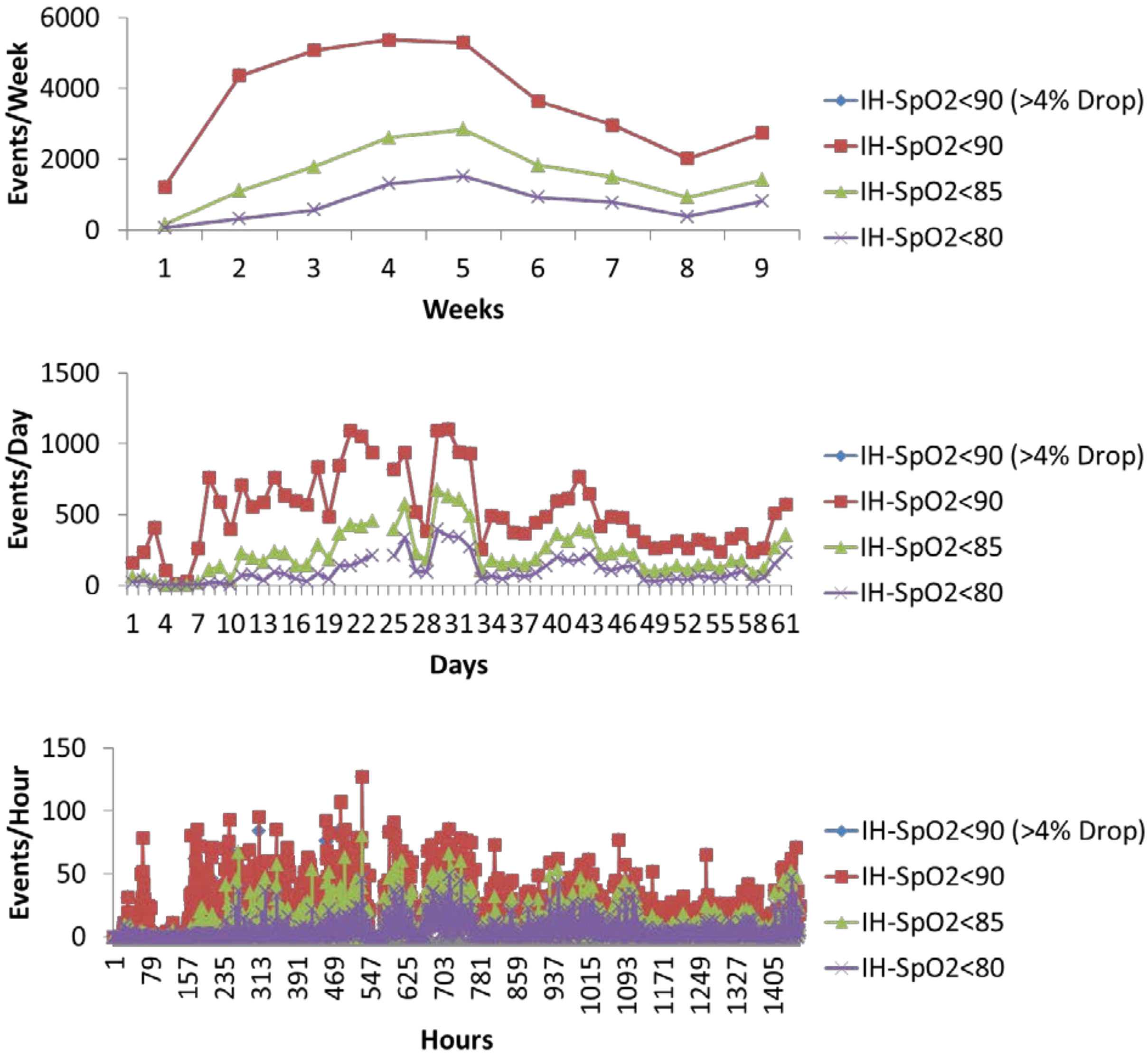

Figure 2. 2: Sample demonstration of frequency of IH events averaged over 3 intervals (weeks, days and hours).

The graphs present IH below multiple thresholds: IH-SpO2<90 (>4% Drop), IH-SpO2<90, IH-SpO2<85 and IH-SpO2<80.

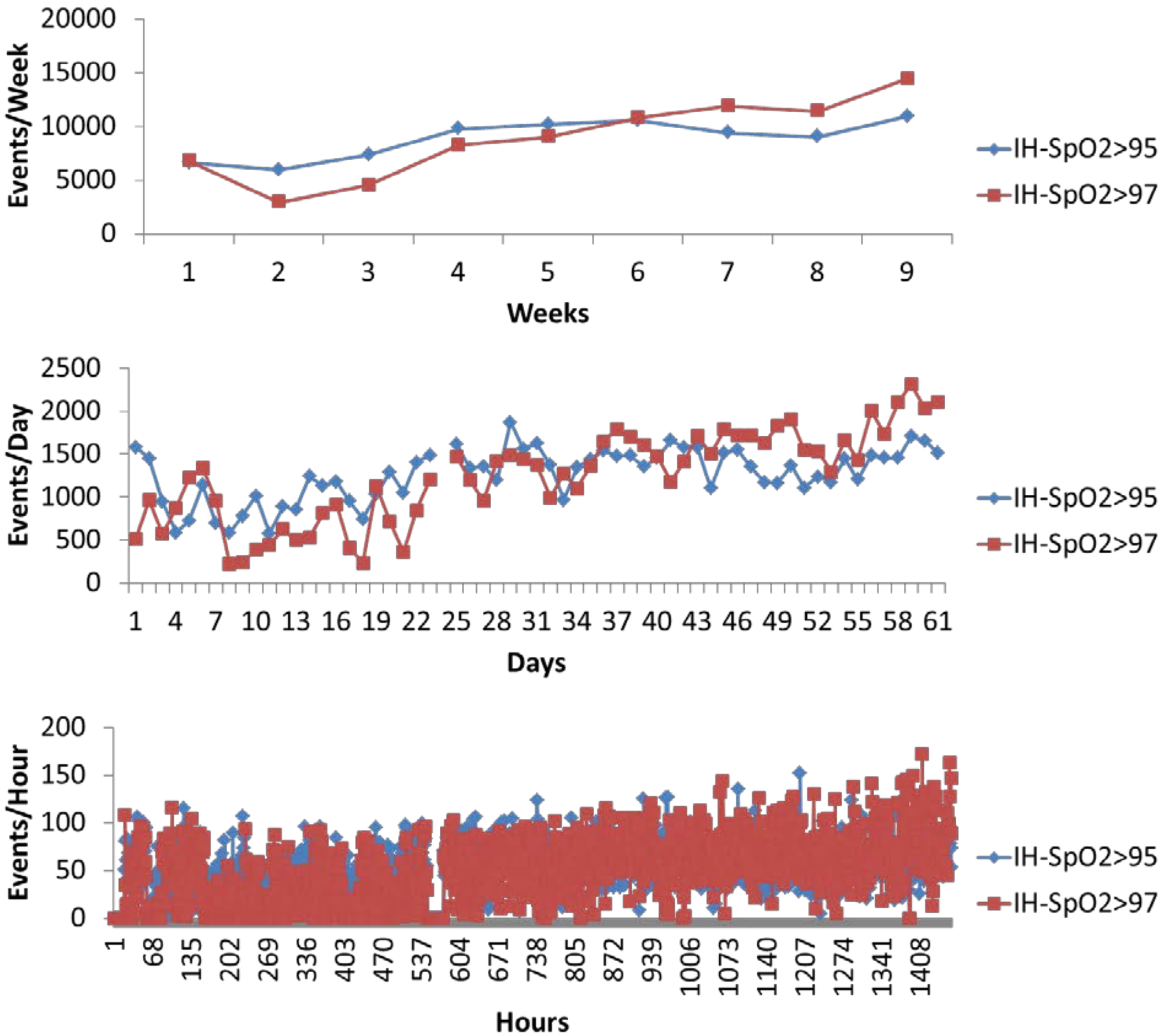

Figure 2. 3: Sample demonstration of frequency of hyperoxemic events averaged over 3 intervals (weeks, days and hours).

The graphs present hyperoxemia events with SpO2 greater than 95% (IH-SpO2>95) and 97% (IH-SpO2>97).

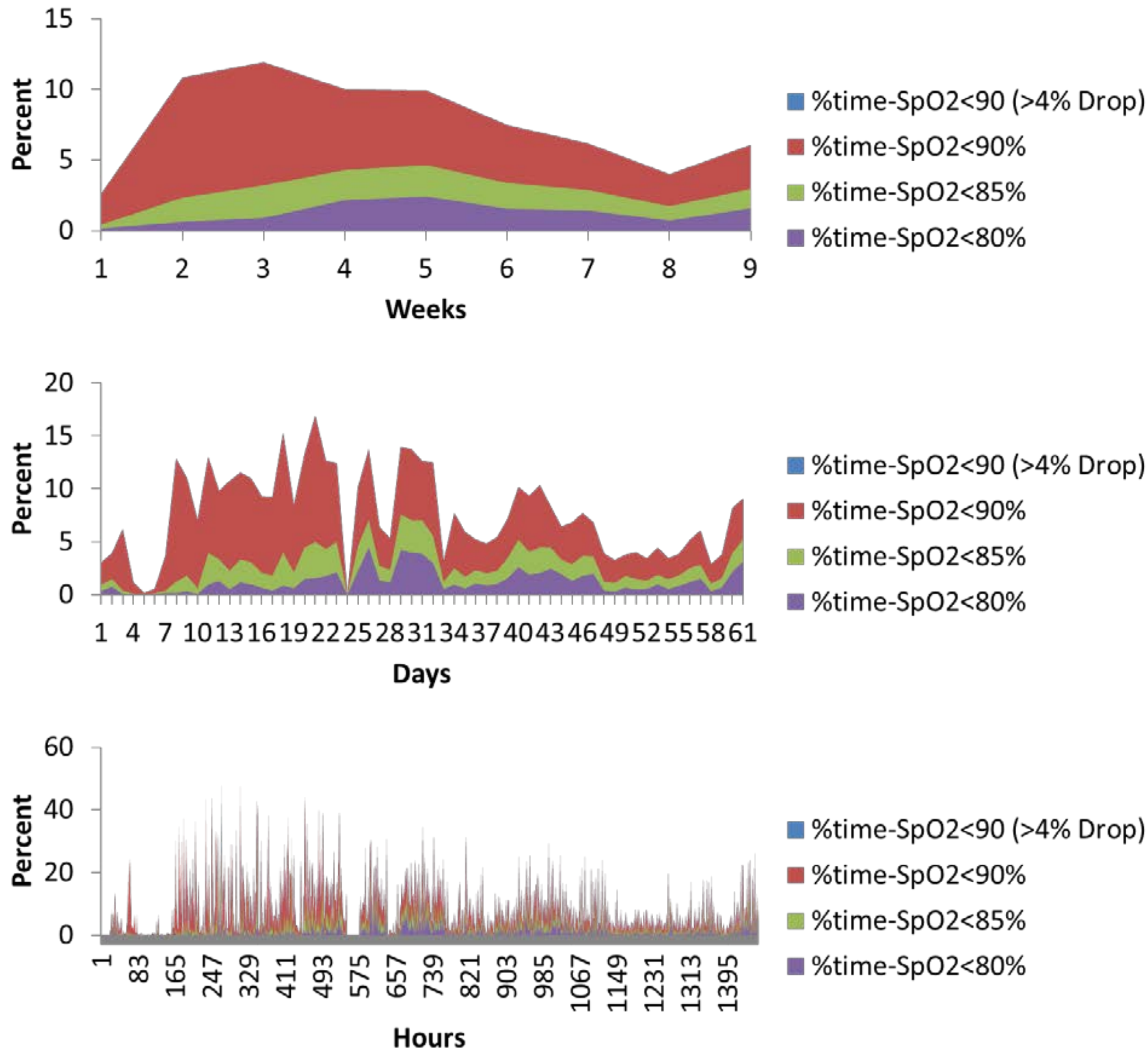

Figure 2. 4: Sample demonstration of percent time spent with SpO2 below thresholds averaged over 3 intervals (weeks, days and hours).

The graphs present percent time below multiple thresholds: %time-SpO2<90 (>4% Drop), %time-SpO2<90, %time-SpO2<85 and%time-SpO2<80.

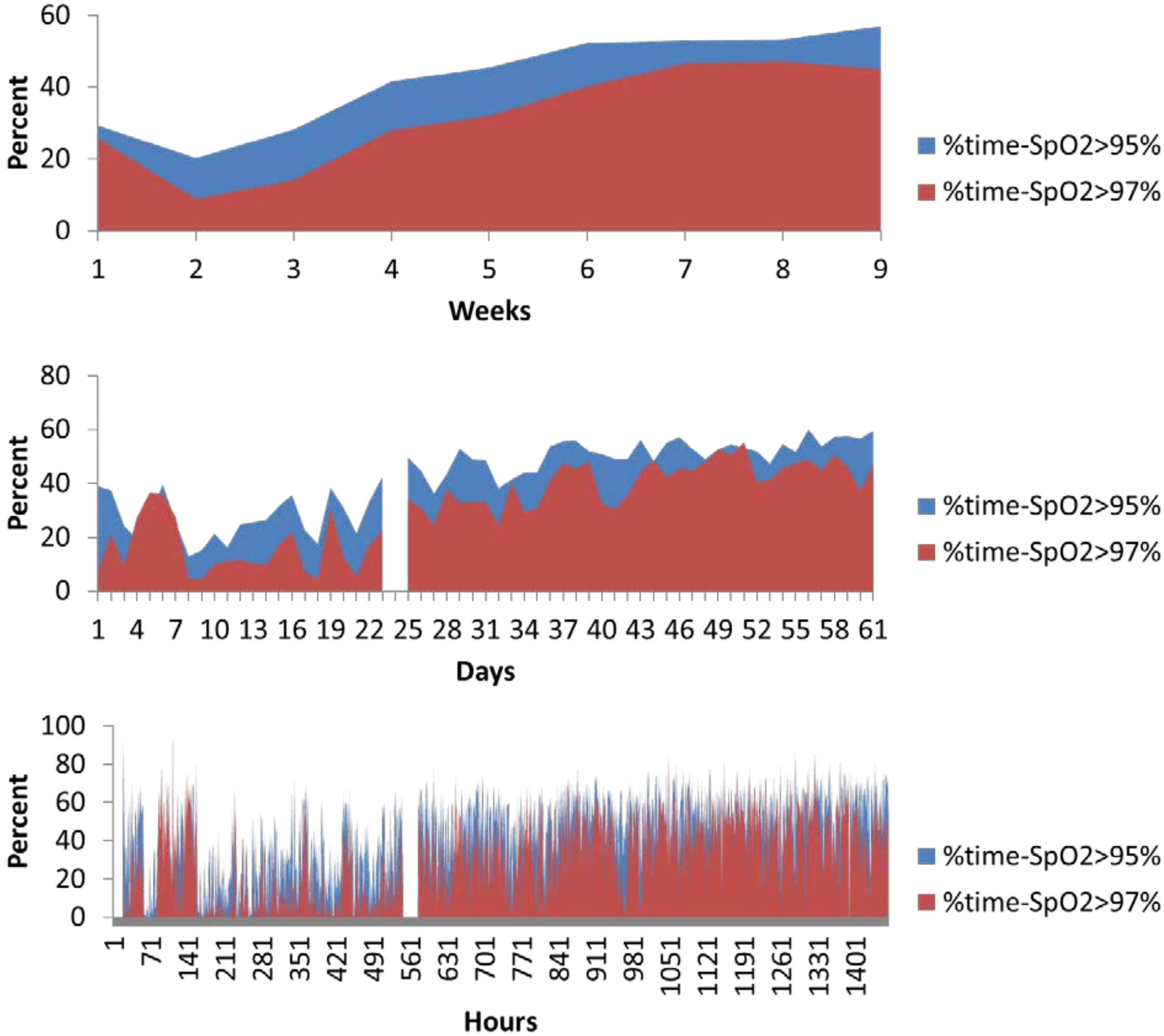

Figure 2. 5: Sample demonstration of percent time spent with SpO2 above thresholds (hyperoxemia) averaged over 3 intervals (weeks, days and hours).

The graphs present percent time with SpO2 greater than 95% (%time-SpO2>95) and 97% (%time-SpO2>97).

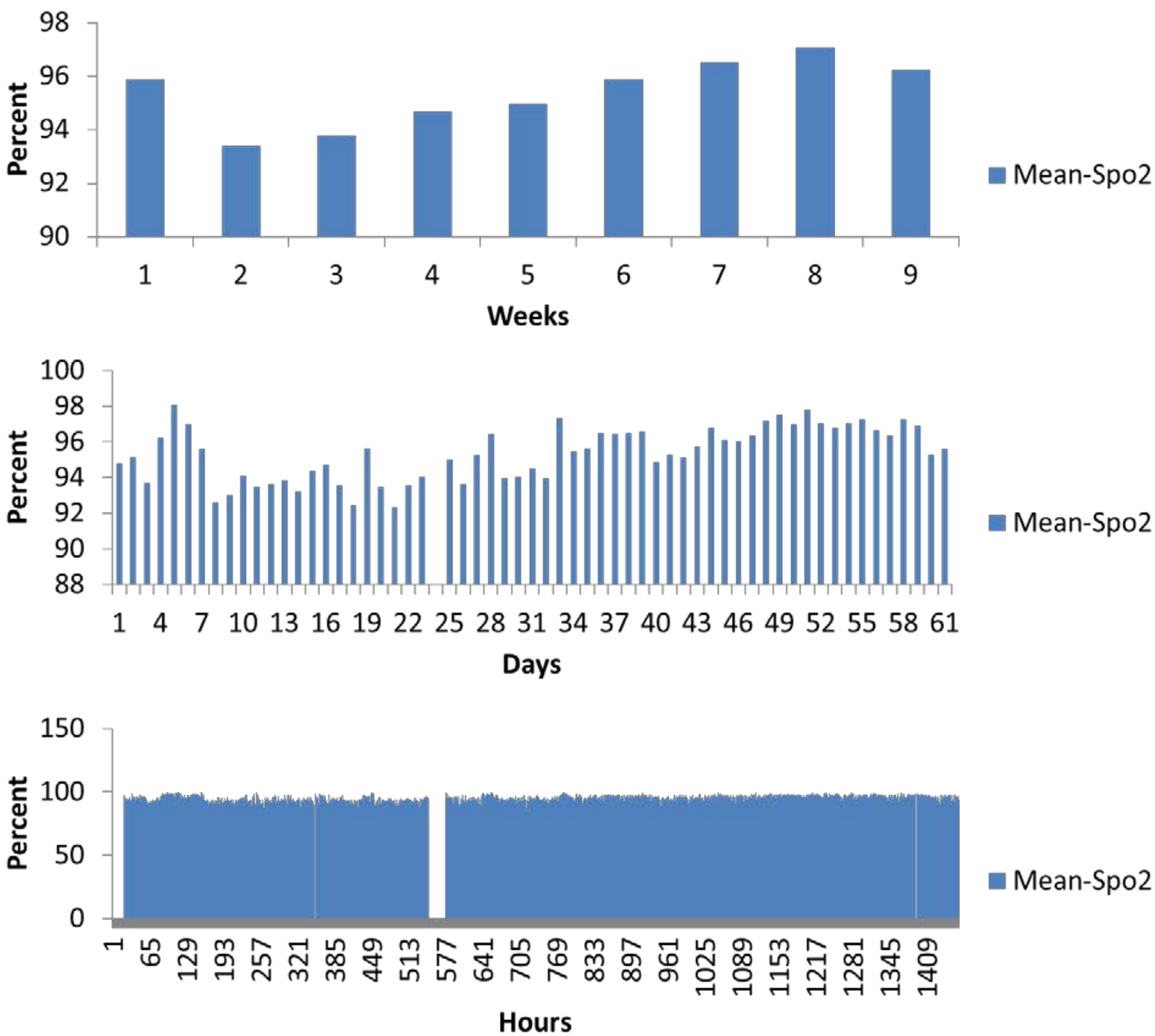

Figure 2. 6: Mean SpO2 presented at different intervals (weeks, days, hours) from a sample patient.

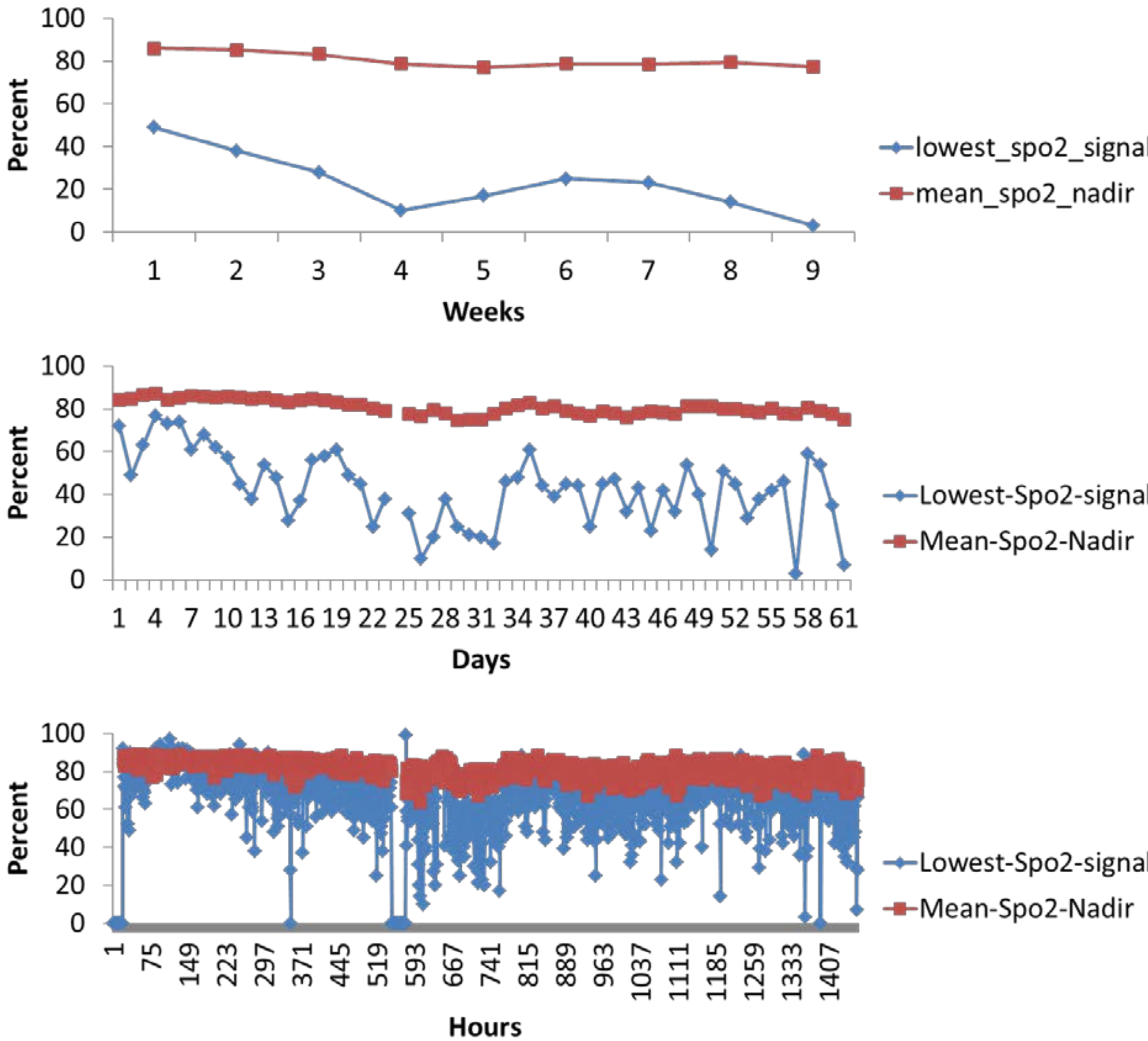

Figure 2. 7: Mean average nadir and lowest SpO2 signal presented at different intervals (weeks, days, hours) from a sample patient.

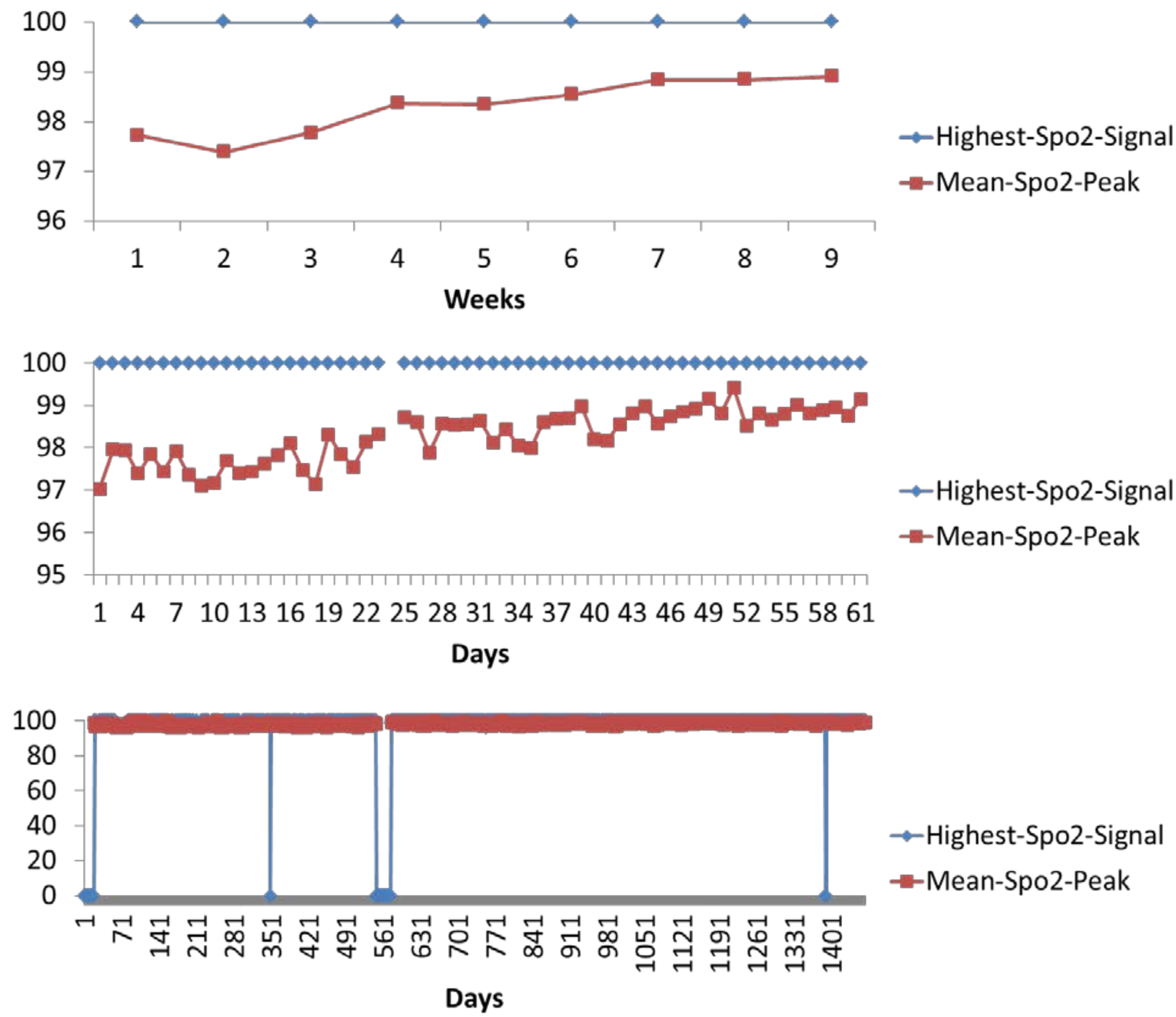

Figure 2. 8: Mean average peak and highest SpO2 signal presented at different intervals (weeks, days, hours) from a sample patient.

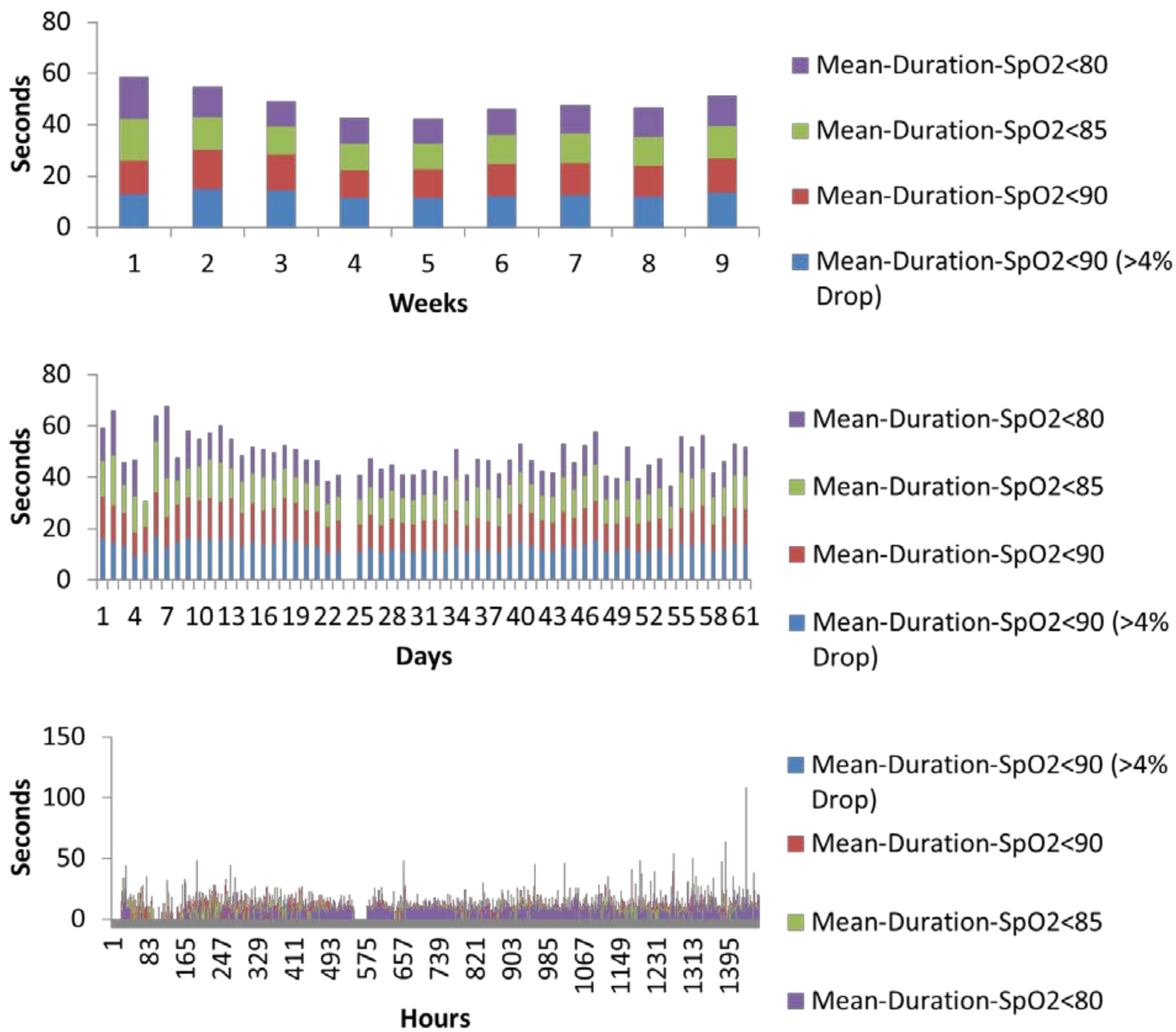

Figure 2. 9: Mean average duration of IH events presented at different intervals (weeks, days, hours) from a sample patient.

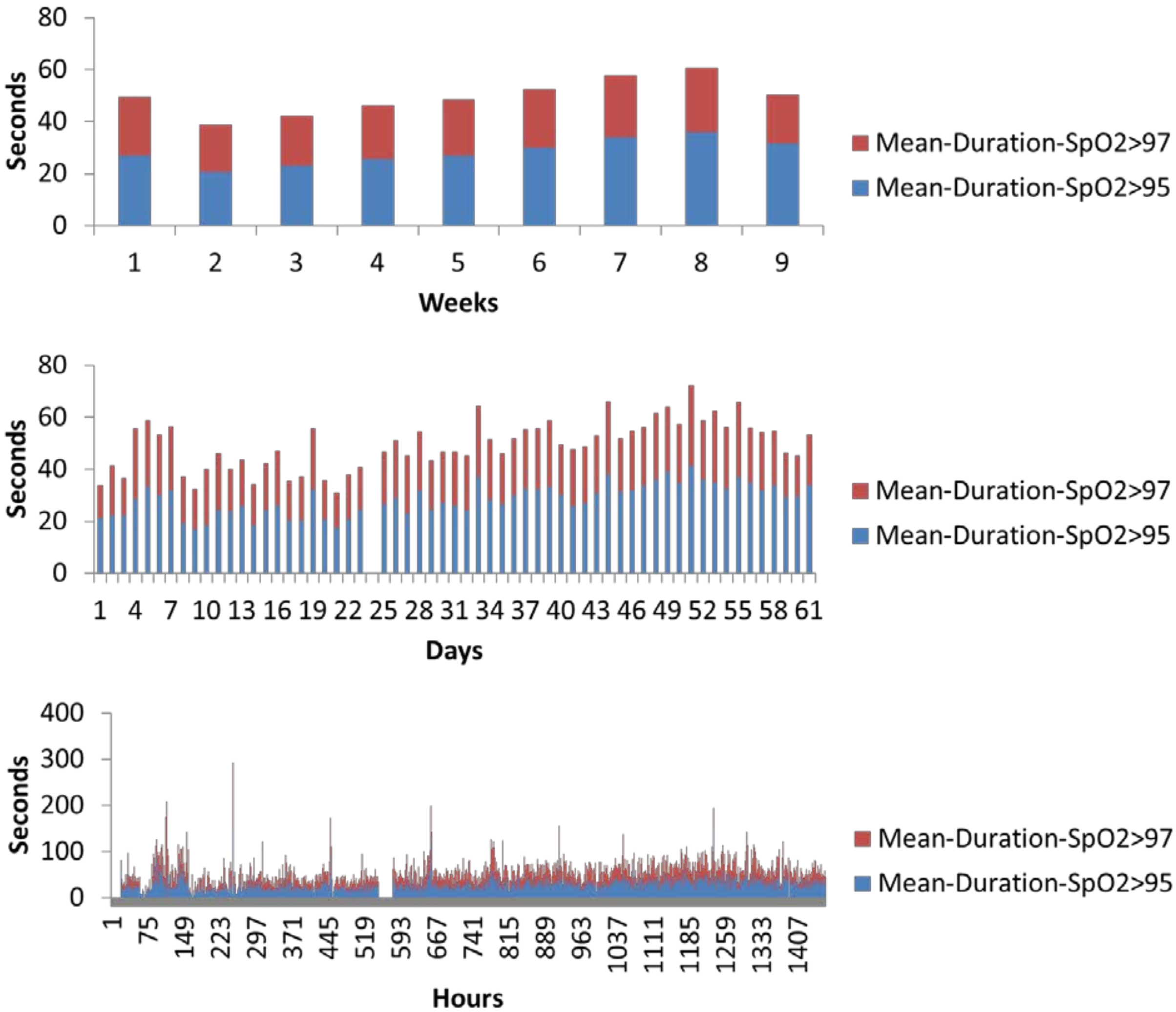

Figure 2. 10: Mean average duration of hyperoxemia events presented at different intervals (weeks, days, hours) from a sample patient.

