Review

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Posted Date: 25 December 2024

doi: 10.20944/preprints202412.2162.v1

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Review

Precision Medicine and Diagnosis of Neonatal Illness

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Abstract: Background/Objectives: Precision medicine is a state-of-the-art medicine tactic that tailors information about people's genes, environment, and lifestyle to aid the prevention, diagnosis, and treatment of various diseases.to provide an overview of the currently available knowledge and applicability of precision medicine in the diagnosis of different cases admitted to the NICU, such as encephalopathies, respiratory distress syndrome of prematurity, hemodynamic instability, acute kidney injury, sepsis, and hyperbilirubinemia. **Methods:** The authors searched the databases such as PubMed and PubMed Central, for the terms, neonatal "precision medicine", "personalized medicine", "genomics", "and metabolomics", precision medicine in the diagnosis of neonatal illness. The related researches were collected. **Results:** The review highlights the diagnostic approach that serves to implement precision medicine in the NICU and provide precision diagnosis, monitoring, and treatment. **Conclusion:** In this review, we projected several diagnostic approaches that provide precision identification of health problems among sick neonates with complex illnesses in the NICU, some are noninvasive and available in ordinary healthcare settings, while others are invasive or not feasible or still in ongoing research as machine learning algorithms. Future researches are needed for the wide implementation of artificial intelligence tools in the diagnosis of neonatal illnesses.

Keywords: precision medicine; neonatal brain injury; neonatal hemodynamics; neonatal RDS; neonatal sepsis; hyperbilirubinemia; AKI; BPD; pharmacokinetics; NICU

Introduction

Precision medicine (PM) is a state-of-the-art medicine tactic that tailors information about people's genes, environment, and lifestyle to aid the prevention, diagnosis, and treatment of various diseases. Precision medicine helps healthcare providers expect which management strategies will be valid with specific populations. Precision medicine aims to provide more accurate prevention of diseases, and point to the diagnosis mainly based on the genetics or molecular interpretation of the disease. Also, it focuses on the appropriate treatments for "the right patients at the right time".-Precision medicine has several advantages; among these, it facilitates recognizing the pathophysiological aspects of the disease and improves the use of specific treatments that best fit individual patients [1,2].

According to the U.S. National Research Council states that [personalized medicine" is an older term that refers to the same concept as "precision medicine" and carries the same meaning [3]. However, personalized medicine is distinct in some ways, as the term 'personalized medicine' infers treatments and prevention approaches adapted to the individual]. Traditionally, the application of personalized medicine has broader aspects [3]. Precision medicine was established in the late 1990s, still, neonatal medicine is late in applying PM.

Neonates admitted to neonatal intensive care unit (NICU) suffer from complex diseases with myriads of different pathology related to several systems and organs as respiratory distress syndrome (RDS), respiratory failure, and hemodynamics instability due to several causes such as patent ductus arteriosus and persistent pulmonary hypertension of neonates (PPHN), they also subjected to acute kidney injury (AKI), electrolytes disturbances, encephalopathy and brain injury, seizures, hyperbilirubinemia, and sepsis. Neonatal guidelines for the majority of diseases are available, which

 doi:10.20944/preprints202412.2162.v1

could be beneficial for some sick neonates, but not for others. Subsequently, the strategy of one-size-fits-all is not always useful. Newborn infants may present with complicated disorders related to immaturity as well as complications due to multiple organ dysfunction. Neonatologists may face difficult times in predicting the specific diagnosis and treatment of such infants. Hence, the precision medicine role will help to implement a well-organized plan of care, that certainly will improve the outcome and prevent the short and long-term serious outcomes and complications.

Precision medicine has emerged recently and utilizes huge amounts of biological data, including; genomics, transcriptomics, epigenomics, proteomics, metabolomics, and pharmacogenomics). These diagnostic tools provide patients with effective treatment related to their genetic, biological, and clinical features [3].

Although some diagnostic tools are used in daily practice, still, some as genomics and metabolomics are restricted to the research due to a lack of current knowledge in neonatology. Conversely, non-invasive tools such as NIRS, portable Ultrasounds, and functional echocardiography have been shown to support diagnosis and guide the management of sick neonates in the NICU [4–6].

This review aims to present an overview of the currently accessible knowledge of precision medicine in the diagnosis of various cases admitted to the NICU, such as encephalopathies, respiratory distress syndrome of prematurity, hemodynamic instability, acute kidney injury, sepsis, and hyperbilirubinemia.

Information Sources: The authors looked at known databases such as PubMed and PubMed Central, mainly using the terms, “precision medicine”, “personalized medicine”, “genomics”, “metabolomics”, precision medicine in the diagnosis of neonatal RDS, precision medicine in the diagnosis of neonatal sepsis, precision medicine in the diagnosis of seizures and precision medicine in the diagnosis of neonatal sepsis precision medicine in the diagnosis of hemodynamic instability and neonatal encephalopathy.

1. Neonatal Human Genome and Precision Medicine

The neonatal genome projects applied the next-generation sequencing (NGS) to recognize curable pediatric onset genetic diseases, this facilitates early diagnosis, permits precise drug usage, and prevents complications [7]. Also, the BabySeq project revealed differences between healthy and sick neonates' genomic sequencing. Subsequently, sick neonates may need further analysis to identify the specific treatment associated with their clinical features and pharmacogenomic variants [8]. 25% of genetic disorders related to death in NICU could receive treatment if were early diagnosed [9]. The intent of the genomic sequencing in acutely ill neonates project was to evaluate the usefulness of rapid next-generation sequencing versus the existing diagnostic practice and treatment guidelines as well as the prognostic data in acutely ill neonates and infants. It was reported that whole genome sequencing (WGS), and rapid whole genome sequencing (rWGS) may improve the diagnosis promptly compared with regular genetic testing [10]. These sequencing techniques seem to have a beneficial value as they encompass millions of tests at a time instead of doing multiple tests. WGS determines the arrangement of all the nucleotides in the human being's DNA and can detect variants within the genome regions [11]. Identification of gene variants may aid in expecting neonates at risk of severe disorders. Moreover, it could assist in the prevention and in providing specific treatment. Nevertheless, there are limitations to the WGS, as it cannot diagnose some disorders such as congenital hypothyroidism, and is an expensive technique, especially in developing countries. Also, there are some ethical concerns related to affected families.

Precision medicine through genetic studies can assess the risk of early complex diseases that neonatologists faced daily. These complex diseases are affected by genetics and environmental factors. Distinguishing the genetic knowledge that contributes to the vulnerability, seriousness, and complications of these diseases will ensure early diagnosis and better outcomes. Variants in Brain-Derived Neurotrophic Factor (BDNF) were connected with the severity of retinopathy of prematurity [12]. The polymorphisms in the high mobility group box 1 protein (HMGB1) are related to the necrotizing enterocolitis predisposition as well as survival [13]. The risk for neonatal sepsis is related to

 doi:10.20944/preprints202412.2162.v1

immature immune mechanisms, environmental and maternal factors, and neonatal genomics [14]. The integration of the genotypic information from newborns with basic clinical data could predict the probability of bronchopulmonary dysplasia [15]. Moreover, the genotype and phenotype can be used for appraisal of the risk and drug use among neonates. Polygenic risk score (PRS) depends on data from genome-wide association study (GWAS) and is composed of risk alleles, risk magnitude for each allele as well as prevalence of the diseases. So PRS can assess the risk for each patient [16]. Moreover, it highlights preventive intrusions, suggests the age of onset, recommends lifestyle modification, and evaluates family risk for the diseases. However, PRS could cause false positive results [7].

Artificial intelligence as Machine Learning (ML), may be used to merge and organize the genotypes and phenotypes for appraisal of the possibility of diseases.

2. Brain Injuries

2.1. Early Brain Injury in Neonates and Neonatal Encephalopathy

One of the important aspects of NICU neonatal care is to preserve brain function and improve the quality of life for NICU graduate neonates. Till now, it is difficult to anticipate the risk factors or to differentiate the various causes of brain injury, especially during the early neonatal period and among complex cases. Early brain injury may be related to different categories; maternal comorbidities, placental abnormalities, encephalopathy whether hypoxic ischemic (HIE) or non-HIE, vascular incidents such as IVH, especially among preterm infants, coagulopathy as arterial ischemic stroke, and periventricular venous hemorrhagic infarction, trauma, perinatal infection such as cytomegalovirus, rubella, chickenpox or toxoplasmosis, jaundice, metabolic disorders, and genetic/epigenetic anomalies. These causes may be fatal or lead to permanent brain damage and cerebral palsy. Neonatal encephalopathy(NE) is one of the top causes of admission to NICU, mainly among full-term infants. The clinical definition of NE is a "clinical syndrome of disturbed neurologic function in the first week after birth in an infant born at or beyond 35 weeks of gestation, manifest by a subnormal level of consciousness or seizures, often accompanied by difficulty with initiating and maintaining respiration, and depression of tone and reflexes [17,18]." The term HIE is usually used as synonymous with NE, however, HIE is just one of the subclasses of NE. NE may be due to other causes rather than perinatal hypoxia-ischemia. NE is interrelated to multiple risk factors such as sepsis, trauma, or vascular events [19]. Neonatal HIE is recognized as the predominant risk factor for brain injury, accounting for 50%–80% of cases that present in NICUs [20].

The definite final diagnosis of HIE is based on retrospective data that include the suggestive history and nature of neurological impairment. The neonatal diagnostic manifestation of HIE is presumptive, the ACOG-AAP task force proposes it includes low Apgar score <5 at 5 and 10 min; fetal umbilical acidemia (pH < 7.0 or base deficit ≥12 mmol/L); with a history of abnormal fetal heart rate before birth, then confirmation by radiological verification of brain damage on MRI or magnetic resonance spectroscopy. Multi-organ dysfunction is among the suggestive diagnostic criteria that succeeding acute peripartum /intrapartum hypoxic or ischemic events occur immediately before or during labor. The evolving outcomes may include spastic quadriplegia or dyskinetic cerebral palsy [17].

Although it seems to be decisive, the ACOG-AAP diagnostic criteria may not be very specific to exclude other causes of NE as sepsis, trauma, or vascular events or to include all HIE cases too. Moreover, there may be a combination of several risk factors, subsequently leading to inadequate management of the non-HIE causes. Furthermore, the previous diagnostic criteria do not specify the timing, duration, extent, or magnitude of severity of hypoxia insult or consider the individual response to the precipitating event. It does not adequately stratify the grades of HIE. The reliance on the diagnosis of the mild stage by Sarnat and Sarnat classification affects the decision for TH; unfortunately, 25 % of those diagnosed as mild HIE had abnormal neurological/developmental outcomes and cerebral palsy [21]. It was shown that the AAP-ACOG definition could miss some cases of HIE, due to the non-inclusion of some aspects; milder acidosis or higher Apgar scores; [22,23] a typical MRI

 doi:10.20944/preprints202412.2162.v1

diagnostic criteria, progressive, unusual course of HIE;[24]. Subsequently, the final diagnosis may not be finalized till a neurodevelopmental delay or impairment is seen later in affected infants /children.

A study by Lee et al. reported that a not negligible percentage of HIE were due to impaired blood flow or oxygen delivery during birth as around fifty percent of the studied cases had no clinical or radiological evidence of perinatal asphyxia [25].Non-HIE NE causes can be misinterpreted as HIE at birth. Respiratory distress and abnormal tone can be seen in congenital neuromuscular diseases, such as congenital myotonic dystrophy, and metabolic and/or genetic abnormalities may be started early with abnormal levels of consciousness, convulsion, and breathing difficulties [19].

A definitive early precision diagnostic criterion for the different causes of NE is still needed to discriminate the wide diseases and abnormalities that cause NE [26]. The distinction between subclasses of NE at birth is difficult. Early signs and symptoms may be similar for diverse etiological diseases. The advancement in TH and other neuroprotection therapy, and the need for early support of specific therapy for non-HIE encephalopathy as metabolic causes or rapid interference for surgical causes necessitate, an urgent individualized approach to save newborn infants from permanent brain damage.

As a result of all of these limitations over-diagnosis, underdiagnoses, or misdiagnosis of HIE as well as other causes of brain injury may ensue. Each neonate's life and quality of life matter to his family and society. This emphasizes the need to personalize precise data on various risk factors, symptoms /signs, nature, and sites of brain damage to plan precision medicinal therapy.

Advances in PM may allow precise diagnosis, and offer proper on-time adequate neuroprotection therapy based on the definitive individualized need during high brain plasticity. Precision medicine may permit recognition of the risk factors, grade of insults, and magnitude of the severity of brain damage based on interpreting the results of ultrasound, MRI, EEG, genetic profile, and developmental millstones. Additionally, the ongoing studies and research that currently are taking place should be encouraged and supported to facilitate the implementation of precision medicine in the NICU.

In this review article, we present some recent promising research related to prognosis and management (PM) in the area of brain injury. These studies show the rules of precision medicine in anticipation, recognition, prevention, and reinforcing appropriate decisions for neonates with brain injury in the NICU.

Genetic testing can help diagnose (HIE) and other causes of neurodevelopmental disorders by identifying genetic and epigenetic abnormalities. Additionally, it can provide insights into the severity and extent of brain damage and inform neuroprotective treatment options.

