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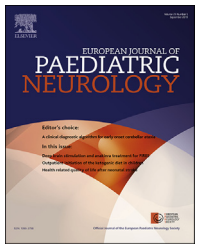

Original article

# Punctate white matter lesions of preterm infants: Risk factor analysis

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ABSTRACT

*Aim:* Punctate white matter lesions (PWML) are frequently detected in preterm infants undergoing brain MRI at term equivalent age (TEA). The aims of this study were to assess prevalence of PWML and to identify risk factors for PWML in VLBW infants.

*Methods:* Brain MRI scans obtained at TEA and clinical charts of a consecutive sample of very low birth weight (VLBW) infants admitted to Gaslini Children's Hospital NICU between 2012 and 2016 were retrospectively analyzed. MRI protocol included Susceptibility Weighted Imaging (SWI) sequence in order to identify hemosiderin depositions as a result of previous microbleeds. PWML were classified according to their number (≤6 lesions and >6 lesions) and signal characteristics (SWI+ lesions and SWI− lesions). Univariate and multivariable analysis were performed in order to identify risk factors for PWML (as a whole) and for each subgroup of PWML.

*Results:* 321 VLBW infants were included. PWML were identified in 61 subjects (19%), 26 of whom (8% of the study population) had more than 6 lesions. Risk factors for PWML (as a whole) were higher birth weight (OR = 1.001; p = 0.04) and absent or incomplete antenatal steroid course (OR = 2.13; p = 0.02). Risk factors for >6 PWML were need for intubation (OR = 11.9; p = 0.003) and higher Apgar score at 5 min (OR = 1.8; p = 0.02). Presence of GMH-IVH was the only identified risk factor for SWI + lesions.

*Conclusions:* Our results confirm the high prevalence of PWML among VLBW infants. Differentiation between SWI+ and SWI− lesions is crucial as they have different risk factors and may likely represent two different entities.



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## 1. Introduction

Survival rates of very low birth weight (VLBW) infants improved dramatically over the last decades, and the incidence of major brain lesions, including cystic periventricular leukomalacia (cPVL), has been constantly decreasing.[1] There is no consensus on the main determinant for this improvement in perinatal care, although several factors have been quoted, like improved ventilation techniques, higher rates of caesarean delivery and widespread adoption of antibiotic prophylaxis and antenatal corticosteroids.[2] On the other hand, milder forms of white matter injury, like punctate white matter lesions (PWML), are frequently diagnosed in preterm infants. This is likely due to the increasingly widespread use of magnetic resonance imaging (MRI) in this population.[3] PWML are seen at brain MRI as small, focal, often multiple, alterations of signal intensity (high on T1 and/or low on T2) in periventricular white matter[3–5] Dedicated MRI sequences like susceptibility-weighted imaging (SWI) can detect small hemosiderin depositions and therefore differentiate haemorrhagic from non-haemorrhagic PWML.[6] Less commonly, PWML can also be identified by cranial ultrasound as focal and persistent periventricular hyperechogenicities.[5]

Long-term outcome of PWML is still debated, although the number of lesions, their pattern, and their localization seem to be important.[7–10] De Bruïne et al. showed that infants with more than 6 PWML are at increased risk of adverse neurodevelopmental outcome,[8] while Guo et al. evidenced a connection between white matter injury in the frontal lobes and adverse cognitive and language development.[10] Moreover, white matter microstructure alteration and maturation delay have been associated with PWML.[11,12] Several risk factors have been proposed for white matter injury in preterm brain, but only few authors have focused their attention specifically on PWML.[13,14] The aims of this study were to assess prevalence of PWML and to investigate risk factors related to the development of different types of PWML in a large population of VLBW infants.

