

# Umbilical cord blood procalcitonin level in early neonatal infections: a 4-year university hospital cohort study

N. Joram, J.-B. Muller, S. Denizot, J.-L. Orsonneau, J. Caillon, J.-C. Rozé, C.

Gras-Le Guen

## ► To cite this version:

N. Joram, J.-B. Muller, S. Denizot, J.-L. Orsonneau, J. Caillon, et al.. Umbilical cord blood procalcitonin level in early neonatal infections: a 4-year university hospital cohort study. European Journal of Clinical Microbiology and Infectious Diseases, 2011, 30 (8), pp.1005-1013. 10.1007/s10096-011-1187-0. hal-00669195

# HAL Id: hal-00669195 https://hal.science/hal-00669195

Submitted on 12 Feb 2012  $\,$ 

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers. L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

- 1 Umbilical Cord Blood Procalcitonin Level in Early Neonatal Infections: A 4-
- 2 year University Hospital Cohort Study.
- 3
- 4 Nicolas Joram, M.D.; Jean-Baptiste Muller, M.D.; Sophie Denizot, M.D.; Jean-
- 5 Luc, Orsonneau, M.D.; Jocelyne Caillon, M.D., P.H.D.; Jean-Christophe Rozé,
- 6 M.D.;Christèle Gras-Le Guen, M.D.,
- 7 **P.H.D.**
- 8
- 9 Corresponding Author: Nicolas Joram, M.D
- 10 Département de Périnatologie, Hôpital Mère Enfant CHU Nantes, Nantes 44
- 11 093, France
- 12

13 **Abstract: Purpose.** Study of Procalcitonin (PCT) measured in cord blood as a

- 14 discriminating marker of early-onset neonatal infection. **Methods**. Monocenter
- 15 retrospective study with prospective collection of data including all babies born
- 16 during the study period. Those presenting infection risk factors had PCT
- 17 measurement. Three groups were defined : certainly infected, probably infected,
- and non-infected. **Results**. 12485 newborns were included, 2151 had PCT
- 19 measurement, 26 were infected. Receiver operating curves of PCT determined
- 20 0.6 ng/ml as the best cut-off, with an area under the curve of 0.96 (CI 95%: 0.95
- 0.98). Sensitivity, specificity, positive and negative predictive value and
- 22 positive and negative likelihood ratios were 0.92 (0.75 0.98), 0.97 (0.96 0.98), 0.97 (0.96 0.98), 0.98 (0.92 0.96), 0.99 (0.99 0.98), 0.97 (0.96 0.98), 0.99 (0.99 0.9
- 23 0.98), 0.28 (0.20 0.36), 0.99 (0.99 0.99), 32 (24 41) and 0.08 (0.02 0.3).
- Post-test probabilities were 28% (23 to 33) if the test was positive, and less than 25 0.010(100 1.1005) if the test was positive has a less than 25 0.010(100 1.1005) if the test was positive has a less than 25 0.010(100 1.1005)
- 0.01% (0 1.10-5) if the test was negative. Gestational age between 28 and 32
  weeks (OR 4.4, 1.2 16.2) and pH at birth < 7.10 (OR 2.9, 1.1 7.4) were other</li>
- independent factor of PCT increasing (p<0.05). **Conclusion**. PCT measured in
- 28 umbilical cord blood is reliable to detect early infected and non-infected
- 29 newborns.

30 Keywords: Early onset neonatal infection; neonate; procalcitonin

# 31 INTRODUCTION

Early-onset neonatal infection (EONI) suspicion is one of the most stressful situations for neonatologists, despite generalized prophylaxis of group B streptococcal (GBS) neonatal infection (1-3). Indeed, despite the reduced frequency of this pathology, the diagnosis remains an emergency because early diagnosis and treatment are critical to optimise the outcome (4). Because clinical 37 signs of EONI are known to be non specific, pediatricians often start early empiric antibiotic treatment in newborns presenting few symptoms or some risk 38 factors of infection before receiving results of bacteriologic cultures and 39 inflammatory markers, but as a result, exposing a considerable number of 40 patients to unnecessary treatment. In a recent study, Stocker et al reported that 41 an empiric antibiotic treatment (ampicillin and gentamicin) > 72h was 42 administered to 82% of a cohort of neonates with suspected EONI, while only 43 44 18% of them were finally classified as probable infections and 1% as proven infections (5). Concerns about subsequent consequences of such treatment 45 strategies are serious and multiple: changes in microbiotal ecology of the 46 neonates and the recently reported role of these changes in the early origins of 47 diseases (6,7); risk of development of resistant bacteria (8); nosocomial 48 49 infection risk; mother/newborn separation; and increased cost.