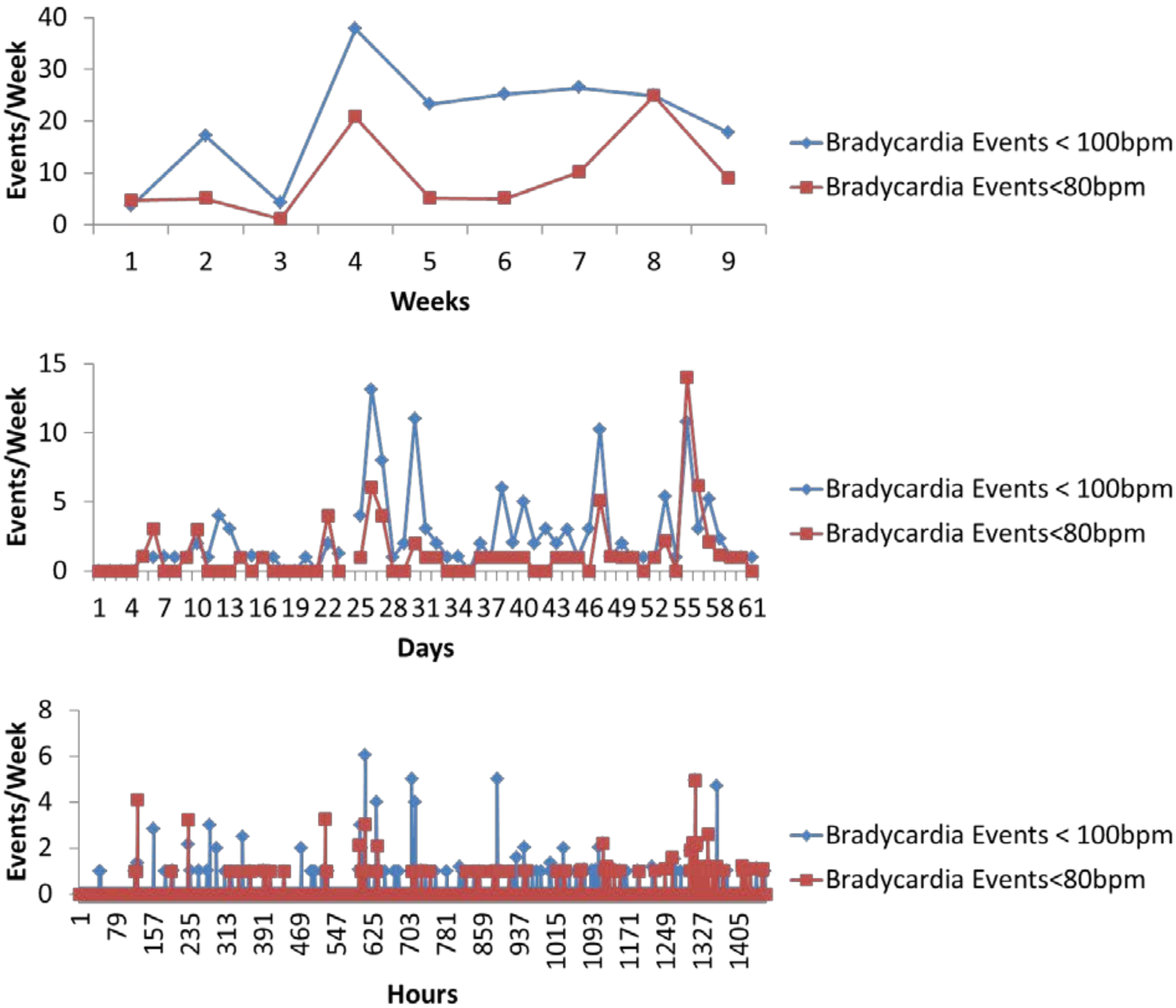

Figure 2. 11: Sample demonstration of bradycardia events averaged over 3 intervals (weeks, days and hours).

The graphs present heart rate deceleration below two thresholds of 100 beats per minute (bpm) and 80bpm.

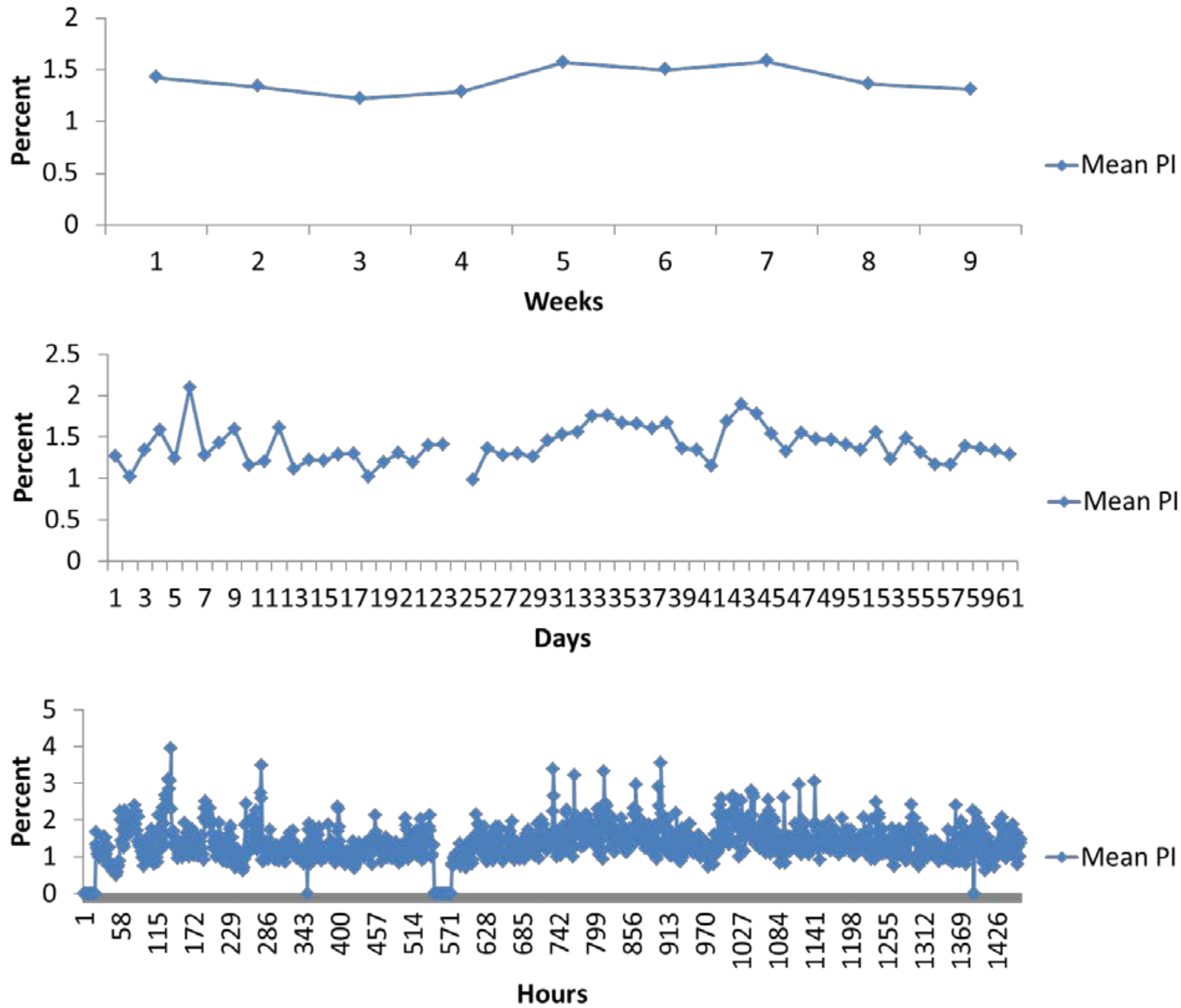

Figure 2. 12: Mean perfusion index (PI) presented at different intervals (weeks, days, hours) from a sample patient.

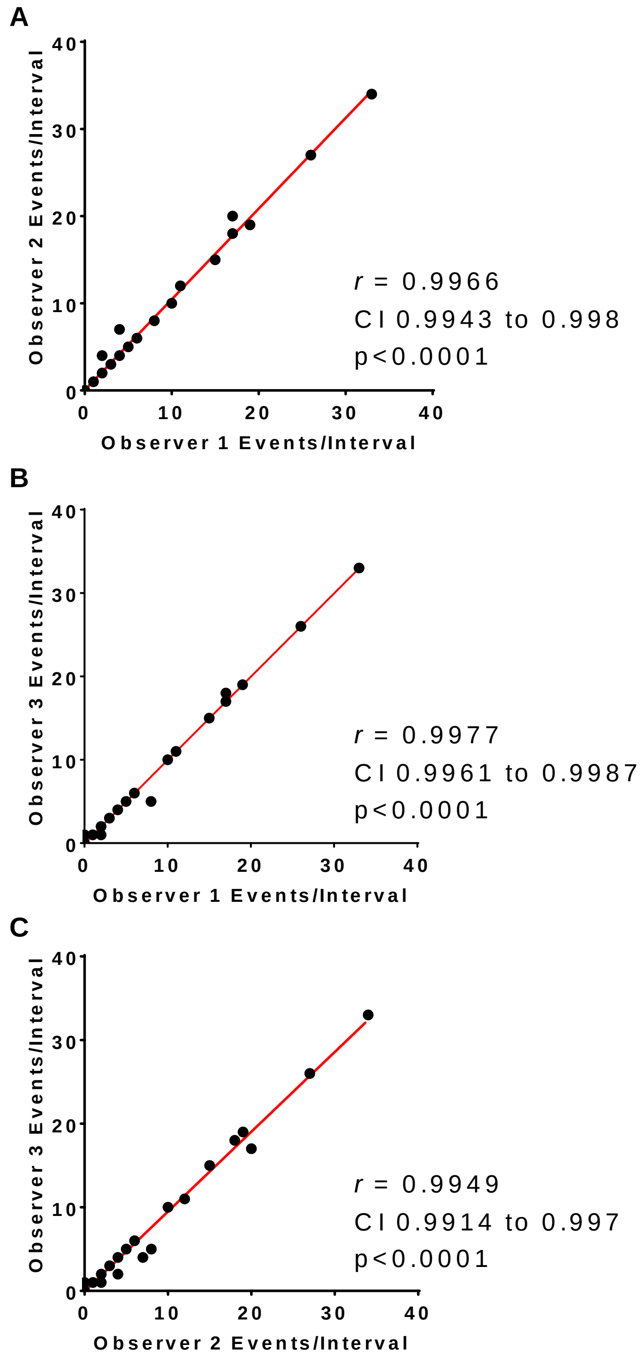

Figure 2. 13: Inter-observer Pearson correlations among observers for the number of IH events (IH-SpO2<80).

(A) Observer 2 vs. Observer 1. **(B)** Observer 3 vs. Observer 1 **(C)** Observer 3 vs. Observer 2.

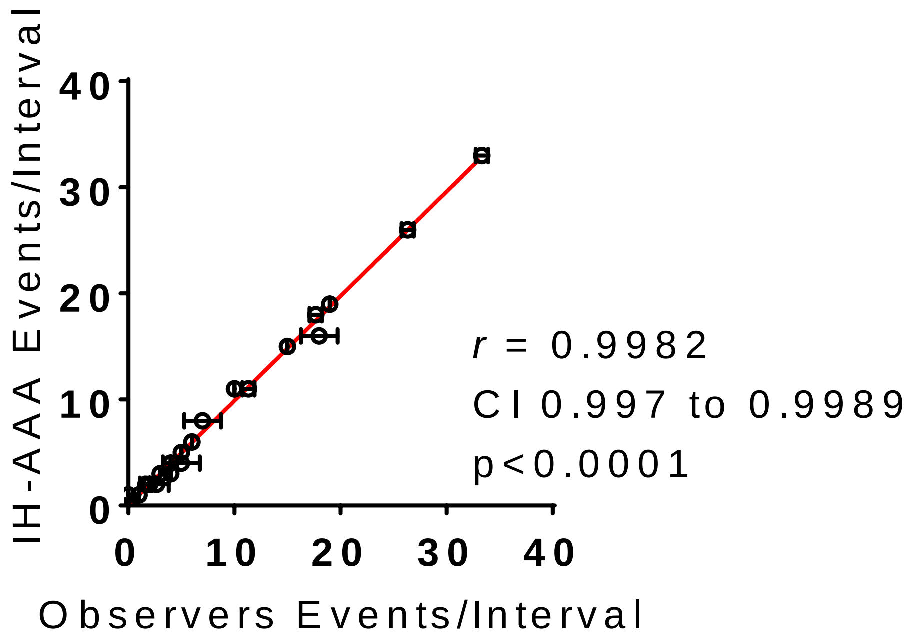

Figure 2. 14: A Pearson correlation comparing mean observer counts versus those calculated by IH Automated Analyses Algorithm (IH-AAA) for IH-SpO2<80

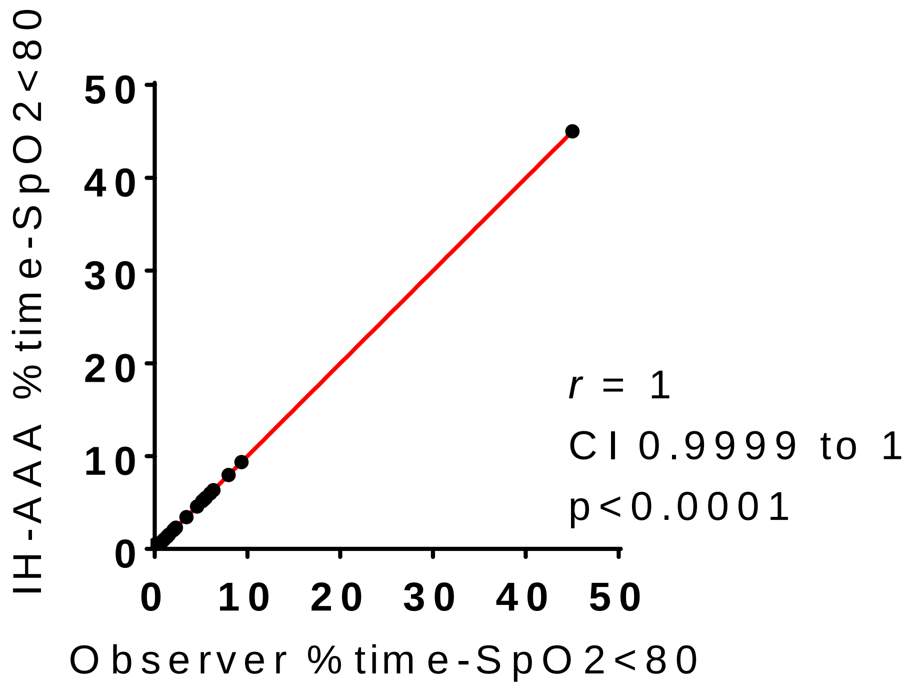

Figure 2. 15: A Pearson correlation comparing observer calculation versus IH Automated Analyses Algorithm (IH-AAA) for %time-SpO2<80

CHAPTER 3: PRENATAL OPIOID EXPOSURE AND INTERMITTENT HYPOXEMIA

Citation: This chapter was published as an original manuscript in Frontiers in Pediatrics: This chapter was not altered from the original publication.

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I. Introduction

Intermittent hypoxemia (IH) is defined as brief, episodic drops in oxygen saturation (SpO_2) (1, 2). Preterm infants are at increased risk for IH due to their respiratory control instability/apnea of prematurity superimposed on immature lung structure/function. Intermittent hypoxemia in preterm infants can persist beyond discharge from the neonatal intensive care unit (NICU) (4). Brief episodes of oxygen desaturations may seem clinically insignificant, but these IH episodes, occurring up to hundreds of times per day, have a cumulative effect on neonatal morbidity and mortality. There is ample evidence showing a significant effect of IH on neurocognitive handicap, decreased neuronal integrity, increased inflammation and oxidative stress, and impaired growth (32, 41). Furthermore, IH has been linked to severe retinopathy of prematurity and long term neurodevelopmental impairment such as worse language and motor outcomes (2, 3, 43, 44) (45). The clinical relevance of IH is a relatively new observation with the advent of high-resolution pulse oximeters and assessing factors that influence IH is imperative.

There is a rise in substance misuse in the USA reaching a nationwide epidemic (65-70). There is an urgent need to understand the impact of prenatal opioid exposure on neonatal outcomes (41). Opioid exposure is associated with long-term neurobehavioral and developmental impairment in infants (71-78). Opioids are known to suppress breathing and respiratory effort especially in neonates (22). Since most mothers who misuse opioids have also been found to smoke and use poly-drugs that affect breathing pattern, it has been challenging to assess the isolated effect of prenatal opioid exposure on respiratory outcomes. Prenatal tobacco exposure alters respiratory control and worsens lung function (25-28, 79). Prenatal exposure to other illicit drugs such as cocaine perturbs maturation of respiratory control, resulting in disruption of postnatal respiration (24). Only few studies were able to assess the effect of isolated opioid exposure on neonatal respiratory outcomes. However, these studies included mostly later preterm and term infants or were limited to short monitoring times and small sample sizes (17, 80). In this study, we utilize continuous high resolution pulse oximeters to assess the relationship between isolated prenatal opioid exposure and IH in preterm infants during the first 2 months of life.

II. Methods

Study Design and Data Collection

Oxygen saturation data were prospectively collected from 130 preterm infants less than 30 weeks gestational age (GA) admitted to our level 4 NICU between November 2014 and April 2017. We used high resolution pulse oximeters (Radical 7: Masimo, Irvine, CA) set at 2 second averaging time and 1Hz sampling rate to continuously monitor patients during the first 8 weeks of life. In order to differentiate intermittent from sustained hypoxemia, we included events between 4-180 seconds (1). The exact threshold below which IH is clinically significant is controversial. A drop in SpO2 to less than 80% is widely considered to be clinically relevant (1-3). Therefore, the primary outcome measure was defined as percent time spent with SpO_2 below 80% (%time-SpO2<80). The secondary outcome measure was defined as the number of severe IH events with SpO_2 less than 80% (IH-SpO_2<80). Other outcome measures such as length of stay and neonatal morbidities were collected.

Pulse oximeters were equipped with serial data recorders (Acumen Instruments Corp) for continuous data collection. Novel programs were utilized to filter and analyze data (Matlab, Natick, MA) (1, 61). Data with artifacts were excluded. Only SpO_2 data with good signal were included in the analyses. Preterm infants less than 30 weeks GA were included. Infants with major congenital malformations were excluded.

Data related to substance misuse and tobacco use were retrospectively collected from medical charts. If a mother chronically used prenatal opioids and/or the maternal/neonatal drug screens were positive for opioids, then the infant was considered for screening. Infants were then excluded from the study if the mother used tobacco, alcohol, or other drugs (such as cannabis); i.e., in order to assess for isolated opioid exposure, patients with any other exposure were excluded. Infants in our cohort who were not exposed to opioids, tobacco, or other drugs served as controls. Neonatal meconium or urine drug screens are performed in the immediate newborn period. Positive drug screens due to opioids and other medications used for pain or sedation during delivery were excluded, as they do not represent prenatal misuse. Tobacco and alcohol use were collected from mothers' medical records, as the toxicology screens at our hospital do not test for alcohol or tobacco exposure. The study was approved by the University of Kentucky Institutional Review Board, and informed consent was obtained prior to SpO_2 data acquisition.

Statistical Analysis

Descriptive statistics for continuous variables are presented as either the mean with standard deviation or median with interquartile range, and frequencies and percentages are given for categorical variables. Two-sample t-tests and

Wilcoxon two-sample tests were used to compare opioid exposure to non-exposure with respect to continuous variables, and chi-square or Fisher's exact tests were used for categorical variables. To compare opioid exposure to non-exposure with respect to IH measures over time, we utilized multivariate Gaussian linear modeling in order to account for repeated measurements from subjects, and to adjust for the potential confounders of gestational age, birth weight, APGAR score at 5 minutes of life, gender, and the use of prenatal steroids. In order to meet statistical assumptions in these models, the square root of the IH measures was taken. Furthermore, weekly observations were weighted by the percentage of time IH was tracked during the given week. Analyses were conducted in SAS version 9.4 (SAS Institute, Cary, N.C.), and all tests were two-sided with a 5% significance level.

III. Results

Of the 127 infants in our database with complete data sets, 19.7%, 29.1%, and 4.7% were prenatally exposed to opioids, tobacco and cannabis, respectively. None were exposed to alcohol, cocaine and other illicit drugs. Opioid exposed infants were positive for buprenorphine metabolites (64%), oxycodone (16%) and other opioids such as heroin and fentanyl (20%). A total of 82 infants qualified for analysis as they were either unexposed to any illicit drug/tobacco (n=68) or exposed to opioids only (n=14). **Figure 3.1** presents the flow diagram for patient eligibility and exclusion.

There were no significant differences in baseline characteristics as presented in Table 1. The mean GA was 27 weeks in both groups. There were no significant differences in birth weight, gender and Apgar scores at 5 minutes of life. The vast majority of infants received prenatal steroids with no difference between groups. There were no significant differences in respiratory outcomes and neonatal morbidities between groups as presented in Table 2. Our cohort included preterm infants less than 30 weeks GA. Essentially all infants had respiratory distress syndrome and received surfactant. Severe bronchopulmonary dysplasia, postnatal steroids use for lung disease, and oxygen need at 28 days, 36 weeks postmenstrual age and at discharge did not differ between opioid exposed and unexposed groups (all p=NS). Other neonatal morbidities such as patent ductus arteriosus, late onset sepsis, and necrotizing enterocolitis did not differ between groups (all p=NS). None of the exposed infants died versus 9 deaths in the unexposed group (p= 0.35). The median length of stay was 17 days longer in the opioid group (85 days) compared to unexposed group (68 days); however, the results were not statistically significant (p=0.32).

There was a statistically significant increase in our primary outcome measure, %time-SpO_2<80, as represented in **Figure 3.2**. The estimated difference in the means of the square root of %time-SpO_2<80 was 0.23 [95% CI: (0.03, 0.43), p=0.03]. The mean number of IH events was estimated to be 2.95 [95% CI: (-0.35, 6.25), p-value = 0.08] higher in the opioid exposed group, as represented in **Figure 3.3**; however, this did not reach statistical significance. Note that these results represent the square root of means in order to meet statistical assumptions in these models; estimated medians for IH measures are calculated using our model results and are presented in **Figures 3.2B and 3.3B**. Given increased death in the unexposed group, we then analyzed data excluding deaths, and results were similar. Specifically, there was a statistically significant increase in our primary outcome measure (%time-SpO_2<80) in the opioid exposed compared to the unexposed group, with an estimated mean difference (square root) of 0.24 [95% CI: (0.05, 0.44), p-value = 0.02]. Furthermore, the mean number of IH events was estimated to be 2.98 [95% CI: (-0.20, 6.16), p-value = 0.07] higher in the opioid exposed group, not quite reaching statistical significance.

IV. Discussion

These results suggest that prenatal opioid exposure is associated with increased IH measures compared to unexposed preterm infants. This study has two main findings. First, interestingly, the increased IH measures in opioid exposed infants persisted beyond the early postnatal period. Preterm infants were continuously monitored with high resolution pulse oximeters during the first 2 months of life. Second, we had the unique opportunity to assess the relationship between isolated opioid exposure and respiratory instability in preterm infants. It was challenging in the past to assess the relationship between isolated prenatal opioid exposure and respiratory outcomes/IH, as the majority of women who use opioids also smoke or misuse poly-drugs. Given our cohort demographics, we had the ability to report this association in infants exposed to opioids only.

Another interesting secondary finding in our study is the steady increase in IH in the first month of life before plateauing and then decreasing. This natural progression of IH has been described before from another cohort of preterm infants less than 28 weeks GA (1, 2). Our study replicates this finding from a new cohort of preterm infants less than 30 weeks GA. The rise in IH may be related to peripheral chemoreceptor dysregulation and development of lung disease (7).

Patients in our opioid exposed and unexposed groups did not significantly vary in terms of baseline characteristics (such as age, weight, gender) and neonatal morbidities (such as lung disease, patent ductus arteriosus, late onset sepsis and necrotizing enterocolitis). In addition, we adjusted in the model for factors that may influence oxygenation in preterm infants such as GA and prenatal steroids. The finding of 9 deaths in the unexposed group compared to no deaths in the opioid exposed group may be due to chance. Secondary analyses excluding deaths showed similar results with increased IH in the opioid exposed group. A significant secondary finding in this study is the high prevalence of tobacco and drug exposure in our cohort of preterm infants. The frequency of opioid exposure in our preterm population is higher than previously reported, thus creating urgency toward addressing this significant problem in this vulnerable patient population (65, 67-70).

There are multiple proposed mechanisms by which prenatal opioid exposure may affect breathing patterns and subsequent persistent IH in preterm infants. Prenatal opioid exposure alters the response to carbon dioxide and depresses central respiratory control centers (17-21); a main driver for respiratory output. Olsen et al demonstrated a blunted response to carbon dioxide in methadone exposed infants compared to controls (17). Ali et al compared the response to hypercarbia among three groups of term patients who were exposed to tobacco/substance misuse, tobacco alone, and unexposed controls. The authors showed a lower increase in central respiratory drive in response to hypercarbia in infants exposed to substance misuse as compared to tobacco alone and unexposed controls (18). Another mechanism that explains our results may be related to in utero hypoxia related to opioids. Prenatal opioids, especially street heroin, cause chronic intrauterine hypoxia leading to brainstem gliosis, resulting in injury to the central respiratory network. This may lead to respiratory instability and subsequent IH (19). Finally, data from animal models showed that exposure to opioid agonists caused down-regulation of placental neurotransmitter receptors (23). Abnormalities or depletion of receptor sites, especially if the same process occurs in the fetal brain, could impair the function of the normal neonatal respiratory control network leading to frequent or prolonged apnea and subsequent IH.

Many studies have assessed the impact of prenatal opioid exposure on sudden infant death syndrome (SIDS) in infants with controversial results. This study does not address SIDS; rather, it focuses on IH, the end result of apnea of prematurity. However, the mechanism by which prenatal opioid exposure is associated with increased SIDS and IH may be similar. Although our study period focused on the inpatient setting, it is plausible that opioid exposed infants continue to have increased cardiorespiratory events/IH after discharge.

Interestingly, compared to unexposed infants, opioid exposed infants had a trend toward longer length of stay (68 versus 85 days, p=NS), which may be related, in part, to persistent cardiorespiratory events.

A major limitation of this study is that data related to exposure were retrospectively collected. Another limitation is a lack of reporting daily caffeine use and daily respiratory support settings. At our center, virtually all infants with GA less than 30 weeks are started on caffeine therapy. Furthermore, our study focused on IH events and lacked reporting of apnea and bradycardia events. Lack of addressing heart rate is a limitation since bradycardia events may be associated with poor long term outcomes (3). Another limitation is the small sample size; however, our sample size of isolated opioid exposure is relatively large compared to existing literature. This is a single center study; hence, our results may not be generalizable. Finally, we did not compare the long term neurodevelopmental outcomes for exposed versus unexposed infants.

V. Conclusion

There is rising evidence linking IH to neonatal morbidities and impairment. However, the exact threshold (frequency, duration, severity) by which IH leads to injury in preterm infants needs further investigation; i.e., any increase in IH may be associated with impairment in preterm infants. Furthermore, there is a need to understand factors, such as prenatal opioid exposure, that may influence IH and subsequently increase neonatal morbidities. In this study, we show an association between prenatal opioid exposure and increased IH measures in preterm infants. Studies to address the relationship between opioid exposures, IH, and long term neurodevelopmental outcomes are imperative. Given the rising epidemic of opioid misuse in the USA, understanding the relationship between opioid exposure, IH and long term impairment is imperative. A larger prospective study aimed at understanding these relationships may have a direct impact on short and long term management of preterm infants.

VI. Acknowledgements

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Table 3. 1: Baseline Characteristics	**Opioid Exposed** N=14	**Unexposed** N=68	**p-Value**
Gestational age (weeks)	27.0 ± 2.1	27.0 ± 1.6	0.97
Birth weight (grams)	948 ± 263	928 ± 247	0.79
Male	6 (43%)	23 (34%)	0.54
Apgar 5 min	7 (6, 7.5)	6 (5, 7)	0.21
Prenatal steroids	12 (86%)	61 (91%)	0.62
Mean ± SD, Median (Interquartile range)			

Table 3. 2: Neonatal Morbidities and Outcomes	**Opioid Exposed** N=14	**Opioid Unexposed** N=68	**p-Value**
Received Surfactant	14 (100%)	62 (91%)	0.58
Respiratory distress syndrome	14 (100%)	67 (99%)	1
Oxygen at 28 days of life	10 (71%)	39 (57%)	1
Oxygen at 36 weeks corrected age	7 (50%)	19 (28%)	0.26
Oxygen at discharge	9 (64%)	30 (44%)	0.18
Severe Bronchopulmonary Dysplasia	9 (64%)	27 (46%)	0.21
Postnatal steroids use for lung disease	6 (43%)	19 (29%)	0.35
Pneumothorax	1 (7%)	2 (3%)	0.43
Patent Ductus Arteriosus	8 (57%)	24 (35%)	0.13
Necrotizing Enterocolitis	0 (0%)	2 (3%)	1
Late Onset Sepsis	3 (21%)	9 (13%)	0.43
Mortality	0 (0%)	9 (13%)	0.35
Length of Stay (days)	85 (59, 101)	68 (56, 91)	0.32
Frequency (%), Median (Interquartile range)			

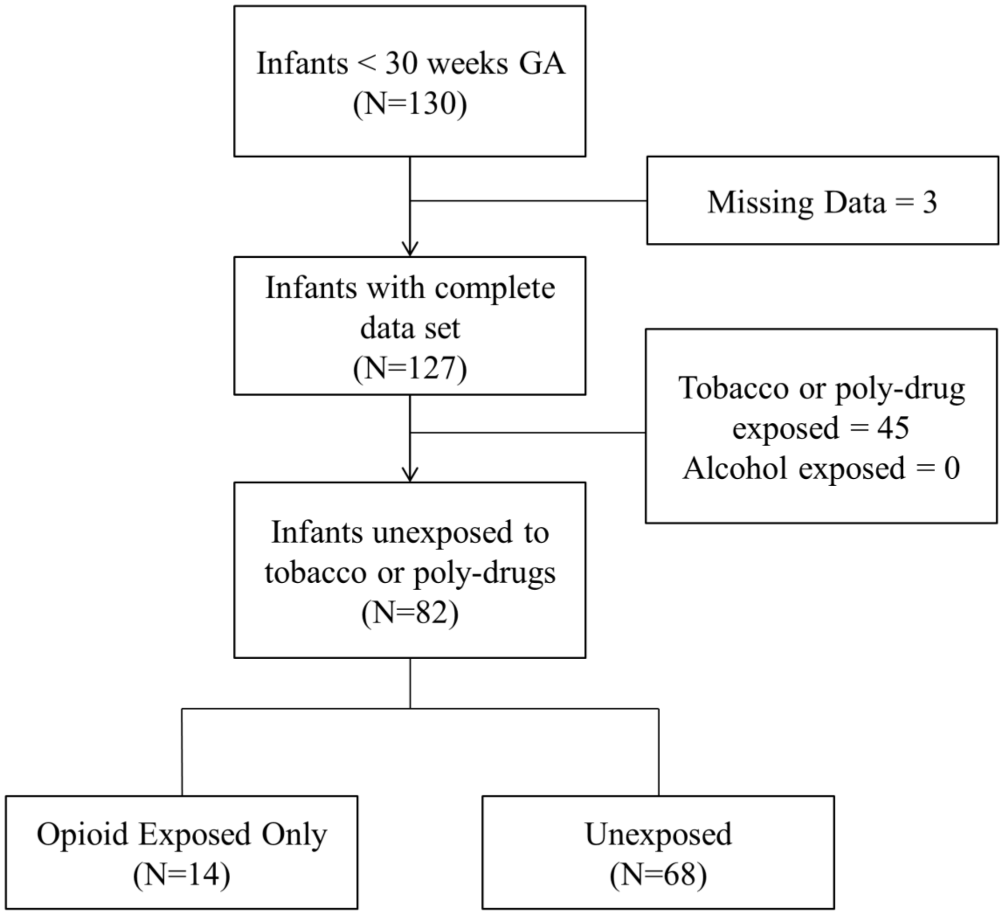


Figure 3. 1: Flow diagram for patient eligibility.

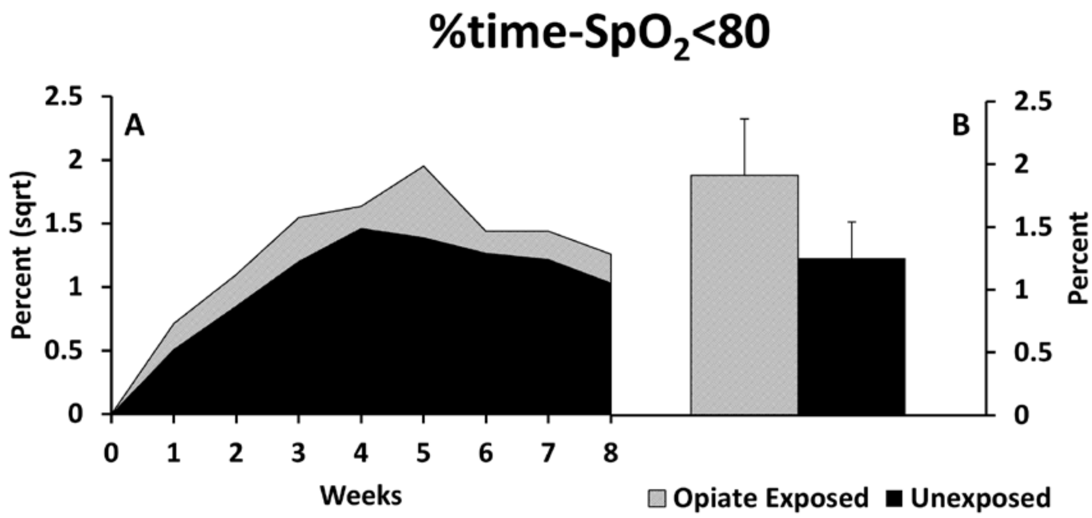


Figure 3. 2: Comparison of %time-SpO2<80 between opioid exposed and unexposed.