## 2. Materials and methods

### 2.1. Patient population

In January 2012, routine brain MRI at term-equivalent age (TEA, between $39^{+0}$ and $41^{+6}$ weeks post-menstrual age) was introduced as part of a screening program for prematurity-related lesions in all VLBW infants admitted to our NICU. In order to select a consecutive sample of VLBW infants who had undergone brain MRI at TEA, all VLBW infants consecutively admitted to our NICU between January 2012 and August 2016 were retrospectively identified: among this group, infants who died before TEA and those whose parents refused MRI were excluded from the study. This retrospective study was approved by the local ethics committee.

### 2.2. Data collection

Demographic and clinical data of included subjects were retrospectively extracted from our institutional electronic database and clinical charts. Collected data included demographic characteristics (birth weight and gestational age, gender and multiple gestation), prenatal data (use of assisted reproductive technologies, gestational diabetes, maternal hypertension, metrorrhagia and/or placental abruption, premature rupture of membranes, intrauterine growth retardation, antenatal steroid course), intrapartum data (type of delivery, Apgar score at 1 and 5 min), and postnatal data (need for intubation, mechanical ventilation and its duration, surfactant use, pneumothorax during first 72 h of life, inotrope use during first 72 h of life, early onset sepsis [EOS, bacteraemia or bacterial meningitis occurring at $\leq$72 h], late onset sepsis [LOS, worsening of clinical conditions occurring at $\geq$72 h treated with antibiotic therapy], multiple LOS episodes, LOS with bacteraemia, LOS with CSF pleocytosis, LOS with positive CSF culture, spontaneous intestinal perforation (SIP), necrotising enterocolitis (NEC), surgical treatment for NEC, pharmacological treatment for patent ductus arteriosus (PDA), surgical treatment for PDA, presence of germinal matrix haemorrhage – intraventricular haemorrhage (GMH-IVH), periventricular haemorrhagic infarction (PVHI), and cerebellar haemorrhage (CBH) on brain MRI (Table 1).

### 2.3. Imaging

MRI scans were performed at term-equivalent age during spontaneous sleep using the "feed and wrap" technique. The need for sedation (oral midazolam, 0.1 mg/kg) to prevent head motion was agreed with the neuroradiologist on the basis of the quality of images after the first sequence and the infant's state of arousal. Hearing protection was used in all patients, and heart rate and oxygen saturation were monitored non-invasively throughout the examination. Scans were performed with a 1.5 T MR system (InteraAchieva 2.6; Philips, Best, The Netherlands) using a dedicated paediatric head/spine coil. Our institutional standard MRI protocol included 3 mm thick axial T2-weighted and T1-weighted images, coronal T2-weighted images, sagittal T1-weighted images, axial diffusion-weighted images (b value: 1000 s/mm$^2$) and axial SWI. Informed consent that included statements about the significance and limitations of MRI at TEA was obtained in all cases.

### 2.4. Data analysis

MRI scans of included subjects were independently reviewed by two neuroradiologists experienced in neonatal neuroimaging (performing more than 200 neonatal brain MRI per year) blinded to the clinical history of the patients. PWML were defined as small areas of high T1 and/or low T2 signal in periventricular white matter.[4] PWML were classified according to their number ($\leq$6 lesions and >6 lesions) and signal characteristics: in the presence of low signal on SWI ("black dot") PWML were classified as SWI+, while in the absence of low signal as SWI−.[6] In case of concomitant presence of SWI+