Currently, a sensitive and specific marker is required in order to make a definite 50 EONI diagnosis. While most clinical and standard biological markers are 51 inadequate (4, 9), procalcitonin (PCT), implicated as a sensitive and specific 52 marker of bacterial infections in various situations in adult and pediatric 53 medicine, may be a good candidate (10, 11). Some studies have shown a 54 physiological increase in the PCT concentration during the first three post natal 55 days, thereby making the interpretation of this marker during this period 56 complicated (12). In this context, we previously conducted a prospective cohort 57 study in neonates with suspected EONI, measuring the serum PCT concentration 58

at birth in umbilical cord blood, which reflects antenatal (transplacental or 59 ascendant) infectious process. PCT appeared in as a discriminating marker of 60 EONI with 87.5% (CI 95%: 71 - 100) sensitivity, 98.7% (CI 95%: 97.7 - 99.7) 61 specificity, 67.3 positive likelihood ratio and 0.13 negative likelihood ratio (13). 62 However, the determination of PCT concentration used a semi-quantitative 63 method with imprecise cut-off, and confidence intervals were large because of 64 the low incidence of EONI and a limited number of neonates with probable or 65 proven infection. 66

67 Consequently, the aim of this study was to evaluate the diagnostic value of a 68 quantitative umbilical cord blood PCT concentration in a large cohort of 69 neonates in a university hospital where PCT determination is routinely 70 performed in newborns presenting risk factors of EONI. We hypothesized that 71 umbilical blood cord PCT quantitative concentration is an early and 72 discriminating marker of EONI.

#### 74

#### PATIENTS AND METHODS

The study reported here follows STARD guidelines (14). We conducted a one center observational retrospective analysis of prospectively collected data from July 2005 to September 2008 in the University Hospital of Nantes, France. Written consent was not required. The study was approved by the local clinical research committee of the University Hospital of Nantes.

80

### Inclusion and exclusion criteria

81 All children born during this period were included in the study, composing an historical cohort. In our center, only those with risk factors for EONI usually 82 have a PCT measurement and were therefore considered in the statistical 83 analysis. These risk factors are those proposed by international and French 84 national consensus guidelines: clinical suspicion of chorioamnionitis (fever, 85 painful uterine contractions), intrapartum fever  $\geq 38^{\circ}$ C, spontaneous 86 premature delivery at < 37 gestational weeks (GW), prolonged rupture of 87 membrane > 12 h, maternal colonization with GBS, signs of fetal asphysia 88 (tachycardia, meconium stained amniotic fluid) and symptomatic newborn 89 (respiratory distress/apnea, tachycardia/bradycardia, arterial hypotension/poor 90 perfusion, seizure/floppy infant, irritability/lethargy, feeding intolerance) (15 -91 17). We add that since 2001, GBS screening in France is generalized and 92 antibiotics are administrated to the mother during labor in case of a positive test 93 in accordance with national guidelines (18). Newborns presenting antenatal 94 95 proinflammatory malformation (gastroschisis and omphalocele) were not 96 included and those whose data were incomplete were secondarily excluded from97 the study.

98 Laboratory test

Procalcitonin measurement and bacteriological samples (gastric and placental 99 culture) were performed in every newborn presenting infection risk factors. 100 Blood samples were obtained from umbilical blood in standard vacuum blood 101 collection tubes (Venosafe®, TERUMO) containing lithium heparin. The tubes 102 103 were carried to the biochemistry laboratory within one hour and then centrifuged at 3000 g for 10 minutes before analysis. We used a time-resolved amplified 104 cryptate emission (TRACE®) technology assay with the KRYPTOR® 105 automated analyzer (BRAHMS, Hennigsdorf, Germany). This assay requires 50 106 107 µl of plasma, and the reaction time is about 20 minutes. The assay has a very 108 low detection limit (0.02 ng/ml) and a functional sensitivity of 0.06 ng/ml. 109 Reproducibility of the test is excellent, with a coefficient of variation of 6% for a value of 0.25 ng/ml and 3% for values higher than 1.0 ng/ml. If the newborn 110 was symptomatic during the first 48 hours of life, or in the case of a positive 111 Gram stain of a bacteriological sample, investigations were extended after birth 112 113 with blood cell count, blood culture and C-Reactive protein (CRP). The latter was performed by an immunoturbidimetric method with a COBAS® 6000 114 analyzer (ROCHE DIAGNOSTICS, Mannheim, Germany). CRP levels were 115 obtained during the initial evaluation of the newborn (CRP1) and between 12 116 and 24 hours after (CRP2). 117