A) Preterm infants exposed to prenatal opioids had increased time spent with oxygen saturation less than 80% (%time-SpO_2<80) compared to unexposed infants (p=0.03). The model adjusted for gestational age, birth weight, gender, prenatal steroids, and Apgar scores at 5 minutes of life. **B)** This figure demonstrates the estimated average %time-SpO_2<80 medians in both groups calculated using the adjusted model results. Sqrt, square root.

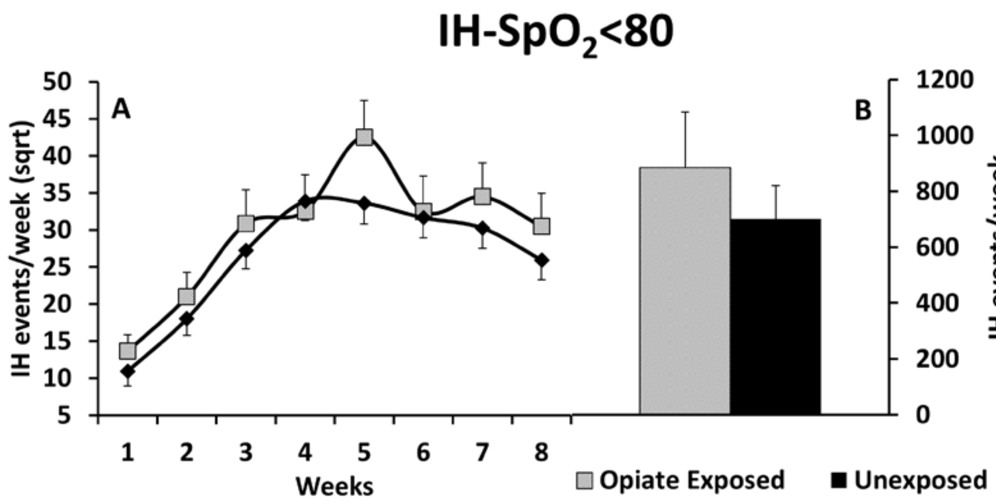


Figure 3. 3: Comparison of IH-SpO2<80 between opioid exposed and unexposed.

A) Preterm infants exposed to prenatal opioids did not have a significant increase in number of intermittent hypoxemia (IH) events per week (IH-SpO_2<80) compared to unexposed infants (p=0.08). The model adjusted for gestational age, birth weight, gender, prenatal steroids, and Apgar scores at 5 minutes of life. **B)** This figure demonstrates the estimated average IH-SpO_2<80 medians of opiate exposed versus unexposed preterm infants calculated using the adjusted model results. Sqrt, square root

CHAPTER 4: INTERMITTENT HYPOXEMIA IS ASSOCIATED WITH INCREASED SERUM C-REACTIVE PROTEIN IN PRETERM INFANTS

I. Introduction

Systemic inflammation perturbs breathing patterns, worsens apnea and cardiorespiratory events. There is ample evidence in preterm infants and animal models demonstrating that systemic inflammation increases apnea and subsequent IH (29, 30, 81-83). Furthermore, apnea and subsequent increase in IH is often an early sign of inflammatory processes such as sepsis and necrotizing enterocolitis (NEC) in the neonatal intensive care unit (NICU) (29, 30, 81-83). However, interestingly, based on animal studies, the relationship between inflammation and IH may be bidirectional, **Figure 4.1** (7). I.e. IH may be pro-inflammatory itself.

Mounting evidence, links IH with both short and long term neonatal morbidities such as retinopathy of prematurity, sleep disordered breathing, neurodevelopmental impairment and increased mortality (2, 3, 16, 45, 46, 54, 55, 84-86). Intermittent hypoxemic episodes due to obstructive sleep apnea in adults are associated with increased levels of inflammatory biomarkers (87-93). Multiple inflammatory markers have been tested in adults. There is ample evidence demonstrating increased C-reactive protein (CRP) serum levels in adult patients with obstructive sleep apnea (87-93). There are no studies in preterm infants to support IH being pro-inflammatory; however, there is rising evidence from neonatal animal models suggesting IH is pro-inflammatory. For example, IH exposed rat-pups had increased serum inflammatory biomarkers such as IFN-γ and IL-1β (32). Chronic IH in rodents increases inflammation (monocyte chemoattractant protein-1, IL1β, TNF-α, and a 5 fold increase in IL-6) in the carotid body chemoreceptors altering their function and subsequently affecting respiratory control and apnea (14, 33-35). We wanted to assess the relationship between IH and serum CRP for the first time in human preterm infants.

II. Methods

Study Design and Data Collection

Oxygen saturation (SpO2) data were prospectively collected from 26 preterm infants less than 30 weeks gestational age (GA) admitted to our level 4 NICU between November 2014 and September 2015. We used high resolution pulse oximeters (Radical 7: Masimo, Irvine, CA) set at 2 second averaging time and 1Hz sampling rate to continuously monitor patients. Pulse oximeters were equipped with serial data recorders (Acumen Instruments Corp) for continuous data collection. Novel programs were utilized to filter and analyze data (Matlab, Natick, MA) (1, 61). Data with artifacts were excluded. Only SpO_2 data with good signal were included in the analyses. Infants with major congenital malformations were excluded.

We collected blood samples at 30 days of life to assess for systemic inflammation at peak of IH. Blood samples for CRP were collected for research purposes and not for clinical purposes; i.e. not because there was a concern for illness or change in status. High sensitivity CRP was analyzed using commercial ELISA kits. Data related to other morbidities that may increase CRP such as sepsis and necrotizing enterocolitis (NEC) were collected. Other demographics, morbidities, and respiratory characteristics were collected from medical records.

Statistical Analyses

Average IH measures (IH profile) were calculated for the week prior to CRP collection. Plots of CRP and IH measures were performed to identify outliers. We assessed the relationship between IH and CRP using GraphPad

Prism 7. Statistical analyses were based on Pearson correlation and linear regressions. Since the exact threshold below which IH causes injury is unknown, we calculated an IH profile reflecting the continuum of the problem. We did not set a lower or upper limit for IH in this study. A drop in SpO2 to less than 80% is widely considered to be clinically relevant (1-3) and therefore was selected as the primary outcome measure. Other thresholds included SpO2 of 85% and 90%. Furthermore, 6 different IH duration intervals were calculated: 1-59 seconds, 60-119 seconds, 120-179 seconds, 180-239 seconds, 240-299 seconds, more than or equal to 300 seconds.

III. Results

Of the 26 infants included, 25 had SpO2 data available during the week prior to CRP collection. Blood samples for CRP analyses were obtained at median day of life (DOL) 30 (IQR 29-32 days). Scatter plots identified 2 outliers with CRP values of 20.829mg/dL and 69.128mg/dL **(Figure 4.2**). One of the outliers had sepsis within 2 weeks (11 days) prior to the CRP collection date. Three other patients had sepsis but occurred more than 2 week prior to our assessment. No patients had NEC. Plots for IH measures to identify outliers are presented in **Figures 4.3**.

Median GA is 27 weeks (Interquartile Range (IQR) 26 - 28 weeks). Median birth weight is 980 grams (IQR 763 - 1230 grams). There were no small for gestational age (SGA) infants. Median weight at the time of CRP is 1220 grams (IQR 900 – 1440 grams). Median CRP is 0.236mg/dL (IQR 0.025 - 1.648 mg/dL). Respiratory support data are presented in **Table 4.1.**

There was strong positive correlation between our primary measure, %time-SpO2<80, and serum CRP levels **(Figure 4.4)**. The positive correlation between percent time below threshold and CRP persisted with higher SpO2 threshold of 85% and 90%. There was moderate positive correlation between our primary measure, IH-SpO2<80, and serum CRP levels **(Figure 4.5).** The positive correlation between IH events and CRP persisted with higher SpO2 threshold of 85%. There was a strong positive correlation between duration of events and CRP; i.e. the longer the IH events the higher the serum CRP **(Figure 4.6)**. Furthermore, there was a statistically significant positive correlation between primary outcome IH measures and CRP at the 6 different duration intervals examined (except for IH-SpO2<80 at 1-59 seconds, p-Value 0.06) **(Figures 4.7 and 4.8)**. The mean SpO2 and CRP had a strong negative correlation; i.e. the lower the mean SpO2 the higher the inflammatory marker **(Figure 4.9)**. There was no statistically significant correlation between IH mean nadir and CRP **(Figure 4.10A)**. There was moderate negative correlation between peak mean IH and CRP as represented in **Figure 4.10B**.

IV. Discussion

Our results show that increased IH is associated with increased systemic CRP. This relationship between IH and inflammatory markers is documented for the first time in human preterm infants. Interestingly, most IH profile measures at all three thresholds and 6 duration categories correlated with worse inflammation. These results are clinically relevant as elevated inflammation during NICU stay, mainly 28 days, has been shown to be associated with worse long term outcomes (94).

Intermittent hypoxemia at all thresholds and durations was associated with increased serum CRP. The strongest correlation was between %time-SpO2<threshold and CRP. This is clinically relevant as percent time below threshold is available to the clinical team from the clinical pulse oximeter histograms. The frequency of IH correlated positively with CRP only with moderate and severe IH. The lower the mean SpO2 is the higher the serum CRP; an important finding with possible impact on the oxygen target saturation controversy in the NICU (2, 8, 37, 46, 95, 96).

C-reactive protein in comprised of five identical, non-covalently associated subunits (approximately 23 kD each) (97). C-reactive protein has both pro-inflammatory and anti-inflammatory characteristics (98). Both acute and chronic inflammation can increase CRP such as infection and metabolic stresses, respectively (99, 100). We chose CRP as our inflammatory measure for multiple reasons. First, compared to other markers of inflammation, CRP is widely used in the NICU with known reference ranges (101-105). Second, CRP is a good and stable marker for low grade inflammation (100, 106). Minor CRP elevations are considered a marker of low-grade inflammation, sometimes called subclinical inflammation or mini-inflammation. Low grade inflammation is the degree of inflammation we expected will be associated with increased IH. We utilized high sensitivity CRP commercial ELISA kits in order to measure low grade CRP changes. Third, multiple adult studies including meta-analyses have demonstrated increased CRP in patients with IH from obstructive sleep apnea (87-93).

Our results suggest that IH may be pro-inflammatory itself. Since IH is pro-inflammatory, that may lead to a spiral or snowball effect (positive feedback loop). Apnea events cause IH and subsequent systemic postnatal inflammation that is transferred to the respiratory control network, peripheral chemoreceptors and lungs. The postnatal inflammation leads to a further cycle of increased apnea events and consequently higher frequency of IH (**Figure 4.1**). Interestingly, this phenomenon may be in part responsible for the IH peak at 4-5 weeks of age (54).

This study has multiple strength including the prospective design and novel results. A major limitation for this study is the small sample size. However, the results were consistent at multiple IH thresholds and duration intervals suggesting a significant relationship between IH and increased CRP. Another limitation is the use of a single inflammatory marker. Future studies should focus on multiple inflammatory markers along the inflammation cascade. Other markers that have been associated with IH in adults with obstructive sleep apnea or IH in neonatal rodent models include, Interleukin (IL)-6, IL-1β, IL-8, Tumor Necrosis Factor (TNF)-α, Intercellular Adhesion Molecule (ICAM)-1, Interferon (IFN)-γ, Vascular Cell Adhesion Molecule (VCAM)-1 (14, 32-35, 89).

We demonstrate in this study, for the first time in preterm infants, that IH is associated with increased inflammation, namely CRP. While there is mounting evidence of adverse effects of IH, there has been no focus on inflammation in the cycle of events in preterm infants. Our findings are significant as the increased inflammation may be the mediator for increased morbidities and impairment in infants with IH (2, 3, 16, 45, 46, 54, 55, 84-86). Future larger studies that examine the role of inflammation as a mediator for long term injury from IH should be examined.

V. Acknowledgments

I thank all the team members mentioned in the acknowledgments section. I specially recognize Hong Huang MD, PhD for blood sample processing for C-reactive protein analyses. I thank the Gerber Foundation and Children's Miracle Network for funding sample analyses.

Table 4. 1: Respiratory Characteristics	**Frequency, n (%)**
Room Air	3 (12%)
Continuous Positive Airway Pressure	7 (28%)
Non-Invasive Nasal Ventilation	6 (24%)
Conventional Ventilation	9 (36%)
Oxygen Supplementation	12 (48%)

These respiratory setting were collected on the day of CRP measurement.

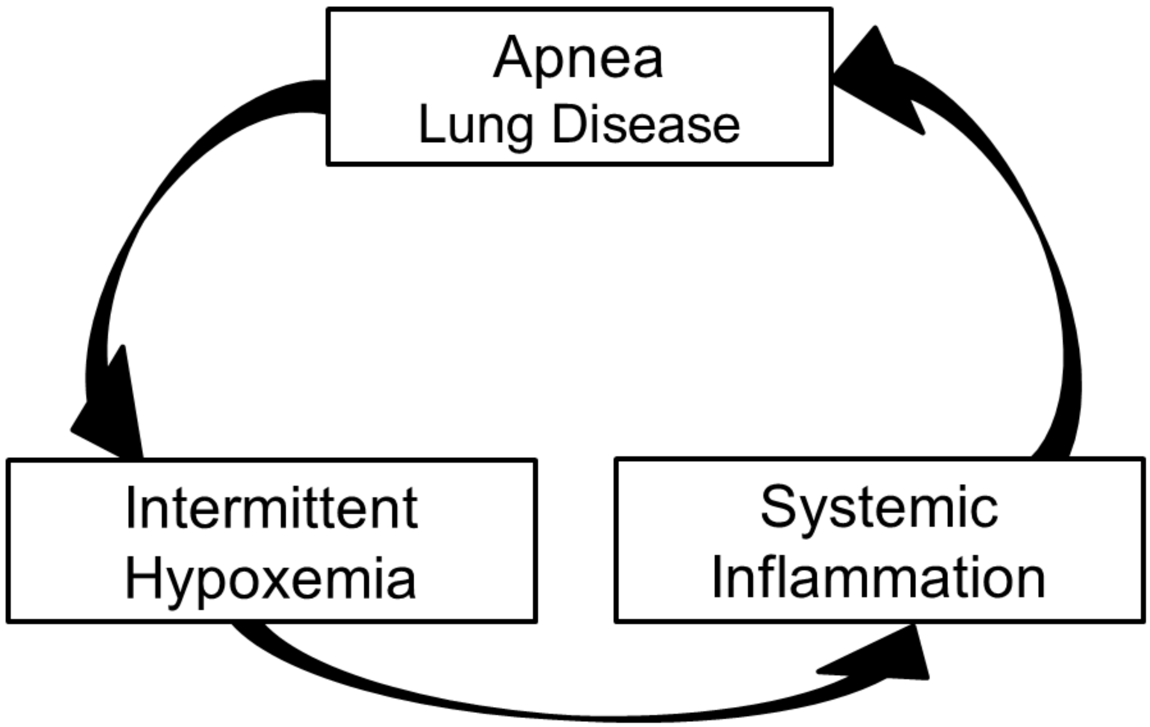


Figure 4. 1: Proposed vicious cycle related to apnea, IH and postnatal inflammation.

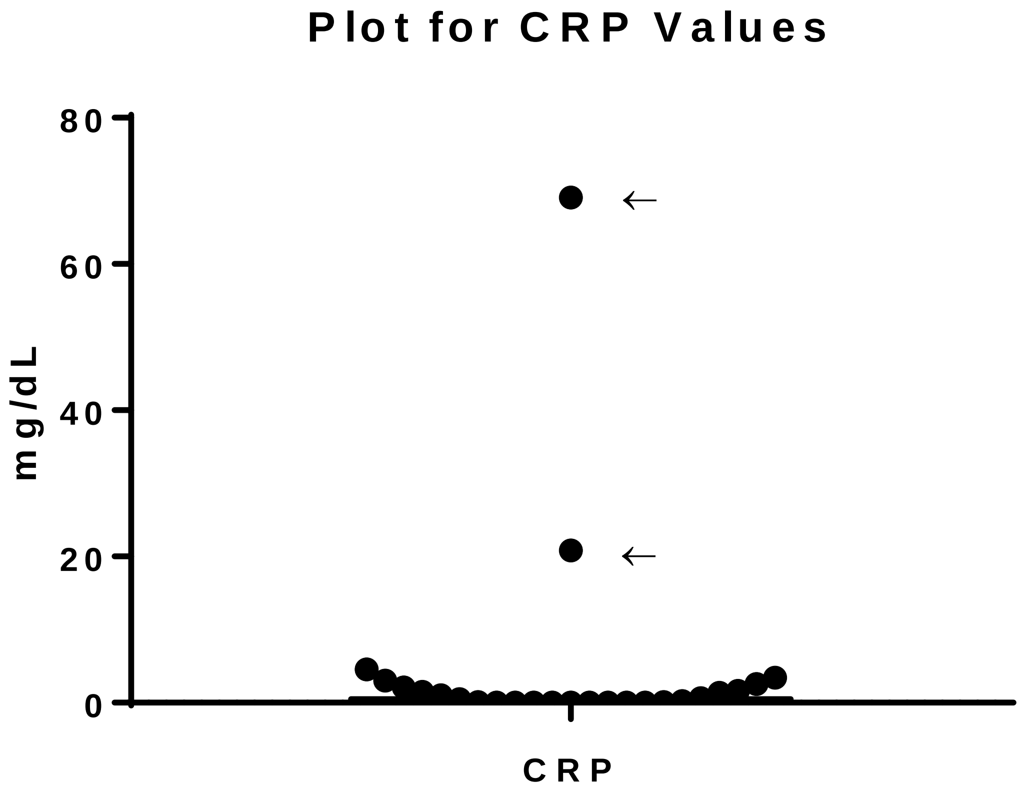


Figure 4. 2: Scatter plot for CRP levels in studied patient population

Two outliers were identified. Arrows identify outliers.

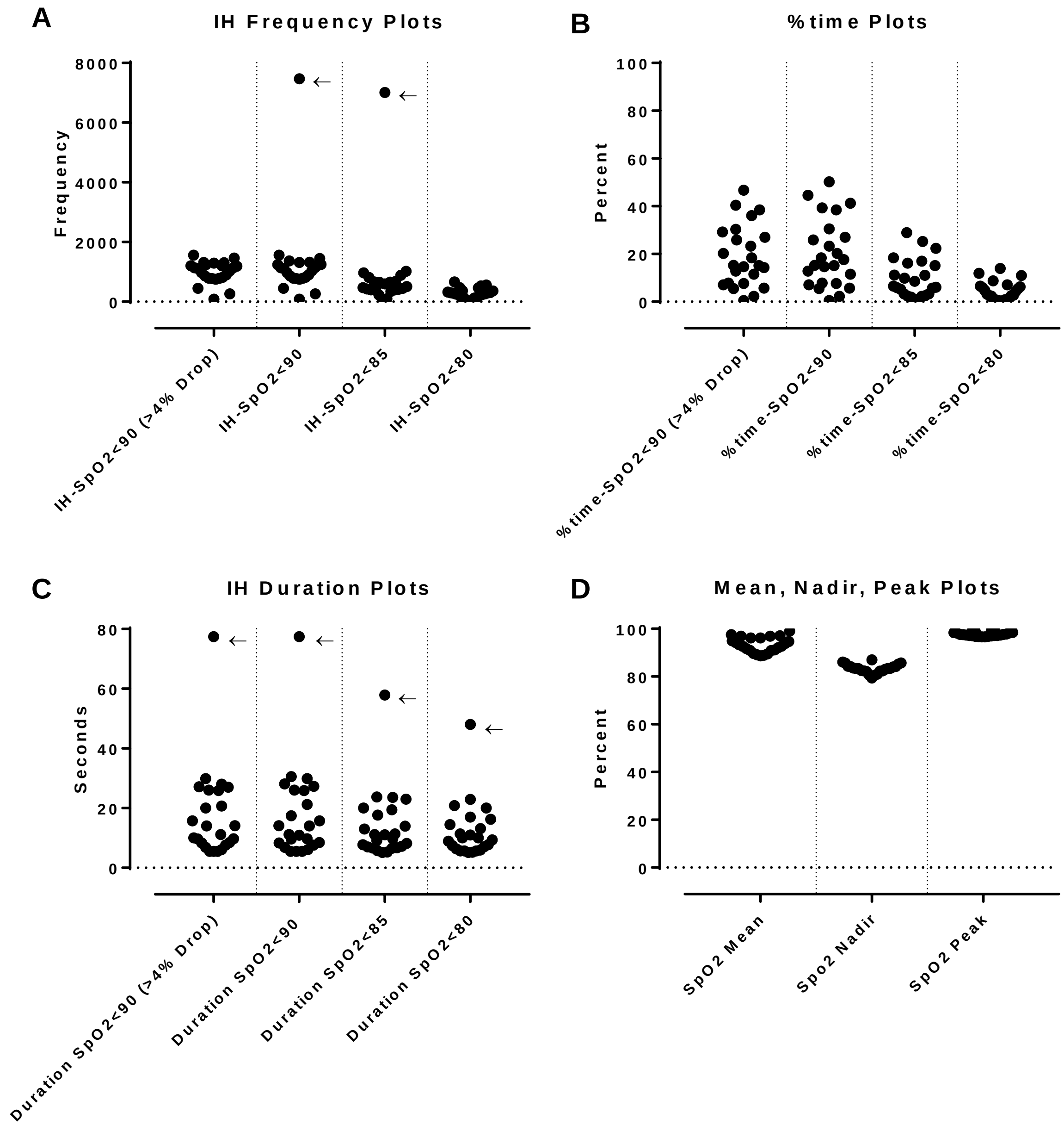


Figure 4. 3: Scatter plots for IH in studied patient population.

A) Frequency of IH. **B)** Percent time with SpO2 below threshold. **C)** Duration of IH. **D)** Mean, nadir and peak of IH. Arrows identify outliers.

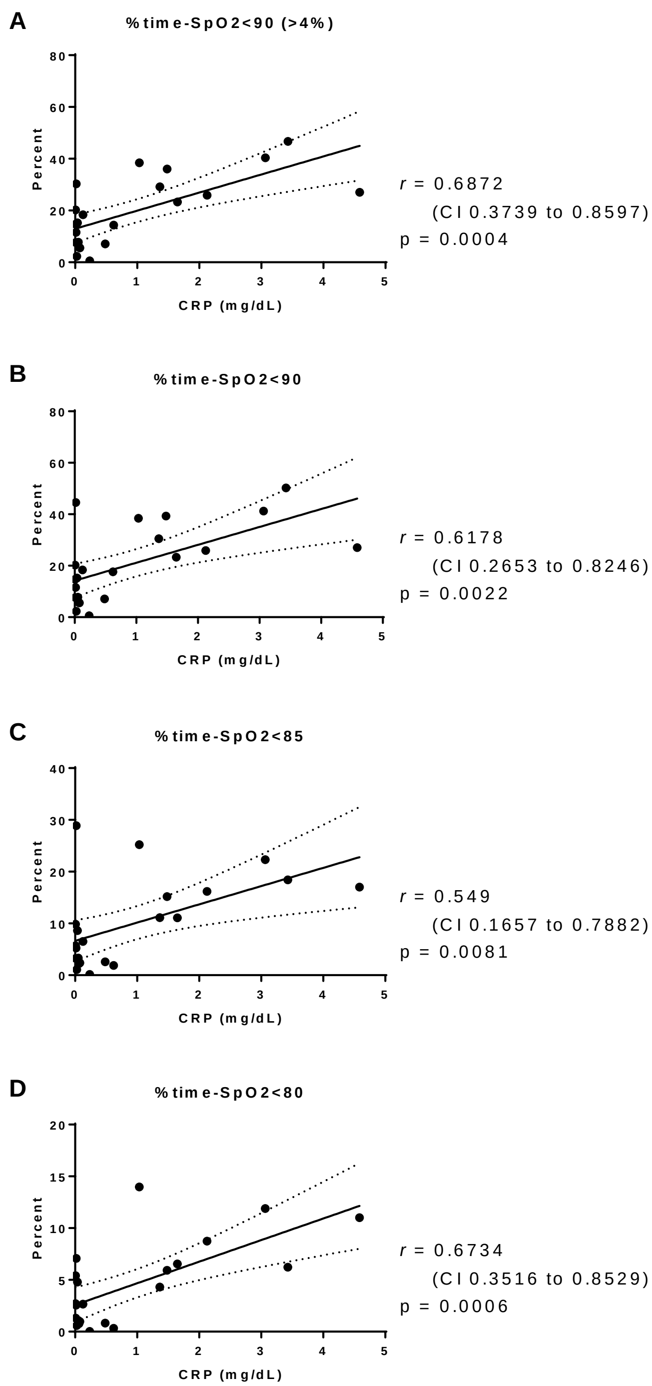


Figure 4. 4: Correlations comparing serum CRP and percent time below thresholds.
A) %time-SpO2<90 (>4% Drop) versus CRP. **B)** %time-SpO2<90 versus CRP. **C)** %time-SpO2<85 versus CRP. **D)** %time-SpO2<80 versus CRP. All correlations were statistically significant with p-values were less than 0.01.

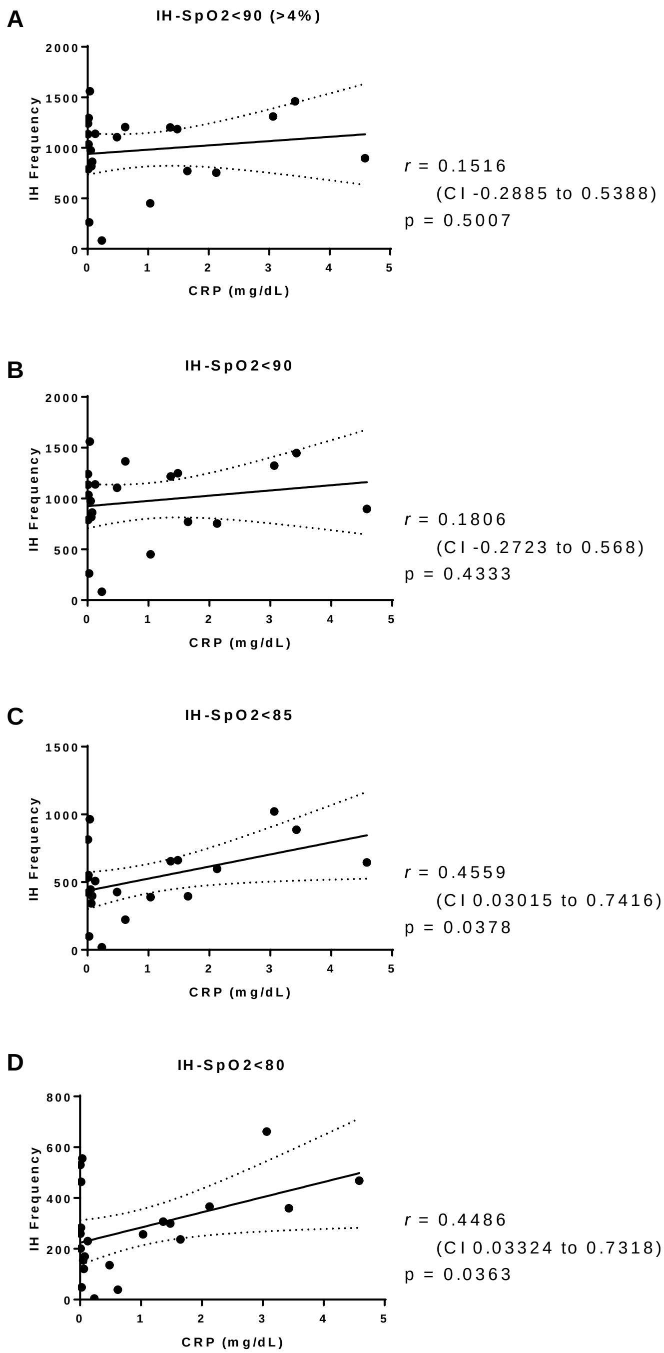


Figure 4. 5: Correlations comparing serum CRP and IH frequency.

A) IH-SpO2<90 (>4% Drop) versus CRP. **B)** IH-SpO2<90 versus CRP. **C)** IH-SpO2<85 versus CRP. **D)** IH-SpO2<80 versus CRP. The positive correlations between moderate (IH-SpO2<85), severe (IH-SpO2<80) IH and CRP are statistically significant (p-value less than 0.05).

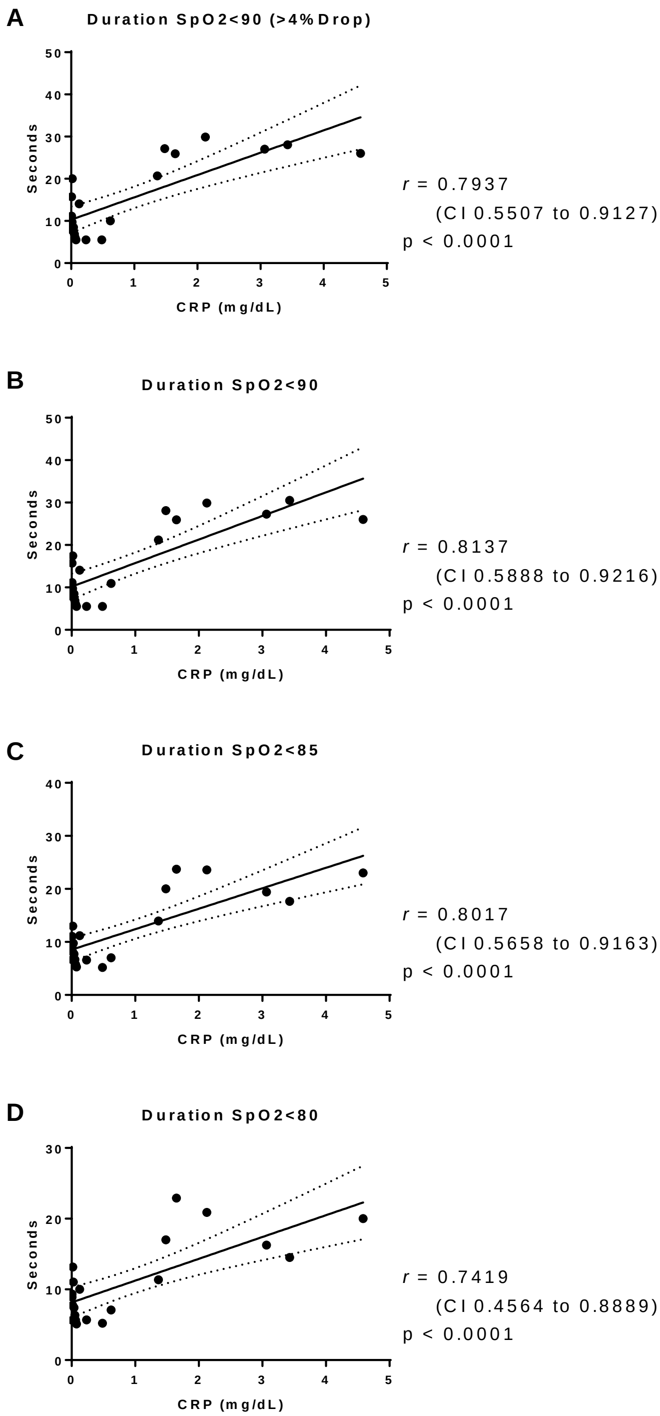
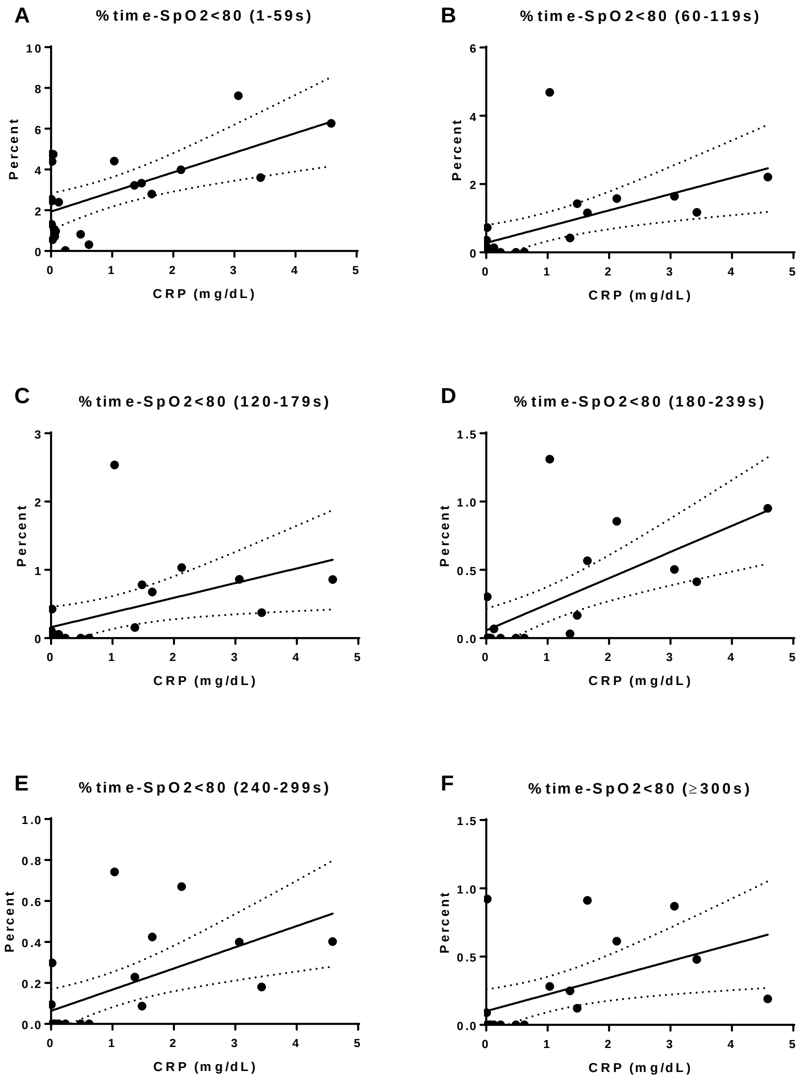


Figure 4. 6: Correlations comparing serum CRP and IH duration.