**Table 1 – Univariate analysis of risk factors for PWML.**

| | Study population (N = 321) | PWML (N = 61) | No PWML (N = 260) | p-value |
|---|---|---|---|---|
| Birth weight (gr) mean ± SD | **1052 ± 265** | **1139 ± 223** | **1031 ± 270** | **0.004** |
| GA (weeks) mean ± SD | 28.3 ± 2.3 | 28.4 ± 1.8 | 28.3 ± 2.3 | 0.71 |
| Apgar score at 1′ mean ± SD | **5.5 ± 2.1** | **6.1 ± 1.6** | **5.4 ± 2.1** | **0.01** |
| Apgar score at 5′ mean ± SD | **7.7 ± 1.3** | **8.1 ± 0.8** | **7.6 ± 1.3** | **0.005** |
| Male gender (%) | **146 (45.5)** | **35 (57.4)** | **111 (42.7)** | **0.04** |
| Multiple gestation (%) | 118 (36.8) | 25 (41) | 84 (37.3) | 0.66 |
| ART (%) | 68 (21.2) | 13 (21.3) | 55 (21.2) | 1 |
| IUGR (%) | 76 (23.7) | 11 (18) | 65 (25) | 0.32 |
| Absent/incomplete antenatal steroid course (%) | **81 (25.2)** | **21 (34.4)** | **60 (23.1)** | **0.07** |
| Gestational diabetes (%) | 14 (4.4) | 2 (3.3) | 12 (4.6) | 1 |
| Maternal hypertension (%) | **59 (18.4)** | **6 (9.8)** | **53 (20.4)** | **0.06** |
| Metrorrhagia/placental abruption (%) | 51 (15.9) | 9 (14.8) | 42 (16.2) | 1 |
| PROM (%) | 89 (27.7) | 18 (29.5) | 71 (27.3) | 0.75 |
| Birth by caesarean section (%) | 246 (76.6) | 43 (70.5) | 203 (78.1) | 0.24 |
| Need for intubation (%) | 241 (75.1) | 51 (83.6) | 190 (73.1) | 0.10 |
| Intubation during first 72 h (%) | 233 (72.6) | 49 (80.3) | 184 (70.8) | 0.15 |
| MV > 2 h (%) | 194 (60.4) | 40 (65.6) | 154 (59.2) | 0.39 |
| MV > 2 h during first 72 h (%) | 174 (54.2) | 34 (55.7) | 140 (53.8) | 0.89 |
| MV > 7 days (%) | 86 (26.8) | 15 (24.6) | 71 (27.3) | 0.75 |
| MV > 14 days (%) | 55 (17.1) | 9 (14.8) | 46 (17.7) | 0.71 |
| MV > 28 days (%) | 27 (8.4) | 4 (6.6) | 23 (8.8) | 0.80 |
| Surfactant (%) | 226 (70.4) | 47 (77) | 179 (68.8) | 0.27 |
| Multiple surfactant doses (%) | 98 (30.5) | 15 (24.6) | 83 (31.9) | 0.28 |
| Pneumothorax during first 72 h (%) | 11 (3.4) | 2 (3.3) | 9 (3.5) | 1 |
| Inotrope use during first 72 h (%) | 28 (8.7) | 5 (8.2) | 23 (8.8) | 1 |
| EOS (%) | 11 (3.4) | – | 11 (4.2) | 0.13 |
| LOS (%) | 142 (44.2) | 24 (39.3) | 118 (45.4) | 0.47 |
| LOS >1 episode (%) | 48 (15) | 8 (13.1) | 40 (15.4) | 0.84 |
| LOS with bacteraemia (%) | 42 (13.1) | 6 (9.8) | 36 (13.8) | 0.52 |
| LOS with CSF pleocytosis (%) | 17 (5.3) | 1 (1.7) | 16 (6.4) | 0.21 |
| LOS with positive CSF culture (%) | 3 (0.9) | 1 (1.7) | 2 (0.8) | 0.47 |
| LOS with CRP rise (%) | 92 (28.7) | 14 (23) | 78 (30) | 0.34 |
| SIP (%) | 4 (1.2) | 2 (3.3) | 2 (0.8) | 0.16 |
| NEC (%) | 21 (6.5) | 5 (8.2) | 16 (6.2) | 0.57 |
| Surgery for NEC (%) | 11 (3.4) | 2 (3.3) | 9 (3.5) | 1 |
| PDA (%) | 196 (61.1) | 40 (65.6) | 156 (60) | 0.47 |
| Surgery for PDA (%) | 33 (10.3) | 6 (9.8) | 27 (10.4) | 1 |
| Any surgical intervention (%) | 56 (17.4) | 13 (21.3) | 43 (16.5) | 0.36 |
| GMH-IVH (%) | 82 (25.5) | 18 (29.5) | 64 (24.6) | 0.42 |
| PVHI (%) | 16 (5) | 2 (3.3) | 14 (5.4) | 0.74 |
| CBH (%) | 53 (16.5) | 9 (14.8) | 44 (16.9) | 0.85 |