Genetic testing using targeted gene panel sequencing can identify the underlying cause of NE and associated hypoxic brain injury, irrespective of the initial clinical presentation. Lee et al. reported that 32.4% of the studied cases had pathogenic variants that involved 9 genes; CACNA1A, KCNQ2, SCN8A, STXBP1, NSD1, SCN2A, PURA, ZBTB20, and ENG.

Personalized treatments developed according to the data of the studied genetic tests, such as the sodium-channel blockers for neonates with KCNQ2 or SCN8A variants and the implementation of a ketogenic diet in cases with STXBP1 or SCN2A mutations, these therapies showed particular usefulness among the studied cases [25].

Zhang et al study magnifies the mutation range of genes related to NE and provides new confirmation for molecular diagnosis in unexplained neonatal encephalopathy. They reported fifteen NE-related pathogenic variants. Seven of the pathogenic variants related to the identified 12 genes were related to premature translation termination, and four variants were destructive to the protein structure of KCNQ2 [27]. A cohort study by Yang et al. identified 30 genes for NE. Epileptic encephalopathy represented the major gene ratio (58.5%) followed by metabolic and syndromic (18.9%) then the mitochondrial in 3.8% of the studied cases. The greatest affected genes were epileptic-related genes *KCNQ2* and *SCN2A*. There are ten genes recognized to be encephalopathy-causing genes, comprising KCNQ2, SCN2A, SCN1A, KCNT1, CDKL5, STXBP1, and ARX related to epileptic

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encephalopathy, SERAC1, and AMT linked to metabolic encephalopathy, and MECP2 in syndromic encephalopathy [28]. It has been shown that epileptic genes ALDH7A1, DEPDC5, and PRRT2, metabolic genes DBT22 [29]and MMACHC [30] mitochondrial-gene NDUFA11[31], and nine syndromic genes cause diverse brain abnormality, and/or nervous system damage, and/or encephalopathy. A study by Paolo et al. showed marked differences in gene expression profile in neonates with neonatal encephalopathy at the time of birth in response to hypoxia compared with healthy control and septic neonates [32]. Accordingly, genetic testing may empower personalized neuroprotective treatment; if a known specific variant is identified and known target therapy is available too**.** A study of the transcriptomic profile of adverse neurodevelopmental outcomes after neonatal encephalopathy disclosed a marked expression of genes associated with melatonin and polo-like kinase pathways in neonates with adverse outcomes. It was proposed that transcriptomic profiling could help for rapid risk stratification in neonatal encephalopathy [33].

There were significant differences in the genome expression profile during the first three days after birth between newborn infants with HIE from developed countries and neonates from South Asia, this may explain the inadequate response to TH among infants in developing countries. The adverse outcome was related to the attenuation of eukaryotic translation initiation factor 2 in the high-income countries neonates while aldosterone signaling in epithelial cells in the South Asia neonates [34]. Subsequently, according to this study, the whole-blood genome expression profile possibly will be beneficial for the diagnosis, and personalized neuroprotection in HIE, and for observing the response to therapeutic remedies. However, we have to look at the race differences when evaluating the difference in response to therapy, in addition, future research will help to elucidate the link of genetic variation in categorizing NE or screening the response to therapy.

Artificial intelligence (AI) was applied in research to add value to neuro-precision medicine. The machine learning algorithm (ML) was used and validated for identifying infants at risk of HIE by Murray et al. The algorithms were applied to compute a probability index for the occurrence of HIE. The model may assist in early diagnosis and hasten recognition of neonates who need continuous neurological or neurophysiological monitoring [35]. The availability of such models assists in timely diagnosis and judgment for therapeutic decisions. The ML models using MRI findings were shown to be able to predict neurodevelopmental outcomes in newborn infants with hypoxic-ischemic encephalopathy throughout all neurodevelopmental domains and the outcomes [36]. A detailed MRI scoring system allows prognostic appraisal for motor, cognitive, and composite outcomes. Damages in the deep grey matter and cerebellum were prognostic of unfavorable outcomes [37].

The recent ML models of cerebral oxygenation (rcSO2) for the detection of brain injury in term neonates with HIE have more power as the algorithm can merge a selection of features to predict outcomes rather than one quantifiable approach to summarize the signal. It is based on automated models of NIRS. The researchers designed distinct ML models applying clinical variables for the comparison of functions. This model looked at the rcSO2 features and their relation with the severity of HIE as classified by modified Sarnat score at 1 h. The study concluded that automated analysis of regional cerebral oxygen saturation rcSO2, using either ML or deep learning methods, was able to determine infants with adverse outcomes [38].

Looking at another diagnostic point for early brain damage and intraventricular hemorrhage in extreme preterm infants, the ML model was used for clarifying the regional cerebral oxygenation ($rcSO_2$) and peripheral oxygen saturation (SpO^2) signals. SpO^2 may not be enough for early prediction of brain injury. Low $rcSO^2$ could indicate hypoxia, decreased blood flow, vasoconstriction, and/or abnormal cerebral autoregulation.

ML model was developed using the combination of early EEG background analysis and clinical data to support the expectation of neonates with HIE who were at the highest risk of seizures, hours before seizures appeared. This reinforces the application of automated measurement tools for early assessment of HIE [39]. Another study showed the role of automated models for the diagnosis of neonatal encephalopathy by applying the aEEG deep neural networks. The model design allows superior feature extraction and classification in a data-driven method, the researchers confirmed that

 doi:10.20944/preprints202412.2162.v1

deep neural network methods may advance the precision identification of neonatal encephalopathy, particularly for describing the EEG background pattern, sleep-wake-cycling, and seizure that help neonatologists for precision diagnosis [40].

However, the diagnostic role of machine-learning methods has to be proven with more studies and large cohorts of neonates. Some studies need to overcome limitations such as the stage of illness, the timing of performance of ultrasound or MRI, the timing of the incident of IVH, and synchronization of the measurement as SPO2 and rcSO2 [41].

Metabolomics provides the metabolic status of cells, tissues, or organisms concerning genetic variations or external stimuli. Metabolomics patterns may be valuable in the early detection of perinatal asphyxia-induced HIE. It could aid in planning individualized neuroprotective and therapeutic hypothermia (TH) strategies [42]. Animal models showed that metabolomics is a promising tool to expand the interpretation of HIE pathophysiology, diagnosis, and therapeutic interventions. The metabolite 2-phosphoglyceric acid was decreased in OGD/R-induced HT-22 cells and participates in the neuroprotection of OGD/R-causes cell demise and HIBD-provoked neuronal injury. Applying 2-phosphoglyceric acid as a therapeutic agent decreases brain injury and neuronal damage by down-regulating Acyl—CoA synthetase long-chain family member4 and increasing glutathione peroxidase 4 expression. They suggest that 2-phosphoglyceric acid could be a promising neuroprotective in brain damage due to HIE [43]. The study of metabolomics is promising for individualized therapy.

Accordingly, recent research studies are helping to modernize the neonatal practice to proceed in implementing personalized and precision medicine for neonatal neurologic illness. The previously mentioned studies were about evolving new methods that were based on ML models and automated exploration of data that were controlled by experts who applied MRI, NIRS, and EEG as well as genetic studies and omics. These new tools may offer a fast, effective tool for the prediction, diagnosis, and categorization of brain injury. However, more studies are still needed to validate the efficacy of these tools with a larger cohort of neonates to support future implementation for the prevention, diagnosis, and treatment of brain injury. before it becomes permanent. Hopefully, these tools be simple and friendly to use especially during the first hours after birth to protect the brain of newborn infants.

These new tools can support decisions for more precise diagnosis monitoring and therapy; genetic testing could assist in the prediction of the possibility of insults and adverse outcomes and can differentiate subclasses of NE and precise therapy. ML models can support decisions by monitoring the progress of illness and the prediction of outcome; metabolomics supports individualized therapy.

2.2. Neonatal Seizures

Neonatal seizures are among the major clinical problems in NICU; they may be too distinct to be the cause of admission or subclinical and detected with monitoring of infants who were admitted due to other diseases. Usually, a seizure is a sign that reflects brain injury [44]. [Seizures are “sudden, paroxysmal, repetitive, stereotypical events, and abnormal alterations of electrographic activity from birth to the end of the neonatal period [45]. It is then further categorized as clinical only, electroclinical, or electrographic only based on the EEG findings” [46]. “The American Clinical Neurophysiology Society defined an electrographic neonatal seizure as “a sudden, abnormal EEG event, defined by a repetitive and evolving pattern with a minimum 2 μV peak-to-peak voltage and duration of at least 10 seconds.” “Evolving” is defined as an unequivocal evolution in frequency, voltage, morphology, or location, for example, increasing amplitude and decreasing frequency of discharges over time. This definition does not require any evident clinical change” [44–47]. According to the classification of the International League against epilepsy ILAE; seizures are motor or nonmotor, whereas motor seizures are predominantly focal or multifocal and include; automatisms, clonic, epileptic spasms, myoclonic, sequential, and tonic. Nonmotor includes; automatisms and behavioral arrest [46,47].]

The Clinical identification of seizure morphology per se is not diagnostic in every infant; some signs may mimic normal behavioral activities or even no physical signs at all, depending upon the

 doi:10.20944/preprints202412.2162.v1

etiology, grade of severity, and course of illness [48,49]. Severe HIE may not show clinical seizures. However, ictal activities may be seen in EEG.

Severe illnesses in newborn infants such as HIE, vascular stroke, hemorrhage, infection, hypoglycemia, or electrolyte imbalance presented with acute symptomatic seizures and accounted for a high percentage of neonatal seizures. Some respond to treatment of the related risk factors such as hypoglycemia or hypocalcemia. However, some seizures do not respond to general management as neonatal epilepsy syndromes related to structural abnormalities, genetic syndromes, or inborn errors of metabolism. These seizures are still difficult to recognize and may miss early proper therapy [46]. The neonatal-onset epilepsies are roughly about 10-15% of neonatal seizures [50].

One of the challenges for early diagnosis of neonatal seizures is the nature of the seizure; the majority of seizures are exclusively electrographic, while clinical seizures have no electrographic findings [51,52], subtle and subclinical seizures possibly will not be noticeable [53]. However, it was suggested that seizure semiology could suggest the primary illness; focal clonic seizures are significantly associated with vascular stroke or infection while focal tonic seizures are related to genetic epileptic encephalopathy. Autonomic seizures could be associated with bleeding and myoclonic seizures signify inborn errors of metabolism [54,55].

One of the diagnostic and monitoring tools in NICU is the aEEG/EEG as well as near-infrared spectroscopy. The aEEG is useful for detecting repeated seizures and status epilepticus, also the trend provides instant and significant data regarding the evolving brain functions over time, and during sleep-wake cycling [56]. Conversely, a systematic review report revealed that the precision of the aEEG alone is not adequate and inconsistent for recognition of seizure [57]. Barely thirty percent of one-time seizures can be perceived in the trend of aEEG, but the precision of the trend increases when seizures are more recurrent and with longer duration [56]. Subsequently, from this data, multi-channel EEG recordings and conventional EEG, continuous video-EEG are important for diagnosis, monitoring, and evaluating the efficacy of therapy [58,59].

Genetic testing as whole exome sequencing may identify about 75% of neonates with neonatal epilepsy [60,61], use of whole exome sequencing to avoid the phenotypic intersect of different genetic epilepsies [61,62]. Self-limited familial neonatal seizures which are AD can be differentiated by a genetic disorder in KCNQ2, KCNQ3 SCN2A, early-infantile epileptic encephalopathy associated with structural brain malformations is linked with genetic variants in ARX, CDKL5, SLC25A22, STXBP1, KCNQ2, SPTAN1, SCN2A, and metabolic disorders. Early myoclonic encephalopathy associated with metabolic disorders, genetic variants are in genes STXBP1, TBC1D24, GABRA1, epilepsy of infancy with migrating focal seizures is related to pathogenic variants of KCNT1, SCN2A, SCN1A, SLC25A22, PLCB1, QARS genes. Self-limited neonatal seizures have a favorable outcome and may disappear within 48 hours, on the contrary, early-infantile epileptic encephalopathy has early-life mortality or severe developmental disabilities [46].

Other challenges for diagnosis of neonatal seizures are the seizures mimic movement such as jitteriness, hyperekplexia, benign sleep myoclonus, rapid eye movement sleep behavior, apnea, various motor automatisms, and dystonic or tonic posturing provoked by stimulation[47,63,64]. Although it could be differentiated by abolishing with touch or repositioning the limb or no abnormal changes in EEG, it may represent contributory disease or could be related to nonelectrographic seizures [64].

Subsequently, precision diagnosis batteries have to include the EEG to detect electrographic activities, and neuroimaging for recognizing disorders in the brain structure, involving hemorrhage, infarction, or defect of cortex development, as well as chemical tests for glucose and electrolytes. Additionally, genetic testing is essential to offer individualized treatment and achieve better neurocognitive development outcomes. Precision diagnosis will direct to specific therapy that suits the individual variability such as phenobarbitone, fosphenytoin, phenytoin, and levetiracetam for treatment of acute symptomatic seizures, or TH in HIE. Medication such as conventional sodium channel inhibitors are proposed for defects in SCN2A and SCN8A genes, on the contrary, may be worse if given to cases with defects of gene SCN2A.Pyridoxal phosphate is suitable for pyridoxal-phosphate

 doi:10.20944/preprints202412.2162.v1

dependent epilepsy, and a low-purine or low-protein diet for molybdenum cofactor deficiency with cyclic pyranopterin monophosphate if the defect in gene MOCS1 [65].