#### 118 Data collection

We systematically recorded data in a customized database at the time of 119 newborn discharge. Clinical data included gestational age at birth, birth weight, 120 sex, prenatal corticosteroid exposure, time of membrane rupture, mode of 121 122 antenatal antibiotics exposure, chorioamnionitis, preeclampsia, delivery. intrauterine growth retardation, death, multiple pregnancy, APGAR score at 5 123 minutes of life and abnormal clinical signs during the first 48h of life. Biological 124 125 data included the result of the mother's GBS screening, gastric fluid and placenta colonization, blood culture and cerebrospinal fluid result, pH at birth, 126 CRP1, CRP2, white blood cell (WBC) count, immature neutrophil rate, and 127 platelets count. 128

129

### Patient classification

130 According to the clinical, biological and bacterial findings, and in accordance with international criteria, three groups were a posteriori defined as certainly 131 infected, probably infected, and non-infected (19, 20). Two neonatologists who 132 were unaware of the procalcitonin value and the medical management of the 133 patient retrospectively classified newborns according to the following criteria. In 134 135 cases of discordance, a third opinion was used. The certainly infected group was defined by a positive central sample (blood culture or cerebro spinal fluid). A 136 newborn was considered as probably infected if he presented clinical signs and 137 at least two abnormal laboratory results among WBC count >  $34 \ 10^{9}$ /l or <  $5 \ 10^{9}$ 138 /l, immature neutrophil > 10%, thrombocytopenia < 100  $10^9$  /l, and CRP > 20 139

mg /l when blood culture was negative. Patients were considered non-infected
when their data did not match these criteria. For the analysis of results, probably
and certainly infected newborns were considered infected.

143

### Statistical analysis

We first described baseline characteristics of all patients presenting infection 144 risk factor. We then analyzed PCT distribution among infected (certainly or 145 probably) and non-infected newborns and we compared PCT median in these 146 147 two groups using Mann and Whitney test. We then performed a ROC curve using Medicalc software in order to determine the best cut-off for PCT measured 148 in cord blood. Sensitivity, specificity, negative and positive predicting values 149 and negative and positive likelihood ratios were assessed using SPSS For 150 Windows version 15.0. Post-test probabilities were determined using a 151 152 nomogram of bayes (21). The abilities of PCT and CRP to discriminate neonates with and without early neonatal infection were compared using the method of 153 comparison of AUC published by Hanley and McNeil (22). Then, in order to 154 assess the ability of antenatal infection risk factors and PCT to predict infection, 155 we analyzed their statistical relationship with infection performing a univariate 156 157 analysis followed by a multivariate analysis by logistic regression. All the following variables were binary and were coded as follows: (0) no, (1) yes for 158 clinical suspicion of chorioamniotis, intrapartum fever >38°C, spontaneous 159 premature delivery < 37 GW, prolonged rupture of membrane > 12 h, maternal 160 colonization with GB, fetal asphyxia, and PCT value higher than the cut-off. 161

162 Then, in order to compare the accuracy of these clinical antenatal risk factors and of PCT for the screening of patients, we built two mathematical models. For 163 the building of these two models, the whole population was randomized into 2 164 subgroups using an SPSS 15.0 random function. We then performed a new 165 logistic regression to select items composing the models, among the first 166 subgroup with a significance limit of p>0.05 to remove from the model, and 167 then validated it in the second subgroup. The first model was called 'clinical 168 model' and only included clinical risk factors. The second model was called 'bio 169 170 clinical model' and was the same as the first model, but also included the PCT value. The ability of each model to discriminate between infected and non 171 infected newborns was evaluated by their AUC. AUCs of the two models were 172 173 compared in each subgroup and to PCT alone using the Hanley and McNeil 174 method (22). Additionally, for each model we compared the AUC between the two subgroups using Delong's method to assess the reproducibility of the 175 models (23). Lastly, in order to describe more accurately all factors increasing 176 PCT value, we performed a logistic regression with SPSS version 15.0 software. 177