**GA:** gestational age; **ART**: assisted reproductive technologies; **IUGR:** intrauterine growth retardation; **PROM:** premature rupture of membranes; **MV:** mechanical ventilation; **EOS:** early onset sepsis; **LOS:** late onset sepsis; **SIP:** spontaneous intestinal perforation; **NEC:** necrotising enterocolitis; **PDA**: patent ductus arteriosus; **GMH-IVH:** germinal matrix haemorrhage – intraventricular haemorrhage; **PVHI**: periventricular haemorrhagic infarction; **CBH:** cerebellar haemorrhage.
Bold signifies statistically significant or borderline significant (p<0.08) in the univariate analysis and thus used for multivariable analysis.

and SWI– lesions in the same patient, prevalent type of lesions was considered. Any discrepancies in interpretation of MRI findings were resolved by consensus.

Descriptive statistics were generated for the whole cohort and data were expressed as mean and standard deviation (SD) for continuous variables. Absolute or relative frequencies were calculated and reported for categorical variables. Differences between groups were evaluated with Student t test for continuous variables and with $\chi^2$ or Fisher's exact test for categorical variables.

Univariate analysis was carried out to determine which demographic, prenatal, intrapartum or postnatal characteristics were significantly more frequent among the patients with a specific lesion. The comparisons were performed in the following groups:

1. Patients with PWML vs patients without PWML
2. Patients with >6 PWML vs all the others (infants without PWML or with 6 PWML or less)
3. Patients with SWI + PWML vs all the others (infants without PWML or with SWI– PWML)
4. Patients with SWI– PWML vs all the others (infants without PWML or with SWI + PWML).

Logistic regression analyses were used for each variable and the results were reported as odds ratio (OR) with their 95% confidence intervals (CI). The absence of exposure to the factor or the variable that was less likely to be associated with the risk of the lesion was used as the reference for each analysis. Multivariable analysis was performed, and only variables that proved to be statistically or borderline significant in univariate

analysis ($p < 0.08$) were included in the model. The model showing the best fit was based on backward stepwise selection procedures, and each variable was removed if it did not contribute significantly. In the final model a P-value <0.05 was considered statistically significant, and all P-values were based upon two-tailed tests. Statistical analysis was performed using SPSS for Windows (SPSS Inc, Chicago, Illinois USA).

## 3. Results

Three hundred seventy-three VLBW infants were admitted to our unit in the selected period. Of them, 38 infants deceased before TEA, while the parents of 14 infants refused MRI for logistic or personal reasons. In the remaining population of 321 infants, GA ranged between 23 and 34 weeks (mean 28.3 ± 2.3) and birth weight between 435 and 1495 g (mean 1052 ± 265). Thirty-seven percent of infants were born from multiple pregnancies, 23% were born by vaginal delivery, and female/male ratio was 1.2.

Consensus was reached about the presence, number and signal characteristics of PWML in all cases. PWML were detected in 61/321 patients (19%). Of these, 26 subjects had more than 6 PWML (8% of study population and 43% of subjects with PWML). Among the 61 patients with PWML, low signal on SWI sequence (SWI+ PWML) was detected in 15 cases (4.7% of study population and 25% of subjects with PWML; Fig. 1A–C), and in two of them minor number of SWI− lesions coexisted. Forty-six patients presented only SWI− PWML (Fig. 1D–F).

