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approved version of the following dissertation:**

**The Effects of Recorded Music Therapist's Singing Voice
and Recorded Mothers' Singing Voice
on the Transition Preterm Infants from Incubator to Open Crib**

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by

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Dissertation

Presented to the Faculty of the Graduate School of
The University of Texas Medical Branch
in Partial Fulfillment
of the Requirements
for the Degree of

Doctor of Philosophy

**The University of Texas Medical Branch
May, 2015**

Dedication

To all preterm infants and their families who have had to spend time in a neonatal intensive care unit. To my tito, Octavio Jimenez: you just left, but it feels like it has been forever. I hope I made you proud.

Acknowledgements

A special thank you to Dr. Alice Hill for taking me in under her wing and allowing me to blossom as a researcher and an educator. Thank you for your constant guidance and for continuing to keep me grounded throughout this journey. I would also like to thank my committee members: Dr. Martin, Dr. Cupit, Dr. Armentrout, Dr. Verklan, Dr. Shattuck, and Dr. Rath. It is thanks to your knowledge and passion for preterm infants and their well being that I have been able to fulfill my dreams of researching within such a vulnerable population.

I would especially like to thank the staff at The University of Texas Medical Branch Infant Special Care Unit for allowing me to conduct my study on their unit. I am indebted to the nursing staff as they were always willing to lend a helping hand and supported the collaborative efforts that I initially presented.

Finally, I would like to thank my mother, Terri Stout, for her patience and dedication to assuring that my daughter was provided with a loving environment as I focused on conducting this study for the past year.

**The Effects of Recorded Music Therapist's Singing Voice
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Publication No. _____

Jacqueline Stout-Aguilar, PhD
The University of Texas Medical Branch, 2015

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A majority of preterm, very low birth weight, and low birth weight infants is thought to be deprived of auditory stimulation needed for normal maturation. It is known that preterm infants by 26 weeks gestation perceive and respond to sounds such as their mothers' voice and general speech (Hall, 2000), and that auditory stimulation may positively influence physiological and growth outcomes after birth (Cassidy & Standley, 1995). While auditory stimulation is known to affect health outcomes such as weight and length of stay in hospital, what is not clear is whether the changes that occur as a result of the stimulation will influence the infants' transition from the incubator to an open crib and whether one type of stimulus (recorded music therapist's singing voice or infants' mothers' singing voice) is more effective than another.

The purpose of this study was to determine the effect of auditory stimulation (recorded music therapist's singing voice and recorded infants' mothers' singing voice) on preterm infants' 1) length of time to transition from the incubator to the open crib, and 2) growth (weight in grams) during this transition period.

The study used an experimental pre-test, post-test design. Ninety preterm infants enrolled in three study groups were randomly assigned to one of two experimental groups (recorded music therapist's singing voice or recorded mothers' singing voice) or to a control group. Music was played for 20 minutes, three times per day for three days per week until the infant transitioned out of the incubator to the open crib.

Data were analyzed using descriptive statistics (central tendency, ranges, standard deviations, and percentages) to characterize the sample and the Analysis of Variance (ANOVA) was used to determine differences among the three groups on study variables of growth and length of time to transition to an open crib. Findings showed no significant differences between the group on weight gain or length of transition time. Although not significant, a trend was found in weight gain with recorded music therapist voice showing a higher mean weight. From this sample of infants it can be concluded that recorded music has no effect on growth and intra-nursery transition.

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List of Abbreviations

ANCOVA	Analysis of Covariance
ANOVA	Analysis of Variance
CRE	Cardiorespiratory Event
dB	Decibel
GA	Gestational Age
ICU	Intensive Care Unit
IRB	Institutional Review Board
ISCU	Infant Special Care Unit
MSS	Maternal Sound Stimulation
MT	Recorded Music Therapist's Singing Voice
MV	Mother's Recorded Singing Voice
NICU	Neonatal Intensive Care Unit
PI	Principal Investigator
PMA	Postmenstrual Age
RHS	Routine Hospital Sounds
UTMB	University of Texas Medical Branch
VLBW/LBW	Very Low Birth Weight/Low Birth Weight

Chapter 1: Introduction to the Study

INTRODUCTION

Chapter one introduces the study and describes the problem, purpose, and significance of the study. It also discusses the theoretical framework, defines the pertinent terms, variables, specific aims, and related research questions and gives a brief overview of the study design.

PROBLEM STATEMENT

The March of Dimes (2013) reported that 13 million babies are born prematurely worldwide each year, including one in every eight babies in the U.S. A majority of these infants is thought to be deprived of auditory stimulation needed for normal maturation. This stimulation deprivation may affect development of auditory and language skills, as well as later socio-emotional development. Recent evidence of auditory learning and voice recognition in the fetus has suggested that this early preference is based on prenatal experience (Kisilevsky et al., 2009). It has been established that preterm infants by 26 weeks gestations perceive and respond to sounds (Hall, 2000). While it is clear that mothers' voices and lullabies can affect preterm infants' health outcomes (Krueger et al., 2010), it is unclear whether auditory stimuli impact these outcomes such that infants' intra-nursery transition become affected or whether one type of stimulus is more effective than another.

PURPOSE STATEMENT

The purpose of this study was to understand how auditory stimuli (recorded music therapist's singing voice and recorded infants' mothers' singing voice) could affect the transition of preterm infants from incubator to open crib.

STUDY AIMS AND HYPOTHESES

The study objective was to determine if a recorded music therapist's singing voice (MT) and recorded mothers' singing voices (MV) played at or below 60 decibels (dB) decreased the time preterm infants spent in incubators by positively impacting growth parameters (e.g., weight), thus allowing for earlier weaning to an open crib. The central hypothesis was that auditory stimuli would influence the rate of transition between incubator and open crib. At study commencement, no research was found that examined how playing a recorded music therapist's singing voice differed from the playing of the infants' mothers' recorded singing voice in the transition of preterm infants from incubator to open crib. The rationale for this research was that if the effects of a recorded music therapist's singing voice or recorded mothers' singing voice had positive impacts on growth and transition from incubator to open crib, then U.S. Neonatal Intensive Care Units (NICUs) may begin to implement approaches that directly focused on incubator to open crib transition when caring for preterm infants.

To accomplish the central objective of this study, the following specific aims were delineated:

Aim 1: Identify the influence of auditory stimuli (recorded music therapist's singing voice singing voice and recorded mothers' singing voice) on the weight of preterm infants.

H1.1: Preterm infants who receive a regimen of recorded music therapist's singing voice (Group I) or recorded mothers' singing voice (Group II) will gain more weight than infants who do not have music played to them (Group III).

Aim 2: Determine the influence of auditory stimuli (recorded music therapist's singing voice and recorded mothers' singing voice) on transition time for preterm infants from incubator to crib.

H2.1: Preterm infants who receive a regimen of a recorded music therapist's singing voice (Group I) or recorded mothers' singing voice (Group II) will transition from the incubator to crib faster than infants who do not have music played to them (Group III).

Regarding expected outcomes, the items proposed in Aims 1 and 2 were expected to yield positive effects on the length of time spent in an incubator and the weight gained by preterm infants. Such results could positively impact neonates and families because early transition to an open crib will allow for an increase in maternal-infant bonding opportunities, which should promote positive growth and development. Identification of a targeted therapy may allow preterm infants initially placed in incubators to transition to open cribs sooner, which could reduce costs associated with lengthened hospital stays.

SIGNIFICANCE OF THE STUDY

For over 25 years researchers have shown that music could improve physiological responses and growth of premature infants (Stadley, 2002). Results have suggested that music stimulation may significantly reduce initial weight loss, significantly reduce length of NICU and total hospital stays, significantly reduce daily group mean of stress behaviors, increase daily average weight, and increase breastmilk or formula and caloric intake (Caine, 1991). Research in NICU practice supports the use of music in critical areas such as sucking, weight gain, sleep, and recovery from painful procedures (Lowey, 2013). Music therapy is a structured intervention that delivers music with the purpose of

achieving specific therapeutic goals (e.g., stress reduction) for improvement of patients' clinical conditions (Allen, 2013). Recent studies have shown that exposure to maternal voice can significantly increase oxygen saturation (Filippa et al., 2013), decrease apnea and bradycardia events (Doheny et al., 2012), improve weight gain (Zimmerman et al., 2013), and improve feeding tolerance (Krueger et al., 2010). Although research in this area remains ongoing, these preliminary results are of clinical relevance because correlations have linked early feeding outcomes and respiratory stability with later-life language skills.

The goal of this study was to determine whether a recorded music therapist's singing voice and recorded mothers' singing voice can affect the transition of preterm infants from incubator to open crib. The research is significant because it will contribute to a broader understanding of auditory stimulation on intranursery transition time. Specifically, if it can be determined that music—recorded music therapist's singing voice, recorded mothers' singing voice, or both—decreases transition time from incubator to open crib, transition guidelines or hospital policies can be established that include music therapy. Additionally, such findings may facilitate a decrease in costs associated with hospital length of stay.

RATIONALE FOR USE OF MUSIC THERAPY IN THE NICU

A systematic review of 10 studies with a total enrollment of 780 participants revealed that music significantly benefits an infant by positively affecting observed behavioral state, decreasing heart rate, decreasing respiration rate, raising oxygen saturation level, increasing weight gain, decreasing days in hospital, and positively affecting feeding rate/non-nutritive sucking rate (Standley, 2001).

For the purposes of this study, the use of music therapy was incorporated in order to assess its effects on weight gain and intra-nursery transition of the preterm infant.

DELIMITATIONS

The data collection phase of this study was conducted from October 2014 to February 2015. The study was conducted in an Infant Special Care Unit (ISCU in Southeast Texas. Ninety (90) preterm infants were enrolled in the study. The results of this study could be generalizable to a) preterm infants, and b) infants in hospital NICUs in southeast Texas.

ASSUMPTIONS

Although the lullaby “Twinkle Twinkle Little Star” has been shown to be a “therapeutic lullaby” because it has a defined applied pitch range “higher” or “lower” for singing (Loewy, 2013), it was assumed that the same lullaby would increase the infants’ weight gain enough to show a difference in transition time for the intervention groups. It was also assumed that mothers in any of the groups may sing to their infants outside of the intervention or control times. However, the prescribed nature of the intervention (i.e., pre-recorded music of the music therapist and mothers’ singing voice, specified length of time and frequency) would make it difficult for mothers to deliver the same interventions informally.

DEFINITION OF TERMS

Music Therapy: The conceptual definition of music therapy is a structured intervention that delivers music with the purpose of achieving specific therapeutic goals for improvement of patients’ clinical conditions (Allen, 2013). The operational definition

of music therapy in this study is the playing of a recorded lullaby “Twinkle Twinkle Little Star” that was sung by a music therapist or infants’ mothers. The music was delivered to the infants three times each day from time of enrollment in the study to transition from incubator to crib.

Very Quiet Sleep: Infants’ eyes are closed, and respiration is relatively regular and abdominal in nature. A tonic level of motor tone is maintained, and motor activity is limited to occasional startles, sighs, or other brief discharges (Brandon et al., 1999). In this study, observations were made using these criteria at the beginning and end of every intervention period.

Active Waking: Eyes are usually open, dull, and unfocused but may close during crying or high-level activity. Motor activity is typically high (Brandon et al., 1999). In this study, observations were made using these criteria at the beginning and end of every minutes throughout the intervention period.

Drowsiness: Eyes are “heavy-lidded,” opening and closing slowly, or open but dazed in appearance. The level of motor activity is low and respiration fairly even.