178 **RESULTS** 

#### 179 *Clinical and bacteriological description of the population*

Overall, 12485 newborns were analysed during the study period. Three of these were certainly infected (0.24‰). As shown in Figure 1, 2178 of these were suspected of having an infection and had a PCT measurement, among which 27 were excluded because of lacking data, 2151 newborns were therefore

considered in the statistical analysis and the following results only concern these newborns. Among these 2151 newborns presenting risk factors, 26 were considered as probably or certainly infected which represented an incidence of 1.2%. In the preterm population the incidence was 2%. Baseline characteristics of the selected patients are described in Table 1.

- 189
- 190

### PCT as a marker of early neonatal infection

191 Distribution of PCT value among infected and non infected patients is shown in Figure 2. PCT median in infected newborns was significantly higher than among 192 non infected patients (3.03 ng/ml vs 0.16 ng/ml, p<0.001). Considering the 193 entire study population, analysis of the ROC curve (Figure 3) determined a cut-194 off of 0.6 ng/ml as the best compromise, with an area under the curve of 0.96 195 196 (CI 95%: 0.95 - 0.98). Performing ROC curves in full-term and preterm populations we selected the same cut-off of 0.6 ng/ml. Twenty-four of the 26 197 infected children had a PCT value higher than 0.6 ng/ml, and PCT was lower 198 than 0.6 ng/ml in 2066 of the 2125 non-infected newborns. Nevertheless, among 199 the 2097 newborns presenting a negative PCT result, 2 were probably infected. 200 201 The first one was a full-term baby with a membrane rupture longer than 24 hours as the only infection risk factor, presenting respiratory distress 24 hours 202 after birth. Escherichia coli was found in gastric fluid culture, PCT was 0.22 203 ng/ml, CRP1 measured 2 hours after birth was negative but CRP2 24 hours later 204 was at 34 mg/l and antibiotics were introduced at the beginning of the 205

206 symptoms. The second newborn was also a full term baby whose mother had fever and a membrane rupture of 14 hours, with meconium stained amniotic 207 fluid. The newborn was asymptomatic with a PCT of 0.17 ng/ml and a negative 208 CRP1 at birth. CRP2 18 hours later was at 87 mg/l, antibiotic treatment was 209 210 therefore started. On the basis of the cut-off of 0.6 ng/ml, statistical values of 211 PCT established in the whole cohort and in preterm and full-term populations are expressed in Table 2. Corresponding post-test probabilities determined with 212 213 the nomogram of Bayes are shown in Figure 4. These results indicate that the probability for a newborn with an infection risk factor of being infected was 214 28% (CI 95%: 23 - 33) if the test was positive, and less than 0.001% (CI 95%: 0 215  $-1.10^{-5}$ ) if the test was negative. In the preterm group, the post-test probability 216 217 of being infected was 30% (CI 95%: 23 - 37) in the case of a positive test, and 218 0% (CI 95%: 0 - 1) in the case of a negative test.

### 219

Comparison of PCT and CRP

ROC curves of PCT, CRP1 and CRP2 are represented in Figure 3. The area under the ROC curve of CRP1 was 0.75 (CI 95%: 0.72 - 0.78), which was significantly lower than that of PCT (p=0.002). Considering the CRP2 value, the area under the ROC curve was 0.88 (0.86 - 0.91) and did not differ from that of PCT (p= 0.184) or CRP1 (p=0.056).

225 *Relationship between infection and antenatal risk factors and PCT* 

As displayed in Table 3, only chorioamnionitis was identified as a statistically significant independent antenatal risk factor of infection related with post natal diagnosis of infection (p< 0.05). PCT measured in cord blood was also independently related with infection (p<0.001).