The diagnostic and supervising methods appear insufficient to precisely predict or reinforce the diagnosis of seizures in the NICU. There is a need for a machine learning model to be developed and include the essential data such as gestational age, age of onset of seizures, category of seizure, duration of seizures, semiology, MRI and other neuroimaging, and electrical activities of the EEG, to support precision diagnosis of aetiological causes of seizures and support therapy. Future researches are needed to support the whole care of neonates with seizures, which is a suitable screening method for the prediction and early detection of seizures in busy NICU, what is the right medicine/therapy for this specific newborn infant, and for how long should this medication will be given?

3. Heamodynamic Disturbances

3.1. Patent Ductus Arteriosus (PDA)

Precision medicine is important to ensure the management of the hemodynamic stability of neonates admitted to the NICU. Patent Ductus Arteriosus(PDA), hypotension, and shock as well as persistent pulmonary hypertension of the neonates (PPHN) are among the common problems that need timely diagnosis and management. The hemodynamic instability among these infants may be due to the response to stress factors, such as hypoxia, infection or hypoperfusion, immature physiological response, and inadequate compensatory mechanisms. Moreover, congenital heart disorders, mechanical ventilation, and medications such as chronotropic, analgesic, and muscle relaxants may also participate in hemodynamic instability. Usually, we depend upon indirect measures to monitor the hemodynamics; such as heart rate, blood pressure, capillary refill time, urine output, and blood gases [66]. The mechanisms of instability and primary pathophysiology are not similar for each neonate. Subsequently, to improve the outcomes of these infants, personalized medicine aims to make a precise diagnosis of the underlying precipitating cause, followed by precise treatment medication and monitoring the effect of treatment.

Assessment of Patent Ductus Arteriosus

Hemodynamic assessment of PDA using transthoracic echocardiography is a golden standard for diagnosis and monitoring of treatment; clinical signs only are undependable. 2D Doppler echocardiography targeted neonatal echocardiography and Cardiac Point-of-Care Ultrasound, can be used for diagnosis and therapeutic precision in NICU [67]. Echocardiography reveals the hemodynamic significantly PDA, (hs PDA) and hence the need for treatments. Echocardiography data display the shape, size, and volume of the shunt as well as the pulmonary pressure and other structural cardiac lesions including duct-dependent cardiac defects [5,68]. Neonatologist-performed echocardiography has proven to enable a longitudinal assessment for early recognition of a PDA in preterm infants with high flow ductal volume, hence assisting in planning for appropriate therapeutic intervention as drug dosages and interval [5].

In preterm infants, it is important to estimate the volume of transductal left-to-right shunt, to recognize those who will need immediate active medical therapy. If not treated shunt may cause increased pulmonary blood flow and decreased systemic perfusion, which could be related to multiorgan comorbidities such as pulmonary hemorrhage, bronchopulmonary dysplasia, Intraventricular hemorrhage (IVH), Necrotizing enterocolitis (NEC), or focal intestinal perfusion as well as brain injury periventricular leukomalacia, and impaired school performance [69,70]. The assessment of PDA has to evaluate ductal characteristics, ductal size, magnitude, and impact of the shunt assessment of pulmonary circulation and systemic hypoperfusion. [PDA size is considered small when less than “1.5 mm, moderate from 1.5 to 1–2 mm, and large more than 2 mm”. Also, evaluation determines the” flow direction; left to right, right to left, or bi-directional, and Doppler assessment; maximum velocity (Vmax) in systole and end-diastole. Assessment of pulmonary over circulation is recognized when dilated left side of the heart and LA/Ao ratio is considered mild <1.4, moderate 1.41–1.6, severe >1.6, or when LVEDD or LPA diastolic velocity—mean velocity >0.42 m/s, end-diastolic velocity >0.2

 doi:10.20944/preprints202412.2162.v1

m/s, or reversal of mitral E/A ratio. Moreover, to verify the significance of intra-atrial shunt. The markers for systemic hypoperfusion are retrograde or absent blood flow during diastole in the descending aorta, or coeliac trunk or superior mesenteric artery, or anterior or middle cerebral artery" [71,72].] However, some of these measurements have interobserver variability [73]. Generally, the diagnosis of the hsPDA has to include at least the following: "1. Diameter of the ductus arteriosus > 2.0 mm 2. Ductal flow pattern ('growing' pattern or pulsatile with Vmax < 2 m/s and Vmax/Vmin > 2) 3. Retrograde post ductal aortic/coeliac/SMA diastolic flow 4. La/Ao > 2 5. LVO > 300 ml/kg/min 6. Mitral valve E/A ratio > 1 [71].

Spontaneous closure of the duct is favored by some molecular, physiological, and structural factors. Some genetic factors are associated with increased risk of PDA as TFAP2B (rs987237), TRAF1 (rs1056567), AGTR1 (rs5186), while others as PTGIS (rs493694, rs693649), ESR1 (rs2234693) and IFN-g (rs2430561 are linked to decrease the risk of PDA. It was reported the possibility of the "rs7557402 SNP" variant with the failure of Ibuprofen treatment of PDA closure [70,71]. This genetic variance may explain the resistance to therapy in some cases.

Other diagnostic tools for PDA include platelet count and indices, cardiac peptides and near-infrared spectroscopy (NIRS), proteomics analysis, and Machine learning prediction models [72]. Recent evidence proposed that low platelet counts and platelet dysfunction are associated with unsatisfactory response to treatment [74]. Cardiac peptides such as BNP/proBNP and cardiac high-sensitivity troponin T correlated with hsPDA [75,76]. Near-infrared spectroscopy is a noninvasive monitoring tool for regional tissue oxygenation and aids in preventing multiple organ comorbidity in other organs such as the brain, kidney, and intestine. [77]. Lung ultrasound can also postulate early detection of hsPDA through evaluation of pulmonary edema levels, in extremely preterm infants [78].

The perfusion index (PI) is a simple, continuous parameter provided by pulse oximetry to access the peripheral perfusion, and it could help to predict a significant PDA [77,79] Electrical Cardiometry/electrical velocimetry can be used in addition to echocardiography since it is non-invasive, monitors the cardiac output, and proves to be safe and feasible [77]. Newly emerging studies showed proteomics analysis differences in 21 proteins and 8 cytokines between neonates with a large PDA and neonates without a PDA, there was an elevation in angiotensinogen, periostin, pro-inflammatory associations, including interleukin (IL)-1β and IL-8, and anti-inflammatory associations, including IL-1RA and IL-10, while complement factors C8 and carboxypeptidases were decreased. The study reports the association between the PDA and the renin-angiotensin-aldosterone system and immune- and complement systems [80]. Another study showed that the level of plasma protein disulfide-isomerase A6 (PDIA6) was downregulated in patients with PDA, it may have potential clinical implications for PDA treatment and provide evidence regarding the etiology and molecular mechanism of PDA [81]. Proteomic studies may implicate a new insight into the pathogenesis and approach for the management of the PDA, especially among refractory cases.

Machine learning was developed to calculate the effectiveness of treatment for hsPDA closure. Artificial intelligence(AI) using deep learning and convolutional algorithm model identified PDA with 0.84 positive predictive value, 0.80 negative predictive value, 0.76 sensitivity, and 0.87 specificity [82]. ML was developed to enable the identification of the signature of sounds of PDA and congenital heart diseases with promising results [82]. Park et al study showed that an AI-based PDA diagnostic support system has 84% accuracy and the ability to detect PDA symptoms up to 3.3 days in advance [83]. The implementation of AI-dependent models can assist early prediction of infants and provide on-time treatment, however, studies are needed to include a dataset that can provide more accuracy for these models.

Treatment is directed to hemodynamically significant PDA. Recently it has been advised to monitor infants for spontaneous closure of the ductus arteriosus and avoid prophylactic treatment, a decision has to be made based on the clinical, echocardiographic, and NIRS evaluations and include the neonate's gestational and postnatal age. The pharmacologic therapy has been proposed for hsPDA and includes drugs that inhibit prostaglandin as cyclooxygenase inhibitors; indomethacin and oral or intravenous ibuprofen, and paracetamol (acetaminophen) [84].

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However, there is an abnormal response to the pharmacological treatment of PDA due to genetic variances. The low clearance of indomethacin and ibuprofen are related to the cytochrome P450-related enzymes; CYP2C9 and CYP2C8. Nonresponsiveness to pharmacological treatment is related to genes associated with prostaglandin and NO action or synthesis, such as SLCO2A1, PTGS2, and NOS3 [85].

Precision diagnosis of PDA depends mainly upon echocardiography which can predict hsPDA, longitudinal assessment for the response for pharmacotherapy, NIRS determines tissue oxygenation assist to support other organs and facilitate stabilization of the hemodynamic of newborn in the NICU. Other diagnostic tools may play a role in the future as proteomics, and machine learning tools for the prediction of PDA. Treatment is reserved for hsPDA and should be based on the integration of neonates' personalized needs, and clinical data in conjunction with echocardiography markers.

3.2. Neonatal Shock

Shock and Crashing Neonates

Sudden unexpected clinical deterioration or cardiorespiratory instability in neonates is often referred to as a "crashing" neonate. It is an apparent life-threatening event. Several risk factors are associated with crashing neonates; trauma (accidental –nonaccidental as abuse), heart disease structural and nonstructural; hypovolemia /arrhythmias/Hypoxia, endocrinopathies (congenital adrenal hyperplasia -thyrotoxicosis), metabolic abnormalities (electrolyte imbalances), inborn errors in metabolism, seizures, formula mishaps(mix-ups/ under or over-dilution), intestinal catastrophes (necrotizing enterocolitis, intussusception, and midgut volvulus) /omphalitis, toxins exposure /poisons, and sepsis (meningitis, pneumonia, urinary tract infection).

Shock is a state when there is a discrepancy between oxygen need and delivery on a cellular level. Subsequently, physiologic compensatory mechanisms are initiated to overcome the insults, and according to the response three shock phenotypes may be seen; compensated shock, uncompensated shock, and irreversible shock [70].

In neonates, the compensatory phase may pass unnoticed. The blood pressure could be normal or even high and not reflect the low cardiac output. This means we cannot rely on blood pressure only to anticipate hemodynamic instability(HI) and shock in newborn infants. Precision diagnosis of shock is a must, sick neonates have limited and immature compensatory mechanisms and may end up with irreversible shock in no time. Thorough cohesive hemodynamic monitoring using clinical data such as blood pressure, pulse pressure, capillary refill time (CRT), and oliguria, NIRS to assess the regional tissue oxygenation and assess tissue oxygen saturation (StO2), fractional oxygen extraction (FOE) will be helpful for diagnosis before the increase of lactic acidosis. Moreover, evaluation of cardiac output by transthoracic echocardiography /targeted /neonatal performed echocardiography as well as biomarkers of organ dysfunction as lactic acid will aid in the early individualized treatment of sick neonates in the NICU and avoid the complications of low cardiac output [5,86]. Furthermore, cardiac output can be assessed with other tools as electrical biosensing technologies, and transpulmonary ultrasound dilution [70].

Recently, point-of-care ultrasound (POCUS) has been integrated into a complex clinical situation and emergencies in the NICU. The first step in the decision tree is ruling out cardiac tamponade followed by pneumothorax, pleural effusion, then acute critical aortic occlusion, acute abdominal complications, and severe intraventricular hemorrhage [87]. Another POCUS protocol by Elsayed et al. was developed, experts take ten minutes to screen life-threatening conditions. However, POCUS screening cannot detect congenital heart diseases, detailed assessment of pulmonary hypertension, ventricular functions, or arrhythmias as it is not the scope of POCUS [88].

A new physiologic-based integrated algorithm for the prediction and assessment of neonatal (HI) was published, this algorithm takes into consideration the heterogeneity of the different physiologic mechanisms causing hemodynamic instability. HI was categorized into 5 classes based on blood pressure, echocardiographic markers, and oxygen indices, which are hemodynamic instability due to vasodilatory physiology, hemodynamics instability due to vasoconstrictive physiology,

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cardiogenic shock, volume depletion, and left to right shunt physiology. The algorithm incorporates a combination of the integrated monitoring markers to facilitate the diagnosis of shock phenotype e.g. hemodynamic instability due to vasodilatory physiology can be predicted when there is a decrease in the diastolic blood pressure or trending down, decrease in systolic and mean blood pressure with normal pulse pressure, low systemic vascular resistance, brisk CRT, PI > 3 and oliguria or urine output < 1 ml/kg/h for at least 12 hours beyond the first 12 hours after birth. NIRS markers show an increase in end-organ oxygen extraction which is a warning of compromised autoregulation and low oxygen delivery, and if amplified extraction moves to full capacity, lactic acidosis is an ominous sign of organ failure [89].

Using such algorithms enables individualized treatment for the shock phenotype according to the HI precipitating trigger. Regarding the previous example of vasodilatory shock, vasopressors such as nor-epinephrine, or vasopressin or its equivalent, and steroids in resistant cases may be the specific therapy. If Echocardiography shows underfilling of both ventricles, anticipate volume depletion shock, and if the heart is dilated, consider the diagnosis of chronic venous congestion.

Integrated monitoring of the HI could decrease adverse events from using inotropes or vasodilators or unnecessary fluids for vasodilatory shock or diuretics and inotrope in volume depletion shock as well as inotrope or extra fluids in case of cardiomyopathy with chronic venous congestion.

4. Respiratory Disorders

4.1. Neonatal Respiratory Distress Syndrome

Respiratory distress syndrome (RDS) or hyaline membrane disease is the common cause of respiratory distress in preterm neonates. caused by deficiency of pulmonary surfactant due to either inadequate surfactant production, or surfactant inactivation in the context of immature lungs. RDS can occur also in near-term and full-term infants due to genetic mechanisms disrupting surfactant metabolism resulting in diffuse lung disease in near-term and full-term infants that mimics RDS in preterm infants**.** The deficiency of surfactant increases the surface tension within the small airways and alveoli and affects the gas exchange by reducing the compliance of the immature lung causing impaired lung function. It is diagnosed clinically based on the onset of progressive respiratory insufficiency shortly after birth in a preterm neonate in conjunction with a characteristic chest radiograph [90].

RDS is a complex matter and considered a heterogeneous syndrome with numerous clinical and pathophysiologic subgroups so the need for precision medicine approaches to both diagnosis and treatment of NRDS is essential and the continuous attempts to apply the same treatment to all RDS patients are unlikely to be useful. The therapeutic agents can be employed in the specific pathophysiology and actual patient requirements that are the future of NRDS and likely to derive benefit [91]. Cortisol levels are correlated with the severity of RDS and can predict respiratory support strategies [92].

Prognostic enrichment in precision medicine focuses on patient stratification based on a combination of clinical (e.g., gestational age, ethnicity, gender) information, genetic pre-disposition, and individual biomarkers, enabling more precise differentiation of different phenotypes within this group of pre-term infants [93].

There are three unique challenges in the implementation of precision medicine approaches in neonates with RDS:

- Surfactant genetic and biological tests
- Advanced oxygenation metrics
- Functional lung imaging

Surfactant genetic and surfactant biology of RDS neonates

Pulmonary Surfactant is a complex mixture of lipids and proteins that covers the inner lining of normal alveoli needed to prevent end-expiratory atelectasis. surfactant is stored in specialized

 doi:10.20944/preprints202412.2162.v1

organelles in alveolar type 2 cells called lamellar bodies, storage particles consisting of packed surfactant phospholipids and proteins, released from type II alveolocytes before its secretion into the airspaces. The phospholipid components of surfactant mainly disaturated phosphatidylcholine (DSPC) are responsible for lower surface tension these lowering are imported into lamellar bodies by ATP binding cassette sub-family A, member 3 (ABCA-3). Sequence variants in the gene (ABCA3) disrupting or limiting the production of ABCA3 may therefore cause surfactant deficiency ABCA3 production is developmentally regulated, increasing with advancing gestation and The exact amount of functional ABCA3 needed to prevent lung disease is not known, [94].

Surfactant Proteins B (SP-B) and C (SP-C) are low-molecular-weight hydrophobic proteins They are encoded by separate genes (called SFTPB and SFTPC) located on human chromosomes 2 and 8, respectively have important roles in surfactant function and metabolism genetic mechanisms disrupting or altering the production of SP-B and SP-C might result in a phenotype of RDS [95].

A summary of the specific genes and proteins and key clinical features are summarized in **Table 1.** Genetic surfactant-related disorders.

Table 1. Genetic surfactant-related disorders.

Protein	**SP-B**	**SP-C**	**ABCA3**	**SP-A**	**SP-D**	**TTF–1**	**GM-CSF Receptor [3,4]**
Gene	*SFTPB*	*SFTPC*	*ABCA3*	*SFTPA1* *SFTPA2*	*SFTPD*	*NKX2–1*	*CSFR2A* *CSFR2B*
Pulmonary Phenotypes	RDS	ILD PF RDS	RDS PPHN ILD PF	PF Lung cancer	None yet known	RDS ILD Recurrent Infection	Alveolar Proteinosis
Inheritance	AR	AD sporadic	AR	AD sporadic	N.A.	Sporadic AD	AR
Prognosis	Rapidly fatal	Variable	~60% rapidly fatal; ~40% variable	Generally adult onset, progressive	N.A.	Variable	Childhood to adult-onset; variable
Incidence	<1 in 1,000,000	Unknown	Uncertain, 1 in 10 K to 1 in 20 K	Unknown	N.A.	Unknown	Unknown

RDS, respiratory distress syndrome; ILD, interstitial lung disease; PF, pulmonary fibrosis; PPHN, persistent pulmonary hypertension of the newborn; AR, autosomal recessive; AD, autosomal dominant [96].

Knowing the genetic mechanisms of RDS is useful for understanding the typical clinical presentations of neonatal RDS, establishing the diagnosis and performing genetic counseling to the Families of neonates with positive genetic results. Such testing is readily available through multiple laboratories with the same limitations due to cost, turn-around time, and difficulties in interpretation. There are many simple, low-cost biological available as bedside tests able to measure both the amount and the function of endogenous surfactant to predict RDS occurrence. which can be done non-invasively through testing of blood, saliva, or buccal samples [97].

1. Quantitative Tests

Lamellar body count (LBC): Lamellar bodies can be found in lung lavages, amniotic fluids, and gastric aspirates as lamellar body-like particles (LBPs). The more number of particles in those fluids,

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the more lung maturity. Consider good candidates as a point-of-care technique for use at the bedside and guide surfactant replacement therapy [98]. **Advantage:** The test is a quick and easy quantitative biological tool, readily available at the bedside, and can be done without any time-consuming sample preparation or dilution. Nevertheless, some limitations as LBC could be unfeasible for around 35% of samples due to blood contamination and the high viscosity of many samples [99], The test only determines the amount of pathophysiological at birth and may suggest the insurgence of RDS, rather than its clinical severity (i.e. need for a surfactant or invasive ventilation). There is low reliability in predicting CPAP failure as the RDS may be affected by other factors such as the degree of alveolarization, the prenatal steroid dose, and extravascular water [98]. Also, surfactant biophysical properties can be affected by the amount of surfactant protein and anionic phospholipids, the effect of reactive species of oxygen on the chemical structure of surfactant lipids and proteins, the rate of secretory phospholipase A2 and the presence of inhibitors surfactant activity substances that can be found in amniotic fluids[100].

2. Qualitative Tests

Two tests can be considered technically quick and easy enough to be employed as point-of-care methods to predict CPAP failure; the stable microbubble test (SMT) and the surfactant adsorption test (SAT) [101].

Stable Microbubble Test

SMT is an old simple and rapidly effective method to predict RDS, the sample can be obtained from gastric aspirate, amniotic fluid, and BAL depending on the fact that surfactant present in amniotic fluids or gastric aspirates, when vortexed, forms numerous small stable microbubbles of at least <15 μm which are less abundant or absent in samples from neonates with RDS [102]. limitation of this test, it is influenced by the subjectivity of inter- and intra-observing variability under a microscope, the drops of amniotic or gastric fluid may contain different amounts of surfactant, and thus the efficiency to create a stable bubble is not normalized for the available surfactant pool, lack of physiopathology studies relating SMT results to lung aeration or other surfactant function measures [102], the presence of meconium in vitro may affect the stability of surfactant microbubbles tested by SMT[103].

Surfactant Adsorption Test (SAT);

SAT is relatively quick, sensitive, and not biased by any intrinsic sample dilution, and a high-throughput fluorescent method to test indirectly both adsorption and stable accumulation of surfactant at the air-liquid interface. SAT consists of two simple steps: a first step of incubation for labeling surfactant from a biological fluid, and the subsequent step of detection of both its capability to move up towards the air-liquid interface crossing the subphase volume and the kinetics of its interfacial accumulation over time [104]. Due to its sensitivity and suitability, SAT has been widely used to test different types of materials. SAT was employed to assess in vitro surfactant activity from animal or cellular sources, therapeutic surfactant preparations, and non-bronchoscopic bronchoalveolar lavages at different temperatures of asphyxiated neonates under therapeutic hypothermia. However, it is not suitable to be used in clinical care, but successfully used for research purposes in BAL fluids obtained from animals and human neonates [105].

For future studies, up to now, no studies have been performed about SP-D levels and other surfactant proteins in different sample matrices to predict RDS in neonates. Genetic SP-D variations seem to be associated with severe RDS in very preterm birth infants[106]. Collaboration between the genetic scantiest, academics, and clinical practitioners is required to make progress toward the development of a quick test, highly reproducible, accurate, low-cost, bench-to-bedside, easy to use, and minimally invasive. Surfactant biological tests need further translational investigations and/or industrial development.

Advanced Oxygenation Metrics

Oxygenation metrics are simple and available, and solid physiopathology background tools may allow the detection of factors influencing oxygenation other than FiO2 and variously identify the

 doi:10.20944/preprints202412.2162.v1

sickest patient. Oxygenation metrics include the alveolar-arterial gradient, oxygenation index (OI), and oxygen saturation index (OSI).

The alveolar-arterial gradient (A-a gradient) measures the difference between the oxygen concentration in the alveoli and arterial system, it can help to narrow the differential diagnosis for hypoxemia [107]. The a/A ratio has been used for many years using various thresholds to indicate surfactant replacement. If the hypoxia is due to any pathology or dysfunction of the alveolar-capillary unit, it will result in a high A-a gradient and if the hypoxia is due to another reason it will result in a low A-a gradient. However, the a/A ratio is not always technically possible and ABG invasive procedures may be ethically questionable in neonates with mild respiratory failure not needing invasive procedures.

Oxygenation index (OI)

OI is considered to be a primary indicator of respiratory disease severity in mechanically ventilated patients [108]. (OI) is commonly used, as it takes the mean airway pressure into account (OI = mean airway pressure × FiO2 × 100/PaO2) and many guidelines recommend surfactant according to mean airway pressure (MAP)/positive end-expiratory pressure (PEEP) and the fraction of inspired oxygen - FiO2 needs [109]. Oxygen index required PaO2 measured by blood gas analysis on samples obtained by indwelling arterial which considered invasive procedures difficult to measure continuously.

Oxygen saturation index (OSI)

OI is modified to include a less-invasive estimation of factors influencing oxygenation by using an oxygen saturation index (OSI) replacing PaO2 with peripheral saturations. The OSI easy to diffuse in neonatal critical care correlates well with OI and A-a gradient uses oxygen saturation - SpO2 instead of PaO2, which is noninvasive and can be measured continuously. The OSI is calculated as follows: OSI = [MAP (cmH2O) × FiO2 (%)]/SpO2 (%)[110] .The influence of foetal haemoglobin on OSI is unknown so estimation of foetal haemoglobin may be included to correct saturation values and personalize the evaluation [111]. For future studies, we need more studies to detect the influence of foetal haemoglobin on this OSI and the ability of oxygenation metrics to predict CPAP failure.

Functional Lung Imaging:

Lung Ultrasound

Lung ultrasound (LUS) has been described as a useful bedside tool used for both descriptive and functional purposes and can be repeated in the event of any clinical change providing the clinician with valuable information in the absence of ionising radiation and with minimal disruption for the patient [112]. The ultrasound image described in respiratory distress syndrome (RDS) consists of interstitial syndrome up to white lung (grouped B-lines), a pathological pleural line, and an air bronchogram together with variably sized consolidations [113]. Using ultrasound to assess lung aeration is a quicker indicator of surfactant need help in detecting neonates with a greater risk of requiring surfactant or mechanical ventilation even before oxygenation criteria [114]**.** Nevertheless, LUS is not specific to loss of lung aeration due to the magnitude of primary surfactant deficiency but can be useful also in all cases of neonatal RD

Echography-guided Surfactant Therapy (ESTHER)

LUS-guided surfactant administration has been already investigated in a quality improvement project and a randomized controlled trial. LUS-guided surfactant administration improves oxygenation after surfactant dosing and reduces oxygen exposure early in life with possible secondary benefits, such as shorter invasive ventilation [115].

Functional Lung Imaging: Electrical impedance tomography (EIT)

EIT is a validated research tool used for dynamically imaging regional ventilation and lung function that depends on individualizing respiratory therapies in RDS. EIT measures the differential properties of air and liquids in tissues via a small non-adhesive electrode belt placed around the infants' chest, the electrical activity is recorded to create an image representing the amounts of ventilation, aeration, gas flow, or perfusion within the area of the chest imaged [116]. EIT also describes

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the effect of surfactants during and after the replacement therapy and describes the effect of surfactants on respiratory support strategies [117]. in neonates with RDS EIT can demonstrate differences in end-expiratory lung volume at different CPAP levels [118].

EIT is an interesting tool but less practical when applied in the clinical setting, and it can't investigate the prediction of CPAP failure. For future studies, there is a need for more studies looking for early ultrasound patterns in the development of bronchopulmonary dysplasia, EIT also needs further industrial development and specific diagnostic accuracy studies. Neonatologists should receive training in lung ultrasound as it is accessible and easy to learn even with relatively little experience in this technique as every NICU has an ultrasound device.

