Published in final edited form as:

Int J Dev Neurosci. 2014 August ; 0: 25–31. doi:10.1016/j.ijdevneu.2014.04.002.

Perinatal biomarkers in prematurity: Early identification of neurologic injury

Maria Andrikopoulou, MD¹, Ahmad Almalki, MD¹, Azadeh Farzin, MD², Christina N. Cordeiro, MD¹, Michael V. Johnston, MD³, and Irina Burd, MD, PhD^{1,3,4,†}

¹Integrated Research Center for Fetal Medicine, Department of Gynecology and Obstetrics, Johns Hopkins University School of Medicine, Baltimore, MD

²Division of Neonatology, Department of Pediatrics, Johns Hopkins University School of Medicine, Baltimore, MD

³Department of Neuroscience, Kennedy Krieger Institute, Baltimore, MD

⁴Department of Neurology, Johns Hopkins University School of Medicine, Baltimore, MD

Abstract

Over the past few decades, biomarkers have become increasingly utilized as non-invasive tools in the early diagnosis and management of various clinical conditions. In perinatal medicine, the improved survival of extremely premature infants who are at high risk for adverse neurologic outcomes has increased the demand for the discovery of biomarkers in detecting and predicting the prognosis of infants with neonatal brain injury. By enabling the clinician to recognize potential brain damage early, biomarkers could allow clinicians to intervene at the early stages of disease, and to monitor the efficacy of those interventions. This review will first examine the potential perinatal biomarkers for neurologic complications of prematurity, specifically, intraventricular hemorrhage (IVH), periventricular leukomalacia (PVL) and posthemorrhagic hydrocephalus (PHH). It will also evaluate knowledge gained from animal models regarding the pathogenesis of perinatal brain injury in prematurity.

Keywords

biomarkers; intraventricular hemorrhage; periventricular leukomalacia; brain injury; prematurity

Preterm birth and neurologic sequelae

The rate of preterm birth rate (<37 weeks) in the United States is one of the highest in the developed world, with a staggering incidence of 11.7%, with greater than 500,000 premature

© 2014 ISDN. Published by Elsevier Ltd. All rights reserved.

[†]Corresponding Author: Irina Burd, M.D., Ph.D., Integrated Research Center for Fetal Medicine, Department of Gynecology and Obstetrics, Johns Hopkins University Hospital, Phipps 228, 600 North Wolfe Street, Baltimore, MD 21287 iburd@jhmi.edu.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

infants born each year (Hamilton et al, 2013). Although improved neonatal intensive care and technological advances have allowed for increased survival of extremely premature infants, preterm birth accounts for over 75% of perinatal mortality and greater than 50% of perinatal and long-term morbidity (Berghella, 2010).

The most common forms of central nervous system (CNS) injury in preterm infants are intraventricular hemorrhage (IVH) and periventricular leukomalacia (PVL). In extremely low birth weight infants weighing 500–999g, IVH occurs in about 45% of neonates (Wilson-Costello et al, 2005), while PVL occurs in 3–4% of infants who weigh less than 1,500 g and 4–10% of those born prior to 33 weeks of gestation (Rezaie & Dean, 2002). PVL it is considered the major cause of cerebral palsy (Bass, 2011). Other long-term medical disabilities associated with preterm birth include cognitive dysfunction, blindness and impaired vision, hearing loss, and disorders of psychological development, behavior and emotion (Sutton & Darmstadt, 2013).

The high incidence of neurological injuries among preterm infants highlights the need for the discovery of biomarkers for the early detection of preterm infants at increased risk for poor neurologic outcomes, in order to allow for the implementation of early neuroprotective and postnatal treatment interventions.

Biomarkers have gained scientific and clinical value in the practice of medicine. In the past few decades, advances in genomics, proteomics, and molecular pathology have generated many candidate biomarkers with potential clinical utility in every field of medicine (Bang et al, 2007, Keller et al, 2008, Gagnon et al, 2009). In perinatal medicine, the pathophysiology of preterm labor is poorly understood. This has fueled increased interest in the identification of biomarkers that can predict preterm birth, as these may allow for the identification of high-risk populations as candidates for further intervention. Such discoveries could also help to define the mechanisms leading to preterm birth. However, current knowledge on pathophysiology of preterm labor and its associated biomarkers have not yet translated into a reduction in preterm birth rates. Additionally, the use of such biomarkers in clinical practice to predict adverse outcomes such brain injury remains challenging. Interestingly, Leitner et al (2014) have recently reported that preterm birth and fetal cortical injury may occur by divergent mechanisms. This stresses the importance of identifying biomarkers targeting the prediction of adverse outcomes such as IVH, PVL and PHH, rather than those associated preterm labor alone.

Some investigators have focused on the detection of proteins in the serum that should only be present in the CNS as possible biomarkers for neuronal injury in prematurity (Gazzolo et al, 1999, Gazzolo et al, 2001). Others consider the detection of serological markers such as pro-inflammatory cytokines and enzymes in maternal and neonatal blood or cord blood as a promising tool for early diagnosis of brain damage (Heep et al, 2003, Kassal et al, 2004, Poralla et al, 2012). Additionally, different groups have used other methods so as to early predict neurologic injury, such as Apgar scores, imaging modalities or EEG abnormalities of neonates as early biomarkers (Ment et al, 2009, Woodward et al, 2009, Forsblad et al, 2007, Watanabe et al, 1999). In our review, we mainly focus on serological biomarkers for early prediction of IVH PVL and PHH as complications of prematurity.

Intraventricular hemorrhage

Intraventricular hemorrhage is a major complication of prematurity which is associated with long term adverse neurological outcomes. The site of origin of bleeding is generally the subependymal germinal matrix, which is located between the caudate nucleus and the thalamus at the level of the foramen of Monro (Volpe, 2001). IVH occurs most frequently in infants born before 32 weeks gestation or who weigh less than 1500g at birth, and its incidence increases with decreasing gestational age. In extremely premature infants weighing 500–999g, IVH occurs in about 45% of neonates (Wilson-Costello et al, 2005). The etiology is multifactorial and is primarily attributed to the intrinsic vulnerability of the germinal matrix capillaries to hypoxic injury and also to impaired cerebral autoregulation. In terms of clinical presentation, intraventricular hemorrhage can be asymptomatic in up to 50% of the cases. However, acute IVH can present with altered level of consciousness, bulging fontanel and neurologic deficits (Bassan, 2009).

IVH has been modeled using several animal species to study the causative factors and evolution of brain damage (Balasubramaniam & Del Bigio, 2006). Understanding the pathogenesis of subsequent brain injury is of utmost importance if IVH is to be prevented or treated. Xue et al (2003) developed a rodent model of IVH by injecting mice with autologous blood in the periventricular region and studied molecular and cellular processes involved in brain injury, as well as long term neurologic abnormalities and outcomes in a similar rat model (Balasubramaniam et al, 2006). Additionally, the same group showed that mouse brain injury was aggravated after injection of LPS, underlying the role of inflammation at the pathogenesis of brain injury (Xue et al, 2005). Differently, McCarty et al (2002) utilized mutant alpha V integrin mice which developed IVH to investigate the role of adhesion of endothelial cells in pathogenesis of the brain injury.