### 230 *Analysis of 'clinical' and 'bio clinical' models*

In each model, the probability of the newborn being infected was  $1/(1+e^{-z})$ . In 231 the 'clinical' model, z was equal to 0.092 \* Gestational age (weeks) + 2.84 \* 232 clinical suspicion of chorioamnionitis (no = 0, yes = 1) + 2.3459 \* APGAR233 score at 5 minutes less than 7 (no = 0, yes = 1) - 2.1453. For the 'bio clinical' 234 model, z was equal to 0.064 \* Gestational age (weeks) + 2.29 \* clinical 235 suspicion of chorioamnionitis (no = 0, yes = 1) + 0.73 \* APGAR score at 5 236 minutes less than 7 (no = 0, yes = 1) + 0.97 log[PCT(ng/ml)] - 2.07. When the 237 two models were built by being applied to the first subgroup, the AUCs were 238 0.90 (CI 95%: 0.84 - 0.95) for the 'clinical' model and 0.98 (0.96 - 1) for the 239 'bio clinical' model, with the latter being significantly higher (p < 0.05). The 240 241 AUCs were also significantly different when the models were applied to the second subgroup for validation, being 0.70 (0.62 - 0.78) for the 'clinical' model 242 and 0.96 (0.92 - 0.99) for the 'bio clinical' model (p<0.001). A comparison of 243 the AUCs of the 'clinical' model and 'bio clinical' model between the two 244 245 subpopulations did not show any significant difference (p = 0.05 and p = 0.54, respectively) and confirmed the reproducibility of the models. AUC of the 246 'clinical' model in the whole population was not different compared to that of 247 PCT alone (p = 0.2)248

- 249
- Analysis of other factors influencing PCT value

All results of the analysis of other factors associated with a PCT value higher than 0.6 ng/ml are expressed in Table 4. Gestational age between 28 and 32 weeks, pH at birth lower than 7.10 and post-natal diagnosis of infection were independently related with a PCT value higher than 0.6 ng/ml (p<0.05).

Evaluation of the impact of the use of PCT on antibiotic administration 254 During the study period, 348 of the 2151 studied babies presented abnormal 255 clinical signs during their first 48 hours of life. Among the 1803 asymptomatic 256 257 newborns, 105 received a treatment with antibiotics, and only 29 of them had a PCT higher than 0.6 ng/ml, and only one asymptomatic newborn with PCT 258 lower than 0.6 ng/ml was a posteriori considered infected. We can conclude that 259 if antibiotic treatment set up had been decided only considering PCT value 260 among these asymptomatic newborns, it could have been avoided in 76 of them 261 262 (72%), for only one infected newborn missed.

#### 263 **DISCUSSION**

This study analysed a larger cohort of newborns than in the previous study (13) 264 and confirmed that PCT measured in umbilical cord blood is an early and 265 discriminating marker of EONI. First, its high sensitivity shows its ability to 266 267 detect early infected patients. Indeed, 24 of the 26 infected patients in our cohort had a PCT value higher than 0.6 ng/ml and only 2 of these had a negative 268 concentration. Another interesting result is the ability of this marker to detect 269 non-infected patients among those presenting risk factors of infection as 270 represented in the cohort of this study. Post-test probability determined with the 271

272 nomogram of Bayes showed that a newborn with at least one of these infection risk factors and a PCT value lower than 0.6 ng/ml had a statistical risk of being 273 infected of about 0%. This confirms the results of the preliminary study carried 274 out in a similar population with semi-quantitative dosage, where the sensitivity 275 and negative predicting values were 0.87 (CI 95%: 0.71 - 1) and 0.99 (0.98 -276 0.99), respectively. The post-test probability was 1% for a negative test (13). 277 This also confirms that PCT can be used in full-term newborns as in preterms, 278 279 with the same cut-off. In comparison, Kordek et al. conducted a study of 286 newborns and found a sensitivity of 0.80, and an NPV of 0.95 for PCT also 280 measured in cord blood with a cut-off of 1.22 ng/ml (24). 281

The main weakness of this marker, not identified in the preliminary study but 282 shown in this large cohort, is its low positive predictive value. Among the 84 283 284 children with a PCT value higher than 0.6 ng/ml, only 24 were infected. A low predicting value for positive measurement was already found in the 285 aforementioned Kordek study (24). The first explanation of these results seems 286 to be the impact of perinatal asphyxia, as shown in the logistic regression, which 287 identified a pH lower than 7.10 as an independent factor. These findings have 288 289 seldom been described previously. In a group of 117 non-infected patients, Chiesa et al. (12) did not find any significant influence of antenatal asphyxia on 290 PCT measurement. The second explanation could be the influence of antenatal 291 inflammation. Indeed, prematurity in this study increases PCT independently of 292 infection. A specific analysis of the perinatal context of these preterms with 293

increased PCT, but without diagnosed infection, shows that most had several
infection risk factors with a probable foetal inflammation response syndrome.
We can suggest the possibility that antibiotics received by the mother avoided
neonatal infection, but not labour.