Alert: Eyes are open and scanning. Motor activity is typically low (Brandon et al., 1999).

Sleep-Wake Transitions: Mixed waking and sleeping behaviors with generalized motor activity. Eyes are typically closed, but there may be rapid opening and closing (Brandon et al., 1999).

Active Sleep: Eyes are closed with intermittent rapid eye movement. Respiration is uneven. Sporadic movements occur, but muscle tone is low between movements (Brandon et al., 1999).

Weight Gain: Increase in infant weight measured in grams.

Transition time between incubator and crib: The total amount of time the infant spent in the incubator. This was measured in days.

ORGANIZATION OF THE STUDY

The dissertation is organized into five chapters. Chapter one introduces the study, purpose of the study, study aims and hypotheses, assumptions, significance, and definitions. Chapter two provides the background literature and analyzes current and previous literature focused on the development of preterm infants' auditory system, sounds in incubators, and music therapy in NICUs. Chapter three discusses the methodology and describes the study sample, location, and interventions applied to study participants. Chapter four presents the results of the study including the characteristics of the sample, data analysis, and results for each research question.

Finally, Chapter five provides a summary of the study and further clarifies the findings. The strengths and weaknesses of the study are discussed as well as nursing implications. Recommendations for future research on music therapy in neonatal intensive care units are presented.

Chapter 2: Literature Review

INTRODUCTION

Chapter two presents a review of the related literature of auditory development in the preterm infant along with a review of the effects of noise on the preterm infant. In addition, sounds in incubators and a rationale for music therapy in the NICU are presented.

AUDITORY DEVELOPMENT

The fetus is surrounded by amniotic fluid and the first sounds it experiences are low-frequency digestive noises and maternal sounds transmitted through its cranial bones (Lecanuet & Schaal, 1996; Querleu et al., 1988; Sohmer et al., 2000). Auditory system development is an elaborate process that begins very early in gestation (Hall, 2000). All major structures of the ear, including the cochlea, are in place at 23 – 25 weeks of gestational age (GA) (Cheour-Luhtanen et al., 1996). The human fetus can perceive and react to auditory information beginning at approximately week 26 of GA (Ruben, 1992). Between 26 – 30 weeks GA, hair cells in the cochlea become finely tuned to specific frequencies and can translate vibratory acoustic stimuli into electrical signals that are sent to the brainstem (Querleu, 1989). Beyond 30 weeks GA, the auditory system is mature enough to interpret complex sounds and distinguish between different speech phonemes (Hepper et al., 1993) that are likely the beginning of language and speech development (Mehler et al., 1988).

After birth, most preterm infants can hear when they are admitted the NICU unless they are born with a congenital anomaly. Immediately following birth, preterm infants lose the fluid they are surrounded by *in utero*, which in turn requires them to hear

via air conduction even though their auditory systems previously used bone conduction. The major difference in the primary mode of hearing (bone vs. air conduction) and the medium of sound transmission (fluid vs. air) presents an acoustic gap between the unnatural acoustic environment of the hospital and the developmental demands of the newborn's auditory system (Lahav & Skoe, 2014). Understanding the fundamentals related to hearing in preterm infants provides a foundation for understanding infants' ability to hear sounds such as music.

EFFECTS OF NOISE ON THE PRETERM INFANT

Physiological Impact

NICU preterm infants are primarily exposed to auditory stimulation that includes noise generated by ventilators, infusion pumps, fans, telephones, pagers, monitors, and alarms. The continual exposure to these high-frequency noise levels and recurrent electronic noises may cause trauma to infants' auditory systems. It is well documented that high noise levels are harmful to preterm infants and result in increased physiologic stress (Lahav & Skoe, 2014). Even brief stimulation from alarms and telephones can result in an autonomic response increase with a subsequent physiologic decline that places infants at risk for both bradycardic and hypoxic episodes (Bremmer et al., 2003). Researchers believe that these sounds may have a long-term harmful effect, thus increasing the risk for auditory, language, and attention disorders later in life (Lahav & Skoe, 2014).

Behavioral State Impact

In addition to physiological changes, researchers have shown that infants' behavioral state, which is a group of characteristic behaviors and physiologic changes that recur together in a regular pattern (Brazelton & Nugent, 1996; Wolff, 1966), may be affected by NICU noise levels. Each behavioral state is organized into patterns that differ from other states, and states are divided into sleep and awake states (March of Dimes, 2003). Seven categories of behavioral state have been studied with preterm infants: quiet sleep, active sleep, sleep-wake transition, drowsy, alert, non-alert waking activity, and crying. These categories were developed by Thoman and modified by Holditch-Davis (Holditch-Davis & Thoman, 1987; Thoman, 1987, 1990; Thoman et al., 1975, 1985). These categories exhibit reliable individual differences and accurately profile behavioral states of young preterm infants during the neonatal period (Holditch-Davis & Edwards, 1998a, 1998b). Development of sleeping and waking states throughout infancy reflects the development of the central nervous system (Ardura et al., 1995; Ingersoll & Thoman, 1999). Deviations from normal state development can indicate neurological pathology (Holditch-Davis et al., 1998, 2004). Wolff (1966) identified several distinct behavioral states including "alert-inactivity," "alert-activity," and "drowsy" states and noted that the percentage of time infants spent in "alert-inactivity" increased steadily over the first month of life. Common categorizations of behavioral states used by researchers include quiet sleep, active sleep, drowsiness, quiet alert, active alert, crying, and indeterminate (Korner, 1972; Thoman, 1990; Wolff, 1966). In both preterm and full-term infants, a predictable pattern of state development has been observed and is characterized by a decrease in active sleep and an increase in quiet sleep, alert states, and sleep state

organization (Foreman et al., 2008; Holditch-Davis, 1990; Holditch-Davis & Edwards, 1998a; Holditch-Davis et al., 2004). Differences in the frequency of the different behavioral states have been noted in infants born at 34, 37, and 40 weeks GA (Mouradian et al., 2000). Immature development of cortical organization, particularly in the frontal and prefrontal regions that control behavioral states, may contribute to these observed differences (Duffy et al., 1990). Compared to full-term infants, preterm infants spend more time in transitional states and less time in alert/awake states (Holditch-Davis, 1990; Holditch-Davis & Edwards, 1998a; Holditch-Davis et al., 1998). Preterm infants have a limited ability to maintain an alert state (Panniers, 2002). However, this limited ability to maintain alertness is important for social interaction and is correlated with the ability to feed orally (Gill et al., 1988; Panniers, 2002; Pickler et al., 2005; White-Traut et al., 2002b, 2005). It is important to fully understand the impact of sound on the infants' behaviors in the NICU, especially as it relates to the transition of the infant from incubator to the crib.

In the past 20 years, the survival rate of preterm, Very Low Birth Weight/Low Birth Weight (VLBW/LBW) infants has improved dramatically (Fanaroff et al., 2007). Yet as more preterm infants reach school age, the high incidence of neurodevelopmental problems becomes increasingly apparent (Marlow et al., 2005). There is growing concern that these problems may, in part, stem from an unfavorable NICU environment, particularly excessive noise exposure (Perlman, 2009). Because of these unfavorable exposures, researchers have suggested that it is necessary to compensate preterm infants for the loss of their own mothers' voice and heartbeat—sounds that are highly preferable to infants (Fifer & Moon, 1990). Safe sound levels within the NICU are essential for the

healthy development of preterm infants, and an understanding of recommended and actual sound levels is important for caregivers.

SOUNDS IN INCUBATORS

Premature newborns are prone to having a wide range of long-term developmental problems, including issues with hearing, attention, speech, language, spatial orientation, motor behavior, and self-regulation (Bhutta et al., 2002). There have been increasing concerns that some of these problems may be due to the impact of the NICU environment on the developing brain. Panagiotidis and Lahav (2010) conducted a pilot study in which they analyzed the safety and feasibility of a specialized audio system created for NICU incubators. This system allowed for the playing of preterm infants' mothers' voices and other meaningful sounds in incubators to provide for auditory stimulation that would have otherwise been provided *in utero*. The proposed audio system included two rechargeable speakers (iHM79, SDI Technologies, Rahway, NJ) that were connected to a small commercially available MP3 player (Sansa1 Clip, SanDisk, Rahway, NJ). The player was installed externally on the railing system behind the incubator. The recording of the voice and heartbeat of a mother of a premature infant (26 weeks postmenstrual age [PMA]) admitted to the Brigham and Women's Hospital NICU was conducted in a digital workstation. Sound level measurements inside the incubator revealed that at baseline with no sounds the level was 48.4 ± 0.3 dB; with mothers' voice only the level was 58.1 ± 0.9 dB; with heartbeat only the level was 51.0 ± 0.7 dB; and with maternal voice and heartbeat together the level was 58.6 ± 0.7 dB. It was determined that the specific system tested was deemed safe and feasible and did not interfere with the routine functionality of incubators.

A research utilization project conducted by Medoff-Cooper (1994) successfully identified the standardization of a protocol for weaning preterm infants (without the use of any other interventions) to open crib. As a result, preterm infants were noted to be safely moved to open crib at a much lower weight than had been shown in the few published clinical trials. Medoff-Cooper (1994) enrolled 270 infants from 10 hospitals, with the mean GA being 29.3 ± 2.8 weeks. The mean birth weight for preterm, VLBW/LBW infants was $1,118.05 \pm 269.64$ grams. The mean weight when infants were moved to an open crib was $1,598 \pm 135.7$ grams. The mean time for temperature stabilization after infants were placed in an open crib was 63 ± 41 minutes. This research utilization project was successful in that it demonstrated that preterm, VLBW/LBW preterm infants could be moved to open cribs at a much lower weight than had been shown in earlier clinical trials, which had found that infants could not be weaned prior to weighing 1,700 grams. Nonetheless, transitional approaches have not been effective in analyzing how any external variables, such as music therapy, affected the transitional phase.

MATERNAL SOUNDS IN NICU INCUBATORS

Very low birth weight infants (<1,500 grams) are at a heightened risk for developing growth restriction due to inadequate nutrition in the first weeks of life (Clark et al., 2003). Aside from being preterm, VLBW/LBW infants, there is also a lack of maternal exposure while inside a closed incubator which does not provide the preterm infant with an optimal environment for maturation and development. One method that has been shown to potentially optimize the NICU incubator is to attempt to reinstate the maternal aspects of the intrauterine environment to the infant by exposing the infant to

their own mother's voice and/or heartbeat that they are typically exposed to while in the womb. McMahon et al. (2012) conducted a study to provide preterm, VLBW/LBW infants with biological maternal sounds (BMS) and measure the effects on weight gain velocity during the neonatal period. Thirty two VLBW/LBW infants were enrolled in this study. Subjects included those that were admitted to the Brigham and Women's Hospital (BWH) NICU in Boston, MA. A specialized studio was used to record the BMS sounds at the selected institution. The voice recordings were obtained using a large-diaphragm condenser microphone (KSM44, Shure, Niles, IL) which captures a wide range of maternal vocalizations, including speaking, reading, and singing. Heartbeat recordings were acquired with a digital stethoscope (ds32a, Thinklabs Digital Stethoscopes, Centennial, CO). The recorded soundtrack was then mixed with soothing sounds and uploaded onto an MP3 player (Philips Electronics, SA2RGA04KS, Amsterdam, The Netherlands) and was played inside infants' incubators or cribs four times per 24-hour period for a total of 45 minutes. The control group did not receive BMS and routine care was provided for each infant assigned to the control condition. The 28-day growth trajectory revealed that, on average, infants exposed to BMS gained significantly more weight ($1,220 \pm 159$ grams) compared with matched controls ($1,204 \pm 137$ grams; $t = 3.35$, $p = 0.002$). Findings suggest that exposure to such biological sounds during NICU hospitalization could improve weight gain velocity within the first 28 days of life in preterm, VLBW/LBW infants.