Birth weight and Apgar score at 1 and 5 min were higher among subjects with PWML than in controls. Male gender and absent or incomplete antenatal steroid course were more frequent among subjects with PWML than in controls, while maternal hypertension was less frequent (Table 1). After multivariable analysis, absent or incomplete antenatal steroid treatment and higher birth weight were significantly associated with PWML (Table 2).

Birth weight ($p = 0.008$) and Apgar score at 5 min ($p = 0.05$) were higher, and absent or incomplete antenatal steroid course ($p = 0.02$) as well as need for intubation ($p = 0.003$) were more frequent among patients with >6 PWML than in controls. Twenty three out of 24 intubated infants in this group were intubated during the first 72 h of life, while one patient was intubated because of a late-onset sepsis. The majority of intubated infants (22/24) were intubated because of RDS and required surfactant administration. Birth by caesarean section ($p = 0.007$) and multiple surfactant doses ($p = 0.03$) were less frequent among patients with >6 PWML. The multivariable model revealed that intubation, higher Apgar score at 5 min, higher birth weight, and absent or incomplete antenatal steroid treatment were significantly associated with the development of >6 PWML, while caesarean section appeared protective (Table 2).

Surgical intervention ($p = 0.03$) and presence of GMH-IVH ($p = 0.001$), PVHI ($p = 0.03$) and CBH ($p = 0.02$) on MRI at TEA were more frequent among patients with SWI+ PWML than in controls. After multivariable analysis, only the presence of GMH-IVH was significantly associated with SWI+ PWML (Table 2).

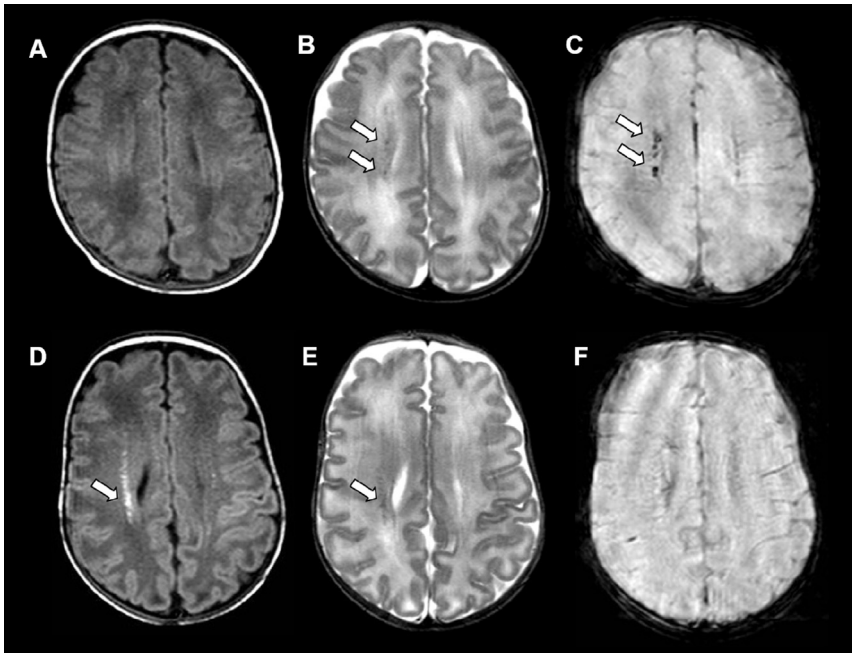


**Fig. 1 – (A–C) Brain MRI performed at term-equivalent age of an ex-preterm infant born at 25 weeks of GA showing SWI+ PWML. SWI+ lesions are barely detectable on the axial T1-weighed image (A), while they are clearly visible as hypointense spots on axial T2-weighted (B) and SWI (C) sequences. (D–F) Brain MRI performed at term-equivalent age of an ex-preterm infant born at 31 weeks of GA showing SWI− PWML. SWI− lesions are better seen on the axial T1-weighted (D) than on the axial T2-weighted image (E), and are not visible on SWI (D).**