As we demonstrated in the comparison of a 'clinical' model with a 'bio clinical' 298 model integrating PCT, this marker seems useful in clarifying the risk of the 299 newborn being infected at birth. This demonstrates that in this situation, the 300 301 clinical approach considering only antenatal risk factors is not sufficient for the evaluation of the risk of infection which is more precise when adding the result 302 of PCT. This prompts to recommend PCT measurement in cord blood in 303 neonates presenting risk factors but this has to be validated by prospective 304 305 studies. Thus, the next step in the use of PCT in clinical practice will be to 306 validate this marker in clinical decision rules. In the context of other pediatric infectious diseases such as meningitis (25) and febrile urinary tract infections 307 (26), PCT seems to be a useful tool for better management by the clinician. In 308 neonatology, Franz et al (27) have shown that a diagnostic algorithm based on 309 CRP and IL-8 reduced antibiotic therapy compared with a standard strategy, 310 311 without increasing the rate of missed infections. According to our results, during the study period, 72% of antibiotic treatment in asymptomatic newborns could 312 have been avoided if the decision had been guided by clinical examination and 313 PCT measurement, missing only one infected patient. This suggests that among 314 these asymptomatic patients, it should be possible to construct a reliable clinical 315

316 rule with a pivotal role for PCT in order to decrease the number of unnecessary 317 neonatal antibiotic treatments. In patients presenting clinical signs of infection, it appears, at the moment, unreasonable to introduce a treatment considering only 318 the PCT result, but this marker should be useful to stop the treatment 48 hours 319 after introduction, after collecting all bacteriological results. This could help 320 limit the number of newborn hospitalisations, to promote parent-infant bonding, 321 and diminish healthcare costs. Moreover, such an antibiotic treatment limitation 322 323 could reduce the adverse effects on gut microbiota and subsequent consequences on future child and adult health. 324

Lastly, as PCT seems to be a reliable infection marker, we can speculate on its ability to be a prognostic marker in order to evaluate early the predicted consequences of neonatal inflammation. Indeed, the relationship between neonatal inflammation, caused among other factors by perinatal infection, and pulmonary and cerebral morbidity has been well described for about ten years (28-36).

In conclusion, our study confirms the ability of PCT measured in umbilical cord blood to detect early neonatal infection, and also to determine non-infected newborns among those presenting infection risk factors. This should allow the neonatologist to integrate this marker into clinical decision rules in order to reduce unnecessary antibiotic treatment during the first days of life.

# **BIBLIOGRAPHY**

1.	Puopolo KM, Madoff LC, Eichenwald EC. Early-onset group B
	streptococcal disease in the era of maternal screening. Pediatrics. 2005
	May;115(5):1240-6.
2.	Ohlsson A, Shah VS. Intrapartum antibiotics for known maternal Group
	B streptococcal colonization. Cochrane Database Syst Rev. 2009 Jul
	8;(3):CD007467.
3.	Trends in perinatal group B streptococcal disease - United States, 2000-
	2006. Centers for Disease Control and Prevention (CDC).MMWR Morb
	Mortal Wkly Rep. 2009 Feb 13;58(5):109-12.
4.	The "ins and outs" of neonatal sepsis. Polin RA. J Pedatrics
	2003;143(1):3-4.
5.	Stocker M, Fontana M, El Helou S, Wegscheider K and Merger TM. Use
	of procalcitonin-guided decision-making to shorten antibiotic therapy in
	suspected neonatal early-onset sepsis : prospective randomised
	intervention trial. Neonatology 2010;97:165-74.
6.	Penders J, Thijs C, van den Brandt PA, Kummeling I, Snijders B, Stelma
	F, Adams H, van Ree R, Stobberingh EE. Gut microbiota composition
	<ol> <li>1.</li> <li>2.</li> <li>3.</li> <li>4.</li> <li>5.</li> <li>6.</li> </ol>