Preterm infants are prone to experiencing frequent cardiorespiratory events (CREs) including multiple daily episodes of apnea and bradycardia. Physiological instabilities, such as the aforementioned entities, are due to the infant's immature

autonomic nervous system which limits the capacity for self-regulation. McMahon et al. (2012) examined whether systematic exposure to maternal sounds could reduce the frequency of CREs in NICU infants. Fourteen preterm infants with gestational ages ranging from 26-32 weeks were enrolled and served as their own controls. The frequency of CRE's was recorded while the infant was exposed to either Maternal Sound Stimulation (MSS) or to Routine Hospital Sounds (RHS). The MSS included recordings of the each infant's mother's voice and heartbeat sounds. MSS was provided four times per 24-hour period via a micro audio system installed in infants' beds. The frequency of adverse CREs was determined based on monitor data and bedside documentation. It was found that there was an overall decreasing trend in CREs in relation to age, with lower frequency of CREs being observed during exposure to MSS versus RHS. This effect was significant in infants ≥ 33 weeks PMA ($p=0.03$), suggesting an effective therapeutic window for MSS when infants' auditory brain development is most intact. This study provides preliminary evidence for short-term improvements in the physiological stability of NICU infants using MSS.

These studies illustrate that sound can be used safely in an incubator with preterm infants, that it can improve weight gain velocity, that it can allow infants to be moved from an incubator at a much earlier age, and that it can provide short-term improvement in physiological stability. These findings lay the groundwork for examining sound in an incubator and promotion of safe transition to open crib at an earlier age.

RATIONALE FOR USE OF MUSIC THERAPY IN THE NICU

Standley (2001) concluded that lullaby selections for premature infants can soothe and calm infants and also convey the human voice, providing much-needed language

stimulation. Research has shown that language development is faster if language is individually directed to infants and if it is provided in “parentese” (speech with song like qualities). Parentese, or the way adults tend to speak with babies, is very similar to lullaby music; this phenomenon seems to be true across all cultures (Trehub, 1993). Standley (2001) reviewed 10 studies with a total enrollment of 780 participants and found that music significantly benefits an infant by positively affecting observed behavioral state, decreasing heart rate, decreasing respiration rate, raising oxygen saturation level, increasing weight gain, decreasing days in hospital, and positively affecting feeding rate/non-nutritive sucking rate.

Loewy et al. (2013) conducted a randomized clinical multisite study where 272 premature infants aged 32 weeks received three interventions per week within a 2-week period. Each infant was randomly assigned to one of four groups: control, lullaby, gatto box, or ocean disk. The lullaby was either parent selected, or if a song could not be identified, “Twinkle, Twinkle Little Star” was used. Results concluded that lower heart rates occurred during the lullaby ($P = .001$) and rhythm intervention ($P = .04$), while sucking behavior showed differences with rhythm sound interventions ($P = .03$). Entrained breath sounds resulted in lower heart rates after the intervention ($P = .04$) and differences in sleep patterns ($P = .001$). Caloric intake ($P = .01$) and sucking behavior ($P = .02$) were higher with parent-preferred lullabies. It was also noted that music decreased parental stress perception ($P = .001$).

Cevasco (2008) conducted a study in which mothers’ recorded voices were played 3 – 5 times per week for 20-minute sessions. Twenty preterm infants were randomly assigned to either experimental or control groups, with those in experimental groups

being played recorded songs that were selected and sung by their own mothers. The results of this study indicated that preterm infants in experimental groups who listened to CD recordings of their mothers' singing left the hospital an average of two days earlier than those who were randomly assigned to control groups.

Polkki et al. (2012) analyzed parents' expectations regarding use of music in NICUs. A cross-sectional, descriptive, and correlational-guided survey was distributed to 197 parents from five hospital NICUs in Finland. Results indicated that most parents agreed that preferred music could have a positive impact on their child, the staff, and parents in NICUs.

Polkki et al. (2012) conducted a separate study to describe expectations of using music for premature infants in the NICU. Two-hundred and ten Finnish nurses were recruited from within the country's five university hospitals that provided care to premature infants in a NICU setting. A valid and reliable questionnaire was distributed to the NICU nurses and the final response rate was found to be 82%. Findings indicated that nurses surveyed in the NICU preferred recorded music over live music. The nurses expected that the music would positively impact the preterm infants, their parents, and the NICU staff. It was concluded that the nurses' expectations were positive regarding the use of music in the NICU and this notion supports the evidence regarding the efficacy of music therapy for premature infants.

Amini et al. (2013) conducted a study to assess and compare the influence of lullaby and classical music on physiologic parameters. Twenty-five preterm infants were randomized into a clinical trial with a cross-over design. Infants with birth weights of 1,000 – 2,500 grams were studied for six consecutive days. Each infant was exposed to

three phases: lullaby music, classical music, and no music (control) for two days each. The sequence of these phases was assigned randomly to each subject. Results indicated that lullabies reduced heart rate ($p < 0.001$) and respiratory rate ($p = 0.004$). These effects extended in the period after exposure ($p < .001$ and $p = 0.001$, respectively). It was also noted that classical music reduced heart rate ($p = 0.018$). The effects of classical music disappeared once the music stopped playing. Oxygen saturation did not change during intervention. Findings suggest that music can affect vital signs of preterm infants, and that effects may be related to a reduction of stress during hospitalization.

Chapman (1978) and Malloy (1979) analyzed the effects of exposure to taped recordings of the maternal voice, an orchestra playing Brahms' "Lullaby," and standard NICU care (control group). Malloy (1979) evaluated weight gain and selected developmental scales from the Rosenblith Behavioral Examination of the Neonate tool and the Bayley Scales of Infant Development tool one day after discharge and at nine months of age. The gestational age of the enrolled infants ranged from 26-33 weeks. The infants in the intervention group were exposed to a recording of their mothers speaking or a lullaby (stimuli presented at 70-75 dB) for five minutes, every two hours, six times daily until their weight reached 1,844 grams. The control group did not receive any auditory stimulation and all NICU standards of care were maintained. Findings indicated no statistical differences in the developmental scales between groups. It was noted that the experimental group gained weight faster because they reached the weight selected for testing (1,844 grams) faster. Infants exposed to the lullaby reached the selected weight significantly earlier (9.9 days; $F = 4.33$, $P = .05$) than those in the control group.

SUMMARY

The literature demonstrates that preterm infants are able to hear by 26 weeks GA and that sound, especially loud noises, can lead to neurological deficits, language deficits, and hearing difficulties in later life. But when sounds, such as music, are used at safe decibel levels with preterm infants in an incubator, infants tend to gain weight and have improved physiological stability. Likewise, music therapy has been shown to improve oxygen saturation, lower heart rate, increase weight gain, and shorten hospital stay. It has been shown that lullabies are effective in helping infants reach their selected weight faster than control groups. Given that lullaby music can help infants reach a predetermined weight faster and shorten infants' length of stay in the hospital, it is important that researchers understand whether music therapy can improve intra-hospital transition from incubator to crib. Understanding whether music therapy improves transition from incubator to crib will allow for further exploration into the possibility of early weaning from the incubator. Medoff-Cooper's (1994) research supports the weaning of an infant from an incubator to an open crib in an accelerated timeframe, and Standley's (2003) research supports the incorporation of music therapy in the NICU to improve oxygen saturation levels, increase weight gain, and shorten hospital stays. The results of these studies reinforce the goals of this study, which was to combine music therapy with an accelerated transition from incubator to open crib.

Chapter 3: Methodology

INTRODUCTION

Chapter three presents the purpose, a brief description of the problem, an overview of the research design, and the methodology that includes the study sample, instrumentation, data collection procedures, and data analysis. The purpose of the study was to: 1) analyze the effects of auditory stimulation on preterm infant weight gain in incubators, and 2) analyze the effects of auditory stimulation on preterm infant time in incubators.

METHODOLOGY

Research Design

An experimental design with random assignment to three groups was used to examine the research hypotheses of this study. The study assessed how preterm infants in a hospital NICU were affected by playing recorded music therapist's singing voice or the infants' mothers' recorded singing voice during the infants' transition from an incubator to an open crib. Preterm infants in two experimental groups were prescribed either recorded music therapist's singing voice or their own recorded mothers' singing voice. Infants in the control group received no prescribed aural stimulation. Treatments were initiated when infants were placed in the incubator and delivered three times per week (Monday, Wednesday, and Friday), three times per day, from admission until infants' transition to an open crib. Music was played a maximum of one hour per day in short intervals of 20 minutes per session and started immediately after vital signs were taken (usually at 0800, 1200, and 1600 hours). The music playing schedule was based on

research suggesting that music should be played at critical periods such as beginning of sleep, quiet times, and immediately after stressful procedures (Standley, 2002). The study used guidelines for sound set forth by Passchier-Vermeer and Passchier (2000) and Panagiotidis and Lahav (2010). These guidelines recommend a safe sound level within the NICU of an hourly sound pressure level of 60 dB.

SAMPLE, SETTING, AND RECRUITMENT

This study included 90 preterm infants admitted to UTMB's Infant Special Care Unit (ISCU). According to the UTMB Division of Neonatology, the ISCU is a level III unit with 21 intensive care beds and 21 intermediate care beds that admits approximately 700 – 720 babies each year.

The sample consisted of preterm infants aged 25 – 36 weeks gestation. Infants were identified as meeting inclusion criteria by using UTMB's electronic system, EPIC. Infants' mothers and fathers were then approached either at bedside or in mothers' postpartum rooms.

A power analysis was conducted using the effect size from previous literature (Cassidy, 2009; Zimmerman et al., 2013). Based on this power analysis, the study required a sample size of 30 subjects per group to reach a power of 80% based on t-test with a 0.05 two-sided significance level and an effect size of 0.757.

Once Institutional Review Board (IRB) approval was obtained, two strategies were implemented to identify and recruit eligible subjects. First, the PI checked UTMB's EPIC electronic medical record daily to assess if any infants meeting the inclusion criteria had been admitted to the unit. Then, the UTMB ISCU Unit Clerk was contacted to determine if the subjects had successfully been admitted to ISCU. Following parental

consent, infants were enrolled. Infants' mothers who spoke English or Spanish and were randomly assigned to group II were asked to record their voices in the conference room at the ISCU or on the postpartum floor of the John Sealy Hospital. Those mothers who were patients in the TDCJ unit remained with their assigned security officer throughout the course of their consent and recording of their voice (if assigned to the mother's recorded singing voice group).

INCLUSION CRITERIA

Preterm infants who met the following criteria were included in the study: birth mother was English or Spanish speaking and a patient at UTMB's John Sealy Hospital or the TDCJ Unit. Infants were included if they were 25 – 36 weeks GA, deemed medically stable by the ISCU physicians, did not require assisted ventilation (CPAP/CiPAP allowable), and were placed in a closed incubator. If enrolled infants became medically unstable at any point (e.g., decrease in temperature, decrease in oxygen saturation levels) they were withdrawn from the study. When inclusion criteria were met, infants were placed in either the control group or one of the two experimental groups via random assignment.

EXCLUSION CRITERIA

Infants were excluded if they had congenital anomalies, central nervous system deficits, were medically unstable, required assisted mechanical ventilation, or were unable to be placed in an incubator.

DATA COLLECTION

Data collection started in October 2014 and ended in February 2015. Infants were randomly assigned to one of three groups using a random assignment calculator (Research Randomizer, 1997-2014) to determine group assignment.

Groups I and II had music interventions. The intervention for the Group I consisted of a commercial lullaby titled “Twinkle Twinkle Little Star” recorded by UTMB music therapist Elizabeth Moore. Lowey et al. (2013) suggested “Twinkle, Twinkle Little Star” as a “therapeutic lullaby” because it has a defined applied pitch range “higher” or “lower” for singing. Scientific studies have suggested that sounds and music should contain neither high nor low voices but suggestive of a range that is “familiar” and “recognized” (Volkova et al., 2006). A female voice was chosen because normally developing fetuses hear women’s voices in the womb and develop a preference for them (Standley, 2002). This music was played for 20 minutes, three times per day for three days a week (Monday, Wednesday and Friday) for infants enrolled in Groups I and II. Logic X Pro software from Apple Inc. (Cupertino, CA) was used to modify sound recordings and trim the noise peaks that are above 65 dBA (60dB). The MP3 file was then transferred onto an iPod Shuffle (Apple Inc., Cupertino, CA) that was attached directly to an iHome (Apple Inc., Cupertino, CA) speaker. Prior to placing the speaker into the incubator, the sound level was tested with an Extech Instruments, Low/High Range Sound Level Meter, Model 407732 sound meter to ensure that decibel levels remained below 60 dB. UTMB’s Bioengineering Department examined the equipment prior to placement in the incubator per hospital policy. The music in combination with environmental noise was not allowed to go over 60 dB for more than 5 seconds.

Music levels were monitored by a sound meter throughout the intervention to ensure that an unsafe decibel level was not encountered when combined with environmental noise. Had the sound level gone above 60 dB the speaker would have been turned off and that session would have been terminated by the PI. Nurses assigned to infants enrolled in the study worked with the PI as they did their routine infant assessments to ensure that vital signs remained stable.

The intervention for Group II consisted of a lullaby with the same title as the commercial lullaby “Twinkle, Twinkle Little Star,” but was made from a recording of their mothers singing. This music was played for 20 minutes for each infant three times per day for three days (Monday, Wednesday, and Friday). Music combined with environmental noise was not allowed to go over 60 dB for more than 5 seconds. These levels were monitored by a sound meter used throughout the intervention to ensure that unsafe decibel levels were not encountered.

In the control group, no music intervention was provided. Sounds heard in the incubator were not altered. Sound meters were placed in incubators and sound levels were recorded at intervals prescribed for the designated time for the experimental groups.

INSTRUMENTATION

An Infant Data Recording Form was used to record infant demographic data, behavioral state, and Apgar scores.

Infant Data Recording Form

Infant data were obtained from the electronic medical record (EMR) and recorded on the investigator developed Infant Data Recording Form (Appendix A). The following data were collected on the form: gestational age (GA), birth weight and length, APGAR

scores at 1 and 5 minutes, maximum sound levels during the intervention (inside and outside of the incubator), vital signs (heart rate, respiratory rate, and oxygen saturations), behavioral state scores and date of intervention. Plans were to collect data on SNAP scores however the NICU data collection site does not routinely conduct SNAP assessments and therefore it was not possible to collect these data from the EMR.

Process for Behavioral State Scoring

Common categorizations of behavioral states used by researchers include quiet sleep, active sleep, drowsiness, quiet alert, active alert, crying, and indeterminate state (Korner, 1972; Thoman, 1990; Wolff, 1966). While there are instruments designed to measure behavioral states (e.g., Anderson Behavioral State Scale, March of Dimes States of a Newborn), for the purpose of this study the infants were observed for behavioral state changes based on the definitions outlined by Holditch-Davis and Edwards (1997). Quiet awake was defined as “the infant’s eyes [being] open or opening and closing slowly. Motor activity is typically low and respirations are fairly even.” Very quiet sleep was defined as “the infant’s eyes were closed. Respirations were relatively regular and abdominal. A tonic level of motor tone was maintained, and motor activity was limited to occasional startles, sighs, or other brief discharges.” Drowsiness was defined as “eyes are ‘heavy-lidded,’ opening and closing slowly, or open but dazed in appearance. The level of motor activity is low and respiration fairly even.” Alert was defined as “eyes are open and scanning. Motor activity is typically low.” Sleep-Wake Transition was defined as “mixed waking and sleeping behaviors with generalized motor activity. Eyes are typically closed, but there may be rapid opening and closing.” Active Sleep was defined as “eyes are closed with intermittent rapid eye movement. Respiration is uneven and sporadic

movements may occur but muscle tone is low between movements” (Holditch-Davis & Edwards, 1997).

Process for Obtaining Apgar Score

Apgar scores were taken from the infants’ electronic medical record and were used to describe the infants’ health status at birth. Practitioners use the Apgar score worldwide to assess the clinical status of neonates during their transition to the external environment. The score measures 5 physical components that begin at 1 minute after birth and are repeated at 5 minutes post-delivery. Studies have found that low Apgar scores (≤ 3) at 1 or 5 minutes can indicate increased risks of infant death and poorer outcomes (Shankaran et al., 2004).

DATA ANALYSIS

The Statistical Social Sciences (SPSS Version 20) software was used for all data analyses. Descriptive statistics were used to determine data range, distribution, and normality of the sample. The sample characteristics were further analyzed using descriptive statistics including means and percentages.

To identify or rule out any differences among the groups in relation to non-study variables it was necessary to statistically control for pre-existing differences in the groups. Analysis of Variance (ANOVA) was performed to explore potential differences between groups on (e.g., gestational age) for extraneous variables across all dependent variables. An alpha of .05 was used.

Aim 1: Identify the influence of auditory stimuli (recorded music therapist’s singing voice and recorded mothers’ singing voices) on the weight of preterm infants.

H1.1: Preterm infants who receive a regimen of a recorded music therapist's singing voice (Group I) or recorded mothers' singing voice (Group II) will gain more weight than infants who do not receive a prescribed auditory stimuli (Group III).

Aim 1 Planned Analyses

To identify or rule out any significant subgroup differences that may have existed non-study variables, preliminary analyses were conducted. Analysis of Variance (ANOVA) was conducted to determine any differences (e.g., gestational age) in extraneous variables across all dependent variables. For significant differences identified, Analysis of Covariance (ANCOVA) was conducted to adjust for these differences and to determine impact of the confounding variables on the outcomes. The adjusted means of time of transition and weight gain estimated from the ANCOVA were reported. If the covariates across groups were well balanced through randomization, an ANOVA was conducted to assess the differences in outcomes. The interactions between groups and GA were evaluated in ANCOVA or ANOVA models. If a significant omnibus test was obtained between the three groups for weight and or length of time to transition, Scheffe's post hoc test was used to explore the differences among the means to show which means were significantly different from each other.

Aim 2: Determine the influence of auditory stimuli (recorded music therapist's singing voice and recorded mothers' singing voice) on transition time for preterm infants from incubator to crib.

H2.1: Preterm infants who receive a regimen of recorded music therapist's singing voice (Group I) or recorded mothers' singing voice (Group II) will transition

from the incubator to open crib faster than infants who do not receive a prescribed auditory stimuli (Group III).

Aim 2 Planned Analyses

The analysis for H2.1 was the same as the analysis described for H1.1.

PROTECTION OF HUMAN SUBJECTS

Permission to conduct the proposed study was obtained from the University of Texas Medical Branch's IRB. Potential risks associated with participation in this study were minimal. Risk of physical or psychological harm was considered to be minimal. All efforts were made to ensure that infant safety was a priority. All personal data were made anonymous and stored in a password-protected database accessible on a password-secured laptop. Involvement and characteristics of this study did not involve any medical procedure. The parents or guardians of the participants were informed of the voluntary nature of the study and withdrawal was possible at any time by simply informing the PI. Involvement did not place participants at risk of civil or criminal liability or negatively affect participants' financial status, employability, personal confidentiality, or personal reputation due to the completely anonymous nature of participation. Prior to enrolling in the study, parents or guardians were asked to sign an informed consent form so that the infants could participate. Individual participants were identified in the nursery by index cards that were blue (recorded music), pink (mothers' voice), or green (control). No participant was excluded based on gender, age, or ethnicity.

Chapter 4: Results

INTRODUCTION

Chapter four presents the results of the study including the characteristics of the sample, analysis, and results for each research question. The purpose of the study was to 1) analyze the effects of auditory stimulation on the weight gained by preterm infants in incubators, and 2) analyze the effects of auditory stimulation on the time a preterm infant spends in an incubator. The aims of the study were to 1) identify the influence of auditory stimuli (recorded music therapist's singing voice and recorded mother's singing voice) on the weight of preterm infants, and 2) determine the influence of auditory stimuli (recorded music therapist's singing voice and recorded mother's singing voice) on the transition time for preterm infants from incubator to crib.

PRETERM INFANT CHARACTERISTICS

Demographic characteristics, sound levels, physiological parameters, and state behaviors were measured for the preterm infants. These measures are reported first for the overall sample and then in a separate section for the three groups (Recorded Music Therapist's Singing Voice (MT), Mothers' Recorded Singing Voice (MV), and Control).

Overall Sample

The overall sample consisted of 90 preterm infants assigned to the three study groups. All subjects initially enrolled in the study remained enrolled throughout the study period. Infants' gestational age ranged from 25 – 36 weeks with a mean of 33.42 weeks (SD = 2.77). The mean birth weight of the infants was 1,959.30 grams (SD = 567.05;

range from 580-3050) and the birth length was 43.20 centimeters (SD = 4.59). In addition, a majority of infants was male and Hispanic/Latino (Table 4.1). Infants' Apgar scores at 1 minute ranged from 1 – 9 with a mean of 7.29 minutes; at five minutes Apgar scores ranged from 4 – 9 with a mean of 8.48 minutes (SD = 0.84).

Sound levels for the overall group were measured outside the incubators and inside incubators during the intervention. Measures were taken at each session for each of the three groups. For the purpose of this study range, mean, and standard deviations for the sound levels inside and outside the incubator are reported for the first day following the initial three intervention sessions (Time 1, Time 2, and Time 3; Table 4.2) and for the last day of the study before the infants were transitioned to the crib (Time 1, Time 2, and Time 3; Table 4.2).

Physiological measures (e.g., heart rate, respiratory rate, oxygen saturation levels) were recorded throughout each session. Again, for the purpose of this study the physiological measures (range, mean, and standard deviation) are reported on the initial day of the study following first intervention sessions and on the last day of the study before infants were transitioned to their cribs (Table 4.3).

The behavioral state measures, recorded as absent or present in a given state, were recorded throughout each session each day. For the purpose of this study the behavioral-states (frequency and percentages) are reported on the initial day of the study following the first intervention session at the three time points and on the last day of the study before the infants were transitioned to the open crib. Observations were made for the six behavioral states. However, for this sample three states were more commonly observed (Table 4.4).

Table 4.1: Demographic Characteristics of Overall Sample (n = 90)

Variables	N	%
Gender		
Male	50	55.6
Female	40	44.4
Race/Ethnicity		
African American	18	20
Caucasian	24	26.7
Hispanic/Latino	47	52.2
Asian	1	1.1

Table 4.2: Mean and SD of Sound Levels (dB) Inside and Outside of Infants' Incubators on First and Last Day of Intervention by Time of Session Each Day for Overall Sample

Variable	Inside				Outside			
	First Day		Last Day		First Day		Last Day	
	M	SD	M	SD	M	SD	M	SD
	Range		Range		Range		Range	
Time 1	55.61	5.77	56.64	5.47	72.93	7.10	78.83	8.61
	44.7-71.5		43.8		57.9-93.8		55.2-100.5	
Time 2	57.3	5.47	55.132	6.72	78.435	7.48	79.40	7.62
	43.8-71.1		42.9-68.2		65.3-94.2		64.0-100.5	
Time 3	55.596	6.76	54.643	5.23	79.22	8.45	78.35	8.50
	23.9-68.2		43.1-65.4		61.5-98.8		61.5-98.8	

Table 4.3: Mean and SD of Physiological Measures of Infants on the First Day Following Initial Intervention and Last Day Following Last Intervention by Time of Session for Overall Sample

Variable	Heart Rate				Respiratory Rate				Oxygen Saturation			
	First Day		Last Day		First Day		Last Day		First Day		Last Day	
	M	SD	M	SD	M	SD	M	SD	M	SD	M	SD
	Range		Range		Range		Range		Range		Range	
Time 1	146	18.52	149	15.82	46	16.14	48	17.21	97	2.26	97	2.40
	101-200		119-197		20-84		23-95		89-100		88-100	
Time 2	146	19.28	150	14.81	47	15.76	49	15.10	96	2.49	97	2.59
	104-197		117-190		24-88		19-81		88-100		87-100	
Time 3	146	17.05	149	16.1	45	15.35	48	15.35	97	2.39	98	2.20
	112-190		106-202		25-78		25-78		90-100		90-100	

Table 4.4: Number and Percentage of Infants for Behavioral State by Intervention Session, for First and Last Day of Study, for Overall Sample

Variable	First Day			Last Day		
	Time 1	Time 2	Time 3	Time 1	Time 2	Time 3
Very Quiet Sleep	n=70	n=70	n=76	n=72	n=67	n=68
	96.1%	92.1%	98.7%	93.5%	4.4%	98.6%
Quiet Awake	n=4	n=4	n=0	n=5	n=3	n=1
	62.6%	5.3%	0%	6.5%	4.2%	1.4%
Crying	n=1	n=2	n=1	n=0	n=1	n=0
	1.3%	2.6%	1.3%	0%	1.4%	0%

Intervention Group Characteristics

For the three groups, 27 (30%) were enrolled in the music therapy (MT) group, 31 (34%) were in mothers' voice (MV) group, and 32 (36%) were in the control group.

Table 4.5 illustrates the demographic characteristics of the infants by group on admission and on weight at enrollment in the study.

Sound levels were measured outside and inside incubators for each study group (i.e. music therapy, mothers' voice, control group). For the purpose of this study sound level inside and outside the incubator is reported for the first day following the initial three intervention times (Time 1, Time 2, and Time 3) and on the final day of the study before infants were transitioned to the open cribs (Table 4.6).

Physiological measures (e.g., heart rate, respiratory rate, oxygen saturation levels) were recorded throughout each intervention session (Time 1, Time 2, and Time 3) for

each study group. For the purpose of this study the physiological measures are reported on the initial day of the study following the first three intervention sessions and on the last day of the study before infants were transitioned to the open crib (Table 4.7).

The behavioral state measures, recorded as absent or present in a given state, were recorded throughout each session each day for each study group. For the purpose of this study the behavioral states (frequency and percentages) are reported on the initial day of the study following the first intervention sessions at the three time points and on the last day of the study before the infant was transitioned to the open crib (Table 4.8)

PRELIMINARY ANALYSES

Preliminary analyses were conducted on the groups to test the distribution and normality of the sample. Findings indicate that the distribution for each group on the dependent variables (weight gain and time spent in incubators) was skewed positively (i.e., tail of the distribution was to the right). Figures 4.1 – 4.6 display the histogram distribution by group. In addition the normality of the sample was examined using a Shapiro-Wilks Test and determined to be statistically significant further indicating a non-normal distribution (Table 4.9).

Table 4.5: Means and Standard Deviations of Preterm Infants Sample Characteristics by Group

Variables	Music Therapy		Mothers' Voice		Control Group	
	Mean Range	SD	Mean Range	SD	Mean Range	SD
Age (Weeks)						
At Birth	33 26-36	3.02	33.4 27-36	2.61	33.9 26-36	2.73
Weight (grams)						
Gestational	1861 890-2950	563.5	1924 580-2980	568.8	2082 700-3050	564.7
Enrollment	1836 825-2927	546	1912 750-2890	503.0	2061 700-2895	547.8
Length (cm)						
Birth	42.77 34-49	4.20	42.74 30-49	5.09	44.07 31.5-51.5	4.45
Apgar Score (1 min)	7.18 2-9	1.96	7.30 2-9	1.98	7.39 1-9	1.69
Apgar Score (5 min)	8.36 7-9	0.78	8.5 6-9	0.78	8.58 4-9	0.99
Time in Incubator (in days)	14.68 1-62	16.25	11.90 1-59	11.90	10.61 1-87	15.70

Table 4.6: Mean and SD of Sound Levels (dB) Inside and Outside of Infants' Incubators on First and Last Day of Intervention by Group and Time of Session

Variable	Inside Incubator				Outside Incubator			
	First Day		Last Day		First Day		Last Day	
	M	SD	M	SD	M	SD	M	SD
	Range		Range		Range		Range	
Music Therapy								
(n=17)								
Time 1	57.12	5.34	58.83	5.43	76.87	6.12	77.92	9.51
	44.8-64.2		50-70		64.3-92.4		65.6-100.5	
Time 2	61.72	8.96	56.94	5.66	75.84	7.82	75.67	7.27
	53.6-85		46.3-64.5		66.6-93.4		64-87.3	
Time 3	59.29	6.13	56.15	4.67	77.75	6.31	75.13	7.98
	47.6-71		47.2-62.8		69.4-93.8		63.2-90.2	
Mother's Voice								
(n=15)								
Time 1	53.91	3.85	57.13	4.99	77.31	9.15	78.19	7.76
	45.9-60.0		49.5-65.6		57.9-93.8		65.5-91.7	
Time 2	56.22	5.02	54.77	9.67	77.42	7.58	79.97	7.57
	48-65.7		43.9-63.1		64.3-90.6		68.6-96.4	
Time 3	54.28	4.88	55.19	4.36	75.15	7.39	77.37	9.48
	48-61.8		50.8-65.4		63.4-91.4		64.1-92.2	
Control Group (n=17)								
Time 1	54.65	8.23	53.46	5.00	82.61	5.60	82.59	8.40
	44.7-71.5		43.8-61		74.6-91.4		66.5-93.2	
Time 2	53.45	7.41	51.51	4.83	80.85	7.50	80.91	6.41
	43.5-72.9		42.3-58.3		65.3-95.3		68.6-90	
Time 3	52.31	5.94	50.39	4.52	80.32	9.51	80.25	7.14
	43.8-66		43.1-57.7		67.7-91.4		66.0-90.2	

Table 4.7: Mean and SD of Physiological Measures of Infants on the First Day Following the Initial Intervention and Last Day Following Last Intervention by Group and Time of Session

Variable	Heart Rate				Respiratory Rate				Oxygen Saturation			
	First Day		Last Day		First Day		Last Day		First Day		Last Day	
	M	SD	M	SD	M	SD	M	SD	M	SD	M	SD
	Range		Range		Range		Range		Range		Range	
Music Therapy												
Time 1	153 121-181	18.1	157 128-178	14.2	53.6 28-84	17.4	51.5 34-79	17.7	97.9 94-100	1.98	98.2 92-100	2.24
Time 2	155 134-185	16.7	159 143-180	9.97	42.3 22-71	15.4	50.7 27-72	16.2	96.8 90-100	3.18	97.3 87-100	3.69
Time 3	142 123-190	18.3	155 126-183	16.2	44.5 25-70	12.9	44.1 27-81	14.1	96.8 90-100	3.13	97.7 95-100	2.17
Mothers' Voice												
Time 1	146 109-166	13.6	153 119-182	14.1	46.9 20-80	17.9	46.2 23-95	21.9	97.5 93-100	2.53	98.5 96-100	1.68
Time 2	149 115-183	18.3	153 138-178	10.6	51.3 38-74	11	45.8 30-70	11.0	92.5 94-100	22.9	98.5 95-100	1.64
Time 3	152 135-190	14.9	150 128-202	18.3	51 24-88	21.4	47 28-67	13.2	96.9 93-100	1.83	98.9 92-100	2.03
Control Group												
Time 1	140 107-164	13.6	156 133-197	14.9	42.2 24-70	13.1	46.7 28-78	15.7	97 89-100	2.78	97.8 88-100	3.02
Time 2	142 104-170	16.8	153 131-190	15.7	46.6 24-85	14.0	44.3 25-70	15.5	97.5 88-100	2.92	97.4 90-100	2.78
Time 3	143 113-163	14.5	154 128-191	14.6	43.8 24-85	13.3	49 19-77	18.1	97.9 92-100	2.54	97.9 90-100	2.70

Table 4.8: Number and Percentage of Infants for Behavioral State by Intervention Session, for First and Last Day of Study, for Group

Variable	First Day						Last Day					
	Very Quiet Sleep		Quiet Awake		Crying		Very Quiet Sleep		Quiet Awake		Crying	
	N	%	N	%	N	%	N	%	N	%	N	%
Music Therapy												
	22	95.7%										
Time 1	23	95.8%	1	4.3%	0	0%	25	100%	0	0%	0	0%
Time 2	24	96.0%	1	4.2%	0	0%	23	95.8%	23	95.8%	0	0%
Time 3			0	0%	1	4.0%	22	95.7%	1	4.3%	0	0%
Mother's Voice												
Time 1	25	92.6%	1	3.7%	1	3.7%	24	88.9%	3	11.1%	0	0%
Time 2	26	92.9%	2	7.1 %	0	0%	24	100%	26	92.9%	0	0%
Time 3	27	100%	1	4.2%	0	0%	24	100%	0	0%	0	0%
Control												
Time 1	27	100%	0	0%	0	0%	23	92.0%	2	8.7%	0	0%
Time 2	21	87.5%	0	0%	2	8.3%	20	87.0%	21	87.5%	1	4.3%
Time 3	25	100%	0	0%	0	0%	22	100%	0	0 %	0	0%

Figure 4.1: Histogram for Distribution of Total Weight Gained by Music Therapy Group

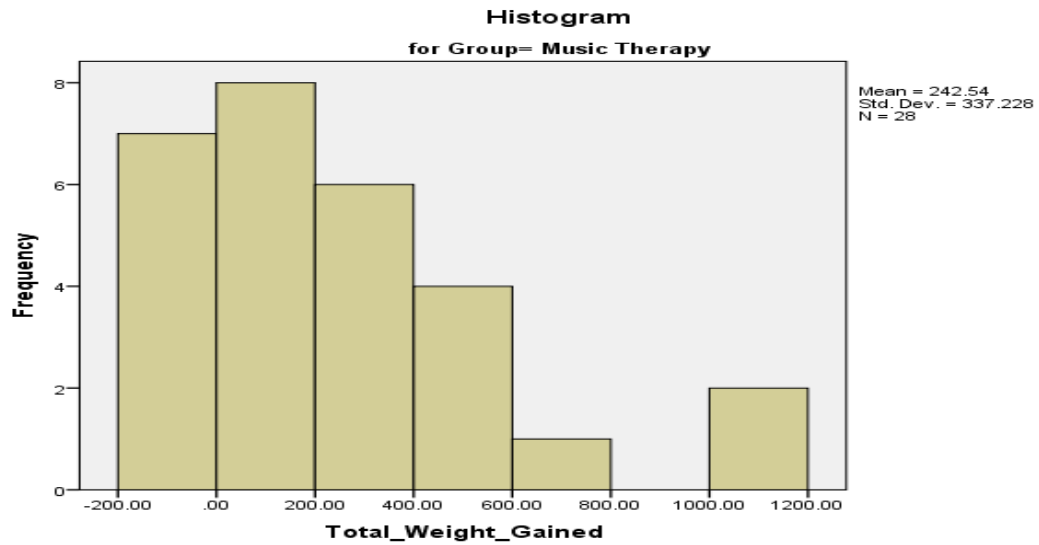


Figure 4.2: Histogram for Distribution of Total Weight Gained by Mother's Voice Group

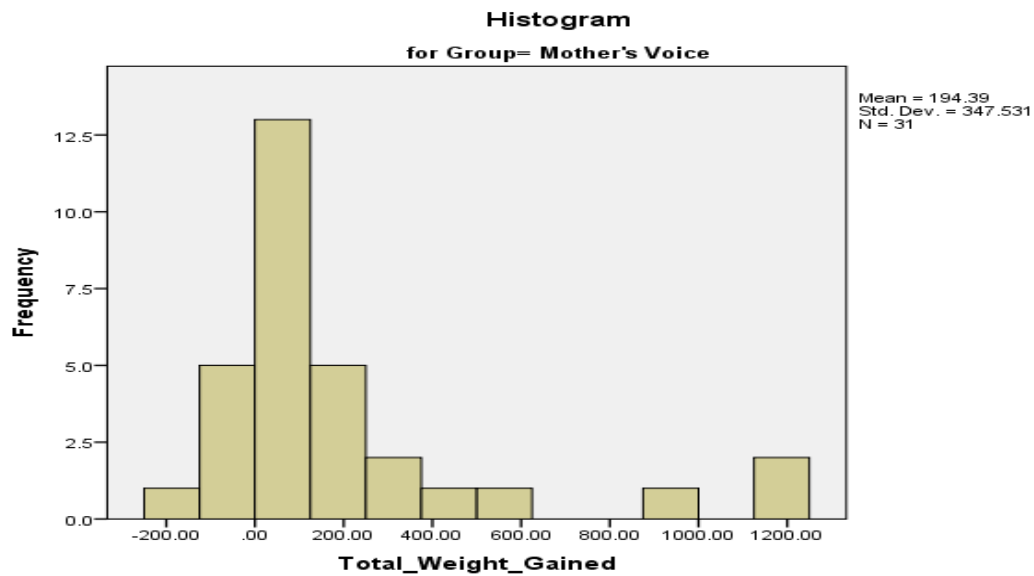


Figure 4.3: Histogram for Distribution of Total Weight Gained by Control Group

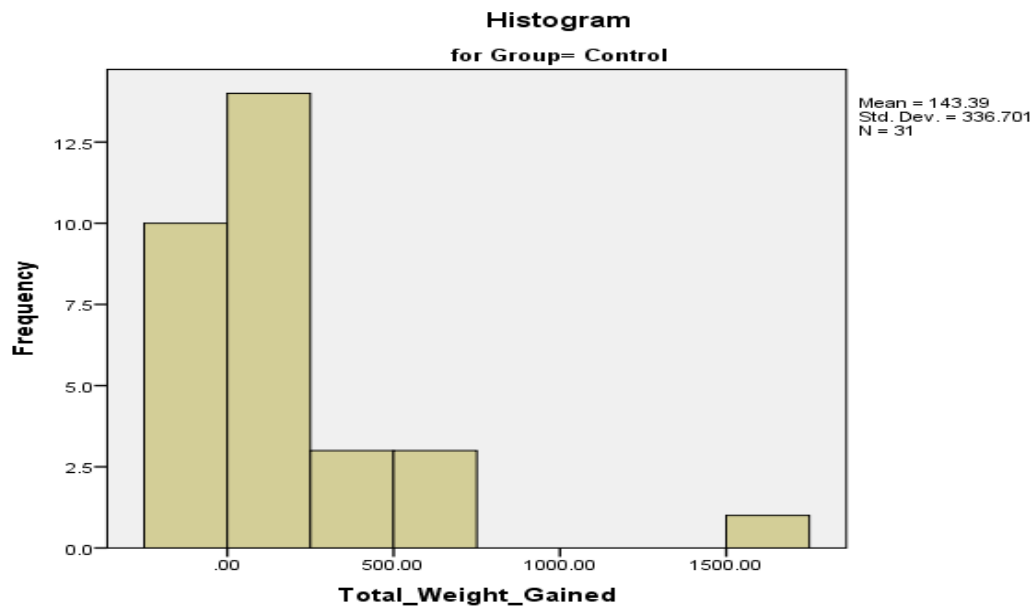


Figure 4.4: Histogram for Distribution of Time Spent in Incubator by Music Therapy Group

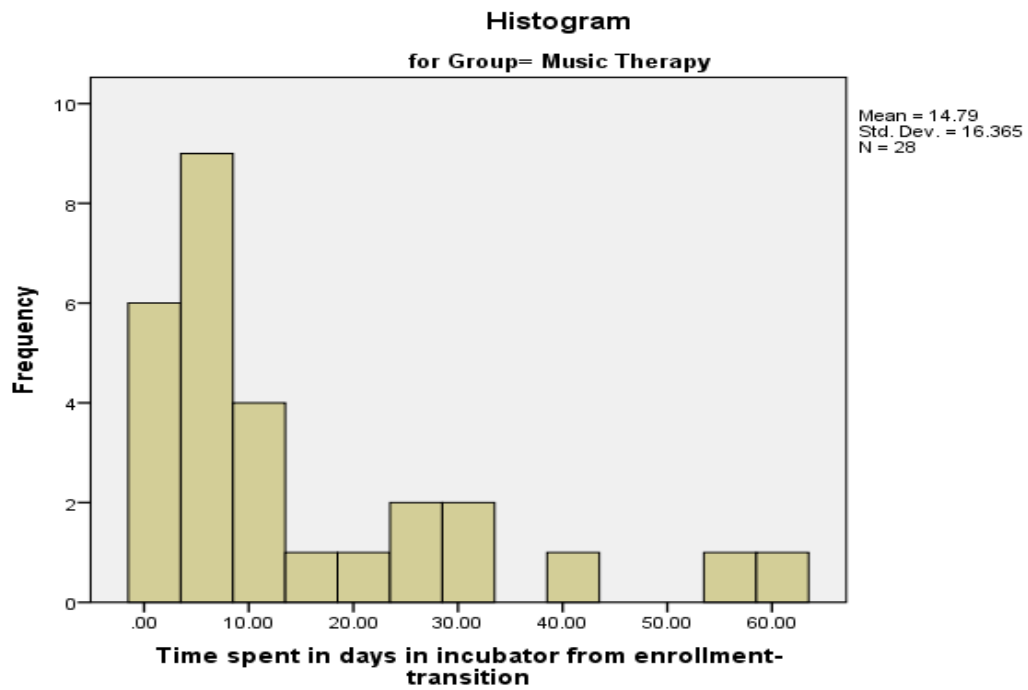


Figure 4.5: Histogram for Distribution of Time Spent in Incubator by Mother's Voice Group

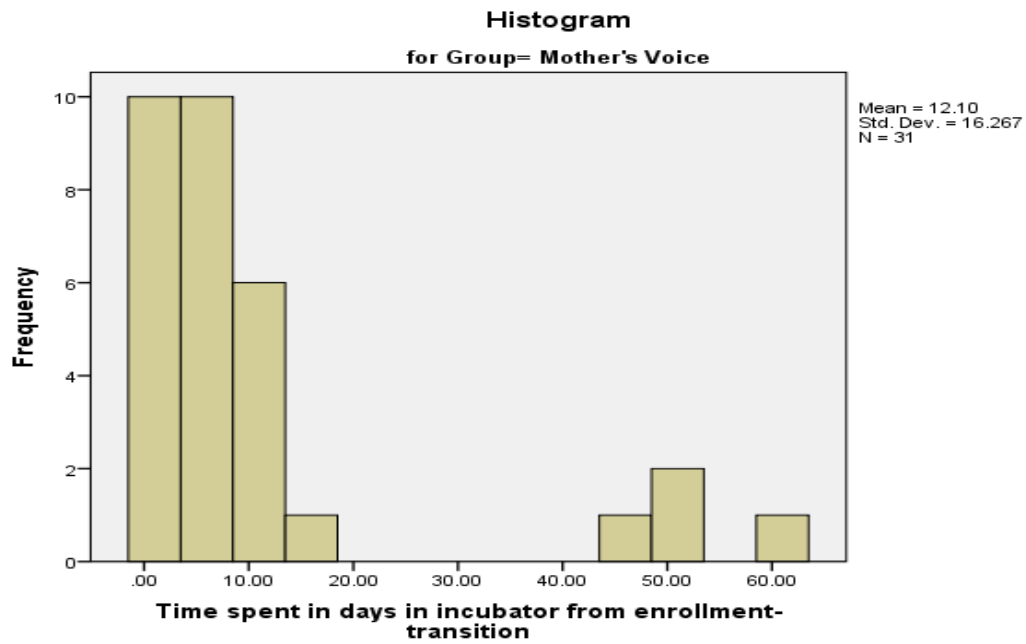


Figure 4.6: Histogram for Distribution of Time Spent in Incubator by Mother's Voice Group

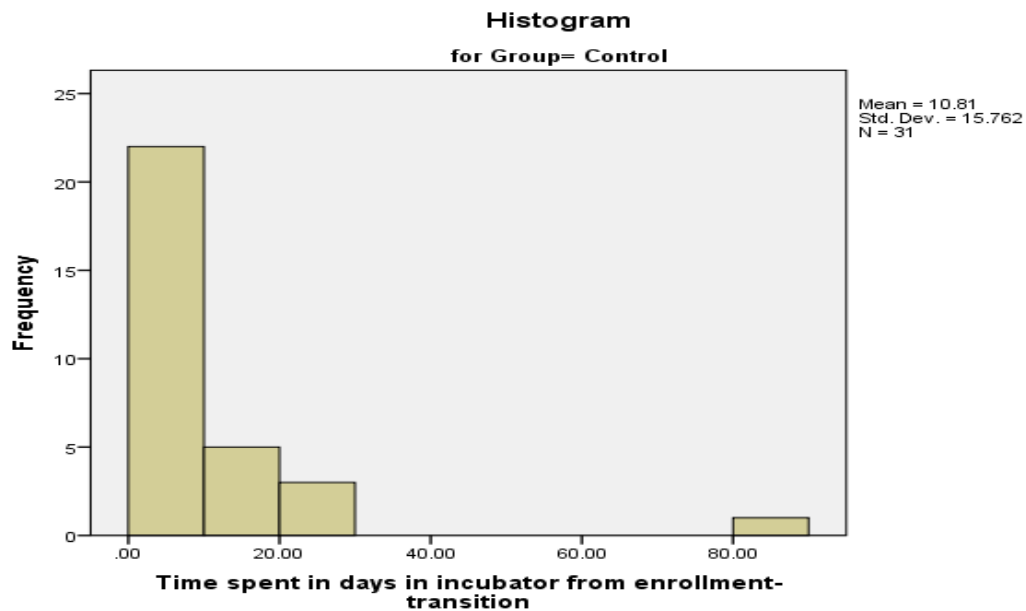


Table 4.9: Shapiro-Wilks Test for Normality of the Three Groups

Variable and Group	Statistic	df	P
Total Weight Gained			
Music Therapy	0.832	28	0.000
Mother's Voice	0.733	31	0.000
Control	0.663	31	0.000
Time Spent in Incubator			
Music Therapy	0.780	28	0.000
Mother's Voice	0.618	31	0.000
Control	0.541	31	0.000

Additional preliminary analyses were conducted to identify the presence of group differences in non-study variables and to statistically control for pre-existing differences if needed. An Analysis of Variance (ANOVA) was performed to determine whether group differences existed on the non-study variable of gestational age. The findings indicate that the mean gestational age was 33 weeks for the MT group, 33.36 for the MV, and 33.87 for control. The Analysis of Variance showed no difference between the groups on gestational age ($F = .734, 2/87, p = .48$), therefore the planned analysis of ANOVA was used to analyze the data for the two main hypotheses. Upon further exploration of the non-study variables, including sound (Table 4.10) and physiological measures (Table 4.11), an ANOVA was conducted and showed significant differences only on sound levels inside the incubator at time 3 on day 1 of the intervention. The post

hoc Scheffe's Test showed a significant difference between the control group and the recorded music therapist's singing group (mean difference=-4.09, $p=0.02$). Although non-normality was noted at time 3 on day 1 inside the incubator on sound level (dB), it can be explored in future analyses since this finding is outside the scope of this dissertation and will not be analyzed with the study hypotheses.

The groups' tests for homogeneity of variance on the time in incubator from enrollment to transition (Levene Statistic = .785, $df = 2/87$, $p = .459$), and total weight gained (Levene Statistic = .433, $df = 2/87$, $p = .650$) showed no significant differences and therefore "equal variance assumed" was used to evaluate these dependent variables in hypotheses 1.1 and 2.1.

Table 4.10: One-Way Analysis of Variance for Sound Levels (dB) Inside and Outside Infants' Incubators on First and Last Day of Intervention

Variable	Inside								Outside							
	First Day				Last Day				First Day				Last Day			
	df	SS	M SQ	F	df	SS	M SQ	F	df	SS	MSQ	F	df	SS	MSQ	F
Time 1																
Between Groups	2	58.9	29.45	.88	2	371.95	185.97	7.52	2	109.04	54.52	1.09	2	161.96	80.98	1.10
Within Groups	67	2235.6	33.37		60	1483.09	24.72		67	3366.57	20.52		61	4504.50	73.84	
Total	69	2294.5			62	1855.04			69	3475.60			63	4666.46		
Time 2																
Between Groups	2	272.3	136.13	2.77	2	339.11	169.56	4.13	2	177.20	88.60	1.61	2	192.46	96.23	1.70
Within Group	71	3494.9	49.22		62	2547.13	41.08		70	3847.76	54.97		63	3577.15	56.78	
Total	73	3767.1			64	2668.24			72	4024.67			65	3769.61		
Time 3																
Between Groups	2	235.1	117.55	4.25*	2	309.08	154.54	6.57	2	27.59	13.79	.189	2	76.87	38.43	.525
Within Group	78	2159.0	27.68		66	1553.71	23.54		77	5607.75	72.83		68	4978.36	73.21	
Total	80	2394.1			68	1862.79			79	5635.33			70	5055.23		

*P=0.018

Table 4.11: One-Way Analysis of Physiological Characteristics on First and Last Day of Intervention

Variable	HR								RR								O2							
	First Day				Last Day				First Day				Last Day				First Day				Last Day			
	df	SS	MSQ	F	df	SS	MSQ	F	df	SS	MSQ	F	df	SS	MSQ	F	df	SS	MSQ	F	df	SS	MSQ	F
Time 1																								
Between Groups	2	1314	657	2	2	50	25	1	2	955	477	1.8	2	1674	823	2.1	2	16	8	1.6	2	8	4	0.8
Within Groups	72	24072	334		75	19218	256		74	19971	296		75	29844	397		72	362	5		74	429	5	
Total	74	25686			77	19262			76	20926			77	31491			74	378			76	438		
Time 2																								
Between Groups	2	167	83	0.2	2	21	10	0.5	2	62	31	1	2	416	208	0.6	2	28	14	2.4	2	33	16	2.6
Within Group	73	27715	379		69	15556	225		73	23409	320		68	24804	364		72	431	5		68	437	6	
Total	75	27882			71	15577			75	23472			70	25220			74	459			70	470		
Time 3																								
Between Groups	2	1114	557	2	2	296	148	0.6	2	9	4	.01	2	689	344	0.9	2	4	2	0.3	2	24	12	2.6
Within Group	75	21261	283		66	17324	262		74	26583	359		66	25856	391		73	422	5		66	304	4	
Total	77	22376			68	17621			76	26593			68	26545			75	426			68	329		

STUDY QUESTIONS

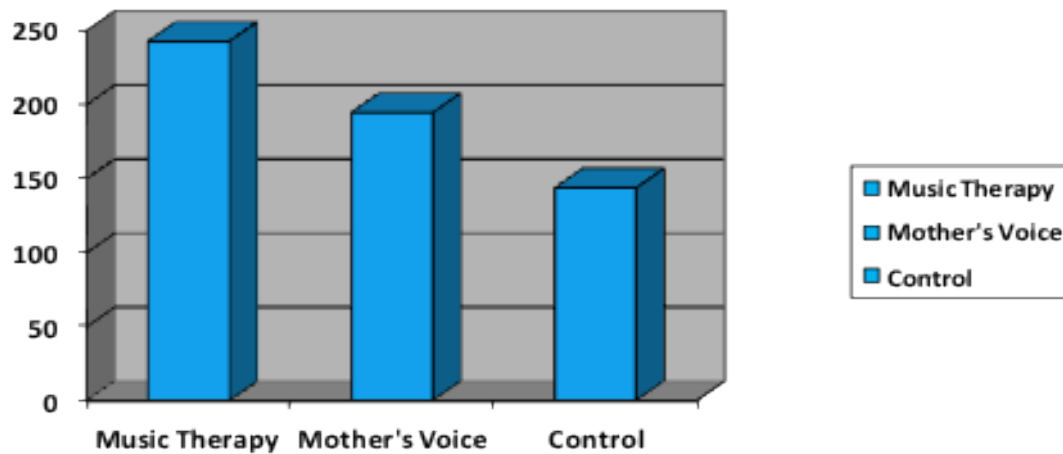
Aim 1: Identify the influence of auditory stimuli (recorded music therapist's singing voice and mothers' singing voices) on weight of preterm infants.

H1.1: Preterm infants who receive a regimen of a recorded music therapist's singing voice (Group I) or recorded mothers' singing voice (Group II) will gain more weight than infants who do not receive a prescribed auditory stimuli (Group III).

This hypothesis was analyzed using a one-way ANOVA comparing groups total weight gain from time in incubator to transition to open crib. Results of the ANOVA showed no statistical significant differences ($F = .625$, $df = 2/87$, $p > .05$) between the groups on the amount of weight gained. The planned Scheffe's post hoc test analysis was conducted and also failed to provide statistically significance differences ($p > .05$). Because the groups showed a non-normal distribution a Kruskal Wallis H was performed. Again, no significant difference was revealed among the groups and therefore these statistics are not reported here. Although no statistical differences were found, the infants in the MT and MV groups had higher mean weights at the time of transition to the crib than infants in the control group (Figure 4.7).

Aim 2: Determine the influence of auditory stimuli (a recorded music therapist's singing voice and recorded mothers' singing voice) on transition time for preterm infants from incubator to crib.

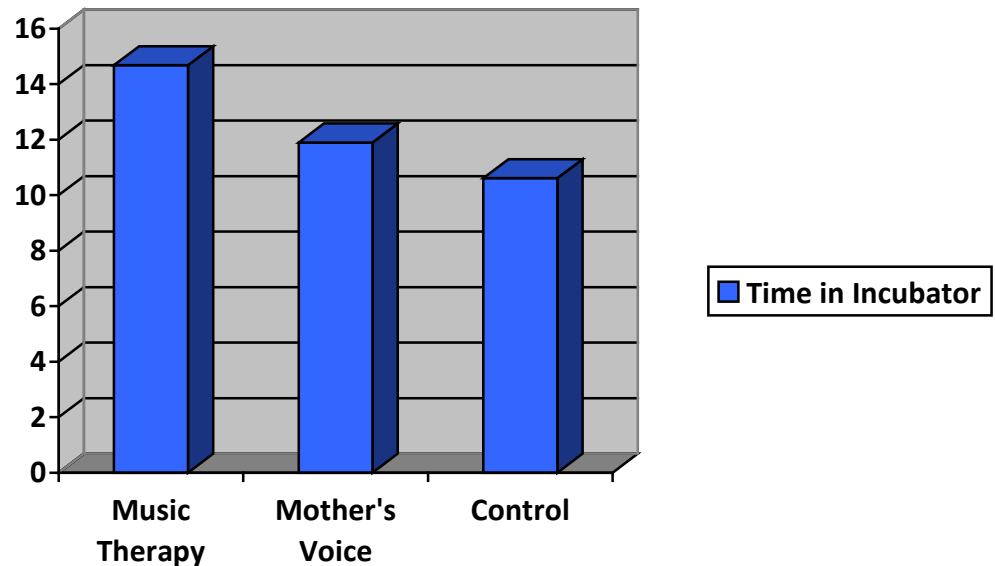
Figure 4.7: Bar Graph for Total Weight Gained by Group



H2.1: Preterm infants who receive a regimen of a recorded music therapist's singing voice (Group I) or recorded mothers' singing voice (Group II) will transition from the incubator to open crib faster than infants who do not receive a prescribed auditory stimuli (Group III).

Hypothesis two was analyzed using one-way ANOVA to determine any differences in length of time needed to transition to the open crib. Results of the ANOVA indicated no statistical significant differences between the groups on the length of time to transition from incubator to crib ($F = .462$, $df = 2/87$, $p > .05$; Figure 4.8). The planned Scheffe's post hoc test analysis was conducted and also failed to provide statistically significance differences ($p > .05$). Because the groups showed a non-normal distribution a Kruskal Wallis H was performed. Again, no significant differences were revealed among the groups and therefore statistics are not reported here.

Figure 4.8: Bar Graph for Total Time Spent in Incubator by Group



SUMMARY

The overall sample consisted of 90 preterm infants with a mean gestational age of 33.4 weeks, mean birth weight of 1,959.3 grams, and mean birth length of 43.2 cm. The majority of the sample was Hispanic (52.2%) and male (55.6%). The one-way ANOVA used to analyze the differences between groups and weight gained showed no significant difference among the groups. The one-way ANOVA used to analyze the differences between the groups and the time the infant spent in an incubator also yielded no significant differences between the groups. However, although not significant the mean weight gain was higher for recorded music therapist's singing voice than for the other groups.

Chapter 5: Discussion, Recommendations and Conclusions

INTRODUCTION

Chapter 5 provides a summary of the study and further clarifies the findings. The strengths and weaknesses of the study are discussed as well as nursing implications. Recommendations for future research on music therapy in neonatal intensive care units are presented.

SUMMARY OF THE STUDY

Music therapy has frequently been cited as an effective intervention when attempting to aid in the weight gain of preterm infants. Research in NICU practice supports the use of music in critical areas such as sucking, weight gain, sleep, and recovery from painful procedures (Lowey, 2013). Parents' voices have also been shown to enhance vocalization in premature infants (Krueger, 2012). In addition, the newborn, up to 8 months in age, can discriminate and show preference for the mother's voice compared with a female stranger's voice (Standley, 1990). While it is clear that mothers' voices and lullabies can affect preterm infants' health outcomes (Krueger et al., 2010), it is unclear whether auditory stimuli impact these outcomes such that infants' intra-nursery transition become affected or whether one type of stimulus is more effective than another.

Purpose Statement and Research Hypotheses

The purpose of this study was to understand how auditory stimuli (recorded music therapist's singing voice and recorded infants' mothers' singing voice) can affect the transition of preterm infants from incubator to an open crib.

The specific aims and related hypotheses of this study were:

Aim 1: Identify the influence of auditory stimuli (recorded music therapist's singing voice and recorded mothers' singing voice) on the weight of preterm infants.

H1.1: Preterm infants who receive a regimen of recorded music therapist's singing voice (Group I) or recorded mothers' singing voice (Group II) will gain more weight than infants who do not have music played to them (Group III).

Aim 2: Determine the influence of auditory stimuli (recorded music therapist's singing voice and recorded mothers' singing voice) on transition time for preterm infants from incubator to crib.

H2.1: Preterm infants who receive a regimen of recorded lullabies (Group I) or recorded mothers' singing voice (Group II) will transition from the incubator to crib faster than infants who do not have music played to them (Group III).

Review of the Methodology

An experimental design with random assignment to three groups was used to examine the research hypotheses of this study. The study assessed how preterm infants in a hospital NICU were affected by playing pre-recorded lullabies (recorded music therapist's singing voice and infants' mothers' singing voice) during infants' transition from incubator to open crib. Preterm infants in two experimental groups were prescribed either recorded music therapist's singing voice or recorded mothers' voices. Infants in the control group received no prescribed aural stimulation. Treatments were initiated when infants were placed in the incubator and delivered three times per week (Monday, Wednesday, and Friday), three times per day, from admission until infants transitioned to open cribs. Music was played a maximum of one hour per day in short intervals of 20

minutes per session and started immediately after vital signs were taken (usually 0800, 1200, and 1600 hours). The music playing schedule was based on research suggesting that music should be played at critical periods such as beginning of sleep, quiet times, and immediately after stressful procedures (Standley, 2002). This study used guidelines for sound set forth by Passchier-Vermeer and Passchier (2000) and Panagiotidis and Lahav (2010). These guidelines recommend a safe sound level within the NICU of an hourly sound pressure level at or below 60 dB.

An Infant Data Recording Form was used to collect data on non-study and study variables. Data were recorded on the preterm infants' weight, physiological measures, and APGAR scores. In addition, the study variables, time to transition from incubator to crib and weights at time enrollment and transition to crib were also recorded on this form.

FINDINGS RELATED TO THE LITERATURE

The overall sample consisted of 90 total preterm infants. The sample was largely Hispanic (52.2%), male (54.6%), and at 34 weeks gestation (23.3%). The mean birth weight of the infants was 1,959.30 grams, mean length was 43.2 cm, and mean Apgar scores at 1 minute and 5 minutes were 7 and 8 respectively. Participants' age ranged from 26 to 36 weeks gestation with a mean gestational age of 33.4 weeks (sd=2.78). Although the sample in this study was diverse, a majority Hispanic sample makes it different from the recent music studies in the literature (Dearn & Shoemark, 2014). In their study of 20 infants a majority of the infants were Caucasian. While race or ethnic background was not used as a study variable in the current study, what this study shows is that Hispanic infants can be recruited to participate in studies focused on music.

The physical environment encountered during the hospital stay directly impacts patient care. The American Academy of Pediatrics (AAP) suggests that the higher noise levels encountered in hospital NICUs may adversely affect growth and development of the preterm infant (AAP, 1974). The AAP recommends that noise levels remain below 45 dB (A) in hospital NICU's (AAP, 1997). Other guidelines recommend a safe sound level within the NICU of an hourly sound pressure level of 60 dB (Passchier-Vermeer 2000 & Passchier, 2010). In this current study, the sound levels inside the incubator remained below 60 dB except for one session with the music therapy group at time 2. When the sound inside the incubator went above 60 dB the music was lowered within 5 seconds. Sound levels, even if above the safe standard for 5 or less seconds, were recorded as the maximum sound levels encountered by the infants inside the incubator. The sound levels encountered outside of the incubators throughout the course of the study ranged from 55.2-100.5 dB, which were significantly above the guidelines and recommendations set forth by the American Academy of Pediatrics for hospital NICUs. The louder sound levels were not constant and appeared to occur during times when physicians and staff were conducting daily rounds, or when a new infant was admitted to the unit.

The extant literature suggests that there is a decrease in heart rate, respiratory rate, and an increase in oxygen saturation when musical interventions are implemented (Amini et al., 2013; Loewy et al., 2013). Since examining physiological parameters following musical interventions was not a focus of this study no attempts were made to determine whether the groups differed significantly on these parameters. However, it was noted that the mean oxygen saturation levels were higher for each group on the last day perhaps a factor of maturity and/or improving health.

Hypothesis 1.1: The effect of auditory stimuli vs. no stimuli on weight gain.

Although the infants in the MT and MV groups gained more weight than infants in the control group, no statistically significant difference was shown. One reason for this may be that researchers in other studies followed their infants over shorter periods of time and the long term effects of the intervention were not tested. That is, the average length of time between incubator and crib in the current study was 12 days whereas most studies in the literature follow their infants for 1 – 5 days (Cevasco, 2008; Dearn & Shoemark, 2014; Lowey et al., 2013; Standley, 2002). This is thought to be a possibility because there are no studies in the literature that examine intra-nursery outcomes that study the effect of music from incubator to crib. Seeing that infants could have potentially become sensitized to the music since it was played for a prolonged period of time, perhaps a secondary analysis of the current weight data from this study examining short-term gain and long-term gains may provide a better understanding of preterm infant growth following a music intervention.

Hypothesis 2.1: The effect of auditory stimuli vs. no stimuli on length time transition time from incubator to crib.

There were no significant differences among the MT, MV, and the control groups on the length of time needed to transition from incubator to open crib. Because there are no studies to date that examine intra-nursery transition time from incubator to crib, it is difficult to evaluate these findings within the context of the extant literature. One speculative reason may be that each infant who is born necessitates different supportive measures that may require a longer amount of time spent in an incubator. Another reason could be that hospital policies and protocols for weaning preterm infants' to open crib

dictate when infants should be moved from incubator these uniform guidelines could mask some of the effects of the interventions. The means of the control group on length of time to transfer to crib was shorter than those of the MT and MV groups. This finding is difficult to explain, especially since the weight gains for the MT and MV groups were greater than that of the control group. Perhaps other confounding variables may have affected the length of stay in the crib and should be controlled for in future studies.

DISCUSSION OF THE RATIONALE FOR THE USE OF MUSIC THERAPY IN THE NICU

A systematic review of 10 studies with a total enrollment of 780 participants revealed that music significantly benefits an infant by positively affecting observed behavioral state, decreasing heart rate, decreasing respiration rate, raising oxygen saturation level, increasing weight gain, decreasing days in hospital, and positively affecting feeding rate/non-nutritive sucking rate (Standley, 2001). The findings of this study, when reviewed within the context of the rationale for the use of music therapy in the NICU, do not support the previous outcomes of weight gain. The failure to show a change in weight gain is possibly related to not being able to show a difference in intra-nursery transition.

STRENGTHS AND WEAKNESSES

A major strength of this research study was the use of an experimental design that included the randomization of infants into experimental or control groups. Although results were not statistically significant, these methods show that it is possible to successfully collect data using music interventions for intra-nursery transitions in preterm infants.

The fact that the sample was over 50% Hispanic is both a strength and a weakness. It is a strength since it shows that a sample from an underrepresented group

can be recruited in research. It can be seen as a weakness because it could make the findings less generalizable to other infants.

IMPLICATIONS

Findings from this study showed no differences among the groups on recorded music therapist's singing, recorded mothers' singing voice and control. Although not significantly different among groups, the mean weight for the preterm infants in the MT group was higher than the mean weights for infants in the other groups. This trend suggests that a closer examination of this variable with music therapy may be warranted.

The findings from this study can provide information for future researchers regarding a necessary adaptation in methodology prior to attempting to further investigate auditory stimulation in preterm infants. The studies that have already been conducted with mothers' recorded singing voice and music therapist's recorded singing voice could improve the timing and delivery of the intervention used in this study. With the incorporation of a revised methodology, in combination with the methods utilized in a study such as this one, perhaps different outcomes on weight gained and time spent in an incubator by a preterm infant maybe realized.

RECOMMENDATIONS FOR FUTURE RESEARCH

The following recommendations are suggested for future studies. The first recommendation is to conduct the study using a larger sample size and to collect and analyze data as repeated measure across time. Increasing the sample size will decrease the likelihood of rejecting the null hypothesis of differences among the MT, MV, and control groups if the hypothesis is false. The benefit of analyzing the data across time will

show any potential differences that may occur at identified time periods. The second recommendation is to block on gestational age and race/ethnicity to ensure that equal numbers from different age and racial/ethnic groups are included in the study. Gestational age was used as a covariate in this study, by blocking on this variable prospectively will add a control to the study during data collection. Blocking on the variable race/ethnicity will ensure that equal numbers of infants from different racial groups are included in the design, which will support the generalizability of the findings.

STUDY LIMITATIONS

Limitations of this study include a small size, over representation of Hispanic infants, and not exploring changes in the dependent variables at predefined intervals across time. The small sample size opens the study up to the possibility of a Type I error, that is, rejecting the null hypothesis when it is true. The over representation of Hispanic infants in the sample may potentially limit the generalizability to other racial or ethnic groups. By exploring the dependent variables for potential changes may help to identify any changes in growth patterns across groups.

CONCLUSIONS

Although a music intervention did not increase the weight gained by preterm infants nor decrease the time they spent in an incubator the trend showing a greater increase in weight for the MT group suggests the need for further exploration into growth outcomes and auditory stimulation. Further, it may be concluded that is possible to recruit Hispanic infants to enroll in a study on music interventions.

Appendix A: Infant Data Recording Form

Participant ID: _____

SNAP Score: _____

Gestational Age: _____

Birth Weight: _____

Gender: ☐ Male ☐ Female

Length: _____

Current Weight:			Current behavioral state: QS AS D QA AA C	
Date:	Heart Rate	Respiratory Rate	Oxygen Saturation	Behavioral State
0800				Before: After:
1200				Before: After:
1600				Before: After:

Current Weight:			Current behavioral state: QS AS D QA AA C	
Date:	Heart Rate	Respiratory Rate	Oxygen Saturation	Behavioral State
0800				Before: After:
1200				Before: After:
1600				Before: After:

Current Weight:			Current behavioral state: QS AS D QA AA C	
Date:	Heart Rate	Respiratory Rate	Oxygen Saturation	Behavioral State
0800				Before: After:
1200				Before: After:
1600				Before: After:

Current Weight:			Current behavioral state: QS AS D QA AA C	
Date:	Heart Rate	Respiratory Rate	Oxygen Saturation	Behavioral State
0800				Before: After:
1200				Before: After:
1600				Before: After:

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