**Table 2 – Multivariable analysis of risk factors for PWML, >6 PWML, SWI+ PWML and SWI– PWML.**

| Risk factors for PWML | PWML (N = 61) | no PWML (N = 260) | OR (95%CI) | p-value |
|---|---|---|---|---|
| Absent or incomplete antenatal steroid treatment | 21 (34.4%) | 60 (23.1%) | 2.13 (1.13–4) | 0.02 |
| Birth weight | 1139 ± 223 | 1031 ± 270 | 1.001 (1–1002) | 0.04 |
| **Risk factors for >6 PWML** | **>6 PWML (N = 26)** | **no >6 PWML (N = 295)** | **OR (95%CI)** | **p-value** |
| Birth by caesarean section | 14 (53.8%) | 232 (78.6%) | 0.22 (0.09–0.55) | 0.001 |
| Need for intubation | 24 (92.3) | 217 (73.6) | 11.9 (2.35–60) | 0.003 |
| Birth weight | 1185 ± 163 | 1040 ± 269 | 1.003 (1.001–1005) | 0.003 |
| Apgar score at 5′ | 8.1 ± 0.8 | 7.6 ± 1.3 | 1.8 (1.12–2.91) | 0.02 |
| Absent or incomplete antenatal steroid treatment | 12 (46.2%) | 69 (23.4%) | 2.53 (1.01–6.33) | 0.05 |
| **Risk factors for SWI+ PWML** | **SWI+ PWML (N = 15)** | **no SWI+ PWML (N = 306)** | **OR (95%CI)** | **p-value** |
| GMH-IVH | 10 (66.7) | 72 (23.5) | 6.5 (2.15–19.6) | 0.001 |
| **Risk factors for SWI– PWML** | **SWI– PWML (N = 46)** | **no SWI–PWML (N = 275)** | **OR (95%CI)** | **p-value** |
| Birth weight | 1203 ± 180 | 1026 ± 268 | 1.003 (1.002–1.004) | 0.0001 |

GMH-IVH: germinal matrix haemorrhage – intraventricular haemorrhage; OR = odds ratio; CI = confidence interval.

Birth weight (p = 0.001) and Apgar score at 1 (p = 0.03) and 5 (p = 0.01) minutes were higher among patients with SWI– PWML than in controls. The multivariable model showed that only higher birth weight was significantly associated with SWI– PWML (Table 2).

## 4. Discussion

In this work, we identified risk factors for punctate white matter lesions on brain MRI at TEA in a large single-centre cohort of VLBW infants. Among more than 50 clinical variables analysed (including demographics, prenatal, perinatal, and postnatal parameters) we identified only two significant risk factors for PWML: (1) a higher birth weight, and (2) absent or incomplete antenatal steroid course. Absence of significant association between other potential risk factors and PWML is in line with previous works. Wagenaar et al. identified only three significant factors for punctate lesions on early MRI scan – that is, a greater birth weight, a grade II-III IVH, and a cohort A.[13] On the other hand, Leijser and coworkers did not identify any significant risk factors for PWML in a TEA scan study.[14] While association between systemic inflammation and white matter injury has been reported in literature,[15] we did not observe any positive correlation between systemic inflammatory/infectious events (i.e. EOS, LOS, NEC, and CSF pleocytosis) and PWML. It is necessary to state, though, that no specific dosage of cytokines was performed in our study.

Higher birth weight as a risk factor of PWML can be regarded as a proxy of infant's brain maturity. Wagenaar et al. have showed that birth weight is a significant risk factor for PWML in a recent work.[13] Similarly, Leijser et al. observed a tendency towards higher birth weight in infants with multiple PWML.[14] In a recent publication describing neurodevelopmental consequences of PWML and involving 506 preterm infants, both a greater birth weight and a significantly higher gestational age were observed in the group of infants with lesions.[7] PWML seem to affect the brain in a certain developmental window, similar to periventricular leukomalacia where "more mature" premature infants are at higher risk.[16] This specific fragility of the white matter has been explained by a vulnerable window in oligodendrocytes development,[17] coinciding with major activation of microglia.[18] In other works, aberrant injury response expressed as arrested preoligodendrocyte maturation in reactive astrocyte-rich lesions has been associated with myelination failure and diffuse white matter injury.[18]

We found a correlation between absent or incomplete antenatal steroid course and the entire group of PWML. Our findings are somewhat different from previously published papers.[14,13,7] Antenatal corticosteroids enhance lung maturation and have been shown to decrease perinatal and neonatal deaths, respiratory distress syndrome, need for mechanical ventilation, early systemic infection, NEC, and IVH.[19] While positive effects of prenatal corticosteroids on risk of intraventricular haemorrhage have been confirmed by multiple studies,[20] the association with white matter damage is still controversial.[21] We believe our findings may add useful data supporting the protective role of steroids on PWML.

In the literature, severe hypocapnia caused by excessive mechanical ventilation has been listed as a risk factor for severe forms of white matter damage, like periventricular leukomalacia.[22] To our knowledge, this factor has not been yet investigated in relation to milder forms of white matter damage, like punctate lesions. No episodes of severe hypocapnia were described in clinical records of the patients included in our study, although we have not performed any specific analysis of $CO_2$ levels derived from different techniques (i.e. arteria cannulation or capillary heel pricks). We have not observed any relation between the duration of the ventilation (more than 2 h, more than 7, 14 or 28 days) and the development of punctate white matter lesions.

We have observed a slightly lower frequency of PVHI (3,3% vs 5,4%, non-significant) in the group of infants with PWML. Still, the relatively small absolute numbers of the patients do not allow us to consider this as a robust finding. We could speculate that PWML are more frequently seen in infants with higher birth weight, while PVHI is associated with higher degree of prematurity and lower birth weights, so these two lesions could just have different vulnerability windows in terms of gestational age at birth.

Total prevalence of PWML in our study population was of 19%; this is similar to previously reported figures from literature.[7,13,8] Only 8% (26/321) of subjects had more than 6 lesions – a condition connected with worse neurodevelopmental outcomes.[8] Results of multivariable analysis in our study demonstrated the importance of absent or incomplete antenatal steroid treatment and need for intubation in the development of these lesions, highlighting the importance of respiratory factors in white matter development.[23] We also observed that a higher Apgar score is a significant risk factor for >6 PWML. This latter observation could seem in contrast with previous ones, but our interpretation is that a typical infant with punctate lesions would be the one born at higher gestational age, with higher birth weight[7] and consequently in better clinical conditions expressed with higher Apgar score at 5 min. Those infants could be in a vulnerable window in oligodendrocytes development, as discussed above; in these conditions the stress connected with intubation could pone them at higher risk of white matter damage. In animal studies, intubation and mechanical ventilation have shown to increase the expression of cytokines and proinflammatory genes, mainly at the level of the lung.[24] We cannot at the moment determine whether in our case the intubation *per se* has contributed to triggering the inflammatory cascade leading to white matter injury, or if respiratory distress requiring intubation is to blame – further studies could help to clear the doubt.

From the point of view of prevention, we wonder if an even more wide use of complete steroid course could have an effect on frequency and severity of PWML, but as our study is a result of retrospective, and not prospective, analysis we are not sure about the strength of this association. We could only speculate that preventing respiratory distress syndrome and avoiding intubation via the use of new non-invasive techniques could have a beneficial effect on white matter development.

Birth by caesarean section seems to be protective against >6 PWML in our cohort. Nevertheless, we remain uncertain whether this observation is determined by intrinsic risks of vaginal delivery or by factors behind preterm spontaneous onset of labour, that are not always easy to define. Tusor et al. found no influence of clinical chorioamnionitis on PWML, while maternal urinary tract infection seemed to be more frequent among infants with PWML.[7] In our work, none of the analysed prenatal factors (use of assisted reproductive technologies, gestational diabetes, maternal hypertension, metrorrhagia and/or placental abruption, premature rupture of membranes, intrauterine growth retardation), except for absent or incomplete antenatal steroid course, resulted as a significant risk factor for PWML. Further studies including data from histological analysis of placenta could be useful in order to better characterise the influence of prenatal factors on white matter development.

When looking at SWI+ PWML only (15/61 cases, or 4.7% of population), we noticed a 6-times higher risk in GMH-IVH exposed infants, corroborating the hypothesis of their venous hemorrhagic pathogenesis, as suggested in the literature.[6,9] The significant association between SWI+ PWML and GMH-IVH is particularly intriguing. GMH-IVH tends to occur in the first 3–5 days of life and in a much shorter time compared to PWML. Venous sludging and microthrombosis have been proposed as potential pathophysiological mechanisms of GMH-IVH.[25] These same phenomena may also involve medullary veins of the periventricular white matter.[26] If bleeding occurs, the hemosiderin deposits along the ependyma may act as a pro-inflammatory stimulus for the adjacent white matter. We previously observed a reduction of fractional anisotropy in the white matter in babies with IVH and lower gestational age,[27] compatible with altered white matter maturation. A similar mechanism may be responsible of SWI+ PWML development in the white matter of preterm babies with GMH-IVH.

Interestingly, the only risk factor for SWI- PWML identified in our work was higher birth weight, suggesting different pathophysiological substrate for SWI+ and SWI– lesions. Although extensive MRI-histopathology correlation studies regarding PWML are lacking, few cases described in the literature seem to confirm existence of different sub-types of PWML. Niwa et al. in their work presented a case of a preterm infant born at 31 weeks and scanned on day 11, who died at 13 days of age. On MRI scan, punctate white matter lesions with low signal on SWI could be seen, corresponding to hemorrhagic changes on post-mortem analysis. On the other hand, the same patient presented lesions hyperintense on T1, but not visible on SWI. On post-mortem, those areas were characterized by histological changes compatible with early gliosis.[6] Another case was described by Rutherford et al.: a preterm infant with a probable neuromuscular disorder who died at 9 days if age. On post-mortem MRI punctate lesions were present, seen as focal low signal areas on T2, corresponding on the autopsy to areas of vascular congestion with infiltration of activated microglia.[3] No SWI sequence was available in that case, to our knowledge. Taken together these observations lead us to consider that the difference between SWI+ and SWI– punctate lesions requires further research.

Our work has some limitations. First of all, because of retrospective design of the study the confirmation of our findings by prospective studies is warranted. Furthermore, we studied a very specific population (i.e. VLBW infants) and extension of our results to other patient groups can be questionable. This is an important aspect to keep in mind considering that PMWL have been described at all gestational ages up to term.[5] Finally, the use of TEA MRI could limit our capacity of identifying those PWML disappearing at earlier stage, even if their clinical significance remains to be defined.[9] On the other hand, we have studied a large single-centre population with homogeneous treatment strategies and distribution of potential risk factors, as different standards of care may influence the data.[13] Furthermore, availability of SWI sequence in all MRI scans allowed us to perform clear identification of PWML with presence of hemosiderin (SWI+) and to analyse them separately from SWI– PWML.

In conclusion, we have identified several risk factors for PWML: the most important one for SWI– lesions is the greater birth weight, and for SWI+ the presence of GMH-IVH. This differentiation is crucial for a better understanding of the etiological factors at the base of these two lesions that, even if classified in the field of white matter lesions, likely represent two different entities. Further prospective studies are needed to corroborate our findings, and to better characterize and define these injuries and their related outcomes.

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## Conflict of interest

The authors declare no conflict of interest.

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