354	and development of atopic manifestations in infancy: the KOALA Birth
355	Cohort Study. Gut. 2007 May;56(5):661-7
356	7. Neu J. Perinatal and neonatal manipulation of the intestinal microbiome:
357	a note of caution. Review. Nutr Rev. 2007 Jun;65(6 Pt 1):282-5.
358	8. Van den Hoogen A, Gerards LJ, Verboon-Maciolek MA, Fleer A,
359	Krediet TG. Long-Term Trends in the Epidemiology of Neonatal Sepsis
360	and Antibiotic Susceptibility of Causative Agents. Neonatology. 2009
361	Jul 2;97(1):22-28.
362	9. Escobar GJ. The neonatal sepsis "work-up". Personal reflections on the
363	development of an evidence-based approach toward newborn infections
364	in a managed care organization. Pediatrics 1999;103 (1 suppl E):360-73.
365	10. Resch B, Gusenleitner W, Muller WD. Procalcitonin and interleukine 6
366	in the diagnosis of early-onset sepsis of the neonate. Acta paediatr.
367	2003;92(2):243-5
368	11. Gendrel D, Assicot M, Raymond J, Moulin F, Francoual C, Badoual J,
369	Bohuon C. Procalcitonin as a marker for the early diagnosis of neonatal
370	infection. J Pediatrics 1996;128:570-3
271	12 Chiago C. Daparo A. Dagoi N. Stagagno M. Da Ciusti M. Osharra IE
3/1	12. Uniesa C, Fanero A, Rossi IN, Stegagno IVI, De Glusti IVI, Osdom JF,
372	Pacifico L. Reliability of procalcitonin concentrations for the diagnosis

- of sepsis in critically ill neonates. Clin Infect Dis. 1998 Mar;26(3):66472.
- 375 13. Joram N, Boscher C, Denizot S, Loubersac V, Winer N, Roze JC, Gras376 Le Guen C. Umbilical cord blood procalcitonin and C reactive protein
  377 concentrations as markers for early diagnosis of very early onset
  378 neonatal infection. Arch Dis Child Fetal Neonatal Ed. 2006
  379 Jan;91(1):F65-6.
- 14. Bossuyt PM, Reitsma JB, Bruns DE, Gatsonis CA, Glasziou PP, Irwig
  LM, Lijmer JG, Moher D, Rennie D, de Vet HC; STARD group.
  Towards complete and accurate reporting of studies of diagnostic
  accuracy: the STARD initiative. Ann Clin Biochem. 2003 Jul;40(Pt
  4):357-63.
- 385 15. ANAES. Diagnostic et traitement curatif de l'infection bactérienne
  386 précoce du nouveau né. September 2002
- 16. Revised guidelines for prevention of early-onset group B streptococcal
  (GBS) infection. American Academy of Pediatrics Committee on
  Infectious Diseases and Committee on Fetus and Newborn. Pediatrics.
  1997 Mar;99(3):489-96.

391	17. Oddie S, Embleton ND. Risk factors for early onset neonatal group B
392	streptococcal sepsis: case-control study. BMJ. 2002 Aug
393	10;325(7359):308.
394	18. ANAES. Prévention anténatale du risque infectieux bactérien néonatal
395	précoce. September 2001
396	19. Haque KN. Definitions of bloodstream infection in the newborn. Pediatr
397	Crit Care Med. 2005 May;6(3 Suppl):S45-9. Review.
398	20. Goldstein B, Giroir B, Randolph A; International Consensus Conference
399	on Pediatric Sepsis. International pediatric sepsis consensus conference:
400	definitions for sepsis and organ dysfunction in pediatrics. Pediatr Crit
401	Care Med. 2005 Jan;6(1):2-8. Review
402	21. Page J, Attia J Using Bayes' nomogram to help interpret odds ratios.
403	Evid. Based Med. 2003;8;132-134
404	22. Hanley JA, McNeil BJ The meaning and use of the area under a receiver
405	operating characteristic (ROC) curve. Radiology. 1982 Apr;143(1):29-36.
406	23. Carsten S, Wesseling S, Schink T, and Jung K. Comparison of Eight
407	Computer Programs for Receiver-Operating Characteristic Analysis.
408	Clinical Chemistry 2003;49: 433–9