4.2. Bronchopulmonary Dysplasia

Bronchopulmonary disease (BPD) is a form of chronic lung disease that is considered the most common complication of pre-term birth with worse long-term outcomes on cardiorespiratory and neuro-development with increased risk of cerebral palsy and developmental delay [119]. This long-lasting sequel requires significant use of resources and funding, so it is essential to identify infants at high risk of developing BPD to prevent disease progression and target them for early intervention to prevent BPD and reduce this healthcare burden because there is no magic cure for BPD, Once BPD has developed, it can only be managed to reduce the severity of BPD and reduce its complications [120].BPD is considered a multifactorial disease with complex interactions of genetic predisposition and environmental exposures. the variation in clinical and biological diversity facilitates the interventions to prevent the development of BPD by using individualizing lung-protective processes. If it were possible to predict those infants who would go on to develop BPD, a targeted and more tailored and personalized approach for treatments would be useful to reach optimizing outcomes of care [121]. In this review, we will discuss novel four non-invasive tools which potential developments in pulmonary research that allow a more individualized optimization to prevent the development of BPD.

1. Pharmacogenetics and Caffeine
2. Electrical Impedance Tomography
3. Electromyography of The Diaphragm
4. Volatile Organic Compounds

Pharmacogenetics and caffeine

Caffeine treatment for pre-term infants is a standard of care treatment for reduced risk of BPD with improved neurodevelopmental outcomes, but there are uncertainties regarding the mechanism of action of caffeine and it is optimal dose [122]. Precision medicine might help to individualize the dosing of caffeine based on the genomic profile of preterm infants. Individual genomic variants and metabolomics heterogeneity are potential indicators for caffeine treatment effectiveness as well as the risk of developing complications. Cytochrome P450 enzymes and adenosine receptors are known genetic associations with caffeine, and cytochrome P4501A2 (CYP1A2) enzyme activity is markedly reduced in pre-term infants, leading to limited Caffeine metabolism [123]. to determine a personalized caffeine dose Future research is needed to evaluate a genomic profile at birth and genomic variation in caffeine metabolism.

Electrical Impedance Tomography

Electrical Impedance Tomography (EIT) is a non-invasive technique, radiation-free, that can visualize regional lung volume and ventilation changes at the bedside and individualize pulmonary treatment. EIT technique uses differences in tissue conductance in response to an electrical current which shows a high correlation to actual intra-thoracic changes in air content [116]). EIT can detect and monitor changes in lung aeration caused by pneumothoraces, atelectasis, incorrect endotracheal tube placement, endotracheal suctioning, (minimal) invasive surfactant administration, and lung recruitment procedures during conventional and high-frequency ventilation in pre-term infants [124]. EIT can assist the clinician in the challenge to optimize ventilator support at an individual level,

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thereby achieving the goal of homogeneous non-injurious ventilation. The importance of EIT in clinical outcomes needs to be investigated in future studies. Development of well-designed EIT hardware and software and easily applicable equipment for neonates is needed for clinical implementation.

Electromyography of The Diaphragm

Changes in airway pressure or flow are used for synchronization of invasive mechanical ventilation but these parameters are not always accurate in the presence of a leak [125].

Transcutaneous dEMG can detect and measure the activity of the diaphragm and provides objective information on the patient's breathing effort breath detection with dEMG is feasible and accurate helping in individualizing the application of respiratory support, titrate, and trigger the mode and level of respiratory support in pre-term infants [124]. Still extensive internal and external validation before submitting to impact analyses in daily practice. In the future, more studies are needed and Neonatologists should receive training on dEMG output parameters to determine which provides the best information on the individual patients' needs.

Volatile Organic Compounds

BPD is considered a multifactorial disease as inflammation and growth failure are risk factors and mediators in its development, based on clinical characteristics or biomarkers, there is no prediction model with an accurate discrimination for early detection of BPD

In adult respiratory medicine, measuring volatile organic compounds (VOCs) is increasingly used [124]. Exhaled breath is a prognostic and predictive test in preterm infants. The collection of exhaled breath is non-invasive and volatile organic compounds (VOCs) which can be separated, qualified, and identified by chromatography-mass spectrometry (GCMS) analysis is rapid with using sensor technology. Several volatile organic compounds (VOCs) are described in exhaled breath and represent metabolic processes in the host, bacterial metabolism, and organ function potential biomarkers [126]. Promising diagnostic or prognostic tools need extensive internal and external validation before their application in practice.

5-Persistent Pulmonary Hypertension of newborn infants

Persistent pulmonary hypertension of the newborn (PPHN) is a severe clinical problem among neonates, which affects their morbidity and mortality significantly. [127]. PPHN results from the failure in the normal transition of circulation after birth and persistent elevation of pulmonary vascular resistance (PVR), which causes right-to-left shunting, inadequate blood flow to the lungs, and severe hypoxemia. The incidence of PPHN is about (2 - 6 per 1000) live births, and with mortality rate ranges from (10–20%) [128]. PPHN is multifactorial and has genetic-based susceptibility and individual risk factors. By understanding the genetic bases and risk factors associated with this condition precision medicine can help in developing targeted prevention and treatment strategies [129]. Identification of Genetic factors in the development of PPHN.

The genetic causes of persistent pulmonary hypertension are complex and involve multiple genes that regulate vascular function and response. BMPR2 Gene: This gene is crucial for regulating cell growth in the pulmonary arteries, and its mutations lead to abnormal cell proliferation, contributing to increased vascular resistance, and has been related to familial and idiopathic elevation of pulmonary arterial pressure [129]. Other genetic variants, including those in SMAD9, NOTCH3, and CPS1, are associated with an increased risk of PPHN also [127]. EDN1 Gene, which encodes endothelin-1 (a potent vasoconstrictor), has been linked to PPHN. A specific variant (rs2070699) in this gene was found to be more prevalent in PPHN patients and associated with higher levels of endothelin-1 in the blood, suggesting a role in the pathophysiology of the disease [130]. Genetic polymorphisms and variations in genes related to pulmonary vaso-reactivity and endothelial function may influence susceptibility to PPHN [128].

Identification of Risk Factors for PPHN

Many factors are associated with an increase in the risk of developing PPHN, including maternal health problems such as obesity and diabetes mellitus, high maternal BMI is linked to an increased risk of PPHN. The use of certain medications (e.g., serotonin reuptake inhibitors) during pregnancy has been associated with higher risks of PPHN. Perinatal conditions such as Cesarean section delivery

 doi:10.20944/preprints202412.2162.v1

have a significantly higher risk of developing PPHN compared to vaginal delivery, meconium aspiration syndrome and congenital diaphragmatic hernia are other risk factors. Sepsis, RDS, and any condition that induces hypoxia or acidosis are neonatal conditions associated with PPHN that can lead to pulmonary vascular remodeling and increase the risk of PPHN [131].

Deep Phenotyping

Precision medicine emphasizes the importance of deep phenotyping, which involves comprehensive profiling of patients at the molecular level. This includes analyzing genomics, transcriptomics, metabolomics, and proteomics to create a detailed picture of the patient's disease state [132]. Using biomarkers for early detection of PPHN is still ongoing for research, with promising potential, for example, N-terminal pro-B-type Natriuretic Peptide (NT-proBNP) may be increased in PPHN and have a positive correlation with the severity of pulmonary hypertension. Endothelin-1 might be elevated in PPHN and could be used as a potential biomarker for early recognition of PPHN [133]. Assessment of the metabolome of lung tissue or intracellular matrix can be easily done by sampling blood, urine, saliva, or even exhaled air. Exhaled air can be transformed into a “breath print” assessed with the use of an “electronic nose”, for non-invasive analysis of metabolic changes in PH. The metabolomics part that can be traced in breath is called “volatolome”, and it consists of volatile organic compounds (VOC) [134]. Plasma asymmetric dimethylarginine might have diagnostic and prognostic value for predicting newborn infants who develop pulmonary hypertension. who developed PH. it may have diagnostic and prognostic values [135].

Immune Profiling and Data Integration:

Recent studies have utilized machine learning to identify immune sub-phenotypes in patients with pulmonary arterial hypertension (PAH), which is closely related to PPHN. By understanding immune responses, integration of clinical data, lifestyle factors, and environmental exposures, we construct a comprehensive health profile. This holistic view can inform treatment decisions and improve patient management [132].

Advanced Monitoring Techniques:

Employing advanced monitoring techniques, such as electrical impedance tomography (EIT), can provide real-time data on lung function and help tailor respiratory support to individual needs. This technology allows for better assessment of lung aeration and ventilation, which is crucial in managing infants with PPHN [124].

Challenges and limitations for precision medicine in PPHN

Precision medicine is a promising advancement in the management of PPHN in neonates but it facing some limitations in applications like genetic and biological complexity of PPHN in its presentations and underlying mechanisms makes it difficult to identify universal genetic signatures effectively. Challenges in biomarker development as many potential biomarkers identified by researchers have not yet been validated for clinical applications [134]. Methodological limitations of the studies regarding sample size and lack of standardization in testing protocols [134]. Risk of overfitting in machine learning approaches used to identify patient subgroups. There is a risk of overfitting models to specific datasets, that may not generalize well to large populations. The need for training and education of health care providers slows the implantation of precision medicine strategies [132].

Recommended strategies for precision medicine in neonatal persistent pulmonary hypertension (PPHN) focus on the following:

Promotion of Genetic and Molecular Profiling [136]. Biomarker development, through research into biomarkers that correlate with disease severity and treatment response, can facilitate early diagnosis and personalized management [137]. Making individualized treatment plans and tailored Pharmacotherapy based on individual responses can enhance treatment efficacy. For example, while inhaled nitric oxide (iNO) is a standard treatment, some infants may not respond adequately. In such cases, alternative therapies like sildenafil or prostacyclin analogs can be considered based on the patient's specific condition and response to initial treatments [137]. Establishing multidisciplinary teams that include neonatologists, cardiologists, geneticists, and pharmacologists can enhance the

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management of PPHN. This collaboration ensures that all aspects of the infant's health are considered when developing a treatment plan [136]. Continued research into the genetic and environmental factors influencing PPHN is essential for developing more effective precision medicine strategies [124]. By implementing these strategies, healthcare providers can enhance the management of neonatal persistent pulmonary hypertension, leading to improved outcomes for affected infants.

The integration of precision medicine into the management of neonatal persistent pulmonary hypertension represents a significant advancement in neonatal care. Focusing on individual profiles such as genetic and molecular insights, healthcare providers can enhance diagnostic accuracy, tailor treatments, and ultimately improve patient outcomes.

6. Neonatal Sepsis

Neonatal sepsis is the cause of morbidity and mortality, especially in very low weight and preterm Its incidence is 3900 for every 100,000 live births with mortality incidence ranging from 10.3 to 28.6% [138]. Development of the neonatal immune system is not perfect as the phagocytosis and other bactericidal effect of innate immune cells (neutrophils and macrophages) are not functioning [139]. Understanding the heterogeneity and dynamic nature of sepsis regarding the neonatal response and three parts of the pathophysiological process of neonatal sepsis (genetic factors and coagulation and fibrinolysis systems) is considered a new pathway in early diagnosis, predicting prognosis, and adapting choice the treatment to evolve from a “one-size-fits-all” to a more personalized and tailored approach to reach optimizing the outcome of care. The gold standard for diagnosis of sepsis is blood and other fluid cultures and sensitivity tests of the organism to antibiotics [140]. Blood culture in neonates has multiple challenges, like difficulty in drawing a sufficient amount of blood for culture, particularly in preterm infants also neonates have low levels of bacteremia which requires up to 5 days to have a positive result so blood culture results are delayed. Also, blood culture may not give positive results due to infection with viruses, fungi, or anaerobes as there are no commercially available anaerobic blood culture bottles for blood volumes less than 3 mL also, the use of maternal antibiotics may lead to reduced sensitivity in blood culture [141]. The organisms that can't easily be cultured under available conditions are called “microbial dark matter”[142].

Current biomarkers used for sepsis diagnosis e.g. c-reactive protein (CRP), interleukin-6 (IL-6), and procalcitonin (PCT) have achieved partial success because most of these biomarkers are associated with inflammation and not specific for infection [143].

New diagnostic markers may be needed for early diagnosis of sepsis e.g. Apelin is proven to compensate for the limited sensitivity and slow speed of the traditional method and specific to infection [144]. Genetic variations in genes involved in bacterial-induced cellular response and those involved in the pathogenesis of sepsis can allow the development of new diagnostic tools and accurate predictors of patient outcomes.

Sepsis and immune response have a genetic component as 10% of human genes can code for mediators. Studies implicated many genes across a lot of immune and coagulation proteins including interleukins and fibrinogen [145].

Novel molecular methods for early detection of neonatal sepsis based on genetic and epigenetic factors support the implementation of precision medicine in neonatal sepsis. We focus on molecular diagnostic techniques with evidence application for clinical translation, not a theoretical foundation that involves evaluating sensitivity, specificity, and accuracy to enable early detection of neonatal sepsis.

- PCR
- microRNA (miRNA)
- T2 Magnetic Resonance (T2MR) Technology
- Bioinformatics Analysis

PCR Techniques

 doi:10.20944/preprints202412.2162.v1

PCR Techniques have potential advantages in both diagnosing and management of neonatal sepsis. These Techniques are primarily based on PCR amplification techniques for detection of 16S or 23S rRNA genes which accurately pinpoint the specific bacteria and the 18S rRNA gene of fungi. [146]. Various novel PCR techniques are available (16S rRNA PCR testing, molecular multiplex PCR, molecular culture, or sequencing the bulk DNA and/or RNA) to identify microorganisms. These novel techniques have many advantages, they can detect small quantities of a variety of pathogens (bacterial or fungal) DNA or RNA, identify species not easily cultured, and provide Rapid results within hours. However, there are limitations, these techniques present challenges and cannot replace blood cultures, such as the need for specialized laboratories and personnel, false-positive results due to contamination, high costs, and no available information on antibiotic susceptibility [147] quantitative PCRs can only identify resistance genes specifically targeted in the assay but any novel mutations responsible for antimicrobial resistance remain undetected [142]. In the future, a large number of research is still needed, training the clinician for sterile sample-collecting techniques, and developing more specialized laboratories.

MicroRNAs in neonatal sepsis

MicroRNAs (miRNAs) are short non-coding RNAs about 22 nucleotides in length, responsible for the regulation of gene expression by inhibiting the translation or transcription of target mRNAs that play a regulatory role in inflammation, immunity, apoptosis, and cell differentiation. Over 2000 circulating miRNAs have been identified in the human genome may help in early diagnostic strategies for neonatal sepsis [148], the expression of circulating miRNAs is regulated in the early stages of sepsis with a positive correlation with the disease severity and progression so blocking pro-inflammatory effects can effectively improve related organ damage caused by sepsis [149]. miRNAs have many advantages as the sample collection is faster and less invasive, more stable in human specimens, and reliable measurements of expression levels. But up to today, the research on miRNA remains limited, and not possible to conduct specific studies on the subtypes of pathogens. The currently available molecular assays have insufficient diagnostic accuracy to replace microbial cultures. But could prove valuable, particularly in conjunction with clinical judgment and routine laboratory parameters. They can be used to help in clinical decisions and to overcome some difficulties in blood culture. In the future, a large number of research is needed to study validation and explore the diagnostic effect of a single miRNA on diseases.

T2 Magnetic Resonance (T2MR) Technology

T2MR is a novel technology molecular assay that is designed and utilizes magnetic resonance technology for direct detection and identifying the presence of pathogens in the bloodstream without the need for blood culture. These nano diagnostic panels have both T2 Bacteria and T2 Candida panels demonstrated a significant advantage over blood culture in terms of time to identification while ensuring consistent sensitivity and specificity [150].

T2MR first amplifies microbial DNA by PCR, enabling detection by the resulting change in the T2 signal of the sample after probes enriched by superparamagnetic nanoparticles hybridize to the amplicon, [151]. T2MR can identify circulating pathogens (either free or white-cell encapsulated), thus avoiding false-positive results associated with freely circulating DNA [152].

There are several advantages such as rapid identification compared to blood culture, cost-effectiveness, and suitability for development as a point-of-care diagnostic test providing reliable support to confirm or exclude neonatal sepsis. Nevertheless, there are limitations as the performance of T2 Bacteria must be placed in context with blood culture to enhance the diagnostic process, with potential complementary roles by each method. Most prospective studies are small or have limited numbers of comparative blood cultures, so integration between T2MR and blood culture could help overcome current diagnostic issues, even in most extreme ages where clinical vulnerability requires a rapid and sensitive approach [153]. In the future larger-scale studies of ongoing investigation for conclusive confirmation and validation are required for their potential application in neonatal sepsis.

Bioinformatics Analysis

 doi:10.20944/preprints202412.2162.v1

Analysis of gene expression profiles of neonatal sepsis patients from public databases to develop a genetic model for predicting sepsis could provide insight into early molecular changes and biological mechanisms of neonatal early onset sepsis(EOS). Four genes, CST7, CD3G, CD247, and ANKRD22, were identified that most accurately predicted neonatal EOS and were subsequently used to construct a diagnostic model. This diagnostic model performed well in differentiating between neonatal EOS and normal infants [154]. Meanwhile, the individual and geographic variability of EOS infants may affect the performance of this model, and the small sample size limits the validation of the model. In the future multicenter randomized controlled studies are needed to evaluate this model regarding expression changes of the four genes in their peripheral blood to determine whether the four-gene signature could identify neonatal sepsis patients with negative blood cultures.

7. Renal Diseases

Precision medicine in neonatal acute kidney injury (AKI) is the approach of research aimed at optimizing the management of AKI in neonates based on genetic, molecular, and environmental factors[155].AKI is defined by rapid reduction of the GFR with build-up of products of protein metabolism, disturbance of electrolyte homeostasis and fluid balance resulting in elevated serum creatinine levels, and/or decreased urine output[156]. Management of neonatal AKI has many difficulties due to the specific physiological characteristics of neonates and their vulnerability to kidney injury from various factors such as prematurity, infections, ischemia, nephrotoxic drugs, and metabolic imbalances. Precision medicine in neonatal AKI aims to identify at-risk infants early, understand the pathophysiology of renal injury, and identify interventions to improve outcomes [155].

Here are some key aspects of diagnostic markers of AKI in neonates:

Serum creatinine(SCr) is considered the "gold standard" of biomarkers for diagnosis of AKI, but there are numerous obstacles with SCr as an indicator of AKI. Most importantly, early serum creatinine in neonates could be a reflection of serum maternal levels and normalizes within days depending on the gestational age. In addition, SCr serves as a functional marker, rather than an injury [157]. Moreover, SCr has begun to increase lately (up to 48–72 h) from the renal insult and may remain at normal level even after the loss of 25–50% of the kidney function [158]. These impediments interfere with the early detection of AKI creating more efforts to discover novel biomarkers that could accurately detect AKI, improve clinical approach, and promote satisfied outcomes. [159]

Cystatin C is a 13-kDa (kDa) cysteine proteinase inhibitor that is formed at a steady rate by all nucleated cells and continuously secreted into the blood. It is freely filtered (>99%) in the glomerulus, then reabsorbed and catabolized in the proximal tubule. It represents a novel marker of glomerular injury as it is not secreted by renal tubules. [160]. Despite being independent of muscle mass, it has many significant limitations regarding its level monitoring as it can be affected by many factors such as age, sex, hypertension, cholesterol levels, thyroid disease, and some medications (steroids) [161].

Neutrophil Gelatinase-Associated Lipocalin (NGAL) is a protein produced in renal epithelia and leukocytes in response to tubular injury and systemic inflammation. There are at least 3 different types in blood and urine. The monomeric form and to some extent the heterodimeric forms are the predominant forms produced by renal tubular epithelial cells during stressful conditions. [161] It has been widely studied and is now considered a strong predictive marker of AKI in a heterogeneous group of critical illnesses even before changes in serum creatinine levels [162]. For example, NGAL was clinically applied for the detection of hypoxic-ischemic AKI [163].In addition, children who developed post-cardiac surgery AKI have rising levels of NGAL in their urine and serum [164]. Also, it was discovered that preterm babies with AKI, those who have high urinary NGAL concentrations, were distinctly associated with fatal outcomes [165]. Finally, using a cut-off of ≥ 400 ng/mL, NGAL in urine was significantly increased in those neonates who subsequently developed severe AKI after receiving nephrotoxic medication in the NICU [166]. Nevertheless, the NGAL assay has some limitations as it is still now not approved by the FDA and there is no cutoff point nor standardized assay specific to it [166].

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Kidney Injury Molecule-1 (KIM-1) is a type I transmembrane protein that is elevated in response to tubular injury and may serve as a sensitive marker in neonatal AKI, especially proximal tubular insult [166].

TIMP-2 and IGFBP7 (Nephrocheck ®) are other structural biomarkers of renal injury are Tissue inhibitor metalloproteinase 2 (TIMP-2) and insulin-like growth factor binding protein 7 (IGFBP7) which are markers of cell cycle arrest. IGFBP7 distinctly initiates expression of p53 and p21, and TIMP-2 accelerates p27 expression. These p proteins inhibit cyclin-dependent protein kinase complexes responsible for cell cycle achievement, leading to G1 cell cycle arrest. Physiologically, if this arrest was temporal it would allow damaged DNA to be repaired and recovered; However, if cell cycle arrest persistently continues, it will cause cellular fibrosis and subsequently develop AKI early and CKD later on [167]. Therefore, those biomarkers are considered powerful indicators of preinjury which is called acute kidney stress.

Interleukins are considered potent biomarkers in predicting AKI and play a fundamental role in its pathophysiology. Due to its anti-inflammatory role, interleukin-10 allows the inhibition of the secretion of the proinflammatory cytokine, hindering the healing process after kidney injury [168]. In addition, Studies have shown that interleukin-18(IL-18) is linked to AKI, inducing acute tubular necrosis, thus rising levels of interleukin-18 could be used as a risk factor for AKI [169].

Beta2-microglobulin (B2mG) is a single-chain, low molecular weight (MW=11.8 kDA) peptide. The small structure of B2mG facilitates its rapid filtration through the renal glomeruli. However, most of the filtrated β2-microglobulin is reabsorbed and catabolized by renal proximal tubular cells. Only trace amounts of β2-microglobulin are excreted in urine. B2mG has been studied as a susceptible biomarker for AKI, being independent of muscular mass, and its rapidly rising level during renal insult granted it a merit compared to serum Cr levels [169]. The rising level of serum B2mG indicates a glomerular insult, whereas tubular disorders are detected when urinary β2-microglobulin is elevated [157].

Limitation: One of the pitfalls during the measurement of those biomarkers is that the exact time of injury to the kidneys cannot be detected. Another embarrassing factor is that some of these markers are not specific to renal tubular cells, but may be produced by other cells, especially in response to infection or inflammation, and can appear in the urine when they exceed the reabsorptive capacity of the renal tubules causing false positive renal injury diagnosis. Moreover, other laboratory abnormalities may interfere with the accuracy of their levels. For example, higher CRP, WBC, and decreased serum albumin are associated with increased levels of Cystatin C and albuminuria > 3000 mg/dL causes invalid TIMP-2*IGFBP7 test. [161]. In addition, the lack of the feasibility of testing kits, reference standards, high cost, and variability in assay techniques and results, create more obstacles for these biomarkers to be clinically applied.

Genetic Factors and Risk Stratification

Genetic factors play a significant role in neonatal AKI susceptibility, and precision medicine seeks to identify genetic variants that predispose neonates to kidney injury.

Genetic Susceptibility: Genetic factors surely have been shared in the susceptibility and severity of AKI, explaining variable AKI manifestation distinctly and different patient responses to treatment. For example, genetic polymorphisms in APOL1 or genes related to kidney development may influence how neonates respond to hypoxia or other insults [170]. Moreover, polymorphisms in inflammation-related genes may increase the vulnerability of an individual to AKI. For example, tumor necrosis factor-α (TNF-α) [171]. and Nuclear Factor Kappa Beta 1 (NFKB1) gene variants may affect the proinflammatory cytokine reaction causing more renal damage, demand for renal replacement therapy, and in-hospital mortality [172].

Genome-wide association study (GWAS) is a mapping method in the identification of genotype-phenotype association and novel disease susceptibility genes in an unbiased manner [173]. Also, It helps in detecting the ethnic variation of complex traits, among others. This method studies the entire set of DNA (the genome) of a large group of people, searching for small variations, called single nucleotide polymorphisms(SNPs). [174] Bhatraju et al studied nine variants discovered to be

 doi:10.20944/preprints202412.2162.v1

associated with AKI susceptibility and reported two variants most strongly associated with AKI mapped to the DISP1-TLR5 locus [175]. Researchers hope that future genome-wide association studies will identify additional SNPs associated with AKI.

Pharmacogenetics: Neonates are more susceptible to the nephrotoxic effects of drugs, and pharmacogenetic testing may help in predicting which infants are at risk for drug-induced AKI, such as from antibiotics (e.g., gentamicin) or anticonvulsants (e.g., valproic acid). [177].

Epigenetic Modifications: Epigenetics is the study of the inherited factors that affect gene expression causing changes to a phenotype without changing the primary nucleotide sequence [178]. Epigenetic factors such as DNA methylation histone modification and non-coding RNAs could influence the neonatal response to kidney injury and recovery. Emerging evidence suggests that epigenetic regulation shares in many renal disorders such as diabetic nephropathy, CKD, and renal cell carcinoma. For example, epigenetic alterations commonly occur in CKD, associated with genes involved in fibrosis, inflammation, and epithelial-to-mesenchymal transition [179].

Limitation: There is a gap of knowledge on the application of the genetic susceptibility of AKI that is the small sample size reduces statistical strength and the ability to determine genetic associations. In addition, the long-term prognosis of AKI remains uncertain owing to the complex interplay with non-genetic factors.

Precision medicine in the management of neonatal AKI has the potential to revolutionize care by tailoring interventions based on genetic, molecular, and clinical data. Early detection through biomarkers, personalized risk stratification, and targeted therapies are all integral components of this approach. As research advances in neonatal genomics, biomarkers, and therapy, the goal is to reduce the incidence and severity of AKI, optimize recovery, and minimize the long-term consequences for affected neonates.

Accurate and precocious diagnosis of AKI is required to detect AKI risk factors and diagnosis so, a personalized therapeutic strategy and follow-up plans can be established. Further studies are required over larger pediatric populations to assess the role of novel biomarkers, and genetic susceptibility factors in different subtypes of AKI. In addition, reference standards for renal biomarkers should be settled to achieve its clinical application. Finally, we suggest the integration of precision medicine into clinical practice to triage patients and optimize the timing and type of interventions designed to improve disease progress and patient outcomes.

8. Hyperbilirubinemia

Precision medicine is an increasingly recognized approach in the management and diagnosis of neonatal hyperbilirubinemia, a common neonatal condition affecting 60% of term and 80% of preterm infants (180), hyperbilirubinemia is either unconjugated hyperbilirubinemia or conjugated hyperbilirubinemia, it is also either physiological or pathological. Physiological hyperbilirubinemia is a transient and self-limited condition resolved during 7-10 days, however pathological hyperbilirubinemia when total serum bilirubin exceeds the 95th percentile for postnatal age in hours is a condition that needs proper diagnosis and treatment to avoid serious neurological complications [180,181] the risk for developing severe hyperbilirubinemia is multifactorial and has genetic-based susceptibility as well as intra and extra uterine exposures. [182] . Understanding these genetic types is crucial for diagnosing and managing neonatal hyperbilirubinemia effectively because it can influence treatment decisions and the need for interventions like phototherapy or exchange transfusion [182].

Precision medicine approaches in the management of neonatal hyperbilirubinemia can involve the following aspects:

1-Genomic insights: Advances in genomic technologies have enabled rapid sequencing and identification of genetic conditions [183].

2- **Genetic testing:** Identifying genetic variants associated with genetic conditions such as **UGT1A1** gene testing or enzymes such as glucose 6 phosphate dehydrogenase [182].

 doi:10.20944/preprints202412.2162.v1

3-Diagnostic algorithms: Current diagnostic pathways are evolving to incorporate genetic testing earlier in the evaluation process, especially in conditions that need rapid interventions such as biliary atresia.

4 -Clinical assessment: Assessing individual risk factors such as blood type and RH incompatibility, prematurity, dehydration, or breastfeeding practices that can increase the risk of developing significant jaundice.

5-Biomarker analysis: Analyzing specific biomarkers, such as levels of unconjugated bilirubin, reticulocyte count, and Combs test, can help predict the likelihood of severe hyperbilirubinemia and guide treatment decisions.

6- ML and data integration: The integration of ML algorithms with clinical data obtained from prenatal screening and genetic analysis with postnatal diagnostic work. This comprehensive data collection can lead to more accurate risk assessment and treatment [184].

7-Advancement in diagnostic technologies: The use of non–invasive bilirubinometry alongside visual assessment to improve the accuracy of diagnosing hyperbilirubinemia, this dual approach can help in making more informed treatment decisions [184].

8-Individualized treatment plans and emerging therapies: Tailoring interventions such as phototherapy or exchange transfusion based on a newborn's genetic predisposition, risk factors, and response to initial treatments can optimize outcomes and reduce the risk of complications [185,186).

The application of precision medicine principles in the management of neonatal hyperbilirubinemia can enhance the accuracy of risk assessment, allow for prediction, early diagnosis, and the ability to tailor a person-specific intervention plan based on specific genetic information and improve long-term outcomes.

Limitations and challenges

Despite the promise of precision medicine, and the potential to improve the diagnosis and treatment of neonatal hyperbilirubinemia, it faces several challenges such as implementation gaps as the field is still catching up [185]. ML data and electronic health records of genetic databases are still insufficient due to a lack of family surveys and screening approaches for genetic databases focused on neonates [183]. Limited evidence for targeted intervention on the efficacy and safety of specific mediations tailored to individual neonates with hyperbilirubinemia [184] limitations in resources and acceptability that decrease their application, especially in source-limited areas and limited long-term follow-up for cases beyond the neonatal period for development of hemolytic anemia, splenomegaly, or impaired liver functions to follow the course of the diseases in infants with suspected risk for genetic diseases with neonatal hyperbilirubinemia [184].

Recommended strategies for precision medicine in the field of neonatal hyperbilirubinemia are to promote monitoring and surveillance through large-scale and diverse genetic databases to enable researchers and clinicians to understand the genetic variants associated with neonatal hyperbilirubinemia and develop targeted interventions. Enhance risk assessment protocols, and develop comprehensive risk assessment tools that combine clinical clues with genetic information from family surveys and genetic counseling to form screening approaches and algorithms. Standardized guidelines that incorporate the latest evidence and best practices. Educate healthcare providers on the latest evidence-based practices that depend on individual genetic bases and environmental factors. Promote further research on the genetic and environmental factors influencing neonatal hyperbilirubinemia and promote the sharing of data among healthcare institutions [182,187].

Precision medicine principles in the management of neonatal hyperbilirubinemia can help healthcare providers enhance the accuracy of risk assessment, improve treatment effectiveness, and minimize the potential adverse effects of excessive bilirubin levels in newborns.

9. Precision drug therapy and Therapeutics Interventions in NICU

8.1. Pharmacogenomics and Pharmacogenetics

Diagnostic Need for Precision Drug Therapy

 doi:10.20944/preprints202412.2162.v1

One crucial aspect of neonatal care in the NICU is to provide meticulous therapeutic strategies and effective medication. The selection of drugs, and determination of dose, interval, route, and concentration is one of the daily hard tasks, for newborn infants with complex presentation. Newborn infants, particularly preterm and very low birth weight have immature pharmacokinetic mechanisms for drug absorption, distribution, metabolic capacity, and excretion that together with the nature of the diseases will adversely affect the final drug concentration in the blood. Subsequently, the therapy may or may not be effective. Moreover, drugs may cause adverse events from either toxic doses or under treatment [188]. Furthermore, the drug doses should be monitored and adjusted to the changes in weight and postnatal age as well as the prospective maturation of the drug metabolism and elimination[189]. The achievement of good outcomes among sick newborn infants requires recognizing the complicated relations between the medication used, patient characteristics, nature, and stage of illness. Identifying the metabolic phenotypes for individuals (combination of haplotypes) assists in the selection of drug therapy and the determination of the appropriate dose according to the metabolic rate and the potential toxicity[190,191]. To implement precision drug therapy, it is important to recognize the ontogenesis process of the drug metabolism and its associated enzymes, transporters, receptors, and hepatic and renal maturity. Some disarrays might happen during the organogenesis of the liver and kidneys, which is related to size and function, capability of the isoenzymes, glomerular filtration rate (GFR), and renal tubular transport activity as well as hemodynamics changes and blood flow. Drugs, such as ACE inhibitors, angiotensin receptor blockers, beta-lactam antibiotics, and nonsteroidal anti-inflammatory drugs, depend upon the organic anion transporters OATs. All these factors may alter the drug concentrations during the neonatal period [189,192]. Potential drug toxicity may arise due to the slow elimination of some drugs or decreased transporters expression or GFR in the neonates that influence the excretion of penicillin, furosemide, and aminoglycosides from the kidney [193]. Immaturity /prematurity impacts the drug response, the low levels of binding protein in newborns lead to higher intracranial concentrations of the drugs than in children and adults which potentiates the risk for drug overdose [189].

Another important aspect, during neonatal and infant periods is a dynamic progressive relationship between metabolic ontogeny and pharmacogenomic aspects. The age has to be considered as well as genetic variations when drug response is evaluated e.g., morphine displays age-related extraction due to age-related rises in OCT1 and UGT2B7 protein and hepatic blood flow [194]. Also, newborn infants display unique hepatic drug metabolism compared to adults, owing to variations in P450 expression. The mechanisms controlling gene expression and induction in neonates differ from those in adults.

The activity of the P450 expression affects the hepatic drug metabolism; the CYP-dependent metabolism in neonates is 50–70% of adults [195]. The CYP2C9 activity and CYP2D6 are very low at birth and increase during the first year of life [196].CYP450 has a role in the metabolism and detoxifications of exogenous xenobiotics and the disintegration of the majority of drugs. Phenytoin which is commonly used for the treatment of seizures needs to adjust the dose in neonates depending on the CYP2C9 activity [197]. Also, (CYP)2C8*3, CYP2C9*2, and CYP2C9*3 polymorphisms have lower ibuprofen clearance [198]. Similarly, the immaturity of UGT1A6 and UGT1A9 affects the glucuronidation of acetaminophen and UGT2B7 interferes with the metabolism of morphine [199]. It is not advisable to prescribe atazanavir therapy among those with the genotype of UGT1A1 $*$ 28/ $*$ 28 as it increases the risk of jaundice [200]. Studies showed that the initial proper dose of warfarin should rely on age, body surface area, and VKORC1 and CYP2C9 genotypes [201].

Precision medicine in drugs addresses and distinguishes the variations in drug responses between patients. Pharmacogenomics (PK) adopts this concept and seeks to ascertain the genetic attribution to inconsistency of the effectiveness and toxicity of the drug. Pharmacogenomics stands for how a person's genes /DNA influences his/her reaction to precise drugs. It involves pharmacology and genomics for specific drugs to plan for a safe drug with appropriate doses that are customized to the person's particular genes[191,202]. Pharmacogenomics recognized genetic variants in the drug-metabolizing genes as well as the gene mutations. Earlier PK looked at the relationship between

 doi:10.20944/preprints202412.2162.v1

ordinary genetic variation and patient reaction to the drugs, to detect the genes caused therapeutics phenotypic variances, however, contemporary it has started to distinguish mRNAs, microRNAs, and other measures that are affected by the genetic variation [203]. The current projects of precision medicine as precision medicine initiatives and the 100,000 genomes, need to incorporate more neonatal clinical syndromes to assess medication of complex neonatal diseases.

Therefore, we can propose that the intent of precision medicine in pharmacogenomics is to provide a specific drug with an exact dose to specific patients whose genes are matched and subsequently minimize the adverse drug reactions. The diagnostic approach of the PK supports the value of the drug precision medicine, and the progress in DNA sequencing and polymorphism depiction tools facilitate the recognition of variants with pertinent outcomes. The process starts with the detection of the genetic variants in metabolizing genes of the drug that have influenced good response or toxic effect. The recent development of high-data sequencing procedures encourages the investigation of the influence of rare variants on drug efficacy. However, these techniques are not routine in the current neonatal practice. Numerous neonatologists do not encourage genotyping due to the high cost, and the technique is not widely available either. Furthermore, a study by Ghaddar et al. showed that physicians have inadequate knowledge regarding PK tests [204], however more recent survey showed that the knowledge level among the studied group was fair to good with positive attitudes [205].

The clinical pharmacogenetics implementation consortium (CPIC) sorted the drugs to A, B, C, and D levels depending on the strength of evidence that the genes affect response to a specific drug. A study showed that genotype-directed treatment using 12 gene pharmacogenomics (PGx- PK) minimizes the adverse drug reaction by 30% in the adult population [206]. Around seventy-five percent of neonates had been subjected to more than one pharmacogenomics drug. There is a complicated association between neonate traits; and pharmacogenomics drugs. Drugs with pharmacogenomics indications such as aminoglycosides, opioids, and nonsteroidal anti-inflammatory drugs, constitute forty percent of the ten most medications prescribed for extremely low birth weight and preterm newborn infants [191]. However, there are several obstacles to the adequate use of pharmacogenetics in the NICU due to scares data on pharmacodynamics and pharmacokinetics and the immaturity of newborn infants. There is a shortage of transformation of justified pharmacokinetics models to adjusted dosing [191].

Therapeutic drug monitoring (TDM) is a traditional method in pharmacokinetics to check the concentration of drugs showing the different variations between individuals [207]. Therapeutic drug monitoring when done at a particular time interval is useful for modifications of the drug dose regimen to avoid drug overdose or toxicity. Preterm infants or neonates with acute kidney injury are vulnerable to medication adverse events. Some drugs such as theophylline, aminoglycosides/gentamicin, vancomycin, and anti-fungal agents such as itraconazole and voriconazole have low safety margins and narrow therapeutic index, neonates may benefit from drug monitoring to preclude ototoxicity and nephrotoxicity [191,208].

Monitoring of the concentration of drugs is ideal for those with genetic polymorphism as CYP2C19 monitoring [209]. A study showed major individual variances in voriconazole metabolism in children. Linking the TDM with *CYP2C19* gene polymorphism disclosed valuable knowledge for customized antifungal therapy in pediatric patients. TDM is also available for phenobarbital and phenytoin which are the most common antiepileptic drugs to avoid their side effect on the developing brain of preterm and newborns [209–212]. Moreover, the use of Time-division multiplexing to measure blood drug levels such as anticonvulsants or antibiotics optimizes the doses and prevents drug side effects that may occur even with normal doses in sick neonates with kidney or hepatic disorders. Pharmacogenomics variants aid in selecting the right drugs for specific patients and calculating the dose[17].

Therapeutic hypothermia (TH) is one of the important, quite recent therapies in the NICU. There are contradicting data regarding the pharmacokinetics of drugs during TH, some studies showed diminished drug clearance by glomerular filtration and declined activity of hepatic cytochrome

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enzymes [213]. Some, drugs as phenobarbital, showed an increase in the plasma concentrations and prolonged half-lives during TH[214]. Nevertheless, other studies showed no noteworthy changes[215]. Conversely, the ParmaCool study increased the phenobarbital dose to 30 instead of 20 mg/kg to achieve the curative concentrations during TH [216]. Also, modification of the doses or duration of antibiotics during TH was advised by several studies; amikacin and gentamicin showed a decline in clearance during TH, and 36-hour dose intervals were recommended Modification of Ampicillin dose to 25–50 mg/kg/day was recommended throughout TH[217].

Introducing new approaches for prescribing therapeutic doses in newborn infants was lagging behind children's. However, there are recent studies for precision dosing in neonates. These approaches depend on advances in diagnostic technology such as mass spectroscopy, pharmacogenomics in neonates, Bayesian assessment, electronic decision support means, and software that incorporates pharmacokinetics and pharmacodynamics. These tools provide pharmacokinetic/pharmacodynamic model-based methods that integrate population and physiology-based pharmacology data.

These recent advances provided appropriate modifications of drug dosages to attain the required outcome without harmful effects from merely depending on age or body weight [218]. The physiology-based pharmacokinetic (PBPK) models merge the physiological and anatomical data of the patients and the biochemical traits of drugs is useful to guide the dose exposure relationship and plan for the best therapeutic dose while population pharmacokinetics modeling (PopPK) defines the time sequence of drug experience in patients and explore causes of differences in patient exposure, it advises the initial prescribed dose, both models have supported our insight of drug metabolism in neonates[219.220.221].

The clinical implementation of these models showed promising results, the precise dose of Fluconazole and Acetaminophen is advised by using the population PK modeling [222]. Mahmood et al. used the PBPK model-based approach to predict the elimination of glucuronidated drugs in newborn infants. [223].

The PBPK was reported to successfully include the ontogeny of drug-metabolizing enzymes involved in acetaminophen metabolism in preterm infants. It was an important finding as acetaminophen is used in the NICU for pain control and ductus arteriosus closure. An inappropriate dose may increase the risk of hepatic toxicity [224].

Another model was described by Vicks et al. to prescribe morphine precision dosing, by utilization of the electronic health record and pharmacokinetic model-informed dosing advice [225]. The model can integrate real-time drug concentration information and allows prompt response for a decision on the appropriate dose. Another model for determining precision morphine dosage for the management of neonatal pain based on informed Bayesian estimation can also, adjust morphine doses in neonates and infants[218].

Tong, et al. work has shown the value of continuous improvement of using the Model-informed precision dosing (MIDP) in neonatal practice. Improvement of gentamicin MIPD in neonates has shown a reduction in the need for several blood samples for drug monitoring of gentamicin [225].

Vancomycin has high variable pharmacokinetics because of developmental changes and grades of disease severity in neonates. The vancomycin model-informed precision dosing software appears to be a promising and safe approach to improving the PKPD dose of vancomycin in newborn infants admitted to the NICU [226,227].The population pharmacokinetic model conjoined with the opportunistic sampling approach showed to be a practical approach for ganciclovir precise dose in newborns with congenital CMV infection [228].

Among the limitations of relying on Model-informed precision dosing is that it does not include the majority of drugs used in NICU. Additionally, those models for analgesics/morphine did not consider the nature of the painful stimuli, whether invasive procedures or prolonged events such as mechanical ventilation. Moreover, the painful stimuli provoke the release of various chemical substances that may affect the dose of morphine [229]. Models that consider the dosage of drugs during TH are not yet available. The NICU patients are subjected to various off-label medications in the

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NICU and are vulnerable to adverse drug events [230]. The financial cost needs to be evaluated and capacity building of the physician to use these technologies is needed to implement the different models.

As we have shown above, genome sequencing is now available for the neonatal population. Combining genomic data with pharmacogenomics data will lead to better therapeutic outcomes[231]. The involvement of neonatologists and related healthcare workers to improve pharmacogenetics knowledge and clinical application of new pharmacology methods in the field of neonatology has to start worldwide[232].

To maximize the advantage of therapeutic precision medicine in neonates more researches need to be done on pharmacogenetics to enhance better outcomes and diminish the expenditures from inadequate treatment. Therefore, future research needs to include the ontogeny of the drug-metabolizing enzymes, transporters, and receptors with associated physiologic changes in postnatal age to the Model-informed precision dosing in NICU to permit precise therapeutic doses. Research involving drug interactions and genetic variations is essential to avoid adverse events among sick neonates who are subjected to multiple drugs and several therapeutic modalities.

Finally, neonatologists have to take into consideration all the recent achievements in pharmacogenomics and pharmacokinetics whenever it is possible and indicate to use the genotyping tests and get the best from applying the available Model-informed precision dosing in NICU. The PM Initiative, which seeks to leverage advancements in genome biology, next-generation sequencing, and digital health, alongside ongoing PK/PD studies, is expected to further enhance the progress achieved through improved drug therapy. Clinical trials to validate the current approaches and to ensure the implementation of ethics and equity are needed.

8.2. Therapeutic hypothermia

Therapeutic hypothermia is the primary neuroprotective strategy applied clinically on neonates with moderate degree to the highest degree of hypoxic-ischemic encephalopathy. It involves cooling the infant's body temperature to around 33-34°C for 72 hours, which helps to reduce metabolic demand and limit brain injury [233,234]. The cooling should ideally start within the first six hours after birth to maximize its effectiveness [234]. In the context of therapeutic hypothermia in neonates, precision medicine aims to provide individualized and targeted interventions based on specific factors that can influence the effectiveness and safety of this treatment approach[235]. Precision medicine approaches in implementing therapeutic hypothermia in neonates may include patient selection and advanced and appropriate diagnostic tools. Cooling therapy is recommended for full-term or near-term infants (≥36 weeks gestational age) who have moderate-to-severe HIE. Specific criteria include; cord blood pH ≤7.0 or base deficit ≥−16, or, Acidotic ABG pH (7.01 to 7.15) with perinatal events of acute decrease of oxygen supply, and low Apgar score ≤5 at 10 minutes (236). The use of advanced diagnostic techniques such as neuroimaging cranial ultrasound(CUS), MRI, EEG, and near-infrared spectroscopy (NIRS) [237]. Moreover, biomarker analysis, can accurately assess the severity of brain injury and predict treatment response. Incorporating genetic and biomarker analyses can help identify patients more likely to respond positively to hypothermia, allowing for a more personalized approach [236]. Heart rate variability (HRV) is considered a potential biomarker for the prediction of the severity of electroencephalogram changes in neonates with hypoxic-ischemic encephalopathy during the 1st twelve hours of life, noninvasive hemodynamic monitoring of cardiac functions e.g. stroke volume and cardiac output during therapeutic hypothermia and re-worming can access the severity of HIE [238]. Studying the kinetics of circulating progenitor cells (CPCs) in infants with encephalopathy can demonstrate the process of cellular repair after injury[239]. Utilizing machine learning algorithms, and AI technologies to analyze large datasets can help predict outcomes and tailor interventions based on individual patient characteristics [236]. Monitoring and Follow-Up of neonates during TH by a multidisciplinary team for brain-oriented care for brain–oriented Care is crucial, they should follow specific protocols and supplied with all diagnostic tools and connected electronic health record (HER) system [236,240]. Implement comprehensive monitoring protocols,

 doi:10.20944/preprints202412.2162.v1

and continuous monitoring of patients during and after hypothermia treatment can help identify complications early and adjust treatment as necessary [241]. Continuous EEG monitoring is recommended to detect and treat seizures and proper follow-up of laboratory parameters during the cooling period to guide adjustments in temperature management and optimize the therapeutic effects[242]. Long-Term Outcome Tracking: Establishing systems for long-term follow-up of patients who undergo therapeutic hypothermia can provide valuable data on efficacy and safety, informing future practices[243].

Limitations and Gap of knowledge for Precision Medicine in therapeutic Hypothermia

The application of precision medicine in TH for neonatal HIE faces several challenges and gaps in knowledge, that can limit the effective implementation of tailored therapies that could improve outcomes for affected infants. There are several challenges as variability in response. There is significant variability in how infants respond to therapeutic hypothermia. Factors such as gestational age, timing of intervention, and severity of HIE can influence outcomes, but the interplay of these factors is not well characterized [244]. Furthermore, the gaps in Knowledge due to the need for reliable biomarkers that can predict which infants will benefit most from therapeutic hypothermia. Current research has not yet established definitive biomarkers to effectively guide treatment decisions [245]. The available data emphasizes the need to adjust patient selection and stratification for TH; tailoring treatment to these populations could enhance efficacy [11]. Need for more research to study the variability in response to TH, such as biomarkers and genetic profiling. Appropriate implementation of AI and ML algorithms. High-quality trials that include diverse patient populations and standardized protocols are essential for validating the efficacy of therapeutic hypothermia.

Precision medicine is a promising approach, In the context of therapeutic hypothermia in neonates, aims to provide individualized and targeted interventions based on specific factors that can influence the effectiveness and safety of this treatment approach, helping early recovery and reducing long-term disabilities.

Conclusion:

In recent years, the integration of precision medicine into neonatal care has gained momentum, supported by a growing body of research that underscores its effectiveness. The evolution of diagnostic measures is vital in managing neonatal disease, especially among neonates admitted to NICU. Traditionally, diagnosis relied on clinical examination, observations, and basic laboratory and radiological assessment. The review explored a shift towards more refined diagnostic criteria. The development of precise diagnostic criteria offers a pathway to more effective and individualized healthcare for neonates admitted to the NICU.

The importance of this topic lies not only in its medical implications but also in its potential to improve the overall quality of life for these newborn infants as timely and accurate diagnosis is essential for implementing early interventions, which can mitigate the effects of brain injury or other organ failure.

The advancements in PM, particularly in the fields of genetics, metabolomics, and machine learning, are revolutionizing the diagnosis and treatment of neonatal complex illnesses. For example, this evolution can distinguish between HIE and non-HIE causes of neonatal encephalopathy as well as causes of seizures and respiratory distress syndrome, hence determining the appropriate course of treatment. With the utilization of sophisticated algorithms and biological data, clinicians can now make more informed decisions, leading to enhanced individualized healthcare. For instance, machine learning models can analyze vast amounts of clinical data to identify patterns that may be imperceptible to human observers and predict outcomes, in this review we discussed some models that use MRI, aEEG, and tissue oxygenation index for early precise diagnosis of NE, seizure, IVH, and implementation of TH. The metabolomics can provide insights into the biochemical changes occurring in the infant's body, further guiding treatment decisions. By analyzing metabolites in blood or urine samples, clinicians can gain valuable information that aids in the diagnosis of HIE, sepsis, RDS, and BPD. The genetic analysis revealed several significant associations between specific gene variants and

 doi:10.20944/preprints202412.2162.v1

the risk of developing HIE, BPD, ROP, NEC, RDS, and AKI. There are differences in genomic sequencing between healthy and sick neonates. The polygenic risk score can assess the risk for each patient. Moreover, the ethnic genome expression profile of neonates with HIE affects their response to TH.

The currently available molecular assays in the diagnosis of neonatal sepsis have insufficient diagnostic accuracy to replace microbial cultures But could prove valuable diagnosis, particularly in conjunction with clinical judgment and routine laboratory parameters.

The implementation of noninvasive tools such as point-of-care ultrasound, echocardiography, and NIRS in the NICU embodies how personalized treatment strategies can be facilitated by advanced monitoring technologies. Precise prediction of hsPDA and identification of shock phenotypes help to provide and monitor individualized therapy.

The review concludes by emphasizing the transformative potential of the new advancement tools in improving outcomes for newborn infants affected by complex illnesses such as neonatal encephalopathy and disturbed hemodynamics due to PDA or shock, respiratory distress syndrome, kidney injury, or hyperbilirubinemia, PPHN, and sepsis. The review also stresses the diagnosis of aspects affecting response to therapy as pharmacokinetics and pharmacogenetics. Also suggests avenues for future directions /research in the topics of neonatal precision medicine.

Future direction

We must remain committed to refining diagnostic criteria, and diagnostic technology that can be implemented in daily use in the NICU with high predictive accuracy and timeliness for early diagnosis on an individual basis. These technologies have to be affordable, friendly used with a reasonable cost, and ensure ethical and equity for all infants to improve the short and long-term outcomes. So neonatologists will be able to personalize therapy and enhance future neonatal care.

Future research must enhance the diagnostic system and monitoring of newborn infants with complex and critical illnesses, incorporating clinical, and biological markers, genetic profiles, metabolomics studies, and imaging tools such as NIRS, MRI, EEG, and echocardiography using AI resources as machine learning algorism models that can analyze complex datasets. For example, large-scale studies that evaluate the effectiveness of machine learning algorithms in diverse clinical settings can help validate their utility and enhance their integration into routine practice. Additionally, ongoing exploration of metabolomics biomarkers may unveil new targets for intervention, ultimately leading to more effective treatment strategies. Development of well-designed hardware and software machine which is easily applicable equipment for neonates and Neonatologists should receive training on these techniques is needed for clinical implementation

Author Contributions: Author Safaa ELMeneza contributed to collecting data, editing, analyzing, and writing topics of brain injury, hemodynamics, precision drug therapy, and therapeutics interventions in NICU/Pharmacogenomics and Pharmacogenetics. Author Naglaa Agebaa contributed to collecting data, editing, analyzing, and writing the topic of persistent pulmonary hypertension in neonates, neonatal hyperbilirubinemia, and therapeutic hypothermia. Author Rasha Fawaz contributed to collecting data, editing, analyzing, and writing topics on respiratory distress of neonates, bronchopulmonary dysplasia, and neonatal sepsis. Author Salwa collected data, edited, analyzed, and wrote about acute kidney injury. All authors have reviewed the final version of the manuscript.

Funding: This research received no funding.

Conflicts of Interest: Authors declare no conflict of interest.

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