In rat models, post hemorrhagic hydrocephalus has been induced by the injection of blood into lateral ventricles in 7-day-old rats (Cherian et al, 2003, Cherian et al, 2004). On the other hand, Alles et al (2010) have used a different model of uni- or bilateral infusion of collagenase into the neonatal periventricular region of 6-day-old rats to recapitulate some aspects of human IVH. Similarly, Letik et al, (2012) have developed an instructive animal model of the neurologic consequences of IVH using stereotaxic injection of collagenase into the ganglionic eminence of newborn rats. These rats exhibited hematoma extension into the ventricles and developed both early and delayed neurobehavioral deficits.

Recently, there has been an emerging interest on the use of biomarkers to predict and early diagnose IVH in preterm neonates even before the onset of clinical presentation and permanent neurologic disability. Most biomarkers studied in prematurity related to IVH since 1998 will be reviewed.

Activin is a growth factor that belongs to the transforming growth factor-beta superfamily. It regulates a variety of biologic processes and its receptors and binding proteins are widely distributed throughout the brain (Luisi et al, 2001). Different studies that have employed models of acute brain injury have shown that activin A plays a very important role in the physiologic response to acute brain injury (Tretter et al, 1996, Lai et al, 1996, Zhang et al,

2003). Florio et al (2006) demonstrated that activin A can be used as a useful biomarker for the early identification of infants with hypoxic-ischemic brain insults who are at high risk for IVH. In the cohort of 53 preterm neonates less than 32 weeks of gestational age, the 11 who developed IVH were noted to have higher activin levels in arterial blood samples at birth than those who did not develop IVH. Similarly, urine activin levels have been found to be higher in preterm infants with IVH as compared with controls without neurologic injury (Sania et al, 2013). Thus, activin A constitutes a promising tool for identifying preterm infants at risk for IVH and larger prospective studies are needed to further evaluate the use of this biomarker as a predictive tool.

S100b, a member of the S100 protein family, is another biomarker well-studied in brain injury. It is synthesized by astrocytes and exhibits neurotrophic or neurotoxic activity. Gazzolo et al (1999) investigated the role of 100b in evaluating perinatal brain distress ultimately leading to IVH in preterm infants. They reported higher concentrations of S100 in blood samples of the 24 preterm infants who developed IVH in the first 24 hours. Additionally, a case control study on urine samples of S100b demonstrated higher concentrations of S100 in urine sampled from preterm infants with IVH (Gazzolo et al, 2001). In a final study, this biomarker was reported to be predictive of neonatal mortality (Gazzolo et al, 2005).

Uric acid (UA) has also been studied as a potential biomarker in brain injury. It is known that the purine metabolite hypoxanthine accumulates in areas of hypoxic/ischemic injury, and as reperfusion occurs, hypoxanthine is catabolized by the enzyme xanthine oxidase to uric acid resulting in the generation of oxygen-free radicals (Wilcox, 1996, Many et al, 1996). Weir et al (2003) demonstrated that high serum urate levels predict poor outcome and higher rates of vascular event among patients with stroke. UA has also been studied as a biomarker for brain injury in prematurity. Perlman et al (1998) reported increased UA concentrations on the first postnatal day of life in preterm infants who developed IVH, although Sysyn et al (2003) failed to demonstrate a statistically significant correlation between UA and IVH in preterm infants.

Many studies have investigated the role of proinflammatory cytokines such as IL-6 as an early biomarker of adverse neurologic outcome in premature infants. In a study conducted by Heep et al (2003), 88 extremely preterm infants (less than 28 weeks gestational age) were grouped according to the maximum level of IL-6 within 12 hours postpartum. These authors reported that serum levels of IL-6 independently predicted development of IVH, and concluded that IL-6 could potentially serve as a biomarker for severe brain injury. These findings are in agreement with the conclusions of Kassal et al, (2004) who measured IL-6 in the cord blood of 69 very low birth weight (VLBW) infants. Kassal et al reported elevated levels of IL-6 among infants with IVH. Furthermore, Sorokin et al (2010), in a large prospective study of 475 women at increased risk of preterm labor, reported elevated maternal levels of IL-6 and C-reactive protein in cases associated with neonatal IVH. In more recent studies, Poralla et al, (2012) demonstrated an association between elevated IL-6 levels and extremely preterm infants with IVH, whereas a prospective study did not demonstrate a statistically significant difference in cord blood IL-6 between preterm neonates who developed and those who did not develop IVH (Bhandari et al, 2011).

Additional biomarkers have also been evaluated by different study groups. Bhadari et al (2011) investigated erythropoietin (EPO), a glycoprotein hormone that controls erythropoiesis, as a potential marker for brain injury. They reported elevation of EPO levels in the cord blood of newborns with increased risk for IVH. A prospective cohort study of 163 infants born before 32 weeks of gestation analyzed 107 cord blood immunoproteins and 12 cytokines from the peripheral blood. CCL18, a member of the CC-chemokine family, emerged as the only cytokine associated with IVH, and was reported as an independent risk factor of this entity (Kallankari et al, 2004). Adrenomedullin, a vasoactive peptide that participates in cerebral blood flow regulation, has also been measured in higher concentration among neonates with IVH. Interestingly, levels of this peptide correlated positively with the severity (grade) of IVH. Finally, studies on total homocysteine (Sturtz et al, 2007) and 2-methoxyestradiol (2ME2), a potent antiangiogenic molecule (Barnes et al, 2010), have failed to demonstrate associations between these biomarkers and the presence of IVH and preterm neonates.

Posthemorrhagic hydrocephalus

One of the most feared complications of IVH is PHH, which is the progressive enlargement of ventricular system due to the blockage of channels for reabsorption of CSF by blood clots. PPH has direct relation with severity of IVH and about ¼ of infants with grade II-IV IVH will develop this complication (Murphy et al, 2002) which progressively leads to periventricular white matter injury. There are currently no biomarkers in clinical use to early identify the group of infants with IVH which are at increased risk for PHH or predict neurodevelopmental outcomes (Merhar, 2010). This article will review the literature on biomarkers of PHH, which might throw light on the illustration of possible pathophysiologic mechanisms of this complication and the subsequent white matter injury.

In clinical studies, one of the most studied biomarkers for PHH is transforming growth factor β (TGF- β). It is a cytokine, released into CSF after IVH, which upregulates the production by fibroblasts of extracellular matrix proteins. These include fibronectin and laminin which can convert a potentially reversible CSF obstruction into a permanent CSF obstruction (Whitelaw & Aquilina, 2012). Whitelaw et al (1999) explored the role of TGF- β 1 in the development of PHH and demonstrated higher CSF levels of this potential biomarker in preterm infants with PHH compared to preterm infants without neurologic injury. However, Heep et al (2004) found that TGF- β 1 CSF concentrations did not differ in infants with PHH from control infants but they did demonstrate a correlation between persisting high TGF- β 1 values and the occurrence of white matter injury. Lipina et al (2010) explored the role of TGF- β 1 in the decision to perform endoscopic third ventriculostomy surgery in infants with PHH. It was shown that the levels of TGF- β 1 were higher in the group of infants with PHH that the surgical procedure failed and needed a shunt, thus they concluded that TGF- β 1 can be a useful biomarker to predict the group of patients that would not benefit from this intervention. A pilot prospective study of Chow et al (2005) demonstrated correlation of CSF TGF- β 2 with PHH and showed that the levels tend to be elevated in patients with poor neurologic outcome. However larger prospective studies are needed to validate the prognostic use of this biomarker.

Whitelaw et al (2001) investigated the role of median neurofilament (NFL), glial fibrillary acidic protein (GFAP) and S100 concentrations in infants with PHH. NFL and GFAP concentrations in the CSF of 18 infants with posthaemorrhagic ventricular dilatation were 20-200 times higher than control values. S-100 protein in cerebrospinal fluid was four times higher than control values. These can be potentially very useful biomarkers but need to be evaluated in a larger cohort of patients. GFAP has been investigated as a potential biomarker also for PVL and has speculated to be a promising biomarker for perinatal brain injury (Blennow et al, 1996, Stewart et al, 2013).

Other studied biomarkers that have been found in increased concentrations in patients with PHH are the matrix metallo-proteinases (MMPs), Non-protein-bound iron (NPBI), plasminogen activator inhibitor 1 (PAI-1) as well as proinflammatory cytokines, underlying the pivotal role of inflammation in this entity (Savam et al, 2001, Okamoto et al, 2008, Hansen 1997, Savman et al, 2002, Schmitz et al, 2007).

Periventricular leukomalacia

Periventricular leukomalacia (PVL) refers to the focal necrosis and gliosis of the periventricular white matter (Volpe, 2001), and it represents the predominant form of brain white matter injury affecting premature infants. PVL is more common among premature than term infants. It occurs more frequently with decreasing gestational age and birth weight, with infants born at less than 32 weeks gestational age at the highest risk for developing PVL (Graham et al, 2002). PVL is the most common cause of cerebral palsy in preterm infants (Bass, 2011). The pathophysiology of this entity is multifactorial and the principal factors involved in the development of PVL are ischemia-induced injury to oligodendrocytes in the periventricular area of the developing brain and infection (O'Shea et al, 1998, Holcroft et al, 2003, Graham et al, 2004). Since the diagnosis of PVL is limited to ultrasound and MRI imaging at later stages of disease, there is emerging interest in the discovery of new biomarkers which will not only help the clinician to identify infants at risk for the disease, but will also shed light onto the pathogenesis and pathophysiology of PVL.

Several animal models have been established to provide a comprehensive understanding of periventricular leukomalacia in prematurity. Two methods are widely used to reproduce selective periventricular white matter lesions in immature experimental animals: the induction of central nervous system hypoxia/ischemia, by combining unilateral or bilateral carotid occlusion and conditions of hypoxia, and the activation of the innate immune system by administration of bacterial lipopolysaccharide (LPS).

Many rodent models have employed hypoxic/ischemic insult to mimic white matter injury in the setting of prematurity. Some studies have used bilateral or unilateral carotid artery occlusion, (Uehara et al, 1999, Cai et al, 2001, Falahati et al, 2013), whereas other groups have combined carotid ligation with a hypoxic environment to induce white matter injury in premature animals (Jelinski et al, 1999, Back et al, 2002). Interestingly, Baud et al (2004) used prolonged prenatal hypoxia to develop a model of PVL. Pregnant rats were placed in a hypoxic chamber (10% O₂) from gestational week 5 to 19. It was demonstrated that prolonged prenatal hypoxia in rats resulted in white matter damage through local

inflammatory responses in addition to the oxidative stress associated with re-oxygenation during the perinatal period.

Animal models of inflammation have also been developed to recapitulate white matter injury in PVL. Guo et al (2002) created a PVL model in rats using intraperitoneal LPS injection at day 15 of gestational age to explore the role of miRNA in the pathogenesis of PVL. Similarly, Tuzun et al (2012) also used an LPS-induced inflammatory model of PVL in rats to study the protective role of the systemic administration of neutrofin in prevention of neuronal apoptosis and hypomyelination, especially when given prenatally.

Other studies have combined hypoperfusion with LPS-induced inflammation to establish a clinically relevant PVL model. They speculated that this combination of insults would better represent the multifactorial etiology of PVL. Eklinds et al (2001) used LPS injection followed by hypoperfusion on the seventh day of life in rats, highlighting that infection can sensitize the immune system for hypoxic/ischemic insults in the brain. Additionally, another model designed by Shen et al (2010) involved unilateral arterial ligation of mice at day 6 of life with exposure to hypoxia (6% O₂) along with intraperitoneal injection of LPS to induce brain injury in mice. It has been suggested that these models which employ both hypoxia/ischemia along with LPS-induced inflammation may be the best choices to induce white matter injury and thereby study PVL (Choi et al, 2011).

In clinical studies, many potential biomarkers have been studied to predict PVL and to investigate the underlying pathophysiology. This paper reviewed all biomarkers identified since 1998. Nucleated red blood cells (NRBCs) have been well studied in prematurity-related brain injury. Although nucleated red blood cells (NRBCs) are rarely found circulating in older children, they are commonly seen in the blood of newborns. Common causes of elevated RBC levels include prematurity, chronic hypoxia, anemia, maternal diabetes and acute stress (Hermansen, 2011). Many studies advocate that the detection of NRBCs at birth reflects a response of the infant to perinatal hypoxia and is a reliable index of perinatal brain damage (Phelan et al 1998, Buonocore et al, 1999) which can be used for the assessment of the severity and the early outcomes of perinatal asphyxia (Goel et al, 2013). This biomarker has also been evaluated for its use in the early prediction of PVL in preterm neonates. Silva et al (2006), in a case control study comparing 176 preterm neonates less than 32 weeks of gestational age who developed cystic PVL or ventriculomegaly to controls without brain injury, reported higher levels of NRBCs among preterm neonates with cerebral white matter injury. However, both chronic and acute hypoxia can result in increased levels of NRBCs, indicating that this marker is not helpful in identifying the time of the hypoxic insult (Perlman, 2006). Additionally, Marom et al (2012), in a retrospective pilot case control study, did not demonstrate a predictive value of absolute NRBC counts in predicting the development of PVL. Thus, the data for this potential biomarker is conflicting and further studies are needed to evaluate its role in predicting brain injury in prematurity.

The glial fibrillary acidic protein (GFAP) is a brain-specific cytoskeletal intermediate filament protein that is found in the astroglia of the central nervous system and is a specific marker of differentiated astrocytes. Studies have explored the use of this marker for the early diagnosis of patients with stroke (Foerch et al, 2006) and in predicting mortality after

traumatic brain injury (Lumpkins et al, 2008, Vos et al, 2010). Bembea et al (2011) demonstrated that high levels GFAP during extracorporeal membrane oxygenation are significantly associated with acute brain injury and death, and GFAP could potentially be used as a biomarker in monitoring neurologic outcomes in those patients. Stewart et al (2013) explored the use of GFAP in white matter brain injury in prematurity and reported that cord blood GFAP levels did not differ significantly among preterm neonates with periventricular white matter injury as compared to those without, but they were significantly higher in neonatal blood for 4 consecutive days as compared to control group. Thus, the possibility to predict PVL with a blood test for GFAP shortly after birth is promising for the rapid identification of infants with PVL, potentially allowing for early intervention. However, further prospective studies are needed to evaluate the use of this biomarker in clinical practice.

It has been speculated that ischemia and systemic infection are implicated in the etiology of cerebral white matter injury (Leviton et al, 2005, Khwaja & Volpe, 2008). This has stimulated interest in evaluating the role of inflammatory cytokines in preterm infants with PVL. Ellison et al, (2005) in a cohort of 146 infants, investigated the cerebrospinal fluid (CSF) concentrations of IL-6, IL-8, IL-10, tumor necrosis factor alpha (TNF- α), and interferon gamma (IFN- γ) to magnetic resonance-defined white matter injury in preterm infants. They reported that preterm infants with MRI-defined cerebral white matter injury had higher levels of CSF IL-6, IL-10, and TNF- α than infants without such injury, suggesting that there exists an altered inflammatory balance in infants with adverse neurologic outcomes. Similarly, Bass et al (2008) studied the role of proinflammatory cytokines (TNF- α , IL-1 β , IL-6) in PVL, but also extended their investigation to the interaction of proinflammatory cytokines with their soluble receptors (sTNF-RI, sTNF-RII, sIL-1RA, and sIL-6R). In contrast to the previous study conducted by Ellison et al, no association was observed between individual cytokine or receptor concentrations and the development of white matter injury. However, modeling cytokines with their soluble receptors significantly improved the predictability of white matter injury in preterm infants.

Further research directions

Several potential biomarkers have emerged from the extensive research on neuronal injury not only related to prematurity but also in every field of brain injury in children and adult population. Some investigators exploring the predictive value of biomarkers in brain trauma have worked on similar biomarkers as those studied in IVH and PVL or have expanded their research on new promising biomarkers (Papa et al 2013, Berger et al, 2006, Goyal et al, 2013, Berger et al, 2012). Additionally there is emerging interest on alteration of biomarkers across different neurologic diseases in children (Shahim, 2013) and a wide range of conditions specifically for pediatric neurocritical care including cardiopulmonary arrest, septic shock, extracorporeal membrane oxygenation, hydrocephalus, and cardiac surgery (Kochanek et al, 2013). Research in the future, in the light of expanding the use of these biomarkers to the field of prematurity, might shed light in early prediction of preterm infants with poor neurologic outcome.

Conclusion

There is a growing body of evidence indicating that biomarkers will constitute a useful tool for the investigation of CNS physiology and will elucidate possible mechanisms of brain injury in prematurity. Although there exists a plethora of studies on possible biomarkers for the early prediction of brain injury in preterm neonates (summarized in Table 1), currently no reliable marker or combination of markers in the amniotic fluid, cord blood, neonatal blood or maternal serum have been exist for clinical use. In the future, further studies on biomarkers may provide information for the early diagnosis of neonates with adverse neurologic outcomes and provide a benchmark for the qualification of new therapies to improve neurodevelopmental outcomes.

Acknowledgments

This work was supported by Aramco Services Company Fellowship Fund (W.F., J.M.R.), NICHDK08HD073315 (I.B.), NINDS28208 (M.J.), NIHRO1EB003543 (S.M.), and a grant support from Brain Science Institute, Johns Hopkins (SM).

References

- Alles YC, Greggio S, Alles RM, Azevedo PN, Xavier LL, DaCosta JC. A novel preclinical rodent model of collagenase-induced germinal matrix/intraventricular hemorrhage. *Brain Res.* 2010; 1356:130–8. [PubMed: 20692236]
- Back SA, Han BH, Luo NL, Chricton CA, Xanthoudakis S, Tam J, Arvin KL, Holtzman DM. Selective vulnerability of late oligodendrocyte progenitors to hypoxia-ischemia. *J Neurosci.* 2002; 22(2):455–463. [PubMed: 11784790]
- Balasubramaniam J, Del Bigio MR. Animal models of germinal matrix hemorrhage. *J Child Neurol.* 2006; 21(5):365–71. [PubMed: 16901440]
- Balasubramaniam J, Xue M, Buist RJ, Ivanco TL, Natuik S, Del Bigio MR. Persistent motor deficit following infusion of autologous blood into the periventricular region of neonatal rats. *Exp Neurol.* 2006; 197(1):122–32. [PubMed: 16271716]
- Bang H, Egerer K, Gauliard A, Lüthke K, Rudolph PE, Fredenhagen G, Berg W, Feist E, Burmester GR. Mutation and citrullination modifies vimentin to a novel autoantigen for rheumatoid arthritis. *Arthritis Rheum.* 2007; 56(8):2503–11. [PubMed: 17665451]
- Barnes CM, McElrath TF, Folkman J, Hansen AR. Correlation of 2-methoxyestradiol levels in cord blood and complications of prematurity. *Pediatr Res.* 2010; 67(5):545–50. [PubMed: 20098341]
- Bass WT, Buescher ES, Hair PS, White LE, Welch JC, Burke BL. Proinflammatory cytokine-receptor interaction model improves the predictability of cerebral white matter injury in preterm infants. *Am J Perinatol.* 2008; 25(4):211–8. [PubMed: 18548393]
- Bass WT. Periventricular Leukomalacia. *Neoreviews.* 2011; 12:76–84.
- Bassan H. Intracranial hemorrhage in the preterm infant: understanding it, preventing it. *Clin Perinatol.* 2009; 36(4):737–62. [PubMed: 19944833]
- Baud O, Daire JL, Dalmaz Y, Fontaine RH, Krueger RC, Sebag G, Evrard P, Gressens P, Verney C. Gestational hypoxia induces white matter damage in neonatal rats: a new model of periventricular leukomalacia. *Brain pathology.* 2004; 14(1):1–10. [PubMed: 14997932]
- Bembea MM, Savage W, Strouse JJ, Schwartz JM, Graham E, Thompson CB, Everett A. Glial fibrillary acidic protein as a brain injury biomarker in children undergoing extracorporeal membrane oxygenation. *Pediatr Crit Care Med.* 2011; 12(5):572–9. [PubMed: 21057367]
- Berger PR, Adelson PD, Richichi R, Kochanek PM. Serum biomarkers after traumatic and hypoxic brain injuries: insight into the biochemical response of the pediatric brain to inflicted brain injury. *Developmental neuroscience.* 28(4-5):327–335. [PubMed: 16943655]

- Berger RP, Hayes R, Richichi R, Beers S, Wang KK. Serum concentrations of ubiquitin C-terminal hydrolase-L1 and α II-spectrin breakdown product 145 kDa correlate with outcome after pediatric TBI. *Journal of neurotrauma*. 2012; 29(1):162–167. [PubMed: 22022780]
- Berghella, V. *Preterm Birth: Prevention and Management*. 1st Edition. Wiley-Blackwell; 2010.
- Bhandari V, Buhimschi CS, Han CS, Lee SY, Pettker CM, Campbell KH, Dulay AT, Oliver EA, Werner EF, Buhimschi IA. Cord blood erythropoietin and interleukin-6 for prediction of intraventricular hemorrhage in the preterm neonate. *J Matern Fetal Neonatal Med*. 2011; 24(5): 673–9. [PubMed: 20937006]
- Blennow M, Rosengren L, Jonsson S, Forssberg H, Katz-Salamon M, Hagberg H, Hesser U, Lagererantz H. Glial fibrillary acidic protein is increased in the cerebrospinal fluid of preterm infants with abnormal neurological findings. *Acta Paediatrica*. 1996; 85(4):485–489. [PubMed: 8740311]
- Buonocore G, Perrone S, Gioia D, Gatti MG, Massafra C, Agosta R, Bracci R. Nucleated red blood cell count at birth as an index of perinatal brain damage. *Am J Obstet Gynecol*. 1999; 181(6): 1500–5. [PubMed: 10601935]
- Cai Z, Pang Y, Xia F, Rhodes PG. Chronic ischemia preferentially causes white matter injury in the neonatal rat brain. *Brain research*. 2001; 898(1):126–135. [PubMed: 11292456]
- Cherian SS, Love S, Silver IA, Porter HJ, Whitelaw AG, Thoresen M. Posthemorrhagic ventricular dilation in the neonate: Development and characterization of a rat model. *J Neuropathol Exp Neurol*. 2003; 62:292–303. [PubMed: 12638733]
- Cherian S, Whitelaw A, Thoresen M, Love S. The pathogenesis of neonatal post-hemorrhagic hydrocephalus. *Brain Pathol*. 2004; 14:305–311. [PubMed: 15446586]
- Choi EK, Park D, Kim TK, Lee SH, Bae DK, Yang G, Kim YB. Animal models of periventricular leukomalacia. *Laboratory animal research*. 2011; 27(2):77–84. [PubMed: 21826166]
- Chow LC, Soliman A, Zandian M, Danielpour M, Krueger RC Jr. Accumulation of transforming growth factor- β 2 and nitrated chondroitin sulfate proteoglycans in cerebrospinal fluid correlates with poor neurologic outcome in preterm hydrocephalus. *Biol. Neonate*. 2005; 88:1–11. [PubMed: 15711035]
- Douglas-Escobar M, Weiss MD. Biomarkers of brain injury in the premature infant. *Front Neurol*. 2013; 3:185. [PubMed: 23346073]
- Eklind S, Mallard C, Leverin AL, Gilland E, Blomgren K, Mattsby-Baltzer I, Hagberg H. Bacterial endotoxin sensitizes the immature brain to hypoxic-ischaemic injury. *European Journal of Neuroscience*. 2001; 13(6):1101–1106. [PubMed: 11285007]
- Ellison VJ, Mocatta TJ, Winterbourn CC, Darlow BA, Volpe JJ, Inder TE. The relationship of CSF and plasma cytokine levels to cerebral white matter injury in the premature newborn. *Pediatr Res*. 2005; 57(2):282–6. [PubMed: 15585689]
- Falahati S, Breu M, Waickman AT, Phillips AW, Arauz EJ, Snyder S, Porambo M, Goeral K, Comi AM, Wilson MA, Johnston MV, Fatemi A. Ischemia-induced neuroinflammation is associated with disrupted development of oligodendrocyte progenitors in a model of periventricular leukomalacia. *Developmental neuroscience*. 2013; 35(2-3):182–196. [PubMed: 23445614]
- Florio P, Perrone S, Luisi S, Vezzosi P, Longini M, Marzocchi B, Petraglia F, Buonocore G. Increased plasma concentrations of activin a predict intraventricular hemorrhage in preterm newborns. *Clin Chem*. 2006; 52(8):1516–21. 2006. [PubMed: 16740650]
- Foerch C, Curdt I, Yan B, Dvorak F, Hermans M, Berkefeld J, Raabe A, Neumann-Haefelin T, Steinmetz H, Sitzer MJ. Serum glial fibrillary acidic protein as a biomarker for intracerebral haemorrhage in patients with acute stroke. *Neurol Neurosurg Psychiatry*. 2006; 77(2):181–4.
- Forsblad K, Källén K, Marsál K, Hellström-Westas L. Apgar score predicts short-term outcome in infants born at 25 gestational weeks. *Acta Paediatrica*. 2007; 96(2):166–171. [PubMed: 17429898]
- Gagnon A, Ye B. Discovery and application of protein biomarkers for ovarian cancer. *Curr Opin Obstet Gynecol*. 2008; 20(1):9–13. [PubMed: 18196999]
- Gazzolo D, Vinesi P, Bartocci M, Geloso MC, Bonacci W, Serra G, Haglid KG, Michetti F. Elevated S100 blood level as an early indicator of intra-ventricular hemorrhage in preterm infants. Correlation with cerebral Doppler velocimetry. *J. Neurol.Sci*. 1999; 170:32–35. [PubMed: 10540033]

- Gazzolo D, Marinoni E, Giovannini L, Letizia C, Serra G, Di Iorio R. Circulating adrenomedullin is increased in preterm newborns developing intraventricular hemorrhage. *Pediatr Res*. 2001a; 50(4): 7.
- Gazzolo D, Bruschetti M, Lituanica M, Serra G, Bonacci W, Michetti F. Increased urinary S100B protein as an early indicator of intraventricular hemorrhage in preterm infants: correlation with the grade of hemorrhage. *Clin. Chem*. 2001b; 47:1836–1838. [PubMed: 11568094]
- Gazzolo D, Florio P, Ciotti S, Marinoni E, di Iorio R, Bruschetti M, Sacchi R, Serra G, Lituanica M, Michetti F. S100B protein in urine of preterm newborns with ominous outcome. *Pediatr Res*. 2005; 58(6):1170–4. [PubMed: 16306188]
- Goel M, Dwivedi R, Gohiya P, Hegde D. Nucleated Red Blood Cell in Cord Blood as a Marker of Perinatal Asphyxia. *Clin Neonatol*. 2013; 2(4):179–182.
- Goyal A, Failla MD, Niyonkuru C, Amin K, Fabio A, Berger RP, Wagner AK. S100b as a Prognostic Biomarker in Outcome Prediction for Patients with Severe Traumatic Brain Injury. *Journal of neurotrauma*. 2013; 30(11):946–957. [PubMed: 23190274]
- Graham, DL.; Lantons, PI. *Greenfield's Neuropathology*. Arnold; London: 2002.
- Graham EM, Holcroft CJ, Rai KK, Donohue PK, Allen MC. Neonatal cerebral white matter injury in preterm infants is associated with culture positive infections and only rarely with metabolic acidosis. *Am J Obstet Gynecol*. 2004; 191(4):1305–10. [PubMed: 15507958]
- Guo K, Yang Y, Qiu J, Kan Q, Zhou XG, Zhou XY. The Expression Profile of microRNAs in Wistar Rats with Lipopolysaccharide-Induced Periventricular Leukomalacia. *J Mol Neurosci*. 2013; 51(3):941–949. [PubMed: 23354881]
- Hamilton BE, Hoyert DL, Martin JA, Strobino DM, Guyer B. Annual summary of vital statistics: 2010–2011. *Pediatrics*. 2013; 131(3):548–58. [PubMed: 23400611]
- Hansen A, Whitelaw A, Lapp C, Brugnara C. Cerebrospinal fluid plasminogen activator inhibitor-1: a prognostic factor in posthaemorrhagic hydrocephalus. *Acta Paediatr*. 1997; 86:995–998. [PubMed: 9343282]
- Heep A, Behrendt D, Nitsch P, Fimmers R, Bartmann P, Dembinski J. Increased serum levels of interleukin 6 are associated with severe intraventricular haemorrhage in extremely premature infants. *Arch Dis Child Fetal Neonatal Ed*. 2003; 88:F501–F504. [PubMed: 14602698]
- Heep A, Stoffel-Wagner B, Bartmann P, Benseler S, Schaller C, Groneck P, Obladen M, Felderhoff-Mueser U. Vascular endothelial growth factor and transforming growth factor-beta1 are highly expressed in the cerebrospinal fluid of premature infants with posthemorrhagic hydrocephalus. *Pediatr. Res*. 2004; 56:768–774. [PubMed: 15319463]
- Hermansen. Nucleated red blood cells in the fetus and newborn. *Arch Dis Child Fetal Neonatal Ed*. 2001; 84:F211–F215. [PubMed: 11320052]
- Holcroft CJ, Blakemore KJ, Allen M, Graham EM. Association of prematurity and neonatal infection with neurologic morbidity in very low birth weight infants. *Obstet Gynecol*. 2003; 101(6):1249–53. [PubMed: 12798532]
- Jelinski SE, Yager JY, Juurlink BH. Preferential injury of oligodendroblasts by a short hypoxic-ischemic insult. *Brain research*. 1999; 815(1):150–153. [PubMed: 9974135]
- Kallankari H, Kaukola T, Ojaniemi M, Herva R, Perhomaa M, Vuolteenaho R, Kingsmore SF, Hallman M. Chemokine CCL18 predicts intraventricular hemorrhage in very preterm infants. *Ann.Med*. 2010; 42:416–425. [PubMed: 20608885]
- Kassal R, Anwar M, Kashlan F, Simulian J, Hiatt M, Hegyi T. Umbilical vein interleukin-6 levels in very low birth weight infants developing intraventricular hemorrhage. *Brain Dev*. 2005; 27:483–487. [PubMed: 16198205]
- Keller T, Zeller T, Peetz D, Tzikas S, Roth A, Czyz E, Bickel C, Baldus S, Warnholtz A, Fröhlich M, Sinning CR, Eleftheriadis MS, Wild PS, Schnabel RB, Lubos E, Jachmann N, Genth-Zotz S, Post F, Nicaud V, Tiret L, Lackner KJ, Münzel TF, Blankenberg S. Sensitive troponin I assay in early diagnosis of acute myocardial infarction. *N Engl J Med*. 2009; 361(9):868–77. [PubMed: 19710485]
- Khwaja O, Volpe J. Pathogenesis of cerebral white matter injury of prematurity. *Arch Dis Child Fetal Neonatal Ed*. 2008; 93(2):F153–F161. [PubMed: 18296574]

- Kochanek PM, Berger RP, Fink EL, Au AK, Bayir H, Bell MJ, Dixon CE, Clark RS. The potential for bio-mediators and biomarkers in pediatric traumatic brain injury and neurocritical care. *Frontiers in neurology*. 2013;4. [PubMed: 23408773]
- Lai M, Sirimanne E, Williams CE, Gluckman PD. Sequential patterns of inhibin subunit gene expression following hypoxic-ischemic injury in the rat brain. *Neuroscience*. 1996; 70:1013–1024. [PubMed: 8848164]
- Leitner K, Al Shammery M, McLane M, Johnston MV, Elovitz MA, Burd I. IL-1 Receptor Blockade Prevents Fetal Cortical Brain Injury but Not Preterm Birth in a Mouse Model of Inflammation-Induced Preterm Birth and Perinatal Brain Injury. *Am J Reprod Immunol*. 2014 Epub ahead of print.
- Lekic T, Manaenko A, Rolland W, Krafft PR, Peters R, Hartman RE, Altay O, Tang J, Zhang JH. Rodent neonatal germinal matrix hemorrhage mimics the human brain injury, neurological consequences, and post-hemorrhagic hydrocephalus. *Exp Neurol*. 2012; 236(1):69–78. [PubMed: 22524990]
- Leviton A, Dammann O, Durum SK. The adaptive immune response in neonatal cerebral white matter damage. *Ann Neurol*. 2005; 58(6):821–8. [PubMed: 16250014]
- Lipina R, Reguli S, Novackova L, Podesvova H, Brichtova E. Relation between TGF-beta 1 levels in cerebrospinal fluid and ETV outcome in premature newborns with posthemorrhagic hydrocephalus. *Childs Nerv. Syst*. 2010; 26:333–341. [PubMed: 19823849]
- Luisi S, Florio P, Reis FM, Petraglia F. Expression and secretion of activin A: Possible physiological and clinical implications. *Eur J Endocrinol*. 2001; 145:225–236. [PubMed: 11517001]
- Lumpkins KM, Bochicchio GV, Keledjian K, Simard JM, McCunn M, Scalea T. Glial fibrillary acidic protein is highly correlated with brain injury. *J Trauma*. 2008; 65(4):778–82. discussion 782–4. [PubMed: 18849790]
- Many A, Hubel CA, Roberts JM. Hyperuricemia and xanthine in preeclampsia, revisited. *Am J Obstet Gynecol*. 1996; 174:288–91. [PubMed: 8572024]
- Marom R, Lubetzky R, Mimouni FB, Bassan H, Sira LB, Berger I, Dollberg S, Mandel D. Neonatal absolute nucleated red blood cell counts do not predict the development of cystic periventricular leukomalacia. *Isr Med Assoc J*. 2012; 14(7):420–3. [PubMed: 22953617]
- McCarty JH, Monahan-Earley RA, Brown LF, Keller M, Gerhardt H, Rubin K, Shani M, Dvorak HF, Wolburg H, Bader BL, Dvorak AM, Hynes RO. Defective associations between blood vessels and brain parenchyma lead to cerebral hemorrhage in mice lacking α_v integrins. *Mol Cell Biol*. 2002; 22(21):7667–7677. [PubMed: 12370313]
- Merhar S. Biomarkers in neonatal posthemorrhagic hydrocephalus. *Neonatology*. 2011; 101(1):1–7. [PubMed: 21791933]
- Ment LR, Hirtz D, Hüppi PS. Imaging biomarkers of outcome in the developing preterm brain. *The Lancet Neurology*. 2009; 8(11):1042–1055.
- Murphy BP, Inder TE, Rooks V, Taylor GA, Anderson NJ, Mogridge N, Howrood LJ, Volpe JJ. Posthaemorrhagic ventricular dilatation in the premature infant: natural history and predictors of outcome. *Arch Dis Child Fetal Neonatal Ed*. 2002; 87:F37–F41. [PubMed: 12091289]
- Okamoto T, Takahashi S, Nakamura E, Nagaya K, Hayashi T, Shirai M, Fujieda K. Matrix metalloproteinases in infants with posthemorrhagic hydrocephalus. *Early Hum. Dev*. 2008; 84:137–139. [PubMed: 17919852]
- O'Shea TM, Klinepeter KL, Meis PJ, Dillard RG. Intrauterine infection and the risk of cerebral palsy in very low-birthweight infants. *Paediatr Perinat Epidemiol*. 1998; 12(1):72. [PubMed: 9483618]
- Papa L, Ramia MM, Kelly JM, Burks SS, Pawlowicz A, Berger RP. Systematic review of clinical research on biomarkers for pediatric traumatic brain injury. *Journal of neurotrauma*. 2013; 30(5): 324–338. [PubMed: 23078348]
- Perlman JM, Risser R. Relationship of uric acid concentrations and severe intraventricular hemorrhage /leukomalacia in the premature infant. *J. Pediatr*. 1998; 132(1):436–439. [PubMed: 9544897]
- Perlman JM. Predicting cerebral white matter injury in preterm infants. *Obstet Gynecol*. 2006; 107(3): 546–8. [PubMed: 16507921]

- Phelan JP, Korst LM, Ahn MO, Martin GI. Neonatal nucleated red blood cell and lymphocyte counts in fetal brain injury. *Obstet Gynecol.* 1998; 91(4):485–9. [PubMed: 9540927]
- Poralla C, Hertfelder HJ, Oldenburg J, Müller A, Bartmann P, Heep A. Elevated interleukin-6 concentration and alterations of the coagulation system are associated with the development of intraventricular hemorrhage in extremely preterm infants. *Neonatology.* 2012; 102(4):270–5. [PubMed: 22906886]
- Rezaie P, Dean A. Periventricular leukomalacia, inflammation and white matter lesions within the developing nervous system. *Neuropathology.* 2002; 22(3):106–132. [PubMed: 12416551]
- Sannia A, Zimmermann LJ, Gavilanes AW, Vles HJ, Calevo MG, Florio P, Gazzolo D. Elevated Activin A urine levels are predictors of intraventricular haemorrhage in preterm newborns. *Acta Paediatr.* 2013; 102(10):e449–54. [PubMed: 23808611]
- Savman K, Nilsson UA, Blennow M, Kjellmer I, Whitelaw A. Non-protein-bound iron is elevated in cerebrospinal fluid from preterm infants with posthemorrhagic ventricular dilatation. *Pediatr Res.* Feb; 2001 49(2):208–12. [PubMed: 11158515]
- Sävman K, Blennow M, Hagberg H, Tarkowski E, Thoresen M, Whitelaw A. Cytokine response in cerebrospinal fluid from preterm infants with posthaemorrhagic ventricular dilatation. *Acta Paediatr.* 2002; 91(12):1357–63. [PubMed: 12578295]
- Schmitz T, Heep A, Groenendaal F, Hüsemann D, Kie S, Bartmann P, Obladen M, Felderhoff-Müser U. Interleukin-1beta, interleukin-18, and interferon-gamma expression in the cerebrospinal fluid of premature infants with posthemorrhagic hydrocephalus--markers of white matter damage? *Pediatr Res.* Jun; 2007 61(6):722–6. [PubMed: 17426654]
- Serpero LD, Bellissima V, Colivicchi M, Sabatini M, Frigiola A, Ricotti A, Ghiglione V, Strozzi MC, Li Volti G, Galvano F, Gazzolo D. Next generation biomarkers for brain injury. *J Matern Fetal Neonatal Med.* 2013; 26(Suppl 2):44–9. [PubMed: 24059552]
- Shahim P, Månsson JE, Darin N, Zetterberg H, Mattsson N. Cerebrospinal fluid biomarkers in neurological diseases in children. *European Journal of Paediatric Neurology.* 2013; 17(1):7–13. [PubMed: 23026858]
- Shen Y, Plane JM, Deng W. Mouse models of periventricular leukomalacia. *J Vis Exp.* 2010; 18(39)
- Silva AN, Smith RN, Lehmann CU, Johnson EA, Holcroft CJ, Graham EM. Neonatal nucleated red blood cells and the prediction of white matter injury in preterm infants. *Obstet Gynecol.* 2006; 107:550–6. [PubMed: 16507923]
- Sorokin Y, Romero R, Mele L, Wapner RJ, Iams JD, Dudley DJ, Spong CY, Peaceman AM, Leveno KJ, Harper M, Caritis SN, Miodovnik M, Mercer BM, Thorp JM, O'Sullivan MJ, Ramin SN, Carpenter MW, Rouse DJ, Sibai B. Maternal serum interleukin-6, C-reactive protein, and matrix metalloproteinase-9 concentrations as risk factors for preterm birth <32 weeks and adverse neonatal outcomes. *Am J Perinatol.* 2010; 27:631–640. [PubMed: 20195952]
- Stewart A, Tekes A, Huisman TA, Jennings JM, Allen MC, Northington FJ, Everett AD, Graham EM. Glial fibrillary acidic protein as a biomarker for periventricular white matter injury. *Am J Obstet Gynecol.* 2013; 209(1):27. [PubMed: 23467054]
- Sturtz WJ, Leef KH, Mackley AB, Sharma S, Bottiglieri T, Paul DA. Homocysteine levels in preterm infants: is there an association with intraventricular hemorrhage? A prospective cohort study. *BMC Pediatr.* 2007; 28:7, 38.
- Sutton PS, Darmstadt GL. Preterm birth and neurodevelopment: a review of outcomes and recommendations for early identification and cost-effective interventions. *J Trop Pediatr.* 2013; 59(4):258–65. [PubMed: 23486391]
- Sysyn GD, Rozycki HJ. Lack of prognostic significance of early elevated serum uric acid levels in low birth weight infants. *Biol. Neonate.* 2003; 83:253–257. [PubMed: 12743454]
- Tretter YP, Munz B, Hubner G, ten Bruggencate G, Werner S, Alzheimer C. Strong induction of activin expression after hippocampal lesion. *Neuroreport.* 1996; 7:1819–1823. [PubMed: 8905672]
- Tüzün F, Gencpinar P, Özbal S, Dilek M, Ergur BU, Duman N, Ozkan H, Kumral A. Neuroprotective effect of neotrofin in a neonatal rat model of periventricular leukomalacia. *Neuroscience letters.* 2012; 520(1):6–10. [PubMed: 22579826]

- Uehara H, Yoshioka H, Kawase S, Nagai H, Ohmae T, Hasegawa K, Sawada T. A new model of white matter injury in neonatal rats with bilateral carotid artery occlusion. *Brain research*. 1999; 837(1): 213–220. [PubMed: 10434005]
- Volpe, JJ. *Neurology of the Newborn*. 4th ed. WB Saunders; Philadelphia: 2001a. Intracranial hemorrhage: Germinal matrix-intraventricular hemorrhage; p. 428
- Volpe, JJ. *Neurology of the Newborn*. 4th ed. WB Saunders; Philadelphia: 2001b.
- Vos PE, Jacobs B, Andriessen TM, Lamers KJ, Borm GF, Beems T, Edwards M, Rosmalen CF, Vissers JL. GFAP and S100B are biomarkers of traumatic brain injury: an observational cohort study. *Neurology*. 2010; 75(20):1786–93. [PubMed: 21079180]
- Watanabe K, Hayakawa F, Okumura A. Neonatal EEG: a powerful tool in the assessment of brain damage in preterm infants. *Brain and Development*. 1999; 21(6):361–372. [PubMed: 10487468]
- Wilcox WD. Abnormal serum uric acid levels in children. *J Pediatr*. 1996; 128:731–41. [PubMed: 8648529]
- Weir CJ, Muir SW, Walters MR, Lees KR. Serum urate as an independent predictor of poor outcome and future vascular events after acute stroke. *Stroke*. 2003; 34(8):1951–6. [PubMed: 12843346]
- Wilson-Costello D, Friedman H, Minich N, Fanaroff AA, Hack M. Improved survival rates with increased neurodevelopmental disability for extremely low birth weight infants in the 1990s. *Pediatrics*. 2005; 115:997–1003. [PubMed: 15805376]
- Whitelaw A, Christie S, Pople I. Transforming growth factor-beta1: a possible signal molecule for posthemorrhagic hydrocephalus? *Pediatr. Res*. 1999; 46:576–580. [PubMed: 10541321]
- Whitelaw A, Rosengren L, Blennow M. Brain specific proteins in posthaemorrhagic ventricular dilatation. *Arch. Dis. Child Fetal Neonatal Ed*. 2001; 84:F90–F91. [PubMed: 11207222]
- Whitelaw A, Aquilina K. Management of posthaemorrhagic ventricular dilatation. *Arch. Dis. Child Fetal Neonatal Ed*. 2012; 97:F229–F223. [PubMed: 21289015]
- Woodward LJ, Anderson PJ, Austin NC, Howard K, Inder TE. Neonatal MRI to predict neurodevelopmental outcomes in preterm infants. *New England Journal of Medicine*. 2006; 355(7):685–694. [PubMed: 16914704]
- Xue M, Balasubramaniam J, Buist RJ, et al. Periventricular/intraventricular hemorrhage in neonatal mouse cerebrum. *J Neuropathol Exp Neurol*. 2003; 62:1154–1165. [PubMed: 14656073]
- Xue M, Del Bigio MR. Immune pre-activation exacerbates hemorrhagic brain injury in immature mouse brain. *J Neuroimmunol*. 2005; 165:75–82. [PubMed: 15964638]
- Zhang G, Ohsawa Y, Kametaka S, Shibata M, Waguri S, Uchiyama Y. Regulation of FLRG expression in rat primary astroglial cells and injured brain tissue by transforming growth factor-beta 1 (TGF-beta 1). *Neurosci Res*. 2003; 72:33–45.

Highlights

Review perinatal serological biomarkers of neurologic injury in prematurity.

Focus on the use of animal models in understanding the pathogenesis of intraventricular hemorrhage and periventricular leukomalacia.

Emphasize the need for future prospective studies aimed at identifying single- or multi-marker strategies to prevent or enable the early diagnosis of adverse neurological outcomes in preterm infants.

Table 1
Biomarkers of intraventricular hemorrhage (IVH) and periventricular leukomalacia (PLV) in prematurity

Biomarker	Function	Location	Change	Association	Sensitivity Specificity PPV, NPV, AUC
Activin	Growth factor	Neonatal Urine	Increase	IVH (Sania et al, 2013)	First urination Cut off >0.08ng/L: PPV:45.8%, NPP:93.4%, Sensitivity: 68,75% Specificity: 84.52% AUC:0.793
Uric acid	End product of purine metabolism, potent antioxidant	Neonatal Blood	Increase	IVH (Perlman et al, 1998)	N/A
S100	Calcium binding protein	Neonatal urine & blood	Increase	IVH (Gazzalo et al, 1999, 2001)	Sensitivity and specificity 100% at a urine cutoff of 0.70µg/l
IL-6	Cytokine	Neonatal blood & maternal blood	Increase	IVH (Heep et al, 2003, Kassal et al, 2004, Sorokin et al, 2010)	N/A
CRP	Protein which rises in response to inflammation	Maternal blood	Increase	IVH (Sorokin et al, 2010)	N/A
EPO	Hormone that controls erythropoiesis	Cord blood	Increase	IVH (Bhadari et al, 2011)	N/A
CCL-18	Member of the CC-chemokine family	Cord blood & Neonatal blood	Decrease	IVH (Kallankari et al, 2004)	Cut off value:4969 fluorescence units: Sensitivity: 74% Specificity:73% AUC:0.72
Adrenomedullin	Vasoactive peptide that participates in cerebral blood flow regulation	Neonatal blood	Increase	IVH (Sturtz et al, 2007)	N/A
TGF β1	Cytokine which upregulates the production by fibroblasts of extracellular matrix proteins	CSF	Increase	PPH (Whitelaw et al, 1999, Lipina et al, 2010)	Cut off 6.5ng/mL: Sensitivity:80% Specificity:78% PPV:80% NPV:78%
TGF β2	Cytokine which upregulates the	CSF	Increase	PHH (Chow et al, 2005)	N/A

Biomarker	Function	Location	Change	Association	Sensitivity Specificity PPV, NPV, AUC
	production by fibroblasts of extracellular matrix proteins				
NFL, GFAP, S100	NFL: essential component of neurofilament core in axons and can be used as a marker of axonal degeneration GFAP: Specific marker of differentiated astrocytes S100: Calcium binding protein	CSF	Increase	PHH (Whitelaw et al, 2011)	N/A
MMP 9	zinc-dependent endopeptidase	CSF	Increase	PHH (Okamoto et al, 2008)	N/A
NPBI	Potent catalyst in the generation of hydroxyl radicals	CSF	Increase	PHH (Savam et al, 2011)	N/A
PAI-1	primary inhibitor of the endogenous plasminogen activators	CSF	Increase	PHH (Hansen et al, 1997)	N/A
TNF- α , IL-1 β , IL-8, IL-18	Cytokines	CSF	Increase	PHH (Sävman et al, 2002, Schmitz et al, 2007)	N/A
GFAP	Specific marker of differentiated astrocytes	Neonatal blood	Increase	PVL (Stewart et al, 2013)	N/A
Nucleated RBCs	Erythroblasts, precursors to reticulocytes and mature erythrocytes	Neonatal blood	Increase	PVL (Silva et al, 2006)	Nucleated RBC count of 18 per 100 WBCs: sensitivity 56.9%, specificity 57.9%, PPV:57.9%, NPV:56.9%
IL-6, IL-8, IL-10, TNF- α , IFN γ	Cytokines	Neonatal CSF	Increase	PVL (Ellison et al, 2005)	N/A
TNF- α , IL-1 β , IL-6 and their soluble cytokine receptors (sTNF-RI, sTNF-RII, sIL-1RA, sIL-6R)	Cytokines & receptors	Neonatal Blood	Increase	PVL (Bass et al, 2008)	IL-6/sIL-6R interaction model: AUC:0.72, IL-1 β : AUC:0.47, sIL-1RA: AUC: 0.656, improving to AUC of 0.78 when combined. Combined

Biomarker	Function	Location	Change	Association	Sensitivity Specificity PPV, NPV, AUC
					modeling: AUC:0.84

Abbreviations: TNF- α :Tumor necrosis factor- α , IL-1:Interleukin-1, IL-6:Interleukin-6, IL-8:Interleukin-8, IL-18: Interleukin-18, INF- γ :Interferon γ , PPV:Positive predictive value, NPV:Negative predictive value, AUC:Area under the curve, CRP:C-reactive protein, EPO:Erythropoietin, CSF:Cerebrospinal fluid, IVH:Intraventricular hemorrhage, PHH:Posthemorrhagic hydrocephalus, PVL:Periventricular leucomalacia, TGF- β 1:Transforming growth factor beta 1, TGF- β 2:Transforming growth factor beta 2, GFAP: Glial fibrillary acidic protein , NFL: median neurofilament, MMP 9:matrix metalloproteinase 9, NPBI: Non-protein-bound iron, RBCs: Red Blood Cells, WBC:White Blood Cells, N/A: not available