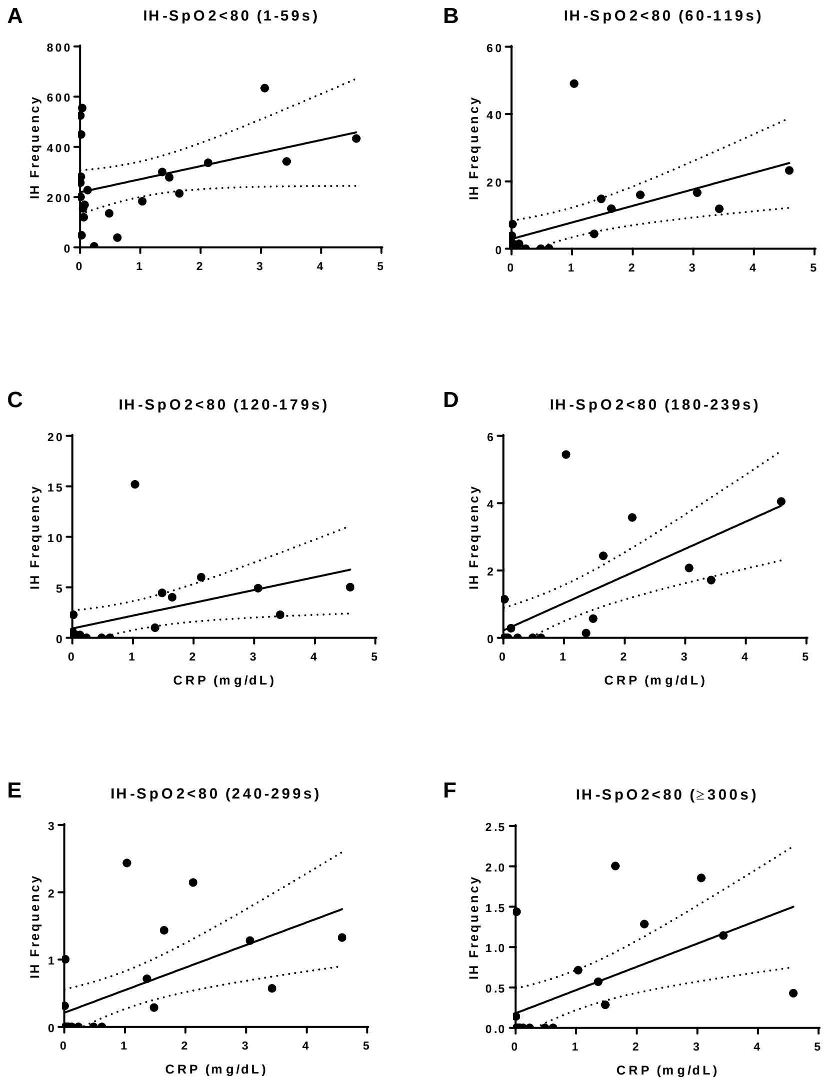
A) Duration SpO2<90 (>4% Drop) versus CRP. **B)** Duration SpO2<90 versus CRP. **C)** Duration SpO2<85 versus CRP. **D)** Duration SpO2<80 versus CRP. All correlations were statistically significant with p-values were less than 0.0001.



G. **CRP versus %time-SpO2<80**	**A (1-59s)**	**B (60-119s)**	**C (120-179s)**	**D (180-239s)**	**E (240-299s)**	**F (≥300s)**
r	0.6215	0.5594	0.4713	0.6632	0.5861	0.4917
95% CI	0.2709 to 0.8265	0.1804 to 0.7938	0.062 to 0.7449	0.3353 to 0.8477	0.2185 to 0.808	0.0884 to 0.7565
p-Value	0.002	0.0068	0.0268	0.0008	0.0042	0.0201

Figure 4. 7: Correlations comparing serum CRP and primary outcome measure %time-SpO2<80 at multiple duration intervals.

A) 1-59 seconds **B)** 60-119 seconds **C)** 120-179 seconds **D)** 180-239 seconds **E)** 240-299 seconds **F)** more than or equal to 300 seconds. **G)** This table presents the correlation coefficients (*r*), 95% confidence intervals (CI) and p-values for all intervals. All correlations were statistically significant.



G. CRP versus IH-SpO2<80	A (1-59s)	B (60-119s)	C (120-179s)	D (180-239s)	E (240-299s)	F (□300s)
r	0.4031	0.555	0.4671	0.6677	0.5819	0.57
95% CI	-0.02225 to 0.7049	0.1742 to 0.7914	0.05663 to 0.7425	0.3426 to 0.85	0.2124 to 0.8058	0.1954 to 0.7995
p-Value	0.0628	0.0073	0.0284	0.0007	0.0045	0.0056

Figure 4. 8: Correlations comparing serum CRP and primary outcome measure IH-SpO2<80 at multiple duration intervals.

A) 1-59 seconds **B)** 60-119 seconds **C)** 120-179 seconds **D)** 180-239 seconds **E)** 240-299 seconds **F)** more than or equal to 300 seconds. **G)** This table presents the correlation coefficients (*r*), 95% confidence intervals (CI) and p-values for all intervals. All correlations (except 1-59 seconds) were statistically significant.

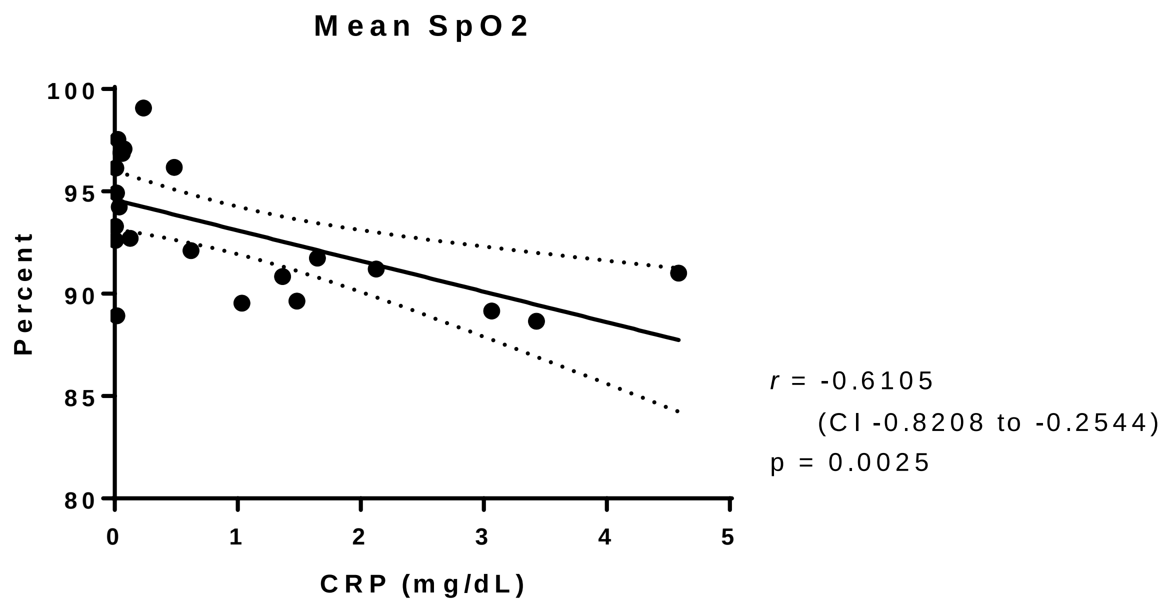


Figure 4. 9: Negative correlation between mean SpO2 and serum CRP.

The lower is the mean oxygen saturation to the higher the CRP level.

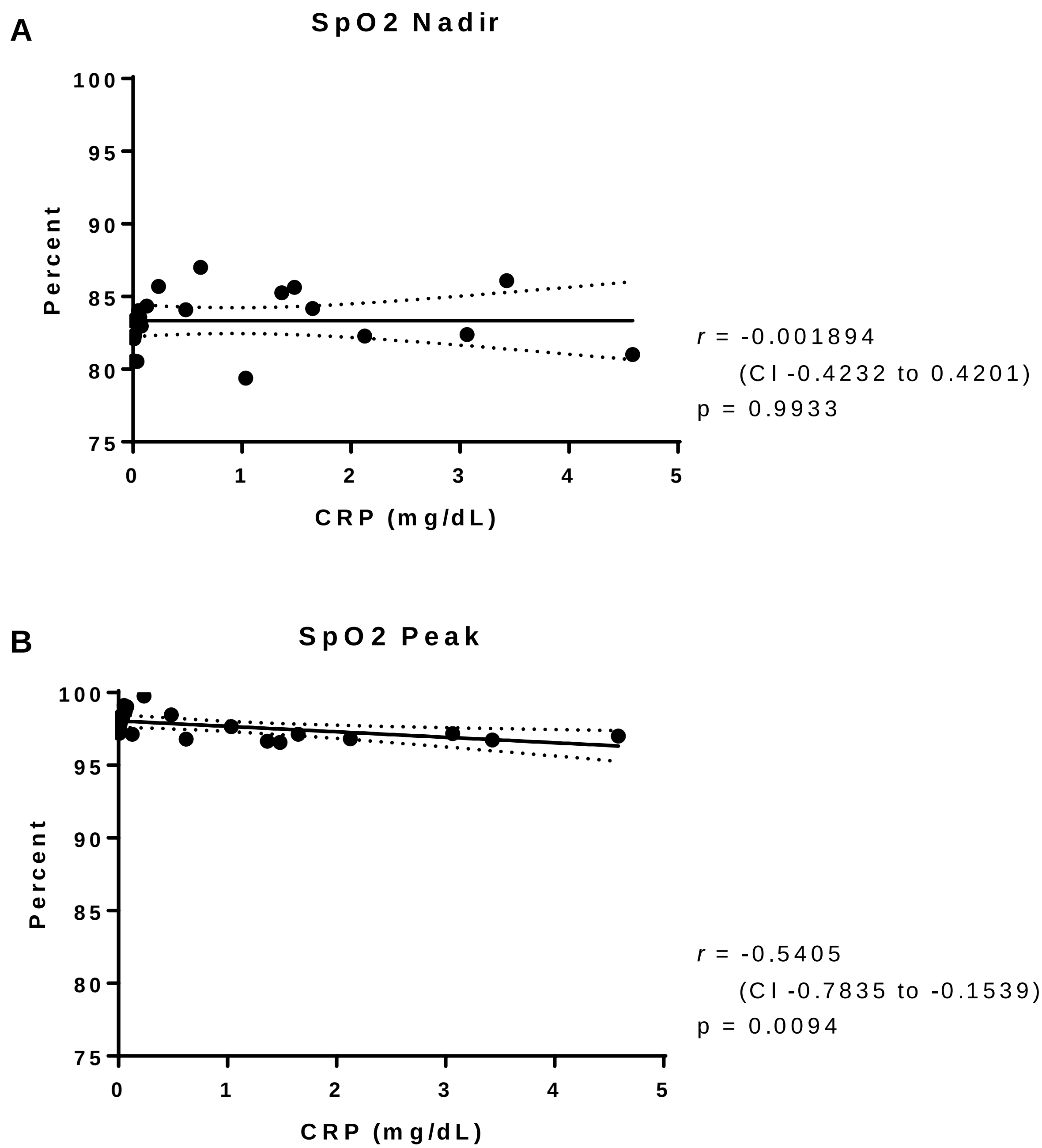


Figure 4. 10: Correlation between serum CRP and IH mean nadir/mean peak

A) Correlation between CRP and IH mean nadir. There was no statistically significant relationship. **B)** Significant negative correlation between mean peak SpO2 and CRP.

CHAPTER 5: MATERNAL CHORIOAMNIONITIS AND INTERMITTENT HYPOXEMIA IN PRETERM INFANTS

I. Introduction

Four million babies are born per year in the United States and close to a half million are premature (<37 weeks gestation) (107). The total societal economic cost of preterm birth is estimated at 26 billion dollars (108-110). Mean costs of care associated with extreme prematurity are nearly a quarter of a million dollars in the first 4 years of life; approximately 20 times higher than late preterm infants (111). Although significant progress has been made in the care of preterm infants, they continue to suffer from significant morbidities such as apnea, chronic lung disease (bronchopulmonary dysplasia (BPD)), retinopathy of prematurity (ROP), and neurodevelopmental impairments (NDI) (112, 113). Intermittent Hypoxemia (IH), contributes to the aforementioned morbidities (54). Brief episodes of oxygen desaturations may seem clinically insignificant, but these IH episodes, occurring up to hundreds of events/day, have a cumulative effect on morbidities and mortality. As presented in Chapter 1, mounting evidence, links IH with both short and long term neonatal morbidities such as ROP, NDI, sleep disordered breathing, and increased mortality (2, 3, 16, 45, 46, 54, 55, 84-86). Laboratory and animal data show IH results in increased inflammatory cytokines, increased free radicals and oxidative stress, increased white matter injury, neurocognitive handicap, and poor growth (4, 41, 54, 114-120).

Several predictors that influence IH have been investigated. Gestational age and IH are inversely related (5); with extremely preterm infants having the highest prevalence of IH. Infants with BPD have increased IH that persists on mechanical ventilation (1, 2, 7). Preterm infants with anemia are at increased risk for IH (1, 36); as hematocrit level decreases the probability of apnea/IH events increases. Intermittent hypoxemia natural progression changes with postnatal age. There is low IH frequency during the 1st week of life, followed by a progressive increase over weeks 2-3, peaks around 4-5 weeks, and decreases at weeks 6-8 (1, 2, 54). The reasons leading to the rise in IH postnatally are poorly defined but likely due to both developing lung disease and chemoreceptor dysregulation possibly due to inflammation and hypoxia (7).

Systemic inflammation increases apnea events and subsequently IH (29). Hofstetter et al. showed that systemic inflammation increased IL-1β that binds to its receptors located on endothelial cells of the blood brain barrier (29). Activation of IL-1β receptors leads to increased prostaglandins in the respiratory control network in the brain leading to respiratory depression/apnea and subsequent IH (29, 121). In addition, systemic inflammation worsens lung disease, decreases lung reserves leading to more IH in the presence of apnea (16). Interestingly, inflammation in the pulmonary system can be transmitted, likely through the vagal nerve, to the central respiratory network in the brain stem leading to further respiratory instability/apnea (30, 31, 122). In summary, inflammation increases apnea, worsens lung disease and subsequently increases IH.

Prenatal (intrauterine) inflammation is a common cause of preterm birth. Prenatal inflammation can happen with or without infection. Recently in 2016 the National Institute of Child Health and Human Development (NICHD) suggested the Triple I terminology referring to Intrauterine Inflammation, Infection or both in order to replace chorioamnionitis. For the purpose of consistency in this document, we will use the terminology of maternal chorioamnionitis (MC) as the main contributor to prenatal inflammation (123). Funisitis is inflammation of the umbilical cord (124). The majority of fetuses exposed to MC develop a systemic fetal inflammatory response syndrome (FIRS), usually defined as elevated serum interleukin-6 (IL-6). Fetal inflammatory response syndrome occurs due to the infant being in direct contact with affected amniotic fluid and/or inflammatory cell or cytokine transfer through placental circulation (125-127). Importantly, prenatal inflammation is reported to be a major contributor to morbidities e.g. apnea, BPD, ROP and brain injury (2, 127-150).

Pilot Assessment

A total of 30 infants less than 30 weeks GA were enrolled in this pilot trial to test the hypothesis that prenatal inflammation is associated with increased IH in postnatal life. Patients were monitored for 4 weeks. The presence of MC was collected from medical records through our Vermont Oxford Network (VON) database. Maternal chorioamnionitis as documented by the clinical team was considered positive in the data base. Blood samples were collected on day of life (DOL) 1 to measure high-sensitivity C-reactive protein (hsCRP); widely used in the NICU and a reliable measure of low grade inflammation (106, 151-154). Data related to MC and blood samples were available for 26 patients and, of those, 6 patients had MC. Median hsCRP on DOL 1 was more than 10 times greater in patients with MC (0.82 mg/dl) compared to no MC (0.071mg/dl), however, these differences were not statistically significant. Patients with MC had statistically significant increased IH during the study period **(Figure 5.1)** that persisted after adjusting for GA, gender, ethnicity, and severity of disease (SNAP-PE) scores.

A limitation of the pilot assessment was that MC was collected per the clinical team and may not meet all clinical chorioamnionitis criteria (123). Hence, we decided to define MC in the following study per placental pathology reports. We wanted to test the hypothesis that pathologic MC or funisitis are associated with increased IH in preterm infants. Since in funisitis, the umbilical cord is affected, we hypothesized a greater impact on IH in those infants.

II. Methods

Study Design and Data Collection

Oxygen saturation data were prospectively collected from preterm infants less than 35 weeks gestational age (GA) admitted to our level 4 NICU between November 2014 and July 2017. We used high resolution pulse oximeters

(Radical 7: Masimo, Irvine, CA) set at 2 second averaging time and 1Hz sampling rate to continuously monitor patients during the first 4 weeks of life. In order to differentiate intermittent from sustained hypoxemia, we included events between 4-180 seconds (1). The exact threshold below which IH is clinically significant is controversial. A drop in SpO2 to less than 80% is widely considered to be clinically relevant (1-3). Therefore, the primary outcome measures were defined as percent time spent with SpO_2 below 80% (%time-SpO2<80) and frequency of IH events with SpO2 drop below 80% (IH-SpO2<80).

Pulse oximeters were equipped with serial data recorders (Acumen Instruments Corp) for continuous data collection. Novel programs were utilized to filter and analyze data (Matlab, Natick, MA) (1, 61). Data with artifacts were excluded. Only SpO_2 data with good signal were included in the analyses. Preterm infants less than 30 weeks GA were included. Infants with major congenital malformations were excluded.

The presence of MC was collected from medical records. We chose our exposure as pathologic MC (inflammation noted in the placenta on pathology reports) or Funisitis (inflammation of the umbilical cord on pathology reports) in attempt to have more objective data. We did not include clinical MC since the data related to MC was collected retrospectively and hence clinical parameters may not be always appropriately documented.

Severe BPD was investigated as a secondary outcome measure given the controversial literature suggesting a relationship between MC and BPD. Severe BPD was defined per the National Institute of Child Health and Human Development (NICHD) criteria for respiratory status at 36 weeks corrected age (155). Respiratory settings and other demographic and baseline characteristics were collected from medical charts.

Statistical Analyses

Descriptive statistics for continuous variables are presented as either the mean with standard deviation or median with interquartile range (IQR), and frequencies and percentages are given for categorical variables. Two-sample t-tests and Wilcoxon two-sample tests were used to compare MC or Funisitis exposed to those not exposed with respect to continuous variables, and chi-square or Fisher's exact tests were used for categorical variables. Patients with exposure or MC or Funisitis were compared to unexposed. In addition, infants with MC only and Funisitis were separately compared to unexposed. To compare MC or Funisitis infants to those not exposed with respect to IH measures over time, we utilized multivariate Gaussian linear modeling in order to account for repeated measurements from subjects, and to adjust for the potential confounders of gestational age, small for gestational age (SGA) and the use of prenatal steroids. In order to meet statistical assumptions in these models, the square root of the IH measures was taken. Furthermore, weekly observations were weighted by the percentage of time IH was tracked during the given week. Analyses were conducted in SAS version 9.4 (SAS Institute, Cary, N.C.) and GraphPad Prism.

III. Results

A total of 151 patient included in our cohort were reviewed. Of those, 121 infants had placental pathology reports and respiratory/IH outcomes data. Baseline characteristics and comparisons between groups are presented in **Tables 5.1 - 5.4**. There was a difference in GA (p <0.0001) and birth weight (p= 0.0019) among groups. Deaths prior to discharge varies among groups (p=0.0011) with increased mortality in the exposed compared to unexposed infants. Other baseline characteristics did not vary among groups.

Contrary to our hypothesis, infants with funisitis had no major differences in IH measures compared to unexposed (**Figures 5.1 and 5.3**). The differences were most pronounced while comparing the MC only group versus unexposed (**Figures 5.2 and 5.4**). After adjusting for GA, SGA and prenatal steroids, statistically significant differences were noted while comparing the MC only versus unexposed (**Figures 5.3 and 5.5**). Severe BPD tended to be higher in any of the exposed groups compared to unexposed; however both unadjusted and adjusted differences were not statistically significant (**Figure 5.6**)

IV. Discussion

Our results related to IH measures in infants exposed to perinatal inflammation were inconsistent. The significant increase in IH in infants with clinical MC noted in our pilot study was not consistently replicated in infants with pathologic definition of MC. There were increased IH measures in infants exposed to pathologic MC and/or funisitis compared to unexposed infants. After adjusting for GA, SGA status and prenatal steroids, differences were statistically significant in the MC only group. Severe BPD did not vary among groups, however tended to be higher in pathologic MC and/or funisitis exposed infants compared to unexposed.

There were no differences in SGA status between groups, a major risk factor for increased IH. Infants with pathologic MC had lower GA and birth weight compared to those unexposed. This is an expected finding given that the incidence of prenatal inflammation is inversely related to GA, ranging from 75% to 35% in 23 and 29 week GA infants respectively (127, 156-159). This difference in GA may be responsible for the increased unadjusted IH in the exposed groups. Statistically significant higher IH persisted in the MC only group after adjusting for GA. Interestingly, infants with MC only group had a statistically significant smaller GA compared to funisitis infants (**Table 4**). This may suggest that the impact of MC on IH is most pronounced in extreme prematurity; in contrast to our cohort that included older preterm infants of less than 35 weeks GA. We did not adjust for birth weight in the model analyses given the collinear relationship with GA.

Our results suggest that the effect of prenatal inflammation due to MC on IH persisted far beyond the perinatal period; an interesting and important finding documented for the first time in human preterm infants. The reasons for persistently increased IH in MC exposed infants at 5-6 weeks postnatal age (**Figure 5.1** and **Figure 5.3**) are unknown. We speculate that perinatal inflammation from MC exacerbates the IH/inflammation cycle by causing chemoreceptor dysregulation and worsening of lung disease (**Figure 5.7**) (7).

A limitation of this study is that data related to MC were retrospectively collected. The choice of pathologic definition of chorioamnionitis is another limitation that likely had an impact on our results. Pathologic chorioamnionitis is a histologic finding that may not be symptomatic with no change in maternal clinical status and subsequently the infant. The placenta is thought to act as a barrier that protects the infant and therefore without clinical symptoms the full impact of inflammation may not have reached the infant. Our choice of pathologic definition relates to inconsistent documentation in medical records of symptoms of clinical chorioamnionitis such as uterine tenderness and foul smelling amniotic fluid; hence we may underestimate the number of clinical chorioamnionitis. The secondary outcome measure of severe BPD was chosen as a dichotomous variable per the NICHD definition (155). The absence of significant differences in severe BPD among groups may under estimate the complexity and continuum of lung disease in preterm infants. Finally, this is a single center study and our results may not be generalizable.

This study investigates relationship between prenatal inflammation due to MC and IH. No other groups have studied this relationship in the past in preterm infants. We demonstrated a persistently increased IH in the MC only group beyond the perinatal period, long after the direct effect of inflammation resolves. Our inconsistent results may be related to the pathologic definition of MC versus clinical chorioamnionitis. Prospective studies investigating the impact of clinical chorioamnionitis on IH may provide mechanistic insights in this understudied relationship between inflammation and IH in preterm infants.

V. Acknowledgements

I thank all the team members as mentioned in the acknowledgements section. Special thanks to Hong Huang MD, PhD for processing blood samples for CRP analyses in the pilot assessment. Special recognition to Audra Stacy (M4), Amrita Pant MBBS and Crystal Wilson LPN for contributions to data collection related to this chapter.

Table 5. 1: Baseline Characteristics for All Infant with and without MC or Funisitis					
	No MC or Funisitis		**MC or Funisitis**		**p-Value**
N	58		61		
Gestational Age	27 3/7	25 6/7-28 5/7	25.6	24 6/7-26 6/7	<0.0001
Birth Weight	1030	765-1155	830	685-980	0.006
Small for Gestational Age	3	5.2%	3	4.9%	1
Prenatal Steroids	53	91.4%	54	88.5%	1
Female	28	48.3%	31	50.8%	0.31
Non-Hispanic/ White	49	84.5%	52	85.2%	1
Deaths	2	3.4%	8	13.1%	0.001
Median IQR, n %, MC: Maternal Chorioamnionitis					

Table 5. 2: Baseline Characteristics for No MC or Funisitis versus MC only infants

	No MC or Funisitis		MC only		p-Value
N	58		19		
Gestational Age	27 3/7	25 6/7-28 5/7	25 1/7	23 6/7-25 6/7	<0.0001
Birth Weight	1030	765-1155	730	640-853	0.001
Small for Gestational Age	3	5.2%	1	5.3%	1
Prenatal Steroids	53	91.4%	16	84.2%	1
Female	28	48.3%	6	31.6%	0.4253
Non-Hispanic/ White	49	84.5%	17	89.5%	0.7893
Deaths	2	3.4%	4	21.1%	0.0263

Median IQR, n %, MC: Maternal Chorioamnionitis

Table 5. 3: Baseline Characteristics for No MC or Funisitis versus Funisitis exposed					
	No MC or Funisitis		**Funisitis**		**p-Value**
N	58		42		
Gestational Age	27 3/7	25 6/7-28 5/7	26 2/7	25 1/7-27 5/7	0.0093
Birth Weight	1030	765-1155	880	700-1145	0.1047
Small for Gestational Age	3	5.2%	2	4.8%	1
Prenatal Steroids	53	91.4%	38	90.5%	0.0067
Female	28	48.3%	25	59.5%	0.0558
Non-Hispanic/ White	49	84.5%	35	83.3%	0.7069
Deaths	2	3.4%	4	9.5%	0.1829
Median IQR, n %, MC: Maternal Chorioamnionitis					

Table 5. 4: Baseline Characteristics for Infant with MC versus Funisitis

	MC		Funisitis		p-Value
N	19		42		
Gestational Age	25 1/7	23 6/7-25 6/7	26 2/7	25 1/7-27 5/7	0.003
Birth Weight	730	640-853	880	700-1145	0.03
Small for Gestational Age	1	5.3%	2	4.8%	1
Prenatal Steroids	16	84.2%	38	90.5%	0.69
Female	6	31.6%	25	59.5%	0.19
Non- Hispanic/ White	17	89.5%	35	83.3%	1
Deaths	4	21.1%	4	9.5%	0.47

Median IQR, n %, MC: Maternal Chorioamnionitis

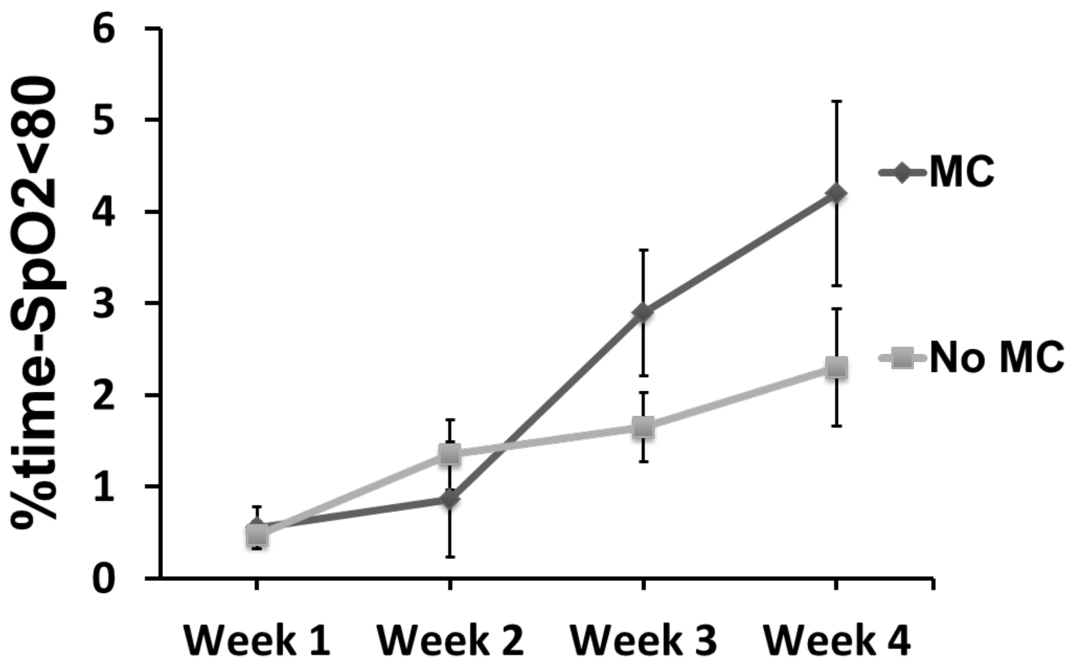


Figure 5. 1: Increase in %time-SpO2<80 in preterm infants less than 30 weeks born with maternal chorioamnionitis (MC).

The %time spent with SpO2<80% was higher in the MC group compared to no MC. Statistically significant difference noted in model analysis (adjusted) between groups during study period, $p<0.05$. This data is from a pilot assessment defining MC per clinical team.

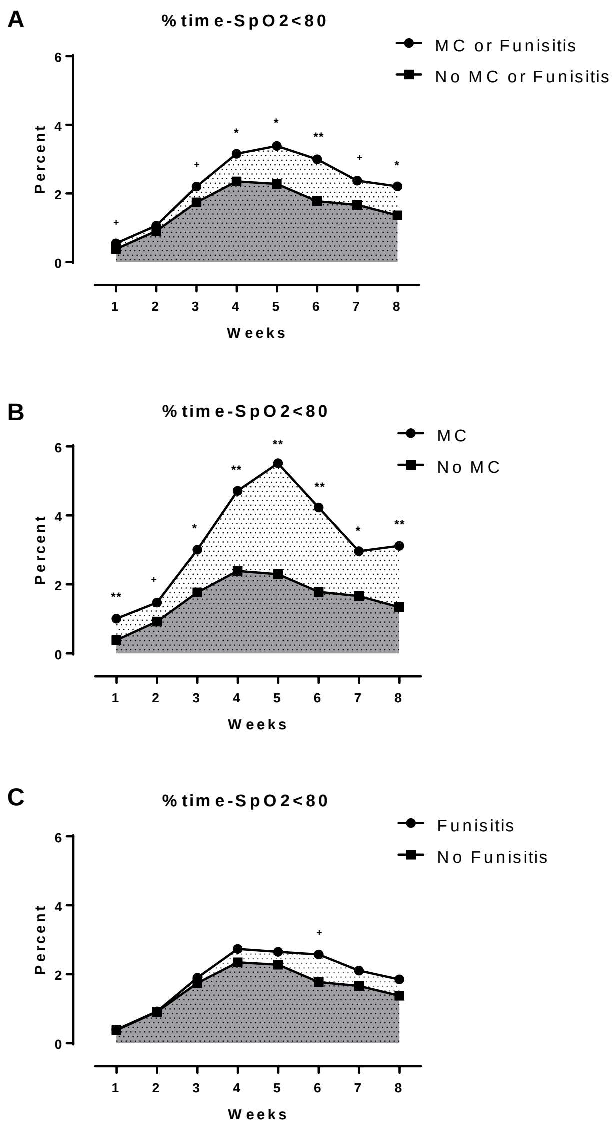


Figure 5. 2: Unadjusted differences in %time-SpO2<80 between pathologic maternal chorioamnionitis (MC) and/or Funisitis versus unexposed

This figure demonstrates unadjusted differences in %time-SpO2<80 between pathologic maternal chorioamnionitis (MC) and/or Funisitis and unexposed (no MC or Funisitis). **A)** The %time-SpO2<80 was higher in MC or funisitis group compared to no MC or funisitis (unexposed). The differences were statistically significant during postnatal weeks 4, 5, 6, and 8. **B)** The %time-SpO2<80 was consistently higher in MC only compared to no MC or funisitis. The differences were statistically significant during all postnatal weeks (except week 2). **C)** There were no statistically significant differences in %time-SpO2<80 in funisitis vs no MC or funisitis groups. **p<0.01, *p<0.05, $^{+}$p<0.1.

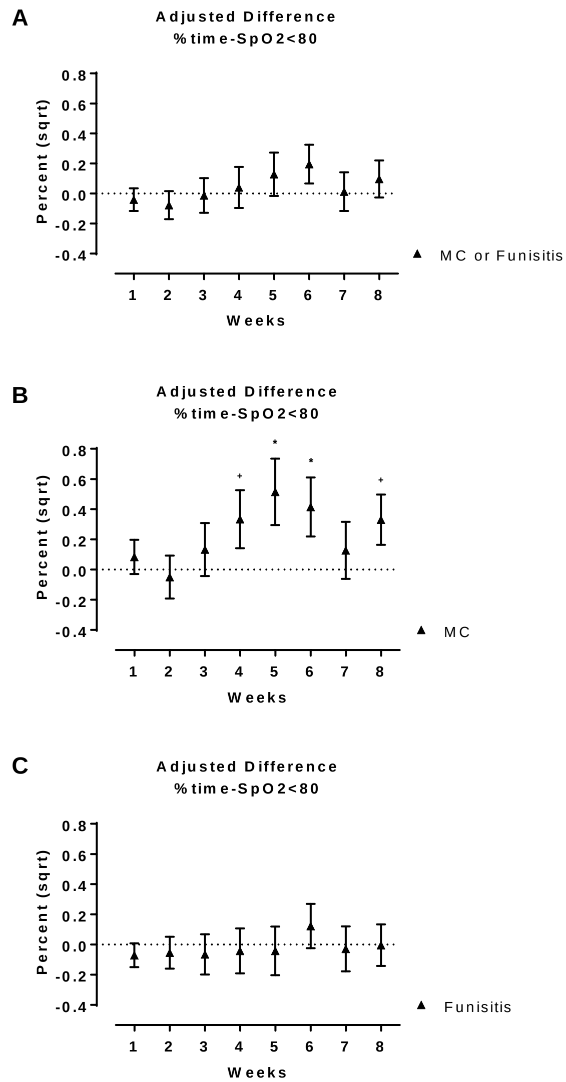


Figure 5. 3: Adjusted differences in %time-SpO2<80 between pathologic MC and/or Funisitis versus unexposed

This figure demonstrates adjusted differences in %time-SpO2<80 between pathologic maternal chorioamnionitis (MC) and/or Funisitis and unexposed (no MC or Funisitis). The graphs presents exposed minus unexposed estimates after adjusting for gestational age, small for gestational age status and prenatal steroids. **A)** There was no difference in %time-SpO2<80 in MC or funisitis group compared to no MC or funisitis (unexposed). **B)** The %time-SpO2<80 was higher in MC only compared to no MC or funisitis. The adjusted differences were statistically significant during postnatal weeks 5 and 6. **C)** There were no statistically significant differences in %time-SpO2<80 in funisitis vs no MC or funisitis groups. *p<0.05, $^{+}$p<0.1.

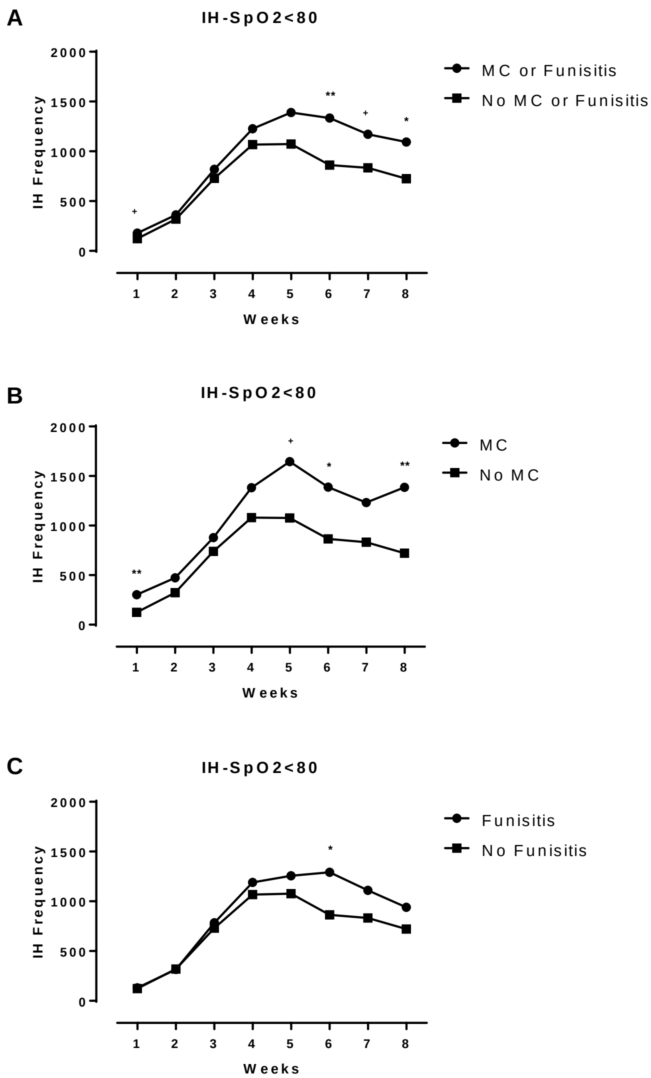


Figure 5. 4: Unadjusted differences in IH-SpO2<80 between pathologic MC and/or Funisitis versus unexposed

This figure demonstrates unadjusted differences in IH-SpO2<80 between pathologic maternal chorioamnionitis (MC) and/or Funisitis and unexposed (No MC or Funisitis). **A)** There was a trend toward higher IH-SpO2<80 in MC or funisitis group compared to no MC or funisitis (unexposed) that was statistically significant during postnatal weeks 6 and 8. **B)** There was a trend toward higher IH-SpO2<80 in MC only compared to no MC or funisitis. The differences were statistically significant during postnatal weeks 1, 6 and 8. **C)** There was a trend toward higher IH-SpO2<80 in funisitis vs no MC or funisitis groups that reached statistical significance during week 6 only. **p<0.01, *p<0.05, $^{+}$p<0.1.

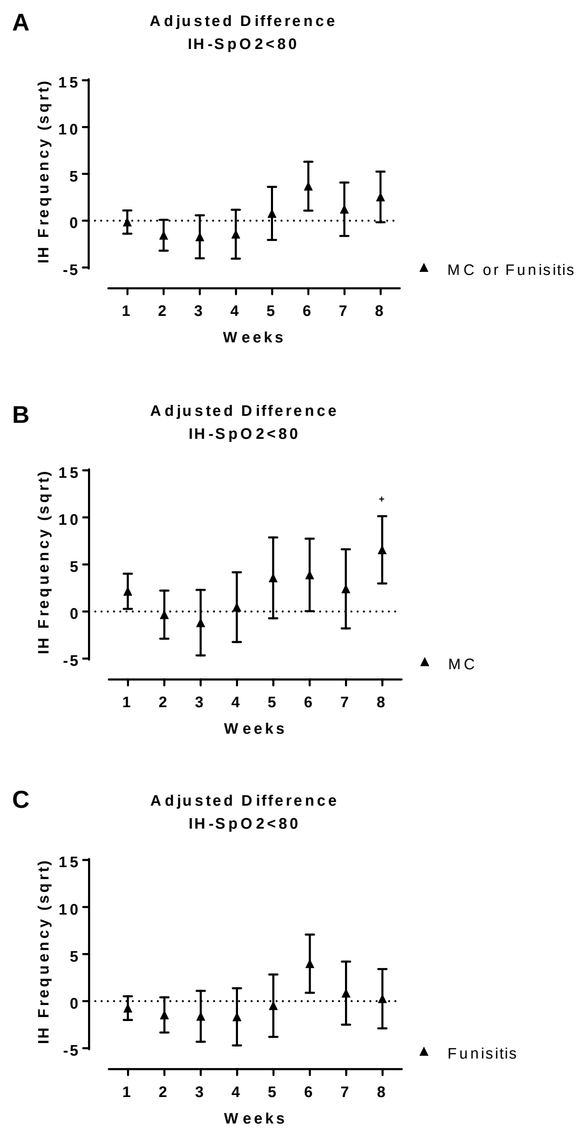


Figure 5. 5: Adjusted differences in IH-SpO2<80 between pathologic MC and/or Funisitis and unexposed (no MC of funisitis)

This figure demonstrates adjusted differences in IH-SpO2<80 between pathologic maternal chorioamnionitis (MC) and/or Funisitis and unexposed (no MC or Funisitis). The graphs presents exposed minus unexposed estimates after adjusting for gestational age, small for gestational age status and prenatal steroids. **A)** There was no difference in IH-SpO2<80 in MC or funisitis group compared to no MC or funisitis (unexposed). **B)** There was no difference in IH-SpO2<80 in MC only compared to no MC or funisitis. **C)** There were no significant differences in IH-SpO2<80 in funisitis vs no MC or funisitis groups. $^{+}$p<0.1.

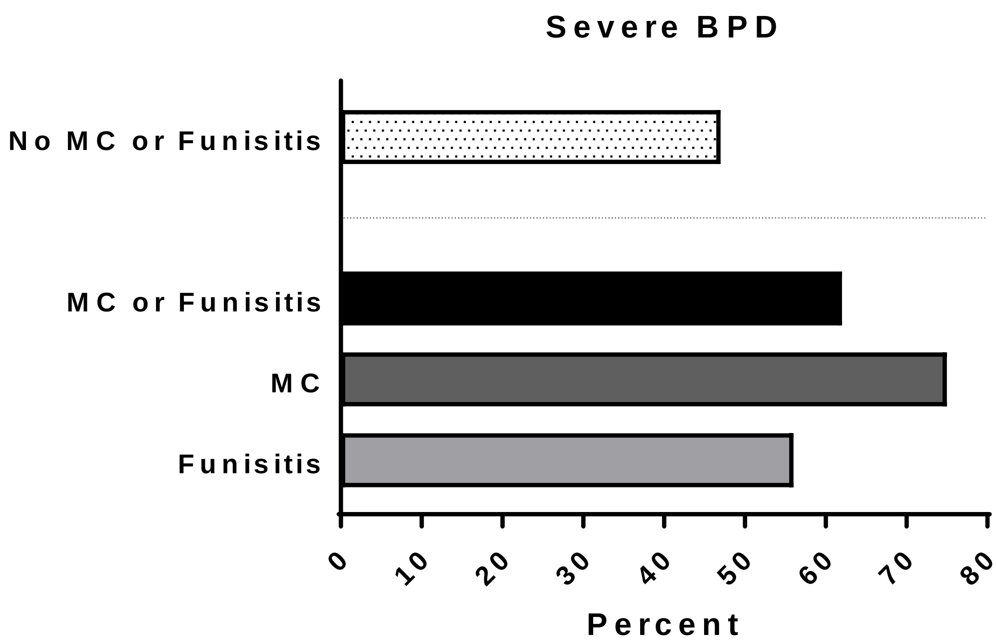


Figure 5. 6: Differences in severe bronchopulmonary dysplasia (BPD) among groups

This figure presents the frequency on severe bronchopulmonary dysplasia (BPD) among groups. There was a trend towards increased severe BPD in the MC and/or Funisitis groups compared to unexposed (No MC or Funisitis); MC or Funisitis p=0.14, MC p=0.057, funisitis p= 0.42. A logistic regression model adjusting for gestational age, small for gestational age status and prenatal steroids showed no a statistically significant difference in severe BPD among groups (p=0.79).

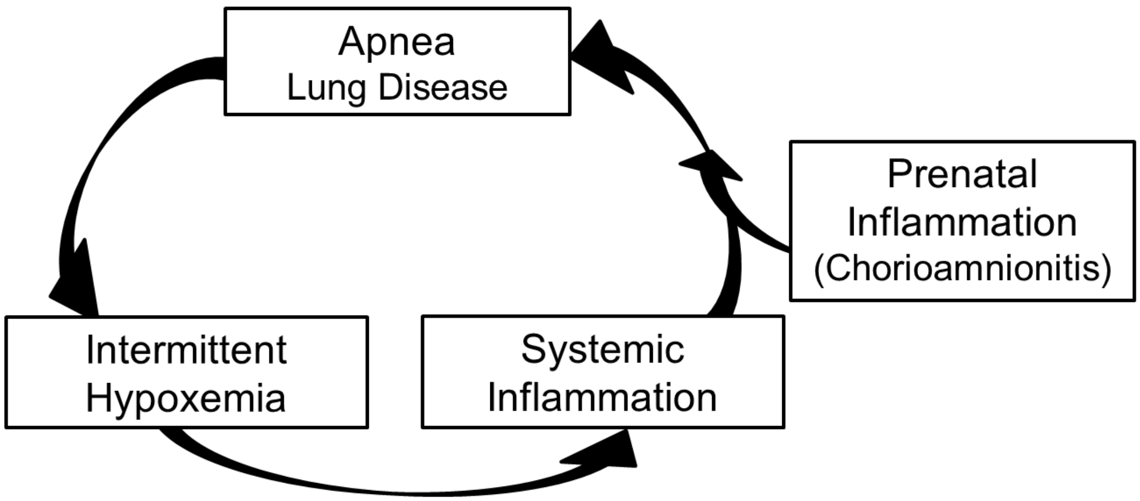


Figure 5. 7: Proposed relationship between intermittent hypoxemia and inflammation and possible role of maternal chorioamnionitis.

The relationship between IH and inflammation is bidirectional with inflammation worsening IH and subsequently IH increases inflammation leading to further respiratory depression. Prenatal inflammation (maternal chorioamnionitis) exacerbates the cycle leading to more IH.

CHAPTER 6: ROLE OF INDOMETHACIN IN REDUCING INTERMITTENT HYPOXEMIA: PRELIMINARY ASSESSMENT

I. Introduction

Although significant progress has been made in the care of preterm infants, they continue to suffer from significant morbidities such as apnea, bronchopulmonary dysplasia (BPD), retinopathy of prematurity (ROP), and neurodevelopmental impairments (NDI) (112, 113). In addition, prematurity is associated with elevated societal economic costs. Four million babies are born per year in the United States and close to a half million are premature (107) with total societal economic cost of approximately 26 billion dollars (108-110). Mean costs of care associated with extreme prematurity are nearly a quarter of a million dollars in the first 4 years of life; approximately 20 times higher than late preterm infants (111).

Intermittent Hypoxemia (IH), episodic drops in oxygen saturations, contributes to the aforementioned morbidities (54). Brief episodes of oxygen desaturations may seem clinically insignificant, but these IH episodes, occurring up to hundreds of events/day, have a cumulative effect on morbidities and mortality. Mounting evidence, links IH with both short and long term neonatal morbidities such as retinopathy of prematurity (ROP), neurodevelopmental impairment (NDI), sleep disordered breathing, and increased mortality (2, 3, 16, 45, 46, 54, 55, 84-86). Laboratory and animal data show IH results in increased inflammatory cytokines, increased free radicals and oxidative stress, increased white matter injury, neurocognitive handicap, and poor growth (4, 41, 54, 114-120). Decreasing IH will lead to decreased associated morbidities and impairment in preterm infants. In addition since cardiorespiratory events delay discharge (6, 160), an intervention to decrease IH will reduce length of stay and the burden on health care dollars.

Currently there are multiple strategies aimed at decreasing IH. Those mainly include methyl xanthine use and respiratory support; i.e. focus on treatment of apnea and management of lung disease. Although effective, the aforementioned strategies do not eliminate IH or lead to lung injury with subsequent long term consequences (4, 161-175). No current strategy focus on other causes of increased IH such as inflammation. Since prenatal inflammation plays a role in increased IH, finding strategies to ameliorate prenatal/perinatal inflammation may be effective at decreasing IH and associated morbidities. Preterm infants are commonly born through a prenatal inflammatory process (123, 151, 176, 177). The increased systemic inflammation at birth worsens apnea and lung disease leading to a rise in IH. Anti-inflammatory agents may ameliorate systemic inflammation and decrease IH. Olsson et al. in rat pup experiments showed that indomethacin reversed the depressive respiratory effects of inflammation (caused by IL-1 β and lipopolysaccharide (LPS)) in addition to hypoxia (144). In this preliminary assessment, we wanted to assess the effect of indomethacin, anti-inflammatory agent, on IH in preterm infants.

II. Methods

Study Design and Data Collection

Oxygen saturation data were prospectively collected from 30 preterm infants less than 30 weeks gestational age (GA) admitted to our level 4 NICU between November 2014 and September 2015. We used high resolution pulse oximeters (Radical 7: Masimo, Irvine, CA) set at 2 second averaging time and 1Hz sampling rate to continuously monitor patients during the first 4 weeks of life. In order to differentiate intermittent from sustained hypoxemia, we included events between 4-180 seconds (1). The exact threshold below which IH is clinically significant is controversial. A drop in SpO2 to less than 80% is widely considered to be clinically relevant (1-3). Therefore, the primary outcome measure was defined as percent time spent with SpO_2 below 80% (%time-SpO2<80).

Pulse oximeters were equipped with serial data recorders (Acumen Instruments Corp) for continuous data collection. Novel programs were utilized to filter and analyze data (Matlab, Natick, MA) (1, 61). Data with artifacts were excluded. Only SpO_2 data with good signal were included in the analyses. Infants with major congenital malformations were excluded.

Infants were randomized to placebo versus indomethacin in this randomized controlled (double blind) trial (RCT). Indomethacin was given within 12 hours of birth and repeated every 24 hours for a total of 3 doses per the current evidence based dosing regimen utilized for other indications (178-183). Neonatal morbidities, including maternal chorioamnionitis (MC), were collected from medical records. In regards to this assessment, after the intervention infants received the standard clinical care per clinical team; except for the additional pulse oximeter.

Statistical Analyses

Statistical analyses for IH were based on linear mixed models, which statistically accounted for repeated measures. Intermittent hypoxemia (%time-SpO2<80) and change of IH over time were compared in indomethacin versus placebo groups using SAS version 9.4 (SAS Institute, Cary, N.C.). Analyses were based on intention-to-treat, and tests were two-sided with a 5% significance level. Comparisons were performed for all infants and in infants with MC only; as we considered the latter most likely group to benefit given they are born through and inflammatory process. Comparisons for baseline characteristics, respiratory support and morbidities were performed using GraphPad Prism 7 (GraphPad Software, La Jolla California USA).

III. Results

Oxygenation data was available on 26 preterm infants with 13 infants each of the indomethacin and placebo groups. Table 1 represents baseline characteristics between indomethacin and placebo groups. There were no differences in GA, birth weight and gender and other baseline characteristics (**Table 6.1**). Table 2 represents respiratory characteristics during and at the end of study period showing no significant differences between groups. More infants were on non-invasive support at 36 weeks corrected age, however these results were not statistically significant (**Table 6.2**).

There were no statistically significant differences in neonatal morbidities between groups as represented in **Table 6.3**. There was one death in the indomethacin group versus none in placebo. Severe IVH was similar in both groups. Infants in the placebo group tended to have more PDA, however, all except for one were non-hemodynamically significant per Gomez et al (61). Late onset sepsis and necrotizing enterocolitis rates were not different between groups.

Although results were not statistically significant, there was a trend toward lower IH rates in the indomethacin compared to placebo group. **Figure 6.1** presents data for all infants. **Figure 6.2** presents data from the patients born with MC. There is attenuation of the peak %time-SpO2<80 at 4-5 weeks of life, however it was not statistically significant.

IV. Discussion

This preliminary data demonstrate that indomethacin, administered shortly after birth, may be a promising new therapy for reducing IH in preterm infants. Infants with increased prenatal inflammation due to MC may benefit the most from this intervention. Perinatal inflammation plays a major role in the pathophysiology of IH and vice versa; and administering an anti-inflammatory agent may break the IH/inflammation vicious cycle in its earliest stages leading to decreased IH **(Figure 6.3).**

Current strategies aimed at decreasing IH focus on treatment of apnea and management of lung disease. Caffeine, a competitive adenosine receptor inhibitor, improves IH (4, 161, 162). Recent evidence suggest that caffeine may also have mild anti-inflammatory effects (184). Caffeine is used in NICUs worldwide and usually discontinued around 34-36 weeks corrected age (185). Recently, Rhein et al. showed that prolonged caffeine use reduces IH frequency until 37 weeks corrected age. Although caffeine is effective in decreasing IH, it does not eliminate IH. Other approaches to ameliorate IH are respiratory support measures such as mechanical ventilation, continuous positive airway pressure (CPAP) and oxygen supplementation (13, 186). However, respiratory support, even with current gentle ventilation strategies, leads to lung injury with subsequent long term consequences (163-174). In addition, oxygen supplementation in preterm infants leads to ROP (major cause of visual impairment) (37). Furthermore, preterm infants continue to have frequent IH events while on respiratory support (1, 2). A strategy that addresses other factors that increases IH (such as inflammation) may have an additive impact on amelioration of IH and hence improve long term outcomes. Although our results are not statistically significant, our trends align with preclinical animal model data. Olsson et al., demonstrated that indomethacin administration reversed the depressive effects of inflammation on breathing patterns(144). Indomethacin is a promising intervention that needs further investigation. Finding a strategy (indomethacin) to decrease inflammation at birth may decrease IH and subsequently decrease associated morbidities in preterm infants.

Prophylactic indomethacin has been tested in preterm infants to reduce other neonatal morbidities such as IVH and PDA. Multiple studies demonstrated that indomethacin decreases severe IVH by more than 30% (183, 187).

However, the decrease in IVH did not translate to improved long term outcomes (187). Similarly prophylactic indomethacin use improves PDA closure (179, 188). However, prophylaxis was not more effective, compared to early treatment of symptomatic PDA, at reducing mortality and respiratory outcomes (189). Both the lack of long term benefit and increased risk benefit ratio, especially in infants without PDA, led to increased practice variation in use of prophylactic indomethacin. However, indomethacin has not been prospectively studied in preterm infants born with MC. Since these infants are born through an inflammatory process, we speculate they may benefit the most from an anti-inflammatory agent. As shown in **Figure 6.2**, infants with MC who received indomethacin tended to have lower IH peak at 1 month of life.

For future larger RCT involving infants with MC, indomethacin should be considered for multiple reasons. *First*, in contrast to postnatal steroids, indomethacin has a good safety profile and is not associated with long term NDI in preterm infants (178, 180, 190-194). Adverse effects associated with indomethacin include transient renal insufficiency (195); which can be ameliorated by interventions to improve renal perfusion. Other reported but rare adverse effects include increased risk of bleeding and intestinal perforation (178, 180, 191, 193, 194). *Second*, indomethacin is associated with decreased morbidities, mainly patent PDA and IVH (178-180, 192, 196, 197); morbidities that may affect cardiorespiratory events in preterm infants. *Third*, indomethacin also regulates blood flow to the brain which may lead to improved respiratory control and less IH (another mechanism to improve IH); as preterm infants have a paradoxical ventilatory depression in response to hypoxia/poor brain perfusion (198, 199). *Fourth*, indomethacin is an effective anti-inflammatory agent that reversibly inhibits cyclooxygenase (COX)-1 and COX-2 enzymes, which results in decreased formation of prostaglandin precursors; main culprits leading to apnea in the setting of inflammation (29, 30). *Fifth*, animal data show indomethacin reverses the effects of inflammation on respiratory patterns(144). *Sixth,* indomethacin is not the standard of care with wide practice variation both locally and nationally creating equipoise and ability to test indomethacin (200). *Finally,* although indomethacin has been studied in preterm infants, the focus of those studies was not in the setting of perinatal inflammation or IH.

A major limitation of this study is the small sample size especially for the subset involving MC patients. However, this was a preliminary assessment aimed at generating pilot data to power future larger studies. Another limitation of this study is the use of indomethacin dosing regimen for IVH and PDA prophylaxis (179, 181-183). Indomethacin was administered in 3 doses (0.2mg/kg/dose on DOL1 and 0.1mg/kg/dose on DOL 2 and 3) in the first 3 days of life. This dosing regimen has documented safety but it may not be adequate to suppress inflammation in infants with born with MC. Longer treatment course may be necessary to have a significant impact on decreasing inflammation and subsequent IH. Ideally, pre and post indomethacin inflammatory markers should have been measured to document a decrease in systemic inflammation.

This is the first study to test the effect of indomethacin in management of IH in preterm infants. This innovative pilot study possibly identified a subset of preterm infants (with prenatal inflammation/MC) who may benefit the most from indomethacin to reduce IH; an important discovery in the era of precision medicine. Future larger studies should focus on investigating indomethacin in patients born with MC.

V. Acknowledgements

I thank all the team members as mentioned in the acknowledgements section. I especially acknowledge the principal investigators (Peter Giannone MD and John Bauer PhD) for the “Comparative effectiveness of preventative strategies for IVH in preterm infants” as this chapter utilized the trial’s infrastructure.

Table 6. 1: Baseline Characteristics	**Indomethacin** N=13	**Placebo** N=13	**p-Value**
Gestational age, weeks	27 4/7 (26 2/7-28 5/7)	27 3/7 (25 3/7-28 5/7)	NS
Birth weight, grams	980 (750 - 1228)	1080 (735 - 1230)	NS
Male	69.2%	69.2%	NS
Apgar 5 min	5 (3-7)	6 (5-8)	NS
Maternal Chorioamnionitis	2 (15.4%)	4 (30.8%)	NS
Prenatal steroids	13 (100%)	12 (92.3%)	NS
Frequency (%), Median (Interquartile range)			

Table 6. 2: Respiratory Characteristics	Indomethacin N=13	Placebo N=13	p-Value
Respiratory distress syndrome	13 (100%)	13 (100%)	NS
Received Surfactant	12 (92%)	11 (85%)	NS
Respiratory Support at 28 days of life			NS
Oxygen Supplementation	11 (84.6%)	12 (92.3%)	
No Support	1 (7.7%)	1 (7.7%)	
Non Invasive Support	8 (61.5%)	7 (53.8%)	
Ventilator Support	4 (30.8%)	5 (38.5%)	
Respiratory Support at 36 weeks corrected age (CA)			NS
Oxygen Supplementation	9 (69.2%)	6 (46.2%)	
No Support	0 (0%)	6 (46.2%)	
Non Invasive Support	9 (69.2%)	5 (38.5%)	
Ventilator Support	1 (7.7%)	1 (7.7%)	
Discharged/Death prior 36 weeks CA	2 (15.4%)	1 (7.7%)	
Oxygen at discharge	7 (54%)	6 (46%)	NS
Frequency (%)			

Table 6. 3: Neonatal Morbidities	**Indomethacin** N=13	**Placebo** N=13	**p-Value**
Severe IVH	3 (23.1%)	3 (23.1%)	NS
Patent Ductus Arteriosus	3 (23.1%)	5 (38.5%)	NS
Necrotizing Enterocolitis	0 (0%)	0 (0%)	NS
Late Onset Sepsis	3 (23.1%)	2 (15.4%)	NS
Mortality	1 (8%)	0 (0%)	NS
Frequency (%)			

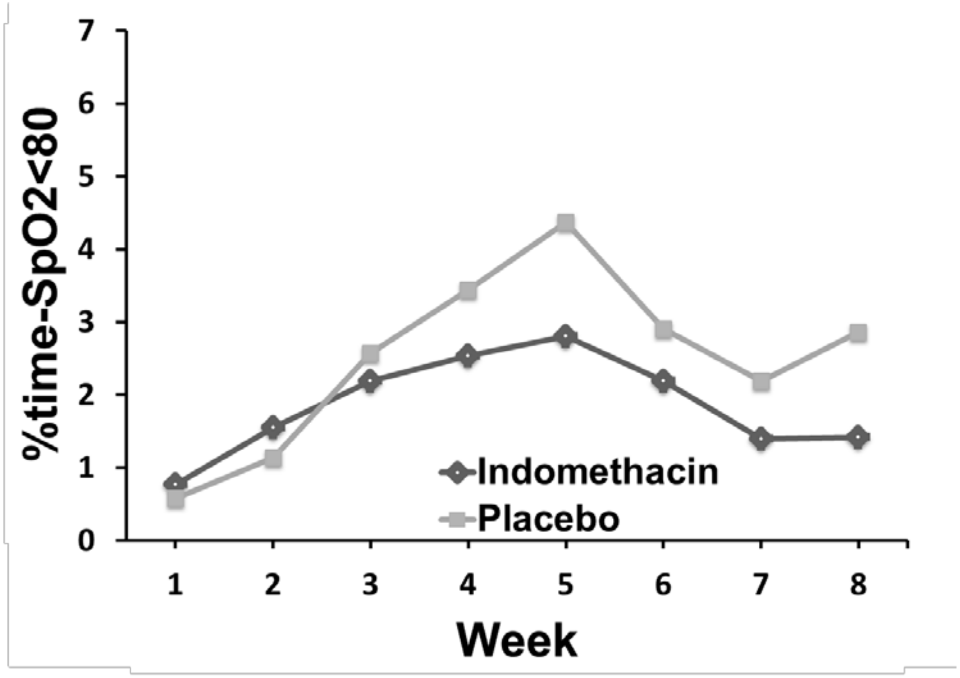


Figure 6. 1: Potential benefit of indomethacin in reducing intermittent hypoxemia (IH) in preterm infants.

Benefit of indomethacin (black) vs placebo (gray) on IH as reflected by percent time spent with SpO2<80% (%time-SpO2<80). N=26, *p=NS*.

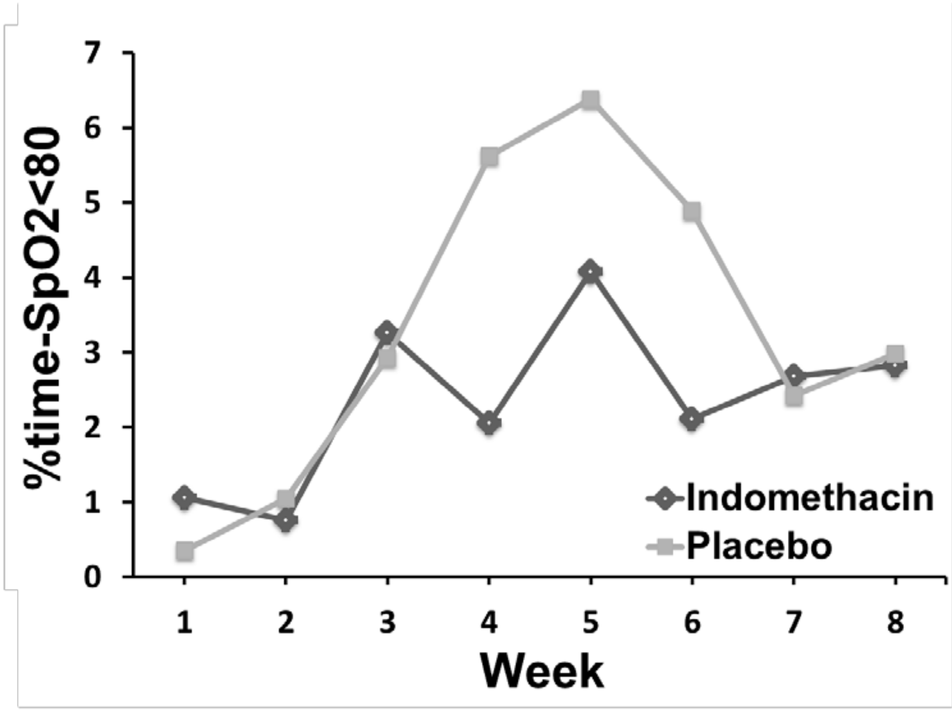


Figure 6. 2: Potential benefit of indomethacin in reducing intermittent hypoxemia (IH) in preterm infants with maternal chorioamnionitis (MC).

Benefit of indomethacin (black) vs placebo (gray) on IH as reflected by loss of 4-5 weeks peak in percent time spent with SpO2<80% (%time-SpO2<80). N=6, *p=NS*.

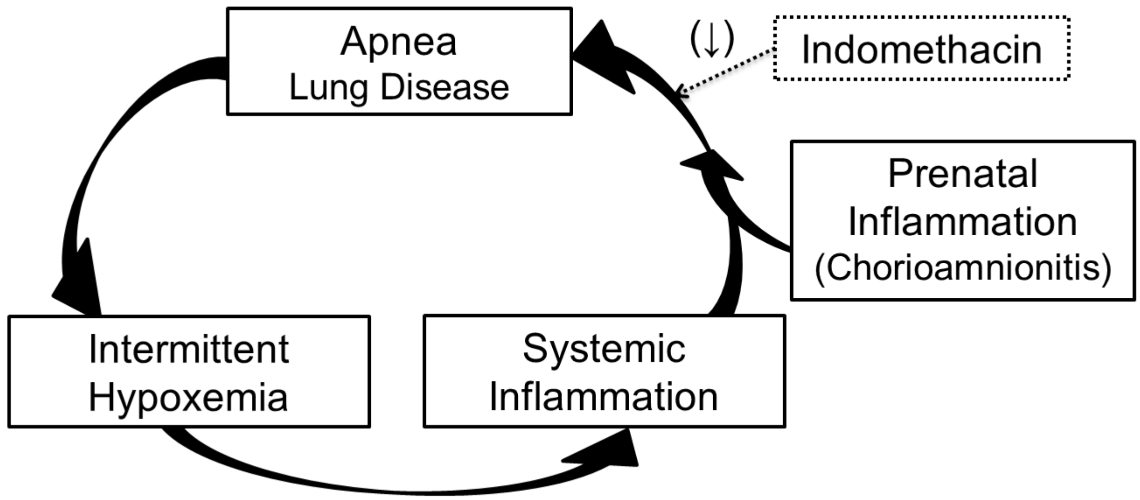


Figure 6. 3: Proposed relationship between inflammation and intermittent hypoxemia (IH) and potential benefit of indomethacin.

The relationship between IH and inflammation is bidirectional with inflammation worsening IH and subsequently IH increases inflammation leading to further respiratory depression. Prenatal inflammation (maternal chorioamnionitis) exacerbates the cycle leading to more IH. We speculate that Indomethacin, an anti-inflammatory agent; improves IH by ameliorating the described vicious cycle.

CHAPTER 7: SUMMARY AND FUTURE DIRECTIONS

Over the past 4 years we have made multiple major contributions to the field; especially as it relates to Pediatrics and Neonatal Perinatal Medicine. We investigated a clinically significant medical problem (IH) that although has profound consequences has not been well studied in preterm infants.

First, we developed novel methods and processes to perform high fidelity studies and accurately assess cardiorespiratory events/IH. We have the ability to efficiently collect, post process and analyze bedside monitoring data. We utilized high resolution pulse oximeters with 2 second averaging time and 1Hz sampling rate. Our analyses IH Automated Analyses Algorithms (IH-AAA) have the capacity to import multiple streams of raw data and export detailed IH profiles from each subject. Furthermore, the IH profile is an innovative and unique method to address this understudied problem. Calculating an IH profile provides an enhanced representation of the continuum of the IH problem. Having a good representation of the spectrum of the problem may help identify thresholds (frequency, duration, severity) beyond which IH leads to neonatal morbidities and impairments. These novel measures can be further developed to become part of the routine monitoring strategies in the NICU for instantaneous feedback to clinical care.

Second, we reconfirmed finding related to the dynamic natural progression of IH in preterm infants. Di Fiore et al. and Abu Jawdeh et al. first reported from a single center study that there is a low frequency of IH in extremely preterm infants (less than 28 weeks GA) during the first week after birth, followed by a progressive increase by weeks 2-3, with a peak around 4-5 weeks then plateau/decrease during weeks 6-8 (1, 2). No other studies have addressed this issue or replicated these findings in order to better understand mechanisms in the future. We now reproduce this finding from a second center, utilizing an expanded patient population of less than 30 weeks GA (versus 28 weeks shown before) showing similar IH dynamic frequency until 10 weeks postnatal life (versus 8 weeks shown before). The reasons leading to the rise in IH postnatally are poorly defined but likely because of both developing lung disease and chemoreceptor dysregulation possibly resulting from inflammation and hypoxia (7). Now that these findings have been reproduced, studies should focus on understanding further mechanisms and causes for the rise in IH during early postnatal life.

Third, our findings demonstrate the importance of prenatal exposures and their effects on postnatal outcomes. We had the unique opportunity to assess the relationship between isolated opioid exposure and respiratory instability in preterm infants. It was challenging in the past to assess the relationship between isolated prenatal opioid exposure and respiratory outcomes/IH, as the majority of women who use opioids also smoke or misuse poly-drugs. Our results suggest that prenatal opioid exposure is associated with increased IH measures compared to unexposed preterm infants. Interestingly, the increased IH measures in opioid exposed infants persisted beyond the early postnatal period. Another important finding is that the prevalence of opioid exposure in our local preterm population is higher than previously reported nationally, thus creating urgency toward addressing this significant problem in this vulnerable patient population

Fourth, we translated and complemented the knowledge we have from preclinical animal and bench studies to the clinical setting in preterm infants. We showed for the first time in preterm infants that cumulative IH is associated with increased markers of inflammation, namely C-reactive Protein (CRP). Our results suggest that IH at any of the selected thresholds is associated with increased CRP. In addition, we demonstrated that the longer IH events are associated with higher CRP levels. These are important findings that shed light on possible mechanisms by which IH causes neonatal morbidities and impairment. Future longitudinal studies that focus on repeated measures of short and long acting markers of inflammation throughout the inflammatory cascade will help define mechanisms and better understand this relationship between IH and inflammation.

Furthermore, our findings support our hypotheses of a bidirectional relationship between inflammation and IH. It is well established that systemic inflammation leads to increased apnea and subsequently IH (29, 30). In this document, we demonstrated that IH may be pro-inflammatory itself. The pro-inflammatory effects of IH may lead to a vicious cycle (positive feedback loop). Apnea events cause IH (oxygen desaturations) and subsequent postnatal inflammation systemically and hence in the respiratory control network, peripheral chemoreceptors and lungs. The postnatal inflammation leads to a further cycle of increased apnea events and consequently higher frequency of IH. Interestingly, this phenomenon may be in part responsible for the IH peak at 4-5 weeks of age.

Fifth, we demonstrated that maternal chorioamnionitis may be associated with increased IH during early postnatal life. No other groups have studied this relationship in the past in preterm infants. We speculate that maternal chorioamnionitis starts or exacerbates the aforementioned cycle early leading to the snowball/spiral effect. Our inconsistent results in this chapter may be related to the pathologic definition of MC versus clinical chorioamnionitis. Prospective studies investigating the impact of chorioamnionitis (clinical and pathologic) on IH may provide mechanistic insights in this understudied relationship between inflammation and IH in preterm infants.

Sixth, our preliminary assessment suggests that indomethacin, a commonly used medication in the NICU, may be used in a novel indication; to decrease IH in patients born with increased inflammation due to MC. This is the first study to test the effect of indomethacin in management of IH in preterm infants. This innovative pilot study possibly identified a subset of preterm infants (with prenatal inflammation/MC) who may benefit the most from indomethacin to reduce IH; an important discovery in the era of precision medicine. A large randomized clinical trial is needed to test the efficacy of this promising intervention, in management of IH in preterm infants born with perinatal inflammation.

Seventh, we present in Appendix A a recent publication showing that red blood cell transfusion (RBC) decrease IH events beyond the first week of life. We also demonstrated a lack of benefit/possible worsening in oxygenation after RBC transfusion in the first week of life; an interesting finding now reported twice from two separate cohorts. This finding requires further investigation especially after possible worsening in oxygenation reported in this study. We also documented factors, other than hematocrit, that should be considered before RBC transfusion administration; including mechanical ventilation, FiO_2 requirement and IH measures. Our publication is a stepping stone towards larger studies aimed at finding objective bedside measures to guide RBC transfusion administration.

Eighth, we present in Appendix B a publication addressing the relationship between perfusion index (PI) and patent ductus arteriosus (PDA) in preterm infants. Perfusion index (PI) is a noninvasive measure of perfusion collected from the bedside utilizing our developed methods. Delta PI (ΔPI) is the difference between PI measured pre-ductal versus post-ductal. We were able to demonstrate that a lower mean ΔPI and pre PI values over a 4-hour period have the potential to detect the presence of PDA in premature infants. We were the first to report a lower variability in ΔPI in infants with PDA compared to those without. This non-invasive measure (PI) is a promising bedside tool to assess for PDA in preterm infants. Future studies are needed to determine the clinical utility of PI in predicting hemodynamic significance and hence need for PDA treatment in preterm infants.

We have multiple ongoing studies addressing IH from various perspectives. A) We are assessing other factors that may influence IH in preterm infant. For examples, we hypothesized that delayed cord clamping may reduce IH through a rise in both hematocrit and progenitor cells. A bolus of blood and progenitor cells from delayed clamping of the umbilical cord may have a lasting impact on IH. This study is funded by the Gerber Foundation and we are near completion of patient enrollment. B) We are assessing the utility of IH as a clinical marker for patient management in the NICU. For examples, among other markers, we are testing IH as a predictor for 1) readiness to discontinue mechanical ventilation (extubation readiness) and 2) thresholds for RBC transfusions in preterm infants. C) We are assessed the relationship between IH and neonatal morbidities. For example, we are investigating the relationship between IH and growth impairment in preterm infants. In addition, we completed enrollment for a study funded by the Children's Miracle Network assessing the relationship between IH and acute kidney injury (IHAKI study) in preterm infants.

Finally, a valuable experience throughout this process is working with a talented and dedicated multidisciplinary team. Our team encompasses multiple divisions, departments, colleges and other institutions and universities. We are a solid example of the value of team science during this new era of clinical and translational research (201). Our respiratory control research program is one of handful programs nationwide able to perform such complex high-fidelity studies related to cardiorespiratory events in preterm infants. The team has established an excellent working relationship and will continue to tackle complex questions involving health of infants.

APPENDIX A

Citation: *Ibonia KT, Bada H, Westgate P, Gomez EM, Bhandary P, Patwardhan A, Abu Jawdeh EG. Blood Transfusions in Preterm Infants: Changes on Perfusion Index and Intermittent Hypoxemia. Transfusion (In press)*

BLOOD TRANSFUSIONS IN PRETERM INFANTS: CHANGES ON PERFUSION INDEX AND INTERMITTENT HYPOXEMIA

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Conflict of Interest: None

Running Title: Transfusion, Perfusion Index, Hypoxemia

ABSTRACT

Background

Red blood cell (RBC) transfusion decreases intermittent hypoxemia (IH) events beyond the first week of life. This benefit may be related to improved perfusion to the respiratory control network. Perfusion index (PI) is a perfusion measure provided by the pulse oximeter. We hypothesized that the benefit in IH after RBC transfusion is associated with a rise in PI. In addition, we assessed the value of PI and clinical measures in predicting the effect of RBC transfusion on IH.

Study Design and Methods

We prospectively enrolled infants less than 30 weeks gestational age. PI and oxygen saturation (SpO_2) were monitored with high-resolution pulse oximeters 24 hours pre and post RBC transfusion. Data was analyzed at three postnatal periods, epoch 1: first week of life (1 to 7 days of life), epoch 2: 2 to 4 weeks of life (8 to 28 days of life), and epoch 3: 4 to 8 weeks of life.

Results

One hundred eighteen transfusions were analyzed. IH measures significantly decreased post transfusion in epochs 2 and 3. PI significantly increased after transfusion, but it did not correlate with the decrease in IH measures. Mechanical ventilation, fraction of inspired oxygen (FiO_2), and IH measures influenced the effects on oxygenation.

Conclusions

RBC transfusion improved IH after the first week of life. The benefit in IH did not correlate with PI increase after transfusion. Pre transfusion respiratory support and IH measures predicted the effect of transfusion on oxygenation.

Key Words: red blood cell transfusion, preterm infants, perfusion, hypoxemia

INTRODUCTION

Intermittent Hypoxemia (IH), defined as episodic drops in oxygen saturation, is common in preterm infants.(1-3) The incidence of IH in extremely low gestational age infants changes during the first 2 months of life.(1, 2) There is low IH frequency during the first week of life, followed by a progressive increase over weeks 2-3, plateaus around 4 weeks, and decreases at weeks 6-8.(1, 2) Intermittent hypoxemia is associated with both short and long term morbidities such as retinopathy of prematurity,(2) neurodevelopmental impairment, and late death.(1, 3-5) Red blood cell (RBC) transfusion results in IH improvement, particularly beyond the first week of life.(1) Perhaps, the main rationale for RBC transfusion in preterm infants is improvement in oxygenation.(6) There are two proposed mechanisms for beneficial effect of RBC transfusion on oxygenation. The first relates to greater cardiovascular stability with increased perfusion to the respiratory control network leading to improved central respiratory drive and subsequent less IH.(1, 7-9) The second suggests greater stability of oxygenation due to a rise in hematocrit leading to less IH in the presence of apnea.(1),(10)

Perfusion index (PI) is a noninvasive measure of perfusion provided by the bedside pulse oximeter. Perfusion index is calculated from the ratio of the pulsatile to non-pulsatile signal at the monitoring site.(11, 12) Perfusion index correlates with superior vena cava flow,(13) detects critical left heart obstructive disease,(14) and patent ductus arteriosus.(15, 16) Furthermore, Kanmaz et al. noted that RBC transfusion is associated with a significant increase in PI and suggested that PI may be a useful marker for the need of transfusion.(17) Therefore, we wanted to assess if the benefit in IH seen after RBC transfusion is associated with a rise in PI in preterm infants at different postnatal ages. In addition, we assessed the predictive value of PI, hematocrit, mechanical ventilation, fraction of inspired oxygen (FiO_2) and IH; in order to identify infants who will benefit the most from the RBC transfusion in terms of oxygenation.

MATERIALS AND METHODS

Study Design and Data Collection

This was a prospective cohort study conducted at the University of Kentucky Medical Center Neonatal Intensive Care Unit between November 2014 and October 2015. The study was approved by the University of Kentucky Institutional Review Board. Infants with gestational age (GA) less than 30 weeks were approached in the first week of life and informed consent was obtained from parent(s). Infants were then followed and oxygen saturation was continuously monitored in the first 2 months of life. Infants who received RBC transfusion per the NICU transfusion guidelines were included in the analyses. The following is a summary of the local NICU transfusion guidelines: Hematocrit threshold of <35% for mechanically ventilated neonates or FiO_2 requirement >40%, hematocrit <28% for infants on non-invasive respiratory support or FiO_2 requirement <40%, and hematocrit <22% for neonates on no respiratory support. RBC transfusion at 15ml/kg was administered over a 3 hour period. Oxygen saturation (SpO_2) and PI were monitored using continuous high-resolution (2s averaging time and 1Hz sampling rate) pulse oximeters (Radical 7: Masimo, Irvine, CA, USA). The target oxygen saturation in our unit is 90-95%. Patients were continuously monitored for the first 8 weeks of life and data was stored on serial data recorders. Novel programs were utilized to filter (Matlab, Natick, MA, USA) and analyze (SAS Institute, Cary, NC, USA) data. Variables related to demographics, weight, respiratory measures and medications were collected.

The primary outcome measures for IH were defined as 1) a drop in SpO_2 to less than 80% for ≥4s and ≤3min duration (IH-SpO_2<80) and 2) overall percent time spent with SpO_2 <80% (%time-SpO_2<80). The lower limit of 4s duration was based on the previous data by Abu Jawdeh et al. and the upper limit of 3 min duration was used to differentiate intermittent from sustained hypoxemia.(1) Other outcome measures included additional SpO_2 thresholds of 85% and 90%.

A RBC transfusion was eligible for analysis if no other RBC transfusion was administered 24 hours pre or post transfusion. We then analyzed changes in IH frequency (IH-SpO_2<80, IH-SpO_2<85, IH-SpO_2<90), percent time spent below threshold (%time-SpO_2<80, %time-SpO_2<85, %time-SpO_2<90), mean PI, and variability of PI during the 24 hours pre and post RBC transfusion. Additionally, we determined the associated changes in hematocrit and respiratory characteristics.

To account for the effect of postnatal age on IH following RBC transfusion(2), the 8-week monitoring period was stratified into three epochs and analyzed separately; epoch 1: first week of life (1 to 7 days of life), epoch 2: 2 to 4 weeks of life (8 to 28 days of life), and epoch 3: 4 to 8 weeks of life.(1) In order to assess which preterm infants benefit the most from RBC transfusion, we evaluated the predictive value of the following pre RBC transfusion variables: PI, hematocrit, mechanical ventilation, FiO_2 requirement, and IH primary measures.

Statistical Analysis

To compare epochs in Table 1, continuous variables were presented as mean ± standard deviation (SD) and categorical variables were expressed as frequencies and percentages. Sample means and SDs were also utilized in Figures 2 and 3 to visually compare pre and post RBC transfusion values for each epoch. Pearson's correlations were used to quantify associations between changes in different variables. To account for statistical correlation arising from repeated measurements, i.e. multiple observations per subject, generalized estimating equations with robust standard errors were utilized for inference. Finally, linear mixed models with robust standard errors were utilized to obtain results for Table 3, in which change in IH measures (IH-SpO_2<80 or %time-SpO_2<80) after RBC transfusion was the outcome of interest. The primary predictors were pre RBC transfusion mechanical ventilation, FiO_2 requirement, and pre RBC transfusion IH measures. The models also controlled for pre RBC transfusion PI and hematocrit. All tests were two-sided at the 5% significance level. Analyses were conducted in SAS version 9.4 (SAS Institute, Cary, NC, USA).

RESULTS

Fifty preterm infants met criteria for enrollment. Thirty-nine infants received RBC transfusions that were eligible for analysis for a total of 118 transfusions (22, 63 and 33 RBC transfusions in epochs 1, 2, and 3, respectively). The median (IQR) of eligible transfusions were as follows: 2(1-2), 5(3-7) and 8(6-10) for epochs 1, 2, and 3, respectively (Table 1). Figure1 shows the flow diagram for patient enrollment, transfusion eligibility, and number of infants who received transfusions during each epoch. There were no significant differences in GA, birth weight, gender, and race across all 3 epochs (Table 1). The majority of infants required respiratory support, supplemental oxygen and caffeine therapy (Table 1). The FiO_2 requirement (mean ± standard deviation) increased to 35.2% ±11.6 (p=0.1), 43.7% ± 19.3 (p=0.9) and 47.8% ± 24.7 (p=0.3) in epochs 1, 2 and 3 respectively but was not statistically significant.

Changes in Measures Pre and Post RBC Transfusion

As represented in Figure 2A, there was a statistically significant but minimal increase in mean 24 hour PI after RBC transfusion across all epochs. There was no difference in variability of PI between pre and post RBC transfusion in all 3 epochs (pre-post: -0.07 ± 0.33, p=0.2; -0.01 ± 0.12, p=0.5; -0.05 ± 0.15, p=0.1 in epochs 1, 2 and 3 respectively). In epoch 1, there was no change in IH-SpO_2<80 and IH-SpO_2<85 post RBC transfusion; interestingly, there was a significant increase in IH-SpO_2<90 (Figure 3). Overall, %time-SpO_2<80, %time-SpO_2<85 and %time-SpO_2<90 did not significantly change in epoch 1 (Figure 3). In epochs 2 and 3, we found a significant decrease in IH-

SpO_2<80 and IH-SpO_2<85 and no change in IH-SpO_2<90 (Figure 3). Overall %time-SpO_2<80% and %time-SpO_2<85 improved in epoch 2 and 3 with no changes in %time-SpO_2<90. As expected, mean hematocrit significantly increased 24 hours after RBC transfusion across all three epochs (Figure 2B).

Correlations of Changes Pre and Post RBC Transfusion

There was no significant correlation between changes in PI and IH pre and post RBC transfusion in any of the 3 epochs (Table 2). There was no correlation between changes in hematocrit and IH pre and post RBC transfusion in epochs 1 and 2 (Table 2). In epoch 3, there was a positive correlation between the change in hematocrit and IH measures that was statistically significant for %time-SpO_2<80 (Table 2).

Factors Associated with the Effect of RBC Transfusion on IH measures

Linear mixed models were utilized to assess factors that influenced the effect of RBC transfusion on IH. The models controlled for pre RBC transfusion PI, hematocrit, mechanical ventilation, FiO_2 requirement and IH-SpO_2<80 or %time-SpO_2<80. The results are presented in Table 3.

DISCUSSION

Our study shows an increase in perfusion (as represented by the rise in PI) after RBC transfusion. However, this increase does not correlate with the improvement in oxygenation. Consistent with Abu Jawdeh et al.,(1) our study shows that IH improved post RBC transfusion only beyond the first week of life.(2) In addition, our results replicate the lack of benefit in oxygenation after RBC transfusion in the first week of life. This study also demonstrates that pre

RBC transfusion mechanical ventilation need, FiO_2 requirement and IH measures influence the effect of RBC transfusions on oxygenation.

Similar to a study by Kanmaz et al.,(17) our results show a significant increase in PI post RBC transfusion. The increase in PI is minimal and may not be clinically significant. The observed increase in PI did not correlate with a decrease in IH measures following RBC transfusion. The effect of RBC transfusion on PI may be related to volume expansion. In contrast, RBC transfusion effect on IH is likely due to changes in oxygen carrying capacity and stabilization of oxygenation.(6, 10, 18)

The effect of RBC transfusion on IH varied based on postnatal age. There was significant improvement in oxygenation after RBC transfusion in epochs 2 and 3. However, there was no significant change in IH measures after RBC transfusion during the first week of life; in fact, an increase in IH frequency occurred for IH-SpO_2<90. This increase in IH events in epoch 1 after transfusion for IH-SpO_2<90 reflects the increase in milder events (SpO_2 ≥85%); although all trended in the same direction. The etiology of this reproducible lack of benefit in oxygenation after RBC transfusion in early postnatal life is unknown, but may be influenced by multiple factors. The lack of benefit may be related to the already low incidence of IH during this period.(1, 2) Other factors may include inadequate compensatory mechanisms to overcome the changes in blood flow, volume status and blood viscosity associated with RBC transfusion during early postnatal life.(6, 18-20) Furthermore, the higher proportions of high-affinity fetal hemoglobin in early postnatal life may have an impact on the effect of RBC transfusion on oxygenation.(10)'(20) The lack of benefit in oxygenation after RBC transfusion in the first week of life raises important concerns regarding liberal transfusion thresholds during early postnatal life and the need to further evaluate any adverse respiratory effects in this time period. In addition, studies to further evaluate mechanisms and factors that influence the effect of RBC transfusion on IH in the first week of life are imperative.

Respiratory support (mechanical ventilation and FiO_2) and IH measures influenced the effect of RBC transfusions on oxygenation (Table 3). As expected, patients on mechanical ventilation benefited more from RBC transfusion compared to extubated infants in epoch 2 and approached significance in epoch 3. Interestingly, in epoch 1, patients on mechanical ventilation had no improvement or worsening in oxygenation after RBC transfusion. We speculate the findings seen in the first week of life in ventilated infants may relate to patient characteristics including immaturity of compensatory mechanisms, severe lung disease with poor pulmonary reserves and subsequent lung fluid overload from RBC transfusion.(6, 10, 20) Increased FiO_2 requirement pre RBC transfusion was associated with a significant decrease in IH measures post transfusion during epoch 1. After the first week of life, higher IH measures pre RBC transfusion were associated with greater benefit in oxygenation that was statistically significant in epoch 2 and approached significance (p=0.053) in epoch 3. Extent of FiO_2 requirement and IH measures are closely related as FiO_2 adjustment is often based on oxygen desaturations. Our sample size may not have been large enough to reach statistical significance in all epochs; however, FiO_2 and IH measures are promising objective tools able to guide transfusion management. Overall, the results of the study show that postnatal age, along with type of respiratory support and IH measures, influence the effect of RBC transfusion on oxygenation. Further studies to evaluate mechanisms as to how these factors influence the effect of RBC transfusion on IH are needed.

Maintaining hematocrit above a certain consensus threshold is the major indication for RBC transfusion in NICUs worldwide.(21, 22) Consistent with previous studies, our results suggest that hematocrit alone is a weak predictor of the effect of RBC transfusion on oxygenation.(1, 5, 7, 9, 23, 24) Although hematocrit significantly increased post RBC transfusion, the change in hematocrit did not correlate with improved oxygenation after RBC transfusion except in epoch 3 where a poor correlation was noted (Table 2). We speculate that hematocrits are closely followed in the NICU and the levels in our infants may not have been low enough to result in significant cardiorespiratory instability.

A limitation to this study is not having evaluated other hemodynamic factors such as blood pressure, heart rate, and volume status. We also lack documentation of other neonatal morbidities that may have affected PI and oxygenation such as presence of intraventricular hemorrhage, patent ductus arteriosus, and sepsis. As our model included multiple variables, the current sample size may have lacked sufficient power to reach significance in certain epochs. The possible variation in RBC transfusion indications among providers is a limitation, but likely minimized by our unit consensus transfusion guidelines.

CONCLUSION

Red blood cell transfusion is associated with decreased IH events after the first week of life. The lack of benefit in oxygenation after RBC transfusion in the first week of life is an interesting finding now reported twice from two separate cohorts. This finding requires further investigation especially after possible worsening in oxygenation reported in this study. Our primary aim to assess the value of PI as an indication for RBC transfusion did not yield positive findings. We documented factors, other than hematocrit, that should be considered before RBC transfusion administration; including mechanical ventilation, FiO_2 requirement and IH measures. Our study is a stepping stone towards larger studies aimed at finding objective bedside measures to guide RBC transfusion administration.

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TABLE 1 Baseline characteristics of enrolled patients among epochs

	Epoch 1 n=22	Epoch 2 n=63	Epoch 3 n=33	*p* value
Gestational age in weeks (n) (Mean ± SD)	(22) 25.8 ± 1.3	(62) 25.6 ± 1.3	(33) 25.6 ± 1.2	0.8
Birth weight in grams (n) (Mean ± SD)	(22) 807 ± 162	(62) 796 ± 171	(33) 803 ± 179	0.94
Postnatal age in days (n) (Mean ± SD)	(22) 4.6 ± 1.6	(62) 18.0 ± 6.4	(33) 43.5 ± 8.9	<0.001
Weight day of transfusion in grams (n) (Mean ± SD)	(22) 808 ± 153	(62) 982 ± 247	(33) 1475 ± 434	<0.001
Number of Transfusions per patient, Median (IQR)	(22) 2 (1-2)	(63) 5 (3-7)	(33) 8 (6-10)	<0.001
Male, n (%)	15/22 (68%)	36/62 (58%)	21/33 (64%)	0.7
Caucasian, n (%)	17/22 (77%)	53/61 (87%)	27/33 (82%)	0.7
Respiratory Support				
Conventional ventilator, n (%)	18 (86%)	50 (86%)	21 (68%)	0.22
Non-invasive ventilation, n (%)	3 (14%)	8 (14%)	10 (32%)	
NIPPV, n (%)	2 (10%)	6 (10%)	7 (23%)	
CPAP, n (%)	1 (5%)	2 (3%)	3 (10%)	
Missing data for type of respiratory support, n (%)	1 (5%)	5 (8%)	2 (6%)	
Supplemental oxygen, n (%)	19/21 (90%)	54/58 (93%)	29/31 (94%)	0.92
Pre RBC transfusion FiO_2 (n) (Mean ± SD)	(21) 30.0 ± 9.3	(59) 42.7 ± 21.0	(31) 44.8 ± 20.8	<0.001
Caffeine, n (%)	21/22 (95%)	62/62 (100%)	31/33 (94%)	0.55

[a]SD, standard deviation; [b]NIPPV, nasal intermittent positive pressure ventilation; [c]CPAP, continuous positive airway pressure; *p for mean difference;*

TABLE 2 Correlations of changes in PI, Hematocrit and IH

	Epoch	ΔIH Events < 80%		Δ%time < 80%	
		r	*p* value	r	*p* value
Δ Perfusion Index	1	-0.05	0.46	-0.18	0.61
	2	0.18	0.13	0.14	0.38
	3	-0.16	0.22	0.07	0.51
Δ Hematocrit	1	0.1	0.33	-0.03	0.88
	2	-0.11	0.37	-0.05	0.86
	3	0.271	0.08	0.322	0.02

[a]Δ represents change in value: post RBC transfusion - pre RBC transfusion. [b]r = correlation coefficient

TABLE 3. Predictors of the effect of RBC transfusions on IH.

Predictor	Outcome Measure	Epoch 1 Coefficient	Epoch 1 (95% CI)		Epoch 1 *p*	Epoch 2 Coefficient	Epoch 2 95% CI		Epoch 2 *p*	Epoch 3 Coefficient	Epoch 3 95% CI		Epoch 3 *p*
Perfusion Index	IH-SpO_2<80	**-26.9**	-74.6	20.7	*0.22*	**-14.8**	-103.6	74.1	*0.73*	**99.4**	-75	273.7	*0.20*
	%time-SpO_2<80	**-1.20**	-3.00	0.58	*0.16*	**-0.50**	-2.96	1.99	*0.69*	**1.86**	-2.95	6.67	*0.40*
Hematocrit	IH-SpO_2<80	**-0.33**	-9.1	8.4	*0.93*	**12.7**	-2.5	27.9	*0.10*	**-5.56**	-14.39	3.27	*0.20*
	%time-SpO_2<80	**-0.03**	-0.15	0.09	*0.60*	**0.40**	0.12	0.72	*0.01*	**-0.14**	-0.49	0.20	*0.40*
Mechanical ventilation	IH-SpO_2<80	**35**	-4.4	74.5	*0.07*	**-60.8**	140	18.5	*0.13*	**-84.1**	-173.7	5.4	*0.06*
	%time-SpO_2<80	**2.40**	0.46	4.48	*0.02*	**-2.30**	-4.69	-0.03	*0.050*	**-2.30**	-5.52	0.73	*0.10*
FiO_2	IH-SpO_2<80	**-1.52**	-3.01	-0.03	*0.047*	**-0.82**	-1.7	0.05	*0.06*	**2.01**	-0.23	4.24	*0.07*
	%time-SpO_2<80	**-0.10**	-0.14	-0.04	*0.002*	**-0.04**	-0.08	0.00	*0.07*	**0.07**	-0.01	0.15	*0.09*
IH-SpO_2<80	IH-SpO_2<80	**-0.49**	-1.1	0.12	*0.10*	**-0.38**	-0.8	0.04	*0.07*	**-0.24**	-0.49	0.003	*0.053*
%time-SpO_2<80	%time-SpO_2<80	**0.55**	-0.19	1.29	*0.13*	**-0.29**	-0.52	-0.06	*0.02*	**-0.28**	-0.65	0.08	*0.10*

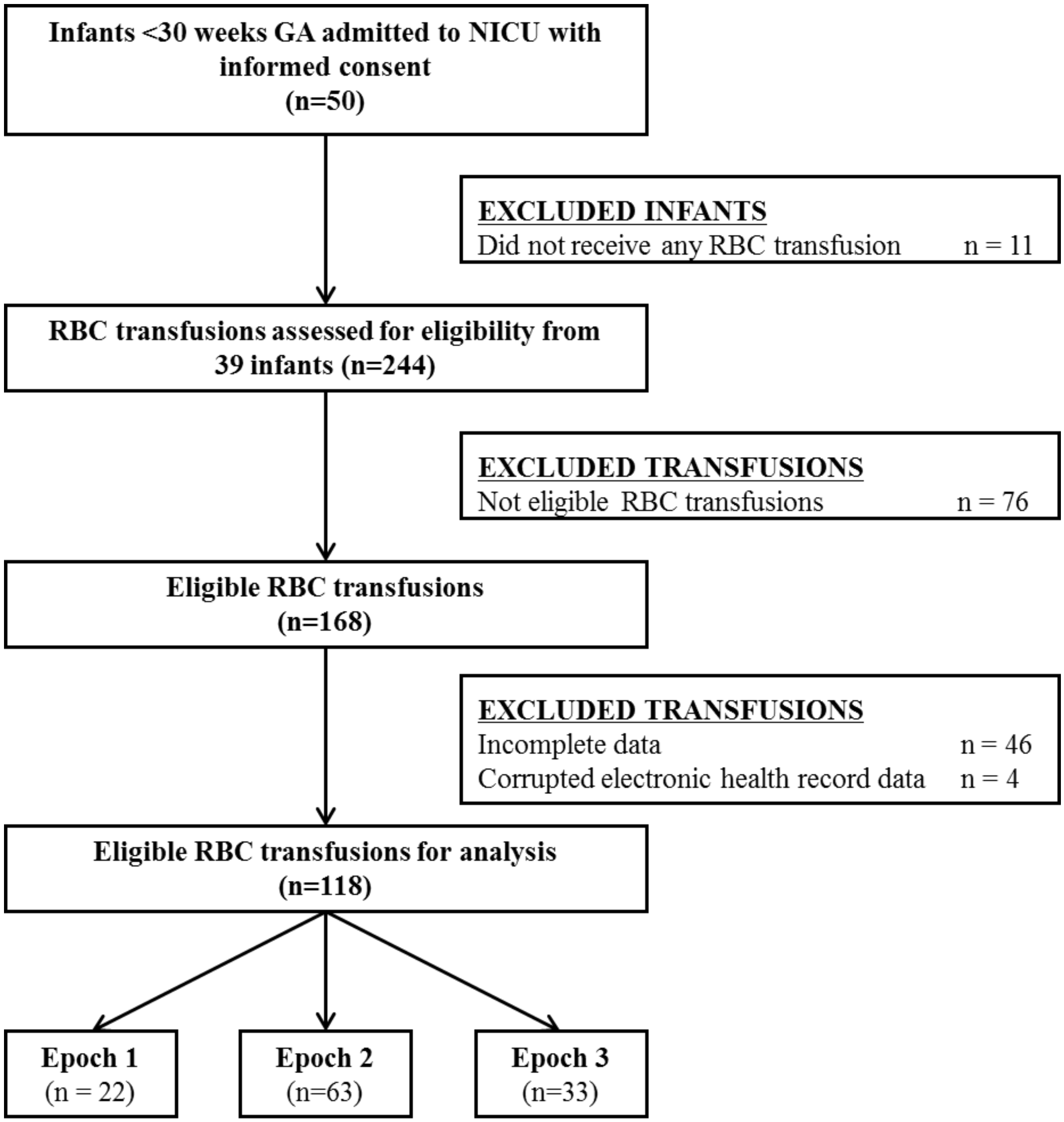

FIGURE 1: Flow diagram for patient enrollment and transfusion eligibility

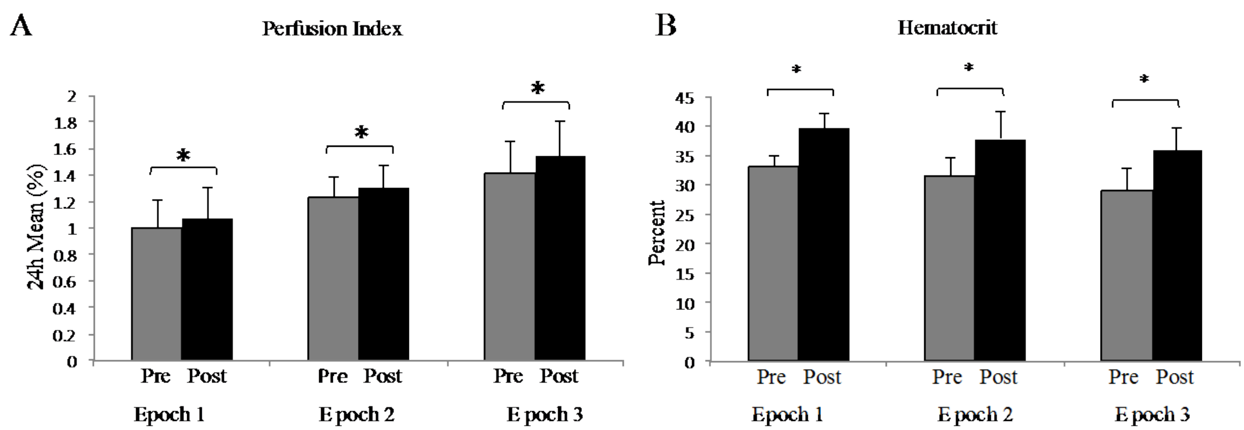

FIGURE 2: Mean PI and Hematocrit levels for all the 3 epochs pre and post RBC transfusion. There was a statistically significant increase in the PI (A) and hematocrit (B) after RBC transfusion in all the three epochs (**p*<0.05). Mean/standard deviation

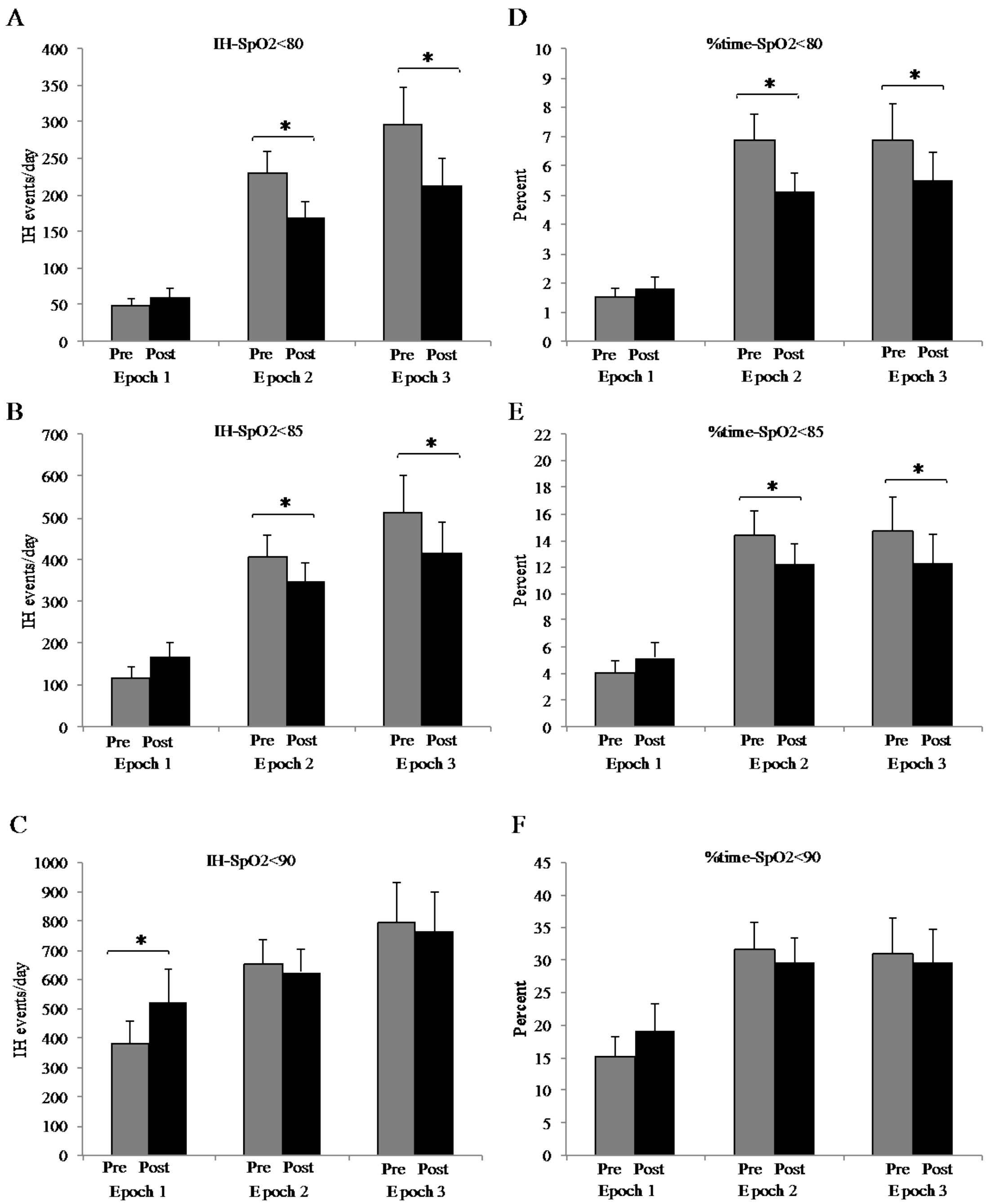

FIGURE 3: IH events/day and % time below threshold pre and post transfusion. 3A-C: IH-SpO_2<80 and IH-SpO_2<85 decreased in epochs 2 and 3 (*p<0.04) while IH-SpO_2<90 increased in epoch 1 (*p=0.04). 3D-F: % time-SpO_2<80 and % time-SpO_2<85 decreased in epochs 2 and 3 (*p<0.04). There was a decrease in % time-SpO_2<90 in epochs 2 (p=0.2) and 3 (p=0.3) and increase in epoch 1 (p=0.07). Mean/standard deviation

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APPENDIX B

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RELATIONSHIP BETWEEN PERFUSION INDEX AND PATENT DUCTUS ARTERIOSUS IN PRETERM INFANTS

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ABSTRACT

Background

Perfusion index (PI) is a noninvasive measure of perfusion. ΔPI (difference between pre- and postductal PI) may identify hemodynamically significant PDA. However, studies are limited to brief and intermittent ΔPI sampling. Our objective is to assess the value of continuous high resolution ΔPI monitoring in the diagnosis of PDA.

Methods

Continuous ΔPI monitoring in preterm infants was prospectively performed using two high-resolution pulse oximeters. Perfusion Index measures (ΔPI mean and variability, pre- and postductal PI) were analyzed over a 4-h period prior to echocardiography. A cardiologist blinded to the results evalu- ated for PDA on echocardiography. Linear mixed regression models were utilized for analyses.

Results

We obtained 31 echocardiography observations. Mean ΔPI (−0.23 vs. 0.16; $P < 0.05$), mean pre-PI (0.86 vs. 1.26; $P< 0.05$), and ΔPI variability (0.39 vs. 0.61; $P = 0.05$) were lower in infants with PDA compared to infants without PDA at the time of echocardiography.

Conclusion

Mean ΔPI, ΔPI variability, and mean pre-PI measured 4 h prior to echocardiography detect PDA in pre- term infants. PI is dynamic and should be assessed continu- ously. Perfusion index is a promising bedside measurement to identify PDA in preterm infants.

BACKGROUND

Patent ductus arteriosus (PDA), a common condition in pre- term infants, leads to shunting of blood between the systemic and the pulmonary circulations. Approximately 65% of infants born between 25 and 28 wk gestational age (GA), and 85% of those born at 24 wk GA will have PDA at first week of life (1). Persistent patency is associated with adverse outcomes, including prolonged assisted ventilation and higher rates of death, bronchopulmonary dysplasia, pulmonary hemorrhage, necrotizing enterocolitis, impaired renal function, intraventricular hemorrhage, periventricular leukomalacia, and cere- bral palsy (1,2). Because of these associated complications, majority of infants < 28 wk GA will receive medical or surgical therapy in an attempt to close the PDA (1–3). Currently, the gold standard for PDA diagnosis is echocardiography (2, 4–8), and often clinical symptoms are not associated with echocar- diography findings (1,4).

Perfusion index (PI) is a noninvasive measure for monitoring the general hemodynamic status of the preterm infant (9–11). Perfusion index provides assessment for the pulse strength and is derived from pulse oximetry. PI, measured by infrared light, is calculated as the ratio of the pulsatile (AC) to nonpulsatile components (DC) of the blood flow in tissue (9,10,12,13). In neonates, PI has clinical application. Granelli et al. (14) corre- lated lower PI values in infants with critical left heart obstructive disease. In addition, De Felice et al. (15) reported that PI was decreased in infants born to mothers with chorioamnionitis.

Reports are inconsistent as to the value of PI in the assess- ment of PDA. This may be attributed in part to location and the duration of PI measurements (12,16). Khositseth et al.(16) hypothesized that the peripheral perfusion of the lower extremities (postductal) is decreased compared to the right arm (preductal) in preterm infants with hemodynamically significant patent ductus arteriosus (hsPDA). This difference is due to left-to-right shunt across the ductus arteriosus into the pulmonary artery. They reported that a difference in PI between the upper and the lower extremity, or delta PI (∆PI), of more than 1.05% strongly correlated with the echocardio- graphic diagnosis of hsPDA (sensitivity: 66.7%, specificity: 100%, positive predictive value: 100% and negative predictive value: 86.4%). Their study was limited by a one-time obser- vation that may not be reflective of the hemodynamic vari- ability of perfusion in infants with PDA. Alternatively, Vidal et al. (12) conducted a study to evaluate the postductal PI of premature infants in order to categorize the PDA status and found that postductal PI did not correlate with PDA and was not influenced by ductal flow pattern.

We conducted a prospective study to assess the value of ∆PI in the diagnosis of PDA in preterm infants, using high resolu- tion continuous pre- and postductal monitoring.

METHODS

This prospective study was conducted at a level IV NICU between November 2014 and July 2015. The study was approved by the Institutional Review Board of the University of Kentucky and parental informed consent was obtained in all cases. Infants with GA ≤ 29 wk were enrolled on the first day of life and followed for a 2-wk period. Infants with major congenital malformations were excluded. Those infants in which we had an echocardiography and adequate PI data for 4 h prior to the echocardiography were chosen for analysis.

Perfusion Index Measurement

Perfusion index was continuously monitored using high resolution (2 s averaging time, 1 Hz sampling rate) pulse oximeters (Masimo Radical Masimo Corporation, Irvine, CA). In order to capture echocardiograms performed for PDA assessment, data were recorded continuously during the first 14 d of life.

Subjects were connected to two pulse oximeters simultaneously; right upper extremity for pre-ductal monitoring and either lower extremity for postductal monitoring. Data from pulse oximeters were continuously stored to serial data recorders. The pre- and postductal PI difference (ΔPI) was defined as the PI measured preductal minus the PI measured postductal (16).

Echocardiography

Two-dimensional, color Doppler, spectral Doppler, and M-mode echocardiography was performed to assess for PDA at the discre- tion of the attending physician using a Phillips IE33 echocardiog- raphy machine with 12-MHz transducer. A cardiologist, blinded to the results of the study, independently examined the echo images and categorized subjects into the following three groups: (i) hemodynami- cally significant PDA (hsPDA); (ii) nonhemodynamically signifi- cant PDA (non-hsPDA), and (iii) no PDA. The definition of hsPDA included infants with at least two of the following: (i) ductal diameter at the pulmonary side ≥ 1.4 mm/kg; (ii) left atrial to aortic ratio ≥ 1.5;(iii) left pulmonary artery (LPA) mean flow velocity of ≥ 0.42 m/s; and (iv) LPA end-diastolic velocity of ≥ 0.2 m/s (3,5,7,17–19).

Sample Size

In order to determine the minimum sample size needed to assess the value of ∆PI in PDA diagnosis, we utilized the results reported in the pilot data by Khositseth *et al*. (16). Assuming the ∆PI (%) mean and SD are 1.00 and 0.70, respectively, for children with PDA and 0.04 and 0.10, respectively, for children with no PDA, we calculated a total required study sample size of 15 infants (power 80%, $p<0.05$).

Data Management and Statistical Analysis

The pulse oximeters serial data recorders were time synced. Perfusion Index sampling rate was 1Hz (every second). However, there were rare occurrences of two values per second. In such cases, the average value for the given second was utilized. In order to better visualize an example of PI values over time (**Figure 2**), we plot PI values that were averaged over each minute. Any given value, at any given second, by itself will not represent a true overall reflection of PI for the duration of several hours, and thus cannot be used to accurately predict PDA. We therefore decided, for predictive purposes, to assess the utility of average ΔPI values during the 4 h prior to an echocardiography as a single measure of PI to predict PDA which could better represent the hemodynamic status of preterm infants. This period of 4 h will capture changes resulting from the ultradian rhythm that has been reported in premature infants (20). Subjects with 4 h of adequate monitoring prior to the echocardiography were considered for analysis. Artifacts and extreme values, found in less than 2% of PI measurements, were removed as they were associated with inadequate signal capture.

Data analyses were conducted by a statistician. The primary outcome of interest is the average ∆PI during the 4 h leading up to echo- cardiography and pre- and postductal PI were secondary outcomes. Furthermore, PI variability was analyzed by using the outcome of the SD of the individual PI values over the 4 h. When comparing mean values for no PDA, non-hsPDA and hsPDA, linear mixed regression models were utilized in order to account for repeated measurements in subjects with multiple echocardiograms. The Kenward and Roger degrees of freedom method was used for inference (21). Generalized estimating equations with the Kauermann and Carroll correction (22) and between-within degrees of freedom were used to evaluate base- line differences among groups defined by PDA status. Analyses were conducted in SAS Version 9.4 (SAS Institute, Cary, NC). All tests were two-sided at the 5% significance level.

RESULTS

A total of 40 infants were enrolled upon admission. Of these, 4 had no echocardiography performed and 16 had missing PI data or artifacts during the study period. Final analyses included data from 20 infants with a total of 31 echocardiography observations (each infant was observed at 1 to 3 occasions) (**Figure 1**). Eighteen infants were found to have PDA on echocardiography. The characteristics at birth of the infants did not significantly differ between those with and without PDA, as shown in **Table 1**. The baseline characteristics of the infants at the time of echocardiography are presented in **Table 2**; no statistically significant differences were noted among groups. As represented in **Figure 2**, PI values were found to be highly variable with changes every minute.

Mean ΔPI differed signifi antly between infants with PDA and without PDA (**Figure 3**). Mean pre- and postductal PI values are presented in **Figure 4**. The preductal PI was significantly elevated in infants without PDA as compared to infants with PDA. Among the PDA subgroups, the preductal PI of those with non-hsPDA was lower compared to infants without PDA (**Figure 4**). The mean postductal PI did not differ among groups (**Figure 4**).

Variability of ΔPI, pre- and postductal PI is presented in **Figure 5**. ΔPI variability was significantly lower in infants with PDA compared to no PDA. Although not statistically significant, the PI variability is consistently low in infants with PDA for pre- and postductal measures.

DISCUSSION

Our study demonstrates that the mean ΔPI, mean pre-PI and the ∆PI variability can identify PDA in premature infants. Mean values of ∆PI, pre- and postductal PI and ∆PI variability were continuously calculated over the 4-h period prior to echocardiography compared to intermittent measures as previously described (11,12,14,23,24). Our observations are somewhat contradictory to initial expectations related to changes in pre-ductal PI and ΔPI. We expected to observe a steady preduc- tal PI and a decreased postductal PI leading to a larger ΔPI in infants with PDA. The negative ΔPI (**Figure 3**) is likely a combination of a decreased preductal PI (reported by Karadag *et al*. (25)) and a postductal PI that is either steady (reported by Vidal *et al*. (12)) or increased (reported by Alderliesten *et al*.(9)). These results have a combined effect towards a negative ΔPI value found in infants with PDA.

We found the preductal PI to be significantly lower in infants with PDA compared to infants without PDA (**Figure 4**). To understand this result, we refer to the definition of PI (AC/DC*100) (15,26), wherein AC is the pulsatile component of the signal and DC is the nonpulsatile component. Infants with PDA can have an absent or reverse flow during diastole in the postductal sites but continuous forward blood flow in the preductal sites (7,8). In infants with PDA, there is also an increase in the cardiac output to compensate for the decreased perfusion in the postductal sites (27–29). This change in cardiac output increases the preductal DC component in infants with PDA compared to no PDA; explaining why the preduc- tal PI is lower in these infants. Our results are consistent with Karadag *et al*. (25) who analyzed the preductal PI in infants treated with surfactant. They found that the incidence of PDA was greater among the infants with a lower preductal PI.

Our study shows no difference between mean postductal PI in infants with PDA and no PDA. Our findings are consistent with Vidal et al. (12) who found no statistically significant difference or correlation between postductal PI and PDA in premature infants. Although not statistically significant, the postductal PI was higher in our infants with PDA compared to infants without PDA (Figure 4). We believe that with PDA there is a decrease in the DC component of the postductal PI due to the overall lower perfusion and decreased mean arterial pressure at the postductal sites (30,31). Furthermore, our find- ings are consistent with the report by Alderliesten et al. (9) who found in a study of 342 neonates that infants with hsPDA had higher postductal PI than infants without hsPDA. They attrib- uted this finding to a hyperdynamic circulation with a widened pulse-pressure resulting in an increase in the AC component. We believe that the increase in postductal PI, if present, is the result of a combination of the effect of the elevated AC component (due to the hyper-dynamic circulation) and a decreased DC component (due to a decreased general perfusion).

Given that the mean ∆PI may not reflect instantaneous hemodynamic changes, we also assessed the variability of the ∆PI over the 4-h monitoring period. Since the correlation of blood fl w and PI has already been established (11, 32), we believe that the ∆PI variability should also correlate with the hemodynamic status of the infant. Our fi dings show that infants with PDA have signifi antly lower ∆PI variability compared to those with no PDA (**Figure 5**). Although trending in the same direction, changes in variability were not statistically significant for pre- and postductal PI (**Figure 5**). The change in ∆PI variability observed in our study is noteworthy since it has not been previ- ously described. De Felice *et al*. (15) speculated that changes in PI variability may be associated with neonatal morbidities, similar to heart rate variability. Decreased heart rate variability in preterm infants with PDA was described by Prietsch *et al*. (33). This decreased heart rate variability resolved after treatment with indomethacin. ΔPI and heart rate variability are valuable at identifying subclinical cardiovascular dysfunction in pre- term infants (15). The variable PI, as a refl ction of the changing hemodynamic status of infants, may also explain the discrepancy among PI values reported in different studies (9,13,14,23,24). Compared to other studies (11,12,14,23,24), we measured PI with high resolution (1s sampling rate) continuous pulse oximetry which gives our study the strength of having high quality monitoring for long periods of time. We advocate for continu- ous measurement of PI compared to spot checks; however, the question that remains to be answered is the optimal monitoring duration needed to detect hemodynamic instability.

The echocardiographic classification of hsPDA used in this study is commonly reported in the literature (8,30,31) but did not correspond to the clinical status of our infants. Those infants designated by echocardiography as hsPDA required less mechanical ventilation, had less FiO2 requirement, and no difference in acidosis compared to non-hsPDA; although not statistically significant (**Table 2**). It is possible that mechanical ventilation may have an effect on PI measures; however, our sample size does not allow to determine an independent effect of ventilation on PI changes. Our study was not designed to establish any correlation between the PI values and the clini- cal severity of the ductus arteriosus. Even though the ductal stealing phenomenon in infants with PDA is well known (17,30,31), its relationship with end organ hypoperfusion and neonatal morbidity remains controversial (34).

Although we achieved the planned observations per our power calculation (accounting for data loss), our sample size is small to evaluate other factors that may affect PI values. Our study has the strength of offering continuous high-quality monitoring throughout the study period. This allowed us to adequately assess the relationship between PI and PDA.

We were able to demonstrate that a lower mean ΔPI and pre PI values over a 4-h period have the potential to detect the presence of PDA in premature infants. We are the first to report a lower variability in ΔPI in infants with PDA compared to infants without PDA. Perfusion index provided by the bedside monitor is a promising bedside tool to assess for PDA in preterm infants. Future studies with a large cohort are needed to determine the clinical utility of PI in predicting PDA and monitoring of its hemodynamic course through days of treatment.

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Table 1. Characteristics of infants at birth*

	PDA $n = 18$	No PDA $n = 13$
GA weeks, median (IQR)	26 4/7 (25 2/7, 27 6/7)	25 2/7 (25 1/7, 26 3/7)
Sex, male (%)	83	46
Weight, median (IQR)	931 (780, 1,030)	730 (660, 855)
Antenatal steroids (%)	89	77
Chorioamnionitis (%)	6	15
Caesarean section (%)	61	62
Surfactant in delivery room (%)	67	77
Time (minutes) to first surfactant, median (IQR)	9 (7, 96)	12 (8,15)
Apgar 1, median (range)	4 (1–8)	3 (2–8)
Apgar 5, median (range)	6 (1–5)	6 (2–8)

*IQR, interquartile range.
All P = NS by Wilcoxon–Mann–Whitney test or chi-square.

Table 2. Infant characteristics at the time of echocardiography, comparing groups, and subgroups*

	No PDA	All PDA	hsPDA	Non-hsPDA
N	13	18	7	11
Day of life, median (IQR)	8 (5, 11)	7 (2, 6)	6 (5, 7)	8 (6, 9)
GA weeks, median (IQR)	25 2/7 (25 1/7, 26 3/7)	26 4/7 (25 2/7, 27 6/7)	27 3/7 (27 3/7, 27 5/7)	26 4/7 (26 3/7, 28 5/7)
Weight, median (IQR)	730 (660, 855)	931 (780, 1030)	1,021 (905, 1030)	880 (775, 1,005)
Heart rate, median (IQR)	163 (144, 173)	165 (156, 172)	165 (157, 173)	164 (156, 168)
Respiratory rate, median (IQR)	49 (32, 61)	54 (38, 69)	68 (50, 81)	46 (35, 63)
Temperature, median (IQR)	98.1 (97.7, 98.4)	98.0 (97.7, 98.3)	97.5 (97.4, 98.1)	98.5 (97.9, 98.7)
Systolic blood pressure, median (IQR)	56.0 (47.0, 66.0)	62.0 (51.5, 74.0)	60.0 (53.0, 73.0)	63.0 (50.5, 75.5)
Diastolic blood pressure, median (IQR)	28.0 (24.0, 45.0)	36.0 (30.0, 43.7)	43.0 (36.5, 43.5)	34.0 (26.0, 43.0)
Mean blood pressure, median (IQR)	42.0 (35.0, 48.0)	41.5 (39.2, 48.2)	42.0 (41.0, 50.0)	40.0 (35.5, 42.5)
% infants on mechanical ventilation	92%	77%	71%	81%
PEEP, median (IQR)	6.0 (5.8, 7.3)	7.0 (6.0, 7.0)	7.0 (6.3, 7.0)	6.5 (6.0, 7.3)
FiO2 requirement, median (IQR)	35.0% (27.0, 60.0)	32.5% (26.5, 50.0)	28.0% (26.0, 40.0)	40.0% (30.0, 52.5)
Blood gas analysis, median (IQR)				
pH	7.3 (7.3, 7.4)	7.3 (7.3, 7.4)	7.3 (7.3, 7.4)	7.3 (7.3, 7.4)
pCO2	43.0 (36.0, 55.0)	44.5 (35.0, 52.2)	42.0 (32.0, 47.5)	45.0 (36.0, 53.0)
HCO3	28.0 (22.0, 29.0)	23.0 (20.2, 25.7)	23.0 (19.5, 24.0)	24.0 (21.5, 26.5)
Base deficit	3.5 (2.1, 5.5)	3.8 (2.2, 6.3)	3.9 (2.5, 5.9)	3.5 (2.3, 6.5)
Urine output (24 h), median (IQR)	3.26 (2.71, 3.41)	2.75 (1.97, 3.90)	3.70 (1.90, 4.76)	2.70 (2.18, 3.39)
Creatinine, median (IQR)	0.78 (0.66, 0.88)	0.71 (0.66, 0.88)	0.71 (0.66, 0.81)	0.70 (0.67, 0.88)
Total fluid volume/kg/day, median (IQR)	135 (130, 150)	130 (122, 140)	130 (125, 135)	140 (125, 145)

IQR, interquartile range; PEEP, positive end-expiratory pressure.
All P = NS by Wilcoxon–Mann–Whitney test or chi-square.

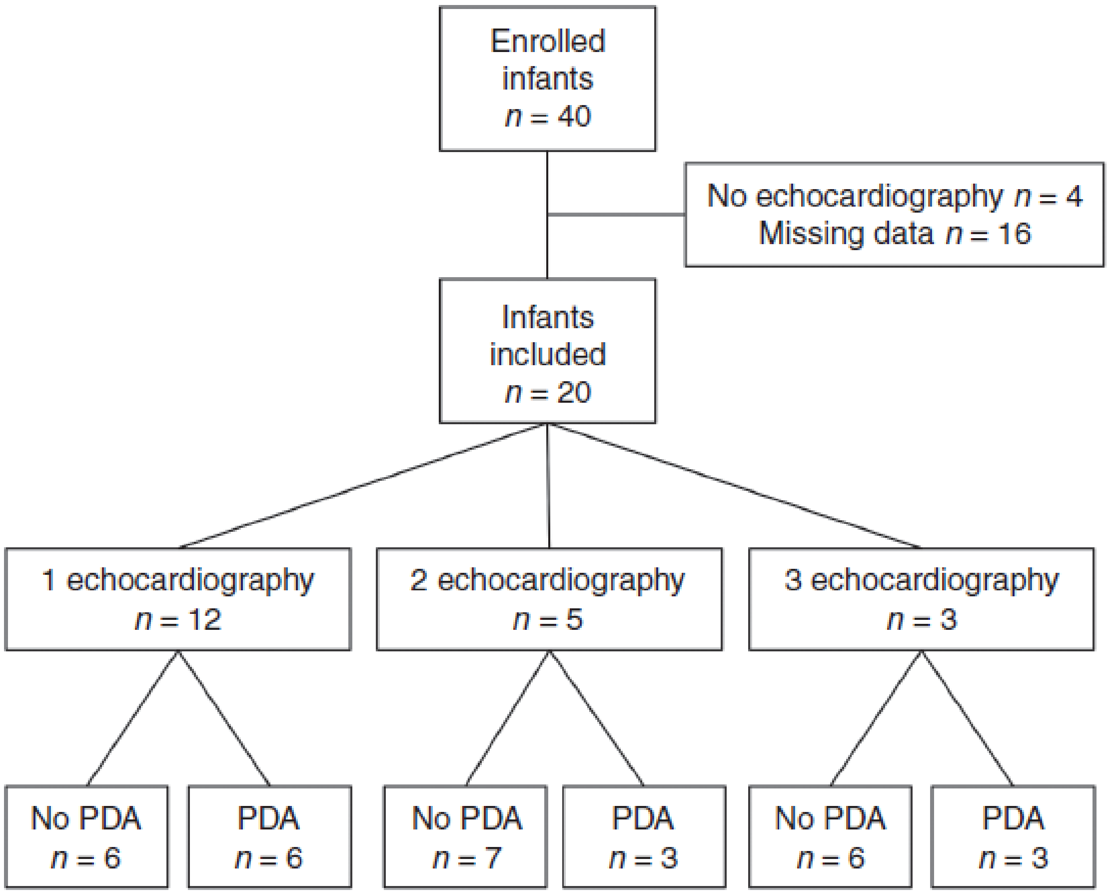

Figure 1. Flow diagram of the enrolled patients.

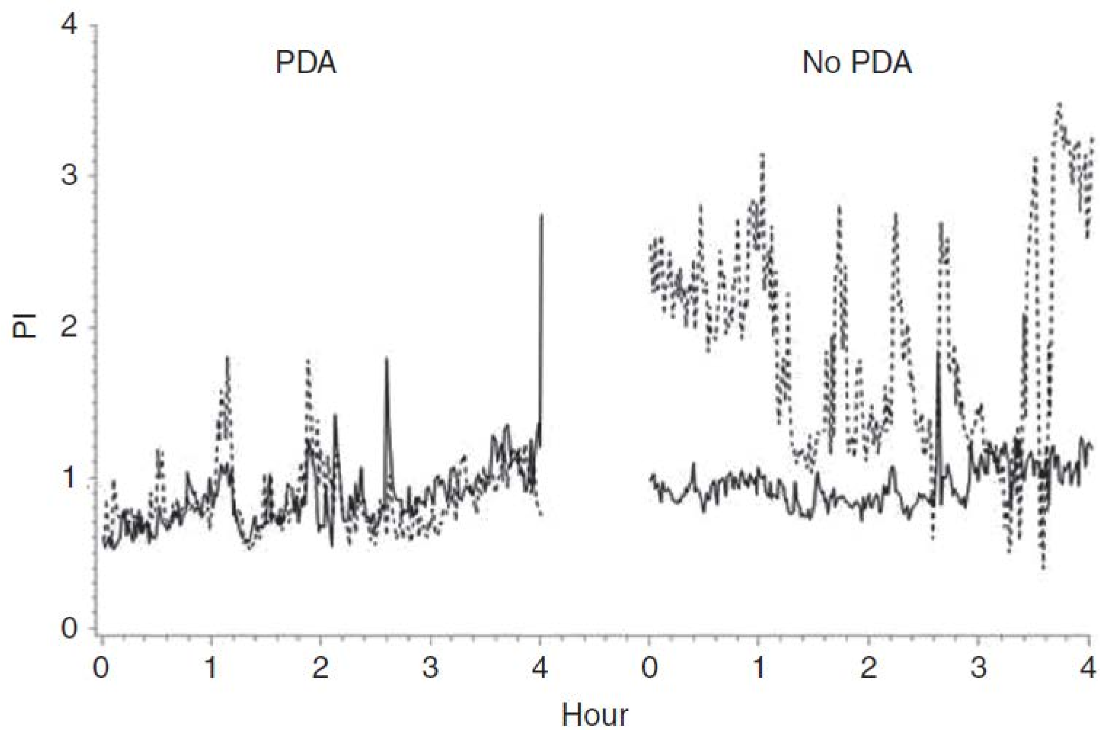

Figure 2. Sample plot representing PI values of one infant with PDA (Variability 0.33, Mean −0.01) and no PDA (Variability 0.71, Mean 0.87) for 4 h prior to an echocardiogram. Dashed lines represent the preductal PI and solid lines represent the postductal PI.

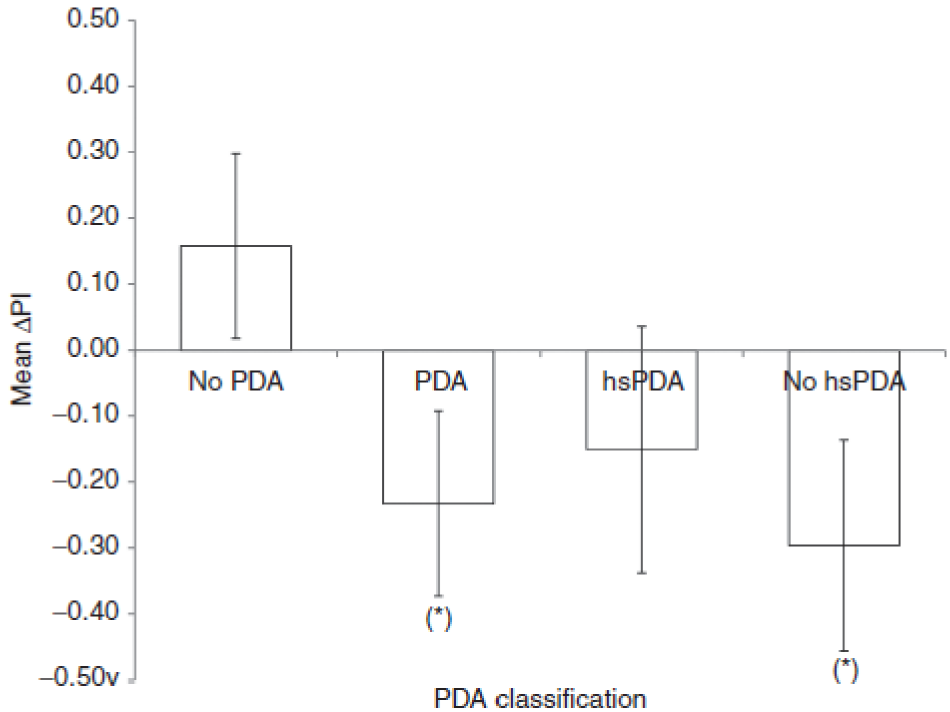

Figure 3. Mean ± SD values of Delta Perfusion Index (ÄPI) 4 h prior to echocardiography. Comparing ÄPI in infants with no PDA vs. infants with PDA, hemodynamically significant PDA (hsPDA) and no hsPDA. **P* < 0.05 compared to no PDA.

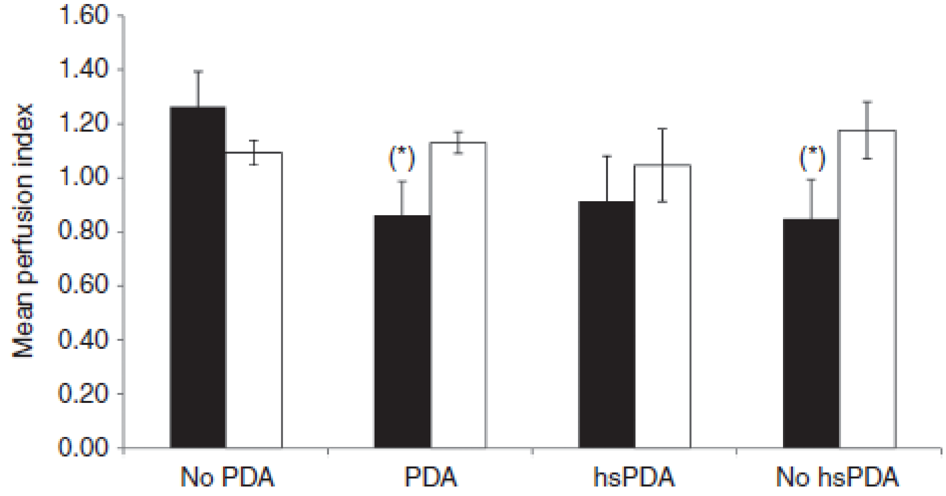

Figure 4. Mean ± SD values of Perfusion Index (PI) 4 h prior to echocardiography. Comparing the preductal (black bar) and postductal (white bar) PI of infants with no PDA vs. infants with PDA, hemodynamically significant PDA (hsPDA) and no hsPDA. *$P < 0.05$ compared to no PDA.

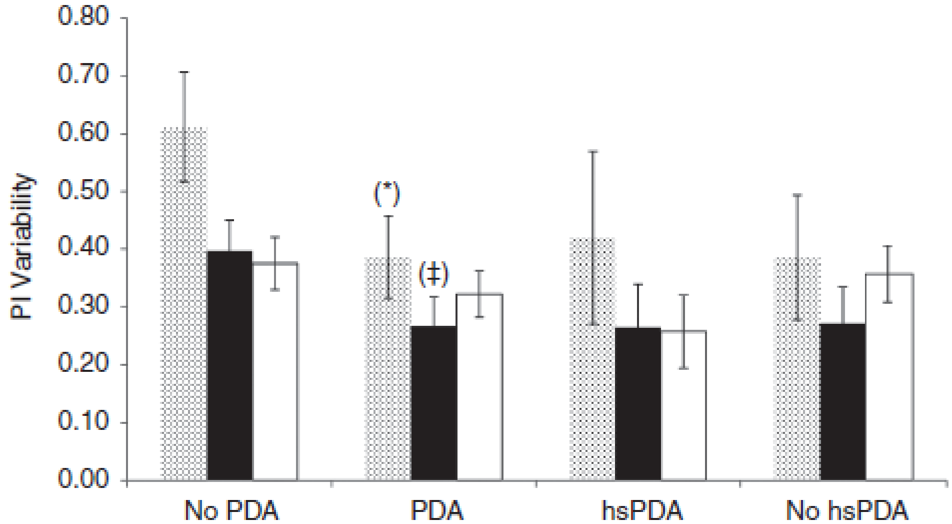

Figure 5. Mean ± SD values of the Variability of Perfusion Index (PI) 4 h prior to echocardiography. Comparing ∆PI (dotted bar), preductal PI (black bar) and postductal PI (white bar) variability for infants with no PDA vs. with PDA, hemodynamically significant PDA (hsPDA) and no hsPDA. *$P < 0.05$ and ‡$P = 0.08$ compared to no PDA.

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185. Abu Jawdeh EG, O'Riordan M, Limrungsikul A, Bandyopadhyay A, Argus BM, Nakad PE, Supapannachart S, Yunis KA, Davis PG, Martin RJ. Methylxanthine use for apnea of prematurity among an international cohort of neonatologists. Journal of neonatal-perinatal medicine. 2013;6(3):251-6. doi: 10.3233/NPM-1371013. PubMed PMID: 24246598.

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195. Basivireddy J, Jacob M, Pulimood AB, Balasubramanian KA. Indomethacin-induced renal damage: role of oxygen free radicals. Biochem Pharmacol. 2004;67(3):587-99. doi: 10.1016/j.bcp.2003.09.023. PubMed PMID: 15037210.

196. Bada HS, Green RS, Pourcyrous M, Leffler CW, Korones SB, Magill HL, Arheart K, Fitch CW, Anderson GD, Somes G, et al. Indomethacin reduces the risks of severe intraventricular hemorrhage. J Pediatr. 1989;115(4):631-7. PubMed PMID: 2677294.

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extremely preterm infants in an all-referral NICU. J Perinatol. 2017;37(8):932-7. doi: 10.1038/jp.2017.71. PubMed PMID: 28617424.

198. Joshi A, Gerhardt T, Shandloff P, Bancalari E. Blood transfusion effect on the respiratory pattern of preterm infants. Pediatrics. 1987;80(1):79-84. PubMed PMID: 3601522.

199. Fanaroff AA, Fanaroff JM, Klaus MH. Klaus & Fanaroff's care of the high-risk neonate. 6th ed. Philadelphia, PA: Elsevier/Saunders; 2013. xiv, 626 p. p.

200. Mirza H, Oh W, Laptook A, Vohr B, Tucker R, Stonestreet BS. Indomethacin prophylaxis to prevent intraventricular hemorrhage: association between incidence and timing of drug administration. J Pediatr. 2013;163(3):706-10 e1. doi: 10.1016/j.jpeds.2013.02.030. PubMed PMID: 23522865; PMCID: PMC3939677.

201. Bennett LM, Gadlin H. Collaboration and team science: from theory to practice. J Investig Med. 2012;60(5):768-75. doi: 10.2310/JIM.0b013e318250871d. PubMed PMID: 22525233; PMCID: PMC3652225.

VITA

ELIE G. ABU JAWDEH, MD, FAAP

Revision Date: April 2018

EDUCATION

Undergraduate

09/1998-06/1999 **Brummana High School**
Brummana, Lebanon
Lebanese Baccalaureate Part II (Equivalent to freshman in USA)
Graduated with Honors

09/1999-06/2002 **American University of Beirut**
Beirut, Lebanon
Bachelor of Science, Biology Major
On Dean's Honors list during senior year

Professional/Graduate

09/2002-06/2006 **American University of Beirut**
Beirut, Lebanon
Medical Doctor (**MD**), Major Medicine,

01/2015-present **University of Kentucky**
Lexington, KY
Doctor of Philosophy (**PhD**), Clinical and Translational Science
GPA 4.0, Passed qualifying exam 01/2017 (PhD Candidate)
[In progress, expected defense 05/2018]
Dissertation Title: Intermittent Hypoxemia in Preterm Infants.

Post Graduate

07/2006-06/2007 **American University of Beirut Medical Center Department of Pediatrics**
Beirut, Lebanon
Post-doctoral Research Fellow (PI, G. Dbaibo)

06/2007-06/2010 **Case Western Reserve University**
Rainbow Babies and Children's Hospital

Cleveland, OH
Pediatrics Residency

07/2007-06/2010 **Case Western Reserve University**
Rainbow Babies and Children's Hospital

Cleveland, OH
International Health Track certificate (Global Child Health)

07/2010-06/2013 **Case Western Reserve University**
Rainbow Babies and Children's Hospital

Cleveland, OH
Neonatal Perinatal Medicine (Neonatology) Fellowship

01/2015-12/2016 **University of Kentucky**
Graduate Certificate in Clinical and Translational Science
Graduate Certificate

ACADEMIC AND CLINICAL APPOINTMENTS

08/2013-present **University of Kentucky,** Lexington, KY
Assistant Professor of Pediatrics
Neonatologist

04/2014-present **University of Kentucky,** Lexington, KY
Director, Infant Respiratory Control (Apnea) Program

04/2014-present **University of Kentucky,** Lexington, KY
Medical Director, Neonatal PA Residency Program

08/2017-present **Baptist Health,** Corbin, KY
Neonatologist

AWARDS AND HONORS

06/2006 *Graduation Ceremony Address*
American University of Beirut, Faculty of Medicine

07/2010 *Ambulatory Care Award*
Case Western Reserve University, Rainbow Babies and Children's Hospital, Awarded to the outstanding graduating resident in continuity clinic.

07/2010 *The Zeithaml Award*
Case Western Reserve University, Rainbow Babies and Children's Hospital, To the Graduating Resident who most demonstrates the characteristics of warmth, thoughtfulness, compassion, a willingness to assist others and a unique ability to relate to children and their families.

06/2014 *Program Directors Award for Excellence in Curriculum Development*
University of Kentucky, Pediatric Residency Program; In acknowledgment of enhancements to pediatric residency education.

06/2015 *New Scientist Travel Award*
American SIDS Institute (AASPP Conference)

06/2016 *Chairman's Research Award*
University of Kentucky; In recognition for outstanding contributions to pediatric research.

12/2016 *Omicron Delta Kappa National Leadership Honorary Society – Nu Circle*

02/2018 *Young Faculty Award*
Southern Society for Pediatric Research (SSPR)

Other Honors & Awards

06/1999 *Old Scholars Award,* Brummana High School
Awarded for leadership, overall academic and extracurricular excellence.

08/2000-06/2002 Member at Large, Biology Student Society, American University of Beirut

06/2004	Corroborated scout, Lebanese Scout Association
10/2008-10/2010	Treasurer, WAAAUB (Worldwide Alumni Association of the American University of Beirut) Northeast Ohio Chapter

LICENSURE AND CERTIFICATION

10/2006	Educational Commission for Foreign Medical Graduates #06900443
07/2010-06/2013	Ohio State Medical Board #095735
06/2013-Present	Kentucky Board of Medical Licensure #46100
09/2006	Lebanese Colloquium in Medicine, Diplomate
10/2010	Diplomate, American Board of Pediatrics
04/2014	Diplomate, American Board of Pediatrics Neonatal Perinatal Medicine

MEMBERSHIPS IN PROFESSIONAL ORGANIZATIONS

07/2006-present	Member, Lebanese Order of Physicians
07/2007-present	Member, American Academy of Pediatrics
07/2010-present	Member, American Academy of Pediatrics, Section on Perinatal Pediatrics
09/2015-present	Member, American Association of SIDS Prevention Physicians
11/2016-present	Elected Member, Society for Pediatric Research
1/2018-present	Board Member, American Association of SIDS Prevention Physicians

NATIONAL/REGIONAL COMMITTEES

09/2002-06/2004	International Federation for Medical Students Association Lebanese Medical Students International Committee Standing Committee of Public Health National Treasurer
06/2004-06/2006	Lebanese Scout Association Mount Lebanon District Commissioner

7/2010 – 6/2013	Member, Ohio Perinatal Quality Collaborative; A statewide quality-improvement collaborative aimed at reducing late-onset sepsis in preterm infants – Rainbow Babies and Children's Hospital.
1/2016 - Present	Planning Committee Member, American Association of SIDS Prevention Physicians (AASPP) Conference

ACADEMIC COMMITTEES

11/2005 – 7/2006	President, Student Representative Committee, American University of Beirut, Faculty of Medicine.
3/2006 – 7/2006	Founding Chairperson, Students Curriculum Committee, American University of Beirut - Faculty of Medicine.
7/2010 – 6/2013	Member, Fellowship Educational Committee, Rainbow Babies and Children's Hospital, Division of Neonatology.
10/2013 – Present	Member, NICU Operations Council Kentucky Children's Hospital, Neonatology
12/2013 – Present	Chairperson, Multidisciplinary Rounding Work Group, Kentucky Children's Hospital, Neonatology.
4/2014 – Present	Member, Tiny Baby Workgroup, Kentucky Children's Hospital, Neonatology.
9/2014 – Present	Member, Fellowship Clinical Competency Committee, University of Kentucky, Neonatology
10/2014 – 10/2016	Member, Family Centered Care, Kentucky Children's Hospital, Neonatology.
2/2015 – 12/2017	Physician Representative, New NICU Design Committee, Kentucky Children's Hospital.

6/2016 – Present	Member, Research, Training and Care Innovation Committee, Obstetrics/Maternal Fetal Medicine/Neonatology Academic Service Line, University of Kentucky.
10/2016 – Present	Member, Clinical Operations and Facilities Committee, Obstetrics/Maternal Fetal Medicine/Neonatology Academic Service Line, University of Kentucky.
10/2016 – Present	Member, Network and Brand Committee, Obstetrics/Maternal Fetal Medicine/Neonatology Academic Service Line. University of Kentucky.
3/2017 – Present.	Chairperson, Neonatology Wellness Board, Department of Pediatrics, Neonatology. University of Kentucky.

MENTORSHIP AND ADVISING ACTIVITIES

	University of Kentucky Lexington, KY
07/2013-06/2016	Mentor (Primary), Chair of Scholarly Oversight Committee Enrique Gomez-Pomar MD, MS. Neonatology Fellow, Department of Pediatrics and Masters in Clinical and Translational Science
07/2013-06/2016	Mentor (Primary), Chair of Scholarly Oversight Committee Katrina Ibonia MD, MS. Neonatology Fellow, Department of Pediatrics and Masters in Clinical and Translational Science
06/2014-05/2017	Resident Advisor Ryan Keith, MD/Pediatrics Resident
9/2015-01/2016	Research Mentor Aayush Gabrani MBBS, Research Staff
09/2015-12/2015	Research Mentor Divya Mamilla MBBS, Research Staff

07/2015-06/2017	Research Co-Mentor / Member of Scholarly Oversight Committee Kelsey Montgomery MD, Neonatology Fellow/Department of Pediatrics
01/2016-6/2017	Mentor Amrita Pant MBBS, Research Staff
04/2016-06/2017	Research Mentor Mandy Brasher, Medical Student
05/2016-09/2017	Research Mentor Jordan Redfield, Medical Student
07/2016-Present	Research Mentor Friederike Strelow, MD, Chief Resident/Pediatrics
02/2017-Present	Research Mentor Audra Stacy, Medical Student
11/2017-Present	Research Mentor Hannah Graff, Medical Student
1/2018-present	Medical Student Advisor Kaitlyn Senay, Medicine 2 Student Lauren Crossman, Medicine 2 Student

REVIEWER

- Pediatric Research, official journal of the Society for Pediatric Research
- BOAJ Pediatrics, editorial board member. Open access journal
- Pediatric Academic Societies (PAS) conference.
- Grant Review, Center for Clinical and Translational Science, University of Kentucky

PEER REVIEWED PUBLICATIONS

1. **Abu Jawdeh EG**, O'Riordan M, Limrungsikul A, Bandyopadhyay A, Argus BM, Nakad PE, Supapannachart S, Yunis KA, Davis PG, Martin RJ. Methylxanthine use for apnea of prematurity among an international cohort of neonatologists. J Neonatal Perinatal Med. 2013 Jan 1;6(3):251-6. doi: 10.3233/NPM-1371013.

2. **Abu Jawdeh EG,** Martin RJ. Neonatal apnea and gastroesophageal reflux (GER): is there a problem?
Early Hum Dev. 2013 Jun;89 Suppl 1:S14-6. doi: 10.1016/S0378-3782(13)70005-7. Review.

3. **Abu Jawdeh EG**, Dick TE, Walsh MC, Martin RJ, and Di Fiore JM. The Effect of Red Blood Cell (RBC) Transfusion on Intermittent Hypoxemia (IH) in ELBW Infants. *J of Perinatology* – 2014 Jun 27;97(12):1240-6

4. Gomez EM, Makhoul M, Westgate PM, Ibonia KT, Patwardhan A, Schanbacher B, Giannone PJ, Bada H, **Abu Jawdeh EG**. The Relationship Between Perfusion Index and Patent Ductus Arteriosus in the Premature Infant, *Pediatr Res*. 2017 May;81(5):775-779. doi: 10.1038/pr.2017.10. Epub 2017 Jan 18.

5. **Abu Jawdeh EG**, Westgate PM, Pant A, Stacy AL, Mamilla D, Gabrani A, Patwardhan A, Bada HS, Giannone P. Prenatal Opioid Exposure and Intermittent Hypoxemia in Preterm Infants: A Retrospective Assessment. *Front Pediatr*. 2017 Dec 6;5:253. doi: 10.3389/fped.2017.00253.

6. Ibonia KT, Bada H, Westgate P, Gomez EM, Bhandary P, Patwardhan A, **Abu Jawdeh EG**. Changes in Perfusion Index and Intermittent Hypoxemia Following Red Blood Cell Transfusion in Preterm Infants. *Transfusion* (In press)

Under review

7. **Abu Jawdeh EG**, Hardin F, Kinnard T, Cunningham MD, Neonatal Post-Graduate Training Program for Physician Assistants: Meeting a Need in Neonatal Care. (Under review)

8. Huang C, Gu Y, Chen J, Bahrani A, **Abu Jawdeh EG**, Bada HS, Yu G , Chen L. A wearable fiberless optical sensor for continuous monitoring of cerebral blood flow in mice. (Under review)

9. RHO Study Group. Use of Home Recorded Oximetry to Safely Discontinue Oxygen in Premature Infants with Bronchopulmonary Dysplasia (Under Review)

INVITED REVIEWS/BOOK CHAPTERS

1. Workbook in Practical Neonatology, 5th Edition – Richard Polin and Mervin Yoder
   Chapter 12: Neonatal Apnea. Ribeiro A, **Abu Jawdeh EG**, Martin RJ

2. How to Help the Children in Humanitarian Disasters, 3rd Edition – Karen Olness, Anna Mandalakas and Kristine Torjesen. *Chapter 1: Care of the Neonate. Chapter 2: Hyperbilirubinemia. Chapter 3: Neonatal Sepsis.* **Abu Jawdeh EG**

3. **Abu Jawdeh EG.** Intermittent Hypoxemia in Preterm Infants: Etiology and Clinical Relevance. NeoReviews.18(11):e637-e46. PubMed PMID: 28099422

Under review

- Workbook in Practical Neonatology, 6th Edition – Richard Polin and Mervin Yoder
  Neonatal Apnea. Ribeiro A, **Abu Jawdeh EG**, Martin RJ

ABSTRACTS / RESEARCH PRESENTATIONS

1. 05/2008 Schnettler L, Solomon M, **Abu Jawdeh EG**, Madden J, O'Riordan MA, Furman LM. Maternal self-efficacy and feeding issues in full term infants in an inner-city pediatric practice. Pediatric Academic Societies (PAS) annual meeting. Baltimore Maryland. Poster Presentation.
2. 06/2009 Schnettler L, Solomon M, **Abu Jawdeh EG**, Madden J, O'Riordan MA, Furman LM. Maternal self-efficacy and feeding issues in full term infants in an inner-city pediatric practice. Rainbow Babies and Children's Hospital 39th Annual Science Day. Podium presentation (Lisa Schnettler).
3. 06/2010 **Abu Jawdeh EG**, Mroueh S, Nabulsi M, Sabra R, Wright M. Development of 360-Degree Evaluations for Medical Students. Pilot Study - Fourth Year Medical Students - Pediatric

Clinical Clerkship. Rainbow Babies and Children's Hospital 40th Annual Science Day 2010; Cleveland Ohio Podium presentation.

4. 06/2010 **Abu Jawdeh EG**, Ciener D, Stryker C, O'Riordan MA, Mercuri-Minich N, Bhola M, Wilson-Costello D. Impact of Inhaled Nitric Oxide Therapy on Very Low Birth Weight Infants. Rainbow Babies and Children's Hospital 40th Annual Science Day 2010; Cleveland Ohio Podium presentation

5. 05/2012 **Abu Jawdeh EG**, O'Riordan MA, Limrungsikul A, Bandyopadhyay A, Argus BM, Nakad PE, Yunis KA, Davis PG, and Martin RJ. Prevalence of Prophylactic Caffeine Use Among an International Cohort of Neonatologists. Pediatric Academic Societies (PAS) annual meeting, Boston Massachusetts. Poster Presentation.

6. 05/2012 **Abu Jawdeh EG,** Martin RJ, and Di Fiore JM. The Beneficial Effect of Red Blood Cell (RBC) Transfusions on Intermittent Hypoxemia (IH) in VLBW Infants Varies with Postnatal Age. Pediatric Academic Societies (PAS) annual meeting, Boston Massachusetts. Poster Presentation.

7. 06/2012 **Abu Jawdeh EG**, O'Riordan MA, Limrungsikul A, Bandyopadhyay A, Argus BM, Nakad PE, Yunis KA, Davis PG, and Martin RJ. Practice Variation in Pharmacotherapy for Apnea among an International Cohort of Neonatologists. Rainbow Babies & Children's Hospital 6th Annual Fellow's Research Day 2012; Cleveland Ohio Podium presentation

8. 12/2012 **Abu Jawdeh EG,** Martin RJ, and Di Fiore JM. The Effect of Red Blood Cell (RBC) Transfusions on Intermittent Hypoxemia (IH) in VLBW Infants. American Academy of Pediatrics (AAP) Section on Perinatal Pediatrics - 81 Perinatal and Developmental Medicine Symposium, Marco Island, Florida. Podium.

9. 03/2015 Gomez EM, Makhoul M, Ibonia KT, Schanbacher B, Patwardhan A, Bauer J, Bada H, **Abu Jawdeh EG**. Perfusion Index for management of hemodynamically significant Patent Ductus Arteriosus (hsPDA) in extremely preterm infants. 10th Annual CCTS Spring Conference. UK Center for Clinical and Translational Science. Lexington Kentucky; Poster presentation

10. 03/2015 Ibonia KT, Bhandary P, Gomez EM, Westgate P, Patwardhan A, Schanbacher B, **Abu Jawdeh EG.** Perfusion Index Predicts the Effect of Red Blood Cell Transfusions on Oxygenation in Preterm Infants. 10th Annual CCTS Spring

Conference. UK Center for Clinical and Translational Science. Lexington Kentucky; Oral presentation (Katrina Ibonia)

11. 03/2015 **Abu Jawdeh EG,** Haynes SS, Westgate PM, Kinnard TB, Garlitz K, Ryzowicz T, Monroe B, Bhandary P. Multidisciplinary Rounding Improves Team Member Satisfaction and Engagement on NICU Rounds. 10th Annual CCTS Spring Conference. UK Center for Clinical and Translational Science. Lexington Kentucky; Poster session

12. 04/2015 **Abu Jawdeh EG,** Haynes SS, Westgate PM, Kinnard TB, Bhandary P. Standardized Rounding Processes Improve Team Member and Parent Engagement on NICU Rounds: Results of the Multidisciplinary Rounding Group. Pediatric Academic Societies (PAS) annual meeting, San Diego California. Podium.

13. 09/2015 Bhandary P MD, **Abu Jawdeh EG** MD, Hanna M MD, Subedi L MD, Gomez Pomar E MD, Barber G NNP, Haynes S BSN, Carpenter A MSN, Hanna M MD. Development of a Golden Hour Protocol for ELBW Infants to Improve Outcomes. Vermont Oxford Network, Chicago Illinois. Poster Presentation

14. 02/2016 Gomez E, Barber G, **Abu Jawdeh, EG**, Subedi L, Haynes S, Carpenter A, Bhandary P. Golden Hour Protocol Improves Quality and Efficiency of Care in Extremely Low Birth Weight Infants. Southern Society for Pediatric Research Annual Meeting, New Orleans, Louisiana February 2016. Poster session.

15. 02/2016 Ibonia KT, Bada H, Gomez EM, Bhandary P, Westgate P, Patwardhan A, Schanbacher B, **Abu Jawdeh EG.** Correlation of Changes in Perfusion Index And Intermittent Hypoxemia Following Red Blood Cell Transfusion In Preterm Infants. Southern Society for Pediatric Research Annual Meeting, New Orleans, Louisiana February 2016. Poster session.

16. 04/2016 Mamilla D, Westgate P, Gabrani A, Pant A, Wasemiller A, Joshi M, Bada H, Bauer J, Giannone PJ, **Abu Jawdeh EG**. Effect of Prenatal Maternal Tobacco Use on Intermittent Hypoxemia and Length of Stay in Preterm Infants: Pilot Study. 11th Annual CCTS Spring Conference. UK Center for Clinical and Translational Science. Lexington Kentucky. Poster session.

17. 04/2016 **Abu Jawdeh EG**, Kinnard TB, Jackson-Belcher L, Cunningham MD. A Neonatology Training Program for Post-Graduate Physician Assistants: Meeting a Need in Neonatal Care. Pediatric Academic Societies (PAS) annual meeting, Baltimore Maryland. Poster session.

18. 04/2016 Huang H, Joshi M, Schanbacher B, **Abu Jawdeh EG**, Giannone P, Bauer J, Bhandary, P. Variation in cord blood hematopoietic stem and progenitor cell subsets in preterm and term infants. Pediatric Academic Societies (PAS) annual meeting. Baltimore Maryland. Poster session.

19. 04/2016 Ibonia KT, Bada H, Gomez EM, Bhandary P, Westgate P, Patwardhan A, Schanbacher B, **Abu Jawdeh EG.** Changes in Perfusion Index And Intermittent Hypoxemia Following Red Blood Cell Transfusion In Preterm Infants. Pediatric Academic Societies (PAS) annual meeting. Baltimore Maryland. Poster session.

20. 04/2016 Gomez E, Barber G, **Abu Jawdeh, EG**, Subedi L, Haynes S, Carpenter A, Bhandary P. Golden Hour Protocol Improves Quality and Efficiency of Care in Extremely Low Birth Weight Infants. Pediatric Academic Societies (PAS) annual meeting. Baltimore Maryland. Poster session.

21. 04/2016 Gomez EM, Makhoul M, Westgate PM, Ibonia KT, Patwardhan A, Schanbacher B, Bada H, **Abu Jawdeh EG**. Perfusion Index does not diagnose hemodynamically significant Patent Ductus Arteriosus (hsPDA) in preterm infants. Pediatric Academic Societies (PAS) annual meeting. Baltimore Maryland. Poster session.

22. 04/2016 Gabrani A, Wasemiller D, Mamilla D, Schanbacher B, Patwardhan A, Giannone PJ, Cunningham MD, **Abu Jawdeh EG**. Extubation failure in preterm infants: A role for monitoring intermittent hypoxemia. 11th Annual CCTS Spring Conference. UK Center for Clinical and Translational Science. Lexington. Poster session.

23. 10/2016 Gomez EM, Makhoul M, Westgate PM, Ibonia KT, Patwardhan A, Schanbacher B, Bada H, **Abu Jawdeh EG**. The Relationship Between Perfusion Index and Patent Ductus Arteriosus in the Premature Infant. Third Annual Neonatal Cardiopulmonary Biology Young Investigators' Forum, Chicago Illinois. Poster symposium (Enrique Gomez-Pomar).

24. 04/2016 **Abu Jawdeh EG**, Pant A, Mamilla D, Gabrani A, Westgate PM, Patwardhan A, Bada H, Bauer J, Giannone PJ Maternal Opiate and Tobacco Use: Effects on Intermittent Hypoxemia in Preterm Infants. Southern Society for Pediatric Research. Lexington Kentucky. Poster session.

25. 09/2016 Bhandary P, Hanna M, Patra A, **Abu Jawdeh EG**, Giannone P. Successful utilization of cord blood for admission testing in very low birth infants. Vermont Oxford Network. Chicago Illinois. Poster session.

26. 09/2016 Patra A, Bhandary P, Hanna M, **Abu Jawdeh EG**, Gomez Pomar E, Barber G, Subedi L, Carpenter A, Haynes S, Giannone P. Evidence Based Standardized Clinical Practice Guidelines Reduce Incidence of Severe Intraventricular Hemorrhage in ELBW Infants. Vermont Oxford Network. Chicago Illinois. Poster session.

27. 02/2017 Patra A, Bhandary P, Hanna M, **Abu Jawdeh EG**, Gomez Pomar E, Barber G, Subedi L, Carpenter A, Haynes S, Giannone P. Reducing Incidence Of Severe Intraventricular Hemorrhage In Extremely Premature Infants: A Quality Improvement Initiative. Southern Society for Pediatric Research (SSPR) Annual Meeting, New Orleans, Louisiana. Poster session.

28. 02/2017 Pant A, Westgate P, Raffay T, Gabrani A, Brasher M, Giannone P, Cunningham MD, Abu **Jawdeh EG**. Extubation Failure in Preterm Infants: A Role for Monitoring Intermittent Hypoxemia. Southern Society for Pediatric Research. New Orleans, Louisiana. Poster session.

29. 02/2017 Redfield J, **Abu Jawdeh EG**, Westgate P, Huang H, Pant A, Bada H, Giannone P, Hanna M. Relationship between Acute Kidney Injury and Intermittent Hypoxemia in Extremely Preterm Infants. University of Kentucky AOA conference. Lexington Kentucky. Poster session.

30. 03/2017 Montgomery KA, **Abu Jawdeh EG**, Goldstein RF, Yozwiak JA, Patra A, Huang H and Ragsdale L. Assessment of NICU Inter-Provider Communication and Patient Safety. Annual CCTS Spring Conference. UK Center for Clinical and Translational Science. Lexington Kentucky. Poster session.

31. 03/2017 Redfield J, **Abu Jawdeh EG**, Westgate P, Huang H, Pant A, Bada H, Giannone P, Hanna M. Relationship between Acute Kidney Injury and Intermittent Hypoxemia in Extremely Preterm Infants. Annual CCTS Spring Conference.

UK Center for Clinical and Translational Science. Lexington Kentucky. Poster session.

32. 03/2017 **Abu Jawdeh EG,** Carpenter S, Wasemiller D, Whitlock H, Savardekar H, Pant A, Schanbacher B, Bada HS, Giannone PJ, Bauer JA, Patwardhan A. Measurement of Intermittent Hypoxemia (IH) Events in Preterm Infants: Development of a Validated Method. Annual CCTS Spring Conference. UK Center for Clinical and Translational Science. Lexington Kentucky. Poster session.

33. 03/2017 Strelow F, Westgate P, Pant A, Patwardhan A, Bada HS, Giannone PJ, Desai N, **Abu Jawdeh EG.** Relationship between Postnatal Weight Gain and Intermittent Hypoxemia (IH) in Preterm Infants. Annual CCTS Spring Conference. UK Center for Clinical and Translational Science. Lexington Kentucky. Poster session.

34. 05/2017 Patra A, Bhandary P, Hanna M, **Abu Jawdeh EG**, Gomez Pomar E, Barber G, Subedi L, Carpenter A, Haynes S, Giannone P. Reducing Incidence Of Severe Intraventricular Hemorrhage In Extremely Premature Infants: A Quality Improvement Initiative. Pediatric Academic Societies (PAS), San Francisco, California. Poster session

35. 05/2017 Bhandary P, Savardekar H, **Abu Jawdeh EG**, Giannone PJ, Hanna M, Patra A, Differences in Sodium Measurements between Point of Care and Laboratory Analyzers in ELBW Infants During the First Week of Life. Pediatric Academic Societies (PAS), San Francisco, California. Poster session.

36. 05/2017 Raffay TM, Dylag A, **Abu Jawdeh EG**, Martin RJ, Di Fiore JM. Neonatal Intermittent Hypoxemia May Predict Bronchopulmonary Dysplasia Risk. Pediatric Academic Societies (PAS), San Francisco, California. Poster session.

37. 05/2017 Bhandary P, Patra A, Hanna M, **Abu Jawdeh EG**, McGee L, Haynes S, Giannone P. Decreasing Phlebotomy in Preterm Infants by Successful Utilization of Cord Blood for Admission Testing. Pediatric Academic Societies (PAS), San Francisco, California. Poster session.

38. 05/2017 Pant A, Westgate P, Raffay T, Gabrani A, Brasher M, Bada HS, Giannone P, Cunningham MD, **Abu Jawdeh EG**. Extubation Failure in Preterm Infants: A Role for Monitoring Intermittent Hypoxemia. Pediatric Academic Societies (PAS), San Francisco, California. Poster session.

39. 2/2018 Abu Jawdeh EG, Westgate P, Pant A, Stacy A, Patwardhan A, Bada H, Giannone P. Relationship between Intermittent Hypoxemia and Inflammation in Preterm Infants: Vicious Cycle. *Southern Society for Pediatric Research. New Orleans February.* Poster session.

40. 4/2018 Strelow F, Westgate P, Pant A, Patwardhan A, Bada H, Giannone P, Desai N, **Abu Jawdeh EG** Evaluation of Postnatal Growth and Caloric Intake in Relation to Intermittent Hypoxemia. Annual CCTS Spring Conference. UK Center for Clinical and Translational Science. Lexington Kentucky. Podium Presentation (Strelow).

41. 4/2018 Stacy A, Westgate P, Patwardhan A, Bada H, Giannone P, **Abu Jawdeh EG**. Pathologic Maternal Chorioamnionitis and Intermittent Hypoxemia in Preterm Infants. Annual CCTS Spring Conference. UK Center for Clinical and Translational Science. Lexington Kentucky. Poster Session

42. 5/2018 Strelow F, Westgate P, Pant A, Patwardhan A, Bada H, Giannone P, Desai N, **Abu Jawdeh EG.** Relationship between Postnatal Growth, Caloric Intake and Intermittent Hypoxemia (IH) in Preterm Infants. Pediatric Academic Societies (PAS), Toronto CA. Poster session.

43. 5/2018 Brasher M, Raffay T, Patwardhan A, Bada H, Giannone P, Westgate P, **Abu Jawdeh EG.** Response to First Dose of Albuterol in Mechanically Ventilated Preterm Infants. Pediatric Academic Societies (PAS), Toronto CA. Poster session.

44. 5/2018 Montgomery K, Goldstein R, **Abu Jawdeh EG**, Yozwiak J, Patra A, Westgate P, Ragsdale L. Impact of Individual Communication Styles on NICU Safety Culture Perception. Pediatric Academic Societies (PAS), Toronto CA. Poster session.

45. 5/2018 RHO Study Group. Use of Home Recorded Oximetry to Safely Discontinue Oxygen in Premature Infants with Bronchopulmonary Dysplasia. Eastern Society for Pediatric Research Meeting. Platform presentation.

RESEARCH SUPPORT

Ongoing support

Title: A Low-cost Compact Diffuse Speckle Contrast Flow-oximeter for Neonatal Brain Monitoring
Source: NIH R21 HD091118-01A1, April 2018 - Mar 2020 (PI, Yu)
Role: Co-Investigator

Title: Comparison of Aerosol delivery of Infasurf to Usual Care in Spontaneously Breathing RDS
Source: ONY, June 2017- June 2019 (PI, Cummings)
Role: Principal Investigator (Site)

Title: A Randomized Trial of Outpatient Oxygen Weaning Strategies in Premature Infants.
Source: Patient-Centered Outcomes Research Institute (PCORI), January 2016 – Dec 2017 (PI, Rhein)
Role: Principal Investigator (Site)

Title: Effect of Delayed Cord Clamping on Chronic Intermittent Hypoxia in Extremely Premature Infants.
Source: The Gerber Foundation, Oct 2014 – Oct 2018. In no cost extension
Role: Principal Investigator

Completed support

Title: Predictors of Intermittent Hypoxia in Premature Infants
Source: Children's Miracle Network, Jan 2014 – Jan 2018.
Role: Principal Investigator

Title: Intermittent Hypoxemia and Acute Kidney Injury (IHAKI study).
Source: Children's Miracle Network. April 2016 – April 2017.
Role: Principal Investigator (Multiple PI, Hanna)

Title: Perfusion index predicts the effect of red blood cell transfusions on oxygenation in preterm infants.
Source: CTSA UL1RR033173 (NCRR). July 2015-July 2016.
Role: Research Mentor

Title: Perfusion Index for Management of Hemodynamically Significant Patent Ductus Arteriosus in Extremely Preterm Infants.
Source: CTSA UL1RR033173 (NCRR). July 2015-July 2016.
Role: Research Mentor

Title: Infant Control of Breathing and Apnea Monitoring Program – Neonatology
Source: WHAS Crusades, July 2014.
Role: Principal Investigator

Submitted grants under-review

Title: Intermittent Hypoxemia in Preterm Infants: Role of Inflammation and Novel Treatment Strategy through a Randomized Placebo Controlled Trial (HIT Study).
Source: NIH R01 Re-submitted March 2018.
Role: Principal Investigator

Pending resubmission

Title: Noncontact High-Density Optical Imaging of Neonatal Brain Function (PI, Yu)
Source NIH R01 (30 percentile, pending resubmission July 2018)
Role: Co-Investigator

COLLABORATIONS AND ACKNOWLEDGMENTS

- Kaplan HC, Lannon C, Walsh MC, Donovan EF; Ohio Perinatal Quality Collaborative: Ohio statewide quality-improvement collaborative to reduce late-onset sepsis in preterm infants. Pediatrics 2011 Mar;127(3):427-35

- Memish ZA, Dbaibo G, Montellano M, Verghese VP, Jain H, Dubey AP, Bianco V, Van der Wielen M, Gatchalian S, Miller JM: Immunogenicity of a single dose of tetravalent meningococcal serogroups A, C, W-135, and Y conjugate vaccine administered to 2- to 10-year-olds is noninferior to a licensed-ACWY polysaccharide vaccine with an acceptable safety profile. Pediatr Infect Dis J. 2011 Apr;30(4):e56-62.

GLOBAL HEALTH

09/2006-06/2007 **Volunteer Outreach Clinic** (Public Service), Non-Governmental Organization associated with American University of Beirut and active in refugee camps in Lebanon. The NGO provides health care and awareness in an outpatient facility located in underserved areas.
Member/Physician volunteer.

03/2010 **Peace Initiative (Iniciativas de Paz)** (Public Service)

Non-Governmental Organization (NGO) active in Central/Latin America
Medical mission for disaster relief following Earthquake in Haiti.
Physician Volunteer

08/2015 **Shoulder to Shoulder (Hombro A Hombro)** (Public Service)
Non-Governmental Organization affiliated with University of Kentucky active in Ecuador. Medical Mission with ambulatory and stationed clinics; we cared for pediatric and adult patients in rural areas.
Physician Volunteer; Supervised UK residents and students.

Other Global Health Related

- Case Western Reserve University, School of Medicine 14th Management of Humanitarian Emergencies, Focus on Children and Families, A Course in Disaster Preparedness – 2010

- Secondary Prevention of Type II Diabetes Mellitus in Lebanon with a Focus on the Practice of Comprehensive Care. Social and Preventive Medicine Public Health Project: field study and report. ***Elie Abu Jawdeh****, Ibhar Al-Mheid, Bilal Ataya, Aline Baghdassarian, Omar Batal, Mohamad Elfakhani, Mentor: Iman Nuwayhid MD, DrPH.* March 2006

- "Exploring Childhood on the Street; When Street Becomes More Homey than Home"; American University of Beirut, Faculty of Medicine, Social and Preventive Medicine, Public Health Project. Field research project and report about street children in Lebanon.

***Elie Abu Jawdeh**, Joelle Abi Rached, Tarek Abou Hamdan, Joelle Amm, Aline Baghdassarian, George Mollayess Mentor: Iman Nuwayhid MD, DrPH.* July 2003

SPECIAL CERTIFICATIONS

- University Hospitals of Cleveland - Rainbow Babies and Children's Hospital,
  ECMO Physician Specialist (2011 - 2015)

- Case Western Reserve University, Collaborative Institutional Training Initiative (CITI), Continuing Research Education Credit Program (CREC) (2008 – 2014)

- American Academy of Pediatrics, Neonatal Resuscitation Program, Provider (2007 - present), Pediatric Advanced Life Support (2007 - 2011)

- University of Kentucky Collaborative Institutional Training Initiative (CITI) Completion Certificate (11/2014-present)

INVITED PRESENTATIONS

08/2014	**University of Kentucky** Department of Pediatrics Lexington, KY Neonatology Grand Rounds: *Neonatal Apnea, Overview*
05/2015	**American University of Beirut** Beirut, Lebanon Rounded with the NICU team and presented to residents/fellows "*Apnea and Reflux in Preterm Infants*"
05/2015	**Contemporary Pediatrics Conference** Lexington, KY Invited Speaker: "*Gastroesophageal Reflux in Infants"*

06/2015 **Case Western Reserve University**
18th Management of Humanitarian Emergencies, Focus on Children, Women and Families.
Cleveland, OH
Invited Speaker: "*Neonatal Resuscitation"*
Moderator: "*Case Discussion"*

09/2015 **American Association of SIDS Prevention Physicians**
Pre-conference research session
Naples, Florida
Invited Speaker: *Intermittent Hypoxemia Research*

11/2015 **UHC/AACN Nurse Residency Program Annual Conference, webinar**
Lexington, KY
Panelist; *Life Adventure Center: A novel approach to improving team* communication *(Webinar)*

12/2015 **University of Kentucky**
Department of Pediatrics
Lexington, Kentucky
Grand Rounds: *Neonatal Apnea and Intermittent Hypoxemia*

02/2016 **University of Kentucky**
Department of Pediatrics, Neonatology
Lexington, KY
Neonatal Grand Rounds: *Management of Gastroesophageal Reflux in the Preterm Infant*

05/2016 **Case Western Reserve University**
19th Management of Humanitarian Emergencies, Focus on Children, Women and Families.
Cleveland, OH
Invited Speaker: "*Neonatal Resuscitation"*
Moderator: "*Case Discussion"*

09/2016 **American University of Beirut**
Department of Pediatrics
Beirut, Lebanon
Grand Rounds: *Neonatal Apnea and Intermittent Hypoxemia*

09/2016 **American Association of SIDS Prevention Physicians Conference**
Naples, Florida
Invited Speaker: *Predictors Intermittent Hypoxemia Research*

01/2017 **University of Kentucky**
Department of Pediatrics, Neonatology
Lexington, KY
Grand Rounds: *GERD? Probably Not*!

02/2017 **National Collaborative for Perinatal Neonatal Network (NCPNN) Conference**
Beirut, Lebanon
Invited Speaker: *Gastroesophageal Reflux in Preterm Infants*
Invited Speaker: *Intermittent Hypoxemia in Preterm Infant*

05/2017 **Case Western Reserve University**
Cleveland, OH
Resident Workshop/Invited Speaker: *Helping Babies Breathe; Neonatal resuscitation in undeserved setting*

09/2017 **American Association of SIDS Prevention Physicians Conference**
Naples, Florida
Invited Speaker: *Intermittent Hypoxemia in Preterm Infants: Consequences*