**Supplemental material**

**Cerebral palsy and polymorphism of the chemokine *CCL18* in very preterm children**

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**1.1. Description of the antenatal and neonatal factors**

GA was confirmed by ultrasound before 20 wk of gestation. Infants with BW below two standard deviations from the mean of gestation-adjusted BW were classified as small for gestational age. The infants underwent serial brain ultrasound assessments. IVH was classified according to Papile [1], and the most severe grade was included in the present analyses. BPD was defined as chronic respiratory disease requiring supplemental oxygen or any form of positive pressure ventilation therapy at the age of 36 postmenstrual wk [2].

1 Papile LA, Burstein J, Burstein R, Koffler H: Incidence and evolution of subependymal and intraventricular hemorrhage: a study of infants with birth weights less than 1,500 gm. J Pediatr 1978;92:529-534.

2 Walsh MC, Yao Q, Gettner P, Hale E, Collins M, Hensman A, Everette R, Peters N, Miller N, Muran G, Auten K, Newman N, Rowan G, Grisby C, Arnell K, Miller L, Ball B, McDavid G, National Institute of Child Health and Human Development Neonatal Research Network: Impact of a physiologic definition on bronchopulmonary dysplasia rates. Pediatrics 2004;114:1305-1311.

**1.2. DNA sample preparation**

Genomic DNA was extracted from buccal cells, umbilical cord blood, or paper blood samples. Extraction methods used were Chelex 100 (Bio-Rad, Hercules, CA, USA) for buccal cells, UltraClean DNA Blood Isolation kit (MO BIO Laboratories, Carlsbad, CA, USA) for umbilical cord blood, and MOBIO UltraClean BloodSpin DNA Isolation Kit (MO BIO Laboratories) for dried paper blood spots on Guthrie cards. A whole-genome amplification was performed for buccal cell and paper blood DNA samples, as described previously [3]. DNA samples that originated in Canada were extracted with the ChargeSwitch gDNA Mini Tissue Kit (Invitrogen, Carlsbad, CA, USA) for umbilical cord tissue and cell samples, QIAamp DNA Blood Midi Kit (Qiagen) for whole blood specimens, and ChargeSwitch gDNA Buccal Cell Kit (Invitrogen) for tracheal aspirate samples.

3 Karjalainen MK, Huusko JM, Ulvila J, Sotkasiira J, Luukkonen A, Teramo K, Plunkett J, Anttila V, Palotie A, Haataja R, Muglia LJ, Hallman M: A potential novel spontaneous preterm birth gene, AR, identified by linkage and association analysis of X chromosomal markers. PLoS One 2012;7:e51378.

## 1.3. Analysis of CCL18 levels in cord blood

Concentrations of CCL18 were analyzed using antibody-based protein microarrays with DNA amplification and the concentrations were reported as fluorescence units [4] .Briefly, a sample protein is captured by a specific antibody that is affixed to an oligonucleotide primer. A second, biotinylated detector antibody then binds to the antibody-DNA conjugate. Thereafter, a universal antibiotin antibody is bound to a second antibody. In the presence of DNA polymerase and fluorescent nucleotides, rolling-circle replication then occurs on the universal antibody. In the process of rolling-circle signal amplification, a circular DNA hybridizes to the oligonucleotide DNA primer. The rolling-circle signal amplification product, the fluorescence, is then detected by a microarray scanner. The amount of fluorescence equals the amount of a sample protein.

4 Schweitzer B, Roberts S, Grimwade B, Shao W, Wang M, Fu Q, Shu Q, Laroche I, Zhou Z, Tchernev VT, Christiansen J, Velleca M, Kingsmore SF: Multiplexed protein profiling on microarrays by rolling-circle amplification. Nat Biotechnol 2002;20:359-365.

**1.4. Acknowledgements**

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| **Supplemental Table 1.  *CCL18* polymorphisms and cord blood levels of CCL18** | | | |
| *CCL18* polymorphism | Genotypes (n) | CCL18 levels  mean (SD) | p value1 |
| rs1102934 | GA+GG (25) | 6818 (2353) | 0.16 |
|  | AA (61) | 5972 (2572) |  |
| rs2015086 | CC+CT (29) | 7055 (2366) | 0.033 |
|  | TT (70) | 5869 (2522) |  |
| rs2015070 | AA+GA (18) | 7213 (2515) | 0.064 |
|  | GG (81) | 5995 (2487) |  |
| rs2735835 | AA+GA (56) | 6378 (2674) | 0.69 |
|  | GG (40) | 6171 (2323) |  |
| rs712044 | GG+AG (39) | 6177 (1756) | 0.93 |
|  | AA (57) | 6218 (3007) |  |
| 1Student’s t-test.  CCL18 concentrations did not correlate with CP outcome among these 99 VLGA children. | | | |