409	24. Kordek A, Hałasa M, Podraza W. Early detection of an early onset
410	infection in the neonate based on measurements of procalcitonin and C-
411	reactive protein concentrations in cord blood. Clin Chem Lab Med.
412	2008;46(8):1143-8
413	25. Dubos F, Moulin F, Raymond J, Gendrel D, Bréart G, Chalumeau M.
414	Distinction between bacterial and aseptic meningitis in children:
415	refinement of a clinical decision rule. Arch Pediatr. 2007
416	May;14(5):434-8. Epub 2007 Jan 26. French.
417	26. Leroy S, Romanello C, Galetto-Lacour A, Smolkin V, Korczowski B,
418	Rodrigo C, Tuerlinckx D, Gajdos V, Moulin F, Contardo M, Gervaix A,
419	Halevy R, Duhl B, Prat C, Borght TV, Foix-l'Hélias L, Dubos F, Gendrel
420	D, Bréart G, Chalumeau M. Procalcitonin to reduce the number of
421	unnecessary cystographies in children with a urinary tract infection: a
422	European validation study. J Pediatr. 2007 Jan;150(1):89-95.
423	27. Franz AR, Bauer K, Schalk A, Garland SM, Bowman ED, Rex K,
424	Nyholm C, Norman M, Bougatef A, Kron M, Mihatsch WA, Pohlandt F;
425	International IL-8 Study Group. Measurement of interleukin 8 in
426	combination with C-reactive protein reduced unnecessary antibiotic
427	therapy in newborn infants: a multicenter, randomized, controlled trial.

428 Pediatrics. 2004 Jul;114(1):1-8. Erratum in: Pediatrics. 2004
429 Dec;114(6):1746

430 28. Gomez R, Romero R, Ghezzi F, Yoon BH, Mazor M, Berry SM.The
431 fetal inflammatory response syndrome. Am J Obstet Gynecol. 1998
432 Jul;179(1):194-202.

29. Yoon BH, Romero R, Kim KS, Park JS, Ki SH, Kim BI, Jun JK. A 433 434 systemic fetal inflammatory response and development of J 1999 435 bronchopulmonary dysplasia. Am Obstet Gynecol. Oct:181(4):773-9 436

437 30. Yoon BH, Park CW, Chaiworapongsa T. Intrauterine infection and the
438 development of cerebral palsy. BJOG. 2003 Apr;110 Suppl 20:124-7.

439 31. Ghezzi F, Gomez R, Romero R, Yoon BH, Edwin SS, David C, Janisse
440 J, Mazor M.Elevated interleukin-8 concentrations in amniotic fluid of
441 mothers whose neonates subsequently develop bronchopulmonary
442 dysplasia. Eur J Obstet Gynecol Reprod Biol. 1998 May;78(1):5-10.

32. Nelson KB, Grether JK, Dambrosia JM, Walsh E, Kohler S,
Satyanarayana G, Nelson PG, Dikens BF, Phillips TM. Neonatal
cytokines and cerebral palsy in very preterm infants. Pediatric research
2003;53(4):600-7

33. Duggan PJ, Maalouf EF, Watts TL, Sullivan MH, Counsell SJ, Allsop J,
Al-Nakib L, Rutherford MA, Battin M, Roberts I, Edwards AD.
Intrauterine T-cell activation and increased proinflammatory cytokine
concentrations in preterm infants with cerebral lesions. Lancet. 2001
Nov 17;358(9294):1699-700.

- 452 34. Paananen R, Husa AK, Vuolteenaho R, Herva R, Kaukola T, Hallman
  453 M. Blood cytokines during the perinatal period in very preterm infants:
  454 relationship of inflammatory response and bronchopulmonary dysplasia
  455 J Pediatr. 2009 Jan;154(1):A2.
- 456 35. Hatherill M, Tibby SM, Turner C, Ratnavel N, Murdoch IA.
  457 Procalcitonin and cytokine levels: relationship to organ failure and
  458 mortality in pediatric septic shock. Crit Care Med. 2000 Jul;28(7):2591459 4.
- 460 36. