Selected Topics in Neonatal Care

Edited by R. Mauricio Barría

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Meet the editor

R. Mauricio Barría, MSc, DrPH, is a Principal Investigator and Assistant Professor at the Faculty of Medicine at Universidad Austral de Chile. He is currently Director of the Nursing Institute and Director of the Evidence-Based Health Office at the university. He was trained as an epidemiologist and received his MSc in Clinical Epidemiology from Universidad de la Frontera in Temuco, Chile, and his DrPH from Universidad de Chile in Santiago, Chile. His research interests lie in the areas of Maternal-Child Health, Neonatal Care and Environmental Health. He is skilled in epidemiological studies designs with special interest in cohort studies and clinical trials.

Contents

Preface

Neonatal care includes a wide range of aspects that are necessary to provide the support that assures the best outcome in the care of premature and sick newborn. The technoscientific advances in this area have led to an increase in these babies' survival, who are sometimes in need of sophisticated care. However, there are essential levels of care that all centers that look after high-risk newborns must include.

During neonatal care, the health team faces a variety of crucial problems in the evolution of neonates. This book includes important topics related to neonatal care grouped into four sections. In 14 chapters that address relevant issues about neonatal care, the book seeks to contribute to the clinical work of the health teams of neonatal units.

The first section of this book focuses on the critical issues in neonatal care presented in the first hours of life. Chapter 1 introduces global and fundamental aspects of neonatal care. Chapter 2 refers to different procedures, scopes and possibilities in the very first hour of life, "the golden hour," both short and long term, and analyzes psychological and physiological aspects during this period. Chapter 3 develops important elements regarding the assessment of the hemodynamic stability of neonates and analyzes different measurement methods and their benefits and limitations. Chapter 4 assesses the relevant actions of respiratory care, analyzing the transition from fetal to neonatal breathing and respiratory support in the delivery room to non-invasive and invasive respiratory support methods. Chapter 5 closes this section by showing the state of the art of the pathophysiology, diagnosis and therapy of neonatal hypoglycemia with emphasis on the pathogenesis of congenital hyperinsulinemic hypoglycemia.

Section II groups three selected topics on neonatal infection. Chapter 6 includes epidemiological, diagnostic, therapeutic and prognostic aspects of neonatal meningitis. Chapter 7 develops neonatal osteomyelitis, a problem that, although infrequent, can cause serious complications; it discusses the clinical presentation, general approach, diagnosis and therapeutic management. This section is completed with Chapter 8 on fungal infections in the neonatal intensive care unit, in which the epidemiology of this type of infection is described, and candida infection, as well as other neonatal fungal infections, is also discussed.

Section III includes selected topics on surgical problems. This section includes Chapter 9 that develops important aspects of neonatal care related to anesthesia. This topic includes general principles, elements of anesthetic preparation and intraoperative management of anesthesia. Chapter 10 describes the most frequent surgical problems, and Chapter 11 updates the topic of neonatal male circumcision in the context of Thailand from a public health perspective.

Finally, section IV includes different miscellaneous topics in the field of neonatal health. Chapter 12 focuses on neonatal gene therapy for inherited disorders and explains through

experimental models the usefulness and potential of this type of therapy, beginning from the analysis of two of these problems: metachromatic leukodystrophy and hypophosphatasia. Chapter 13 develops a review of the current knowledge regarding the effects of selected endocrine active compounds in the neonatal period. Chapter 14 concludes this section and the book with an interesting report on approaches carried out in Nigeria that could have a positive effect on early neonatal mortality reduction in that country.

As described, this book includes various topics of interest for all those health professionals who are dedicated to neonatal health care. It is necessary to thank each one of the authors who through their contributions, taken from both experience and context, have allowed the development of this interesting book, which it is hoped will be useful for professionals and students dedicated to neonatal care.

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Critical Issues in Neonatal Care

Introductory Chapter: Essential Issues in Neonatal Care

R. Mauricio Barría

Additional information is available at the end of the chapter

1. Introduction

 Mortality in infants and under 5-year olds has been a constant health concern. In 2015, of the total number of deaths in children under five worldwide, 45.1% corresponded to neonatal deaths, with the three leading causes of death being prematurity (15.9%), causes related to childbirth (10.7%), and sepsis or meningitis (6–8%) [1]. By the year 2030, one of the sustainable development goals (Goal 3) was to put an end to the avoidable deaths of newborns and children under 5 years of age, and it was defined that all countries must try to reduce mortality to at least 12 and 25 for every 1000 births, respectively [2].

Neonatal deaths are unequally distributed worldwide, with 99% occurring in low- and middle-income countries. The lack of basic neonatal care technologies in low-resource countries is considered a relevant contributing factor to this inequality [3]. However, prevention of mortality due to the three main causes of death (complications associated to premature birth, causes related to childbirth, and sepsis) is possible with the implementation of simple and low-cost interventions, even in countries with limited resources [4]. In addition to increasing survival, the primary objective of obstetric and neonatal care is to reduce morbidity by emphasizing the need for interventions that improve the outcome of immature babies.

The frequency of premature births, occurring before 37 weeks of gestation, also shows regional and global differences. The results show that low-income countries have the highest rate of premature births, with figures such as 15.5 and 15.8% of the total number of births in Pakistan and Indonesia, respectively, in contrast to 12% in the United States, who also has a high percentage of premature babies compared to other developed and high-income countries; providing an important contribution to infant mortality and morbidity [5].

However, extremely preterm infants disproportionately contribute to the burden of neonatal morbidity, mortality, and long-term neurodevelopmental disability, even though a significant

increase in survival without increased neonatal morbidity has been observed in preterm infants born between 25 and 28 weeks [6]. Therefore, increased survival of these newborns makes them more susceptible to developing acute and chronic morbidities such as intraventricular hemorrhage, necrotizing enterocolitis, bronchopulmonary dysplasia, chronic lung disease, and neurosensory disorders, among others.

About 80% of premature children are born between 32 and 37 weeks of gestation, which is known as moderate/late preterm. About 10% of these children are born between 28 and 32 weeks of gestation, the rest correspond to births before 28 weeks. Among the former, which has a comparatively lower risk compared to those of a lower gestational age, some die unnecessarily due to the lack of simple and essential care, such as heat and nutrition [7]. Babies born between weeks 32 and 37 have at least seven times the risk of neonatal mortality and a 2.5 times higher risk of postneonatal infant mortality [8]. Similarly, in the 28–32 week group in lower income countries, more than half of the children die; many could survive without intensive care [7].

Consequently, for neonates to face the risk of death, as well as their short- and long-term morbidity, basic prevention measures such as timely diagnosis and early treatment should be included, taking into consideration the most frequent risks and problems in these patients.

2. Neonatal care and the main problems in newborns

The risk of death is greater on the first day of life and it has been confirmed that the proportion of deaths occurring during the first week of life are constant in all regions and economic environments. This highlights the urgent need to provide timely and high-quality care from the moment of birth. The time between a potentially harmful event and death can be very short and the first minute after birth (golden minute) is the crucial window for neonatal resuscitation.

In general, all newborns are vulnerable after childbirth, which is a key point for growth and development. The highest risk of death is concentrated immediately after birth and in the first days of life. For most newborns, essential care is required, which the mother will ideally provide, such as providing warmth, a clean and safe environment, and providing nourishment through breastfeeding. However, in premature or low birth weight babies, the care requirements are greater since these children are especially vulnerable to thermal instability, difficulty breathing, feeding limitations, and the risk of infections.

In particular, premature babies are especially vulnerable and can become hypothermic in a matter of minutes, which increases the risk of respiratory distress, hypoglycemia, infections, and death. Problems related to childbirth and premature delivery are predominant causes of early neonatal mortality, while infections are more common in the late neonatal period [9].

Due to the abovementioned, temperature is an aspect of basic care for newborns. This is considered fundamental because hypothermia increases the probability of early and late neonatal death [10–12]. Consequently, by providing appropriate thermal conditions during delivery, immediate care, and in intensive care rooms, the risk of hypothermia is reduced and neonatal survival is improved. In this sense, differences have been observed in admission temperatures between extreme and moderate preterm infants, and it has been confirmed that extreme

premature infants have more frequently low and high temperatures at admission. In addition, an inverse relationship between temperature at admission and intrahospital mortality has been observed [13].

Other frequent problem in preterm and very low birth weight infants is hypoglycemia as a consequence of their limited reserves of glycogen and fat, and the inability to use alternative substrates for energy production. In these babies, recurrent and prolonged episodes of hypoglycemia are associated with severe brain damage and poor neurodevelopmental outcome [14–16]. The incidence of neonatal hypoglycemia has been reported between 5 and 15% in healthy children, although it exceeds 50% in risk neonates, while severe hypoglycemia reaches 20% of these children [17].

However, as previously noted, neonatal hypoglycemia not only affects high-risk infants but also continues to be a cause of significant morbidity in term and near-term newborns [18], so this problem should be a priority within preventive actions and early treatment in neonatal care.

In knowledge of the conditions and risk factors of newborns, differential detection and control measures have been proposed. For example, in term neonates, control has been proposed for 12 h after childbirth, while in smaller children the evaluation is continued for 48 h. In other conditions, such as the diabetic mother's son, 24-h monitoring has been recommended [17, 19].

Lately and considering the potential deleterious effects of hypoglycemia, measures of continuous monitoring of glycemia have been proposed, which has shown good tolerance in the newborns and absence of complications. This can detect episodes of neonatal hypoglycemia and hyperglycemia that would not otherwise be detected with intermittent measurements [20]. The use of continuous glucose monitoring in newborns can reduce the frequency of blood sampling and improve glycemic stability, with more time in the euglycemic range. It has also shown a better and timelier detection of episodes of hypoglycemia compared with conventional methods such as intermittent capillary glucose testing [21].

Respiratory disorders are also frequent problems in newborns. Respiratory distress syndrome (RDS) is the most common cause of respiratory distress in preterm infants and the most of children born extremely prematurely evolve rapidly with RDS after birth. The incidence is higher while the baby's gestational age is lower. Up to 98% of babies born at 23 weeks and 86% of babies born at 28 weeks develop RDS [22]. However, the incidence of RDS has decreased in recent decades as a consequence of the use of continuous positive airway pressure (CPAP), conventional ventilation, high-frequency ventilation, and replacement of surfactant for newborns [23].

3. Conclusions

The scenario shown with some examples of frequent problems and interventions involved in neonatal care highlights the need for a complex and comprehensive therapeutic approach. Neonatal care interventions that have been effective in improving the survival of hospitalized newborns and considered a priority are: the use of antenatal corticosteroids to prevent neonatal RDS, early initiation of breastfeeding, umbilical cord care, and kangaroo care in premature babies [24]. Additionally, processes to improve the quality of neonatal care are considered key

points for nutrition, use of medicines, central line care, respiratory care, and care in the delivery room with the aim of reducing the incidence of necrotizing enterocolitis, growth deficit, mortality, sepsis, chronic lung disease, brain damage, and retinopathy of prematurity [25]. Other aspects of special attention in timely neonatal care, particularly in neonatal intensive care, include hemodynamic monitoring and respiratory care.

Conflict of interest

The author has no conflict of interests to declare.

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Neonatal Care in the First Hour of Life

Teja Škodič Zakšek, Anita Jug Došler,
Ana Polona Mivšek and Petra Petročnik

Additional information is available at the end of the chapter

Abstract

The very first hour in a baby's life can have a significant—lifelong—impact on the health of the baby and on the bond between the mother and a baby. Keeping mothers and babies together is a safe and healthy birth practice. Childbirth and the first hour after birth is a time of many changes for both mother and child. Changes are also physiological, as well as psychological. Creating an optimal environment for birth boosts the right hormones for natural birth, which reduces the need for interventions that could cause early mother-baby separation. One of the major challenges in the birth hospital is how best to combine a midwifery care and those medical procedures that are not necessary, to right form the birth as a family intimate and privacy event, if, of course, the child and maternal health would allow this. The first hour after birth is a once-in-a-lifetime occasion for both the baby and the parents, a unique experience, and once lost, it can never be relived.

Keywords: newborn, transition, golden hour, breastfeeding, skin-to-skin contact

1. Introduction

The transition to extrauterine life is a remarkable physiological event that involves a series of modifications that depend on the degree of maturation in late gestation, the process of delivery itself and establishment of independent physiological processes for regulating homeostasis after placenta lost its function. These processes are establishment for respiration, change from parallel to serial circulation, oral feeding, thermoregulation and glucose homeostasis [1]. Respiratory and cardiovascular changes occur simultaneously and are mutually dependent. The triggers of initial first breath are complex and not fully understood yet. Many factors play role in the initiation of breathing, and some of them derive already during the birthing process [2].

We must admit that these are great and demanding changes that need to occur in a short period. However, it is not the purpose of this chapter to describe the processes that occur in the body of the newborn. The main purpose is to remind the readers how to support these natural processes and not disturb them with unnecessary interventions.

When we speak of mature healthy newborn, midwives have to be alert to observe possible complications; however, the newborn in this case does not need any special interventions. On the contrary, the most precious ingredient for the baby in this immediate postpartum period is time. Midwife has to permit natural processes to occur spontaneously and not force them.

The smooth physiological transition can be promoted already by enabling natural processes of the first and second stages of labor; however, we can claim that physiological third stage is even more directly connected to the newborn. Expectant (physiological) third stage of labor is connected to many advantages; because of the delayed cord clamping, baby gets more red blood cells and hematopoietic stem cells and 30% of additional blood volume that is important for respiratory function. At birth, this blood moves into the infant's lung; the cardiac output to the lung changes from 8–10% in utero to 45% in the immediate newborn period and demands an increased blood volume. An adequate red cell volume is necessary for oxygen delivery and consequently effective tissue functioning, normal pH and circulator integrity.

Right after the birth, remarkable changes in respiration and circulation are occurring in the newborn body. Therefore, midwife has to give the baby time for these adjustments. First minute after the birth of the baby, midwife has to observe and wait, and not overstimulate the baby and manipulate with him/her in order to provide the preconditions for these major and dramatical physiological changes. The decision for procedures of stabilization are suggested to be done after the 1st minute Apgar estimation.

When there is no need for resuscitation, the best place for the baby is by her mother. Separating mother and baby can have harmful effect on breastfeeding and their relationship [3]. Skin-to-skin prevents heat loss. Ludington-Hoe et al. [4] confirmed that mother and baby can synchronize body temperatures, when skin-to-skin is practiced; the energy saved can be used to stabilize heart and respiration rates. With kangaroo method also the initiation of breastfeeding is eased. Evidence suggests that the baby, when undisturbed, usually takes about 45–55 min to find the way to its mother's breast, using the primal reflexes [5]. With the birth environment that provides warmth, safety and intimacy, the baby is able to make essential physiological adaptations. Midwives need to follow these physiological transitional processes.

World Health Organization and United Nations Children's Fund [6] say that all mothers and babies should be kept together after the birth and should be encouraged to practice skin-to-skin in the first hour after delivery, even if mothers do not intend to breastfeed. This opportunity should be offered to all, also mothers and babies after cesarean section or vacuum extraction.

World Health Organization and United Nations Children's Fund [6] recommended that all healthy mothers and babies, regardless of feeding preference and method of birth, have uninterrupted skin-to-skin care beginning immediately after birth for at least an hour, and until after the first feeding. All other procedures of initial newborn care can wait until the end of the fourth stage of labor (3 h after the birth), when the woman and the baby are to be discharged

to the postpartum ward [7]. As Gunn et al. [8, p. 765] acknowledge 'in a situation where both, mother and a child are healthy and well, any actions on the part of the midwife should be made unobtrusively and with fully informed consent of the parents.' More importance should be given to the establishment of mother-infant bond, since contact with mother and baby in the hours after the birth not only fosters attachment, but at the same time fosters child's development [8].

2. Keeping mothers and babies together beyond the moment of birth

The first hour after birth, it is extremely sensitive and important for the stabilization of vital functions (breathing, saturation, blood pressure, thermoregulation, blood sugar stability, the newborn must establish pulmonary and cardiac function, etc.) in both mother and child, as well as the process of attachment between them and father. That is why the first hour after birth some call the golden hour [9].

Family bonding and baby's first breastfeed is very important act after delivery. If mother or baby needs some help or medical advice during first breastfeed, then medical staff should help them at this essential time of birth, for both vaginal and cesarean births. If the mother has general anesthesia, we can put a newborn immediately after birth on father's chest. This increases the effectiveness of breastfeeding, the process of attachment between mother and child, and reduces stress in their child [10].

At the moment of birth time, a mother needs a quiet, dim lighting, warmth and calm environment. She is still in labor. Her uterus needs to contract down. With smooth first hour after birth and mother's skin-to-skin contact to a newborn, we allow the newborn to pass through nine instinctive phases in their behavior. These phases are innate and naturally given to every newborn (**Table 1**).

Phase	Naming	Baby's instinctive behaviors—explanation
1	Crying during the birth	Because of lungs expansion, baby starts crying
2	Relaxation	Baby shows relasxed hands without mouth movements
3	Awakening	Baby shows some movements with hands, heads and shoulders
4	Baby' activity	Baby shows mouthing, suckling and way of movements
5	Baby's rest	Phase without baby's activities
6	Baby's crawling	Baby's recognizing the breast and nipple
7	Recognizing with familiarization	Baby familiarizes the nipple and breast. He also licks, touches and massages it
8	Sucking nipples	Baby is attached and is sucking the nipples
9	Baby's sleeping	Baby's restful sleep

Table 1. Baby's instinctive behaviors during bonding and 'skin-to-skin care' after delivery.

The first stage is the birth cry. This distinctive cry occurs immediately after birth as the baby's lungs expand. The second stage is the relaxation stage. During the relaxation stage, the newborn exhibits no mouth movements and the hands are relaxed. This stage usually begins when the birth cry has stopped. The baby is skin-to-skin with the mother and covered with a warm, dry towel or blanket. The third stage is the awakening stage. During this stage, the newborn exhibits small thrusts of movement in the head and shoulders. This stage usually begins about a few minutes after birth. The newborn in the awakening stage may exhibit head movements, open his eyes, show some mouth activity and might move his shoulders. The fourth stage is the activity stage. The newborn begins to make increased mouthing and sucking movements as the rooting reflex becomes more obvious. This stage usually begins about 8 min after birth. At any stage of the phase, the baby may rest. He may have periods of resting between periods of activity throughout the first hour or so after birth. The sixth stage is the crawling stage. The baby approaches the breast during this stage with short periods of action that result in reaching the breast and nipple. This stage usually begins about 35 min after birth. The seventh stage is called familiarization. During this stage, the newborn becomes acquainted with the mother by licking the nipple and touching and massaging her breast. This stage usually begins around 45 minutes after birth and could last for 20 minutes or more. The eighth stage is suckling. During this stage, the newborn takes the nipple, self-attaches and suckles. This early experience of learning to breastfeed usually begins about an hour after birth. It may take more time with skin-to-skin for the baby to complete the stages and begin suckling, especially for mothers who gave birth by cesarean section. The final stage is sleep. The baby and sometimes the mother fall into a restful sleep. Babies usually fall asleep about 1½–2 h after birth [11].

Continuous skin contact between newborn and mother should not affect on the work of the professional staff in the birth hospital. For example, procedures as it is control postpartum bleeding or disruption of the umbilical cord should be carried out without separation of the mother and newborn. If the birth was spontaneous and the child is not under the influences of medicines, keeping mother and newborn together beyond the moment of birth enables the child to be in a state of openness and vigilance and the most susceptible to the first impressions of the outside world. In the opinion of many eminent scientists of the child to design a basic response patterns, intimacy and sociality, which are matrix for all life [9, 12], one of the major challenges in the birth hospital is how best to combine a midwifery care and those medical procedures that are not necessary, to right form the birth as a family intimate and privacy event, if, of course, the child and maternal health would allow this [9, 11, 13]. Preventing separation except for compelling medical indications is an essential safe and healthy birth practice and an ethical responsibility of health-care professionals [14].

During the first hour after birth, many of hormones are releasing: dopamine, oxytocin, prolactin and estrogen. All these hormones initiate maternal instincts. Skin-to-skin contact allows that the mother and child are more relaxed and connected to each other. Whatever promotes the attachment between mother and child: touching, dermal contact, frequent eye contact and so on also promotes the development of a child's brain. Skin contact activates the amygdala, which is a part of the limbic system in the brain that regulates emotional learning, memory processing and detection appetite. This part of the brain is the most developed in just the first

two months of a child's life. Oxytocin receptors in a woman's brain increase during pregnancy. When baby is born, mother is more responsive to this hormone that promotes maternal behavior. Oxytocin is produced in large amounts when breastfeeding and holding babies are close skin-to-skin. Initial attachment has a positive effect on the formation of self-esteem of both parents, because the parents more quickly identify the child's needs and can respond on it. From the child's perspective, the separation from his mother is life-threatening. Keeping mothers and babies together beyond the moment of birth protects the child against the negative consequences of segregation. The frequency of crying and the quantity of stress hormones are lower if child is in skin contact with his mother. In this way, the mother's body heat is also transmitted to the newborn, who is better able to regulate self body temperature and respiration [15]. Skin-to-skin contact heightens response, stimulates behaviors that help to meet the newborn's basic biological needs, activates neuroprotective mechanisms and enables early neurobehavioral self-regulation. Skin-to-skin care reduced maternal physiologic stress and depressive feelings after hospital discharge, which may help to empower women in their role as mothers [16].

3. Behavioral hormonal effects

Blackburn [17] sees hormones as a chemical messengers which either in the body fluids or in blood excert a physiological effect on other cells in other places in the body. The hormones interplay in labor and birth is often compared to an orchestra where every instrument knows exactly how to play perfect notes to create a beautiful melody. If the melody is played well, it sets the stage also in a more immediate way for the postpartum process for both the mother and her baby, because all the different hormones released by mother and fetus during the first and second stages of labor are not yet eliminated during the hour following birth.

One of such hormones is already mentioned oxytocin, which is relatively well studied in relationship to behaviors after birth [18] but still not fully understood [19]. As Phillips [18] notices, it has been shown to increase relaxation, attraction, facial recognition and maternal care-giving behaviors which are all necessary to ensure infant survival. Odent [19] recognizes that oxytocin is never released in isolation. It is always part of a complex hormonal balance in our metaphor part of an orchestra. That means that in the hour following birth, in physiological conditions, the high peak of oxytocin is associated with a high level of prolactin, which is also known as the 'motherhood hormone.' It is known to affect mothering behavior in animals. In humans, oxytocin induces a state of calm and reduces stress [20]. Love and affection between the mother and a child is enhanced, and bonding is optimal. These pleasant moments stimulate the secretion of oxytocin, and also prolactin, and skin-to-skin contact between mother and baby after delivery helps both breastfeeding and emotional bonding [6]. Odent [9] sees this as the most typical situation for inducing love of babies. Oxytocin and prolactin complement each other and are released in response to stimulation by the baby's sucking at the breast. When a baby suckles at the breast, sensory impulses pass from the nipple to the brain. In response, the anterior lobe of the pituitary gland secretes prolactin and the posterior lobe secretes oxytocin [21]. If a mother is in severe pain or emotionally upset, the oxytocin reflex may become inhibited, and her milk may suddenly stop flowing well. In

animals also prolactin is responsible for mothering behaviors [18]. During the first few weeks, the more a baby suckles and stimulates the nipple, the more prolactin is produced, and the more milk is produced. This effect is particularly important at the time when lactation is becoming established, right after the birth.

Oxytocin is responsible for increasingly strong and effective contractions during the labor. And when, during the labor, levels of oxytocin rise, endorphins (sometimes called natural opiates) are released. Beta-endorphin is secreted by the pituitary gland in times of pain and stress. It activates the mesocorticolimbic dopamine reward system and produces pleasure in association with sex, birth and breastfeeding. It is known by now that after birth, both mother and a baby are saturated with natural opiates if the birth is physiological. They reinforce the mother-infant bond and contribute to ecstatic feelings for both [21]. Endorphins also help make the transition to extrauterine life easier for the baby, facilitating relaxation and calm [18].

As the baby descends during the labor, in fact close to the actual birth also catecholamines are released. Sometimes they are called 'fight or flight' hormones: epinephrine (adrenaline) and norepinephrine (noradrenaline).They are secreted from the adrenal gland above the kidney in response to stresses such as fright, anxiety, hunger or cold, as well as excitement, when they activate the sympathetic nervous system for fight or flight. During birth, when women are scared or have difficulty coping with pain, they can be overproduced and can inhibit production of oxytocin. However, normal values ensure mother is alert when baby is born; also, baby is alert, with eyes wide opened and trying to make eye contact with mother [19].

To our current knowledge, many *different hormones* can *influence* several *types of behavior*, but for the purpose of getting to know the behavioral effects of different hormones involved in the birth process, four most important ones were described. It is known by now that all the different hormones released by the mother and by the baby during labor and delivery are not eliminated immediately. By knowing that, we realize it is essential to promote best practices already in labor processes.

4. First hour and maternal attachment behaviors

Maternal attachment and bonding does not start at birth; from psychological point of view, the system has been prepared during the whole pregnancy, when mother imagines her baby and when the baby gets to know the odor, voice and smell of mother [22]. However, after the birth, she encounters him/her for the first time and therefore this time is so crucial for the establishment of the relationship between the baby and the mother/parent. The space for this intimate process must be given to the family and as Gunn et al. [8, p. 765] write: 'the midwife should never undermine the role of the mother who is transitioning into her new role.'

When left undisturbed, mothers demonstrate 'species-specific behavior' [23]. Mother explores her baby with her fingertips, then strokes the child and even then cuddles him/her into her arms, facing her. She establishes eye contact, talks to her baby and then introduces him/her to partner [8]. She progresses through three major steps:

- Her first preoccupation is the survival of the baby.

- Then, she needs to know that everything is fine with the baby.

- Once reassured that the baby is healthy, mother wants to make the baby her own. She seeks physical resemblance.

These steps are crucial for every new mother; however, this is not yet a relationship [24]; attachment is much more complex and takes more time to be established. The initial emotional connection that mother establishes with her newborn baby is called bonding [25]. It was believed that bonding is one-way relationship (from parents toward child), under the strong influence of important maternal and infant oxytocin that promotes empathy. Other neurotransmitters such as opioids and dopamine also play role in the bonding process [26–28]. It can be therefore concluded that bonding is eased when the birth process is natural and all these hormones are expressed. Within the context of the results of latest research, the experts began to question whether bonding is really a one-way relationship. Feldman et al. [29] found synchronic levels of oxytocin in infants and mothers who interacted with them. These high levels of oxytocin in baby help her/him to adapt to extrauterine life [16]. Despite the fact that babies communicate nonverbal, they respond to parents.

Also, the baby responds facially to mother's voice, especially in the case of physiological birth, right after the birth, when the baby is in a quiet alert state, aware of the surrounding and uses all his/her senses. The baby has competencies to develop ties with parents [24]. Besides voice, his/her strongest sense is scent, necessary to find the mother's breast. After the first feed, baby usually gets to sleep that can last even 6 h [30].

All these (nonverbal, mostly facial) responses of the baby evoke interaction with parents that sets grounds for developing a bond among them. They were acknowledged already by Bowlby [31]. He proposed that there is an attachment system that is biologically based and promotes survival. He claimed that infants have specific behaviors that attract proximity of the caregiver in order to survive or to be emotionally connected, so-called proximity-seeking behavior. Repetitions of such interactions by the caregiver lead to 'internal working model' or internal representation of the attachment relationship [32].

The infant, despite that he/she is not verbal yet, generates these affective, sensorimotor activities from parents. He cannot self-regulate yet, but can learn this capacity through parental care-giving behaviors and his own ability to self-regulate [32]. It is therefore of crucial importance that woman is relaxed and in touch with her own feelings.

Reid and Freer [33] wrote that maternal role develops smoothly when mothers' self-esteem in mothering abilities is enhanced. Midwife can strengthen her perception with different interventions. If they make parents aware of babies' behavioral and autonomic cues, they can be more confident in caring for their newborn, taking into account the child's individual tolerance (for habituation to noise, light, etc.) [34].

Benefits of skin-to-skin for attachment, breastfeeding and thermoregulation are well known, and new insights, however, revealed even other advantages. Colonization of the baby with the mother's microbiome occurs first during vaginal delivery, later on with her skin microbes,

and during initial breastfeeding, also the newborn gut is colonized with microbes that built normal gastrointestinal flora.

5. A metabolic perspective

When newborn transits to a life outside a womb, it must adapt to many new circumstances. One is metabolic transition, which is not as dramatic as, for example, changes in cardiopulmonary systems, but equally complex and essential for survival. As Colson [35] notices cardiopulmonary, immune and thermal adaptations are well documented, but most texts fail to describe the normal physiological metabolic transition from fetus to neonate.

Just after birth, as soon as the umbilical cord ceases to pulsate, placental circulation stops. This means that the constant supply of maternal nutrition especially glucose transferred via the placenta stops. Before the birth, no significant production of glucose has been demonstrated [36]. In utero insulin is being used as a growth hormone instead of being a metabolic regulator. Colson [35] explains that the processes of lipogenesis (formation and storage of fat in the form of adipose tissue) and glycogenesis (formation and storage of glucose in the form of glycogen in the liver, cardiac muscle and brain) are replaced by the metabolic pathways of neonatal life. These are glycogenolysis (breakdown of glycogen), lipolysis (breakdown of fats), gluconeogenesis (endogenous glucose production) and ketogenesis (formation of ketone bodies).These pathways imply a metabolic switch at birth from glucose to fat and therefore a diet initially lower in carbohydrate and high in fats. It is true that while neonatal blood glucose levels immediately fall in almost all healthy infants, it must adapt to intermittent feeding, digestion and intestinal absorption of nutrients (adapted from Colson, p. 13). The fetus prepares for his transition mainly by storing glycogen, producing catecholamines and depositing brown and white fat [37]. After the birth, hepatic glycogen stores are mobilized and hepatic synthesis of glucose from noncarbohydrate substrates enuses. This substrate enters the citric acid cycle and produces adenosine triphosphate, which serves as the energy source for the brain [37]. These events actually allow baby to gradually mobilize glucose to meet energy requirements. So-called transient neonatal hypoglycemia is a process of normal adaptation to extrauterine life, and it is important that we realize that in first 3–4 h healthy newborn could have low blood glucose levels.

Colson [35] exposes several practices that stand behind understanding of the normal metabolic physiology:

1. Metabolic transition is not generally taught in midwifery and medical curricula as part of normal postnatal adaptation from fetus to neonate. Descriptions of metabolic changes are absent or sparse. When present, they are usually rooted in pathology.

2. Research has shown that patterns of metabolic adaptation are different according to whether the baby is breastfed or has artificial feeds and this is largely ignored in midwifery and pediatric assessment. Mixed feeding is common in the first three days postnatal even when the mother wants to breastfeed exclusively.

3. Furthermore, in the early postnatal days, current breastfeeding definitions disregard dose. A baby is considered to be breastfed when receiving any amount of mother's milk, however small. Not knowing whether the baby is exclusively breastfed blurs the understanding of those clinical characteristics associated with a baby who is wholly breastfed.

4. Mothers are often encouraged to swaddle their babies from birth and to keep them in the cot unless they are actively feeding. This practice assumes that the continuity of maternal nutrition ends at birth as in bottle-feeding. Immediate swaddling also accentuates the discontinuity of postnatal transition, as mothers are physically separated from their babies even when they are in the same room. The early physical separation negates the continuity and postnatal effectiveness of the maternal body to maintain a homeostatic neutral/thermal environment from fetus to neonate. Keeping babies in the cot in between feeds instead of holding them during the first three days postpartum may have a negative effect upon early nurturing and breastfeeding.

5. Maternal choice rather than physiology provides the framework that underpins midwifery assessment. When there are breastfeeding problems in the first three days postnatal, a bottle-feeding solution is often offered. For example, when the baby demands breastfed and is unsettled, it is often believed that mother's early colostral milk is insufficient. Mothers are often told that they can give the baby a bottle if they want. The irony is that maternal choice then appears to motivate supplementation. One often sees written in the notes 'baby unsettled, mother requested bottle' (Colson, p. 12).

In order to optimize metabolic adaptation, babies and mothers must be kept closely together after birth. Health workers must encourage mothers to maintain close body contact with their babies as often as they want in an undisturbed environment [38].

6. Impacts on infant microbiome assembly

The human body is colonized by a vast number of microbes, collectively referred to as the human microbiota. The average human has over 100 trillion microbes in and on their body, and many of the latest discoveries are challenging previously held ideas about good and bad bacteria. Funkhouser and Bordenstein [39] say that the human microbiota comprises only 1–3% of an individual's total body mass, outnumbering human cells 10 to 1 and adding over 8 million genes to our set of 22,000. At the beginning of the twentieth century, French pediatrician Henry Tissier said that human infants develop within a sterile environment and acquire their initial bacterial inoculum while traveling through the maternal birth canal but now the sterile womb hypothesis remains dogma. The intrauterine environment during healthy pregnancy has been presumed to be free of, although recent evidence of microbes presents in the amniotic fluid, umbilical cord blood, fetal membranes and placenta of healthy term pregnancies after both vaginal and C-section delivery has challenged this belief [40]. It is known by now that human infants are colonized with maternal vaginal and fecal microbes as they exit the birth. The way how is known to have long-term consequences on mothers and child

health. This is especially important considering immune-mediated diseases. For example, children born via C-section are significantly more likely to develop allergic rhinitis, asthma, celiac disease, type 1 diabetes and inflammatory bowel disease [40].

Besides mode of delivery, breastfeeding also provides a route of maternal microbial transmission. Breast milk was considered sterile at first, but in colostrum collected aseptically already harbors hundreds of bacterial species [39].

To ensure the best maternal transmission of beneficial microbes, Reed [41] has made following suggestions:

• Baby should be naked on mothers chest immediately following birth for at least an hour and a lot in following frost days.

• Avoid bathing baby for at least 24 h after birth [42]. Use own linen from home for baby if in hospital.

• Minimize the handling of baby.

• Exclusively breastfeeding. If not, probiotics should be considered.

• Avoid unnecessary antibiotics for the baby [43, 44].

• Probiotics may also be beneficial for babies suffering from colic.

The complex symbiosis between humans and microbes is important for our health, and breastfeeding benefits the health and well-being of infants. Maternal transmission is also a key factor in shaping the structure of the microbiome in animal species over evolutionary time, since microbes that promote host fitness, especially in females, will simultaneously increase their odds of being transferred to the next generation; therefore, it is essential to create optimal conditions to achieve the transmission.

7. First hour and breastfeeding

Breastfeeding has many advantages for the child, mother and the environment. The smooth first hour after birth and mother's skin-to-skin contact to newborn have positive impact on the effectiveness and duration of breastfeeding. Shorter intervals between birth and the start of skin-to-skin care and longer times spent skin-to-skin after birth improved breastfeeding exclusivity and duration. In the first month, woman has to breastfeed as often as the newborn wants or even more. With this the production of milk is assured. Many females are meeting with problem of too small amount of milk. The most frequent reasons are too little ingested liquid and disorderly diet. The problem can also be tiredness, increased amount of stress and rare short passing feeds. Relaxed and satisfied mother, who lives in a pleasant and tranquil environment, will have much better conditions for smooth milk lactation. The production and amount of milk is determining by the law of demand and offer. Birth environment and each health professional in their professional action may be more or less supportive impact on the ability and confidence of women to be born, breastfeeding and care for the baby, and baby's

ability to effectively breastfeed. Full breastfeeding can of course be established successfully after a cesarean section also. The beginning of the milk secretion can be delayed after caesarean section, however, there is no rule. It is important to know that a child for successful feeds need to have a search reflexes, reflexes of swallow and sucking reflex which is instinctive. The way of the childbirth does not have influence per this. It is true that the first feed is postponed as a mother is put to sleep during an intervention. Clinical staff has to help her to add a child only after a certain time, when a mother is wake and she is aware of herself and surroundings. There are qualified professionals working in various medical institutions that hold a specific and additional knowledge about lactation and breastfeeding. With breastfeeding, the 'good bacteria' from the mother's body with a calm environment create good conditions for the development and strengthening of the child's immune system. The WHO and UNICEF are recommending three important breastfeeding activities: (1) early breastfeeding and skin-to-skin contact with mother just after the birth; (2) exclusive breastfeeding to baby's age of 6 months without other food or liquids and (3) continued breastfeeding to baby's age of 2 years or even more. Meantime, the child can get complementary foods like soft foods and liquids, etc. [45–47]. Early breastfeeding and skin-to-skin contact immediately after birth keep a baby warm and have positive influence on their immune system. Despite that breastmilk is the best food with antibodies for baby's development, it also has effects on mother's ability of continuing exclusive breastfeeding. Mothers who breastfeed also have a (1) early initiation of breastfeeding—place newborns skin-to-skin with their mother immediately after birth, and support mothers to initiate breastfeeding within the baby's first hour of life; (2) exclusive breastfeeding—provide only breastmilk to infants from birth until 6 months of age, with no other food or liquids (including water); (3) continued breastfeeding—breastfeeding until age 2 or longer, in addition to adequate and safe solid, semisolid or soft foods (also called complementary foods) [45–47]. Immediate skin-to-skin contact and starting breastfeeding early keeps a baby warm, builds his or her immune system, promotes bonding, boosts a mother's milk supply and increases the chances that she will be able to continue exclusive breastfeeding. Breastmilk is more than just food for babies—it is also a potent medicine for disease prevention that is tailored to the needs of each child. The 'first milk'—or colostrum—is rich in antibodies to protect babies from disease and death, lower risk of developing breast and ovarian cancers. Breastfeeding can also delay mother's ovulation [13, 47].

One of the important factor that contributes to good establishment of breastfeeding is adding of the newborn to the mother's chest as soon as possible after birth, advisably to first half an hour or at least in hour after childbirth. If mother needs an advice or help during this time, it is very important that she gets it. First of feeds is introducing the first food and the first immunization to a child which further encourages the production of colostrum. Baby's sucking reflex is expressed the most during the first hour after a birth. It is awaken during skin-to-skin contact and care. If the mother is breastfeeding the newborn immediately after birth, the hormonal balance during pregnancy is established for a long time, which very favorable impacts on mother's overall health being. In addition, under the influence of the hormone oxytocin, which is secreted, while a newborn stimulates mother's nipples, the uterus intensively cramps and quickly returns to its original size. And this reduces the likelihood of severe bleeding after childbirth. Oxytocin, which increases significantly during skin-to-skin care, promotes

newborn attachment, reduces maternal and newborn stress and helps the newborn transition to postnatal life. With breastfeeding in first hour, the child also has a stable heart rhythm without bradycardia. The possibility of apnea is reduced by 75%, since the depth of each breath becomes more stable [9, 13, 48].

Colonization by mother's bacteria and first lactation colostrum, which creates an optimal intestinal flora, is an optimal protection of the child immunity from possible allergies which might otherwise can be developed later in child life. Breastmilk is more than just food—it is also a potent medicine. It protects the child against disease, regulates the child's immune system and helps child to digest the food [47]. This process helps to program the healthy development of the infant's gut microbiome for life. There is evidence, for example, that breastmilk can help to counteract an infant's genetic predisposition for obesity and other chronic diseases. So the first hour after birth is a critical period with irreversible consequences from the point of bacteriological view [12, 48]. More on that can be found in impacts on infant microbiome assembly chapter.

Mother's breasts are natural thermoregulator to maintain the body temperature of a child. They regulate the temperature of a child. If the child is cold, the breast temperature increases, or if the child is warm, the breast temperature falls. Mother's breasts are also natural thermoregulator for child's respiratory and heart rates [8, 10, 48].

An undisturbed first hour with skin-to-skin also reduces the risk of hypoglycemia (see chapter about metabolic adaptation). Newborn babies can produce glucose from their body stores of energy until they are breastfeeding well and are more likely to do so when they remain skin-to-skin with their mothers. Breastfeeding extended period of natural immunity against mumps, measles and polio. Colostrum has a laxative effect and helps to facilitate the elimination of the first child's stool. It is extremely easy to digest and does not cause constipation. The child has also less troubling with abdominal cramps. As long as the child is breastfed, it is protected against many infections, because breast milk receipt of child antibodies that protect against diseases that can overcome its mother. The child is also protected against ear infections, diarrhea, gastrointestinal infections and diseases of the respiratory tract. For breastfed infant, it is less likely to be diagnosed with meningitis and childhood diabetes. An active intake (compared with passive swallowing bottle) promotes the proper development of the jaw, the mouth muscles and cheek bones, resulting in a very favorable impact on the development of children's speech.

8. Conclusion

First hour following birth for mature newborn is without doubt the most critical hour in life of human beings. During this time, a lot of changes happen. When woman gives birth, all the hard work she does generate changes in the chemistry in the brains. It makes women want to nurture her child. These hormones also cause the uterus to contract, shrink and stop bleeding.

Based on decades of evidence, the World Health Organization and United Nations Children's Fund [6] recommended that all healthy mothers and babies, regardless of feeding preference

and method of birth, have uninterrupted skin-to-skin care beginning immediately after birth, lasting for at least an hour.

There is still a lot of unnecessary interventions in the first hour after birth in many maternity hospitals. Routine procedures are being carried out starting from early cord clamping to vitamin K injection, eye prophylaxis antibiotic ointment, navel prophylaxis, foot and hand printing, weighing, measurements and bathing and others. All health-care providers should know that immediate skin-to-skin contact is the best way for a newborn and mother to bond. Healthy newborns should be placed in 'skin-to-skin' contact with the mother until the first round of breastfeeding is established. Skin-to-skin care means placing dried, unclothed newborns on their mother's bare chest, with warmed light blankets or towels covering the newborn's back. Women who have a planned or unplanned C-section would not be in the ideal position for intimate bonding right away. Baby could be taken to a warming table for a quick assessment first. Authors [18] claim that there is no reason why stable mothers should not have the experience of skin-to-skin contact after cesarean births, to collect the same short- and long-term benefits of it. Even from psychological point of view, it helps them mourn the loss of a normal vaginal birth.

The first hour should be focused on baby's first breastfeed and mother-baby and family bonding. The manner in which a new baby is welcomed into the world during the first hours after birth may have short- and long-term consequences.

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Hemodynamic Monitoring in Neonates

Petja Fister and Štefan Grosek

Additional information is available at the end of the chapter

Abstract

Sick neonates are often hemodynamically unstable, hence their organs are inadequately supplied with oxygen. In order to maintain blood flow to vital organs, a number of compensatory mechanisms divert the blood flow away from the non-vital organs. If hemodynamic changes are detected early, the cardiovascular compromise can be recognized in compensated phase and thereby the escalation to decompensated phase of low cardiac output syndrome might be prevented. In the treatment of hemodynamically unstable neonate venous filling, contractility of the heart muscle, blood pressure in the aorta, systemic blood flow, and regional distribution of blood flow should be evaluated. There are many evaluation and measurement methods based on different physical basis, each of them having their advantages and disadvantages. For most of them, it has not been demonstrated that they improve outcomes of sick neonates. Using these methods, useful hemodynamic data for the treatment of sick neonates can be obtained. Using new techniques will clarify the pathophysiology of cardiovascular failure in sick neonates, assess the effects of drugs on blood pressure and perfusion of the heart and other organs.

Keywords: neonate, hemodynamics, oxygenation, perfusion, arterial blood pressure, cardiac output, peripheral vascular resistance

1. Introduction

In neonatal intensive care unit (NICU), hemodynamic instability is an important cause for admission and treatment after respiratory distress syndrome (RDS) and most common problems of prematurity [1]. Therefore, hemodynamic monitoring is important especially in transitional period to extrauterine life and during the next following days. Hemodynamic monitoring is also crucial in neonates with congenital heart defects (CHDs) and other complex surgical anomalies of neonates. During early transitional period, cardiac output (CO) is not dependent solely

on the performance of the neonate's left ventricle but also on pulsating blood flow through the umbilical vein. This pulsating blood flow is extremely important especially if newborn is under fetal distress, and it is usually prudent to postpone ligation of umbilicus for a short period of time to add additional pulsatile and volume support to neonate's CO [2, 3]. Even later in the life, neonate with RDS requires hemodynamic monitoring due to disturbances related to RDS or other common problems of prematurity or immaturity. Besides prematurity, immaturity of cardiovascular system in the first days and weeks of life and altered physiology of systemic and pulmonary circulation in neonates with CHD may need frequent hemodynamic monitoring, whether invasively or noninvasively.

2. Methods

We conducted electronic searches of articles on hemodynamic management and care of neonates, using key terms: neonate, hemodynamics, oxygenation, perfusion, arterial blood pressure, cardiac output, and peripheral vascular resistance in the PubMed data base from the years 2000 to 2017 and reported the most relevant ones. The article lists the methods for the evaluation of the venous filling, cardiac output, blood pressure, regional blood flow, and microcirculation, from the clinical methods to the noninvasive and invasive ones. Some methods for monitoring the cardiovascular status of neonates are mainly used for research purposes. This chapter includes the impact of optimal arterial blood pressure, tissue perfusion, and persistent ductus arteriosus on the hemodynamic management in neonates. Also, the short- and long-term outcomes in respect of hemodynamic management of neonates in the intensive care unit are addressed.

3. Physiology of hemodynamics

Hemodynamics describes the dynamics of blood flow in the body. Blood flows in the circulatory system, which is composed of the central pump—the heart—and the blood vessels, and is controlled by homeostatic mechanisms. The pulsatile rate of blood flow out of the heart is called the cardiac output. The main role of the cardio-circulatory system is to match the oxygen and nutrient needs of the organs and tissues and elimination of the metabolic wastes. The cardio-circulatory system should provide an appropriate blood flow to the organs and by that an appropriate tissue perfusion. In the physiologic conditions, the tissue oxygen and nutrient needs are matched by their supply. In cardiovascular compromise, the compensatory mechanisms allow the redistribution of blood flow to the vital organs—the brain, heart, and suprarenal gland in the neonate—at the expense of decreased blood flow to the non-vital organs.

3.1. Hemodynamic monitoring

Hemodynamic monitoring encompasses the observation and measurement of hemodynamic parameters over time. The ultimate goals of hemodynamic monitoring is to alert the

health-care team of impending cardiovascular crisis, to obtain information specific to the disease processes, which may facilitate diagnosis and treatment and allow one to monitor the response to therapy, and also to derive estimates of performances and physiological reserve that may in turn direct treatment [4]. The purpose of hemodynamic monitoring is to attain the optimal goals of cardiovascular therapy. Three functional-based questions should be addressed: (1) Will blood flow to the body increase (or decrease) if the neonates' intravascular volume is increased (or decreased), and if so, by how much? (2) Is the decrease in arterial blood pressure due to loss of vascular tone or merely due to inadequate blood flow? (3) Is the heart capable of maintaining an effective blood flow with an acceptable perfusion pressure without going into failure?

Physiologically, hemodynamic parameters can be divided into central or macro-hemodynamic parameters, which assess blood flow and pressure in the heart, vena cava, pulmonary artery, and the aorta, and the peripheral or micro-hemodynamic parameters, which assess the regional microvascular blood flow and tissue oxygenation (**Table 1**). The majority of existing hemodynamic monitoring assesses the central part of the cardio-circulatory system.

In the past, hemodynamic monitoring data were attained in neonates with CHD after heart surgery when the invasive and frequently inaccurate pulmonary artery catheters have been inserted [5]. Recently, efforts have been made to monitor the newborns as noninvasively

Hemodynamic parameter	Physiologic parameter	Clinical assessment	Noninvasive measurement	Invasive measurement
Central or macro	Preload—venous filling	Estimating jugular venous pressure, hand pressure on the liver, elevation of the legs	Inferior vena cava diameter and its collapsibility, lung ultrasonography	Central venous catheters*
	Cardiac output	Palpating peripheral pulses, heart rate*, capillary refill time, peripheral-core temperature difference	Echocardiography, cardiac magnetic resonance, electrical cardiometry*, arterial pulse waveform analysis*, electrocardiogram	Arterial catheters*
	Afterload—arterial blood pressure	Palpating peripheral pulses	Cuff oscillometry, Doppler ultrasound	Arterial catheters*
Peripheral or micro	Regional blood flow	Skin color, lactate	Perfusion index*, near-infrared spectroscopy*	
	Microcirculation		Laser-Doppler method, video microscopy, xenon clearance techniques	

*Continuous method, all others are intermittent.

Table 1. Physiologic hemodynamic parameters in neonates, assessed clinically, noninvasively, and invasively.

as possible and many new techniques have been applied in this vulnerable population to measure the central (arterial blood pressure and systemic blood flow) and peripheral hemodynamic parameters (peripheral vascular resistance). Arterial blood pressure is measured either noninvasively by sphygmomanometer or invasively through arterial catheters. Systemic blood flow is noninvasively assessed by echocardiography, cardiac magnetic resonance, electrical cardiometry, and arterial pulse waveform analysis. Invasive methods for measuring the systemic blood flow are applied through centrally inserted vascular catheters. It is not known whether laser-Doppler and spectroscopy in the near-infrared spectrum (near-infrared spectroscopy, NIRS) can reliably monitor peripheral vascular resistance (**Figure 1**) [6].
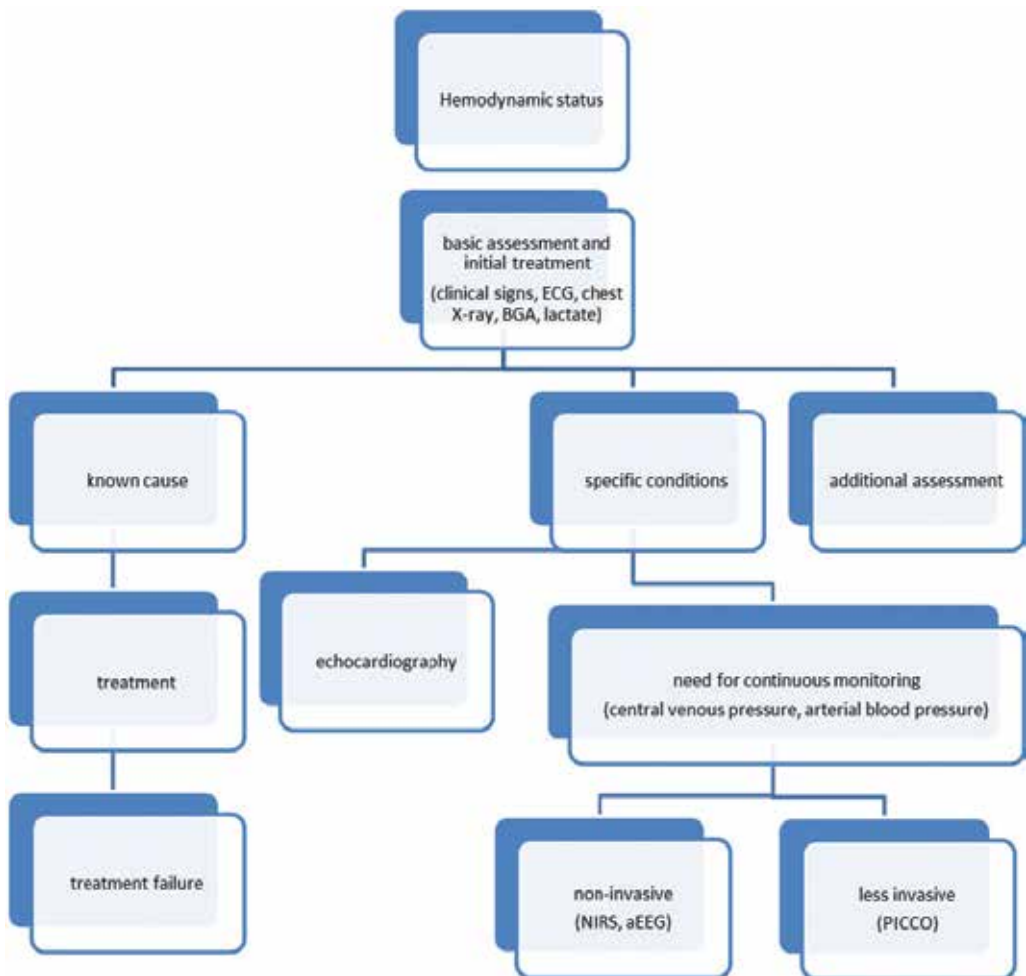

Figure 1. Assessment of hemodynamic status in neonatal intensive care unit. Adapted from Ref. [64]. aEEG, amplitude-integrated electroencephalogram; BGA, blood gas analysis; ECG, electrocardiogram; NIRS, near-infrared spectroscopy; PICCO, pulse-induced contour cardiac output.

4. The central hemodynamic monitoring

The central hemodynamic monitoring assesses the blood flow and the blood pressure in the heart and major vessels. The heart function and by that the stroke volume (SV) are determined by the preload—the venous filling, contractility of the heart muscle, and the afterload—which can be estimated only partially with the pressure in the aorta. The CO is the product of the SV and the heart rate (HR). Neonates increase their CO mainly by increasing the HR as they cannot sufficiently increase the SV. The HR is influenced by the body temperature, catecholamine secretion, and the autonomic nervous system.

4.1. The preload assessment

Clinically, the preload can be assessed visually assessing the jugular venous pressure, which is rarely possible in neonates with short neck. Using the hand pressure on the liver or elevation of the legs increases preload and is a simple method of assessing preload but less used in the NICU. The venous filling in neonates can be measured by inferior vena cava (IVC) diameter, and its collapsibility during respiration indicates volume responsiveness [7, 8]. IVC assessment is not an accurate marker of volume status and fluid responsiveness in cases of (1) increased right atrial pressure, (2) tricuspid or pulmonary valve regurgitation, (3) pulmonary hypertension, or (4) right ventricular dysfunction. Another method for assessing volume status of a neonate is lung ultrasonography (US) [9, 10]. Sonographic visualization of B-lines and measurement of extravascular lung water may aid diagnosing early volume overload in neonate. In a neonate with RDS after receiving surfactant, the B-lines are still visible [11].

4.2. Cardiac output

The measurement of CO is the most important parameter of the central hemodynamic monitoring, assessing the perfusion of organs. It is vital for the etiopathogenic diagnosis of low cardiac output syndrome, being due to either hypovolemia, myocardial dysfunction, vasodilatation, tamponade, pneumothorax, obstructive shock, pulmonary hypertension, or acute RDS. CO should be measured in the following clinical conditions: congenital and acquired heart diseases, shock, multiple organ failure, cardiopulmonary interactions during mechanical ventilation, clinical research, and assessment of new therapies [12]. The following three questions guide us in the interpretation of the adequacy of CO: (1) Is the delivery of oxygen adequate to meet the metabolic need of the patient? (2) Is oxygen delivery occurring with an adequate perfusion pressure? (3) Is the patient able to utilize the oxygen delivered, and if not, why so? [13]. Not only the measurement but also adequacy of CO and oxygen delivery is important, reflecting in clinical (capillary refill and core-peripheral temperature difference) and laboratory (lactic acid) parameters. But caveat is needed; normal values do not mean that regional perfusion is adequate, as well as abnormal values do not provide us with etiologic clue.

Clinically, CO with the systemic blood flow and perfusion can indirectly noninvasively be assessed by palpating the peripheral pulses and heart rate, capillary refill time, and measuring the peripheral-core temperature difference [14]. None of clinical methods for the evaluation

of CO is definitely reliable [15]. The HR in neonate is affected by many factors and so it is not necessarily a good indicator of hemodynamic status. In hypovolemic state, the immature heart muscle and the autonomic nervous system impact the cardiovascular response differently in neonates in comparison to adults. The capillary refill time is affected by the pressure technique, variability among investigators, ambient temperature, drugs, and maturity of neonatal skin. The capillary refill time of >3 s has 55% sensitivity and 81% specificity for the prediction of the low systemic blood flow [16].

Noninvasive methods that are used in clinical practice to assess neonatal CO are echocardiography, cardiac magnetic resonance, electrical cardiometry, and arterial pulse waveform analysis. Functional echocardiography (fECHO) enables visualization of the shape and size of the chambers of the heart, the heart valves, contractility, and relaxation of the heart muscle. Various forms of Doppler echocardiography, such as color, continuous and pulsed Doppler echocardiography, enable the determination of both the blood flow direction and velocity. By assessing the velocity of blood flow, two more parameters can be calculated: the difference in pressure above and below the narrowing with the modified Bernoulli's equation and the blood flow through the blood vessel diameter and a mean flow velocity of the blood [17].

fECHO is daily used in the neonatal intensive medicine and constitutes an important part of an integrated bedside hemodynamic monitoring of a neonate. An appropriate training and experience is necessary for performing fECHO; so the obtained measurements are accurate and reliable. It should be noted that fECHO is not a substitute for formal echocardiographic assessment of the neonate by a pediatric cardiologist, especially if the newborn is suspected to have CHD or the latter has already been diagnosed [18, 19]. fECHO enables discontinuous hemodynamic assessment of neonate in real time. With fECHO, we can estimate the central venous pressure, and thus the volume of blood, and the contractility and filling of the right and left cavities, pulmonary arterial pressure, and the SV of the both ventricles, thereby CO. With neonatal fECHO, we can assess the presence, direction, and measure the blood flow velocity through the shunts, such as persistent ductus arteriosus (PDA) and open foramen ovale, the pressure in the pulmonary artery and the superior vena cava (SVC) and thereby assess heart function. The drawback is the limited accuracy of the measurements (10% with a single investigator and 20% among various investigators). Acquired measurements also depend on the blood flow: from the transitional to neonatal, shunts through connections between the pulmonary and systemic blood flow and immaturity or lability of the lung vasculature, and thus the pressure in the pulmonary artery [20].

fECHO is helpful in the treatment of neonates with cardiovascular instability, PDA, persistent pulmonary hypertension, preterm premature rupture of membranes, perinatal asphyxia, sepsis, and general management in intensive care for a sick neonate. In the case of hemodynamically significant PDA, fECHO has significant limitations in measuring CO. Significant left-right shunting through PDA apparently increases SV of the left ventricle, which is a measure of pulmonary blood flow; outflow from the right ventricle is a measure of the systemic blood flow. Similarly, the measurement of flow from the right ventricle is affected by the presence of left-right atrial shunting which apparently increases SV and the outflow from the right ventricle. For this reason, the assessment of the systemic blood flow is made through the measurement

of the venous return to the heart through the SVC. The flow in the SVC is normally between 30 and 50% of the total systemic blood flow. Similarly, to assess the pulmonary blood flow, the blood flow velocity in the left pulmonary artery is measured [21].

Measuring the ejection fraction (EF) and fractional shortening (FS) in hemodynamic monitoring of the newborn is not very useful measurement, except in the case of severe myocardial dysfunction, when measurements are not really necessary, since the poor contractility of the heart muscle is obvious. We calculate them on the basis of measurement differences in dimensions of left ventricle in the long axis between diastole and systole. The problem of measurement in neonates is that the anterior wall of the ventricle is relatively stiff in comparison to the posterior and lateral wall of the left ventricle. To avoid this, they proposed measuring the dimensions of the left ventricle in short axis [22]. Two new US methods for assessing cardiac function are the measurement of diastolic function and tissue Doppler US. Diminished diastolic relaxation effects systolic function of the heart muscle, and thus the SV and CO. We may assess the diastolic function from the shape of Doppler wave of the inflow of blood into the ventricle. The filling of the ventricle has two phases: early ventricular filling during relaxation (E wave) and late active ventricular filling due to atrial systole (A wave). In a healthy neonate, 80% of the blood fills the left ventricle early in diastole, so the predominance of A wave indicates impaired diastolic function [23]. But when assessing impaired diastolic function, we have to be aware that diastolic function in premature and mature infants is already impaired because of immature contractile system of cardiac muscles [22]. The method of tissue Doppler echocardiography detects low frequency of high energy resulting from the movement of the ventricular wall which cannot be assessed by the standard Doppler investigation. In the four-chamber view of the heart in the longitudinal axis, we observe three different variables of ventricular wall motion: velocity, acceleration, and displacement [24]. Even if the method is advantageously used in adult patients, their clinical utility in the treatment of sick newborns is not yet fully understood [25].

Cardiovascular magnetic resonance (CMR) imaging is a method of nuclear magnetic resonance based on the spinning of hydrogen nuclei in a magnetic field, which are most numerous in the human body. CMR is the gold standard for the assessment of CO. Special expensive equipment, trained personnel, sedation, and transportation of a neonate are needed to perform CMR. In adults, CMR is used to assess the function of the ventricles, in complex congenital heart disease and cardiomyopathy. Compared with adults, it is necessary to increase the image resolution in neonates, both spatially (because of the size of the heart), as well as the time (due to the relatively high heart rate of newborn). The essential advantages of this method are detailed assessment of CO, cardiovascular anatomy, and good repeatability. CMR cannot measure the oxygen need and consumption [26]. Images obtained by CMR in real time have lower quality. Kino CMR enables improved anatomy imaging and the ventricular wall motion, by which the heart is imaged at specific phases of the cardiac cycle, depending on the electrocardiographic (ECG) recording. This creates a series of images that can be played as a movie (kino). End-diastolic and end-systolic endocardial and epicardial borders are followed and we reconstruct three-dimensional models of ventricles: end-systolic and end-diastolic volume, ejection fraction, and SV. CMR with a phase contrast allows the measurement of blood flow to the heart throughout the cardiac cycle [27]. This method quantifies the flow in IVC and descendent aorta,

which indicates the systemic perfusion in the premature neonate. Similarly, we can measure the flow in the internal carotid and basilar arteries and thus the blood flow in the brain [28].

Electrical impedance cardiometry is the only available method that enables continuous noninvasive monitoring of SV and CO in a neonate [29, 30]. The method is based on a model of the electrical velocimetry, using four-surface ECG electrodes attached to the left side of the neck (two electrodes), and to the chest (two electrodes). Alternating electric current (AC) of constant amplitude flows through the pair of external electrodes toward the direction of the aorta. The ratio of the current and measured voltage is equal to the conductivity (or bioimpedance). Each tissue in the chest has its bioimpedance: that of the blood is very low, whole bone and lungs, filled with air, have high bioimpedance. Moreover, bioimpedance of bone is static, and bioimpedance of the lungs, which are filled and emptied of air, and of the heart and large blood vessels, which are filled with blood, is dynamic, in accordance with the respiratory or cardiac cycle. In case of sudden acceleration of blood flow into the aorta in systole, the conductivity dramatically increases. Electrical bioimpedance of the chest is strongly increased with every heartbeat. The neonate's movement causes artifacts in measuring the electrical impedance [31, 32].

Arterial pulse waveform analysis is a relatively good method for monitoring the dynamics of arterial blood pressure and assessment of CO. It is based on the analysis of the curve of arterial pulse waveform derived from arterial catheter and on the fact that the pulse pressure is proportional to SV [33]. The arterial pulse waveform changes in the case of arrhythmia, shock, or hypothermia, when it comes to peripheral vasoconstriction. Typically, the devices for calculating the SV and CO are based on the analysis of arterial pulse waveform, and require periodic, and in advance calibrations. An additional limitation of the method is that it assumes a constant rate of systemic vascular resistance [34]. So far, the methods of arterial pulse wave analysis have not been studied in neonates.

Invasive methods for assessing the CO have been developed in adults, and then applied in sick neonates, where their use is limited. Limitations of these methods are the invasiveness, complexity, and relatively long process (need for central vascular approach and taking sequential blood samples for laboratory analysis), for which reason they are seldom used in practice. Invasive methods for estimating average CO over time are based on a few physical principles. Fick's law is the law of mass/mass flow conservation. The amount of oxygen in the pulmonary artery and the amount of oxygen in the capillaries, flowing from the alveoli, is equal to the concentration of oxygen in the pulmonary vein [35].

Clinically, the O_2 consumption can be measured by measuring the concentration of oxygen in the inhaled and exhaled air and pulmonary ventilation. Consumption is estimated as the difference between the amount of oxygen in the inhaled and exhaled air. The concentration of oxygen in the peripheral arteries is the same as in the pulmonary veins. Pulmonary arteries have mixed venous blood. Samples for the analysis of O_2 are obtained from the pulmonary artery or the right ventricle through the cardiac catheter. Thus, we can calculate the CO (CO = O_2 consumption/AVΔO_2). Neonates compensate the reduced release of oxygen from fetal hemoglobin in tissues with higher hemoglobin concentrations, a larger volume of blood per unit of body weight, and increased CO. Instead of measuring O_2 consumption, we can measure the formation of CO_2 using capnography and assume that it is equal to the exchange of CO_2 in

the lungs [36]. In neonates with a healthy alveolar-capillary membrane, the partial pressure of CO_2 in arterial blood is equal to the partial concentration of CO_2 in the exhaled air [37].

Stewart principle is a method of dilution of the indicator and provides calculation of the CO on the basis of change in the concentration of the dye (lithium-based dilution devices) or a change in the temperature of the solution (thermodilution-based devices). We inject a known amount/temperature of a substance proximally and measure its concentration/temperature distally. We calculate the total flow rate ($Q = m/\int cdt$) from the time profile (integral) curve and a known quantity of the injected substance [38]. Clinically, a known amount of dye or isotope is injected rapidly into a large central vein, or the right side of the heart. We measure the concentration of dye or isotope in arterial blood: the larger the CO the greater the dilution. The most common indicator is a small volume of cold saline; we calculate flow from the temperature change. The average blood flow through an organ can also be calculated from the change in the volume of the hollow body in time: dV/dt (volume of a cavity is imaged within a specified time sequence: by ultrasound, magnetic resonance, X-ray).

4.3. Arterial blood pressure

Arterial blood pressure in the newborn can be measured noninvasively using appropriate cuffs or through the invasive arterial catheters. In principle, the matching of both methods is good [39]. We currently do not know what the normal arterial blood pressure is for a given gestational and postnatal age. Also, we do not know what the value of blood pressure is when the blood flow to vital organs is diminished and the auto-regulatory mechanisms in the brain fail. Thus, neonatal hypotension is not precisely defined. In addition, there is currently no evidence that the treatment of hypotension has significant impact on the clinical outcome in neonates [40–42].

Global perfusion pressure is measured via invasive arterial blood pressure monitoring. However, "adequate" blood pressure does not signify an "adequate" CO; therefore, an increase in blood pressure does not necessary mean an increment of CO. However, without measuring CO we can only assume that adequate mean blood pressure means also adequate CO, which is not always true. Second, the increase in blood pressure does not always mean the elevation of CO (failing myocardium poorly responds to a high vascular resistance). In neonates, the threshold heart rates and perfusion pressure are different depending on gestational and postmenstrual age of premature neonates [42, 43].

5. The peripheral hemodynamic monitoring

Hemodynamic failure results in low cardiac output syndrome with inadequacy of oxygen and nutrients delivered to peripheral tissues and cells. Clinically, peripheral perfusion of organs can be assessed by observing the skin color of a neonate. The method is vastly subjective. A laboratory method of defining the peripheral perfusion is the measurement of the concentration of lactic acid in the peripheral blood. In case of lowered peripheral perfusion tissue hypoxia occurs, which leads to anaerobic metabolism and lactic acid formation. Lactic acid is present in the less perfused tissues and therefore it is primarily not present centrally

in systemic circulation; therefore, lactic acidosis (>2.5 mmol/L) is a late sign of low cardiac output syndrome. Consequently, the concentration of lactic acid in the systemic blood rises after the reanimation. Moreover, the production of lactic acid is increased in the case of adrenaline treatment, which increases glycogenolysis and glycolysis in the liver [44]. The plasma concentration of the lactic acid is associated with the stage of the disease and a higher mortality in neonates [45].

Noninvasively, the peripheral perfusion may be assessed by the perfusion index [46]. It is based on an analysis of the pulse oximetry signal. The perfusion index is the ratio between pulsatile and non-pulsatile pulse oximetry signal (AC/DC × 100). It reflects the difference between the amount of blood in the tissue between systole and diastole. The perfusion index correlates well with the capillary refill time, the surface-core body temperature difference [47], and also the severity of the disease in neonates [48].

Assessing the function of peripheral organs is another way of assessing the peripheral hemodynamics. Using amplitude-integrated electroencephalogram (aEEG), we assess the brain function. Observing respiratory rate and pattern indicates the lung function and myocardial contractility indicates the heart function. By measuring the urine output and cleared substances, we assess the renal function. Measurement of diuresis in the neonate for hemodynamic monitoring has many caveats, especially in premature neonates, since immature renal tubules are not able to concentrate the urine. Moreover, the accurate monitoring of diuresis is an invasive method. Laboratory measurements of liver enzymes, clotting factors, and ammonia concentration assess liver function, and measurements of muscle enzyme assess the muscle function [49].

The peripheral monitors enable measuring the micro-vascular blood flow to the peripheral organs by perfusion index and laser-Doppler method and regional tissue oxygenation by NIRS.

5.1. The regional blood flow monitoring in the peripheral organs in the neonate

Regional blood flow is a complex and dynamic variable that changes depending on the functional activity of the body. In pathological conditions, like, for example, a sudden change in blood pressure or hypoxia, blood flow through the body can change very quickly. Like CO also the regional blood flow is measured in mL/min and expressed either normalized by body weight either by 100 g of tissue, or as a percentage of the CO. Assessing and measuring the blood flow is not a part of the permanent clinical practice in neonatal intensive care units, because the methods are generally complicated, inaccurate, invasive, and expensive, and currently their use is not showing clinical welfare for sick newborns.

With the help of the abovementioned physical principles and listed methods for CO monitoring, it is also possible to estimate the regional blood flow in peripheral organs. Using Fick's law, we can calculate blood flow through the peripheral organ, knowing its oxygen consumption. By applying this principle, several methods for the evaluation of blood flow in the brain—through the inhalation of 15% nitrous oxide [50] and the clearance of a radioactive

isotope of xenon [51]—have been developed. Regional blood flow to peripheral organs can be measured using Doppler ultrasound [52]. The method is based on measuring the blood flow velocity through the blood vessel. The product of the flow rate and maximum vessel diameter allows us to evaluate blood flow in the vessel, and if the vessel supplies the oxygen and nutrients to the organ we can estimate the blood flow to the organ. Blood flow to the brain can also be measured using magnetic resonance imaging (MRI) techniques. Blood flow to the brain is supplied by four arteries in the neck; hence, it can be evaluated using a scan through the arteries: the cross section of the vessels is multiplied by the blood flow velocity. The blood flow velocity is measured by the loss of magnetization caused by the fresh blood flowing into the plane of imaging (imaging with phase contrast) [28]. For quantitative measurement of the flow, a contrast agent containing gadolinium can be used [53]. The blood flow in each blood vessel can be measured with an electromagnetic flowmeter: the moving particles-ions are deflected to right angles to the direction of movement in an electric field and by that deflection the electric current is measured. Blood flow can also be measured by thermodilution method: the faster the blood flow, the cooler the tip of the heated sensor. Using the plethysmography, the displaced blood volume from the vessels is measured [38].

Frequently used noninvasive and continuous method for the indirect assessment of blood flow in various organs is the spectroscopy in the near-infrared spectrum [54–56]. The method is based on the principle of different absorption patterns of oxyhemoglobin and deoxyhemoglobin. It measures the index of tissue oxygenation and enables the calculation of the extraction of oxygen of the tissues in the target organ. If we assume that changes in the regional tissue oxygenation are not accompanied by changes in the arterial blood oxygen saturation (SaO_2), oxygen consumption, the amount of blood in the arteries and veins, and hemoglobin concentration, then the measured tissue oxygenation can be used to assess organ perfusion. NIRS optodes can be placed practically anywhere on the surface of the body, they measure tissue oxygenation approximately 4 cm below the surface (in the brain, kidneys, intestines, liver, and muscle) [57].

5.2. Microcirculation monitoring in the neonate

Microcirculation encompasses arterioles, capillaries, arteriolar-venous connections, venules, and lymphatic vessels. It supplies the target organs with oxygen and nutrients. Capillary filling or opening depends on the tone of arterioles and precapillary sphincters, whose diameter changes in parallel to the contractions/relaxations of smooth muscle in the vessel wall. The humoral factors, endothelial vasodilators (NO, CO, and H_2S), vasoconstrictors (epinephrine, norepinephrine, and endothelin-1), myogenic mechanism, local metabolites, and the parasympathetic nervous system affect the tone of smooth muscles. Blood flow in the microcirculation reflects the function of the central elements of the blood flow and is thus the ultimate indicator of cardiovascular efficiency. Microcirculation in individual organs also operates globally and represents one of the largest virtual organs in the body. In adults, it is estimated that 5% of the total blood volume is located in the capillaries, which may increase its capacity to fourfold [58].

Microcirculation in the newborn can be assessed using a variety of methods, the most commonly used is the evaluation of flow by laser-Doppler method, video microscopy (dynamic capillaroscopy), and xenon clearance techniques [59]. Flow measurement by laser-Doppler method is based on the fact that the frequency of the light beam, which passes through the tissue, changes as a result of reflection from the moving parts—red blood cells (the Doppler effect). The flow is proportional to the concentration and speed of moving red blood cells in the microcirculation. Since the flow in microcirculation is highly variable, we usually assess microcirculatory response to some of the challenge tests and monitor the dynamics of change. The most commonly used provocation methods are the postocclusive reactive hyperemia (hyperemia after a transitional cuffing of the proximal artery), thermal methods (local heating or cooling), and iontophoresis of vasoactive substances [60, 61]. In the first days after birth, the blood flow is very fragile and the peripheral blood flow in the microcirculation is unstable. The myogenic and nervous controls of skin blood flow enable thermoregulation. The blood flow to the skin in the first days after birth is reduced. The blood flow is related to gestational and postnatal age and the incidence of morbidity and cardiovascular function [62]. The deterioration of peripheral blood flow regulation in microcirculation causes vasodilation and decreased peripheral vascular resistance and contributes to the vulnerability of blood flow. The peripheral blood flow in the first days after birth differs in boys and girls; boys have stronger vasodilation; the mechanism is possibly associated with an increased incidence of hypotension in newborn males [63].

6. Limitations

The described methods for hemodynamic monitoring of neonates have many limitations. The clinical ones are vastly subjective and do not correlate well with the laboratory methods. The continuous bedside noninvasive methods are less accurate and sometimes demand complex deducement to what is happening. There are no trials on resuscitation using the noninvasive methods. The more sophisticated noninvasive methods require expensive equipment and are time consuming, measuring the parameters in the moment of measurement and not continuously.

7. Conclusion

Hemodynamic monitoring, which was for a long time not available in neonates and prematures, is becoming an indispensable tool for understanding how cardiovascular system adapts to extrauterine life. This is especially important when treating the smallest premature with peculiar and very vulnerable hemodynamics. This article adds some of the latest information on hemodynamic monitoring in neonates with specific emphasis on the methods which are available, cost-effective, noninvasive, and easy to manage and understand.

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Respiratory Care of the Neonate

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Additional information is available at the end of the chapter

Abstract

The respiratory distress is a very common condition both in term and in preterm neonates and the most frequent reason for admission to the neonatal intensive care unit (NICU). The aetiology greatly depends on the maturation of neonate's organs and perinatal events. The clinical picture is sometimes scarce and very nonspecific for the etiologic determination. Treatment of neonatal RD begins first with the application of a mixture of oxygen and air, then with different modes of non-invasive respiratory support methods. Non-invasive respiratory support can be sustained with nasal continuous positive airway pressure, bi-level positive airway pressure and high-flow nasal cannula ventilation. Non-invasive ventilation with high-frequency oscillations through nasal cannula or masks is also possible with some respiratory devices. Non-invasive ventilation is usually combined with the application of natural surfactant and other therapeutic means, like methylxanthine therapy, prevention and closure of patent ductus arteriosus, and control of infection. In the case of non-invasive ventilation failure, different kinds of invasive ventilation methods are available and being practiced in NICUs. The invasive respiratory support can be maintained by controlled or intermittent mandatory ventilation combined with different supportive synchronous positive inspiratory ventilation, offered by modern respirators.

Keywords: neonate, respiration, ventilation, oxygenation, perfusion, evidence-based therapy, surfactant, respiratory distress

1. Introduction

The lung is the only organ during foetal development that remains dormant, although foetal breathing occurs; gas exchange is performed by the placenta, but the lung is completely ready for transition to extra-uterine life. Its development and maturation are precisely regulated

during foetal life and needed to accomplish readiness for extra-uterine exchange of gases. During labour, different obstetric and non-obstetric factors may affect lung inflation in transitional period to extra-uterine life.

Even mature neonates have smaller diameter of the airways in comparison to infants, children and also adults; their chest wall is more compliant and the lung volume at the end of expiration is the same as the closing volume. Therefore, their lungs are prone to collapse. Besides, the neonates have fewer alveoli with ventilation perfusion mismatch, but a two–three–fold increase in oxygen demand in comparison to adults [1]. They have relatively inefficient respiratory muscles due to lack of red respiratory muscles and more white ones, which get tired faster. Their pulmonary vascular wall contains more muscle fibres and is therefore more prone to vasoconstriction. The sudden falls in partial oxygen pressure result quickly after short hyperpnoea in hypopnoea or apnoea. The foetal haemoglobin binds oxygen with greater affinity than adult haemoglobin. Neonates also have immature immune system with lack of the acquired immunity against microorganisms and are thus more prone to infection. In a premature infant, all these differences in lung development are more prominent in comparison to the term neonate and further influence the transitional period of extra-uterine life.

Because of all the above-mentioned developmental immaturity of the lungs, heart, blood vessels, circulation and immune system, neonates are more prone to develop idiopathic respiratory distress (RD) at the beginning of the life but also in later days and weeks and these differences are even more pronounced in premature and very premature neonates. The aetiology and pathophysiology of the RD differ between mature and premature infants [2]. In the most extreme prematurity, several forms of RD syndrome or hyaline membrane disease (HMD) may develop because of lack of surfactant and underdevelopment of the lung.

RD is very common in neonates, affecting about 10% of them [3]. Some of them have disorders of transition from foetal to extra-uterine life; others have RD caused by congenital or acquired infection or congenital malformations of different organs (thoracic and extra-thoracic). Regarding perinatal and labour history, gestational age and appropriateness of birth measures, the aetiology of the neonatal RD could be suspected. Clinical picture of neonatal RD is rather nonspecific in regard to the aetiology of it and also the management is quite universal. RD may develop immediately and acutely after birth or more slowly in the next few hours depending on the cause of neonatal breathing difficulties [4].

According to the situation in the delivery room, we have to decide how to approach the neonate with breathing difficulties. A well-equipped and trained neonatology team should be available during all difficult or premature deliveries which have to follow recommended resuscitation care of the neonate. The first golden minutes are the most important not only to properly recognize a neonate who needs our support but also to apply appropriate, sufficient and not too aggressive support if not needed. Appropriate inspiratory pressure and oxygen therapy during artificial ventilation in the delivery room may prevent immediate and later complications especially in the most vulnerable extreme premature infants. Prevention of hypothermia, in premature neonates with plastic bag, is also one of the important preventable methods because it may prevent severe metabolic derangements due to hypothermia, harmful for all organs [5].

2. Methods

We conducted electronic searches of articles on respiratory management and care of neonates with RD, using key terms: neonate, respiration, ventilation, oxygenation, oxygen, evidence based therapy, caffeine, surfactant, respiratory distress in the PubMed data base from the years 2000 to 2017 and reported the most relevant ones. Also, consensus guidelines on neonates with RD were reviewed [6]. The article describes transition from intra- to extra-uterine life, lists the methods for assessment of neonates with RD, describes the respiratory support in the delivery room, treatment with methylxantines, oxygen and surfactant, and the non-invasive and invasive tools for artificial ventilation.

This chapter includes the impact of optimal ventilation, tissue oxygenation and perfusion on the non-invasive and invasive respiratory management of neonates with RD. Also, the short- and long-term outcome in respect of respiratory management of neonates in the neonatal intensive care unit (NICU) is addressed.

3. Transition from foetal to extra-uterine life

The transition from foetal to extra-uterine life is a process of rapid physiologic changes in the neonate that begin in utero as the foetus prepares for transition from intrauterine placental support to extra-uterine self-maintenance. The process can last for up to 12 h after birth and depends on the gestational age, placenta, maternal health and congenital anomalies of the neonate. The foetus is preparing to the transition by foetal breathing, producing surfactant storing glycogen in the liver, producing catecholamines and depositing brown fat. After the umbilical cord is cut, placenta no longer does the work of lungs as they begin to exchange gasses. The first breath inflates the lungs and causes circulatory changes: the resistance to blood flow through lungs falls and more blood flows through pulmonary arteries to the lungs. Three foetal shunts close and this results in neonatal circulation. The three processes of the transition—(1) the replacement of amniotic fluid in the alveoli with air, (2) the onset of regular breathing and (3) the increase in pulmonary blood flow—may all be deranged soon after birth and RD ensues [7]. The failure of alveoli fluid clearance results in transient tachypnoea of the newborn (TTN) and causes in turn decreased lung compliance. The deficiency of surfactant mainly in preterm neonates or in term infants of diabetic mothers causes collapse of the alveoli and diffuse and progressive atelectasis that result in decreased lung compliance and small functional residual capacity (FRC) and cause HMD. The abnormal persistence of elevated pulmonary vascular resistance either due to underdevelopment, maldevelopment or maladaptation of pulmonary vessels results in persistent pulmonary hypertension of a newborn (PPHN). PPHN is often associated with congenital anomalies (i.e. congenital diaphragmatic hernia) or chronic intrauterine stress (meconium aspiration syndrome (MAS)).

At the time of birth, the umbilical cord is still pulsating. For term and preterm neonates, delayed cord clamping is recommended. The preterm neonates born before 32 weeks of gestation with delayed cord clamping had better neuro-developmental outcomes at the age of 18 months [8, 9]. Too early umbilical vein clamping, even before the first breath of the neonate,

may lead to a prolonged period of low cardiac output, which along with the undeveloped self-regulation system leads to a reduction of brain blood flow. Delayed cord clamping allows blood to enter the neonate's circulation and by that enhances the performance of the left ventricle, which is the most important for the normal cardiac output and stable haemodynamics especially in foetal distress with compromised haemodynamics at birth [10].

4. Assessment of the respiratory distress of the neonate

The evaluation of a neonate with RD is based on clinical, laboratory and radiologic investigations. In clinical evaluation, the respiratory effort, breathing efficacy and the breathing effect on other organs are assessed (**Table 1**). Signs of respiratory effort may be less pronounced or not visible in three cases: (1) the neonate, who has had RD for a long time, eventually becomes tired and signs of respiratory effort are reduced. Tiredness is a pre-terminal sign of respiratory failure. (2) A neonate with central nervous depression because of either intoxication, or encephalopathy, brain malformation, or increased intracranial pressure, breathes insufficiently without increased respiratory effort. These neonates breathe insufficiently due to reduced breathing impulse. (3) A neonate with neuromuscular disease may have respiratory failure without a significantly seen respiratory effort.

Classic laboratory signs of respiratory failure include acidosis (pH < 7.25), hypoxia (PaO_2 < 50–60 mm Hg (6.7–8 kPa), FiO_2 0.6–0.8) and hypercapnia ($PaCO_2$ > 60 mm Hg (8 kPa)) which are late signs of RD. Radiologically, neonatal chest may be scanned by X-rays [11, 12] or, lately more in use, by ultrasound (US) waves [13–17]. In utero, routine foetal US scan following development and screening for malformations may reveal intra- or extra-thoracic malformations, better differentiated by magnetic resonance imaging (MRI). Combining the risk and etiologic factors for neonatal RD, gestational age and radiologic investigations, the pulmonary disease causing the RD may be diagnosed (**Table 2**), since the clinical picture and laboratory signs are almost not of any help in defining the aetiology of neonatal RD.

	Respiratory effort	Breathing efficacy	Effect on other organs
1.	Breathing rate	Chest movement	Heart rate
2.	Intercostal, jugular, supraclavicular or subcostal retractions	Chest auscultation	Skin colour: cyanosis, paleness, mottled skin
3.	Auscultatory phenomena	Arterial blood oxygen saturation	Disturbance of consciousness
4.	Feeding difficulties		
5.	Expiratory grunting		
6.	Use of auxiliary respiratory muscles		
7.	Nasal flaring, head nodding		
8.	Gasping		

Table 1. The clinical assessment of airway and breathing in a neonate.

Pulmonary disease	Gestational age, risk factors	Aetiology	Roentgenogram	Ultrasound
Hyaline membrane disease	• Preterm • Neonates of diabetic mothers	Lack of surfactant	Diffuse reticulogranular ground glass pattern with air bronchograms and small-lung volume; grading from mild (grade 1) to the most severe pattern (grade 4)	Coalescent B-lines-'white lung', thickened and irregular pleural line
Transitional tachypnoea of the newborn	• Late preterm • Elective caesarean section	Failure of adequate alveoli fluid clearance at birth	Bilateral perihilar linear streaking, patchy infiltrates	Compact B-lines in the inferior and less compact in the superior fields (double lung point) or bilateral numerous noncompact B-lines and normal pleural line and pleural sliding
Persistent pulmonary hypertension	• Term > preterm • Perinatal depression	Persistence of elevated pulmonary vascular resistance	Clear lung fields with decreased pulmonary vascularity	Echocardiography
Meconium aspiration syndrome	• Term • Perinatal depression	Meconium-stained amniotic fluid	Patchy infiltrates	Pulmonary consolidation with air bronchograms, pleural line anomalies, pleural effusion, B-lines in non-consolidation area
Pneumonia	Infection of the mother	Bacterial or viral infection	Non-symmetric bilateral patchy infiltrates	Subpleural consolidation
Pneumothorax	Preterm > term	Air entrance into the pleural space	Edge of collapsed lung	Absence of pleura sliding and B-lines, subpleural consolidation
Congenital heart defect	Term > preterm		May be diagnostic	Echocardiography diagnostic
Congenital malformation of organs	Term>preterm		Diagnostic	
Metabolic, neuromuscular diseases	Term > preterm		Normal	
Genetic defect of surfactant proteins	Term, preterm	lack of surfactant proteins	Diffuse reticulogranular ground glass pattern with air bronchograms and small lung volume; grading from mild (grade 1) to most severe (grade 4) Chronic lung disease of different patterns	

Table 2. The etiologic and radiologic assessment of respiratory state of a neonate.

5. Respiratory support in delivery room

Routine care in delivery room depends on whether we expect and take care of extreme premature neonates or near term or term neonates with breathing difficulties. In case of near term or term neonates, care starts with providing warmth, clearing airway if necessary, drying and stimulating the neonate. Care of preterm infants is different because we immediately place him/her into a heated polyethylene bag with small opening for the nose and mouth and further stabilize him/her under the heater to assure normal body temperature, prevent hypothermia and desiccation [18]. The stabilization of the neonate with RD in the delivery room comprises proper head positioning with wiping of the mouth, and rarely, in the case of more secretion, not removed by wiping, we may perform gentle suction of the neonate's mouth, then nose. We have to avoid the deep insertion of the catheter and vigorous suctioning of mouth which may cause reflex bradycardia. In the case of meconium staining and aspiration, suction of trachea under visual inspection with laryngoscopy guidance is needed first and then intubation and further washing out meconium from the trachea. In case of apnoea, gasping or bradycardia of <100 beats/minute, it is necessary to ventilate neonate's lungs with positive pressure ventilation considering the use of lowest effective inspiratory pressures and volumes to prevent damage to the lungs (barotrauma and volutrauma). Ventilation should fill neonatal lung with a gas mixture of air and oxygen. To prevent and reduce the oxidative stress, caused by excessive use of oxygen in the inspired air, it is necessary to be careful with the use of oxygen. For measuring the arterial oxygen saturation (SpO_2) in the peripheral blood, the pulse oximeter should be attached to the right wrist [19]. Latest guidelines recommend starting ventilation of term neonates with FiO_2 0.21 and later increasing the FiO_2 according to the value of the measured SpO_2. Ventilation of preterm neonates should be started with FiO_2 of 0.21, gradually rising the FiO_2 in accordance with the measured value of SpO_2. Ventilation of the extremely premature infants (born before 28 weeks gestation) should be started with FiO_2 0.30, while of very premature infants (28–31 weeks of gestation) with the FiO_2 from 0.21 to 0.30. If neonate is spontaneously breathing, the constant positive pressure is applied for the stabilization of respiration (continuous positive airway pressure) of 6 cm of H_2O through a mask, nasal tubes or endotracheal tube using neonatal respirator (neonatal resuscitator; T-piece device), intended for stabilization of the neonate in the delivery room. The maximum positive inspiratory pressure should not exceed 20–25 cm H_2O, which is used only in the case of apnoea or bradycardia [6]. No differences in mortality and morbidity of premature neonates have been demonstrated in comparing resuscitation starting with low (FiO_2 ≤0.30) or high levels of oxygen (FiO_2 ≥0.6) [20]. Currently, there is insufficient evidence of sustained lung inflation efficacy and safety in the cardiopulmonary resuscitation and stabilization of the neonate in the delivery room [21, 22].

6. Methylxanthines

Methylxanthines stimulate the respiratory centre to increase its responsiveness to the partial pressure of carbon dioxide in the blood and reduce respiratory depression by hypoxia. They also improve respiratory muscle strength. Therapy with caffeine has proven to reduce the

duration of artificial ventilation and oxygen demand and decrease risk for bronchopulmonary dysplasia (BPD) and the need for patent ductus arteriosus (PDA) ligation [23, 24]. At the age of 2 years, positive effects on cognitive development were observed, but in the same group of children at the age of 5 years they were no longer detected [25]. In a premature neonate with RD, the less invasive artificial ventilation and surfactant therapy are usually combined with early prophylactic intravenous administration of caffeine to achieve the highest level of respiratory support in the least invasive form [26, 27]. Since methylxanthines affect the diaphragmatic activity and increase the tidal volume, we may use them to increase the muscle strength in floppy neonates [28].

7. Treatment with oxygen

Oxygen is necessary for aerobic metabolic processes in the body. The excess of oxygen is detrimental to neonates, particularly the premature infants with immature antioxidant, anti-inflammatory mechanisms and a greater amount of free iron. Hyperoxia affects not only the lung but also other organs, with the greatest effects to central nervous system (convulsions) and eyes (retinopathy of prematurity (ROP)). In comparison with a 'higher' level of SpO_2 (91–95%), the 'lower' level (85–89%) has been shown to diminish the risk of ROP and BPD, but unfortunately at the same time increase mortality, the incidence of necrotizing enterocolitis and poor neuro-developmental outcome [29]. Based on research, current recommendations propose the SpO_2 of preterm infants who require oxygen therapy to be between 90 and 94%, setting alarm limits to 89 and 95% [6]. The SpO_2 of term neonates who require oxygen therapy should be above 92%.

Hypercapnia is associated with acidosis and compromised cardiovascular function, while hypocapnia decreases cerebral blood flow. There is some conflicting evidence on higher $PaCO_2$ levels and the impact on mortality, severe intraventricular haemorrhage (IVH), BPD, ROP and neurodevelopmental outcome [30, 31]. Therefore, the optimal target carbon dioxide levels are not established; based on available data, it should be between 46 in 60 mm Hg (6.1–8 kPa) for ventilated neonates.

Blood gas is monitored in arterial samples, so an indwelling arterial line is necessary in taking care of a neonate with moderate or severe RD. Venous and capillary samples are not appropriate for PaO_2 measurements. They may be of use for $PaCO_2$ monitoring, although they slightly overestimate it, and pH monitoring, although they slightly underestimate it.

8. Surfactant

Pulmonary surfactant, a macromolecular lipoprotein complex, secreted by the alveolar epithelial cells type II, reduces the surface tension in the pulmonary alveoli at the end of exhalation. Sufficient amount of surfactant in the mature lungs prevents complete collapse of the lungs at the end of exhalation. A part of the inhaled air remains to be 'trapped' in the pulmonary alveoli, what is called the FRC. In each subsequent breath, it is not necessary to re-open

the lungs from the total collapse, which greatly reduces the work of breathing and with that fatigue and respiratory failure. A thin layer of a surfactant in the walls of the pulmonary alveoli at the end of each inspiration is not completely waterproof, some liquid passes through the pores being in contact with the air in the lung alveoli, increasing the surface tension and preventing the overdistension of the alveoli at the end of inspiration. The lung surfactant is rapidly adsorbed and easily distributed in the form of a thin film on the surface between the liquid layer and the air in the lung alveoli [32].

Several animal surfactants of bovine or porcine origin are used in Europe (**Table 3**).

Currently, a double-blind study of a synthetic surfactant, CHF5633, with the same effect in the treatment of RD as poractant alpha, but a stronger anti-inflammatory and a more favourable effect on the cerebral haemodynamics, is being conducted [33].

Early publications recommended the application of surfactant in developed RD as a 'rescue' or therapeutic administration or prophylactically for the prevention of RD in very premature infants in the first few minutes after birth. Criticism of the prophylactic administration of surfactant was that likely 27–60% of preterm infants receive surfactant unnecessarily [34, 35]. Recommendation nowadays is to stabilize the respiration of a spontaneously breathing neonate by using CPAP and early selective surfactant administration. When an endotracheal intubation is needed due to progressive RD, the neonate should obtain surfactant as soon as possible [6, 36].

Another way of avoiding lung barotrauma and especially volutrauma, techniques to shorten the duration of artificial ventilation or even completely avoiding it, has been developed in recent years. The first of such methods of fast and non-invasive surfactant application was **IN**tubate—**SUR**factant—**E**xtubate (INSURE). The extubation was followed by non-invasive respiratory support [37, 38]. In comparing early INSURE method with CPAP without the administration of the surfactant, certain advantages were found in the INSURE group. Not enough evidence was found to conclude that one of the two methods is better than the other [39].

Type of surfactant	Company name	Origin of surfactant	Lipids (%)	Proteins (%)	Dose (body mass)
Poractant alfa	Curosurf®	Extraction from porcine lungs	99	SP-B, SP-C 1%	100–200 mg/kg (1.25–2.5 ml/kg) in suspension
Bovactant	Alveofact®	Lavage from bovine lungs	99	SP-B, SP-C 1%	50 mg/kg (1.2 ml/kg) in suspension
Beractant	Survanta®	Extraction from homogenized bovine lungs	88–90	SP-B, SP-C 1%	100 mg/kg (4 ml/kg) diluted before use

Table 3. Surfactants, registered for therapy of RD in Europe.

Less **I**nvasive **S**urfactant **A**dministration (LISA) and **M**inimally **I**nvasive **S**urfactant **T**reatment (MIST) are other two less invasive methods of surfactant application. With LISA, the preterm neonate who is supported by CPAP via nasal prongs, the larynx is opened by laryngoscope and by the Magill forceps a thin and soft aspiration tube is inserted below the level of the vocal cords, and then the surfactant is applied in two to four aliquots while the neonate is spontaneously breathing. With MIST, the tube for surfactant application is somewhat stiffer and bent so it can be inserted into the larynx without the Magill forceps. According to the authors, MIST is an easier method for less skilled doctors [40]. To confirm the hypothesis that less invasive surfactant administration combined with the respiratory support by CPAP is more successful in neonates who have some natural surfactant and not effective in those who have too little of their own surfactant, a blind, multicentre, randomized study in premature neonates between 25 and ≤28 weeks gestation, requiring CPAP and a low percentage of oxygen in the inhaled air (FiO_2 from 0.30 to 0.45) during the first 6 h after birth, is currently underway. The research group is being treated with poractant alpha at a dose of 200 mg/kg body weight, and the control group receives placebo [41]. None of the above-described methods can avoid laryngoscopy.

The methods of surfactant aerosolization have up to date been more or less unsuccessful. With the **C**atheter **A**nd **L**aryngeal **M**ask **E**ndotracheal **S**urfactant **T**herapy (CALMEST) the surfactant is administered by a catheter and the laryngeal mask [42].

The oxygen requirement higher than the $FiO_2 > 0.3$ after 2 h of breathing with CPAP has a high positive-predictive value of a respiratory failure at 6 h after birth. Therefore, the recent recommendations suggest an early less invasive surfactant administration before the neonate requires high proportions of oxygen in the inspired air [43]. If no improvement is seen after the first dose, the surfactant application is repeated for the second or third time. In this case, the poractant alpha at a dose of 200 mg/kg body weight is supposed to have better effect as a lower dose of poractant alpha or beractant (100 mg/kg body weight).

Late-preterm and term neonates besides rarely having a primary surfactant deficiency due to genetic defect of surfactant proteins, they more often suffer from secondary surfactant deficiency in conjunction with MAS, pneumonia and pulmonary haemorrhage. In those cases small studies have shown improved oxygenation, gas exchange and a reduced need for extracorporeal membrane oxygenation (ECMO) [44].

9. Non-invasive respiratory support

The best and most frequently used treatment of neonatal RD nowadays is CPAP through nasal spouts (nasal mask, nasal cannula and nasal tube) with the addition of the interfaces by using various physical processes to insufflate and exhale the mixture of air and oxygen into and out of the respiratory tract of a neonate [45]. It has been proven to reduce side effects that neonates could suffer if they were ventilated by the invasive methods of artificial ventilation. Until now, a variety of techniques applying positive pressure of constant pressure

(CPAP), the intermittent insufflation of positive pressure (nasal intermittent positive pressure ventilation, NIPPV), which can be time determined or synchronous triggered by inhalation (synchronous nasal intermittent positive pressure ventilation (SNIPPV)) and ventilation at two levels of positive pressure (bi-level positive pressure ventilation, Bi-Level) or even with high-frequency oscillations (high-frequency oscillation ventilation (HFOV)) have been developed. Different randomized studies have explored the advantages or disadvantages of one method of non-invasive ventilatory support over the other. In comparing the non-invasive ventilation with NIPPV to the nasal CPAP, fewer respiratory failures and the need for intubation in the NIPPV group were found [46]. Meta-analysis of the use of different devices and interfaces for CPAP has elucidated differences in outcome depending on the use of nasal adapters or interfaces, requiring further research [47]. Similarly, there is an open question whether breathing with the help of bi-level CPAP is better than breathing with CPAP alone and does it pose an advantage of better exhaling CO_2, better oxygenation or other physiological indicators [48].

If higher mean inspiratory pressures are required for the lungs to remain inflated, the potential non-invasive ventilation using HFO via nasal tubes or cannulas may be used since even long-term studies have confirmed advantages of HFO non-invasive ventilation over other invasive ventilation methods.

Ventilation of neonates using high-flow rates (high-flow nasal cannula (HFNC)) has some advantages over CPAP due to less damage to the nose and nasal septa and less pneumothorax (PTX) [49]. A multicentre study being conducted in nine centres in Australia and Norway might give answers as to which breathing support is better in very preterm infants, CPAP or HFNC [50].

Nose requires special attention because the prolonged nasal respiratory therapy may cause decubitus and malformations of the nose. Regular changes of devices and protection of the nose skin and mucosa with skin-protective strips and/or creams prevent those problems. Gastric distension has to be prevented by an opened nasogastric tube and regular checking of gastric over distension which decrease compromise of diaphragm contractions. Neonatal care in term neutral environment incubators or warm beds and preventive positions like Cocoonababy® Nest or similar home-made products besides frequent changes of neonate's positions improve ventilation during the period of acute respiratory problems. Kangaroo care is a useful method to improve bonding between the neonate and the mother or father but has to be carried out cautiously during non-invasive ventilation [51]. During kangaroo care, observations have to be made whether apnoeic spells are more frequent and whether bradycardia occurs.

Non-invasive respiratory ventilation enables non-aggressive approaches, without sedation, analgesia, tracheal intubation and mechanical ventilation. Complications of non-invasive ventilation are mainly pressure sores of skin around the nose, ulceration and necrosis of the septum, much less likely hyperinflation of the lungs, restlessness, PTX, stomach distension or food intolerance. Non-invasive ventilation failure may be predicted by the use of neonatal chest US [52].

10. Invasive artificial ventilation of the neonate

Nowadays, the invasive artificial ventilation of the neonate represents a continuation of treatment in cases where non-invasive ventilation with or without the use of surfactant is not possible or successful. In invasive mechanical support ventilation with a respirator, CPAP is usually supplied in combination with intermittent mandatory or synchronized artificial ventilation (i.e. intermittent mandatory ventilation (IMV); synchronized intermittent mandatory ventilation (SIMV)). Ventilation can be sustained at two different positive pressure levels (variable/bi-level positive airway pressure, bi-level (BiPAP)). Other forms of artificial ventilation include ventilation by releasing the pressure (airway pressure release ventilation (APRV)), neuronal-mediated respiratory support (neurally adjusted ventilatory assist (NAVA)), and so on. In the case of the artificial ventilation, one should always set the concentration of a mixture of the inspired oxygen and air, the frequency of the ventilation, the ratio of duration of the inspiration and expiration or time of inspiration, the end-inspiratory pressure or the tidal volume and the end-expiratory pressure. Each respirator is equipped with the heater and humidifier in order that neonates breathe moist and warm mixture of air and oxygen. In cases of severe pulmonary disease with severe RD, high-frequency oscillating ventilators, which use very low inspiratory volumes that do not damage the lung tissue, may be used.

High-frequency oscillation ventilation is a method of artificial ventilation, which in cases of severe RD can be the least harmful way of ensuring good oxygenation and due to the active exhalation wash out carbon dioxide as well. High-frequency oscillation ventilation will only be successful if the pulmonary alveoli are optimally opened prior to the start of oscillations. In HFO, the optimization of lung volume is achieved by small increments of continuous positive distending airway pressure and the pressure in lung alveoli. Gradually, the increase of continuous positive distending airway pressure leads to opening of the small, collapsed non-ventilated lung and by that to the increment of the FRC of the lungs that ensure good ventilation and oxygenation. Consequently, the optimally opened lung tissue is then oscillated with very small tidal gas volume (order of the neonate's dead space of the lungs), which regulates the exhalation of carbon dioxide from the lungs. Oscillations with small volumes are less harmful for the delicate lung parenchyma and do not damage it, thereby preventing the secondary injury such as barotrauma and volutrauma. When lung function improves, the neonate's ventilation can be switched to conventional artificial ventilation again [53].

The most important factor in invasive artificial ventilation of the neonate is to prevent the lung over-distension, because it injures the delicate lung tissue and causes air leakage outside the airway with the development of pulmonary interstitial emphysema (PIE), PTX or pneumomediastinum (PM) and other even more severe forms the air leakage into the chest. When lungs become more compliant, the pressure-guided artificial ventilation may lead to lung over-distension, therefore many of the neonatal respirators are programmed to the restriction of tidal volume (volume-targeted ventilation; volume-guaranteed ventilation (VGV)). Care should be taken not to cause hypocapnia during ventilation since it decreases the brain blood flow and causes periventricular leukomalacia (PVL) and IVH which jeopardize the neurological development. The volume-targeted artificial ventilation was shown to shorten the duration

of artificial ventilation and hypocapnia, lessen the incidence of BPD, IVH grade III/IV, PTX and PVL in comparison to the pressure-controlled ventilation in preterm neonates. The mortality rate was unaffected by the mode of artificial ventilation [54].

During intubation and invasive ventilation, neonates are prone to cardio- and cerebrovascular instability. The intensive invasive therapy subjects neonates to more infections and the invasive ventilation to volu-, barotrauma and shear stress. Common complications due to intubations and invasive ventilation are hoarseness, aphonia, tracheal stenosis, and feeding and perioral sensation disorders.

11. Additional supportive therapies for neonatal respiratory distress

For treating pulmonary hypertension in different pulmonary diseases of the neonate the inhaled nitric oxide, pulmonary artery dilator, has been shown to have some beneficial effects [55]. On the other hand it has not been shown to be beneficial in preterm neonates with RD in reducing BPD or mortality [56]; though most NICUs are using it nowadays when hypoxic respiratory failure in extreme premature cannot be solved by other means [57].

The ECMO pumps the blood through an artificial lung back into the bloodstream, providing heart-lung bypass support outside of the neonatal body. ECMO is applied in neonates with severe RD due to congenital diaphragmatic hernia (CDH), meconium aspiration syndrome (MAS), pneumonia, air leak problems or PPHN [58]. Veno-venous ECMO is preferred to be used in infants with hypoxic respiratory failure unless an arterio-venous ECMO is needed due to combined cardio-respiratory failure. ECMO support should be used only in neonates weighing ≥2 kg of body mass.

General supportive care of a neonate with RD encompasses optimization of thermal neutral environment, fluid and nutritional management and a stable hemodynamic state ensuring adequate oxygenation and perfusion of neonatal organs.

12. Conclusion

RD of a neonate has almost identical clinical picture irrespective of many etiologic entities it originates from. The perinatal history, labour course, the gestational age and appropriateness of birth measures for the gestational age should all be taken into account in diagnosing the aetiology of the RD.

The modern management of neonatal RD is minimally invasive. In the delivery room, neonates are being stabilized. The respiratory support is primarily non-invasive ventilation as well as the surfactant is applied with less invasive methods not involving intubation and artificial ventilation. If intubation is required, the time of artificial ventilation should be as short as possible. Hyperoxia and hypocapnia should be avoided. Further studies will show whether such non-invasive treatment is also going to affect the incidence of BPD, neurodevelopmental outcome and other long-term consequences of intensive neonatal therapies.

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Neonatal Hypoglycemia

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Additional information is available at the end of the chapter

Abstract

Hypoglycemia is the most frequent metabolic abnormality in the newborn, but no consensus exists on what level of blood glucose is able to protect the brain and influence the child's neural development and which is the best course of management in cases labeled as hypoglycemia. Early diagnosis, urgent treatment, and prevention of future episodes of hypoglycemia are the cornerstones of management, now supported by recent advances in molecular genetics and in our understanding of the pathophysiology of neonatal hypoglycemia, particularly the pathogenesis of congenital hyperinsulinemic hypoglycemia.

Keywords: hypoglycemia, newborn, molecular mechanisms, hyperinsulinemia, actual treatment

1. Introduction

Hypoglycemia is the most frequent metabolic abnormality in the newborn, and although it is the most common biochemical disorder in this age group [30], it is still a source of clinical concern and controversy, as no consensus exists on what level of blood glucose is able to protect the brain and influence the child's neural development [6, 22, 53] and which is the best course of management in cases labeled as hypoglycemia. Early diagnosis, urgent treatment, and prevention of future episodes of hypoglycemia are the cornerstones of management, now supported by recent advances in molecular genetics [48] and in our understanding of the pathophysiology of neonatal hypoglycemia, particularly the pathogenesis of congenital hyperinsulinemic hypoglycemia [12].

Hypoglycemia occurs in 1.3–4.4 per 1000 full-term newborns and 15–55 per 1000 preterm newborns. This suggests that gestational age has enormous influence on its onset; in certain groups, adaptive mechanisms are not adequately developed, which predisposes them to increased risk of hypoglycemia. According to current evidence, the prevalence of hypoglycemia is approximately 10% in full-term neonates [45]; 6.5% in appropriate for gestational age (AGA), 8% in large for gestational age (LGA), and 15% in small for gestational age (SGA) newborns; and 15.5% in late-preterm infants [7].

2. Metabolic aspects

The maintenance of physiological concentrations of glucose in newborns plays important roles, including protecting the brain from damage caused by insufficient glucose intake and preventing the consequences of hyperosmolarity caused by high glucose concentrations. Although glucose is the preferred energy source of neurons, other sources, such as lactate and ketone bodies [23], seem to exert a neuroprotective effect. However, in hypoketotic states, such as hyperinsulinism or fatty acid oxidation disorders, ketone and lactate concentrations are not high enough to replace glucose, and the risk of a cerebral energy deficit is greater [60, 61].

It is known that, during fasting, several metabolic systems are activated to prevent hypoglycemia, which may be seen as a failure in one of these systems or as an abnormality affecting one or more of the hormones that control these systems [24].

As the brain mass of newborn infants in relation to body size is larger than that of adults, the rate of glucose utilization per kg body weight in newborns is two- to threefold than that of adults (4–6 mg/kg/min) [61].

2.1. Maternal provision of glucose to the fetus

The first half of pregnancy is characterized by marked anabolism. In early pregnancy, increased caloric intake not only supports fetal development but also facilitates fat deposition in the mother in preparation for the second half of pregnancy, a period of accelerated fetal growth in which maternal stores are mobilized to meet the needs of the fetus. To this end, increased insulin secretion also occurs in early pregnancy, as a way to store energy.

From the midpoint of pregnancy onward, high levels of circulating maternal insulin are also observed, but high levels of anti-insulin factors override this effect, ensuring the provision of nutrients to the fetus during the postprandial period. Thus, in pregnant women with preexisting diabetes, the effects of these anti-insulin factors are potentiated, causing excess provision of glucose and other energy sources to the fetus and thus triggering the abnormalities observed in infants born to diabetic mothers.

In the expectant mother, glucose is found at levels 25–30% higher than in the fetus, and it is transported to the fetus by concentration gradients and simple diffusion and through the action of transporters. In the fetus, the predominant transporter is GLUT-1, which has a high affinity for glucose and facilitates its passage through tissues [28].

Most glucose in the fetus undergoes oxidation to supply its energy needs, while another part contributes significantly to a buildup of glycogen, protein, and fat in triglyceride form. Glucose is the most important source of energy for the fetus and the major substrate for brain metabolism.

2.2. Glucose uptake in the newborn

At birth, the fetus becomes dependent on itself to obtain energy and meet the metabolic needs of its vital organs, particularly the central nervous system (CNS). Each mole of oxidized glucose provides 38 moles of adenosine triphosphate (ATP) [54].

Cerebral glucose transport takes place through a facilitated diffusion process, which is dependent on glycemia and is not regulated by insulin. Protection against hypoglycemia is coordinated by the autonomic nervous system by means of hormones that stimulate the production of glucose (through glycogenolysis and gluconeogenesis) and limit peripheral glucose utilization [54].

Glycogen is the only glucose storage medium in the body. Its deposits are found in the liver, striated muscle tissue (including cardiac muscle), kidneys, bowel, brain, and placenta.

The fetal liver contains a complete enzyme system for the synthesis and breakdown of glycogen, levels of which are low in early pregnancy but rise slowly and steadily from gestational weeks 15–20, before peaking in the third trimester. At this time, fat deposition also increases. Thus, part of the energy and substrates used for fetal growth is redirected for storage, which will play an important role in the peripartum and postpartum periods.

Hepatic glycogenolysis is the major mechanism of glucose release in the immediate neonatal period, which leads to depletion of glycogen stores. It is induced by an increase in glucagon and catecholamines and a reduction in insulin. This exhaustion of glycogen stores promotes activation of gluconeogenesis, which occurs largely as a result of free fatty acid oxidation in the liver.

Glucose homeostasis will thus depend on glucose intake; gluconeogenesis; glycogen, protein, and fat stores; and hormonal and neural factors.

Glucose produced from the breakdown of dietary lactose into galactose and glucose, for instance, is not taken up by the liver in the neonatal period; the newborn is thus dependent on hepatic gluconeogenesis to sustain glucose production.

Once glycogen stores are low, gluconeogenesis induced by glucagon, catecholamines, cortisol, and growth hormone mobilizes fat and protein substrates. Insulin, thyroid hormone, cortisol, and glucagon systematically promote induction of specific enzymes, thus adapting the neonate to the abrupt cessation of the supply glucose that was provided continuously before birth.

Upon clamping the umbilical cord, the maternal glucose supply, which was 54 mg/dL during pregnancy, ceases abruptly, and the neonate's blood glucose levels decline rapidly and precipitously—from a concentration similar to that of the mother to approximately 41 mg/dL within the first 6 h of life. Physiologically, glucose concentration decreases to approximately 30 mg/dL in the first 2 h after birth, subsequently rises, and plateaus at approximately 45 mg/dL 12 h after birth.

3. Definition

Current evidence is still unable to define a specific glucose concentration that is safe to prevent acute neurological damage or chronic, irreversible neurological injury in the neonate. Weight and gestational age, as well as the age at onset, severity, duration, and number of episodes of hypoglycemia, are all determinants of the blood glucose level most appropriate for protection of the neonatal brain [54]; thus, doubts persist as to whether any single level may represent a red flag for neurological safety.

A plasma glucose level below 30 mg/dL (1.65 mmol/L) in the first 2 h of life or below 45 mg/dL (2.5 mmol/L) after these first 2 h has been considered diagnostic of hypoglycemia [54].

Various situations can influence the appropriateness of a blood glucose level for use as a cutoff point for treatment initiation, including nutritional timing and the presence and absence of symptoms [64]. Thus, in 2011, the American Academy of Pediatrics proposed that neonatal hypoglycemia be defined as a blood glucose level of 2.5 mmol/L before routine feeding [1, 20]. Other studies suggest a limit of 2 mmol/L in asymptomatic newborns and 2.5 mmol/L in symptomatic neonates [42]. Although cutoff values below 2.6 mmol/L have been cited in various studies as defining of neonatal hypoglycemia, there is no guarantee that such a concentration is the most appropriate choice for establishing a diagnosis of this disorder and prompting initiation of treatment. An important finding reported by McKinlay et al. [34] has encouraged neonatologists to consider a glucose concentration >47 mg/dL as the level at which no impairment of appropriate neurological development was observed at age 2 years.

These proposed levels serve to provide a margin of safety until additional data are available to support a more accurate definition. However, the potential risk of neurologic sequelae has led many authors to consider blood glucose values <50 mg/dL in infants as the limit beyond which treatment should be instituted [61].

In practice, blood glucose levels below 50 mg/dL as measured by a glucometer should warrant careful monitoring, and plasma glucose levels below 45 mg/dL should prompt initiation of diagnostic measures and immediate treatment.

4. Etiology and pathogenesis

Overall, neonatal hypoglycemia is caused by one of the three main mechanisms: situations associated with hyperinsulinemia, situations associated with low or depleted glycogen stores, and situations associated with excessive glucose consumption. These mechanisms may also be compounded by the effects of certain drugs used in pregnancy.

4.1. Situations associated with hyperinsulinemia

4.1.1. Infant born to a mother with diabetes

Offspring of diabetic mothers may be abnormally large at birth (LGA), even when the mother was able to keep blood glucose within normal or near-normal range throughout pregnancy.

The risk of birth defects is two to four times higher in fetuses of pregnant women with diabetes, particularly when the disorder is poorly controlled during the period of fetal organ development (i.e., gestational weeks 6–7), and the neonatal mortality rate is fivefold than that of infants born to women without diabetes.

Intermittent maternal hyperglycemia causes fetal hyperglycemia, which, in turn, stimulates excess insulin production by the fetal pancreas. On the one hand, this increased fetal insulin synthesis stimulates excess organ growth (except of the brain and liver, which are not dependent on insulin supply for growth), thus causing fetal macrosomia. On the other hand, it is associated with a high incidence of neonatal hypoglycemia and marked lipolysis during the first few hours after birth. Hyperinsulinism and hyperglycemia may also cause fetal acidosis, which results in an increased rate of stillbirths. Although hyperinsulinemia is probably the leading cause of hypoglycemia, reduced epinephrine and glucagon responses can also be contributing factors. Levels of cortisol and growth hormone are normal [11].

Increased levels of glycated hemoglobin in fetal blood appear to precipitate tissue hypoxia, as this form of hemoglobin has high affinity for oxygen molecules.

Furthermore, chronic fetal hyperinsulinemia increases metabolic rates, thus increasing oxygen consumption and inducing relative hypoxemia; this, in turn, boosts red blood cell production, causing polycythemia and, consequently, hemolysis and neonatal hyperbilirubinemia. Severe hypoxemia can ultimately lead to fetal death.

After birth, the supply of glucose to the fetus is cut off, but hyperinsulinemia persists, speeding both exogenous glucose utilization and endogenous glucose production; this phenomenon may last approximately 3 days, until normal insulin secretion is established. Hypoglycemia may manifest in the intervening period.

4.1.2. Large for gestational age status

LGA neonates may also develop hypoglycemia [44], through the same mechanism observed in infants born to diabetic mothers; however, in these infants, blood glucose reaches normal levels within the first few hours of life [32].

4.1.3. Congenital hyperinsulinemic hypoglycemia

Hypoglycemia associated with congenital hyperinsulinism (CHH), also known as persistent hyperinsulinemic hypoglycemia of infancy (PHHI), is the result of inappropriate insulin secretion or hyperinsulinism. In infants with this disease, hypoglycemia is triggered by fasting and is always accompanied by an increase in plasma insulin concentrations, which are usually inappropriately high for the concomitant low blood glucose concentration. The disease appears to be more closely related to an increase in global endocrine functional activity of the pancreas rather than an increase in the number of pancreatic beta cells.

CHH is an important etiology that should be considered in cases of persistent and difficult-to-control hypoglycemia. It is a medical emergency that requires precise etiological diagnosis and represents a serious therapeutic challenge. The term PHHI was first proposed by Glaser in 1989 [19] and has since come to replace the now-outdated terms nesidioblastosis and islet-cell

dysmaturity syndrome to describe pancreatic abnormalities associated with hypoglycemia and hyperinsulinism.

Most cases of CHH are sporadic (1:40,000–50,000 live births), but a higher prevalence has been described in communities with a high degree of consanguinity. This familial form associated with inbreeding may occur in up to 1:2500 live births. Thus, an autosomal recessive inheritance pattern has been posited to explain it. There is no evidence of sex predominance, and the maternal history is generally negative; however, a careful history may reveal prior neonatal deaths or unexplained seizures or mental retardation in other siblings.

Patients with CHH are mostly LGA, as a consequence of hyperinsulinism, but without significant hepatomegaly. They exhibit persistent symptoms of hypoglycemia, including hypotonia, cyanosis, apnea, and difficult-to-control seizures, as early as the neonatal period. Sudden infant death is also seen in patients with CHH. Although the condition is rare, the high frequency of brain damage and developmental delay as a result of severe, treatment-refractory hypoglycemia in these patients justifies the need for early etiologic diagnosis and immediate treatment.

Currently, the most accepted etiogenic hypothesis for the dysfunction of CHH is inappropriate insulin secretion by pancreatic beta cells. The molecular basis of congenital hyperinsulinism involves defects in key genes that regulate the complex mechanism of insulin secretion control [12]. Nine genes have been identified and classified within the potassium channelopathies (ABCC8, KCNJ11) and metabolic disorders (GLUD1, GCK, HNF4A, HNF1A, SLC16A1, UCP2, HADH) [47, 52]. Genetic defect mutations involving the ABCC8/KCNJ11 genes, which encode the SUR1/Kir 6.2 components of the ATP-sensitive potassium channels (K_{ATP}) in pancreatic beta cells, are the most common [13, 27]. In normal cells, the K_{ATP} channels remain open or closed in response to variation in blood glucose levels, which leads to changes in the action potential of the cell membrane. An increase in blood glucose raises the rate of glucose metabolism in beta cells, resulting in increased adenosine triphosphate (ATP) and decreased adenosine diphosphate (ADP) within the cell, triggering closure of K_{ATP} channels and subsequent depolarization of the beta-cell membrane. This change in potential opens voltage-dependent calcium channels and leads to calcium influx. The subsequent increase in the cytosolic calcium concentration stimulates exostosis of insulin secretory granules; thus, insulin is released continuously.

The potassium channel is a complex of two proteins: SUR1, a receptor with high affinity for sulfonylureas, and Kir 6.2, which forms the inner pore of the channel and maintains its alignment [39, 58, 59]. The regulatory genes of the sulfonylurea receptor and potassium channels were recently mapped to region 11p15.1 of chromosome 11. Individually; none of these proteins has the ability to act as a potassium channel. Depending on the type of mutation affecting the genes that regulate these proteins, CHH may manifest with three distinct phenotypes. The first represents the familial form, with truncation of SUR1 and the absence of K_{ATP}. These patients have the most severe form of CHH and, in most cases, respond poorly or not at all to clinical treatment. In the second type, which accounts for sporadic cases, there is loss of K_{ATP} function but partial response to clinical treatment, due to formation of new potassium

ion channels. In the third type, onset is delayed and severity is mild, as these patients have functional K_{ATPs} and respond to clinical treatment.

A diagnosis of CHH is usually considered when hypoglycemia develops shortly after birth and requires glucose infusion at high rates, usually exceeding 10 mg/kg/min and occasionally up to 15–20 mg/kg/min. Typically, these infants have high blood levels of insulin, sometimes exceeding 10 μU/mL, and the insulin (μU/mL)-to-glucose (mg/dL) ratio is 1:4 or higher.

Beta-cell adenomas are characterized by marked, early onset hyperinsulinemia. These tumors require surgical removal or partial pancreatectomy. They are uncommon in the neonatal period. Definitive diagnosis can only be established through histopathology, and immuno-histochemical study may be required.

4.1.3.1. Beckwith-Wiedemann syndrome

Beckwith-Wiedemann syndrome is one of the most common overgrowth disorders and can be identified in more than 75% of neonates above the 90th percentile for weight and length. It is estimated to occur in 1 in 13,700 births, but mild cases may lead to underestimation of its true frequency [31]. There is no gender predominance. The syndrome is characterized by gigantism, omphalocele, and macroglossia, a triad that occurs in over 80% of cases. Other abnormalities that occur less frequently include earlobe creases and posterior helical ear pits, microcephaly, wide fontanels, a prominent occipital protuberance, facial nevus flammeus, nonspecific cardiac defects, abdominal wall defects (umbilical hernia, diastasis recti), visceromegaly, and hyper-plasia of the kidneys, pancreas, adrenal cortices, gonadal interstitial cells, and pituitary [50].

Neonatal hypoglycemia occurs in at least 50% of cases of Beckwith-Wiedemann syndrome. It is generally serious and may be associated with future mental retardation. Thus, early diag-nosis is important for proper treatment of low serum glucose levels, to prevent neurological damage. It is believed that hypoglycemia in this syndrome is secondary to hyperinsulinism caused by beta-cell hyperplasia and hypertrophy, but glucagon deficiency and a reduction in somatostatin-producing delta cells have also been documented.

There is a clear evidence of genomic influence in the development of Beckwith-Wiedemann syndrome. A mutation in 11p15.5, a region that encompasses multiple gene loci, has been implicated [37].

4.1.3.2. Congenital hyperinsulinemic hypoglycemia and other syndromes

CHH has been described in other diseases and syndromes, including congenital hypothyroid-ism [29], Sotos syndrome [4], Costello syndrome [21], Donohue syndrome [63], and Kabuki syndrome [62].

4.2. Situations associated with low or depleted glycogen stores

Prematurity and intrauterine growth restriction are among the situations that can influence neonatal blood glucose levels [35].

4.2.1. Prematurity

As very low-birth-weight preterm infants have limited glycogen stores, gluconeogenesis is their main source of glucose production. Gluconeogenesis is induced by decreased glucose intake, as well as by high cortisol, catecholamine, and glucagon levels.

Increased neurologic morbidity is particularly common in children with severe, recurrent hypoglycemia. Experimental observations have stressed the resistance of the immature brain to damage caused by hypoglycemia. This resistance is a consequence of compensatory increase in blood flow to the brain, reduced energy needs, increased endogenous carbohydrate stores, and ability to take up and consume alternative organic substrates while saving glucose for energy production [36].

4.2.2. Intrauterine growth restriction

As a result of intrauterine growth restriction, SGA neonates may exhibit several abnormalities shortly after birth, including increased susceptibility to infections, pulmonary hemorrhage, hyperbilirubinemia, and hypoglycemia. The widely varying incidence of the latter may reflect the different etiologies of intrauterine growth restriction (e.g., poor maternal nutrition, mothers with advanced age, uteroplacental insufficiency, derangements in maternal metabolism, or fetal infection). Furthermore, polycythemia and fetal and neonatal hypoxemia, which are often seen in SGA infants, can themselves contribute to development of hypoglycemia [49].

SGA infants are most at risk of hypoglycemia. Of those who do develop it, 65% are premature and 25% are post-term. Hypoglycemia can be asymptomatic or symptomatic and is generally observed in the first 24 h of life.

The factors contributing to low blood glucose levels include inadequate hepatic glycogen stores due to the high brain-to-body-mass ratio of SGA infants, the glucose-dependent nature of cerebral oxidative metabolism, and high overall metabolic rates. Furthermore, a reduction in rates of gluconeogenesis is probably responsible for 1% of episodes of prolonged hypoglycemia in SGA infants, as these infants exhibit high concentrations of gluconeogenesis precursors (such as alanine); this suggests an inability to convert these exogenous precursors into glucose.

Hypoglycemia combined with asphyxia is more damaging to the immature brain than either condition alone.

4.3. Situations associated with increased glucose consumption

Various situations can increase glucose consumption in the neonate. These include severe birth asphyxia [9], severe respiratory distress, and sepsis.

Perinatal asphyxia may initially feature hyperglycemia secondary to cortisol and catecholamine release; this is followed by hypoglycemia secondary to depletion of hepatic glycogen stores, mobilized in response to this excess glucose consumption. The association of hypoglycemia with transient hyperinsulinism has been described [18].

The association of severe respiratory distress, from various causes, and hypoglycemia caused by increased glucose consumption has often been described.

Neonatal sepsis is defined as a clinical syndrome characterized by systemic signs of infection and bacteremia (detected by positive blood cultures) during the first month of life. It is becoming increasingly important due to the reduction of neonatal mortality among the most premature newborns and to the prolonged care of these infants in neonatal units.

Decreased glycogen stores, impaired gluconeogenesis, and increased peripheral glucose utilization appear to be the factors responsible for hypoglycemia associated with sepsis, although the usual response to sepsis observed in animal models has been an increase in the rates of glucose *turnover* and gluconeogenesis, as the result of a counter-regulatory hormonal response [33]. Blunting of this process is observed only during the final stage of illness and serves as a marker of fulminant sepsis [38].

4.4. Drugs used in pregnancy

Drugs such as beta-adrenergic agonists [57], corticosteroids, thiazide diuretics, oral antidiabetics, propranolol, labetalol [3], valproic acid [10], antidepressants (SSRIs) [40], phenytoin, and terbutaline [55], among others, can cause hypoglycemia in infants.

B. Zhu et al. (2016) [67] reported an association between metformin use by diabetic patients during pregnancy and a reduction in incidence of neonatal hypoglycemia when compared to mothers who used insulin. Metformin has proven an effective alternative for use in this patient population, although it can cross the placenta.

5. Manifestations and clinical diagnosis

In most cases, infants—even those at risk—are asymptomatic. Nevertheless, an infant who is apathetic and refusing feeds and has a feeble cry should heighten suspicion of hypoglycemia. In high-risk infants, major findings include fine tremors, acrocyanosis, seizures, and apnea; if left untreated, coma and death may follow.

After birth, neonates born to mothers with diabetes develop complications related to their hyperinsulinemic state. In the first 3 days of life, these infants may exhibit episodes of irritability, tremor, and hyperexcitability or may present with hypotonia, lethargy, and weak suckling—manifestations consistent with early development of hypoglycemia and late onset of hypocalcemia. However, one must bear in mind that these infants are sometimes asymptomatic and the absence of symptoms should not delay testing for hypoglycemia.

The presence of tachypnea in the first days of life may be a transient manifestation of hypoglycemia, hypothermia, polycythemia, heart failure, cerebral edema secondary to traumatic delivery (particularly in macrosomic infants), or asphyxiation. The incidence of respiratory distress syndrome is high in these infants, since hyperinsulinemia may alter fetal lung maturation, inhibiting the development of enzymes required for the synthesis of pulmonary surfactant.

6. Laboratory diagnosis

Glucometry is the method of choice for initial screening of glucose levels, due to its use and minimal blood sample required; however, levels should be confirmed through laboratory measurement in plasma, especially when the glucometer reading is very low, as this method is rather imprecise at the lower limit of detection. Several factors can affect the values obtained by glucometry, such as the expiration date of the test strip, ambient temperature and humidity in the storage environment, the presence of sugars other than glucose, metabolic acidosis, high PO_2, hyperbilirubinemia, high hematocrit, and edema, among others [25, 66]. Several devices have been tested with the aim of demonstrating that their results may be unreliable and influence the management indicated by a reading [14].

7. Diagnostic imaging

A particular vulnerability of the occipital lobe to hypoglycemia has been observed on MRI [16], with no plausible explanation. Other authors have raised the possibility that variant anatomy of the circle of Willis and occipital lobe infarct may be implicated in these cases [2].

8. Treatment

As mentioned previously, there are still no clearly set values to define hypoglycemia in the first 2 h of life. It is known that, in the healthy, full-term neonate, blood glucose levels are lowest between 30 and 60 min of life and rise thereafter to normal baseline values of 60–90 mg/dL between 90 and 180 min of life. This threshold should be considered the physiological goal or therapeutic target at which blood glucose levels should be maintained.

Although one may consider a diagnosis of hypoglycemia when plasma glucose levels are below 45 mg/dL, this is not an absolute cutoff. Depending on the etiology of hypoglycemia and, consequently, on the availability of alternative pathways for gluconeogenesis, patients may be symptomatic in the 45–60 mg/dL range, as in cases of fatty acid oxidation defects.

SGA and late-preterm infants should be fed every 2–3 h and screened before each feeding in the first 24 h. After 24 h, screening needs only be continued in those whose glucose levels remain below 50 mg/dL.

8.1. Newborns asymptomatic in the first 2 h of life

The need for treatment in these children has been called into question, as hypoglycemia may be transient and may resolve spontaneously through stimulant counter-regulatory mechanisms. In general, if the infant is asymptomatic, to start early breastfeeding without the need to draw blood for glucose measurement, formula feeding, or other special care.

However, in some newborns, this physiological process may fail, which may facilitate the development of hypoglycemia; therefore, the American Academy of Pediatrics suggests that

in the first hour of life, asymptomatic at-risk infants should have a glucose check 30 min after feeding; if the blood glucose level remains below 25 mg/dL and the infant is asymptomatic, it should be fed again and blood glucose reassessed 1 h after the first check [67].

8.2. Asymptomatic high-risk newborns

Late-preterm, LGA, SGA, and intrauterine growth restriction (IUGR) infants, as well as those born to diabetic mothers, are at particular risk of hypoglycemia. However, they are often asymptomatic. Breastfeeding followed by repeated glucose measurement has been the standard of care. However, if hypoglycemia persists despite frequent feedings, continuous intravenous infusion of glucose may be indicated.

A dextrose infusion rate of 3–5 mg/kg/min may be used in infants born to diabetic mothers, both to prevent overstimulation of glucose secretion and because of the greater fat mass of these infants. A dextrose infusion rate of 4–7 mg/kg/min may be used in most full-term and late-preterm neonates. In IUGR neonates, a glucose infusion rate of 6–8 mg/kg/min is often necessary. A study in an animal model of IUGR revealed increased peripheral insulin sensitivity, which may be associated with increased glucose infusion requirements. However, some children with IUGR should be followed closely, especially preterm infants, who may develop hyperglycemia due to reduced insulin secretion and less muscle mass for glucose utilization. Continuous intravenous glucose infusion, usually preceded by an IV bolus of dextrose (200 mg/kg over 5 min), is also indicated if these newborns develop symptomatic hypoglycemia. However, the need for such massive glucose administration is hotly contested due to the risk of undesirable effects, particularly in very-low-birth-weight preterm infants. Complete or partial resolution of symptoms once glucose concentration is corrected is considered definitive proof that symptoms were caused by hypoglycemia. Nevertheless, IV dextrose infusions are not an entirely appropriate treatment; they cause discomfort to the infant, which is made worse by the need for placement of a deep IV catheter, the need for NICU admission, and physical separation of the newborn from the mother, which hinders timely initiation of breastfeeding. However, when administered safely so as to prevent these complications, IV infusion of dextrose at low concentrations can be beneficial even in asymptomatic high-risk neonates.

8.2.1. Dextrose gel

Oral administration of glucose in gel form has been considered appropriate and should be part of any protocol to prevent episodes of hypoglycemia in asymptomatic newborns [41]. Current studies have shown that oral administration of 40% dextrose gel may reduce the occurrence of neonatal hypoglycemia by up to 70% [5] and should thus be considered as the first-line treatment in these patients [65].

8.3. Symptomatic newborns

8.3.1. Glucose

Symptomatic neonates should be treated with glucose intravenously, not orally. A 200 mg/kg bolus of glucose should be administered over 1 min (10% dextrose at 2 mL/kg). This should be followed by IV infusion at 6–8 mg/kg/min. Glucose levels should be monitored after 30–60 min,

with a therapeutic target of >45 mg/dL. Control measurements should be obtained every 1–2 h. Once levels are stable, they can be reassessed every 4–6 h. If values do not reach a normal range, the rate of glucose infusion is increased by 1–2 mg/kg/min every 3–4 h. In cases of hyperinsulinism, a rate of 15–30 mg/kg/min may be necessary. Oral feedings should only resume once blood glucose levels have been stable for 6 h.

High glucose concentrations (20–25%) may be necessary to maintain a rate of infusion of 15–30 mg/kg/min; concentrations above 12.5% will require a central venous catheter [56].

8.3.2. Glucocorticoids

Physiologically, glucocorticoids promote increased resistance to insulin action, reduce the secretion of insulin, and activate enzymes involved in gluconeogenesis, mobilizing amino acids for this purpose. Thus, although such effects should theoretically induce an increase in blood glucose, there is no evidence to support glucocorticoid therapy in the treatment of hypoglycemia other than that caused by primary or secondary adrenal insufficiency.

Except in cases of hypoglycemia of self-limiting etiology (e.g., infants born to diabetic mothers), blood and urine samples should be drawn at the time of hypoglycemia for investigation of possible changes in energy and hormone metabolism (lactate, free fatty acids, ketones, insulin, cortisol, growth hormone, urinary organic acids) before any specific medications are administered.

8.3.3. Glucagon

Endogenous glucagon is the counter-regulatory hormone of insulin, secreted by pancreatic beta cells. Physiologically, hypoglycemia induces glucagon secretion to raise glucose levels [43]. The administration of glucagon has proven to be quite effective in full-term and preterm neonates without hyperinsulinism. Serum sodium levels should be monitored during glucagon infusion. Hyponatremia, thrombocytopenia, and a rare paraneoplastic phenomenon, called necrolytic migratory erythema, have been associated with continuous infusion of glucagon. Hypertonic saline solution (3% sodium chloride) may be indicated to treat glucagon-associated hyponatremia.

A dose of 0.02 mg/kg/dose has been recommended [43]. A 24-h continuous infusion has been used at doses of 20–40 µg/kg/h up to a maximum of 1 mg/day. A 50% rise in blood glucose is expected in normal infants. The effect is transient. Long-acting preparations are employed in patients with glucagon deficiency and, in combination with somatostatin, in the treatment of congenital hyperinsulinism. When the expected rise in blood glucose does not occur, the diagnosis of hepatic glycogen storage disease should be suspected.

8.3.4. Diazoxide

This agent is indicated in cases of hypoglycemia associated with hyperinsulinism.

Diazoxide is a benzothiazine derivative that acts by opening ATP-sensitive potassium channels, causing inhibition of insulin secretion by pancreatic beta cells. Therefore, patients with

genetic defects that affect SUR1 and Kir 6.2, the constituent proteins that form the ATP-sensitive potassium channel, may not benefit from administration of this drug. The recommended dose ranges from 10 to 15 mg/kg/day, divided in two or three oral doses, up to a maximum dose of 30 mg/kg/day. It promotes an increase in hepatic glucose production and decreases peripheral glucose utilization. Most of the drug is eliminated by glomerular filtration, and 90% of diazoxide is bound to albumin. Sodium and water retention, plasma volume expansion, edema, thrombocytopenia, anorexia, vomiting, ketoacidosis, and hyperuricemia are possible complications of the use of this drug [17].

When the drug is effective, blood glucose levels will return to normal range within 2–4 days. Any trial of diazoxide therapy should last at least 1 week before the possibility of treatment failure is considered. Onset of action occurs within 1 h of administration, and the duration of action is approximately 8 h, as long as renal function is normal.

Failure of diazoxide therapy suggests an abnormality in ATP-sensitive potassium channels. In these cases, a course of octreotide therapy, which acts further downstream on the insulin secretion pathway, is advised.

8.3.5. Octreotide

Octreotide was the first somatostatin analogue approved for clinical use, due to its more prolonged effect. This substance inhibits the secretion of glucagon, insulin, growth hormone, and thyrotropin, as well as the exocrine secretions of the bowel. Due to its ability to inhibit hormones, this drug can be used in infants with congenital hyperinsulinemic hypoglycemia [19]. A dose of 5–35 mcg/kg/day via subcutaneous injection has been recommended.

8.3.6. Sirolimus (rapamycin)

The management of diffuse hyperinsulinemic hypoglycemia, which does not respond to diazoxide, is a major therapeutic challenge. The successful use of sirolimus, both alone and as adjunctive therapy with octreotide, appears to be a potential alternative to subtotal pancreatectomy. Sirolimus is an immunosuppressant that inhibits the activation and proliferation of T lymphocytes, with effects *downstream* of the IL-2 receptor and other T-cell growth factor receptors.

In a study involving four patients with diffuse hyperinsulinemic hypoglycemia [46], therapy with sirolimus allowed discontinuation of intravenous infusions of dextrose and glucagon in all for patients and maintenance treatment with octreotide alone. At the end of the first year of life, the four patients continued to receive sirolimus and were normoglycemic, without any apparent major adverse events. Sevim Ünal et al. [63] reported the use of sirolimus in a neonate with CHH due to a KCNJ11 gene mutation who had already failed treatment with continuous infusions of glucose (14 mg/kg/min) and prednisone (2 mg/kg/day). Addition of intensive therapy with multiple medications (diazoxide, chlorothiazide, octreotide, glucagon, and nifedipine) also failed to produce an adequate response. However, before partial pancreatectomy was attempted, at age 30 days, sirolimus therapy was instituted at a dose of 0.5 mg/m^2/day.

Improvements in glycemic control were achieved, enabling progressive dosage reduction of the other drugs. At the time of publication, at age 5 months, the infant was on minimal doses of hyperglycemic agents and continued to receive twice-daily sirolimus at a dose of 0.3 mg/m^2/day, without any complications.

8.3.7. Exendin

Recently, exendin-(9-39), a GLP-1 receptor antagonist that raises blood glucose levels in adults, has been introduced as a possible novel therapy for management of hypoglycemia in patients with CHH. However, further studies on its effectiveness and safety are needed [8].

8.3.8. Other drugs

Growth hormone is used in cases of hypoglycemia associated with deficiency of this hormone or with hypopituitarism.

In cases of hypoglycemia due to persistent hyperinsulinemic hypoglycemia that does not respond to treatment with diazoxide, glucose, and sirolimus, partial pancreatectomy may be indicated.

9. Consequences

Recurrent or sustained hypoglycemia can cause neurological damage, mental retardation, epilepsy, and personality disorders [54]. Transient episodes of hypoglycemia are also associated with deficits in math learning around age 10 years [26].

Severe hypoglycemia can lead to impairment of cardiovascular function and is associated with high rates of neonatal mortality in very low-birth-weight infants [15].

Permanent brain damage is found in 25–50% of patients with recurrent severe symptomatic hypoglycemia under age 6 months. Furthermore, hypoxemia and ischemia may potentiate the permanent damage caused by hypoglycemia. The pathological changes described include gyral atrophy, reduced white-matter myelination, and cerebral cortical atrophy. It bears noting that the cerebral infarctions characteristic of hypoxic-ischemic processes are absent in hypoglycemia-associated brain injury [51].

10. Final considerations

Newborns with risk factors for neonatal hypoglycemia or those which, although not considered at risk, exhibit poor suckling or inadequate breast milk intake should receive follow-up monitoring and care after hospital discharge, so as to prevent possible undetected hypoglycemia.

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Selected Topics on Neonatal Infections

Neonatal Meningitis

Selim Öncel

Additional information is available at the end of the chapter

Abstract

Neonatal meningitis continues to be a problematic issue of neonatology and pediatric infectious diseases with its incidence of 0.8–6.1 in 1000 live births, high case fatality rate, and neurological sequelae. Major risk factors for contracting meningitis in the newborn period include maternal peripartum infection, premature rupture of membranes, premature birth, fetal hypoxia, septic or traumatic birth, low birth weight, and galactosemia. The leading causative agent is group B streptococci (in almost half of the cases), and a quarter of cases are due to *Escherichia coli*. Vertical transmission from the mother is often the route of infection. Neonatal meningitis may not be distinguishable clinically from neonatal sepsis without meningitis. Meticulous care should be taken to perform lumbar puncture whenever the patient's status permits since it is an indispensable tool for diagnosis. Initial empirical therapy may consist of ampicillin and cefotaxime, ampicillin and gentamicin, or ampicillin + gentamicin + cefotaxime during the first week of life. Ampicillin + gentamicin + cefotaxime for nonhospitalized infants and the same combination with the replacement of ampicillin with vancomycin for infants still in hospital are suitable options after the first week.

Keywords: infants, neonate, newborns, lumbar puncture, spinal tap, meningitis

Core tips: The incidence of meningitis in newborn period is so high that is incomparable to any other period in human life. The case fatality rate varies between 13 and 59% with respect to country of origin. Neurological sequelae, primarily hearing loss, continue to be an important issue with rates of 20–58% in neonates who manage to survive this relentless disease. Enteroviruses and *Enterobacter sakazakii*, which has been detected to contaminate infant formula, are emerging pathogens of neonatal meningitis. cerebrospinal fluid (CSF) glucose to serum glucose ratio is not a reliable indicator of meningitis in the first 28 days of life, because newborns often receive intravenous glucose infusions and serum glucose concentrations can rise abruptly with stress. Lumbar puncture should always be performed as soon as the infant becomes clinically stable

and suitable for the procedure. If *Listeria monocytogenes* grows in CSF or is suspected as the causative organism from the Gram smear, it is advisable to add ampicillin to vancomycin + gentamicin combination, because CSF concentrations of vancomycin are not bactericidal for Listeria.

1. Introduction

Neonatal meningitis continues to retain its importance all over the world as an infectious disease because of its morbidity. Medical facilities that enable physicians to keep more and more premature infants alive in economically developed countries and, on the contrary side, limitations in access to healthcare systems in economically developing countries keep neonatal meningitis on the medical agenda in an era of highly developed antimicrobial management and immunization.

2. Epidemiology

The incidence of meningitis in newborn period is so high that is incomparable to any other period in human life. Accurate determinations of incidence may not be possible due to lack of reporting by healthcare personnel and difficulties encountered by patients in access to healthcare institutions in economically underdeveloped countries. Nevertheless, it is estimated that 40,000–900,000 new cases of neonatal meningitis occur annually in these countries [1]. The incidence of neonatal meningitis, which is thought to be roughly one in 1000 live births, was reported to be 0.8–6.1 in 1000 live births in an article in which the results of 32 studies, carried out after 1990, have been reviewed [2].

A great progress has been made in this field of infectious diseases, at least in economically developed countries, with the decline of mortality rate from 50% of the past 40 years to that of 10–15% of today; however, almost no change has occurred in neonatal meningitis in terms of mortality in economically developing countries and morbidity worldwide [3]. The case fatality rate varies between 13 and 59% with respect to country of origin. Neurological sequelae, primarily hearing loss, continue to be an important problem with rates of 20–58% in the neonates who manage to survive this relentless disease [1, 4].

Turkey, once an economically developing country, where meningitis constitutes less than 1% of the reported causes of infant mortality, sets a good example of how natural health indices are affected favorably by slight increases in national income. According to the World Bank data, as of 2015, neonatal mortality rate in Turkey is seven per 1000 live births [an 80% decline from the rate (33) in 1990] [5, 6]. Yapıcıoğlu and colleagues reported the meningitis incidence as 1.4% among healthcare-associated infections in their university hospital's neonatal unit in Turkey [7].

3. Risk factors

Major risk factors of neonatal meningitis are low birth weight (<2500 g), premature birth (before 37th week of gestation), premature rupture of membranes (before the onset of labor

or regular contractions), septical or traumatic birth, fetal hypoxia, maternal peripartum infec-
tion, galactosemia, and urinary tract infection [3].

4. Etiology

In economically developed countries, owing to implementation of intrapartum antibiotic pro-
phylaxis beginning in the second half of 1990s, the incidence of early-onset group B streptococ-
cus infections declined, whereas that of late-onset group B streptococcus infections remained
the same [8]. Group B streptococci (GBS) and *Escherichia coli* are responsible for about half
and a quarter of neonatal bacterial meningitis cases, respectively. These agents are succeeded
in order of frequency by *Listeria monocytogenes* or Gram-negative bacteria other than *E. coli*
in some texts, *Streptococcus pneumoniae*, group a streptococci, and nontypable *Haemophilus
influenzae*. Gram-positive organisms other than GBS are encountered as pathogens more often
in very-low-birth-weight (<1500 g) infants. Although rarely, *Neisseria meningitidis* may cause
meningitis in newborns [3, 9].

Despite the data from economically developed countries, GBS predominance in neonatal
meningitis has been observed to be replaced by Gram-negative bacteria in economically
underdeveloped countries. *Klebsiella pneumoniae* is the most common Gram-negative bacillus
and is followed in frequency by *E. coli*.

The most likely causative pathogens in the first three days of life are GBS, *E. coli*, other enteric
bacilli, and *L. monocytogenes*. In addition to these, other Gram-negative organisms, such as *Serratia
marcescens*, *Pseudomonas aeruginosa*, and *Citrobacter koseri*, should also be listed as likely patho-
gens in neonates of four days of age and older. In neonates who have left their first seven days
in life behind, *Acinetobacter, Stenotrophomonas*, multidrug-resistant *Klebsiella*, and Gram-positive
organisms that have not been mentioned above should be considered as causative agents [10].

Neonatal infections caused by herpes simplex virus (HSV) occur one in 3200–10,000 live births.
These infections, which may manifest as neonatal meningitis, constitute 0.2% of neonatal hos-
pitalizations and 0.6% of neonatal deaths in hospital in the United States of America [11].

Fungal meningitis occurs on the grounds of risk factors, such as prematurity, central venous
catheter, congenital immunodeficiency, and long-term antibiotic therapy. The most common
fungal cause of neonatal meningitis is *Candida albicans* [12].

Enteroviruses and *Enterobacter sakazakii*, which has been detected to contaminate infant for-
mula, are emerging pathogens of neonatal meningitis [13, 14].

5. Pathophysiology

Neonates are apt to develop sepsis and meningitis more than all other individuals of all ages
due to relative deficiencies in humoral and cellular immune responses. Preterm and term
infants are deficient in complement in terms of quantity and quality, which leads to suscepti-
bility to infections by encapsulated bacteria. In infants younger than 32 weeks of gestational

age, transfer of maternal circulating antibodies through placenta occurs only in minute quantities. Neutrophil reserves of a neonate can easily become exhausted, since they are 20–30% of that of an average adult [15].

The causative agent of neonatal meningitis is usually transmitted to fetus vertically during labor. When the etiology is bacterial, histopathologic findings of meningitis in newborns are similar, irrespective of the specific agent. Another similarity is observed in the inflammatory responses of newborns, older children, and adults, the only exception being the paucity of plasma cells and lymphocytes in the subacute phase of meningeal reactions in newborns. The most common finding is purulent exudate in meninges and ependymal surfaces of ventricles. Perivascular inflammation is also observed in some patients. This may further proceed to arteritis of various degrees with phlebothrombosis and thrombophlebitis in the subependymal region. Hydrocephalus and encephalopathy were detected in half of the infants died from meningitis. Unlike older infants (3–12 months of age), subdural effusion is rare in neonates. Interleukin-1β is present in high concentrations in the meninges and brain tissue of the infants who have succumbed to the infection [16].

6. Physical examination

Clinical findings of neonatal meningitis are similar to those of neonatal sepsis with or without meningitis. It is not possible to predict with physical examination alone whether the infant has sepsis, meningitis or both. The most common (60%) finding is the alteration in body temperature. This alteration may become manifest as either fever (>38°C) or hypothermia (<36°C). Fever is usually observed in term infants, whereas preterms have a stronger tendency to develop hypothermia [3]. Skin vesicles should suggest HSV in the etiology of meningitis,

Finding	Frequency (%)
Alteration in body temperature (either hypothermia or hyperthermia)	60
Irritability or lethargy	60
Seizures	20–50
Bulging of fontanel	25
Nuchal rigidity	15
Poor feeding	50
Dyspnea	33–50
Apnea	10–30
Jaundice	28
Diarrhea	20

Table 1. Physical examination findings found in neonatal meningitis and their frequencies [1, 3].

but vesicles may not appear in the early stages or may not occur at all through the course of the disease as is the case in 20% of the newborns with systemic HSV infection. In the absence of vesicles, it is impossible to differentiate HSV meningitis from bacterial meningitis or meningitis due to other agents [11]. Seizures are seen more often in Gram-negative bacterial meningitis rather than in meningitis caused by Gram-positive bacteria. It is inadvisable to rely on the presence of bulging anterior fontanel or nuchal rigidity, because only a few infants (25 and 15%, respectively) with meningitis demonstrate these signs [3]. Neurologic signs usually appear after the second day, whereas mainly systemic signs predominate in the first 48 h. Physical examination findings in neonatal meningitis and their frequencies are summarized in **Table 1**.

7. Laboratory tests

First tests to be performed include complete blood count with differentials and cultures (urine and blood). Detection of growth in urine culture could be a reflection of metastatic dissemination of the organism to the bladder, thus cannot be relied upon as a locator of infection in young infants [1].

Lumbar puncture (LP) is an irreplaceable diagnostic tool in neonatal meningitis. Cerebrospinal fluid (CSF) obtained through LP should be examined directly and as Gram- and Giemsa-stained smears under microscope, cultured, and, if needed, sent for polymerase chain reaction. Direct microscopy should be performed as soon as possible, because the later it is performed, the more likely the erythrocytes and leukocytes undergo cellular lysis and escape detection. LP should ideally precede the initiation of antimicrobial therapy, but if, delayed for any reason, such as deteriorating clinical status of the patient, empirical antibiotic therapy should be started immediately.

Interpretation of CSF findings is more difficult in neonates than in older children, since the glucose, protein concentrations, and cell count of CSF are higher due to the high permeability of the blood-brain barrier (**Table 2**).

Age	Erythrocytes (μL/L)	Leukocytes (μL/L)	Protein (mg/dL)	Glucose (mg/dL)
Preterm: <7 d	30 (0–333)	9 (0–30)	100 (50–290) (mostly <200)	54 (27–99)
Preterm: >7 d	30	12 (2–70)	90 (50–260) (mostly <150)	54 (27–99)
Term: <7 d	9 (0–50)	5 (0–21)	60 (30–250)	54 (27–99)
Term: >7 d	<10	3 (0–10)	50 (20–80)	54 (27–99)
d: day(s).				

Table 2. Means and normal ranges of cerebrospinal parameters in neonates [12].

Many experts accept 20–30/μL as the cutoff value for pleocytosis. Decreased CSF glucose, increased CSF protein, and pleocytosis may indicate either bacterial or viral (especially HSV) meningitis. If only one of these parameters is in the normal range, this cannot be accepted as an evidence against the presence of meningitis. If all three parameters are normal, then it can be presumed that meningitis is not present; nevertheless, keeping in mind that completely normal CSF findings may be observed during the very early course of neonatal meningitis, the most prudent approach would be to repeat LP after 24–72 h in such borderline cases. If the infant had meningitis, pleocytosis and other abnormalities consistent with meningitis would be detected in CSF obtained in this second LP [3]. Ample number of erythrocytes in CSF may be interpreted as a clue to HSV meningitis if the physician is sure that the LP was not traumatic. Pleocytosis is more marked in bacterial and Gram-negative meningitides than in viral and Gram-positive meningitides [1].

CSF protein concentrations higher than 100 mg/dL in term infants and 150 mg/dL in preterm neonates are consistent with bacterial meningitis. CSF protein may also be found to be high in parameningeal infections like brain abscess, congenital infections, and intracranial hemorrhage [3].

The glucose concentration is said to be consistent with bacterial meningitis if it is below 30 mg/dL in term newborns and 20 mg/dL in preterm infants. CSF glucose to serum glucose ratio is not a reliable indicator of meningitis in the first 28 days of life, because newborns often receive intravenous glucose infusions and serum glucose concentrations can rise abruptly with stress [3]. In case of a bloody tap, assessing the CSF leucocyte count by correcting it with respect to that of the peripheral blood is not recommended in that it decreases the sensitivity and provides only a slight increase in specificity. When LP is traumatic, the wisest thing to do is to assume the patient as if she/he had meningitis and start empirical therapy [17]. Since sitting position with the legs flexed provides the widest interspinous spaces and it is sufficiently safe, it should be favored for sick neonates whenever the infant's condition permits a spinal tap [18].

Although, as noted above, signs of sepsis and meningitis intertwine in the newborn period, some neonatologists consider that it is unnecessary to perform LP on neonates evaluated for sepsis, especially those with early neonatal sepsis [19, 20]. Blood cultures are negative in one-third of neonates with meningitis who are very low birth weight and born over 34 weeks of gestation [1]. Thus, in case LP is not performed, a significant portion of neonates with meningitis would not get a correct diagnosis and would not be observed for the likely complications of meningitis; for that reason, the author is in favor of the opinion that LP should always be performed as soon as the infant becomes clinically stable and can tolerate the procedure if it has not been possible to be performed at the first suspicion of meningitis. It should be kept in mind that findings of CSF inflammation last for a considerably long duration (days, sometimes weeks), which allows the clinician diagnose or exclude the diagnosis of meningitis.

Ultrasonography is valuable in the follow-up, especially for the cases, in which hydrocephalus has developed as a complication of meningitis. If the disadvantage of radiation exposure is left aside, computed tomography can accelerate the decision making of ventriculostomy in cases of hydrocephalus and surgical drainage in patients with cranial abscesses. Magnetic resonance (MR) is the imaging modality of choice in conditions, such as focal neurologic

abnormalities, resistant infection, and clinical deterioration. MR is the most precise tool for the diagnosis of complications, like sinus vein thrombosis, ventriculitis, and subdural deposits. Electroencephalography has no diagnostic value in neonatal meningitis [12].

8. Diagnosis

History of premature or prolonged labor, intrauterine scalp monitorization, traumatic birth, and maternal peripartum infection should be noted.

Physical signs may be subtle in neonatal meningitis, in which either fever or hypothermia may be the only clue to diagnosis. Pleocytosis under direct microscopy or the presence of bacteria in Gram smear suggests meningitis. Definitive diagnosis is made with the isolation of causative organism in CSF.

The differential diagnosis includes other causes of neonatal seizures, partially treated meningitis, intracranial abscess, intracranial hemorrhage, intracranial aneurysm, cerebral vein thrombosis, head trauma, and congenital metabolic diseases.

9. Management

In the meningitides with the onset in the first 3–6 days of life, the empirical therapy should be ampicillin + cefotaxime, ampicillin + gentamicin, or, if there is a very high probability that the causative organism is Gram negative, as in the case of detection of Gram-negative bacilli on smear, it should be ampicillin + gentamicin + cefotaxime [10].

After the first 3–6 days of life, ampicillin + gentamicin + cefotaxime for infants from outside of a healthcare facility, and vancomycin + gentamicin + cefotaxime for previously or currently hospitalized newborns would be appropriate choices [1, 10]. If *L. monocytogenes* grows in CSF or is suspected as the pathogen from the Gram smear, it is advisable to add ampicillin to vancomycin + gentamicin combination, because CSF concentrations of vancomycin are not bactericidal for Listeria [10].

Dosages of recommended drugs are depicted in **Table 3**.

Dexamethasone therapy, which is used for older children, is not recommended for neonatal meningitis [1, 10].

Acyclovir (20 mg/kg/dose, every 8 h, for 14–21 days) should be administered to all neonates with HSV disease, regardless of manifestations and clinical findings [21].

For newborns whose CSF shows growth of a pathogen, the duration of therapy should be 14 days for Gram-positive organisms and 21 days for Gram negatives if neither any complication nor resistance to therapy is present. If a growth is detected in blood culture but not in CSF, while CSF shows signs of inflammation, a therapy duration of 10 days for Gram-positive organisms and 14 days for Gram negatives would suffice. Empirical antimicrobial therapy

	Weight < 1200 g	Weight = 1200–2 000 g	Weight = 1200–2000 g	Weight > 2000 g	Weight > 2000 g
Antibiotic	Age: 0–4 wk	Age: 0–7 d	Age > 7 d	Age: 0–7 d	Age > 7 d
Ampicillin	50, every 12 h	50, every 12 h	50, every 8 h	50, every 8 h	50, every 6 h
Gentamicin	2.5, every 18 h	2.5, every 12 h	2.5, every 8 h	2.5, every 12 h	2.5, every 8 h
Cefotaxime	50, every 12 h	50, every 12 h	50, every 8 h	50, every 12 h	50, every 8 h
Vancomycin	15, every 24 h	10, every 12 h	10, every 12 h	10, every 8 h	10, every 8 h

wk: week(s); d: day(s); h: hour(s).

Table 3. Dosages (mg/kg) of some antibacterial drugs used for neonatal meningitis [15].

for well-appearing infants with negative blood and CSF cultures and negative inflamma-tory findings in CSF may safely be discontinued 48–72 h after receiving negative CSF culture results from the microbiology laboratory [10].

LP should be repeated after 24–48 h after the beginning of antimicrobials for CSF is expected to become sterile in 24–48 h with appropriate therapy. In some centers, successful therapy for HSV meningitis is confirmed with a negative polymerase chain reaction in CSF, obtained with a repeat LP at the end of a 21-day antiviral therapy.

10. Complications and follow-up

Early complications of neonatal meningitis are increased: intracranial pressure, ventriculitis, cerebritis, hydrocephalus, brain abscess, cerebral infarction, and subdural effusion or empyema [22]. Late complications include hearing loss, abnormal behaviors, developmental retardation, cerebral palsy, focal motor deficits, seizures, and hydrocephalus, some of which may develop due to neonatal sepsis or cerebritis in infants without meningitis. Neonates recovered from meningitis should be referred for brainstem evoked auditory response audiometry 6–8 weeks after completion of therapy and then followed regularly for visual, auditory, and cognitive functions.

11. Prognosis

Case fatality rate is highest in meningitides due to Gram-negative organisms (17%), which are succeeded by those caused by fungi (12%) and Gram-positive bacteria (6%) [10, 23].

12. Conclusion

Taking Turkey as an example, a PubMed search by the author using the Medical Subject Headings terms groups "infant meningitis Turkey," "newborn meningitis Turkey," "neo-nate meningitis Turkey," "infant meningitis Turkish," "newborn meningitis Turkish," and

"neonate meningitis Turkish" revealed only two recent, relatively large-scale studies on the epidemiology of neonatal meningitis in Turkey, which points to the need for more local data in this field [24, 25].

Conflict-of-interest

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Author contribution

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Neonatal Osteomyelitis

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Additional information is available at the end of the chapter

Abstract

Osteomyelitis in neonates is relatively uncommon, but burdened with an increased hospital stay and possible long-term sequelae if not diagnosed on time. It differs from that of older children for etiology, clinical and radiological findings, and treatment. Due to anatomic contiguity, osteomyelitis may coexist with septic arthritis. Soft-tissue swelling or joint effusion is often associated. Our aim is to review the literature to provide the most recent data related to epidemiology, clinical presentation, diagnosis, treatment, and outcome.

Keywords: neonatal osteomyelitis, septic arthritis, antibiotics, imaging studies, micro-organisms

1. Introduction

Osteomyelitis (OM) refers to an infection of the bone that affects around 8/100,000 children [1]. For neonatal OM, an estimated incidence of 1–7/1000 hospital admissions has been reported [2, 3]. Due to their immature immune response, neonates are more susceptible than older children. Preterm infants are at a higher risk because of frequent blood withdrawal, invasive monitoring, diagnostic and treatment procedures, parent's nutrition, ventilatory support, perinatal hypoxia and prolonged NICU stay [4–7]. The long bones are the most frequently affected sites, especially of lower extremities, femur and tibia. Sites less commonly involved include the upper limbs, the pelvis, the clavicle, and the rib [8]. The presence of interosseous collateral arteries makes vertebral bodies less susceptible to infarction from septic emboli and more able to clear bacteria secondary to septic embolization. This explains the low incidence of vertebral body infection in neonates compared to older children and adults [9]. Few studies have focused on race differences. In low-risk neonates with OM, an accompanying

fracture should be considered [10]. Sternal OM is extremely rare, but has been reported [11]. Neonates are most vulnerable to multifocal infection [12, 13]. **Pathophysiology.** Osteomyelitis in neonates is usually due to hematogenous spread of bacterial infections or less frequently to direct inoculation as a result of a trauma or puncture wounds or surgery, infected cephalo-hematoma [14–16]. In preterm infants, direct injection of bacteria can result from heel or veni-puncture and artery or vein umbilical catheterization. Indirect contamination from a nearby infection, for example, cellulitis is also possible. Premature rupture of membranes, transpla-cental infection, and urinary tract infections has been described as risk factors too [17, 18]. A few cases of neonatal Gram-negative germ osteoarthritis have been reported, associated with a vesico-ureteral reflux (VUR) or hydronephrosis by the same microorganism [19, 20]. The most susceptible areas to haematogenous seeding of infection are metaphyseal of long bones, in particular the areas adjacent to the cartilaginous growth plate (physis) that is highly vascu-larized with slow intravascular flow. Abscess can result from the passage of bacteria through gaps from the sinusoidal veins to the capillaries into the tissue, where they are provided an ideal environment to grow. These abscesses frequently rupture into the joint [21]. Acute hae-matogenous OM and septic arthritis of the adjacent joint coexist in up to 76% of all cases as a result of a unique vascular anatomy characterized by the presence of vascular connections between the metaphysis and the epiphysis, particularly before the appearance of a secondary ossification center. Involvement of the shoulder or hip joints is noted when the intracapsular metaphyseal end of the humerus or femoral are involved from infection.

2. Symptoms

Asymmetric movement of extremities, irritability and poor feeding may be often the early and unspecific findings of OM [9, 22]. Common symptoms include bone pain, swelling, red-ness, guarding and failure to move the affected body part (pseudoparalysis). Fever may or may not be present because of an immature immune system. It is important to ascertain joint involvement through the detection of pseudoparalysis and pain during passive movements and signs of local inflammation [22]. Dierig et al. [23] reported a newborn with combined OM and suppurative arthritis caused by *Streptococcus pyogenes* giving rise to right brachial plexus palsy. Acute haematogenous OM is usually defined acute if the signs or symptoms are pres-ent for less than 14 days, and subacute if signs or symptoms are present for more than 14 days. In neonates, acute forms predominate [1].

3. Causative microorganisms

The causative agents of OM reflect those of neonatal sepsis, which vary from country to coun-try and in many cases remain unknown. The most common pathogen is *Staphylococcus aureus*, found in 70–90% of culture positive cases [24, 25]. Other pathogens include *Streptococcus* mainly group B (*Streptococcus agalactiae*) and Gram-negative enteric bacteria (*Escherichia coli and Klebsiella pneumoni*a) *Pseudomonas aeruginosa* [26, 27]. Multi-drug resistant *S. aureus*

(MRSA), community-acquired strains of methicillin-resistant *Staphylococcus aureus* (CA-MRSA) and *Kingella kingae* have emerged as being relevant in recent years and are responsible for serious infections [28]. *Candida albicans* OM needs to be considered in neonates, especially if preterms with specific risk factors. The presentation is more subtle and subacute, even in the absence of fever and elevated inflammatory markers. The progression is prolonged. OM from *Haemophilus influenzae* type b (Hib) has declined significantly; thanks to the introduction of the Hib vaccine.

4. Differential diagnoses

Differential diagnosis may be difficult and cellulitis, septic arthritis, subcutaneous abscess, fractures, and bone tumors should be taken into account. CNS disease (cerebral hemorrhage), trauma, scurvy, and child abuse are to be considered in the case of pseudoparalysis. Allagui et al. [29] described a case of acute OM of the clavicle in a 30-day-old newborn, with clinical symptoms simulating obstetric brachial plexus palsy. Laboratory tests are necessary to confirm a clinical diagnosis of OM. Neonates with OM may have a normal leukocyte count that is elevated in only half of the patients with or without thrombocytosis. The C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR) are almost always elevated (except in small bone infections). It is important to obtain blood, bone, or joint aspirate cultures if necessary, to identify the causative organism, before any antibiotics are given. Because OM usually is a consequence of sepsis, hence lumbar puncture should be considered. Any potential source of infection should be examined, including intravascular catheter tips. Serum procalcitonin may be used as a sensitive and specific marker in the diagnosis of acute OM [30] more suitable as an aid for rule-in diagnosis rather than for exclusion. Its diagnostic performance is better for a lower cut-off value compared to a conventional cut-off of 0.5 ng/ml which is specific but less sensitive [31]. A bone biopsy is advisable if the patient does not respond to the standard therapy.

5. Imaging studies

Imaging (computed tomography (CT) scan, radiography, bone scan, US and/or MRI) is used to identify the site of an infection, the presence of liquid collections for diagnostic aspiration and/or biopsy, to differentiate a unifocal from multifocal disease and to identify present or impending complications, such as joint or extradural involvement. Montgomery et al. [32] showed that the use of advanced imaging (CT scan, bone scan, and/or MRI) in infants younger than 4 months of age may shorten hospital stays, decrease the number of operative procedures required, and possibly limit infection-related sequelae. MRI has become the gold standard to evaluate musculoskeletal infection. It has the capability of assessing the osseous, articular and muscular structures simultaneously and does not require ionizing radiation. In particular MRI plays an important role in defining the extent of soft tissue involvement, defining drainable fluid collections and bone biopsy sites pre-operatively and thereby decreasing

the need for repeating the surgery. Increased marrow intensity with surrounding inflammation are the most suggestive signs of OM. MRI of the spine is useful in children not responding to therapy and/or to detect complications, such as extradural and paraspinal collections that will require surgical treatment because of causing spinal cord compression. The enhanced uptake of the radioisotope, distinguishes OM from deep cellulitis Gadolinium-enhanced fat-saturated T1-weighted sequences increase the confidence of the diagnosis of OM and may help also to distinguish edema from an abscess [33] and allows one to see the isolated involvement of the epiphyseal growth cartilage that is occult on radiographs and bone scintigraphy because of the paucity of growth cartilage ossification. The finding of hypo-enhancing foci in the growth cartilage suggests cartilage ischemia, necrosis or abscess as a consequence of infectious chondritis or septic thrombosis [34]. MRI may not be appropriate for monitoring the evolution of the lesions. Technetium bone scanning has a false-negative rate of as much as 20%, particularly in the first few days of illness. Indium-labeled leukocytes have limitations in newborns. Gallium scanning is not recommended because of lower specificity and exposure to higher levels of radiation. Ultrasonography: Although ultrasonography is an operator dependant technique with an inability to differentiate infectious fluid from traumatic ones, in able hands, it allows the detection of changes of acute OM as early as 48 h after the onset of infection. In the early stages ultrasound document deep soft tissue swelling (1–3 d), then the elevation of the periosteum by a thin layer of fluid, a definite subperiosteal collection, joint effusion and finally cortical erosion (2–4 w). In this last case it is used to guide needle drainage aspiration if necessary. Ultrasound images normalize by 4 weeks in the case of response to treatment [35]. Doppler venous ultrasonography is the first imaging study indicated in the case of clinical suspicion for deep vein thrombosis in patients with OM caused by CA-MRSA. A normal ultrasound scan does not exclude OM. Radiography is usually the first radiological investigation in a neonate with suspected OM, although it is reported that only 20% of the radiographs are abnormal at 10–14 days [36, 37]. Despite the low prevalence of abnormal features at presentation, it allows the exclusion of fractures and is useful to show long-term follow-up of complications. Initially it may reveal normal results, after 10–15 days signs of bone destruction, osteopenia, lytic lesions, and periosteal changes. Metaphyseal irregularities and periostitis (both non-specific) may be documented [37]. It has low sensitivity toward detection of a joint effusion or deep soft tissue swelling; the diagnosis of suspicion may include widening of the joint space with or without subluxation and soft tissues protruding that can be detected as early as 48 h after the onset of infection. Radiography may detect bone destruction when at least one-third of the matrix has been involved. Findings related to the spine may be limited to a loss of the normal lumbar lordosis, disc space narrowing, end plate erosions, pressure erosion of the superior and inferior margins of the adjacent vertebral body if the infection prolonged. Normally, other imaging tests are required. A bone scan is reserved for the cases in which radiographs and/or ultrasound are unclear, for suspected multifocal infection, chronic multifocal OM, and discitis. Bone scintigraphy is highly sensitive to the detection of OM in the early stages of the disease. In the first week of the disease, technetium (99mTc)-labeled bone scans revealed positive in 87% of the cases as compared with 42% diagnosed with radiography. Scintigraphy is useful for detecting multifocal diseases that are more common in neonates. A 99mTc-labeled phosphonate complex is the most used isotope. Scintigraphic study, even if non-specific, is useful to document through increased uptake of

all three phases: perfusion, blood pool activity and bone metabolism. Cold spots occur as a result of decreased blood flow secondary to edema and subperiosteal or articular infection. It may be a discriminating diagnosis in secondary bone infection in the case of persisting coagulase negative staphylococcus (CONS) bacteremia [38]. Computed tomography (CT) scan use is limited in the neonate. It is superior to MRI in chronic OM with cortical destruction, air and sequestra. It may also be used to guide aspiration and biopsy, especially when the spine and paraspinal soft tissues are involved [9].

6. Procedures

If signs and symptoms do not begin to resolve within 48–72 h of initiation of appropriate antimicrobial treatment, bone aspiration may be necessary to identify the pathogen and to drain the pus in accordance with the orthopedic surgeon. Bone and/or joint fluid aspirate for culture, can be bactericidal. Bone biopsy is necessary in the suspicion of tumors.

7. Management

It is a necessary antibiotic treatment, as soon as possible, in order to prevent the potentially adverse anatomic and functional consequences, preferably after obtaining blood and bone aspirates for culture. As cultures may be negative or difficult to obtain, empirical treatment is based on the local prevalence of organisms, resistance patterns taking account of the change over the years of the spectrum of organisms causing OM. The choice of an agent is generally either a penicillinase-resistant penicillin (e.g. nafcillin, oxacillin, flucloxacillin), which will be effective against S. aureus but may be of limited value against other organisms, or a broad-spectrum cephalosporin, which could have reduced the activity against *S. aureus*. Antibiotics against methicillin-sensitive *S. aureus* (MRSA) and streptococci (a penicillinase-resistant penicillin, first generation cephalosporin or clindamycin) must be incorporated into any empiric regimen for OM because *S. aureus*, *group A Streptococci* and group B streptococcus (GBS) and *S. pneuomoniae* together account for more than 90% of the cases of osteoarthritis in neonates [39]. Immunization rates worldwide have obviated the need to use antibiotics against Hib in many countries. Cefuroxime, a second-generation cephalosporin can be used as a single agent against both methicillin-sensitive *S. aureus* (MSSA) and Hib, if they are the suspected pathogens. The increasing incidence of penicillin-resistant S pneumoniae warrants the use of a clindamycin and cefotaxime/ceftriaxone combination. When treating neonatal OM, consider nafcillin and tobramycin or vancomycin and gentamicin combinations to provide coverage of bacteria from the Enterobacteriaceae family, in addition to group B streptococci and *S. aureus*. Vancomycin is preferred for proven or suspected MRSA-related septicaemia or known multi-drug resistant MRSA infection. The suspicion of enteric organisms justify additional therapy with an aminoglycoside, such as gentamicin, tobramycin or amikacin or an extended-spectrum, a Pseudomonas-active agent, such as cefepime [40]. In the case of acquired MRSA infections should be started vancomycin, rather than a penicillin antibiotic [5]. Daptomycin, Linezolid, and Quinupristin-dalfopristin have not been fully

evaluated or approved for use in neonates and should be employed when the neonate cannot tolerate vancomycin [40]. If B streptococcical infection is confirmed, combination therapy with penicillin G (or ampicillin) and gentamicin should be given for 2–5 days, after which time penicillin G (or ampicillin) alone is adequate. Monitoring serum acute-phase proteins, particularly the C-reactive protein, has been proposed as a useful way to determine resolution of infection and duration of therapy [40]. Management of *C. albicans* OM requires prolonged antifungal therapy. Society of America practices guidelines, including surgical debridement in selected cases and fluconazole therapy for 6–12 months, intravenous initially and then orally [26]. Amphotericin B is the most commonly used antifungal therapy. They are not provided clear guidelines on the optimum duration of treatment to eradicate infection. Even if by many are advocated shortened courses of antibiotic therapy because of morbidity and cost implications related to the prolonged therapy, the recommended entire duration of treatment still consists of at least 4–6 weeks until normalization of the C-reactive protein level [41]. Moreover it is typically recommended that infants under 3 months are given the full course of antibiotics parentally due to concerns over absorption and efficacy of oral antibiotics and to ensure adequate serum levels of the antibiotic. Recently some studies reported about oral therapy after a few days of intravenous therapy. Jagodzinski et al. [42] treated 70 children with intravenous therapy, converted after 3 (59%) or 5 (86%) days and continued for three weeks using temperature and C-reactive protein as parameters to response to therapy. The use of third-generation cephalosporins alone to treat OM is not recommended because they are not optimal for treating serious *S. aureus* infections. An earlier study by Vinod et al. [43] suggested that a reduced course of antibiotic therapy could be effective in the treatment of acute OM. Ecury-Goossen et al. [44] resolved clinical symptoms, microbiologic, and radiologic signs by using a short course of two weeks of intravenous antibiotics followed by 4 weeks of oral clindamycin, in selected preterm neonates with OM. The signs of continuing infection are persisting pain, fever, and rising hematologic markers that need prolonged antibiotics and repeated surgical intervention. While in children >3 months an early transition from intravenous to oral therapy (3–4 days) is suggested and a total course of 3 weeks in the treatment of acute OM, there are insufficient data on neonates to alter the current recommendation that a full course of at least 4 weeks of antibiotics be given parentally for neonatal OM due to concerns over absorption and efficacy of oral antibiotics [5]. Intraarticular administration of antibiotic is unnecessary. Some authors reported about successful treatment of newborns with oral dicloxacillin [45–47] fluclocacillin, fusidic acid and penicillin V for an additional period ranging from 14 to 42 days after an initial course of intravenous therapy. Despite everything, large, randomized controlled trials are needed to clarify the best practice in treating acute OM in children. It is important underline that OM and septic arthritis have a potential for life-long disability if treated insufficiently. Vertebral OM of the upper cervical spine requiring surgical treatment in children is rare. Glotzbecker et al. [48] described a surgery of stabilization of the upper cervical spine due to progressive instability caused by OM.

8. Consultations

The involvement of a multidisciplinary team of pediatricians, orthopaedists, and infectious disease specialists is helpful in the management of OM and results in a more efficient

diagnostic workup, and improved adherence to recommendations. An orthopedic and an intervention radiologist would be very helpful in determining the surgery for diagnosis and treatment and to obtain a bone biopsy under fluoroscopic guidance. The involvement of physiotherapists allows individualized rehabilitation programs, designed to improve the anatomical and functional characteristics of the affected bones. A prompt approach to obtaining tissue and blood specimens for the culture led to a higher rate of organism identification. Additionally a multidisciplinary team led to a shorter total length of hospital stay and a lower hospital readmission rate [3].

9. Prognosis and outcome

Considerable morbidity may be associated with neonatal OM. Joint effusion may lead to subluxation or dislocation of the affected joint, accumulation of inflammatory exudate within the joint causes vascular compression and may result in avascular necrosis of the affected epiphysis. Vein thrombosis and fractures are recently reported. Osteomyelitis may become responsible for permanent sequelae in 6–50% like joint disabilities, change in bone growth due to the damage of the cartilaginous growth plate, limb length discrepancies, arthritis, pathologic fractures, and rarely complete destruction of joints. Multiple risk factors are associated with bad outcomes in the long run. A delay in diagnosis and treatment can result in complications that include: damage to the growth plate with premature and/or asymmetrical closure of the growth plate; avascular necrosis of the femoral head with or without complete dissolution of the femoral head and neck; pseudoarthrosis; limb length discrepancies, angular deformities at joints; joint dislocations; joint arthrodesis; vertebra magna (with narrowing of the spinal canal); and block vertebrae. Other factors are: late in surgical drainage and appropriate antibiotic coverage, involvement of hip or shoulder, culture positivity and *S. aureus* isolation [3]. It has been documented that as many as 40% of children with septic arthritis of the hip will develop a serious complication, and a long-term follow-up is mandatory. Especially for concomitant septic arthritis and OM, the final outcome may be not evident until 9–10 years of age. Copley et al. [49] proposed criteria for discharge based on having a CRP < 2 mg/dL prior to discharge along with clinical improvement, resolution of fever, and having two sets of negative blood cultures for at least 48 h following any initial findings of bacteremia. Given the reported peripheral inserted central catheter complications (adverse drug reaction, a return emergency department visit or rehospitalization for adverse outcome), it should be considered the practice of oral antibiotic therapy instead of prolonged intravenous antibiotics after hospital discharge [50].

10. Conclusions

Acute OM, although rare in neonates, is a condition associated with morbidity and possible functional sequelae that need a prompt diagnosis and treatment. The implementation of evidence-based, clinical practice guidelines, a lower rate of initial bone scans, a faster change to oral antibiotics, a lower rate of presumptive drainage, and a shorter length of hospital stay are challenging objectives [51].

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Fungal Infections in Neonatal Intensive Care

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Additional information is available at the end of the chapter

Abstract

Neonates represent a unique and highly vulnerable patient population. Advances in medical technology have improved the survival and quality of life of newborns, particularly those with extreme prematurity or with congenital defects. Furthermore, immunologic immaturity and altered cutaneous barriers play some role in the vulnerability of neonates to nosocomial infections. In this context, the incidence of invasive fungal infections has increased significantly worldwide, representing an important infective complication in patients hospitalized in intensive care units. Invasive fungal infections in Neonatal Intensive Care Unit (NICUs) show high mortality; being species of *Candida,* the most isolates etiologic agents. The better prognosis of the patient is associated with the early diagnosis and fast treatment. However, guidelines to facilitate the optimal therapy choice for the treatment of neonatal fungal disease do not exist. The current antifungal agents that are available to treat fungemia among newborns and children are based on clinical trials in adults, since there are few comparative studies of antifungal agents in infants. The most commonly used drugs for the treatment of invasive fungal infections in neonates are classified in four different classes: polyene, azoles, analogs of pyrimidines and echinocandins.

Keywords: antifungal therapy, fungal infections, intensive care, neonates, sepsis

1. Introduction

During the last two decades, invasive fungal infections in preterm infants have become an increasing problem, mainly when hospitalized in a Neonatal Intensive Care Unit (NICU). Thus, for the last years, 6.3 million children under the age of 5 died each year are estimated,

and more than 40% of these deaths occur in the neonatal period [1, 2]. These data have several causes and particularly the neonates are at high risk due to fungal infections, mainly by yeasts of the genus *Candida* [3, 4].

In addition, species of the genus *Candida* have the capacity to inhabit several niches, thus, as part of the skin, mucosa and gastrointestinal tract [5]. Therefore, is considered that there is an association between colonization and systemic candidiasis mainly in seriously ill patients [6].

Although *Candida albicans* are more frequent, species of *Candida* non-*albicans* are also cause of diverse clinical pictures important in neonates, especially those that are in NICUs. The main Candida non-albicans species included are *Candida parapsilosis* complex, *Candida glabrata* and *Candida krusei*. However, uncommon species as *Pichia fabianii* and *Kodamaea ohmeri* may occur [7–10].

The newborns clinical course in the Intensive Care Units usually complicates after the onset of fungal infections [11]. In this hospital environment, adverse events may occur due to the complexity of the patients [12]. In this sense, the treatments and procedures instituted for primary disease may be an important factor for the onset of fungal infections; birth weight between 1000 and 1500 g is also a predictive factor and a way for the clinical worsening [13].

The predisposing factors to fungal infections include prolonged use of antibacterial and use of medical devices, among other conditions that lead to fungal disease. In addition, biofilms are frequent on the surface of medical devices, being consider a negative event, since it characterizes greater pathogenicity and antifungal resistance of fungi.

Commonly, amphotericin B, azoles and echinocandins are used for the treatment of various invasive fungal infections. However, antifungal therapeutic failures contribute to a higher mortality rate and may occur due to intrinsic resistance, so it is important to perform antifungal sensitivity tests. These tests can predict the ideal antifungal or contribute in the choice according to the use of other medicines and the condition of the neonate [9].

2. Epidemiology and incidence of neonatal invasive fungal infections

Neonates represent a unique and highly vulnerable patient population. Advances in medical technology have improved the survival and quality of life of newborns, particularly those with extreme prematurity or with congenital defects. Immunologic immaturity and altered cutaneous barriers play some role in increasing the vulnerability of neonates to infections. In this context, neonatal infection is a major cause of mortality and morbidity in newborns. Estimates suggest that >1.4 million neonatal deaths worldwide annually are due to invasive infections [14, 15].

The occurrence of invasive fungal infections has increased significantly worldwide, representing an important infective complication in patients hospitalized in intensive care units. Premature infants in NICUs are at particular risk of these invasive fungal infections, and unfortunately, the incidence of fungal septicemia appears to be increasing [16, 17]. In this

context, *Candida* and *Malassezia* species are the most prevalent pathogen involved in fungal infections in NICU [18].

The incidence of bloodstream infections due to *Candida* species in the overall population ranges from 1.7 to 10 episodes per 100,000 inhabitants. An estimated 33–55% of all episodes of candidemia occur in intensive care units and are associated with mortality rates ranging from 5 to 71% [16].

Invasive candidiasis is an important cause of sepsis in the NICU. Candida infections in infants are associated with significant mortality and morbidity, including neurodevelopmental impairment. The incidence of invasive candidiasis in NICU ranges from 2.6 to 13.2% in very low birth weight infants (1500–1000 g) and from 6.6 to 26.0% in extremely low birth weight infants (<1000 g) [19].

C. albicans has been the most frequently isolated species; however, infections caused by others species have been diagnosed with increased frequency. In the NICU in the 1990s, the overall incidence of candidemia increased because of the increased survival and intensive care of extremely preterm infants. During that time period, the proportion of candidemia decreased because of *C. albicans*, whereas increased because of *C. parapsilosis* [20, 21].

Invasive infections associated with *C. parapsilosis* cause fewer acute lethal events in premature newborns than systemic infections with *C. albicans*; nevertheless, *C. parapsilosis* fungemia significantly increases the morbidity and mortality of severely ill infants who require care in a NICU [22].

Laboratory studies have documented that *C. parapsilosis* is less virulent than *C. albicans*. However, the capability to adhere tenaciously to prosthetic materials forming biofilm and to proliferate rapidly in high concentrations of glucose are factors that facilitate the infection in the hospital environment. This trait may contribute to its ability to adhere to plastic catheters and cause systemic infections in premature newborns receiving total parenteral nutrition, blood pressure transducers or other invasive devices. Such a route of transmission may account for the occurrence of epidemic outbreaks of *C. parapsilosis* bloodstream infections [21].

Other emerging *Candida* species such as *C. haemuloniii*, *C. pelliculosa* and *C. tropicalis* have also been associated with infections in NICU. *C. pelliculosa* and *C. haemuloniii* caused clonal infection in NICU [11]. An outbreak of *C. tropicalis* fungemia in a NICU was traced to receipt of total parenteral nutrition and antimicrobial agents [23].

Malassezia species in immunocompromised patients may be associated with several skin conditions and systemic diseases, including folliculitis, seborrhoeic dermatitis, catheter-related fungemia and sepsis. However, this yeast may also cause invasive infections in critically ill low birth weight infants. *Malassezia* fungemia is predominantly caused by *Malassezia furfur* and *Malassezia pachydermatis*. *M. furfur* has been described predominantly in conjunction with nosocomial outbreaks in NICU, particularly in neonates and infants receiving intravenous lipids solution. Additionally, *M. pachydermatis* has been associated with bloodstream infection in preterm with very low birth weight and the prolonged use of indwelling catheters and parenteral lipid formulations [18, 24].

3. Neonatal *Candida* infections

Candida species are correlated to invasive fungal infections among at-risk groups as neonatal patients admitted NICU in and have been ranked third to seventh as a cause of nosocomial bloodstream infection, defined as candidemia, depending on geographical patterns [25–28]. Studies on invasive candidiasis infections and candidemia are frequently focused on specific diagnoses and/or specific populations. In all published studies, ICU was the most frequent localization of the patients, even with different frequencies [29].

Candidemia is associated with high rates of illness and death and has an attributable mortality rate that varies widely in the literature, ranging from 29 to 76%, both in adult and pediatric patients [30, 31]. Furthermore, *Candida* species are common gastrointestinal flora that causes a wide range of severe manifestations when disseminated into the bloodstream. Thus, candidemia has been described as the most common manifestation of invasive candidiasis [32].

These yeasts are less frequent than those infections caused by Gram-positive or Gram-negative bacteria; nonetheless, they are higher rates of morbidity and mortality. Particularly, among newborn with extremely low weight, 10% may to develop candidemia that has until 30% mortality in this patient group. Among infants who survive these infections, several long-term neurological impairments such as cerebral palsy, blindness, hearing and cognitive deficits and periventricular leukomalacia may occur [9, 27].

Neonatal candidemia during the first week of life is less common and less well described than the later onset of this group of infections. According to Barton et al. [33], risk factors for candidemia among neonates had not been studied before their research, in early onset disease (EOD, ≤7 days) or compared to late onset disease (LOD, >7 days). After a 2-year study, the authors concluded that risk factors such as birthweight <750 g, gestation <25 weeks, chorioamnionitis and vaginal delivery were strongly associated with EOD. Infection with *Candida albicans*, disseminated disease, pneumonia and cardiovascular disease were significantly more common in EOD than in LOD. Also, neurodevelopmental impairment and mortality were also higher than controls.

Extremely low birth weight is considered a risk factor related to a poor prognosis in EOD. Also, the role of perinatal transmission is supported by its association with chorioamnionitis, vaginal delivery and pneumonia. Dissemination and cardiovascular involvement are common, and affected infants often die. Empiric treatment should be considered in these risk situations [33, 34].

Pereira et al. [34] developed a retrospective observational study to investigate the risks for sepsis in neonates, including *Candida* infections, and verified an association between healthcare-associated sepsis, antibiotic therapy in day 1, the duration of parenteral nutrition and the use of central vascular catheter. For each extra week on gestational age, the risks declined in 20%, and for each day of parenteral nutrition, the risk increased 22%.

Kung et al. [35] affirm that infants in NICU have a higher incidence of *Candida* infections than any other pediatric or adult population. The predisposing factors were evaluated in Taiwan

using a retrospective matched case-control study conducted in the NICUs of a teaching hospital from July 2003 to June 2006. A total of 164 infants with culture-proven bloodstream infections were identified and the common etiologic pathogens included coagulase-negative staphylococci (28.7%), *Staphylococcus aureus* (16.5%), *Klebsiella pneumoniae* (14.6%) and *Candida* species accounting for 11 (6.7%) episodes. According to these authors, parenteral nutrition was a significant and independent risk of late-onset neonatal sepsis, including those caused by *Candida* species. This risk should be considered when implementing early parenteral nutrition in NICUs.

The collected potential risk factors consisted of: (1) prenatal and maternal history such as toxemia, multiple gestation, intra-uterine growth retardation and perinatal infections; (2) perinatal history such as premature rupture of membrane greater than 18 hours and delay in initial crying; (3) invasive procedures such as instrument insertion and its duration (e.g., placement of nasogastric tubes, endotracheal tubes, mechanical ventilation, peripherally inserted central catheters, chest tubes, blood transfusion or exchanged blood transfusion and lumbar puncture); (4) the concomitant use of medications such as parenteral nutrition and intravenous lipid, antibiotics and steroids and (5) comorbidities such as meconium aspiration syndrome (MAS), persistent pulmonary hypertension of the newborn (PPHN), intraventricular hemorrhage (IVH), necrotizing enterocolitis (NEC), respiratory distress syndrome (RDS), inborn error of metabolism and cardiac anomalies (except for patent ductus arteriosus and secundum type of atrial septal defect). However, transfusion, antimicrobial treatment, use of steroids and the presence of other comorbidities were not associated with *Candida* infections in neonates [35].

Invasive neonatal candidiasis presented an overall mortality rate of 35% during a study in Los Angeles (USA) in a neonatal intensive care unit. In general, every infant used a central venous catheter (CVC), required mechanical ventilation and previous administration of antibacterial agents. According to the authors, delayed institution of antifungal therapy was associated with increased mortality as well as length of hospitalization and the duration of prior antibacterial therapy [36].

Frequently, *C. albicans* is the most fungal clinical isolate; however, the incidence of bloodstream infections caused by *Candida* non-*albicans*, mainly *C. parapsilosis* complex and *C. glabrata*, has increased over the past 15 years. The current high rate of *Candida parapsilosis* infections may be attributed the capacity of this isolate to form biofilms and contaminate solutions, as those used in parenteral nutrition [8].

In recent Italian study, *C. albicans* was the most frequently identified strain, but nearly 20% of infections were due to *Candida* non-*albicans*, mainly *C. krusei* and *C. glabrata* [10]. Since both these strains can be resistant to fluconazole, that is the antifungal drug with the best urinary penetration [37], treatment of these patients could be challenging, despite a recent report that showed effective concentrations of micafungin in the urinary tract [38]. In these search, fungemia was the second most frequent diagnosis and was more frequent in children with malignancy/hematopoietic stem cell transplantation, those undergoing abdominal surgery and in low birth weight neonates, also in this case, confirming other recent pediatric data such as Ota et al. [39] and Steinbach et al. [40].

Invasive fungal infections in NICUs show high mortality. The better prognosis of the patient with invasive candidiasis/or candidemia admitted in NICU is associated with the early diagnosis and fast treatment. Evidence suggests an estimated mortality rate of 40% if therapy is not initiated early. Therefore, it is not a good practice to wait for cultures to become positive. This need for early therapy must be balanced against the need to use antifungal agents to avoid selection of resistant strains. Early empiric therapy guided by stratification systems for high-risk patients should help address these cases [41].

The score for exact risk measurement of invasive candidiasis has yet to be developed. The "Candida Score" presented by Spanish group in 2006 provides an easy-to-use tool to assist the health professionals with critically ill adults [42]. However, we believe that will should be adapted to pediatric patients, in the near future. In this stratification, the selected variables by logistic regression model with increasing weight are total parenteral nutrition, surgery, multifocal *Candida* species colonization and severe sepsis [42].

Recent Infectious Disease Society American guidelines suggest that "empirical antifungal therapy should be considered in critically ill patients with risk factors for invasive candidiasis and no other known cause of fever." Risk factors for invasive candidiasis are well identified. When analyzing clinical data, surveillance culture and levels of anti-*Candida* antibodies plus β-D-glucan in the serum, the same Spanish researchers showed a positive correlation among increasing values of the "*Candida* score" and the rate of invasive *Candida* infections. Such a score was calculated by variables such as total parenteral nutrition, surgery, multifocal *Candida* colonization and severe sepsis. Thus, *Candida* score ≥3 suggest patients at high risk for invasive candidiasis and enable to differentiate patients who would benefit from early antifungal treatment from those for whom invasive candidiasis is highly improbable [43].

4. Other neonatal fungal infections

Despite *C. albicans* is known to be primarily responsible for most neonatal fungal diseases, the prevalence of infections caused by other fungi in neonates and young infants is not significant, except for *Malassezia* species, which may occur in epidemic outbreaks [44–46].

Since 1980, this genus has been recognized in sepsis and systemic infections involving neonates receiving lipidic parenteral nutrition using a central venous catheter. It is believed that lipid supplementation facilitates the colonization of the catheter that used to infuse the nutrients. In newborns, colonization by *Malassezia* can progress to fungemia. The removal of the infected catheter is sufficient to limit infection in most cases [46, 47].

The vast majority of cases of fungemia occur in children less than 12 months old. In this population, this *Malassezia* infection rarely remains asymptomatic. Interstitial pneumonia and thrombocytopenia are common clinical manifestations in this group of patients, and the most frequent symptoms in systemic infections are fever and respiratory dysfunction with or without apnea [46, 48].

Other less common symptoms include lethargy, malnutrition, bradycardia and hepatospleno-megaly. However, no signs of erythema, swelling or purulence appear at the catheter entry site. Signs of skin rash are also not evident in children with systemic infections. Interstitial bronchopneumonia can be found in 40% of children [44–46].

The diagnosis of fungal infection by *Malassezia* is made by isolating the microorganism from blood collected through the catheter or by culturing the catheter tip after its removal. In suspected sepsis by *Malassezia*, the tip of the catheter should be cultured in broth enriched with lipids [45, 46].

The standard therapeutic management for systemic infections by *Malassezia* is still not well defined, since the fungemia by this microorganism is relatively unusual. However, some authors recommend the use of amphotericin B to treat these infections [45, 46, 49]. Morrison and Weisdorf [50] found that all patients enrolled in their study were cured without the administration of systemic antifungal therapy.

Studies have indicated that the most important factor for therapeutic success against systemic infection is the removal of the infected catheter and the interruption of lipid infusion, with or without antifungals [18, 45, 46].

5. Treatment of neonatal fungal infections

The appropriate use of antifungals agents is of particular importance in the prevention and treatment of invasive fungal infection in neonates; however, guidelines to facilitate the optimal therapy choice do not exist. The current therapeutic options that are available to treat fungemia among newborns and children are based on clinical trials in adults, since there are few comparative studies of antifungal agents in infants. The optimal treatment of fungal infection in this special population requires detailed studies on pharmacokinetics, safety and efficacy of antifungal therapies [51–54].

Similar to neonatal invasive infections by species of *Candida*, the management of *Malassezia* sp. fungemia requires the removal of any catheter as soon as the first positive blood culture occurs and the temporary discontinuation of parenteral nutrition in combination with an intravenous antifungal therapy. The most commonly used agents for the treatment of invasive fungal infections in NICU are classified into four different classes: polyene, azoles, analogs of pyrimidines and echinocandins. Among many years, the drugs of choice in this group of patients were amphotericin B alone or in combination with fluocitosin, liposomal formulation of amphotericin B or fluconazole. However, the development of a new generation of azoles and echinocandins, such as micafungin, has increased the therapeutic options for the treatment [45, 54].

Amphotericin B deoxycholate and lipid preparations are traditional choices for invasive fungal infections being active against a majority of clinical important *Candida* species and with reported use for *Malassezia* [45, 55]. Amphotericin B deoxycholate is well tolerated

by neonates who do not exhibit many of the toxicities seen in older children and adults. However, liposomal amphotericin B has been found to be safe and efficacious in newborns with renal impairment. Another polyene agent, nystatin suspension, is administered orally to infants with gestational age ≤ 27 weeks or birth weight less than 750 g until removal of central venous catheters; this is shown to reduce colonization of the gastrointestinal tract and the rate of invasive candidiasis [55].

Among the azoles, fluconazole is more frequently used in NICUs for the treatment of oropharyngeal and systemic candidiasis, but has no inherent activity against the genus *Aspergillus*, which is rare pathogen in neonates. This antifungal agent is commonly recommended as prophylactic therapy in NICU with a high incidence in fungal infections. Fluconazole prophylaxis is effective in reducing the rate of colonization and progression to systemic infection in nursery; on the other hand, some studies have revealed that prophylactic or empiric therapy with antifungal agents may be associated with changes in *Candida* ecology and antifungal agent susceptibility. Actually, the fluconazole dose recommended for neonates is 6 mg/kg/day, and maintenance doses currently used in NICUs in Europe is often higher, between 6 and 12 mg/kg [53, 56–58].

New azoles such as voriconazole, posaconazole and ravuconazole have limited utility in the nursery and are rarely used to treat neonatal infections. Voriconazole is a second-generation triazole that has excellent activity against *Candida* and *Aspergillus* spp.; however, data on its use in neonates are limited. Posaconazole and ravuconazole are the newest agents of the triazole family with added action against zygomycetes, however there are scarces of survey involving these antifungal agents in infants and the use of ravuconazole is not already approved by the Food and Drug Administration (FDA) [59, 60].

The echinocandins (micafungin, caspofungin and anidulafungin) are increasingly used for treatment of *Candida* sp. infections. Their role in the nursery is not so clear, although accruing evidence suggests they may be safe and effective, especially for the treatment of invasive infections caused by *Candida* spp. Some point have to be taken under consideration before the use of echinocandins in NICUs: first, limited clinical data also suggest that these agents may be effective for the treatment of central nervous system infections. Second, a high incidence of *C. parapsilosis* in NICUs is usually reported and this species is related to higher minimum inhibitory concentration (MIC) front of echinocandins [56, 57].

Among the three representatives of the group, micafungin is the most recommended and its use is approved for adults, children and newborns, being considered the one with better description for neonatal population. The use of caspofungin is approved by the FDA, but only for adults and children over 3 months of age. There were no relevant clinical trials that support the administration of anidulafungin among neonates and children [56, 57].

Invasive fungal infections are devastating pathologies that still result in death or serious long-term morbidity in neonates; however, the management of this mycosis has progressed greatly, with the azole agents playing a significant role. Effective prophylactic strategies have recently become available; therefore, the choice and use of appropriate antifungal drugs need careful assessment of neonatal characteristics, the epidemiology and drug pharmacokinetics [53].

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Selected Topics in Surgical Problems

Neonatal Care for Anesthesiologists

Esra Caliskan

Additional information is available at the end of the chapter

Abstract

In recent years, developments in obstetrics and neonatology have significantly improved the survival and quality life time of neonates. Therefore, anesthesiologists are more confronted with these patients due to surgical and non-surgical procedures. For a safe anesthetic approach and safe care, anesthesiologist must have necessary knowledge and equipment on the physiology of the newborn and should be better understand how immature organs respond to surgery and anesthetic. The purpose of this section is to present spot information that will allow clinicians and anesthesiologists to better understand the problems of neonatal patients and to perform safe care for these patients in the light of the physiologic characteristics of neonates. General principles of anesthetic management of neonatal patients are also reviewed and discussed.

Keywords: neonatal physiology, anesthetic management, perioperative evaluation, postoperative care, neonatal surgery

1. Introduction

Differently from the adult patients, pediatric, especially neonatal age group is a patient population that is difficult to assess. It should not be forgotten that children are not miniaturized adults. Therefore, the preoperative evaluation of the neonate is admittedly the most important part of anesthetic evaluation. The developments in the field of neonatal medicine over the past two decades have increased our probability of encountering a healthy and unhealthy neonatal population [1]. Surgical procedures especially emergency surgical conditions can be represent a significant anesthetic challenge and life threatening in neonatal period [2, 3].

Therefore, the anesthetist needs to take into account the immature function of many vital organ systems, the normal course of development, the effects of the underlying pathologic disease processes, which can frequently lead to serious physiological instability [4, 5].

In this chapter will be discussed issues that related to the anesthetic approach and postoperative care to neonatal patients.

2. Physiological feature in neonatal period

2.1. Organ and system differences in neonate

The neonatal period shows anatomical and physiological differences when compared to adult period. In this section, will be mentioned to physiological characteristics of neonates, also anatomical differences of neonates will be discussed in the part of anesthetic management.

It is important to know the anatomical airway differences between adults and children and newborns for a complete anesthesia assessment [6]. These differences can be listed as follows:

- Proportionately larger head and tongue,

- Narrower nasal passages,

- An anterior and cephalad larynx,

- A longer epiglottis,

- Shorter trachea and neck.

Because of these anatomical features, nasal breathing is dominant in neonates and infants until about 5 months of age [6].

In the newborn period, many vital organ and system functions are immature [7]. In respiratory system, high pulmonary vascular resistance (PVR) which is characteristics in fetal circulation decreases by approximately 10% after labor, but the apparent decrease occurs with the first breath [8, 9]. When the transition period to the adult circulation is examined, pulmonary blood flow, increased alveolar and arterial PO_2 appear to be important factors.

The characteristics of the neonatal respiratory system can be listed as follows: [1, 6].

- Immature hypoxic response, lower functional residual capacity (limits oxygen reserves during period of apnea, rapid desaturation in prolonged intubation attempt),

- Central control of ventilation is not fully develop,

- Peripheral feedback mechanisms are not sufficiently mature,

- Neonatal oxygen consumption 6 cc kg^{-1} per minute is twice that of resting adult (this value increases 10 cc kg^{-1} in the first week of life).

Cardiovascular system changes are also observed during neonatal period. The circulation of a newborn infant is a dynamic state which may revert to a transitional characteristic at any time. This means that the adult series circulation accompanies fetal parallel circulation [10].

The onset of spontaneous breathing is the main factor that reduces high PVR which is characteristic of fetal circulation. This decrease in pulmonary pressure is followed by an increase in systemic vascular resistance and left atrial pressure. Some clinical situations such as hypoxia and acidosis may lead to reduction of pulmonary blood flow which causes a return the fetal circulation features that is called persistent fetal circulation (PFC) [11].

Cardiac function is heart rate dependent in healthy neonates because the immature myocardium has limited compliance [2, 12].

The parasympathetic system is predominant in neonates [10], so that vagal stimulus by laryngoscopy and hypoxia may cause bradycardia. Therefore, it should be avoided bradycardia and treated aggressively when it is observed.

Liver and kidney function are also closely related to the anesthetic management of neonates. In the neonatal period, both organs have not yet completed maturation. The liver serves a critical role in carbohydrate, protein, lipid metabolism, coagulation. And is the primary site for biotransformation of drugs [13]. Hepatic oxidation, reduction and hydrolysis maturation process is rapid and these functions reaches adult rates around 6 month of ages [14]. Because of this, newborns may metabolize drugs (include anesthetic drugs) and toxins less efficiently than adults in the early months of their lives, as the pathway of degradation is immature [1]. Also, one of the changes in early infancy is the lower concentration of total serum protein, albumin and α1-acid glycoprotein. These proteins reach the adult level at about 1 year old [14].

One of the main routes of clearance of drugs and metabolites are the kidneys too. Glomerular filtration rate (GFR), active secretion and passive reabsorption are functions that determine renal excretion of drugs [15].

The kidneys also play an important role in the maintaining of acid base homeostasis and fluid electrolyte balance. GFR and reabsorption are increase age dependent manner. A neonate's kidney takes approximately 6–12 months to achieve adult performance.

In the perioperative period, fluid electrolyte balance is affected and the metabolism of commonly used anesthetic drug altered [10]. Drug metabolism and protein binding are also reduced due to immature organ (hepatic and renal) function [16].

Maturational physiologic changes are most prominent in neonatal period and infancy. The distribution of total body fluid component in neonates is different from adults. Body fluid constitute a greater proportion of body weight in the neonate (approximately 70–75%) and higher than adults. In neonate, the intracellular and extracellular fluid compartments are approximately 45 and 33% of body weight, respectively [17]. These body component changes affect volumes of distribution of drugs.

2.2. Thermoregulation

The newborn baby has a greater surface area relative to weight and a thin layer subcutaneous fat tissue [3]. These properties cause more heat loss to the environment in neonates than older

children and adult. Nonshivering thermogenesis is regulated by brown fat is the primary mechanism in heat generation in the neonates [1]. Brown fat tissue is located in the posterior neck along the interscapular and vertebral regions and it is responsible for heat generation in newborns.

There are four different mechanisms of heat loss [18]:

• Evaporation,

• Convection,

• Conduction,

• Radiation,

Cold intravenous fluids, exposure to cold sterilization solutions, drying of anesthetic gases and the direct effect of anesthetic agents on temperature regulation are the factors that increase this loss.

For this reason mentioned above, caution should be exercised in serious temperature changes in the perioperative period. Hypothermia may cause undesirable effects such as acidosis, myocardial depression, and delay recovery from anesthesia [19, 20].

Maximum precautions should be taken to prevent hypothermia in the perioperative period. These precautions are mentioned in the intraoperative management section.

Characteristics of neonates and infants that differentiate them adult patients are summarized in **Table 1**.

Anatomical differences	Physiological differences	Pharmacological differences
Head and neck	Increasing parameters	Hepatic function
• Relatively larger head and tongue	• Heart rate (dependent cardiac output)	• Immature biotransformation
• Narrower nasal passage	• Respiratory rate	• Decreased protein, decreased binding for drug
• Anterior and cephalad larynx	• Metabolic rate	Respiratory changes
• Relatively longer epiglottis and shorter trachea	• Total body water content	• More rapid rise in *FA/FI
Circulation	• Chest wall compliance	• more rapid induction and recovery from inhaled anesthetics
• Left ventricle –noncompliant	• Ratio of body surface area to body weight	• Increased minimum alveolar concentration
• Transitional circulation	Decreasing parameters	Body fluid changes
Respiratory	• Blood pressure	• Relatively larger volume distribution for water- soluble drugs
• Weak intercostal and diaphragmatic muscles	• Functional residual capacity	• Immature neuromuscular junction
• Increased resistance to airflow	• Lung compliance	

*FA/FI: Fractional alveolar concentration/fractional inspired concentration.
Table has modified by Lange Clinical Anesthesia 2015.

Table 1. Characteristics of neonates and infants that differentiate them adult patient.

3. General principles of anesthesia in neonate

Neonatal period, which is the most vulnerable time period in terms of anesthetic risk and perioperative mortality, is a challenging period for pediatric anesthetists.

Drug interactions, physiological and anatomical differences, and knowledge of risk factors are also important to reduce these risks.

What are the risks associated with neonatal anesthesia? In addition to the neonatal differences, prematurity, congenital anomalies, asphyxia at birth, and emergency situations that required surgery are risk factors. At the same time, there is a very narrow margin of error, including airway management, vascular access and drug administration. Some of these risk factors are summarized in **Table 2**.

Therefore, a safe aesthetic approach depends on a good understanding of the variables and physiological and anatomical changes taking place in the transition from fatal to neonatal life.

Anatomical differences

Airway anomalies
   Difficult airway: Micrognathia, macroglossia, cleft palate-lift
Prematurity, Immature organ and system function
   Respiratory
   Apnea (especially premature neonates)
   Respiratory failure, mechanical ventilatory support, supplemental oxygen
   Oxygenation: avoid high FiO_2 (high FiO_2 risk of retinopathy, NEC)
Cardiac
   Transitional circulation
   Persistent pulmonary hypertension
   Immature myocardium
   Parasympathetic dominance
Neurological problems
   Intraventricular/periventricular hemorrhage (IVH, PVH)
Congenital syndromes and disorders (especially associated with risk of difficult airway, comorbidities of cardiac defect)

- Pierre-Robin syndrome

- Treacher-Collins syndrome (mandibulofacial dysostosis)

- Goldenhar syndrome

- Klippel-Feil syndrome

- Down syndrome

Pharmacological differences
   Anesthetic drugs – dilutional changes, prolonged effect
Fluid management
   Glucose: avoid hypoglycemia
   Intolerance for rapid fluid infusion
   Problems of vascular access
Temperature management
   Prevent hypothermia
Emergency surgery

Table 2. Risk factors in neonatal anesthesia.

3.1. Effects of anesthetic drugs and inhalation anesthetics on neonates

Effective and safe drug administration in neonates should be based on detailed knowledge on the physiological characteristics of the neonates and pharmacokinetics and pharmacodynamics of given drug. Clinical pharmacology in neonates is recognized with extensive variability. The effect of anesthetic agents varies with organ maturation, body water content and metabolic differences and, coexisting disease in neonatal patient population.

Inhalation anesthesia is a commonly used anesthetic technique in neonatal population. However, intravenous and regional anesthesia methods also can be applied as anesthetic technique in this patient population. The minimum alveolar concentration (MAC) of an inhaled anesthetic is the alveolar concentration that prevents movement in 50% of patients in response to a standardized stimulus (e.g., surgical incision) and changes with age [1, 6, 21]. MAC for inhalational agents is less in neonates than in infants (up to 6 months of age [12, 21]. In newborns, cardiac and respiratory functions and blood pressure are very sensitive to volatile anesthesia. Immature myocardium and less developed compensatory mechanisms are responsible for this result. Therefore, these anesthetics can cause dose dependent cardiac and respiratory depressant effect and can be seen bradycardia, hypotension and postoperative apnea. These effects are more pronounced during the induction period [12, 22].

Intravenous anesthetic agents may be used for induction of anesthesia when inhalational anesthetics are not preferred and/or not practices.

Factors affecting the administration of intravenous anesthesia can be listed as follows:

- Larger intravascular and extracellular fluid compartments,

- Immaturity of hepatic biotransformation pathways,

- Decreased protein binding,

- Increased organ blood flow,

- Higher metabolic rate.

These factors mentioned above affect the properties of the intravenous anesthetics such as duration of effect (onset and termination), doses and toxicity limits of drugs.

An opioid agent causes fewer hemodynamic changes but it should be noted that increase the risk of postoperative apnea and late-onset re-sedation in recovery in preterm and term neonates [2, 4].

Thus, it is crucial that intravenous lines are flushed and cleared of any medication (e.g. opioid, neuromuscular blocker).

Muscle relaxant may be required for some surgical procedures (especially abdominal surgical procedures such as necrotizing enterocolitis, abdominal wall defect). The response of neonates to nondepolarizing muscle relaxant is quite variable. Neuromuscular junction is immature in preterms and this increases the susceptibility to muscle relaxants. However, the large

extracellular distribution volume will dilute the drug concentration, while those metabolized in the liver will prolong the duration.

4. Anesthesia preparation in neonates before surgical procedure

4.1. Preoperative evaluation

Anesthetic management in neonatal patients can be challenging but careful preoperative assessment means that a safe and smooth preparation process for the neonates and anesthetic team. This evaluation is also an approach that encourages confidence for their parents.

A careful preoperative examination allows the anesthetist to determine the risk of anesthesia based on the general state of health of newborn. It is aimed to establish an appropriate plan for anesthesia, postoperative care and analgesia in the light of acquired information.

Preoperative assessment in neonates should include a comprehensive examination of the neonate's perinatal history (such as prematurity, apnea-hypoxic episode history, duration of oxygen dependency, neurological damage etc.), congenital anomalies, previous anesthesia applications, and general state of health (acute, chronic lung disease, cardiac pathologies, etc.) [23].

Premature, ex-premature and term neonates are highly heterogeneous group within newborn period and needs various surgical interventions. Most of them come directly from the neonatal intensive care unit (NICU) and require urgent intervention. Many unusual congenital syndromes occur in childhood. These congenital anomalies in newborn period may accompany emergency surgical procedures [24]. Congenital anomalies that may cause difficult intubation should be noted and the anesthesia plan should be made accordingly.

In addition, a more detailed examination should be performed to reveal early postnatal problems that may be seen (such as hypoglycemia, infection and sepsis, etc.).

Risk factors which are important in neonatal anesthesia are summarized in **Table 2**.

A systematic approach to preoperative evaluation will prevent important problems from being overlooked. Many healthy neonates undergoing elective simple surgery and procedures are fit and do not require compressive physical examination. However, evaluation of the head and neck (such micrognathia, limited neck mobility, macroglossia, etc.) and cardiorespiratory examination (cardiac congenital anomalies, breathing sounds, respiratory pathologies associated with increased secretion and inflammation etc.) should be performed before surgical procedures. Also in cardiac examination, previously undiagnosed murmur with pathological qualities (e.g. loud, continuous, diastolic and or/associated with a trill) may be noticeable.

Evaluation of the airway is critical to the delivery of safe anesthesia. In neonates with a syndrome associated with airway anomalies, the potential presence of a difficult airway (**Table 2**) (*e.g. Pierre-Robin, Goldenhar, Treacher-Collins Syndrome, cleft palate and lip, maxilla mandibular synechia, etc.*) can be identified preoperatively.

It is also important to identify any concomitant (such as necrotizing enterocolitis, hyaline membrane syndrome, cardiac pathologies, sepsis, etc.) disease which might increase the risk of anesthesia and surgery. Familial, genetic, and neuromuscular diseases are important for anesthetic management. The presence of these diseases should be questioned.

Body weight, height, head circumference size must be recorded in order to evaluate newborn's physical status. For example, microcephaly is associated with some genetic disorders and may cause neurological sequel, which may pose a risk for anesthesia.

In laboratory evaluation, a baseline hemoglobin measurement should be obtained for minor surgical procedures [24]. If there is no indication for emergency surgery, anemia should be treated before surgical procedure. Because, even if it is a minor surgery, anemia may precipitate postoperative apnea and bradycardia in neonate. In addition to hemoglobin, in infants with fluid and electrolyte deficit, initial electrolyte values are also important. In major surgeries (e.g. oesophageal atresia, necrotizing enterocolitis, congenital cardiac surgery, neurosurgery) full blood count, blood electrolyte values, renal function and clotting test are recommended [25].

Fasting time before elective surgery should be pointed out. Clear liquid, breast milk and formula milk intake should be prohibited at according to the recommended fasting times (2 hours for clear liquids, 4 hours for breast milk, 6 hours for formula/non-human milk) before elective surgery [12, 14, 25]. Since prolonged period of fasting time pose a risk for hypoglycemia and dehydration, dextrose containing intravenous maintenance fluids should be instituted early period.

A systematic approach the preoperative assessment is summarized in **Table 3**.

Gathering of patient information
History of neonate
Perinatal
Medical conditions (co-morbidities, congenital syndromes, genetic disease)
Medications
Previous anesthesia and surgical experience
Examination
Body weight, height
Physical observation
Airway (especially pathologies that can cause difficult intubation, micrognathia)
Respiratory System
Prematurity
Bronchopulmonary dysplasia
Laryngeal pathologies, stridor
Breathing sounds
Cardiovascular system
Auscultation of heart
Neurological (intracranial hemorrhage history-treatment, seizure activation)
Previous resuscitation history
Previous anesthesia experience
Optimize medical condition
Determination of anesthesia risk
Determination of anesthetic plan

Table 3. Systematic approach to preoperative assessment.

4.2. Preparation of the operating theater

Anesthesia preparation should be based on the information obtained from the preoperative visit. But, anesthesia preparation does not include preparation of anesthetic equipment, drugs and anesthesia station only. It also includes preparations for the operating theater, such as proper heating of the surgery room, preparations for aspiration materials and arrangements for the patient's position.

The newborn baby has a high ratio surface area to body weight and a thin layer subcutaneous fat tissue. Therefore, neonates lose heat rapidly more than older children and adult [1]. The operating room temperature must be set to maintained neonate's thermoneutral temperature in order to minimize heat loss. In addition, the following applications can be used to reduce heat loss:

- Humidify and warm inspired anesthetic gases

- Many devices such as warmed blanket, overhead radiant heater, a forced air warmer mattress, clear plastic drapes

- Using heating solution for cleansing the skin

- Warm blood and intravenous solutions

- Transport the neonate in a heated incubator

Gas resources and suction materials must be checked.

The patient position is one of the issues to be considered. Different materials such as soft foam padding, rolls and tapes can be used safely for the position.

4.3. Assessment of patient and anesthetic equipment

Proper anesthesia preparation is very important for safe anesthetic management. This is one of the goals of the preoperative visit.

4.3.1. Airway

No matter what anesthetic method is used (sedation or general anesthesia), the airway must be guaranteed in every situation. Difficult, immature airway and respiratory system can lead to airway obstruction during sedation or mask ventilation. Therefore, appropriately size face mask, oropharyngeal airway, laryngoscope blades, endotracheal tube and suction catheters must be available. The use of transparent face masks are recommended for early recognition of cyanosis, vomiting and secretions.

Endotracheal Intubation Study Group [26] demonstrated that the use of Microcuffed tracheal tubes is effective and safe in neonates and young children. But, most anesthetist prefer to use according to the clinical condition of the patient rather than routine.

The laryngeal mask airway (LMA) can be used as an alternative to endotracheal intubation in the presence of difficult airway and for some short surgical procedures. Against possibility of

difficult pediatric airway, advanced airway devices (such as Glidescope, Airtraq) which allow indirect visualization of the larynx should be available [27].

4.3.2. Anesthesia devices

The anesthesia device must be pre-checked before surgical procedure and anesthesia practice. It is essential that the anesthesia ventilator has the ability to provide neonatal compliance, small tidal volume *(*usually in pressure controlled ventilator mode) and positive end-expiratory pressure [24]. In recent years, new generation anesthesia machines have been developed which is able to small tidal volumes [28] and to provide the ability to ventilate using pressure support ventilation (PSV) [29].

4.3.3. Monitoring

Heart rate-electrocardiography, non-invasive blood pressure, pulse oximetry, and temperature monitorizations are sufficient for basic monitorizations in a healthy newborn.

In neonates whom more risky for surgical procedures, in major surgical procedures and a special circumstances (such as congenital cardiac surgery, oesophageal atresia, congenital diaphragmatic hernia, expected ventilation-perfusion anomalies, hemodynamic changes, and acid-base imbalance), invasive arterial blood pressure and central venous pressure should be monitored.

Two oxygen saturation probes (the right arm probe – pre ductal, the leg probe - post ductal) must be plugged. This approach may provide the diagnosis as, with reverting to transitional circulation and is important to evaluate possible PDA mediated shunt development.

In last three decades, continuous monitoring of respiratory gases (inspiratory oxygen and expiratory end-tidal carbon dioxide and monitoring of gas flows) and continuous use of ventilator disconnection devices are observed routinely in most anesthesia clinic [27].

The precordial oesophageal stethoscope is a traditional and still valid method of monitoring the newborn. With this method, changes in heart rate and respiratory parameters can be identified in early phase.

5. Intraoperative management of anesthesia

5.1. Induction of anesthesia

The anesthesia station is the working area of the anesthetist. Before the surgical procedure, all drugs and equipments required for anesthetic administration should be checked. The neonate should be properly monitored and placed on a forced-air warming mattress before the induction of anesthesia. A safe intravenous access should be established for drug use and fluid therapy. The use of a topical anesthetic such as EMLA cream facilitates awake placement of intravenous cannula [30].

In addition to routine monitoring, direct observation of the neonate is also an important monitoring method. This observation allows the anesthetist to recognize early signs of certain clinic situations (such as cyanosis and pallor).

Inhalation agents or intravenous anesthetics may be used for induction of anesthesia. Although depending on the choice of anesthetist, induction with inhalation anesthetics is a more preferred method. One of the characteristics of the newborns that are different from older children and adults is that they have relatively high alveolar ventilation but low functional residual capacity [6, 14, 24]. This higher minute ventilation to FRC ratio with relatively higher blood flow to vessel rich organs contributes to a rapid increase in alveolar anesthetic concentration. These features enable rapid induction and rapid recovery in general anesthesia [6].

Inhalational induction has the advantage of protection of spontaneous ventilation (especially, important in neonates with potentially difficult airway).

With the advantages we have mentioned above, volatile anesthetics cause dose dependent cardiac and respiratory depressant effect in neonates. These effects can result bradycardia, hypotension and postoperative apnea. This hypotensive effect is also more pronounced in neonates and preterms with cardiovascular instability than older children.

Also, immature airway and respiratory system may cause airway obstruction in mask ventilation during induction phase. Laryngospasm is a frequent occurrence in neonatal anesthesia in this period [31] and if it is intervened early is usually easy to manage. However, laryngospasm, hypoventilation, hypoxia are among the causes of apnea during induction (especially in cases of late notice) [32].

In short surgical procedures, mask ventilation or laryngeal mask airway may be suitable to support respiration. However, endotracheal intubation is frequently performed in patients with emergency surgical procedures, long and major surgical interventions, conditions that requiring muscle relaxation and aspiration risk.

In situation that where inhalation anesthetics are contraindicated or not preferred, intravenous anesthetics may be used to induce and maintain anesthesia.

As we have already mentioned, total body water and extracellular fluid are increased in neonates [16]. These different fluid component affect volumes of distribution of intravenous anesthetic drugs (this means increasing the volume of distribution, especially for water-soluble drugs).

In addition, due to the immaturity of hepatic functions, duration of action will be prolonged in neonates if the drug depends on hepatic metabolism [6].

Muscle relaxant may be required for some neonatal surgical procedures. However, due to immaturity of the neuromuscular junction, neonates have an increased sensitivity to the effects of nondepolarizing neuromuscular blocking drugs [33]. For this reason, prolonged effects may occur at additional doses.

5.2. Intraoperative period

Balanced general anesthesia management is usually achieved by inhalation anesthesia supplemented with different class and wide range of drugs and/or muscle relaxants and regional techniques or achieved by total intravenous anesthesia.

Barbiturates, opioids, propofol, ketamine listed among these intravenous drugs [14]. Long-acting agents such as morphine should be avoided especially in day-case surgery or procedures and postoperative apnea risk should be kept in mind.

In neonatal patients, reduced hepatic glycogen stores, inadequate muscle glycogen reserve and gluconeogenesis enzyme activity require close monitoring of blood glucose concentration [34].

Intravenous glucose infusion may be required to maintain normoglycemia (serum glucose concentration of 40–90 mg dL [1, 24].

All maintenance fluids and blood products used intraoperatively must be warmed before use. Intravenous fluids should be titrated with an infusion pump or a fluid-adjusted burette to avoid excessive fluid loading and to give a controlled fluid.

Following the first few days of baby's life, in all newborns of gestational age, adequate intake of sodium is essential for continued normal developmental activity [1]. In full term neonates, it is not usually necessary to add sodium in maintenance fluids in the first 24 hours of life. However, sodium is added to the maintenance fluids after the second day to replace the sodium losses from the renal and gastrointestinal tract [35]. Also, non-hypotonic, dextrose-containing fluids for sodium replacement may also be used. But, hypotonic fluids should be avoided. These fluids are most common cause of potentially lethal postoperative hyponatremia [35, 36].

Intraoperative fluid requirement should also be met, depending on pre-existing fluid deficits, quality and duration of operation, and the extent of blood loss.

It should not be allowed hypothermia during the intraoperative period. Hypothermia may cause stress in the newborn, leading to postoperative respiratory insufficiency and ventilatory support [10]. On the other hand, the only stressor factor that to avoid in newborns is not hypothermia. Many stress factors (such as hypotension episodes, hypoxia, hypercapnia, acidosis, anemia) can occur during intraoperative period. These factors leads increase PVR and can cause return of the transitional circulation [Ivanova-24]. Different therapeutic maneuvers such as hyperventilation, deepening anesthesia, increased inspired oxygen, and volume expansion may also help in treatment [10].

5.3. Postoperative period and pain practices in neonate

Many neonates undergoing day-case surgery can be anesthetized using mask ventilation and/or LMA by continuing spontaneous breathing without the use of muscle relaxants and intubation.

However, in some situations (such as in major surgical procedures, urgent surgery, full stomach, etc.) endotracheal intubation must be done to secure the airway.

In these patients, in these patients there are many factors that determine postoperative extubation (including surgical conditions).

In major surgeries, (such as NEC, oesophageal atresia, diaphragmatic hernia) endotracheal intubation safer for these neonatal patients. In these neonates usually require elective postoperative mechanical ventilator support in early postoperative period in NICU.

Estuation period is a critical as intubation. Likewise, it requires to be careful and attentive.

It should be take care with regard to residual neuromuscular block and the neuromuscular blocker drugs should be reversed. In extubation period, we may encounter with side effects such as bradycardia, hypoxia, bronchospasm, which can be seen in induction of anesthesia. Intubation equipments and drugs should be available in the anesthesia theater for immediate intervention.

Post-conceptional age (PCA) is important factor in ex-premature babies, for day case surgical procedures. Most ex-premature babies are suitable for day case surgical procedures at greater than 60 weeks post-conceptional age (PCA). Prior to 60 weeks PCA, especially in premature infants less than 44 weeks PCA, postoperative apnea risk increased. In these neonates is recommended postoperative saturation and apnea monitoring in postoperative 24 hours period.

Pain in neonatal patients has for many years been ignored. However, the theory that newborns do not feel pain has become a subject that has lost its validity nowadays.

Recent studies have shown that neuroanatomical and neuroendocrine systems of premature newborns are sufficient for the transmission and perception of pain [37].

Untreated pain causes restlessness, increased oxygen consumption, ventilation/perfusion deficiency, and reduced food intake in early period of neonatal life. Long-term effects are learning disability and developmental retardation. Surgical trauma causes pain and a hormonal stress response that is directly related to the severity and urgency of surgery. Opioids (such as fentanyl, ultra-short acting remifentanil or traditional longer acting morphine) reduce the stress response and catecholamine release in response to painful stimuli. In addition, opioids provide effective pain control so decrease the pulmonary vasoconstrictor responses to painful stimuli in NICU.

In recent years, intravenous paracetamol has begun to be used in the treatment protocol of pain in neonates [38].

6. General anesthesia and developing brain. A special highlight for neonatal anesthesia

One of the most interesting topics in the last 20 years is the general anesthetic effect on the developing brain and neurocognitive functions.

Animal studies show that exposure to general anesthetics (especially these acting through the NMDA and GABA receptors) during the critical period of neuronal development in the developing brain leads to an apoptotic cell death and neurocognitive impairment especially in immature brain [39, 40]. It is reported that this effect to be related to drug dosages and exposure time.

Long-term neurobehavioral effects of sedative and anesthetic agents should be supported by long-term human studies. In recent years, early results of international, multidisciplinary, prospective clinical trials such as GAS and PANDA have begun to be published [41, 42].

In these studies and in Wilder et al. study, it has been reported that multiple exposures to anesthesia, rather than single exposure to anesthesia, pose a risk factor for later development learning disability in children [41–43].

However, to withhold sedation and anesthesia when necessary, would result in ethical consequences, in addition will trigger many negative consequences of painful stimulation in neonatal patients.

7. Conclusion

Approximately 1.5 million neonates receive general anesthesia each year for a surgical procedure according to literature data on neonatal patients [44].

In recent years, it has seen significant improvements in pediatric and neonatal anesthesia practice such as patient safety, advanced monitoring methods and new anesthesia equipments. Nevertheless, the neonatal period is still challenging for the anesthesiologist.

Anesthetic approach in neonatal population has to take into consideration the many immature vital organ functions as well as the effects of the underlying disease processes, which can frequently lead to severe physiological derangements.

Therefore, reliable anesthesia management is possible with a good understanding of the anatomical, physiological and pharmacological differences of the neonatal period.

Also, basic understanding of risk factors in neonatal anesthesia is a starting point for the reduction of risk. A careful preoperative examination of the child and the child's medical record, careful and close perioperative monitoring and follow-up and effective pain treatment would reduce mortality and morbidity.

The basics of safe anesthetic management in neonatal patients are summarized in this chapter, based on feature of neonatal period. The approach in neonatal emergency surgical procedures has not been mentioned in this chapter, since it was described another chapter.

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Pre- and Postnatal Surgery, Most Common Conditions, Diagnosis and Treatment

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Additional information is available at the end of the chapter

Abstract

The authors make a general but relevant description of the most common surgical problems occurring both in the human being that develops in the uterus and in the one that has already been born.

Keywords: prenatal surgery, postnatal surgery

1. Introduction

Congenital malformations, apart from the achievements in the prenatal care of women with high-risk pregnancies, are still a subject of great importance in our country. As Mexico is a developing country, due to various factors, an expeditious attention to newborns with malformations has not been achieved. An additional issue that affects the treatment of these children is the alarming increase in the prevalence of structural anomalies. Health statistics indicate that the birth rate is among the world's highest, which means that sometimes the state is incapable of providing the required assistance. Socioeconomic and educational factors are among those that have contributed to the increase in congenital malformations in the past 10 years, and the desired rate of recovery of these children has not been reached. Because of the above situation, specialists have had to refine their skills in elaborating the clinical diagnosis and instituting the appropriate therapeutic measures. In Mexico, neonatal surgery has become a specialty recognized by general pediatric surgery and by the Autonomous National University (UNAM).

2. Prenatal

Maternal-fetal medicine and intrauterine intervention in defects of the fetus, or unborn child, emerged and has evolved as a result of the technical advancements in medicine. The scope of these specialties is encouraging and promises advanced interventions with improved results. The refinement of ultrasound has made a great contribution to the field [1]. The use of ultrasound to visualize the fetus in its environment created a great interest to be able to identify malformations that could be corrected through intrauterine intervention. The most important antecedents of fetal surgery date back to the early 1960s, when Liley devised a technique for fetal transfusion [2]. In 1965, Adamson reported the first fetal surgery in humans, using a hysterotomy [3]. In this research line, experiments were carried out on lamb and monkey fetuses to perfect instruments and techniques, evaluate the results in these models and study their feasibility and reproducibility in humans.

The modern history of fetal surgery began at the University of California, San Francisco, with Michael Harrison. He first conceived his interest in intrauterine intervention for the treatment of congenital diaphragmatic hernia when he was an intern at Massachusetts General Hospital. Harrison's work continues today and is a valuable resource in the field [1–3]. In 1982, Harrison noted that diaphragmatic hernia, congenital hydronephrosis and hydrocephalus were defects which could be susceptible to prenatal treatment, as they were simple structural defects that prevented the normal fetal development of the structures involved. In that same year, the first open fetal surgery was performed at 21 weeks of gestation on a fetus with congenital hydronephrosis, which consisted of performing a hysterotomy to expose the lower abdomen of the fetus and place a double pigtail catheter to communicate the bladder with the amniotic cavity to promote the free flow of urine, thus relieving the obstruction causing the hydronephrosis [4]. The intervention was successful, and pregnancy continued for 14 weeks after intervention; however, at birth, the kidneys already showed irreversible damage, and it was clear that intervention had to be at an earlier stage of intrauterine life [5].

Neonatal surgery was not well known as a separate specialty of general pediatric surgery but has a history dating back to 1955 when P. Rickham, a pediatric surgeon from Switzerland, started a newborn surgery program in the city of Liverpool. Rickham's program began the systematization of neonatal care, especially for congenital anomalies. It was to be hoped that, having established a program of neonatal surgery, it would be recognized as a specialty. In spite of his prestige and worldwide fame, the specialty was not yet established. He published two editions of his classic book Neonatal Surgery, where in addition to the classically studied congenital defects, he narrated his experience operating on children with neurological conditions such as congenital hydrocephalus associated, or not, with pituitary-spinal malformations and musculoskeletal disorders such as club foot.

Neonatal surgery in the last third of the twentieth century has made remarkable advances in diagnostic resources. Postoperative management development has been especially important, with the implementation of ventilators and intravenous nutrition. In Mexico (specifically in the unit where the authors work), after years of managing hundreds of malformed newborns,

a successful postgraduate project in neonatal surgery has been developed at the Universidad Nacional Autónoma de México, which is already being replicated in other institutions in Mexico and other Latin American countries.

2.1. Obstructive uropathy

Congenital hydronephrosis was the first condition to attract attention within the scope of prenatal interventions, especially in bladder outlet obstructions due to urethral valves [4]. Congenital obstruction of the lower urinary tract (bladder neck) comprises a set of conditions of which atresias and posterior urethral valve obstructions are representative. With an approximate incidence of 2.2 per 10,000 live births, it results in oligohydramnios, pulmonary hypoplasia and irreparable renal damage, with 45% mortality. The role of prenatal obstetric ultrasound is crucial as the defect can be detected in 85% of the cases. Ultrasonographic features suggestive of lower urinary tract obstruction are an enlarged fetal bladder and dilated proximal urethra with or without associated hydronephrosis. It has been observed that when the anomaly is present before 25 weeks of gestation and secondary oligohydramnios prevails for more than 14 days, this malformation is associated with neonatal mortality > 90%. The canalicular phase of lung development, crucial to the development of the human lung, occurs during 14–25 weeks of gestation. Morris et al. demonstrated that prenatal intervention to treat bladder obstruction by hydronephrosis using percutaneous or open bladder shunt by a vesico-amniotic shunt increases overall survival in neonates affected by this abnormality, however, the studies published in this regard are few [6].

2.2. Sacrococcygeal teratoma

Sacrococcygeal teratoma is the most common tumor in the newborn. This neoplasm arises between the base of the spine and the rectum and protrudes from inside of the pelvis outwards. The tumor is more frequent in females than in males (3:1), with an incidence of 1 per 40,000 births. The presentation may be grossly frank and is seen at birth. Type 1 has the best prognosis, as the tumor is predominantly external with a small pre-sacral component (**Figure 1**). The tumor corresponding to type 4 is invasive, deforming the pelvis and part of the abdomen. The risk of malignancy depends on two factors: the site and extent of the tumor and age at the time of diagnosis. The prenatal appearance of the sacrococcygeal teratoma on ultrasound is of a mixed solid lesion with a cystic component that arises from the sacrum, with hyperechogenic zones corresponding to calcifications. Recently, ultrafast fetal nuclear magnetic resonance imaging has been used to assess the vascular component of these neoplasms, as there is risk of fetal intrauterine hemorrhage with secondary anemia, hemodynamic decompensation due to the formation of arteriovenous fistulas, hydramnios, hydrops fetalis, placentomegaly and eventually, in severe cases, intrauterine death. The effect of the mass of a sacrococcygeal teratoma has been related to uterine distension, dystocia and tumor rupture at birth, as well as causing mirror syndrome and severe hypertensive states.

The hemodynamic effect of the coccygeal sacrum tumor can be evaluated with echocardiographic study, measuring the relationship between ventricular outflow and the diameter of the vena cava, descending aorta and umbilical vein. A poor prognosis is related to

Figure 1. A giant sacrococcygeal teratoma in a newborn patient.

disproportionate tumor size (more than 150 cm per week is a very poor prognosis), high vascularity, predominantly solid component, placentomegaly, heart failure and concomitant maternal complications. The main objective of prenatal care is to identify the most reliable predictor of poor prognosis in fetuses with sacrococcygeal teratoma. The gestational age at the time of decompensation will determine the type of intervention; if it occurs after the 27th or 28th week of gestation, an emergency cesarean section or EXIT procedure (ex utero intrapartum treatment) will be chosen. If the hemodynamic imbalance is identified before week 27 in the cases of teratoma classes 1 and 2, open maternal-fetal surgery and partial resection of the tumor followed by definitive surgical intervention at birth should be chosen. There are other procedures that consist of disrupting tumor vascularity with dissimilar results, such as thermoregulation, radiofrequency ablation, alcohol sclerosis or laser ablation. The complications or side effects of these methods can result in extensive pelvic muscle damage, hemorrhage, nerve injury and hip dislocation [7].

2.3. Myelomeningocele

Within the congenital anomalies of the central nervous system, myelomeningocele is the most common variety of spina bifida and consists of the extrusion of the spinal tissue into a sac (meninges) occupied by cerebrospinal fluid. Its frequency is about 3.4 per 10,000 live births. Mortality is 10%, and survivors show varying degrees of disability depending on the height of the neural damage, including paralysis of the lower extremities and bladder and bowel dysfunction. In addition to exhibiting the anatomical neurological abnormalities characteristic of

Arnold-Chiari malformation.This anomaly is a generally congenital disease, consisting of an anatomical alteration of the base of the skull, which produces herniation of the cerebellum and brainstem through the foramen magnum to the cervical canal. This anomaly ultimately causes abnormal circulation of cerebrospinal fluid, culminating with hydrocephalus, central apnea, stridor and swallowing disorders. The embryonic pathophysiology of the defect is due to the combination of two strands or the two-hit model. The first effect consists of failure in the formation and closure of the neural tube, with an open defect, which exposes the neural elements to amniotic fluid (the second effect). It is important to note that in experimental models with fetal lambs, where an open neural defect was created and immediately surgically covered, effects on the lower extremities and on urinary continence and bowel movements were not observed. On the other hand, fetuses of lambs who underwent surgical repair of the myelomeningocele after 4 weeks of having experimentally created the defect showed good outcome after fetal intervention, urinary continence was maintained and preserved the neuromuscular function of their limbs, Which supports the hypothesis that prenatal intervention prevents neurological and associated confusion. Adzick in 2011 in a randomized clinical trial [8].

2.4. Pulmonary cystic adenomatoid malformation

Congenital pulmonary malformations have been divided for their study into those related to the bronchial tree: agenesis, aplasia and pulmonary hypoplasia and those of pulmonary parenchyma: congenital lung cystic adenomatoid disease, pulmonary sequestration, lobar emphysema and bronchogenic cyst. The high definition and quality provided by current sonographic studies have achieved better prenatal identification of the fetal lung lesions in the majority of cases detected on routine ultrasound between 18 and 20 weeks of gestation. These malformations have an incidence from 1 per 10,000 to 1 per 35,000 pregnancies. Sonographic study provides information on variables such as volume, location, arterial blood supply and venous drainage and consistency of the adenomatoid malformation, which can vary widely from solid (microcystic) to frankly cystic (macrocystic) [7].

Adenomatoid malformation, consisting of small cysts, generally grows unpredictably and causes compression on surrounding structures, as well as an effect of mass on the mediastinum, esophagus and lungs, causing pulmonary hypoplasia, obstruction of the vena cava, cardiac insufficiency, hydrops fetalis and polyhydramnios. As one method to identify serious cases, several investigators have proposed predictive indices and measures such as the cystic volume ratio (CVR), obtained by dividing the volume of the cystic lesion (length X width X height X 0.52) by the circumference of the head. In a retrospective study by Crombleholme, it was concluded that a CVR greater than 1.6 was associated with a 75% chance of developing hydrops fetalis, which justified prenatal intervention [7, 9]. In cases with a better prognosis, EXIT therapy is recommended, with resection of the mass before birth. Postnatal stabilization and thoracotomy are also acceptable. Macrocystic lesions may be prenatally decompressed by thoracentesis with a single needle or with drainage guided by amniotic thoracic ultrasound. At this time, microcystic lesions are not candidates for drainage and will require fetal surgery [10].

3. Postnatal

3.1. Esophageal atresia

Esophageal atresia is one of the most common congenital malformations and perhaps the most classic of structural abnormalities that exclusively involve the pediatric surgeon, as only a specialist can resolve the problem. Other specialists such as the urologist usually operate on an undescended testicle and a general surgeon on a pyloric stenosis, but even a thoracic surgeon would not attempt to correct an esophageal atresia. The frequency of the anomaly is one case per 3000 to 5000 births, predominately in males, and the prevalence may depend on the region or country being analyzed. How esophageal atresia occurs is unknown, but there are many theories, none of which has withstood the test of time. Esophageal atresia is usually accompanied by other defects, so acrostics are formed to list the components such as VACTERL, for vertebral, anorectal, cardiac, tracheo-esophageal, esophageal, renal and limb anomalies [11]. At times, it is sporadically associated with other defects of the digestive tract, such as duodenal atresia. In Mexico, the frequency measured in cases per year is between 12 and 16 cases a year in regional and specialty hospitals. The Moctezuma Pediatric Hospital, a specialty hospital of the Secretary of Health of Mexico City, sees the most cases per year [12].

3.1.1. Diagnosis

No specific biological teratogen is known, but the anomaly may be produced by the experimental administration of Adriamycin. More often than not, it is a sporadic malformation that does not obey genetic or hereditary patterns, although it is usually accompanied by trisomies such as 13 and 18 or those associated, without the atresia being the axis of clinical expression. It usually occurs in the products of conception of young women. Polyhydramnios can be found during the prenatal visit, which should be the most important warning sign [13, 14]. In pediatric surgery, it is said that sialorrhea in the newborn child is equivalent to an esophageal atresia until proven otherwise. If this is accompanied by cyanosis and asphyxia during feeding, the diagnosis is confirmed. Although it should be noted that it is currently the obligation of the obstetrician in the delivery room to make the clinical diagnosis of suspicion. In other words, waiting for the child to choke on ingested milk signifies malpractice and tardiness to integrate the diagnostic impression.

The routine introduction of a nelaton catheter into the stomach is a maneuver that should be performed in every delivery room. It is the most effective, advisable and efficient way for a child with esophageal atresia to achieve a good condition without additional risk during intervention. With the diagnosis confirmed, the next step is to inject 0.5 ml of water-soluble contrast medium for radiological study (**Figure 2**), which, in addition to highlighting the height of the atresia, will demonstrate a proximal fistula if it exists and for which a surgical procedure would be indicated [13]. When the defect is detected, a probe must be left in the proximal pouch so the saliva is prevented from spilling over into the respiratory tract. The tube should be constructed as a trap so as to provide continuous suction without pinching the esophageal mucosa and avoid blockage. This is achieved by introducing a thin probe through the counter-aperture in the middle third of a larger probe. The thicker probe should be very close to the

bottom of the pouch, and the thinner probe will serve to continuously instill saline solution to keep the system efficient.

From the clinical point of view, esophageal atresia has been divided into four varieties and classified into letters or numbers. The system with letters is the most used: type A, two blind pouches, but no fistula; type B, two segments with the upper one connected to the airway; type C, two segments with the distal esophagus in the airway; and type D, both segments that contain a fistulous path. Radiological studies, in addition to revealing the aforementioned findings, may show a cardiac silhouette that suggests right aortic arch cardiopathy, as the presence of some skeletal malformations such as hemi-vertebra or spina bifida can be detected. The prognosis may be definitive when an area of pulmonary consolidation is identified. The presence of air in the digestive tract indicates the existence of the most frequent variety, an atresia type C or D. The absence of air in the digestive tract identifies variety A or B. Once the diagnosis has been confirmed, studies to reduce the risk to the child should be made. Thus, when a condition that has a high risk for esophageal atresia is detected, such as hydrocephalus or obstruction of the urinary tract, it should be given priority [15].

3.1.2. Treatment

The management of esophageal atresia requires zeal and closeness to the patient as with few other anomalies. In no other condition is it so important for the pediatric surgeon to be aware that the suction device works and the child's vital signs and laboratory values have been restored. Antibiotics are essential. In ideal conditions, an end-to-end esophageal anastomosis
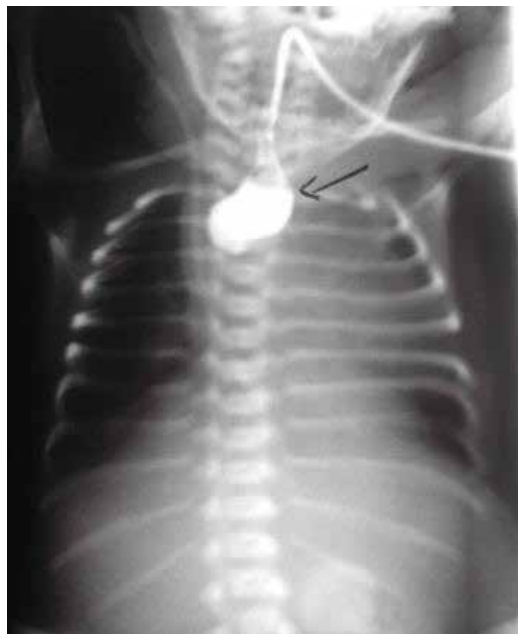

Figure 2. Simple thoracic X-ray, revealing the fundus of the proximal sac contrasted with a water-soluble medium. This is an esophageal atresia type A.

should be performed by right thoracotomy, regardless of whether it is in one or more anatomical planes. It is important that it is done with fine, inert material and the placement of the stitches is firm and their number and location are correct. Ideally, it should be done outside the pleural space, but if it is not feasible, a pleural tube should be placed and removed on the day of the esophagogram to prove that there is no dehiscence and/or refistulization. In approximately a week, they may begin oral feeding; meanwhile, they must be maintained intravenously.

Prognostic criteria can identify those who are included in the favorable group and those who will survive. The best-known criteria have to do with the weight of the product of conception, the presence of other malformations and if bronchial aspiration pneumonia is present. Based on these criteria, those classified as risk type A are those weighing more than 2500 grams, lacking significant malformations and without pneumonia, and more than 90% of these cases survive. Mortality is 50% or more in the opposite, high-risk conditions. Complications, which can be classified as immediate and mediate, are frequent. In the former, it is the dehiscence of the anastomosis, which always has a reserved prognosis, and during late onset, it is the pathological gastro-esophageal reflux [15].

3.2. Duodenal atresia

Atresia of the duodenum is the congenital obliteration of any portion of the small intestine, which lies between the most distal pylorus and the most proximal part of the ligament of Treitz. This is a very common malformation and almost a permanent occupant of neonatal intensive care units, as one case appears for every three to five thousand births [16]. It almost always appears as a sporadic condition but may be part of some chromosomopathies such as trisomy 21. It has been assumed that it appears between 5 and 12 weeks of gestation. The most accepted theory is that of Tandler. Known as recanalization, it consists of the appearance of the concentric histolysis phenomena due to the invasion of vacuoles into the solid cord, an essential characteristic of the duodenum in the embryonic stage, to the degree that the invasion converts the cord in its definitive form, into a tube [17]. Other theories, such as ischemia may have a basis, especially in segment atresia accompanied by a loss of duodenal continuity with or without intermediate fibrous cord.

Anatomically, duodenal atresia has been divided into two forms: the proximal and distal to the ampulla of Vater and divided into three forms clinically—when it appears as a septum or as "wind sock," when there is an interruption of continuity and when these segments are joined by a thin fibrous cord. Regarding the wind sock variety, it should be emphasized that duodenal blockage does not appear at the transition site of the dilated proximal segment and the thin distal segment but that the membrane always emerges a few millimeters above the narrow area.

3.2.1. Diagnosis

Duodenal atresia usually appears early in pregnancy, accompanied by polyhydramnios. When performing sonographic screenings in prenatal consultation, the presence of polyhydramnios should alert the specialist to look for data suggesting blockage of the digestive tract, such as atresia of the duodenum or of other distal structures. Once the product has been born, the correlation can be made by gastric lavage with a catheter as a routine measure and obtaining 20–25

ml of transparent or biliary fluid, which indirectly evidences alterations in the dynamics of the movement of amniotic fluid. If the mongoloid facies resulting from trisomy is added as relevant data, the suspicion of a duodenal block is strengthened. On inspection, there is an increase in volume of the upper abdomen, the skin is taut and shiny and there is early vomiting. Depending on the level of the blockage, the expelled liquid will be yellow, thick and abundant if the blockage is below the ampulla of Vater and the vomitus will be transparent if the obstruction is proximal to the main bile duct outlet. Due to alterations in the enterohepatic circulation, this anomaly is almost always accompanied by jaundice, which disappears as soon as the problem is resolved through surgical intervention. If several days elapse before identifying the anomaly, from the clinical viewpoint, there are usually hydroelectrolytic alterations, exacerbation of jaundice and signs of a systemic inflammatory response. Regarding radiology, simple radiography of the abdomen in an upright position is sufficient, as air is the best contrast medium in those cases. When children have a tube inserted in the stomach, 15–20 ml of air should be injected at the time of the study in order to maintain the contrast. Observing the image of the "double bubble" (**Figure 3**), it is a certainty that the duodenum is totally or partially blocked, and there is a presupposition of the existence of defects such as annular pancreas, malrotation, presence of a narrow angle mesenteric artery and the preduodenal portal vein. However, the presence of the facies, umbilical hernia and macroglossia are usually diagnostically determinant factors so that before the surgical intervention, with a good degree of certainty, it is assumed that the baby suffers from duodenal atresia. No endoscopy or introduction of liquid contrast material is necessary.

3.2.2. Treatment

In our country, this type of patient usually arrives in a serious condition. Vomiting is early, and in less than 24 h, there is an important loss of fluids and electrolytes, so the patient must be immediately transferred to an intensive care unit, where a gastric tube must be installed to allow the exit of all the liquid that is lodged in the stomach in utero, a quantity much superior to the normal gastric capacity. Antibiotics should always be used, as there is often respiratory infection aggravated by microaspirations. Once the gastric tube has been installed and the patients' condition improved, the patient must be transferred to the operating room where, depending on the findings, the intervention of choice will be instituted. If confronted with an atresia without loss of duodenal continuity (variety I), a slight concentric depression should be located on the surface of the duodenum. That indicates the point where the obstructive diaphragm is inserted. Once this reference is placed, a horizontal incision is made on the depression, the septum is removed, the mucosa is sutured with a continuous line of absorbable fine material and the duodenal wall is repaired vertically. This method is known as the Heinecke-Mikulicz technique. If, on the contrary, atresia shows separation of segments, then it must be resolved through two types of access: through a minimally invasive procedure consisting of the introduction of very thin laparoscopy tubes and laparotomy [17]. In both methods, the surgical technique will be the duodenal-duodenal diamond-shaped anastomosis, proposed by Kimura et al. [18], which consists of joining both blind segments, making a horizontal incision in the proximal and a vertical one in the distal so that when anastomosis is terminated, intercommunication is wide enough for the intestinal flow.

Figure 3. Simple abdomino-thoracic radiological study showing great gastric dilation and obstruction of the second portion of the duodenum.

The original report of the diamond anastomosis technique says that the oral feeding route in the postoperative period could be initiated in the first 5–7 days; however, in our experience [19], the waiting period for tolerance of fluid intake can be extended up to 4 weeks, which means that those children must undergo intravenous nutrition for at least 15 days. We believe that duodenal dilatation resulting from the receipt of large amounts of fluid results in alterations in emptying movements, which is corrected spontaneously but later in postnatal life. An additional problem is that these infants suffer from recurrent nosocomial infections due to their prolonged hospital stay and the use of a catheter, which makes their recovery more difficult. Management also includes a gastric probe that protects the anastomosis, the use of antimicrobials and analgesics.

3.3. Jejunoileal atresia

The small intestine is a complex and vital organ. Jejunoileal atresia accounts for almost one-third of all cases of intestinal obstruction in the neonatal period.

The pathogenesis of jejunoileal atresia is still controversial. In the middle of the last century, it was proposed that the atresia was due to a failure in recanalization; however, Louw and Barnard, through experimental studies, showed that this anomaly occurs as a consequence of an ischemic phenomenon that culminates with segments of mesenteric ischemia and in atresia. Subsequently it was shown that abdominal catastrophic events in utero were responsible and that the most common conditions were invagination, perforation, volvulus, strangulation by an internal hernia or thromboembolism. The spectrum of atresia depends on the extent, severity and duration of the ischemia, since the mucosa and submucosa are more susceptible. An incidence from 1 to 5 per 10,000 live births has been estimated. It affects both sexes equally. It affects both the jejunum and the ileum similarly. Compared to duodenal atresia, associated anomalies are less frequent. Multiple intestinal atresias can be an autosomal recessive disorder, more commonly seen in combination with some degree of immunodeficiency and in Canadians. Mutations have been observed in the TTC7A protein, important for the development and function of the intestinal epithelium. This mutation has also been associated with early presentation of inflammatory bowel disease [20].

Based on their anatomical characteristics, four types of atresias have been described. Type I is an intramural membrane of mucosa and submucosa that is continued with a cord, with no mesenteric defect. In Type II, there is intestinal discontinuity but no mesenteric defect. Type III has two subtypes; in suwbtype IIIA, continuity of the intestine is interrupted and there is also a V-shaped defect of the mesentery. In subtype IIIB, there is a lack of continuity of the intestine and it is wrapped around the superior mesenteric artery, giving the appearance of a Christmas tree or apple peel. Type IV consists of a segment with multiple atresias that resemble a string of sausages. We have designated a type V, which consists of a combination of atresias types I, II and IIIA present in the same intestinal segment [21].

3.3.1. Diagnosis

The typical presentation is that of a newborn on the first or second day of life with biliary vomiting, a history of polyhydramnios and abdominal distension, which will be more severe the more distal the obstruction. The most serious form of presentation is intestinal perforation. The diagnosis can be made prenatally through the use of ultrasonography. Findings suggesting atresia, in nearly one-third of cases, include dilated bowel and polyhydramnios. If there is strong suspicion, fetal magnetic resonance usually determines the diagnosis. Once the product of conception is born and suspected, simple vertical and horizontal x-rays of the abdomen should be taken. When a jejunal atresia is present, X-ray will show multiple hydroaerial levels (**Figure 4**) and the "triple bubble" sign. The presence of calcifications suggests the catastrophe and may be present in about 12% of cases. In that scenario, confirmation of atresia is obtained by colonic enema, when a microcolon is observed [21, 22].

3.3.2. Treatment

Once the diagnosis has been made, a gastric tube is placed to empty the proximal digestive tract, the hydroelectrolytic alterations are corrected, antibiotic management is added and a central venous catheter is installed. The operation can be performed by an open or a laparoscopic

approach, in order to resect the proximal atresic bulbous segment and perform a primary anasto-mosis with the distal segment. A precautionary transoperative routine measure is the instillation of saline solution to the distal intestine in order to rule out the presence of other obstacles [22, 23].

Mortality is related to prematurity, associated anomalies, infections and short bowel syn-drome. Postoperative complications that may occur are leakage, stenosis of the anastomosis site and short bowel, if there was extensive bowel resection. Food intolerance may be a mediate complication. Success is rated by the time of initiation of enteral feeding, postsurgi-cal complications and the duration of feeding with total parenteral nutrition. The prognosis of neonates with jejunoileal atresia is very good, with a survival rate greater than 90%.

3.4. Necrotizing enterocolitis

Necrotizing enterocolitis (NEC) is one of the leading causes of death of gastrointestinal origin in the preterm infant. It is a devastating disease, and at present, it can be considered as part of a spectrum of diseases acquired in the neonatal period characterized by necrosis of the ileum and/or colon. The incidence is 1–5 per 1000 newborns [24]. In multicenter studies, it has been

Figure 4. Simple abdomino-thoracic radiological study in horizontal position showing a great dilatation of the small intestine. Note the pelvic opacity.

estimated that NEC is present in 7–13% of neonates younger than 33 weeks of gestation and weighing less than 1000 g. Mortality is almost 50%, mainly in those requiring surgical treatment. The majority of cases occur in premature babies and a lower percentage in term births [24].

The intestinal epithelium of premature infants is predisposed to have an exaggerated inflammatory response to bacterial colonization, allowing destruction of the mucosa and damage to mesenteric perfusion. This inadequate inflammatory response triggers the emergence of TLR4 (toll-like receptor 4), a response receptor found in the premature gut epithelium that recognizes the lipopolysaccharides of membranes of Gram-negative bacteria. This regulation of the signaling pathway includes nuclear factor kappa beta 1 and IL-1 receptors. Other important risk factors that have been implicated in its development include intestinal immaturity, inadequate bacterial colonization of the intestine, asphyxia, anemia, presence of congenital defects such as gastroschisis and persistent ductus arteriosus, use of medications such as indomethacin, low birth weight, Apgar score of five or less, use of mechanical ventilation, feeding with milk formula and, more recently, the presence of an exaggerated inflammatory response [24, 25].

3.4.1. Diagnosis

The typical newborn with NEC is a premature infant who suddenly develops food intolerance, bloating, bloody stools and signs of sepsis (changes in heart rate, breathing, temperature and blood pressure). An important consideration in the diagnosis of NEC is gestational age and age of presentation. In other words, preterm infants born at 27 weeks of gestation are at greater risk, and symptoms occur at 4–5 weeks of life, compared to term infants. This may be because preterm infants have later colonization of the digestive tract, a prolonged hospital stay and have had broad-spectrum antibiotics used [25].

Bell's classification is widely used to classify the diseases: stage I is non-specific, and stage II is characterized by abdominal distension, wall edema, thrombocytopenia and metabolic acidosis. Radiologically intestinal and hepatic pneumatosis is usually observed. In stage III, signs and symptoms of stage II plus hypotension are present, signs of peritonitis, metabolic acidemia and shock are present. Radiologically, there is pneumoperitoneum (free air in the cavity) as an unequivocal sign of intestinal perforation (**Figure 5**) [25].

Currently, certain biomarkers have been valued in the diagnosis of NEC, which include C-reactive protein and pro-inflammatory cytokines (IL-6, IL-8, TNF-α). In addition, organ-specific markers can indicate damage to the enterocyte as proteins bound to intestinal fatty acids, hepatic, fecal calprotectin and claudin-3. It has been mentioned that the use of Doppler ultrasound is a useful tool to avoid unnecessary surgical operations, since it can verify perfusion of the intestinal wall and eventually identify necrotic intestinal segments before perforation occurs, a benefit that is not obtained with simple radiological studies [26].

3.4.2. Treatment

The initial treatment of patients with necrotizing enterocolitis includes fasting with gastric drainage, broad spectrum antibiotics, intravenous fluids according to the requirements and

Figure 5. Abdomino-thoracic radiography with total abdominal opacity known as "frosted glass sign," showing enterocolitis in its extreme stage.

water balances, correction of metabolic alterations and adequate management of acid-base imbalances, and inotropic support will be used when there is Clear signs of hypoperfusion.Mortality may range from 15 to 63% according to the revised series. Surgery is necessary in more than 50% of the cases, and the objective is to result in the least mutilation possible. Peritoneal drainage without laparotomy may be sufficient and has been reserved almost exclusively for those infants weighing less than 750 g or in those with increased intra-abdominal pressure and compromised ventilation. A laparotomy will be required in 74% of cases. In a typical scenario, there are two alternatives, to perform a primary anastomosis or an enterostomy. In our experience, a primary anastomosis should be performed, particularly if the patient's condition is good and the extirpation of the diseased bowel was extensive (**Figure 6**).

The prognosis of children affected by NEC is characterized by a morbidity of 20–50%; but in developing countries, this percentage is higher, and recurrence after the first event (4–10%), retarded growth (10%), bowel stenosis, mainly of the colon (12–35%), short bowel syndrome (20–35%), neurodevelopmental disorders (30–50%) and stoma complications (50%) are emphasized. The mortality ranges from 15 to 63% according to the series studied [27].

3.5. Neonatal hirschsprung disease

Hirschsprung disease (HD) or congenital agangliosis is a disease of unknown origin, first described in 1888, but it took 60 years for researchers in the field to find the first surgical measure that was more or less effective. This condition is relatively frequent, with one case for every 5000 births and with greater prevalence in males. This abnormality is characterized by the absence of enteric ganglion cells in the distal portion of the colon and, depending on the level of arrest of the neuronal migration, it has been classified as classic, when the ganglionic absence reaches up to 80% in the rectosigmoids and 17% involving sigmoids, splenic angle and transverse colon. The long segment is all forms that go beyond the blind pouch, and total colonic agangliosis is called total agangliosis coli and that which extends from the pectinate line to the duodenum is very rare [28].

Normally, the enteric ganglion cells, which emerge from the neural crest, must innervate from proximal segments of the digestive tract to the terminal colon, and this phenomenon takes place between 5 and 12 weeks of gestation. These cells are responsible for the mobility, not only of the small intestine but also particularly of the colon, which when absent makes the propulsive movements ineffective and the distal intestine function as a deposit. Regarding the etiology and pathogenesis of the disease, many studies have been done; however, conclusions have not been clear. It has been assumed that it appears as a consequence of defects in differentiation due to environmental changes, but there are also studies suggesting that it occurs due to an ectopic expression of class II antigens [29]. Likewise, the influence that genes could have on the disease has been speculated, and it has been found that when the extension is greater,
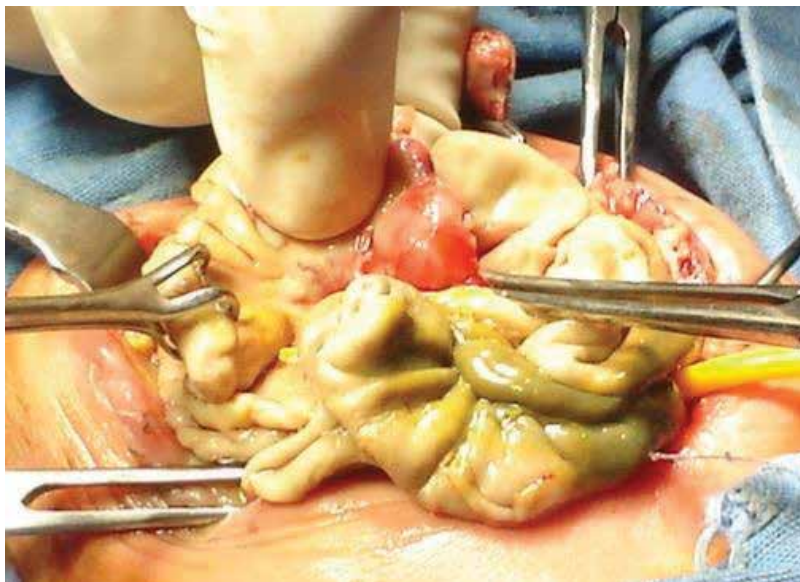

Figure 6. A transoperative image of the previous case. The contrast of the necrotic intestinal tissue with the normal appearance of the proximal (arrow).

the hereditary tendency increases. Thus, when agangliosis is total, the family tendency may be up to 50%, and if it is in the entire colon, the tendency descends to 15–21%. In relation to chromosomal phenomena, segregation studies have shown that Hirschsprung's disease is a genetic condition transmitted as autosomal dominant, autosomal recessive and polygenic forms. In one karyotype, an autosomal dominant gene was found to cause the disease in the chromosomes 10q11.2. [30]. At the experimental level, it has been found that agangliosis has been able to be reproduced in chicken embryos by extirpation of a segment of intestine, which stopped the neuronal migration [31]. In relation to genetic interference, it is known that the off-spring of a woman suffering from the condition is 360 times more likely to transmit the disease than the normal population. The most important constant is that the greater the extension of the aganglionar segment, the greater the risk of inheriting the condition. The conditions that usually accompany HD are neonatal appendicitis [32] and those it shares embryological origin with, such as Wardenburg syndrome, neuroblastoma, pheochromocytoma and Ondine's disease.

3.5.1. Diagnosis

Although HD can occur in non-newborn infants [33], it usually manifests and is frequently identified in the first days of postnatal life. An antecedent of delay in the first meconium emission is useful information, a fact that is usually present in more than 90% of the cases. In the case of a neonate who emits his first evacuation after 48 h of postnatal life and with the passage of days suffers constipation, it must be assumed that the cause is congenital agangliosis. Vomiting and abdominal distention are also typical, causing discomfort that can be improved with the application of a suppository or a probe. After showing improvement, they relapse. They are only able to be well for a few days and then suffer from the same picture. There is usually diarrhea, which is almost unequivocally a manifestation of enterocolitis, the most frequent complication of the condition. Radiological study, according to the majority of the authors, is very useful in more than 80% of the cases. A vertical study is recommended, and several findings can be highly suggestive: hydroaerial levels, edema of the intestinal wall, total pelvic opacity and intestinal dilatation. With these data, the next step is to perform the colonic enema (**Figure 7**), which should be done with water-soluble material, without previous enemas and with the introduction of a nelaton tube no more than 1 cm deep and manually gently inject the contrast medium, while obtaining an x-ray in the lateral position. Images such as jagged rectal wall, a transition zone between a contracted narrow distal portion and a proximal dilatation are highly suggestive. A late study is very useful at 24–48 h. The golden rule for making the diagnosis is undoubtedly rectal biopsy. Performed during or prior to the operation, it will reveal the absence of ganglion cells. Manometry is often useful, especially in older children. One should be very careful to not transgress the normally aganglionar area (a distance from the pectinate line of 10 mm) when performing the biopsy as it leads to false-positive interpretation.

3.5.2. Treatment

Non-surgical management is important prior to any procedure, since some of the parameters of the metabolic state of the neonate must be restored. Once the diagnosis is established, a transrectal catheter should be introduced if the patient is very distended and uncomfortable.

Figure 7. The colon shown by enema in lateral position. The spastic area is indicated by an arrow, which is equivalent to the aganglionic segment.

Antimicrobials are indicated as well as intravenous solutions suitable for the patient's age and condition. De la Torre and Ortega [34, 35] have proposed the surgical method of choice in these children, which should ideally be done during the first week of life and in a single operation, that is, without protective colostomy, in order to avoid the disadvantages of bacterial colonization. Unfortunately, this operation requires several requirements so that it can be implemented and three are the most important are to have a confirmed diagnosis within a few days of postnatal life, not to have an aganglionosis coli and to have a reliable pathologist, as much of the success of the procedure depends on them. It is widely demonstrated that at that age and with those requirements met, success is assured in more than 80% of the cases. The classic operations, Swenson, Duhamel and Soave, are left for later. The complications are diverse and include among the mediate complications, stenosis and fecal incontinence, and among the immediate ones, toxic enterocolitis, which is the one most responsible for death. It appears almost indistinctly as a violent infectious diarrheal syndrome, which immediately aggravates the condition of the baby, requiring a neonatal intensive care unit for correction, since the risk of death is very high.

3.6. Gastroschisis

Gastroschisis is a congenital defect of the abdominal wall consisting of evisceration of the midgut and other intra-abdominal organs, such as the stomach, through a small defect of 2–8 cm usually located to the right of the umbilical cord and in direct contact with the amniotic fluid, which gives them the characteristic serous aspect. It occurs between 5 and 10 weeks of gestation and can be identified in utero from week 20. The pathogenesis is still uncertain, with multiple theories. However, it is mentioned that it can be the result of a hemorrhage or thrombosis, which causes an alteration of the flow during the closing process of the wall, causing the exposure of the intra-abdominal organs. A significant increase in global prevalence has been observed in recent decades and ranges from 3 to 20 per 10,000 live births. This may be due to epigenetic changes [36, 37].

Multiple risk factors are mentioned: such as a mother younger than 20 years old or an adolescent, intake of certain medications such as acetylsalicylic acid, pseudoephedrine or ibuprofen, use of drugs, alcohol and tobacco, use of hormonal contraceptives, anemia, exposure to aromatic hydrocarbons, urinary tract infection and, in recent years, the intake of omega-6 fatty acids and alterations in lipid metabolism and pro-inflammatory cytokine substrates [38, 39].

Often the form of presentation of gastroschisis is sporadic and simple, that is, without major or minor malformations, but in a small percentage of cases, it may be part of other syndromes or chromosomal alterations, as well as other conditions such as skeletal dysplasias, Hirschsprung's disease, hydrocephalus and heart disease [40].

3.6.1. Diagnosis

Once the product is born, by either delivery or cesarean section, a preterm newborn between 35 and 37 weeks of gestation with low weight for gestational age is usually identified. In most cases, basic resuscitation is performed, and with respect to the local management of the defect, it is essential to immediately cover the exposed organs with a sterile, non-adherent, resistant, flexible and soft material to reduce loss of fluids and exposure of the peritoneal serosa to the environment. Avoid covering the exposed organs with gauze or compresses [41]. The most severe form of this malformation is a variety called "closed gastroschisis" (**Figure 8**), through the apparently integral abdominal wall, or with an millimetric orifice to the right of the umbilical cord, there is a black or green structure, turgid or fibrous, pediculate and mobile, of a soft consistency, 2–5 cm in length corresponding to the terminal ileum, cecum, cecal appendix and obliterated right colon. This segment is anchored to a fine fibrovascular network that, like a mesentery, keeps it irrigated. There is also intestinal malrotation and an umbilical membrane that is directed proximally toward a very dilated segment that sometimes ends in jejunal atresia variety IIIA. The other end of the digestive tract corresponds to the left colon, which is also obliterated. This occurs between 4.5 and 9% in cases of gastroschisis. There is always almost total absence of the jejunum and ileum [42]. Another variety is evanescent gut. In these cases, the gastroschisis is identified prenatally and at the moment of birth the abdominal wall is integrated; however, the patient presents intestinal occlusion data that requires an exploratory laparotomy that finds jejunal atresia with no more than 20 cm of small intestine and left colon. These children, in our experience, have a fatal outcome.

3.6.2. Treatment

It is important to initiate large amounts of intravenous fluids, electrolytes and antibiotics (**Figure 9**), place a 10–14 gauge Fr. orogastric probe, perform an evacuation enema with a warm solution at 10 ml/kg, avoid hypothermia, correct the acid-base state and, if necessary, provide ventilatory support. Regarding surgical treatment, there are two options: in the case of non-complex gastroschisis, the first alternative is umbilicoplasty, the introduction of exposed organs into the abdominal cavity without enlarging the defect, suturing the aponeurosis and skin and preserving the umbilical cord, preferably in the first 24 h of extrauterine life. We recommend that it should be attempted in all cases. However, success depends on whether there is viscero-abdominal disproportion, and therefore, it would not be possible to introduce all the intestinal loops or if there is compartmental syndrome. If so, a gradual, delayed closure is preferred between 6 and 10 days, covering it with a PVC membrane or a prefabricated silo. In a comparative study of the two surgical techniques, we found no difference, except that with umbilicoplasty, in-hospital stay time and that the days that the patients required mechanical ventilation and total parenteral nutrition were shorter than with delayed closure [6]. The complex varieties of gastroschisis always require a laparotomy, removal of the necrosed segment or atresia and a primary anastomosis between the residual and viable segments [42].

The prognosis in non-complex cases is generally good with a survival rate greater than 90%; however, in developing countries, mortality may be up to 60% of cases. The main complications are related to prematurity, sepsis, ischemia and intestinal perforation, renal insufficiency or multiple organ dysfunction as well as difficulties for enteral feeding. The prognosis in closed gastroschisis and evanescent intestine is fatal, and both varieties lead to short bowel syndrome, cholestasis, liver failure, sepsis and death.

Figure 8. The clinical aspect of a "closed gastroschisis" in which should be noted the absence of parietal defect, necrosis of the middle intestine and normal aspect of the distal colon, what we call "Antenatal-Paraumbilical-Intestinal-Strangulation" (APIS).

Figure 9. A clinical picture immediately after birth, in which evisceration of the stomach, a good part of the middle intestine and a small portion of the colon can be observed. There is no great serositis because the defect occurred late in gestation.

Acknowledgements

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Update on Neonatal Male Circumcision: A Public Health Perspective

Kriengkrai Srithanaviboonchai

Additional information is available at the end of the chapter

Abstract

Male circumcision (MC) is an effective preventive health intervention. WHO and UNAIDS jointly recommend that the international community considers MC as a potential long-term HIV prevention measure. Neonatal male circumcision (NMC) is a type of MC performed within 1 month after birth. There are several advantages in favor of NMC over circumcision at a later age; it is simpler, safer, and cheaper. Maximum benefits of MC are achieved through NMC. NMC is also more convenient and risk compensation after the surgery is unnecessary. Concerns over NMC include child rights, pain during the surgery, possibility of reduced sexual pleasure, and a long timeframe before achieving HIV reduction benefits. The local HIV epidemic and medical guidelines, policies and strategies, public education and demand creation, finance, readiness of health system, staff training, monitoring and evaluation (M&E), and quality assurance should be considered before and during the implementation of NMC. This chapter uses Thailand as an example of how a country might benefit from introducing NMC as a public health measure. Parents should be informed about the benefits and risks of NMC where service is available to allow them to decide whether their children should be circumcised.

Keywords: circumcision, neonatal male circumcision, HIV prevention, STI prevention, Thailand

1. Introduction

Whether to promote neonatal male circumcision (NMC) as a preventive health measure has long been a contentious subject among health professionals and the general public. NMC has been performed as a modern health intervention in English-speaking countries for health and hygiene since the mid-nineteenth century [1]. Although NMC lessened in popularity in the United Kingdom before World War II [2] and later in the United States over the last few

decades [3], recent findings in several randomized controlled trials (RCT) confirming that male circumcision reduces heterosexual HIV acquisition [4–6], have renewed interest in NMC and debates continue on whether it should be promoted as a public health measure.

NMC, similar to other clinical procedures, has associated medical benefits and risks. However, current academic debates seem riddled with subjective feelings on the issue rather than a dispassionate analysis of recent scientific findings. Internet searches reveal many scientific articles written from the point of view of certain mind-sets, either concurring with NMC or deprecating NMC. These biases are also true regarding dedicated websites discussing circumcision. These articles and websites fall prey to social acceptability biases, which are not surprising given the sensitive nature of MC due to religious beliefs, cultural or religious rites, and sexuality. This conflicting information creates a lot of confusion among physicians and parents of newborn males. Many parents decide not to circumcise their babies, while many NMCs are routinely performed without support of factual scientific knowledge.

Other factors must be considered besides the theoretical medical benefits and risks for each individual. These include the characteristics of targeted localities, for example, real-life clinical circumstances, readiness of health staff, the local HIV epidemic, finance and costs, and related laws and regulations. The unique situation of each area will determine whether NMC should be promoted as a public health measure. Ethical and legal issues such as child rights are also important to explore.

After circumcision was confirmed as an effective HIV prevention measure, the author led a series of studies to evaluate the possibility of using NMC as a public health measure in Thailand [7–10]. The results and conclusions of those studies have been included in this chapter. The lessons learnt from Thailand might be useful for other countries with similar contexts.

This chapter starts by elaborating on how NMC is different from other forms of circumcision and why it is worth be considered as a public health measure. Traditional circumcisions practiced under religious rituals are beyond the scope of this article. An objective summary of the pros and cons of NMC from up-to-date scientific evidence follows. This chapter touches upon important aspects of NMC as a public health measure. The author also discusses his views toward implementation of NMC impartially. Readers should consider this information with care and adapt it to suit their local context. The article ends with recommendations and conclusions. Finally, the author hopes that this article is valuable for those who hope to gain more insight on this very interesting health intervention.

2. Distinguish NMC from other forms of circumcision

2.1. Types of circumcision

Many varieties of circumcisions are currently performed. They can be classified according to their characteristics and purposes. Acknowledging the whole spectrum of circumcision will help distinguish NMC from other procedures. **Figure 1** shows how different types of the
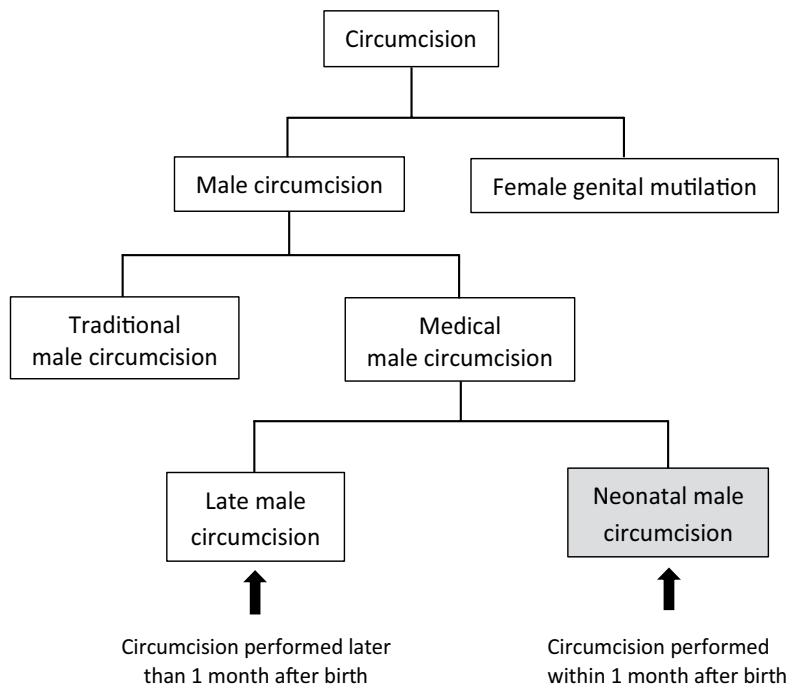
Figure 1. Schematic presentation of types of circumcision.

surgeries are categorized and pinpoints the whereabouts of NMC within the broad range of circumcision types. NMC is highlighted and discussed because of its favorable characteristics over other types of circumcision. It should be noted that other terms are used to refer to NMC such as "early infant male circumcision" and "newborn male circumcision."

2.1.1. Exclusion of female genital mutilations

First, the word "male" is intentionally added into the term to make sure that we are specifically talking about circumcision in males. All forms of female genital mutilations, described by some as female circumcision, procedures that intentionally alter or cause injury to the female genital organs for non-medical reasons [11] are excluded from this article. These procedures, which occur in some regions of the world, have no medical benefit and harm girls and women in many ways. The complications can be short and long term and include excessive bleeding, infection, urinary problems, keloid, sexual problems, and psychological trauma [12]. Female genital mutilations are a clear example of human rights violation. Collective efforts to prohibit and eliminate this practice are fully warranted.

2.1.2. Traditional MC

The prevalence of global male circumcision is estimated at 39% [13]. MC can be classified as traditional and medical. About half of the circumcisions are performed traditionally and are generally associated with certain religious or cultural beliefs [13]. In Muslim and Jewish

cultures, MC is mandatory and conducted as part of a religious ceremony. Circumcision in Muslim male child varies from birth to puberty. For instance in Thailand, Muslim boys are usually circumcised between the ages of 6 and 15. The circumcision rate among men in the Philippines is very high. Circumcision is performed as a rite of passage to welcome boys to the next phase in life and is believed to be a remnant of pre-colonial Islamic influence. Jews traditionally conduct a circumcision ceremony on the eighth day of a male infant's life. Traditional MC normally takes place outside of established medical settings. Those performing the circumcision mostly are not trained health professionals, so there are concerns about possible complications such as excessive bleeding and infections. Efforts have been made to reduce the risk of adverse events associated with traditional circumcision. Traditional MCs are ruled out from this article since they are inevitable and not considered public health interventions.

2.1.3. Medical MC

Medical MC was introduced in English-speaking countries in the mid-nineteenth century. The primary purpose was to reduce masturbation which was considered an unhealthy behavior [14]. It was also performed to improve genital hygiene since the foreskin was believed to serve as a pouch that allows the accumulation of smegma. Medical MC is usually performed in established health facilities by trained medical practitioners. It is generally carried out a few days after birth. The surgery is performed only if the infant is healthy and may be postponed if the infant has a medical condition. Currently, medical MC is relatively prevalent in the United States, Canada, and Republic of Korea. Almost universal MC in the Republic of Korea is the result of the influence of the United States [15].

2.1.4. Focus on NMC

MC performed under 1 year of age is called infant MC. NMC is a specific type of infant MC which is administered within 1 month of birth. There are several advantages in favor of NMC over circumcision at a later age. In this chapter, we focus on NMC performed as a preventive medical measure in established health facilities.

2.2. Why NMC is worth being considered as a public health measure?

2.2.1. NMC carries potential health benefits of MC

Since NMC is performed early in life, it generally includes all the potential health benefits of MC. In this section, proven medical benefits of MC are presented and discussed.

2.2.1.1. Improved genital hygiene

Penile cleanliness is easier to maintain for a circumcised penis because there is no pocket underneath the prepuce that needs to be exposed before cleaning, which could be problematic in young boys. While most children eventually learn to retract the foreskin and cleanse the area as routine hygienic practice before puberty, some might find this difficult. There is a great variability in when the foreskin is fully retractable, with about 40% of boys having

a fully retractable foreskin by age 10 [16]. Parents and boys are sometimes unclear whether they should try to force the foreskin open to clean the under area or leave it alone. This is not a dilemma for circumcised boys. The pouch under foreskin's inner surface is where smegma is compiled. Smegma is the accumulation of sebum combined with dead skin cells produced by the foreskin's inner surface. A build-up of smegma due to lack of routine cleaning may produce foul odors which are caused by the colonization of bacteria and chemical transformations.

2.2.1.2. Elimination of the chance to have phimosis and paraphimosis

Phimosis is defined as the inability to retract the foreskin over the glans penis in uncircumcised males. Male infants are born with congenital physiologic phimosis resulting from adhesions between the epithelial layers of the inner prepuce and glans. As mentioned earlier, this condition will go away in most boys with age after intermittent foreskin retraction and erections. Un-retractable foreskin that occurs after previously retractable or after puberty is considered a health problem and called pathologic phimosis when nonretractability is associated with local or urinary complaints caused by the phimotic prepuce [17]. Most pathologic phimosis in adults is the result of distal scarring of the foreskin due to poor hygiene, balanitis, balanoposthitis, and forceful retraction of the foreskin. It can also occur because of infections or inflammations. For the elderly, increased risk of phimosis is caused by the loss of skin elasticity and infrequent erections. Symptoms of phimosis are difficulty or pain during urination, collection of urine in prepuce, painful erection, and paraphimosis.

Paraphimosis is the condition where the foreskin is trapped behind the glans penis for a long time and can no longer be pulled forward over the tip of the penis. Paraphimosis is common among children who have forgotten to retract their foreskin after voiding or bathing [18]. Other causes of paraphimosis are infection, physical trauma, trying to retract the foreskin back too forcefully, and leaving the foreskin in a pulled back position for an extended period of time. Persons who have phimosis are at risk for developing paraphimosis. When paraphimosis occurs, the prepuce and the distal part of glans may be swollen and painful. Paraphimosis is considered a medical emergency as blood supply to the tip of the penis is diminished [18]. The condition requires immediate medical attention. If left unattended, paraphimosis may lead to serious complications such as severe infection and loss of the penis due to gangrene.

2.2.1.3. Reduction of balanitis and exclusion of balanoposthitis

Balanitis refers to inflammation of the glans penis and can occur at any age. Data from meta-analyses showed that circumcised males have a 68% lower prevalence of balanitis than uncircumcised males [19]. Symptoms may include tight and shiny skin on the glans, redness on the glans, itchiness, unpleasant smelling discharge, painful urination, and localized pain. There are many possible causes of balanitis including poor hygiene, irritation, physical trauma, skin conditions, phimosis, and various infections. Pathologic phimosis is a possible complication of balanitis. It can occur especially when balanitis is frequent since the preputial orifice may be scarred and reduced elasticity. Meatal stenosis is another possible complication of balanitis but is uncommon.

If the foreskin is also affected together with the glans penis, the condition is called balanoposthitis [20]. Therefore, balanoposthitis affects uncircumcised males only. Balanitis usually leads to balanoposthitis except in circumcised males. Signs and symptoms of balanoposthitis are similar to those of balanitis only if they also involve the foreskin, not just the glans. Diabetes is an important underlying cause of both balanitis and balanoposthitis especially in the patients with poorly controlled blood sugar [21]. Prophylaxis circumcision reduces the risk of balanitis and eliminates the chance of developing balanoposthitis. Circumcision is also an effective treatment for both conditions.

2.2.1.4. HIV reduction

Reduction of the risk of HIV acquisition during heterosexual sexual intercourse is perhaps the most significant and most discussed benefit of MC. Researchers have speculated about this benefit early on in the HIV epidemic as it was observed that the rates of circumcision inversely correlated with the rates of HIV infections [22, 23]. In Asia, for example, the prevalence of HIV was high where the rate of circumcision was low (e.g., Thailand and Cambodia) and the prevalence of HIV was low where the rate of circumcision was high (e.g., the Philippines and Korea).

Three randomized controlled trials conducted in South Africa [4], Kenya [5], and Uganda [6] later confirmed that MC reduces the risk of female-to-male HIV transmission by 51–60%. All three studies were stopped early by their respective data and safety monitoring boards due to the obvious differences in HIV incidence between the intervention and the control arms. The protective effect seemed to be sustainable as the effect was maintained at 58% for 72 months of follow-up compared to 60% at 24 months of follow-up among Kenyan trial participants [24]. MC is seen as surgical vaccination as it can be done once and does not rely on consistent health behaviors.

There is scientific evidence that explains why circumcised males would have lower risk of acquiring HIV infection through heterosexual intercourse. Unlike the glans penis, the inner surface of the prepuce is lined with mucosal epithelium with no protective keratin layer. Histologically, the lining of inner foreskin is similar to the lining of nasal mucosa and vagina which are the common entry points of infectious organisms. Thin mucosal epithelium and lack of a protective keratin layer also make the foreskin more susceptible to minor trauma during sexual intercourse [25]. Therefore, the existence of foreskin serves as an entry point for HIV. Langerhans dendritic cells are antigen-presenting immune cells. They are abundant close to the mucosal lining surface of the inner foreskin [26]. In general, their primary function is to take up and process microbial antigens to become fully functional antigen-presenting cells. Langerhans cells in the foreskin and other HIV target cells are the major targets for the HIV, since they have surface CD4 receptors and cofactors that HIV bind to when infecting cells. It is possible that HIV may stay alive longer in the preputial cavity between the non-retracted foreskin and the glans penis since the micro-environment is suitable for its survival [25]. Lower rates of other sexually transmitted infections (STIs) among circumcised men may indirectly reduce the risk of HIV infection [27]. Results from an RCT found a reduction of symptomatic genital ulcer disease and herpes simplex virus type 2 (HSV-2) infections due to circumcision accounted for an 11.2% and 8.6% reduction in the contraction of HIV infection, respectively [28].

2.2.1.5. Reduction of other STIs

Reduction of the risk of acquiring other STIs in circumcised men is less pronounced compared to HIV. Early observational studies revealed conflicting results. Respectable information came from a meta-analysis that concluded that circumcised men are at lower risk for chancroid and syphilis [29]. RCTs conducted in Uganda and South Africa found a 35% and 34% lower prevalence of high-risk HPV genotypes in circumcised men [30, 31]. The study in Uganda also found a 28% lower incidence of herpes simplex virus type-2 (HSV-2) [31]. The South African trial also found protection for *Trichomonas vaginalis* [32]. There is no concrete evidence supporting the preventive effect of MC on the risk of contracting gonorrhea or Chlamydia.

The female partners of circumcised men receive indirect health benefits from MC. Female partners had a lower prevalence of genital ulcers, *T. vaginalis* infection, and bacterial vaginosis compared to female partners of uncircumcised men [33].

2.2.1.6. Cancer reduction

Penile cancer is quite rare in developed countries but is more prevalent in developing countries [34]. Being uncircumcised is a strong risk factor for penile cancer. A systematic review found a 67% reduced risk of invasive penile cancer in circumcised men compared to uncircumcised men [35]. This preventive effect probably occurs through the elimination of phimosis, a strong risk factor for penile cancer [36]. Another explanation is that circumcised men are less likely to acquire HPV as mentioned earlier. High-risk HPV is suspected to be involved in the causation of penile cancer as it is found in a large proportion penile cancer cases [37]. The odds of detecting HPV are lower in circumcised men compared to uncircumcised men [38]. Chronic relapsing balanitis and balanoposthitis due to poor hygiene and circumcision may also increase the risk of invasive penile cancer.

It is well established that most cervical cancer cases are caused by high-risk HPV. Given that circumcised males are less likely to contract HPV, their female partners also are less likely to be infected with HPV. There is evidence showing that these associations help reduce the risk of cervical cancer. A meta-analysis of case-controlled studies found that monogamous women whose male partners had six or more sexual partners and were circumcised had a lower risk of cervical cancer than women whose partners were uncircumcised [38].

2.2.2. Advantages of NMC over circumcision at later age

There is unanimous consensus from the scientific community that MC, if implemented as a non-therapeutic preventive health measure, should be done as early as possible. The procedure is preferably performed within a few days after birth for healthy boys. Following are the list of advantages supporting the rational to perform MC early in life rather than waiting.

2.2.2.1. Maximum benefits of circumcision are achieved

Late circumcision reduces the risk of urinary tract infections (UTIs) which occur mostly during the first year of life by almost 10 times [39]. Thus, there is a loss of this preventive health

benefits when circumcision happens late in life, from adolescent onward. Protection against UTIs and kidney damage in infancy is lost if it is not performed during the first year of life.

Avoiding childhood phimosis and balanoposthitis is also lost if circumcision is not performed early. Some benefits associated with STI prevention would be lost if circumcision is done after sexual debut. As penile cancer is associated with phimosis and HPV infection, late circumcision means increased risk of cancer.

From a public health perspective, a disease prevention measure has to be implemented on a scale large enough to have an impact at the population level. Circumcision could resemble a vaccination program against HIV. The indirect benefits of MC in women would not occur unless a sufficient number of men are circumcised to allow the effect of herd immunity [40]. Higher MC coverage can be achieved easier through NMC. All pregnant women and their husbands could be educated about NMC during antenatal care sessions and decide about the procedure prior to delivery. Circumcisions can be conducted within a few days of birth for healthy infants while the mother recovers from labor and delivery. This would allow mothers and infants to be discharged together.

2.2.2.2. Simpler, safer, and cheaper

Circumcision is much easier to perform during the neonatal period than at a later age. General anesthesia is not required which diminishes the possible adverse events associated with it. The procedure also takes less time, usually just a few minutes to complete. The tissues and blood vessels involved are so tiny that there is no need for stitches.

Bleeding and infection are the two main serious side effects of MC. When performed by trained health personnel at well-equipped health care facilities, NMC is safe and has a low rate of complications [41].

It is cost saving to conduct MC during the neonatal period than at an older age. A study in the US found that NMC was about 10 times cheaper than circumcision performed later [42]. This advantage is especially important in developing countries.

2.2.2.3. More convenient

Circumcising infants during their first few days of life is a lot more convenient than to circumcise older boys or adolescents. Neonates are ready for surgery and would not require the counseling required for older boys. There is no need to do HIV counseling and testing for newborns, since they are considered HIV negative, except for those born to HIV-infected mothers. Older boys need to be informed of the benefits and risks of the procedure and must give their informed consent. Confounding factors include fear of the surgery and psychological difficulties. If the procedure is done during school age, students will have to take time off school. Healing is also faster at around 1 week for NMC compared to at least 2–3 weeks for circumcision in adults [43]. Sexually active persons must abstain from sexual intercourse for 6 weeks to ensure proper healing. Having sex during this period would make patients prone to infections including HIV thus negating the main benefit to be gained from procedure.

Other benefits of NMC over circumcision at later age include no long-term memory of the surgery and a better cosmetic outcome.

2.2.2.4. No risk compensation

Risk compensation is a phenomenon by which people adjust their health behavior in response to the perceived level of risk. People become more careful where they sense greater risk and less careful if they feel more protected. Sexually active males who were circumcised as an adult may engage in greater sexual risks due to a perception they have less HIV risk following MC. This could occur among female partners of circumcised men as well if they perceive lower risk. There is little chance of risk compensation for NMC. Boys who are circumcised very early in life would not sense any change in risk as they have been circumcised their whole lives.

2.2.3. Concerns over MC

Besides the low probability of medical risks associated with the surgery such as bleeding, infection, and unsatisfactory cosmetic result, several concerns have been raised. Some of the issues raised are considered controversial, while others have no evidence to support them. In following section, each topic is discussed citing the ongoing conversation and debate as well as up-to-date scientific information.

2.2.3.1. Ethical issues

Since the surgery is performed on an infant who cannot provide consent, NMC has repeatedly raised ethical-related concerns [44]. The decision to circumcise children is usually taken by the parents who act in their child's best interests. People argue that the authority to perform interventions on a child should be limited to ones proven to be medically necessary. Scholars who do not view NMC as a necessary medical intervention suggest that it should be delayed until boys are mature enough to decide for themselves. Thus, this ethical issue boils down to whether people perceive NMC as a medical necessity based on the available scientific evidence. The guidelines and recommendations issued by relevant authorized bodies and medical committees might help determine its necessity.

There are a lot of discussions and debates in the literature whether NMC is a violation of child rights to bodily integrity [45–49]. The principle of bodily integrity refers to the right of each human being to autonomy and self-determination over their own body. Scholars and activists who favor an intact penis and oppose circumcision promote a concept of "Genital Integrity" which refers to the condition of having complete and unaltered genital organs. In their view, NMC is an unconsented physical intrusion and a human rights violation.

2.2.3.2. Pain during NMC

There is concern that NMC introduces unnecessary pain to the newborn. Children who have undergone NMC do experience pain as evidenced by increased heart rate, decreased oxygen

saturation, and facial expressions indicative of pain during the procedure [50]. Pain experienced during the procedure has long-lasting effects on the circumcised child. Circumcised infants have been observed to have a stronger pain response to subsequent routine vaccination than uncircumcised infants [51]. Various interventions, in single and in combinations, have been used to minimize pain during NMC, e.g., sucrose syrup, oral acetaminophen, topical analgesic cream, and local nerve block. Sucrose alone has not been proven effective in reducing pain from circumcision [52], while topical analgesia may have some effect [53].

2.2.3.3. Reduced sexual pleasure

There are abundance of neurones which are sensitive to the touch in the foreskin. This leads many to believe that circumcised men might have less sexual pleasure than uncircumcised men [54]. This issue is not limited to NMC but is relevant to circumcision at any age. However, most studies testing this hypothesis have not found this to be true. Recent reviews concluded that loss of the foreskin by circumcision had no adverse effect on sexual pleasure during sexual acts [55, 56].

2.2.3.4. Effectiveness in preventing HIV in men who have sex with men (MSM)

MC reduces the risk of heterosexual HIV transmission in men, but its effect on male-to-male sexual transmission is uncertain. As the current HIV epidemic is concentrated in men who have sex with men (MSM) in many parts of the world, the lack of evidence to support that MC could reduce the risk of HIV acquisition among this population raises concerns over the implementation of this measure as a public health intervention. Observational studies on the protective effect of MC against HIV infection among MSM revealed conflicting results, with some studies showing positive results [57, 58] and others negative results [59–61]. A systematic review concluded that MC might offer HIV protection only among MSM who practice primarily insertive anal sex, but not for those who practice primarily receptive anal sex [62]. An RCT is needed to confirm this finding.

2.2.3.5. Long timeframe to see the HIV reduction benefits

Another concern on the implementation of NMC is a long waiting period to see the HIV prevention effect. Since NMC is conducted among newborns, it may take at least 15 years before they are sexually active and for the NMC to yield HIV prevention benefits. However, if this public health intervention is to be done, sooner is better than later. The only intervention that could have yield similar effect on the HIV epidemic is a preventive vaccine. According to the current status regarding HIV vaccine development, it is probable that NMC will yield an effect before an HIV vaccine is available.

3. NMC as a public health measure

A lot of factors come into play in deciding whether to promote NMC as a public health measure in a country or at a specific locality. Following are the issues that need to be considered before implementation of this health intervention.

3.1. Local HIV epidemic

Promoting NMC as a public health measure depends largely on the characteristics of the local HIV epidemic. Generally, the main reason to step up NMC service in health care facilities is to reduce the risk of HIV infection in the population. It may be not worth promoting NMC, if HIV is not a major public health problem in the country or of the target locality. As MC is proven to reduce only the risk of HIV acquisition through heterosexual transmission, it will not be appropriate to promote NMC where new HIV infections occur mainly through other routes of transmission, namely through same sex intercourse in males or intravenous drug use. An economic study using information on the local HIV epidemic as part of the analysis would help determine if NMC is justifiable for the country in the event that the country has to bear the majority of the costs of the intervention.

3.2. Medical guidelines on NMC

Soon after MC was proven to reduce the risk of HIV contraction among heterosexual males, global health authorities such as the World Health Organization and the Joint United Nations Programme on HIV/AIDS recommended that MC, including NMC, be adopted as part of comprehensive national HIV prevention strategies in countries with high prevalence of heterosexually transmitted HIV infection and low rates of male circumcision. The American Academy of Pediatrics determined that the preventive benefits of neonatal circumcision outweigh the risks of the procedure and should be offered to the families who have boy infants [63]. This statement has also been endorsed by the American College of Obstetricians and Gynecologists [64] and the American Urologic Association [65].

While these international recommendations influence some local practitioners, most health care personnel need national guidelines on NMC before they can start providing the service. Hence, local medical professional organizations should be included as important stakeholders while planning for the intervention. Their statements in favor of NMC would assist program implementation in a big way.

3.3. Policies and strategies

Clear policies and strategies will guide the respective details regarding NMC service. The magnitude and characteristics of the program will depend on the policies from a high-level authorized body, usually the Ministry of Health. The NMC implementation strategy can range from promotion of routine NMC to offering NMC only on a case-by-case basis or per request. The overall strategic plan should translate into action plans for each time interval including target numbers of NMC. The policies will also clearly specify the financial aspect of the program mainly on the payment scheme for the service.

3.4. Public education and demand creation

The general public needs to be educated about NMC before the service is promoted. Public education can be rolled out in a variety of ways. Educational materials should be straightforward, comprehensive, and attractive. The main contents of the media should include benefits

and risks of NMC as well as other supporting information such as availability and cost. The design of the education campaign should follow dedicated steps starting with a formative research targeting different audiences such as the general population and pregnant mothers. The formative research and subsequent baseline survey will define the content of the message and explore its suitability along with other relevant issues such as the dissemination channels.

Clear and scientifically accurate information about NMC can prevent misconceptions and rumors about the procedure in the community. Perceived benefits of NMC at the societal level together with high level of satisfaction among early adopters will help increase the demand for the services.

3.5. Finance

NMC represents an added expenditure for the health care system even though the unit cost of NMC is lower than for circumcision at a later age. How the expenses are covered will depend on the nature of the program. However, the parents of the newborns should not have to pay for the total cost if NMC is promoted as a public health intervention. In developing countries, where resources are limited and a high number of recruitments are targeted in a short period of time, local governments usually do not have to pay, as the programs are supported by international agencies. In some other circumstances, the health insurance system may pay for the service. Partial payment by the parents is another possible option.

3.6. Readiness of health system and staff training

A baseline assessment of the readiness and capability of the health system should be conducted. For health facilities, the issues to be assessed should include the antenatal and child delivery service and availability of necessary medical equipment and supplies. Knowledge and attitudes toward NMC should be explored among health care personnel. The information gained from the surveys will help identify the basic elements needed for the service that are still lacking and the aspects that should be emphasized while training health care personnel.

Safety is the first priority when implementing NMC as a public health measure. The surgery should be performed by trained health staff. The health staff who performs the surgery should be educated comprehensively on all aspects related to delivery of NMC, and not just the operation. A training package on NMC developed by the WHO and its partners is available online [66].

3.7. Monitoring and evaluation (M&E) and quality assurance

A system to routinely monitor the performance of the NMC program should be established. The indicators should cover both medical and public health aspects of the intervention. Newly trained staff should conduct first NMC cases under the supervision of more experienced persons. The results of the monitoring system will indicate whether the program is being implemented as intended and should be used to adjust the program to meet the goals in a timely manner. On an interval basis, the outcomes of the program should be comprehensively

evaluated. The evaluators could be from internal and/or external entities as appropriate. The information from the M&E system should be analyzed and used to improve the service and guarantee the quality of the service.

4. Thailand and NMC

Soon after it was confirmed that MC was effective in reducing the heterosexual transmission of HIV among men, the author formed a research team at Chiang Mai University, in Northern Thailand, to investigate the possibility of adopting NMC as a public health intervention in Thailand. A critical review compiling related information and the author's thoughts was the first publication [7]. A series of surveys have been conducted to gain insight on related issues in the country including knowledge and opinions of health staff on the intervention [10], the experiences of health staff on the procedure and capability of the Thai health care system to implement NMC [8], and acceptability as well as concerns of NMC among postpartum mothers [9].

Following are the findings of the abovementioned studies. This case study can serve as an example for other countries that share a similar context to Thailand and are considering adopting NMC as a public health measure.

4.1. Background on Thailand

Thailand is an upper middle income country [67] and located in Southeast Asia. The country was categorized as a "high" human development country according to the most recent Human Development Index (HDI) report [68]. Thailand has a well-established health care system. The infant mortality rate is considered low at 11:1000 live births [69]. The coverage of antenatal care among pregnant women and rate of delivery at the health facilities are high. The country has three major health insurance systems that cover almost all of its citizens namely the Universal Health Coverage Scheme, the Civil Servant Medical Benefit Scheme, and Social Security Scheme. People who can afford or have private health insurance could go to private hospitals for convenience.

Thailand was one of the Asian countries hardest-hit by the HIV epidemic during the early 1990s. The AIDS Epidemic Model (AEM) estimates that more than 1 million Thais were infected by the HIV virus. High quality and good coverage of antiretroviral treatment has saved a lot of lives. It is estimated that there are currently more than 400,000 people living with HIV in the country [70]. Thailand's MC rate in general is quite low [13]. It is prevalent only in Muslim communities especially in the southern most provinces.

4.2. Why should Thailand consider promoting NMC?

Thailand is a good candidate country for promoting NMC as a public health measure for many reasons. Most HIV infections in Thailand occur through heterosexual transmission, of which the risk is substantially reduced by MC. Theoretically, the effect of MC on HIV risk

reduction should be of significant magnitude in countries where the baseline MC rate is low and achievement of high coverage of MC is possible. Thailand has a relatively strong health care infrastructure as evidenced by the relatively low infant mortality rate [69]. An NMC program could possibly be built on this existing capability. The country has high coverage of maternal and child healthcare services including family planning, antenatal care, and deliveries that almost universally occur in the hospitals [71]. Thailand was the first developing country to eliminate mother-to-child HIV infection [72]. Health care personnel can educate pregnant mothers and their families about the benefits of having their children circumcised. Offering NMC could occur during the antenatal period if the fetus is identified as a boy intrapartum. Mothers can also be advised on how to care for their child after NMC and can be discharged from the hospital with the infant. Lastly, most Thais are Buddhists. Buddhism does not have any prohibition or negative beliefs toward circumcision. Further details on this issue were published in a critical review [7].

4.3. Readiness of the Thai health care system to implement NMC

A nation-wide survey was conducted in 2011 to gather baseline information on the capability of hospitals in Thailand to provide NMC [8]. Two questionnaires were sent to all hospitals in Thailand providing obstetric services and considered potentially able to perform NMC. The first questionnaire requested information about the facility's characteristics and its provision of NMC in 2010. The second questionnaire, directed to doctors or nurses who are familiar with delivery and postpartum care, contained opinion questions about NMC, and whether the procedure should be offered in the respondents' hospital. Of the hospitals that had deliveries in 2010, only 8.2% provided at least one NMC. Thirty-eight percent of private hospitals and 2.3% of government hospitals provided the service during 2010. The primary reason for performing NMC was parental request. Only a minority of the respondents thought that NMC was easy to perform (31.3%), was safe (39.1%), and should be offered in their health care facilities (29%). Ninety-two percent stated that physicians should perform the procedure instead of nurses. When asked about who should decide whether or not to conduct NMC, 55% indicated the choice should be left to parents. Forty-three percent believed that the service should be free of charge, while the same proportion felt that the parents should pay for some or all of the cost.

In another study conducted during 2011–2012, Thai health care providers' knowledge and attitudes toward NMC were assessed using face-to-face interviews [10]. The participants were physician administrators, practicing physicians, and nurses whose jobs involved NMC clinical procedures or oversight. The subjects were drawn to represent various hospital sizes and regions of the country using a multi-stage sampling technique. The participants were initially asked whether they agreed that MC had an effect on HIV prevention. Subsequently, printed educational materials on the benefits of NMC were presented to the participants for review. The participants were then asked whether NMC should be implemented at their facilities. Of the 133 health staffs who participated in the study, only 38% initially agreed that NMC reduced the risk of sexual HIV transmission. After reviewing the written information about the benefits of NMC, 59% of the participants thought that NMC should be offered in their

hospitals. HIV aside, 96% recognized the benefits of MC on hygiene, 74% knew about the effect of MC on cancer prevention, and 65% recognized that NMC could prevent STIs. Major concerns about NMC raised were safety and child rights. Physicians and nurses who had previous experience in performing circumcision on patients of all ages were more reluctant to have NMC performed in their hospitals.

4.4. Thai postpartum mothers' acceptability of NMC

In 2011–2012, a survey was conducted among 593 postpartum Thai mothers to determine their perceptions, concerns, and acceptability of NMC [9]. The study found that 70% of postpartum mothers knew or had heard of MC. Their biggest concerns were safety and pain of the procedure. After receiving information about the benefits and risks of NMC, one-third of the participants would choose to have their infants circumcised, one-third would not allow their sons to undergo NMC, and the last third were undecided. Mothers were the most influential person in deciding about NMC followed by fathers. Having a higher level of formal education, a circumcised husband, and knowing of at least one circumcised child were independent predictors of acceptability of NMC among postpartum mothers.

4.5. NMC in Thailand, the way forward

The study showed that NMC was rarely performed in government hospitals where the intervention meant to take place. The health care staffs were unaware of the health benefits of NMC. Most health staffs were concerned about the difficulty and safety of the procedure. However, the fact that a large proportion of health staff agreed that NMC should be offered in their health facility after being educated about the benefits of NMC revealed an opportunity for the program to be adopted by the health staff. The majority of postpartum mothers did not know about the benefits of NMC and were concerned about safety issues. The results suggest that NMC may be culturally neutral with evidence that even modest educational efforts can impact mothers' decisions in favor of NMC.

An education campaign on NMC for health professionals and general public is needed before NMC is promoted as a public health practice in Thailand. An economic study demonstrating the cost-effectiveness of NMC in Thailand is also needed in order to effectively advocate for policies to introduce NMC as an established, offered in health facilities.

Though Thailand might gain considerable benefits from the implementation of NMC as a public health measure, its actual occurrence seems to be impassable according to our research findings. Knowledge and technologies on HIV prevention have evolved rapidly and there are a lot of other options to choose from. For Thailand, other interventions are regarded as more attractive strategies, e.g., routine HIV testing among key populations at higher risk of HIV infection, treatment as prevention, and pre-exposure prophylaxis. Another reason why NMC is overshadowed by other interventions is the lack of proof of an HIV prevention effect on MSM who are the current target population. Unless all necessary things have been done, NMC will not be administered as a public health intervention in Thailand and will only be performed by request and on a case-by-case basis.

5. Conclusion

NMC is as an effective preventive health measure. The medical benefits of NMC outweigh the risks, especially after the effect on HIV prevention has been added. Nevertheless, NMC has not been used to its full potential due to controversies and concerns over the subject. Many issues need to be addressed, if NMC is to be implemented as a public health measure. Implementation of NMC shall vary according to the local context. Parents should be informed about the benefits and risks of NMC where the service is available so that they can decide whether their children should be circumcised. NMC is and will be a health intervention under debate in the years to come.

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Miscellaneous Topics in the Field of Neonatal Health

Neonatal Gene Therapy for Inherited Disorders

Koichi Miyake, Noriko Miyake and Takashi Shimada

Additional information is available at the end of the chapter

Abstract

In spite of developments of neonatal intensive care medicine, it is still difficult or impossible to treat several inherited genetic disorders using conventional pharmacological methods. Gene therapy is a promising alternate approach for treating a variety of genetic disorders. By the time the patient reaches adulthood, however, it is often too late for effective treatment. But in several of these cases, neonatal gene therapy appears potentially useful against inherited disorders that are not obviously treatable through any other methods. This chapter describes the strategy for neonatal gene therapy for inherited disorders and presents preclinical neonatal gene therapy data for two inherited disorders, metachromatic leukodystrophy and hypophosphatasia. We also discuss the utility, advantages, problems and potential of neonatal gene therapy for inherited disorders.

Keywords: neonatal gene therapy, AAV vectors, metachromatic leukodystrophy, hypophosphatasia

1. Introduction

Although there have been significant advances in neonatal intensive care medicine, several neonatal disorders remain major causes of mortality and morbidity. Consequently, there is an urgent need for development of new safe and effective therapies to improve the outcomes of these intractable and devastating neonatal disorders. Gene therapy is an exciting and promising approach to treat many diseases for which there are still no effective therapies. To date, more than 2400 clinical trials of gene therapy protocols have been attempted in effort to treat various genetic diseases as well as many types of cancers and infectious diseases (http://www.abedia.com/wiley/continents.php). The results of preclinical studies suggest that neonatal gene therapies represent potentially effective treatments for currently intractable neonatal

disorders [1–6]. However, although neonatal gene therapies have several advantages over similar therapies used in adult patients, there is as yet no clinical protocol for use of gene therapy in newborn infants. This chapter describes a strategy for the use of neonatal gene therapy in the treatment of inherited disorders and presents preclinical neonatal gene therapy data for two inherited disorders, metachromatic leukodystrophy (MLD) and hypophosphatasia (HPP). We also discuss the utility, advantages, problems and the potential of neonatal gene therapeutic approaches for the treatment of inherited disorders.

2. Adeno-associated virus-mediated gene transfer to neonate

Among the numerous viral and nonviral vectors that have been developed to deliver genes of interest into target cells, adeno-associated virus (AAV) vector has emerged as a particularly promising tool for gene delivery, thanks to its safety (AAV is not pathogenic) and its ability to transduce nondividing cells [7–9]. We are now using several AAV vector serotypes (mainly 1–12), depending on the target [10–13]. **Figure 1** shows the results after intravenous injection into neonatal
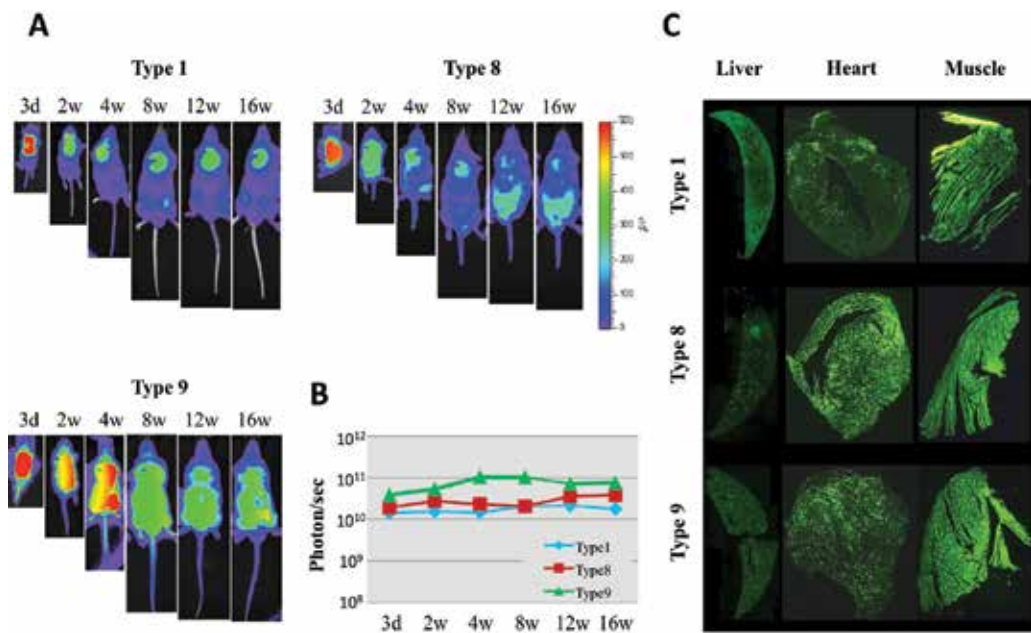
Figure 1. Systemic intravenous injection of AAV vectors into neonatal mice. (A) Approximately 5.0×10^{11} vector genomes (vg) of recombinant AAV vectors encoding the luciferase gene (AAV/Luc) (serotype 1, 8, 9) were injected into the external jugular vein of neonatal mice using a syringe with a 29-G needle. Bioluminescent images of mice were obtained using a Xenogen IVIS imaging system 3 days and 2, 4, 8, 12 and 16 weeks after administration. Color scale bar indicates radiant efficiency (photons s^{-1} cm^{-2} steradian^{-1} per μW cm^{-2}). (B) Radiant efficiency of serotype 1 (blue), 8 (red), and 9 (green) AAV vectors injected mice was quantified. (C) Approximately 5.0×10^{11} vg of AAV vectors encoding green fluorescent protein (serotype 1, 8, 9) were injected into the external jugular vein of neonatal mice. Sixteen weeks after injection, liver, heart and muscle were stained with anti-GFP antibody.

mice of AAV vector serotypes 1, 8 and 9, harboring the luciferase gene. Expression of luciferase was detected within 3 days and continued for more than 16 weeks with no decrease in expression. Serotype 9 mediated the highest expression during the observation period (**Figure 1A, B**). In addition, using an AAV vector encoding green fluorescent protein (GFP), we determined that the organs most efficiently transduced are the liver, heart and muscle (**Figure 1C**). Moreover, although transduction efficiency was not as high, the central nervous system (CNS) was also transduced after intravenous injection of AAV vector, which apparently passes through the blood-brain barrier (BBB) [14] in neonatal mice [15]. Thus, a systemically administered AAV vector was able to transduce several important target organs in neonatal mice, including the CNS, and mediate expression of a gene of interest for a prolonged period of time.

3. Advantages of neonatal gene therapy

Systemic gene transfer to neonates has several advantages over treatment of the adults (**Table 1**). First, as mentioned above, neonatal gene therapy has the potential to overcome the limitation imposed by the BBB on treating genetic disorders of the CNS. Because the BBB is developmentally immature during the perinatal period, AAV-mediated neonatal gene therapy is a highly promising strategy for treating genetic neurological diseases. Second, because the immune system is immature, neonates are immunologically tolerant of the transgene and/or viral vector [16–18]. Immune rejection of the transgene product by neutralizing antibodies is a severe problem for gene therapy in adults. Third, treatment administered soon after birth may enable prevention of early-onset genetic disease. Finally, neonates can be effectively treated with a smaller amount of viral vector than adults. Using smaller amounts of viral vector is superior with respect to both safety and cost. Taken together, these advantages make systemic neonatal gene therapy a promising method for treating systemic genetic diseases.

- Penetrates the blood-brain barrier
- Induces immune tolerance
- Prevents early-onset genetic diseases
- Enables the use of smaller amounts of vector

Table 1. Advantages of neonatal gene therapy.

4. Application of neonatal gene therapy

4.1. Neonatal gene therapy for metachromatic leukodystrophy

Metachromatic leukodystrophy is an inherited, autosomal recessive lysosomal storage disease (LSD) caused by a deficiency in the lysosomal enzyme arylsulfatase A (ASA), which

catalyzes the degradation of galactosyl-3-sulfate ceramide (sulfatide (Sulf)), a major myelin sphingolipid [19]. This disease is characterized by myelin degeneration, mainly in the CNS, and clinically by progressive motor and mental deterioration that is ultimately lethal. Therefore, the major target organ for treatment of this disease is the CNS, and the aim is to arrest or reverse the progression of the neurological symptoms. A major obstacle, however, is the BBB, which limits delivery of systemically administered therapeutic molecules to the brain [14]. It is therefore hoped that systemic administration of an AAV vector harboring ASA during the neonatal period would be useful for treating the CNS. We previously showed that a single systemic injection of AAV vector encoding human ASA (AAV/hASA) into neonatal ASA knockout (MLD) mice results in the wide distribution of ASA in the brain and correction of the biochemical and neurological phenotypes [20]. **Figure 2A** shows that a single systemic injection of AAV/hASA enables transduction of the CNS in neonates but not
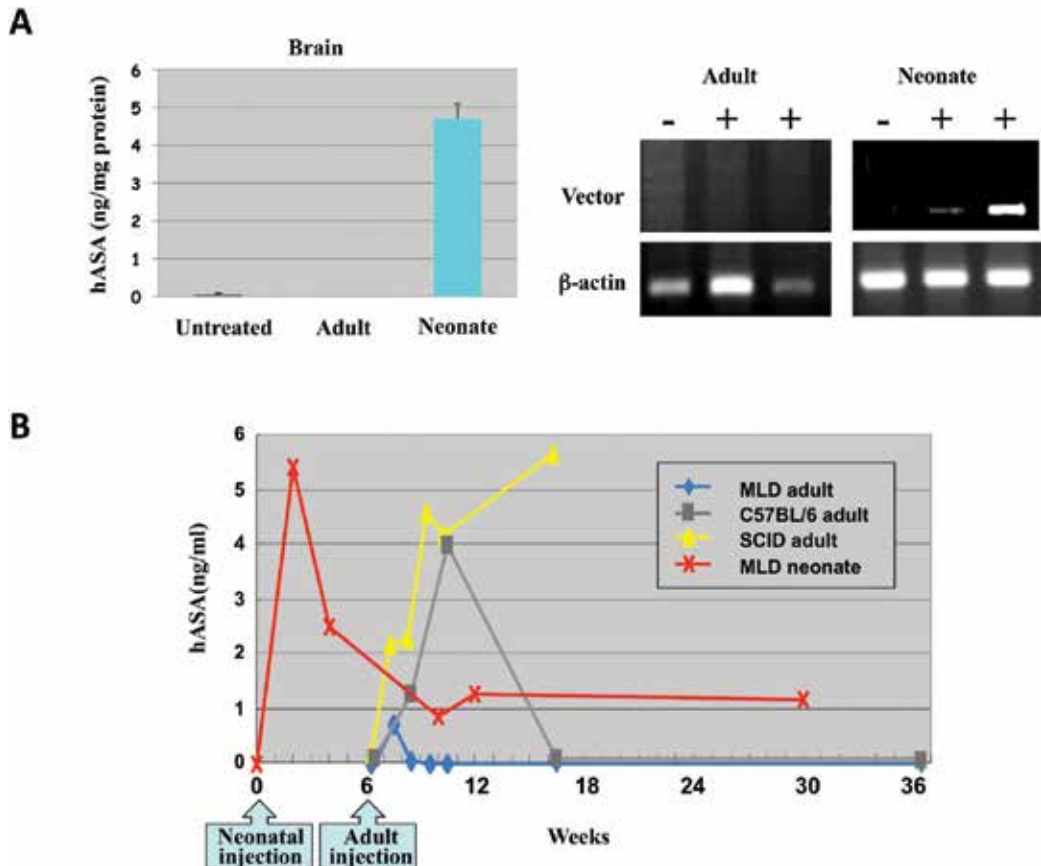
Figure 2. hASA expression of MLD mice following neonatal systemic administration of AAV/hASA vectors. (A) Fifty-two weeks after AAV/hASA injection, hASA concentration in the brain was determined by an indirect sandwich enzyme-linked immunosorbent assay (ELISA) (left panel). DNA from the brain was extracted and analyzed using PCR with hASA-specific primers (right panel). (B) hASA expression in plasma of AAV/hASA-injected mice. hASA concentration in plasma was determined by ELISA. Sustained expression was observed after neonatal injection of AAV/hASA.

in adults. Efficient hASA expression was detected in the brain of AAV/hASA treated at the neonatal period of MLD mice. PCR analysis confirmed that AAV vector genome was observed only in neonatal-treated MLD mice. Moreover, sustained expression of hASA in plasma was detected for at least 30 weeks after intravenous injection into neonatal MLD mice, while only transient increase in plasma hASA was obtained when injected into either adult MLD mice or wild-type C57Bl/6 mice (**Figure 2B**). Vector injection into adult NOD-SCID mice led to sustained secretion of hASA into the circulation, suggesting that immune responses to hASA are a major hurdle for successful gene therapy in immunocompetent adult MLD mice. It thus appears that the systemic injection of AAV vector during the neonatal period is a potentially useful means of treating neurological disorders.

4.2. Neonatal gene therapy for hypophosphatasia

Hypophosphatasia is an inherited disease caused by a deficiency of tissue-nonspecific alkaline phosphatase (TNALP) [21, 22]. The major symptom of human HPP is hypomineralization,
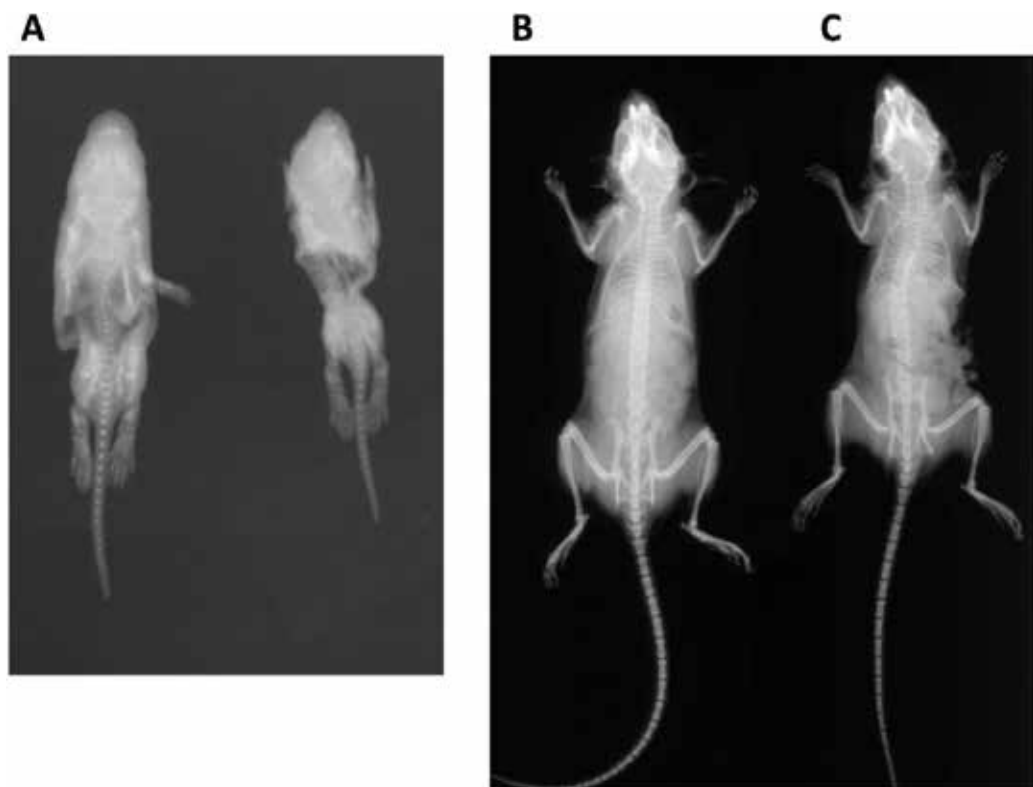
Figure 3. X-ray images of the whole bodies of TNALP knockout mice. Radiographic images were obtained on lFX1000 film (Fujifilm Corp., Tokyo, Japan) using a setup for analysis of small animals. The energy level was 25 kV, and the exposure time was 90 s for 10-day-old untreated TNALP knockout (A), normal wild-type (B) and AAV/TNALP-D10-treated TNALP knockout mice (C).

rickets or osteomalacia, although the clinical severity is highly variable. Patients with infantile HPP may appear normal at birth but gradually develop rickets before reaching 6 months of age. Neonatal gene therapy is a promising strategy for treating infantile HPP by preventing early onset. We have shown that the phenotype of TNALP knockout mice [23–25], which mimics the severe infantile form of HPP, can be prevented by a single neonatal injection of AAV vector encoding bone-targeted TNALP in which a deca-aspartate tail is linked to the C-terminus of soluble TNALP (AAV/TNALP-D10). Sustained expression of TNALP and phenotypic correction of TNALP knockout mice were observed following the neonatal gene therapy [26]. X-ray analysis showed that treated TNALP knockout mice grow as well as normal wild-type mice (**Figure 3**).

5. Problems of neonatal gene therapy

There are several problems that must be overcome before neonatal gene therapies can be used in humans. First, safety concern must be addressed, as there is the possibility of tumor development and of germ-line transmission. It was reported that liver and lung cancers appeared in some mice treated using AAV-mediated neonatal gene therapy [27, 28]. In addition, differences in developmental stages of organs in mice and humans may be another problem. The immune system in mice is less mature at birth than that in larger animals, and the human BBB is functionally mature before birth. It is therefore not clear whether the same beneficial effect of neonatal gene therapy seen in mice would be achieved in human infants. These problems must be overcome before there can be clinical trials of neonatal gene therapy.

6. Summary and future developments

We have shown that AAV-mediated gene transfer in neonatal mice has characteristics that could potentially overcome the problems encountered with current gene therapy protocols. However, before applying neonatal gene transfer to humans, several important issues must be addressed. In particular, the safety of neonatal gene transfer must be carefully evaluated using large animal models, including nonhuman primates. Nonetheless, because of its advantages over gene therapies used to treat genetic disorders in adults, safe and effective neonatal gene therapy has the potential to be an invaluable method for treating genetic diseases.

Acknowledgements

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Endocrine Active Compounds Actions during Neonatal Period: Effect on the Ovary

Maria Slomczynska, Malgorzata Grzesiak and
Katarzyna Knapczyk-Stwora

Additional information is available at the end of the chapter

Abstract

Many female reproductive disorders observed during adulthood originate from the neonatal period, which is a critical stage toward the reproductive potency. Human and animal fertility are determined by the pool of primordial follicles that are established during fetal or neonatal life. The earliest stages of follicle development are under control of a variety of factors, including sex steroids. Neonatal period is a "critical developmental window" in which organisms are susceptible to the environmental chemicals that may affect the reproductive health. Endocrine active compounds (EACs) are found abundantly in the environment and interfere with sex steroids (predominantly androgens and estrogens) by either mimicking or blocking their functions. This review covers the current knowledge about the effects of selected EACs with androgenic (testosterone propionate), anti-androgenic (flutamide), estrogenic (diethylstilbestrol, bisphenol A, 4-tert-octylphenol, phtalates and genistein), anti-estrogenic (ICI 182,780 and parabens), or mixed activity (methoxychlor) on the ovary of neonates, focusing on their effects on the early stages of folliculogenesis. These chemicals have been shown to affect oocyte survival, follicle formation, and growth, as well as steroidogenic functions. The better cognition of mechanisms underlying the long-term consequences of the neonatal EACs exposure may in future lead to an understanding of human health risks and developing prevention strategies.

Keywords: endocrine active compounds, estrogens, anti-estrogens, androgens, anti-androgens, ovary, neonatal window

1. Introduction

Considerable evidence demonstrates that inappropriate steroid exposure during pre- and postnatal developmental periods may have long-term effects on the adult reproductive functions [1]. Female fertility is determined by the pool of primordial follicles within the ovary that is established during fetal or neonatal life. The earliest stages of follicle development are tightly coordinated by numerous factors, including steroids. Importantly, these fetal/neonatal periods are "critical developmental windows" in which organisms are susceptible to the environmental chemicals that may affect ovarian formation and disturb reproductive functions during adulthood, including conception rates, maintenance of pregnancy, and reproductive disorders [2]. Endocrine active compounds (EACs) comprise a wide variety of synthetic or natural chemicals found in the environment arising from anthropogenic, industrial, agricultural, and domestic sources. EACs may interfere with the natural regulation of endocrine system by either mimicking or blocking the function of endogenous hormones and also they may act directly on gene expression by means of epigenetic modifications [3]. This is of concern because women are exposed to EACs on a daily basis, and some EACs are known to target the ovary and cause reproductive health problems. However, the harmful effects of EACs during neonatal period may occur with exposure to much lower doses than those considered harmful to adults [4]. Although many disrupting chemicals are excluded from the everyday use since their dangerous properties have been described, they may persist in the environment for long periods of time. Furthermore, new products with similar properties are still introduced to the market. Therefore, these compounds have currently been one of the greatest concerns all over the world. To assess the effect of EACs on human ovarian and consequently reproductive functions, the animal models are extensively used in experimental studies.

In this chapter, we aim to point out a possible impact of androgens/estrogens excess or deficiency on neonatal ovary following EACs action. The effect of EACs with androgenic (testosterone propionate), anti-androgenic (flutamide), estrogenic (diethylstilbestrol, bisphenol A, 4-tert-octylphenol, phtalates and genistein), anti-estrogenic (ICI 182,780 and parabens), or mixed activity (methoxychlor) will be discussed.

2. Mechanism of steroids action

The action of androgens/estrogens, and their agonists or antagonists, depends on the presence of specific receptors in target cells. The types of receptors that are involved in the signal transduction decide on the signaling pathway. The effect of sex steroid hormones action within the cell may be genomic or nongenomic. A genomic response is usually induced by nuclear/cytoplasmic receptors, while nongenomic pathway is activated by membrane receptors, mostly G-protein-coupled receptor. It is now evident that there is crosstalk between nongenomic and genomic signaling pathways [5].

Steroid hormone receptors, which belong to the superfamily of nuclear receptors, are hormone-activated transcriptional factors. They are modular proteins consisting of C-terminal ligand-binding domain (LBD), highly conserved DNA-binding domain (DBD) with centrally located zinc fingers, and N-terminal domain [6]. Activation of these receptors is mediated by two transcriptional activating domains, AF-1 and AF-2. AF-1, characterized by a ligand-independent transcriptional activation, is localized in the N-terminal region of the receptor, while AF-2, activated by ligands, is localized within the LBD. The LBD and DBD of nuclear receptors are conserved regions, whereas the N-terminal domain is highly variable but important for full transcriptional activity [7]. The latter contains many sites for Ser/Thr phosphorylation, which may be involved in mediating cross-talk with other signaling pathways leading to modulation of AF-1 activity and interaction with coregulators [8].

Sex steroid hormones can freely diffuse across the plasma membrane and bind to their cognate receptors inducing dissociation from the heat-shock proteins. Ligand binding to LBD region causes conformational changes followed by the dimerization of receptors and its translocation into the nucleus. The dimer binds to the hormone response elements (HREs) typically located in the promoter of the target gene and leads to the recruitment of coregulators, either coactivators or corepressors [9]. However, some nuclear receptors (i.e., estrogen receptors, and ERs) are capable of binding to DNA sequence, lacking the classic HRE sequence. Such receptors associate with the components of the AP-1 transcription factor complex (i.e., c-Jun and c-Fos). Additionally, in the low steroids concentration, the ligand-independent signaling pathway occurs allowing the activation of receptors. This process depends on the growth factor receptors and involves MAPK/ERKs pathway, which enhances transcriptional activity through direct phosphorylation of steroid receptors [10]. These pathways are known as "genomic pathway" and are characterized by the regulation of the expression of specific sex steroid receptor-regulated genes [11].

Although steroids exert most of their biologic activity through direct or indirect genomic effects, there are several examples that do not fit to this regulatory scheme. This pathway known as "nongenomic" is much more rapid and involves receptors localized in the plasma membrane or in "lipid rafts" [12]. All sex steroid hormones may stimulate rapid effects in signal transduction pathway through the production of second messengers, ion channels transport, and protein kinase cascades. Rapid effects of androgens and estrogens have been reported in many cell types [13, 14]. Conventional steroid receptors located in the membrane trigger the signal transduction cascade by activation of the Src/Ras/Raf/MAPK/ERK1/2 pathway [15]. Furthermore, steroids may mediate rapid signaling by binding to transmembrane receptors unrelated to nuclear hormone receptors (usually, G protein-coupled receptor, GPCR) and to the modified forms of steroid receptor or to receptors for neurotransmitters. The involvement of the type of receptor may depend on the steroid and cell type [16]. GPCR activates a protein kinase cascade or acts at the level of secondary messengers such as PI3K/AKT/mTOR, PI3K/AKT/cAMP/Ca^{+2} or ERK-mediated pathways like PI3K/AKT/MAPK-ERK/Elk1 [17]. Among GPCR, there are GPR30 for estradiol, mPR for progesterone, and GPRC6A and ZIP9 for androgens [18–21]. All the abovementioned mechanisms of steroid action are presented in **Figure 1**.
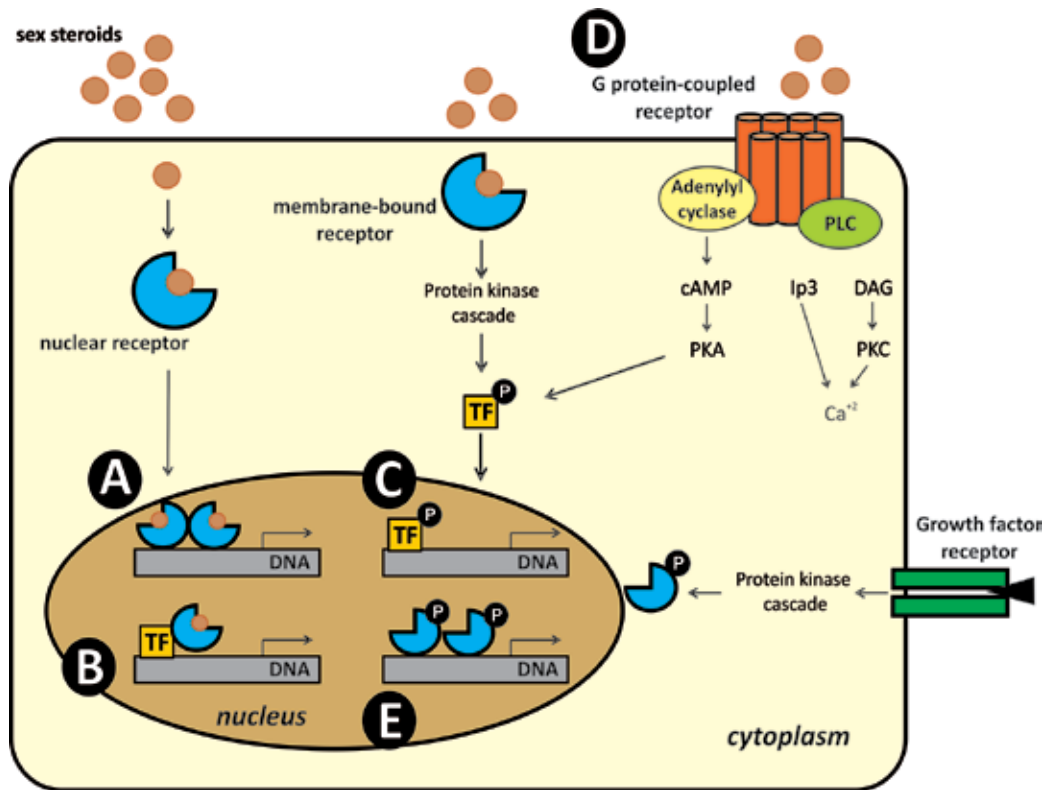
Figure 1. Molecular mechanism of the action of sex steroid hormone receptors. (**A**) Direct genomic effects. (**B**) Indirect genomic effects. (**C**) TF activation triggered by membrane receptor. (**D**) Nongenomic effects triggered by G-protein–coupled receptor. (**E**) Ligand-independent effects. TF, transcription factor; cAMP, cyclic AMP; PKA, protein kinase A; PLC, phospholipase C; IP3, inositol 1,4,5-triphosphate; DAG, diacylglycerol; PKC, protein kinase C. Based on Laurentino et al. [22].

3. The role of androgens and estrogens in the ovarian development

Primordial follicle assembly and the primordial-to-primary follicle transition are the major developmental events within the ovary [23]. The primordial follicle pool represents the total population of germ cells available during reproductive life and, therefore, determines the fertility potential of the female. In mammals, mitosis of oogonia and initiation of meiotic prophase I occur before birth, but the timing of oocyte meiotic arrest, follicle formation, and the initiation of follicle growth varies among species. During fetal ovarian development, primordial germ cells (PGSs) migrate from the endoderm of the yolk sac to the bipotential genital ridges. In mice (reviewed in Ref. [24]), which is a valuable animal model for studying embryogenesis, PGSs migrate into the undifferentiated gonad from around 8.5 day *post coitum* (dpc) to 11.5 dpc. The genital ridge is identical in both males and females and remains bipotential until sex determination (12.5 dpc). PGSs continue to proliferate mitotically during the migration and after arrival in the genital ridge until 12.5–13.5 dpc in mouse and 6th week of pregnancy in human. Then, at 13.5 dpc, in the mouse and between 8 and 13 weeks of gestation in human, meiosis is initiated, allowing oogonia to develop into oocytes that are arrested at the early diplotene phase of meiotic prophase I (reviewed in Ref. [25]). Oocytes

initially develop as large, interconnected clusters of cells called oocytes nests. They are organized into long ovigerous cords surrounded by somatic or pregranulosa cells [24]. In rodents, oocyte nests break down in neonatal ovary that leads to the formation of primordial follicle. This is mediated by oocyte apoptosis starting at 16.5 dpc in mice. In addition, somatic cells can mediate nest breakdown by moving into the nest and intersperse between the remaining oocytes to form primordial follicles [26]. In the mouse ovary, the primordial follicles consist of an oocyte surrounded by a single layer of squamous pregranulosa cells that are formed fairly synchronously between 17.5 dpc and 4 postnatal day. Once assembled, some of the primordial follicles are immediately stimulated to growth, but most of them remain quiescent until selected follicles are gradually recruited into a growing follicle pool throughout reproductive life [27]. The recruitment of primordial follicles into growth (primordial-to-primary follicle transition) involves a change in the shape of the granulosa cells from squamous to cuboidal and the initiation of oocyte growth. The primordial-to-primary follicle transition is an irreversible process. Most of the growing follicles (99.9%) are destined to become atretic, and the rest will ovulate [28]. Around the end of the first postnatal week, some secondary follicles are present in the mice ovary [29].

In contrast, in humans, ruminants, and pigs, follicle formation and the initiation of follicular growth are much less synchronous than in rodents and take place during fetal life. Although cattle and human have similar gestation lengths, follicle assembly in cattle was observed earlier (around 80 dpc) than in human (112–133 dpc). Besides, in bovine ovaries, the first primary and secondary follicles were observed around 140 and 210 dpc, respectively. No secondary follicles were observed in the fetal human ovary [30]. In pig, primordial follicle formation was observed between 70 and 90 dpc, while primordial to primary follicle transition was between 90 dpc and postnatal day 1 [31]. Interestingly, even antral follicles were present before birth in the sheep ovaries [32].

The early stages of folliculogenesis are believed to be gonadotropins-independent. All events related to early follicular development during fetal/neonatal period are mostly regulated by paracrine growth factors originating from the growing oocyte itself and from the somatic cells that surround it [33], and also by ovarian steroid hormones (i.e., progesterone, androgens, and estrogens). Although the pivotal role of androgens in the development of male reproductive organs is well understood, growing evidence supports the direct involvement of these hormones in female reproduction, including early follicular development [34]. The androgen receptor (AR) role in the female is implicated from the studies of various global and tissue-specific AR knockout (ARKO) mouse models. Granulosa cell-specific ARKO (GCARKO) mouse models have demonstrated that granulosa cells are important sites for androgen action and strongly suggested that the AR in these cells is an important regulator of androgen-mediated follicular growth and development. On the other hand, AR inactivation in the oocyte, as shown in the OoARKO female mouse model, appears to have no major overall effect on female fertility [35]. Fowler et al. [36] reported that the oocyte of the primordial follicles was able to synthesize androgens and that pregranulosa cells expressed AR in human fetal ovaries. Initiation of primordial follicle growth was stimulated in mouse, bovine, and primates ovaries by testosterone and dihydrotestosterone [37–39], while in sheep by dehydroepiandrosterone [40]. In general, androgens stimulate the primordial-to-primary follicle transition but impede the later stages of follicle development.

Studies using ERα knockout (αERKO) mice have demonstrated that the lack of ERα does not appear to affect folliculogenesis until the preantral stage of follicle development [41]. Similarly, in ERβ knockout (βERKO) mice, 17β-estradiol (E2) does not appear to be essential for the establishment of germ cell number or ovarian development [42]. However, Kezele and Skinner [43] have shown that both progesterone (P4) and E2 inhibit oocyte nest break-down. It has been shown that fetal ovarian P4 and E2 concentrations decreased during the time as follicle assembly was initiated in the bovine ovary, while the maternal blood P4 and E2 levels were stable [44]. Recent work by Dutta et al. [45] also revealed that fetal mice produce their own steroid hormones to coordinate primordial follicle development. Notably, E2 promotes primordial follicle formation in hamster [46]. These data indicate that both P4 and E2 are important regulators of ovarian follicle assembly. Furthermore, E2 is likely to inhibit primordial-to-primary follicle transition, which was revealed in neonatal rat ovaries [43, 47]. In contrast, the lack of E2 in aromatase knockout (ArKO) mice was associated with decreased numbers of primordial and primary follicles [48]. Similarly, the primordial follicles number was reduced in estrogen-depleted fetal baboon ovaries [49]. These results indicate that although E2 is needed for early events in folliculogenesis, its excessive level inhibits these events. Abnormal control of primordial follicle assembly may affect the reproductive potential of the female and can lead to pathological conditions such as premature ovarian failure.

4. Effects of endocrine active compounds within the neonatal ovary

Many female reproductive disorders observed during adulthood originate from the neonatal period, which is a critical stage toward the reproductive potency. Consequently, it is important to study the effects of neonatal exposure to chemicals that mimic or antagonize the effects of estrogens and androgens to establish their role in ovarian function. Recent attention has been especially focused on environmental factors interfering with endogenous steroids action. There are synthetic and natural environmental compounds such as pesticides (i.e., dichloro-diphenyltrichloroethane (DDT); methoxychlor (MXC), vinclozolin, and atrazine); detergents and surfactants (4-tert-octylphenol, nonylphenol, and bisphenol A); plastics (phtalates); industrial compounds (polychlorinated biphenyl, PCB; 2,3,7,8-tetrachlorodibenzo-p-dioxin, TCDD); and fitoestrogens (genistein and coumestol) [4]. In the light of growing body of evidence that demonstrates the presence of EACs in the environment, understanding the mechanism of selected EACs action within neonatal ovaries provides a basic data for further research of the female reproductive potency. The susceptibility of target tissues is related to the stage of development, the exposure dose, and the individual immune status. Mammals are more sensitive to EACs during fetal and postnatal life than in adulthood. However, environmental influence during fetal and neonatal development may lead to adult onset pathology.

4.1. EACs with androgenic and anti-androgenic properties

Previous data on primates clearly showed the essential role of androgens in promoting early follicular growth [50]. These results were confirmed by *in vitro* study on bovine ovarian follicles, indicating that testosterone stimulated the primary-to-secondary follicle transition

[38]. Importantly, testosterone increased the number of secondary follicles with no impact of estradiol that suggested the direct effect of androgens without conversion to estrogens. Testosterone promotes the growth of bovine follicles *in vitro,* and its stimulatory effect was mediated through ARs in the follicular cells [38]. Thus, reprogramming of early folliculogenesis by androgens excess or deficiency seems to be crucial for understanding their adverse effects on female fertility.

Studies on sheep [51, 52] and primates [53] have shown that excess of androgens in fetal life resulted in reduced ovarian functions in adult life and may provide models for polycystic ovary syndrome (PCOS) in women. Research by Tyndall et al. [54] was undertaken to investigate the effects of different windows of testosterone propionate treatment during fetal or neonatal life in female rats to determine whether and when excess androgen exposure would cause disruption of adult reproductive functions. The findings from fetal exposure to testosterone propionate suggested the possibility of reducing the sensitivity threshold to neonatal androgens exposure in female animals. The results support the concept that androgen programming of adult female reproductive function occurs only during specific time windows in fetal and neonatal life with implications for the development of PCOS in women.

Neonatal androgenization by testosterone propionate induced early misprogramming of ovarian functions in the female rats [55]. The obtained results strongly suggested that transient neonatal hyperandrogenemia led to ovarian dysfunctions marked by altered steroidogenesis and folliculogenesis. Exposure to testosterone propionate resulted in the increase of primary and the decrease of antral follicle frequencies. Moreover, the higher proportion of atretic follicles and the lack of corpora lutea within the ovaries from testosterone propionate-treated rats were observed. Another research on rats revealed that exposure to excess of testosterone in neonatal period increased the LH and testosterone serum levels, the LH/FSH ratio, ovarian theca-interstitial area and expression of steroidogenesis-involved genes, and Lhr and Cyp17a1 in ovaries of adult animals [56]. These results provide some insight into the role of androgens on reproductive development and on the manifestations of clinical disorders such as PCOS.

Among EACs, there is a large group of chemicals exerting anti-androgenic effects and blocking endogenous androgens action. In our previous experiments, the androgen involvement in early stages of folliculogenesis was confirmed using *in vivo* animal model generated for studying androgen deficiency. We have utilized flutamide (2-methyl-N-[4-nitro-30-(trifluoromethyl)-phenyl] propamide), which is a nonsteroidal anti-androgen acting at the AR level and blocking androgen action. Flutamide promotes AR translocation to the nucleus and DNA binding, but nevertheless fails to initiate transcription, inhibiting the AR signaling pathway [57]. Although this is a pharmaceutical compound, it is regarded as a model anti-androgen in experimental studies [58].

In our recent research, flutamide (50 mg/kg body weight) was injected into pregnant gilts during gestational days 20–28 and 80–88 and into female piglets on postnatal days 2–10. We have found ovarian morphological changes indicating delayed folliculogenesis in neonatal pigs exposed *in utero* to flutamide [59]. Abnormal folliculogenesis was also seen in prepubertal pigs, following prenatal period of anti-androgen exposure [60], whereas in adult

animals, only neonatal time of flutamide treatment exerted disturbances marked by altered luteinization and corpus luteum cyst formation [61]. Apart from morphological disturbances, the altered follicular functioning in adulthood was manifested by changes in production of steroids (estradiol and androgens) and gonadotropins (FSH and LH), as well as in expression of genes involved in proper ovarian functioning, including AR, FSHR, aromatase, connexin 43, β-catenin, and aquaporin 5 [61–63]. Furthermore, gestational or neonatal exposure to flutamide affected proliferation and apoptosis rates in large antral follicles of adult porcine ovary, which might influence the normal development of the follicles and pigs fertility as a consequence [64].

It seems that disturbed androgen action during gestational and neonatal periods by using flutamide led to reprogramming of the trajectory of ovarian development in pigs; however, neonatal window of exposure leads to long-term effects observed in adulthood.

4.2. EACs with estrogenic and anti-estrogenic properties

Compounds that mimic estrogens action through an interaction with ERs continue to receive considerable attention. One of them is 4-tert-octylphenol, which belongs to alkylphenol polyethoxylates derivative, which is a nonionic surfactant widely used in a variety of industrial applications [65]. It was demonstrated that pre- or postnatal exposure to 4-tert-octylphenol can advance the onset of puberty in rats [66] and pigs [67]. In addition, in a three-generation study, 4-tert-octylphenol given prenatally to sows induced some effects, including reduction in litter size, observed in the next generations [67]. Furthermore, 4-tert-octylphenol exposure during fetal and postnatal life accelerated the onset of puberty but did not disrupt FSH concentration and the dynamic of ovarian follicular growth in ewes [68]. On the other hand, research on rats revealed that neonatal administration to 4-tert-octylphenol altered hypothalamo-pituitary-ovarian axis resulting in atrophic and polycystic ovaries without corpora lutea [66]. In our most recent experiments using neonatal pigs treated with 4-tert-octylphenol, the advanced folliculogenesis has been shown. We assumed that these specific effects on folliculogenesis in neonatal pigs were characterized by changes in proliferation and apoptosis rates during initial follicular recruitment [69].

Both bisphenol A and diethylstilbestrol are another of the extensively studied EACs with estrogenic activity. Bisphenol A is a plasticizer used commonly in a wide range of consumer products such as food and drink containers, epoxy resins, plastics, baby bottles, thermal receipts, and dental sealants (reviewed in Ref. [70]). Zhou et al. [71] revealed that bisphenol A exposure significantly inhibited germ cell nests breakdown by altering the expression of key ovarian apoptotic genes in cultured neonatal mouse ovaries. Interestingly, exposure of neonates to estrogenic compounds like bisphenol A significantly increases the number of multiple oocyte follicles (MOFs) in adult mice ovaries. It was proposed that neonatal exposure to exogenous estrogens blocked cysts breakdown resulting in oocytes survival [72]. Apart from abovementioned influence of bisphenol A on oocyte nests, additional effects on the follicle development were found. Bisphenol A may interact with multiple factors produced by oocyte and granulosa cells to alter neonatal follicular dynamics. [73, 74]. It was shown that this compound reduced the primordial follicle pool by stimulating the initial recruitment in neonatal

rats associated with an increased proliferation rate, which is likely mediated by an estrogenic pathway [75]. It could also decrease follicle activation in neonatal mice [76]. Additionally, the effect of neonatal exposure to bisphenol A on the reproductive axis was observed in adult rats, which may lead to the development of PCOS [77].

Diethylstilbestrol is a synthetic nonsteroidal estrogen. Studies on neonatal rodents injected with diethylstilbestrol demonstrated results similar to those of bisphenol A. Neonatal exposure to diethylstilbestrol of mice and hamster inhibited germ cell nest breakdown and induced MOFs formation, likely by interfering with the ERβ pathway and inhibiting programmed oocyte death and germ cell loss [78, 79]. Rodríguez et al. [75] used a relatively low concentration of diethylstilbestrol (20 µg/kg body weight) in the rat and found that primordial follicle activation was increased compared to studies of Karavan et al. [80] on mice, where the higher diethylstilbestrol concentration (100 µg/kg body weight) decreased the follicle activation. Both diethylstilbestrol and bisphenol A reduced follicle growth and development, resulting in the presence of more primordial follicles and fewer primary and secondary follicles in mouse neonatal ovary [80]. In prepubertal lambs treated with bisphenol A or diethylstilbestrol during neonatal period, the reduction of the primordial follicle pool and increased atresia was observed. That was surprising since nest breakdown in sheep is completed before birth. However, it seems that the reduction of the primordial follicle pool is due to stimulation of their initial recruitment and subsequent development until antral stage. These alterations may affect the ovarian function in the adulthood [81].

Genistein is a natural phytoestrogen with estrogenic activity found in soy products. The belief that estrogens can regulate nest breakdown is supported by a study of Jefferson et al. [82]. They have demonstrated exposure of neonatal mice to genistein, which inhibited the oocyte nests breakdown and increased oocyte survival. Moreover, ovaries from adult mice treated as neonates with genistein have an increased occurrence of MOFs, suggesting incomplete nests breakdown [83]. This indicates that cell survival/death pathways are altered following neonatal genistein treatment. The inhibition of oogonia/oocyte nests breakdown by EACs action was shown in the **Figure 2**.
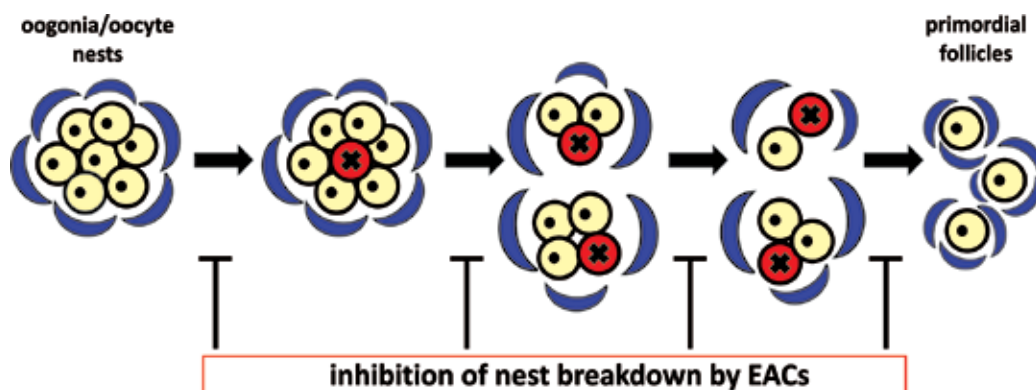

Figure 2. Schematic model of oogonia/oocyte nests breakdown and its inhibition by endocrine active compounds (EACs) with estrogenic activity (bisphenol A, diethylstilbestrol, and genistein).

Phthalates are commonly used as plasticizers in the manufacturing of flexible polyvinyl chloride products. Little information is available on the effects of phthalates on neonatal ovary. These compounds have been actually shown to affect germ cell nest breakdown and primordial follicle assembly in cultured newborn mouse ovaries [84]. In addition, early postnatal treatment with diethylhexyl phthalate accelerated folliculogenesis by decreasing the number of primordial follicles and increasing the number preantral and antral follicles in mice [85]. The decrease in primordial follicle pool indicated estrogenic action of these compounds.

ICI 182,780 is a widely used anti-estrogen that can bind to both ERα and ERβ with very high affinity and completely antagonize the effect of estrogens [86]. It was found that ICI 182,780 in combination with estradiol allowed oocytes cyst breakdown neonatal rat ovaries [87]. On the other hand, Wang and Roy [46] revealed that ICI 182,780 significantly increased apoptosis and caused a modest reduction in primordial follicle formation in neonatal hamster ovary. Likewise, ICI 182,780 accelerated primordial follicles formation from oocyte nests in the neonatal pig. However, the initial follicle recruitment marked by the number of growing was delayed [69].

The compounds that exert both estrogenic and anti-estrogenic properties are parabens, which are widely used as anti-microbial agents in the cosmetic and pharmaceutical industries. Parabens inhibited the early phase of folliculogenesis and steroidogenesis in the ovaries of neonatal rats [88]. Moreover, it appears that parabens through inhibition of transcriptional repressor Foxl2, regulated the levels of steroidogenic enzymes [88].

4.3. EACs with mixed properties

Methoxychlor (MXC) is an organochlorine pesticide metabolized predominantly to 1,1,1-trichloro-2-(4-hydroxyphenyl)-2-(4-methoxyphenyl) ethane (MOH) and the bisphenolic compound 1,1,1-trichloro-2,2-bis(4-hydroxyphenyl) ethane (HPTE) in the organism [89]. MXC along with its metabolites acts through ERs and possesses estrogenic, anti-estrogenic, and anti-androgenic activities depending on the receptor subtype [90]. In a variety of experimental studies, MXC administration in early pregnancy or during the prenatal and neonatal periods has been shown to cause adverse abnormalities in adulthood of female rats, such as reduced ovarian functions and ovulatory rates, as well as pregnancy outcome [1]. Uzumcu et al. [91] demonstrated that MXC administration during the primordial-to-primary follicle transition period in mice (postnatal days 3–10) inhibited follicular development and reduced antral follicle numbers. In addition, MXC treatment increased the level of AMH protein production, which in turn inhibited folliculogenesis within the ovary. Another research suggested that exposure to MXC during fetal and neonatal ovarian development led to adult ovarian dysfunction, including an increase in the number of preantral and early antral follicles and a reduced number of corpora lutea and female infertility in rats [92]. Furthermore, there are several reports suggesting the role of MXC in the induction of epigenetic modifications within the ovary. Rats exposed to MXC (20 or 100 μg/kg body weight) between embryonic day 19 and postnatal day 7 revealed altered methylation pattern in the promoter region of ERα that suppressed the ERα expression and caused ovarian dysfunction. Developmental exposure to MXC led to significant hypermethylation in the ERβ promoter regions, whereas the ERα promoter was unaffected [93].

5. Summary

The plausible link between EACs exposure during critical periods of early development and risk of chronic diseases in adulthood, including premature ovarian failure and PCOS, has been reported. The harmful effects of compounds with androgenic, anti-androgenic, estrogenic, and anti-estrogenic activities during neonatal window may occur with exposure to lower doses than those harmful for adults. These chemicals have been shown to target the ovary of neonates and adversely affect oocyte survival, follicle formation, and growth, as well as steroidogenic functions. There is a concern over EACs due to their common use and persistence within the modern living environment. The better cognition of mechanisms underlying the long-term consequences of the neonatal EACs exposure may in future lead to an understanding of human health risks and developing prevention strategies.

Acknowledgements

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Reducing Early Neonatal Mortality in Nigeria—The Solution

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Additional information is available at the end of the chapter

Abstract

The West African nation of Nigeria seems to have run out of ideas on how their neonatal mortality rate may be lowered. This situation has become dare as the country could not make any significant progress even with the great supports of the last 10 years of Millennium Development Goal. Presently, one in every two deceased child under 5 years of age in Nigeria is a neonate. Literature reveals that most of these deceased neonates are classified preterm or low birthweight, of which nearly four in five must die within first 7 days. This clearly identified the categories and stages of highest mortality; however, it is disappointing that the authorities of the Nigerian health care system have for too long been unable to devise a solution for the neonates. Probably, inadequacy of climatic and cultural compatibilities might partly be responsible for the failure of their current conventional ideas and technologies—these being predominantly imported. Yet, there seems to be lack of interest in some home-grown unconventional ideas that have achieved the needed reduction at few centers. In this chapter, we present the unconventional approaches and encourage across-the-nation translation of the applications to achieve accelerated end to this situation.

Keywords: neonate, Nigeria, neonatal mortality, innovative technique, thermal distress

1. Introduction

Neonatology in the West African sub region, especially in Nigeria, will remain in a state of "scientific comma" until a decisive solution is found to reverse her high neonatal mortality that has continued to be the highest in the world. The solutions required might not necessarily be conventional, as practiced in developed countries of the world, since such imported

scientific methods and systems have failed to completely eliminate or drastically reduce the mortality rates. These certified foreign solutions have been probably rendered ineffective within the Nigerian practice due to complications of poor infrastructure, climate, and above all poor work ethics and cultural inadequacies. The effective solution might not necessarily be the "state-of-the-art" procedures that are applied in the United States of America, United Kingdom, and Europe as Nigerian practitioners continue their endless importation of all kinds of relevant and irrelevant ideas but without the necessary infrastructural base to effectively operate these. This country requires a new breed of scientists and neonatologists that can believe in themselves, look inwards, and apply research methods to develop culturally compatible neonatal solutions using easy-to-acquire and locally available materials.

The Nigeria neonatal record is among the worst in the world. There is no evidence to suggest that more neonates are surviving in Nigeria today as compared to 10 years ago, even with the very celebrated millennium development goal (MDG) campaign. Presently, neonatal contribution to mortality rate below 5 years of age in Nigeria has risen in the last 10 years from 40 to nearly 50% despite the huge expenditure of MDG in Nigeria on importation of ideas and systems [1]. The big questions before anyone who might attempt to solve the high neonatal mortality in Nigeria are:

1. What is *really* the primary factor(s) behind the failure to achieve improved overall neonatal survival in Nigeria?

2. Up to 18 morbidity factors contribute to neonatal deaths in Nigeria but are these contributing equally?

3. Is it needful to save time and resources from some factors and dedicate these to synthesize absolute solutions just for one factor or few that could bring about huge impact, and what could this one factor be?

4. Are there any cluster point(s) of highest mortality that might require in-depth investigation within the spectrum of neonatal life-span of 28 days from birth?

5. How may any locally grown "appropriate technological" solution be integrated into Nigeria's health care system; how may this be applicable both at the tertiary and primary health care levels and be made available to the hardest-to-reach neonates across the hinterlands of the country?

In this chapter, our research group of a team of young Nigerian neonatologists and technologists will discuss our vast experiences and progressive syntheses of ingenious local-content ideas that have drastically reduced the neonatal mortality rate at pockets of neonatal centers across Nigeria; thus, achieving an average facility-based mortality rate of 33/1000 presenting neonates as compared to the national average of 248/1000. We shall discuss the various applications that have restored hope to neonates within our practice—including the handy-approach and initial-setpoint-algorithm (ISA) techniques—and how all the applications have contributed to achieve nearly 100% facility-based survival of premature and low birthweight neonates (including 600-g birthweight) within their first 7 days of life.

2. Many are dying

In 2016, an estimated average of 248 neonates out of 1000 presenting at special care baby units (SCBUs) in Nigeria died. Most of these babies reportedly died of various causes during their first 1 week of life. These data were extracted from a collection of independent outcome publications during the 47th national conference of Paediatrics Association of Nigeria (PANCONF) in January 2016. It is a common practice in Nigeria that SCBUs try to use this annual conference to showcase their discoveries, best practices, and outcomes. Therefore, data that were presented could be taken to be what they considered the most impressive or the best of what the centers were prepared to let others know about. The beauty of the content of the proceeding of the PANCONF 2016 on this subject was that the seven coincidental reports came from centers spread equally across the entire country. This includes:

(1) Yariman Bakura Specialist Hospital (YBSH) Gusau in the North-west [2]

(2) University of Abuja Teaching Hospital (UATH) Gwagwalada in the North-central/middle-belt [3]

(3) Lagos State University Teaching Hospital (LASUTH) Ikeja in the South-west [4]

(4) Stella Obasanjo Specialist Hospital (SOSH) Benin-city in the Mid-west [5]

(5) University of Port Harcourt Teaching Hospital (UPTH) in the South-south [6]

(6) Federal Medical Centre (FMC) Owerri in the South-east [7]

(7) Federal Medical Centre (FMC) Asaba in the South-south [8]

It is worthy of note that six of these independent outcomes came from data sets that were generated based on the conventional techniques of newborn care in Nigeria. This resulted in a national mortality average of 248/1000 [2–6, 8]. However, two of these independent centers presented outcome data that had been influenced by their adoption and practice of the various unconventional methods that were developed through the collaborative research of Neonatal Concerns for Africa [9]. In-between these two institutions, the average neonatal mortality crashed below 34/1000 [3, 7]. This translates to a national average reduction of facility-based mortality by a whopping 86%.

Previous publications on conventional practices within the last 10 years have reported facility-based averages such as 254/1000 and 250/1000 [9, 10]. These figures are quite similar to the present 248/1000; hence, this raises the question of why the custodians of neonatal health in Nigeria have been unable to articulate decisive solutions for such a national emergency situation. The scientists seem far too busy with other things than to own the blame, put on their thinking caps, and synthesize an affordable and sustainable home-grown solution to save their neonates. Instead, the over-dependence on unsustainable importation of foreign technologies and ideas have left the Nigerian health care professionals so scientifically lazy that the neonates are still far away from their hope for survival. It was expected that the high publicity and available funds during the last 10 years of the millennium development goals

(MDG) would have empowered a great success. Since this was an unfortunate failure for the neonatal sector at the national level, Nigeria could restore hopes by a humble study of what constituted the pockets of successes recorded by some few centers that adopted unconventional techniques.

3. Change is compulsorily needed

Neonatal health care professionals in West Africa, especially Nigeria, need to understand that the world expects them to apply whatever science they can manage to deliver a drastic reduction in the neonatal mortality rates as it has been known for the last 10 years. It ought to be a challenge to the health care providers in this country that no significant improvements have been achieved, even after ten years of accelerated campaigning and spending—from the 2007 demographic data of the World Health Organization (WHO) to that of Nigeria's Federal Ministry of Health (FMoH) of 2010, to that of UNICEF's 2012, and perhaps up to the indices of 2017 (**Figure 1**) [1, 11–14].

The question that remained unanswered is whether there are any more untried ideas left for the Nigeria's FMoH and the other custodians of neonatal health in Nigeria that can help lower their well-known horrible indices, including:

(a) Neonatal deaths accounting for nearly 50% of all deaths of children under 5 years of age [1, 15]

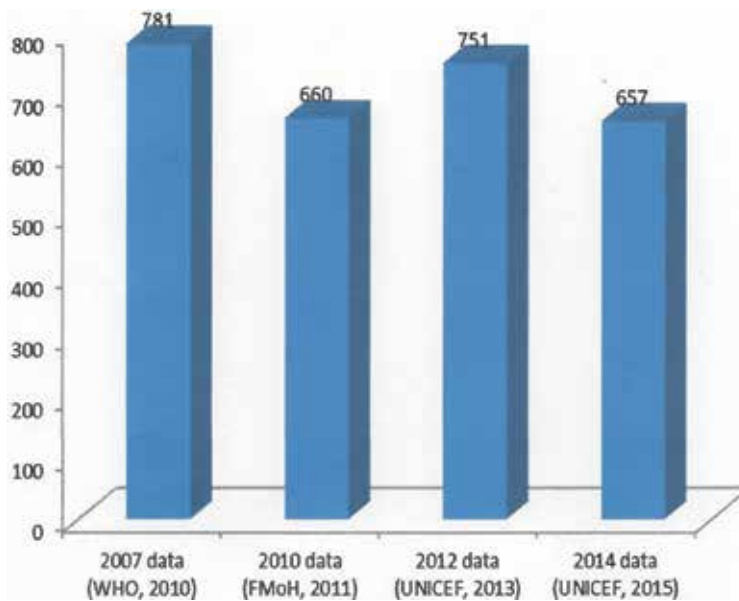(b) Nearly 80% of all deceased neonates dying within their first 1 week of life [14, 16]

Figure 1. Daily newborn mortality over last 8 years of MDG.

This showed that most deceased neonates died before their 7th postnatal day, beyond which most surviving neonates would successfully go home alive. We hypothesize that after the first quarter of neonatal life, i.e., 7 of 28 days, most Nigerian neonates are strong enough to contribute their minimal quotas in resisting and fighting their various debilitating morbidities, howbeit, within a conducive and stable physiological state. The first few days after birth are their greatest period of need for external life support systems and procedures. Inadequate support and procedures during this period would normally be associated with such high rate of death as reported in Nigeria. The most vulnerable of the neonates is the premature ones who may necessarily require knowledgeable support for survival. Nigerian authors have shown that mortality for very preterm and extremely low birthweight neonates at some hospital centers can be quite high, even as high as 933/1000 [17–20]. It is our opinion that the current Nigerian conventional facility-based practices during early neonatal life is inadequate or fundamentally faulty. Our resonating questions remain: Has Nigeria got any other ideas for preventing such high rate of early neonatal deaths that has not yet been tried since the last 10 years as these are urgently needed? Is it time for the FMoH and the establishments to come out openly to accept the failure? Is it time to have compassion on the neonates and search for other sustainable, and perhaps, unconventional methods as have been demonstrated in few pockets of Nigerian centers? The time cannot be any sooner than now.

4. Wholesale importation is unsustainable

It is always attractive to import latest technologies for application in Nigeria. However, poor infrastructural development makes such applications unsustainable. Modern medical equipment is quite expensive and unaffordable to many medical institutions that must look after the neonates. However, the procurement of these systems is not necessarily the main problem. Sadly, the age long pattern at these mostly government-owned referral centers is such that after many years of impoverished neonatal outcomes and political harassments, the government manages to provide appropriate funds to purchase only few of the required equipment. However, no sooner this is done, the center goes back into comatose due to inability to maintain the systems. The efficiency of a system or procedure can often depend on factors relating to infrastructural base, climate, peoples' culture, work attitude, manpower, maintenance supply chain, etc. It is essential that these factors are carefully considered, or else a wholesale adoption of a foreign idea may not yield same result as expected. Nigeria's 100% reliance on importation of needed technologies and ideas are unsustainable due to these factors. The best options forward are either adaptation or synthesis of own solutions.

5. Adaption requires scientific thinking

Most of Nigeria's neonatal systems, techniques, and procedures are imported from the United Kingdom, Europe, or America where these are well-proven to be very effective, reliable, and sustainable. Unfortunately, similar outcomes would depend on the abilities of the importing

country to adapt the application within their own variables of culture, climate, and infra-structure. This requires some scientific thinking. There may be a need to understand the basic physics of the application in order to know the aspect of the application that needs tweaking; and how best to do this to make the application reliably operable in the setting. The Nigerian scientific and research community must be alive, active, and confident to create such interme-diate technologies. We think that a quicker success could be made if technologies are tweaked to fit the Nigerian weather and culture rather than the more difficult route of "tweaking cul-ture for a piece of imported technology". This challenge is both for the Nigerian government but much more for the Universities, research institutions, and their Professors.

6. Syntheses of unconventional techniques

Our hypothesis created the drive to search for the possible factor that could be secretly at work or that might provide a huge positive impact for the neonates if isolated. The search began with a nationally circulated questionnaire that identified lack of functional incuba-tors as a major factor militating against neonatal survival in Nigeria [17]. However, advanc-ing consistently with various careful comparative investigations, our research can presently reveal a strong association of "thermoneutral instability", hiding behind a host of co-morbid conditions that had been said to be responsible for high rate of neonatal mortality in Nigeria [15]. Consistent thermal stability within body temperatures of 36.5–37.4°C is essential for the neonate's physiological wellness to fight disabling conditions, maintain positive progress, and for survival. Lack of thermal stability leaves the neonates so moribund that the other presenting co-morbid factors become excessively too powerful for the neonates in their fights for survival. Hence, unbroken provision of neonatal thermoneutral environment—without excuses of unfavorable weather, climate, cultural, and work attitudes—could be the one-fac-tor game-changer that has eluded neonatal practice in Nigeria.

We developed and tested applications as we investigated various phenomena that were very evident in the Nigerian setting. Methods that applied locally available materials were more easily sustainable, affordable, and maintainable. We opted for these, thereby introduc-ing applications, procedures, and advocacies at various times as published in Ref. [9]. We hence created a package of stand-alone activities that supplemented each other to produce the reported great outcomes at Special Care Baby Units of the Federal Medical Centre Owerri and University of Abuja Teaching Hospital in 2016 [3, 7]. These applications were:

6.1. The Recycled incubator technology (RIT)

In a period of economic recession such as Nigeria is currently experiencing, neonatal incu-bators are increasingly unaffordable due to skyrocketing foreign exchange. Dysfunctional and broken-down incubators litter the hospitals, yet the preterm babies are arriving in their thousands every day. We developed some techniques that Nigeria could use to re-engineer and activate these dysfunctional systems, offering up to 10 fresh years of life-expectancy at costs that are less than 20% of selling price of modern incubators. Recycled incubator technol-ogy (RIT) has worked very well for our collaborating hospitals centers, ensuring consistent

availability of over 15 functional incubators at all times. This grants them an outstanding position when compared with the other government centers that hide behind lack of funds to deny the neonates of this vital system. Any Nigerian hospital with any available casing of broken-down incubator does not actually have excuses to give in support of their denial and neglect for this basic neonatal need. The RIT upgrading or conversion is affordable and does not necessarily require special government subvention as many of our collaborating centers fund this from their internally generated revenue. This simple unconventional technology of the RIT is saving huge amount of lives in Nigeria without huge spending; hence, the Nigerian government and the custodians of neonatal health in Nigerian should pay attention.

6.2. The failure-preventive audit culture (FAC)

One wonders why routine maintenance of these items of neonatal equipment in Nigeria is so neglected. Neonatal systems such as the incubator, as a life support machine, works nonstop, day-in and day-out, yet the conventional Nigerian practice does not involve a compulsory regular maintenance by qualified personnel. Regular professional maintenance of neonatal systems, even in their countries of manufacture, is a routine never to be compromised. Yet anyone wonders why there are so many broken-down systems at Nigerian centers where machines are run continuously without servicing until they are destroyed. The systems at our centers are always functionally available because the hospitals run a six monthly mandatory failure-preventive audit culture (FAC) for the incubators. Similar assiduous care must be made compulsory at all SCBUs operating in Nigeria. Suffice to mention that many of the incubators are homes for the microbes that infect the dying neonates as a result of lack of routine technical cleaning and decontamination. As this is an unlawful life-threatening exposure of a neonate to harmful environment, the seriousness of the Nigerian government should be measured by their willingness to prosecute defaulting professionals and institutions.

6.3. The power banking system (PBS)

The widespread exposure of the neonate to hypothermia even when functional incubators are available can be due to epileptic power supply or outright power failures that last hours unending. The incubator becomes useless when there is no power to run it. Grid power failure can even last for days in Nigeria such that a standby fossil fuel generator becomes inadequate and unsustainable to provide uninterrupted incubator care. Nigeria is yet to provide a decisive solution for her inability to deliver uninterrupted grid power to the nation. Many centers rely on this limitation to offer excuses for the high neonatal mortality. In our few centers, however, the incubators run uninterrupted by the application of our power banking system (PBS) initiative. We used batteries to store up power from the grid and generators whenever these were available. This was in addition to trickling recharging of the batteries using photovoltaic cells mounted on the rooftops, especially when there was failure of both grid and fossil generator powers that lasted longer than necessary.

6.4. Evening-fever syndrome (EFS) antidote

The Nigerian climate can get excessively hot with a higher temperature of up to 47°C in some cities during the dry season [21, 22]. Preterm neonates are often unable to regulate their body

temperatures downward from the high room temperatures in excess of 38°C. This leads to periodic fever that sets in during late afternoons and abates later in the evening, long after sunset. This phenomenon of evening-fever syndrome (EFS) was investigated leading to the proposal of a weather-resilient building technique for neonatal nurseries in Nigeria [22, 23]. We discovered that the periodic hyperthermia due to EFS, often misdiagnosed as a disease process, has the consequences of neonatal thermal distress that drastically slowed down baby's response to treatment. It was hence essential that all our nurseries were renovated or properly relocated within the hospital complexes so as to eliminate or minimize this phenomenon. Neonates in our centers were hence kept within allowable physiological temperatures even when climatic ambient temperatures soared higher above 45°C.

6.5. SCBU nurses retention advocacy

One obnoxious common practice in Nigerian tertiary hospitals is the periodic reshuffling of nurses around various departments of the hospital irrespective of specialities. This practice has such huge negative impact on neonatal outcome that some managers have refused to pay attention to. The practice might be useful in adult wards, some may argue, but this is definitely counterproductive in the special care baby units. It takes much longer than 1 year to transform a general nursing practitioner into one with enough skills to effectively manage the neonates. Learning on the job comes along with its own consequences of blunders that are often committed, some leading to neonatal deaths. However, just as some of these, on-the-job trainees are about to become useful, they are asked to move on while a new bunch of starters are brought in to continue with the devastation. This practice of replacing trained and knowledgeable nurses with learners and starters might sound unbelievable to readers of this article from developed nations; however, this is really one of those practices that have quietly contributed to huge numbers of preventable neonatal deaths in Nigeria. The parochial views of the nursing establishments in Nigeria, including such arguments as "any qualified nurse could work in any department of the hospital", are so powerful that nursing managers seem to have completely lost their senses of reasoning to protect these poor neonates. The management of our few hospital centers has standing rules, after many advocacies, to limit this nurses' reshufflings within only 10% of the entire SCBUs nursing crew. This posting is, however, forbidden to affect the best of the crew members. The challenge here is that members of the SCBU crew are so overworked that many often seek to be posted away. So to minimize this, special incentives other than what all nurses received were given to SCBU crew members in appreciation. This allowed incentives and privileges are still hotly contested in some of our hospitals by supporters of other greedy nurses who would not even want to work at the SCBUs themselves.

6.6. Breathing/apnoea monitors

Lack of adequate number of nursing staff in Nigerian hospitals is an age-long problem that has remained unsolved. This has a huge negative impact on neonatal outcome at Nigerian newborn centers. Hospital managements quickly agree with the need for more staff whenever confronted. However, they easily argue their ways through with the use of Government's endless

embargo on employment. This problem is as bad as having only two nurses to look after 38 neonates during a work-shift in one of Nigeria's busy University Teaching Hospital where this corresponding author once worked as a visiting consultant. How on earth would anyone explain what happens to four neonates that could be undergoing apnoeic attack simultaneously during such a shift? Sure, many of them died from the apnoeic attacks without ever being noticed - most of these from easily preventable causes. Yet, the nursing managements would not reason that the SCBUs needed more nurses relative to the other adult and adolescent wards. The use of affordable simple devices for neonatal chest and body movements monitoring hence became inevitable to use in our centers. Our standard practice of the use of only incubators and cots with installed apnoea monitors for all critically ill neonates and all other premature neonates—especially during their first-seven-days (F7D) of life—must, hence, never be compromised. By this practice, nurses were easily alerted as soon as such neonates go into apnoeic mode. The job of keeping such neonate alive, awaiting proper intervention, becomes the job of any available staff and not just the overworked nurses. For our centers, the apnoea monitor has become the inevitable "third eye" of our nurses to frustrate neonatal "sudden death syndrome" until proper intervention is administered to correct the underlying fault.

6.7. The handy approach

Careful examination of various plots of neonatal body temperature against lifetime, especially during the first few days of life, revealed a startling consistent physiological inadequacy. We created a "regional band" on the graphs, representing the physiological safe-zone temperatures of 36.5–37.4°C (**Figure 2**). This enabled easy assessment of how well the neonates had been assisted to maintain this safezone temperature that is essentially required for thermoneutrality, and hence neonatal survival. We investigated this in many hospitals across the regions of Nigeria—from the South to the North—and we concluded that this was a national problem [9]. It was however more disturbing that the nonphysiological temperature profile also emerged from some neonates that could be seen to have undergone incubator care.

We began to investigate this and quickly discover the complete absence of any standard protocol or algorithm for achieving neonatal normotherm in the Nigerian practice. This led to the synthesis of the handy-approach algorithm, developed as a set of rules that could guide the normalization of a deviating neonatal body temperature back to its safezone temperatures [21]. Ability to use this application became compulsory, and, a necessary requirement for all the nursing and clinical staff of the SCBUs within our neonatal research network (**Figure 3**). Subsequently, a validation study was carried out. This showed that the technique led to greatly improved neonatal temperature profiles and also for better overall outcome [9]. The handy approach is an easy-to-handle tool that practitioners at the rest of Nigeria's SCBU should study for the sake of the multitudes of Nigerian neonates under their care.

6.8. The ISA

The initial set-point algorithm (ISA) was developed as a supplementary temperature control protocol to maximize the impact of the handy approach. This targets a quick attainment of normotherm at the onset of incubator intervention (**Figure 4**). This effectively removes the
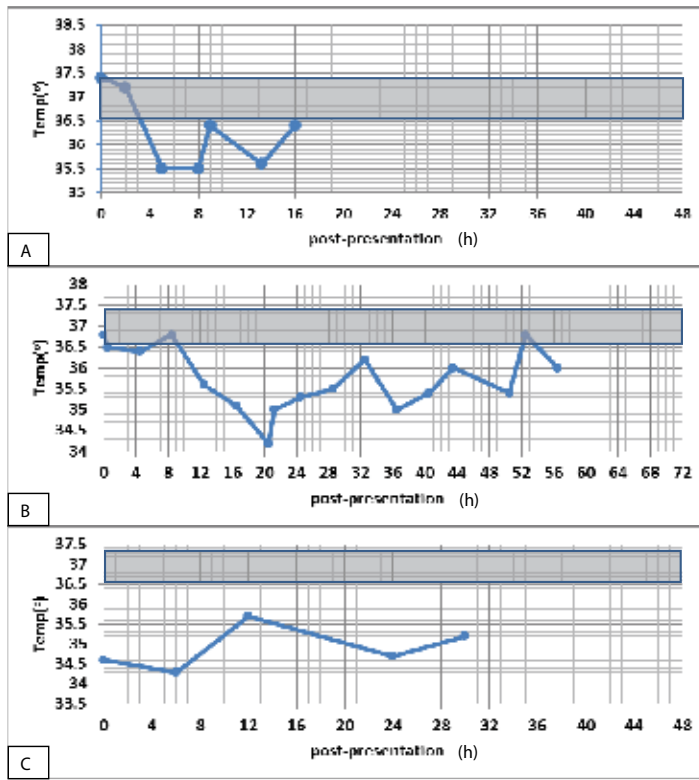
Figure 2. Temperature—Lifetime plot revealing safezone band. (Plots of deceased incubator-nursed neonates from 3 different regional teaching hospitals in Nigeria, A—south: BW = 2000 g admitted 16/01/2013; B—middlebelt: BW = 1580 g admitted 17/11/2012; C—north: BW = 600 g admitted 02/08/2012).

often long postnatal exposure of neonates to temperatures outside the safezone, as this has been argued to inflict a lasting damage that trails some of them to death within their first week of life [15]. The ISA was such a powerful tool that might have coincidentally led to the reduction of F7D contribution to mortality, from 71 to 0%, in a set of studied neonatal cohorts at the University of Abuja Teaching Hospital Nigeria [15]. The devastating postnatal thermal shock described in the research was widespread across Nigerian centers and explained the reason behind the WHO figure of 79% for the mortalities happening before the seventh postnatal day; this figure has never changed between the 2008 and 2013 demographic survey reports [14]. ISA application enabled clinicians at our center to achieve neonatal normotherm in an average of 28 min as compared to conventional average outcome of 12 h and 24 min. The thermal devastation of almost 12½ h long in a hostile environment is unbearable for the poor neonates, especially the preterms, as they are coming from the well-regulated comforts of the womb [15]. Thus, the ISA has been demonstrated to be an effective tool for the care of preterm neonates and a dependable response to this problem. Having also been scientifically validated and peer-reviewed, this has remained a compulsory tool to apply while admitting premature neonates at many of our centers. This has led to drastic overall reduction in the mortality of premature neonates at these few centers; figures falling from 63 to 7% at
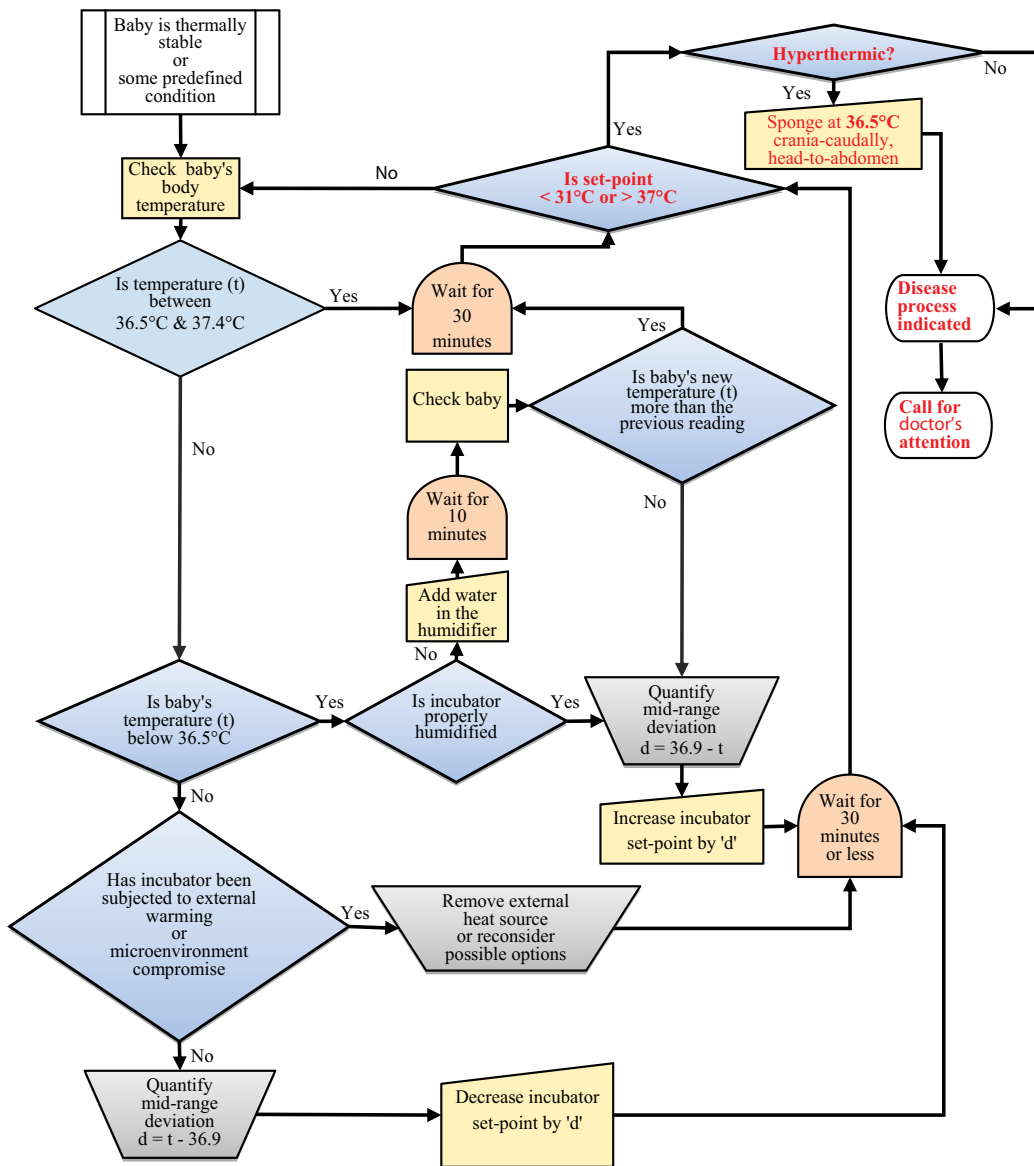
Figure 3. The handy approach. Modified from Amadi [21].

the University of Abuja Teaching Hospital, Gwagwalada Nigeria, for example. The ISA is an unconventional step, all Nigerian and West African centers might have to adopt if they must lower their high facility-based mortality rate of premature neonates. In addition to this excellent overall outcomes with the handy-approach and ISA procedures developed, the Federal Medical Centre (FMC) Owerri Nigeria, became the hospital that nursed the tinniest baby ever to survive in a Nigerian hospital with the use of these applications—baby "Majesty," as they nicknamed her, was born after 26 weeks GA weighing 550 g and went home after 83 days of nursing care on the 26th of April 2015 (**Figure 4**).
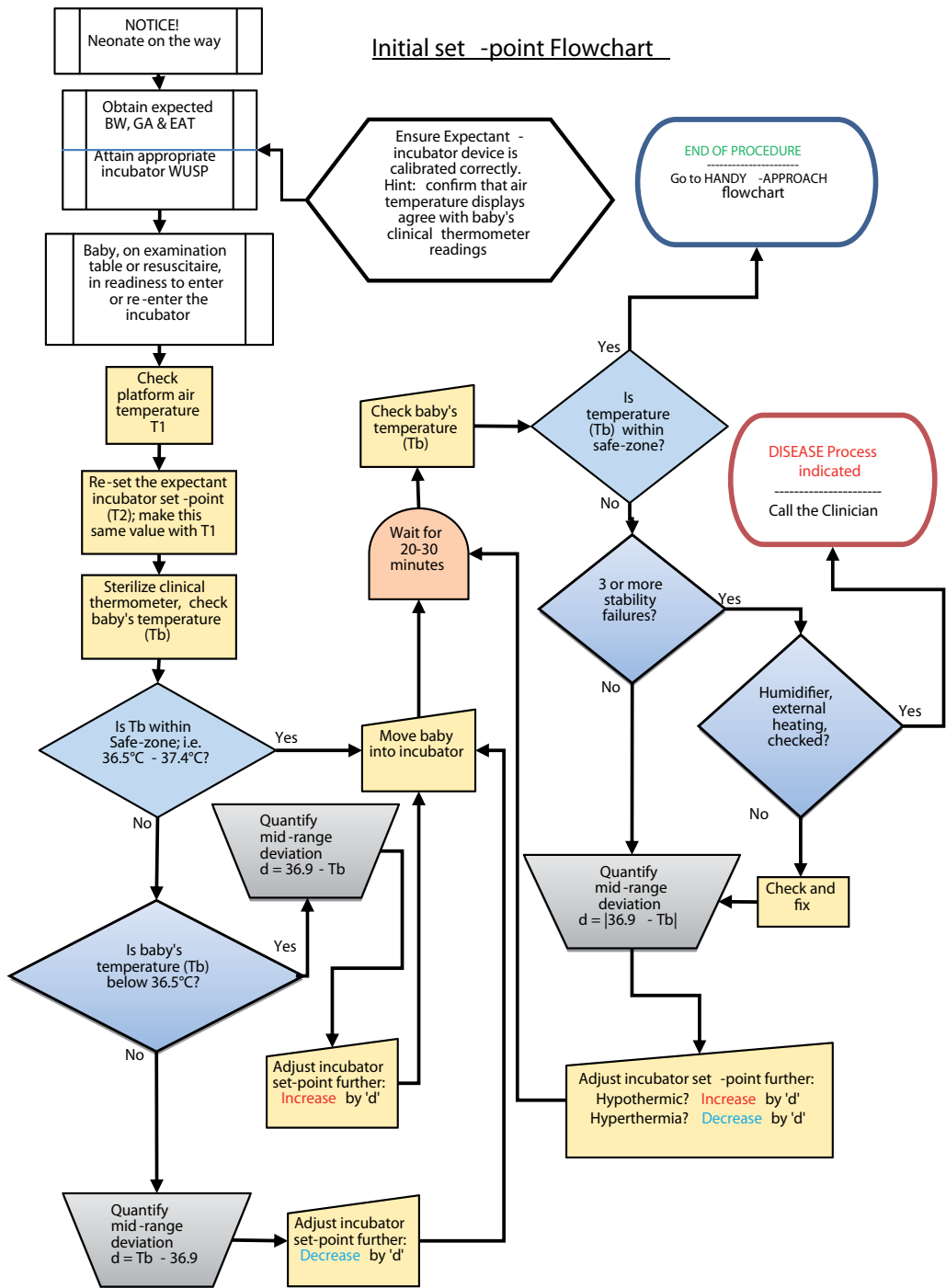
Figure 4. The ISA flowchart. Modified from Amadi et al. [15].

6.9. Pediatrics incubation technique (PIT) courses

Within the limits of our research findings, we can strongly affirm that poor thermal control is largely to blame for the high rate of mortality of premature neonates in Nigeria, especially during the first few days of life. It therefore became essential that the theories and etiology of neonatal thermal shock was well-understood and communicated to would be and practicing neonatal nurses and doctors. These were the roles expected of the pediatrics incubation technique (PIT) courses that were introduced very early in this research group. The PIT courses isolate the Nigerian peculiar climate, weather, culture, and financial dispositions, relating these factors to the neonatal need of adequate warmth for survival. The courses were used to teach practitioners on the best incubator practices within the Nigerian tropical contexts. In Nigeria, being a technologically dependent country in medical devices including incubators, the PIT discusses the tweaks that are appropriately necessary in order to get the best out of these foreign systems. The courses are elective at the Pediatrics Departments of our partner hospitals; however, it is mandatory for the SCBU staff of our centers. There is the desire for zero tolerance to the devastating effect of early neonatal thermal shock at our centers. Therefore, all staff of some of our SCBUs are required to achieve a minimum grade of "Merit" (65% or above) at both PIT level-1 and PIT level-2 courses to guarantee a good career progress at the centers.

7. Conclusions and recommendations

The Nigerian neonatal records are among the worst in the world. This has remained high for far too long, and there seems to be no fresh ideas—those that had not been previously tried or any immediate sustainable solutions from the government, health institutions, or the academia. In our work, we have carefully investigated the mortalities and struggles of the early days of premature neonates in Nigeria and can conclude that this is more than a "nightmare" for them. In the course of our various investigations and publications, we have identified that thermal inadequacies is one factor that must be tackled and decisively resolved before the Nigerian neonates that can be liberated.

We have synthesized unconventional solutions, tried, and validated these using rigorous methodologies to generate our often multicenter- and center-controlled data. The various comparative analyses against conventional techniques in Nigeria have also revealed that our unconventional approaches have succeeded where these failed irrespective of disabling policies and social factors at work. In a center-controlled cohorts study that tested a package of the developed unconventional techniques on extremely premature neonates (**Figure 5**), overall mortality rate (NNMR) fell from 63/100 for conventional techniques to 7/100; the fraction of neonates that would not see their seventh day after birth (f7dMR) fell from 51/100 to 0; and the fraction of the deceased neonates that died before the seventh day (f7d-in-NNM) in the conventional technique cohorts was quite similar to the World Health Organization demographic figure for this [15]. It is still a puzzle that the Nigerian health authorities and their corporate institutions have turned a blind eye to a package of methods so reliable; thereby
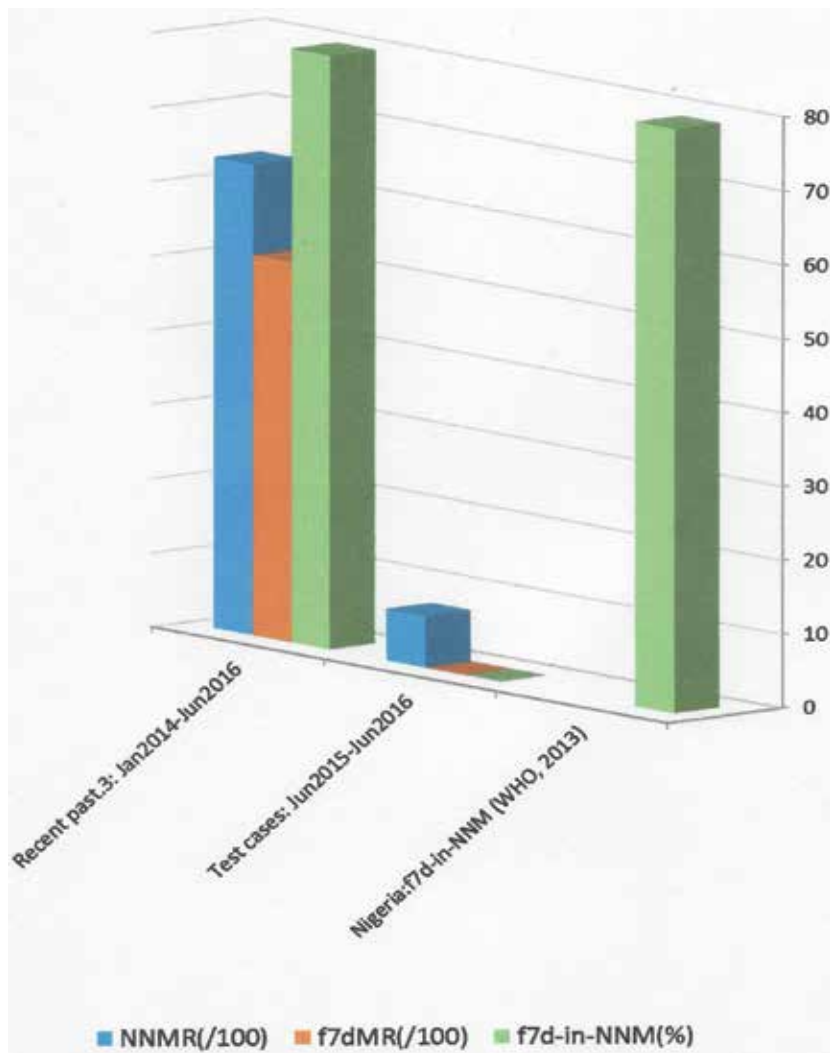
Figure 5. Comparative outcome of a centre-controlled cohort study of ELBW neonates managed with conventional techniques in "recent past" as against unconventional techniques on "test cases"; modified from Amadi et al. [15].

being uncompassionate as thousands of these neonates are sent to their untimely graves. If a set of techniques could so-smash the W.H.O.'s "f7d-in-NNM" demographic figure of 79% for Nigeria, literally eradicating this on a cohort of extremely premature neonates, what are they still waiting for before translating this idea into their national practice?

In the face of ever dwindling and crashing Nigerian economy, we think that our unconventional techniques as presented in our various publications have a great role to play if Nigeria must emancipate from this age long neonatal situation. The use of these affordable systems and ideas are recommended to replace the insatiable crave for unsustainable importation of ideas and technologies that are not consistent with the Nigerian climate, weather, and infrastructural development.

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Conflicts of interest

Both authors declare no conflicts of interests concerning this publication.

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Neonatology is one of the areas of greatest development and evolution within pediatrics. The technoscientific advances in this area have led to an increase in the survival of premature infants who sometimes require sophisticated care. However, there is essential care that must be included in all centers that care for high-risk babies. This book includes important topics related to neonatal care grouped into four sections. In 14 chapters that address relevant issues about neonatal care, the book seeks to contribute to the clinical work of the health teams of neonatal units. Specialists in the field of neonatology from different countries have developed these chapters and through them they hope to share part of their experience.

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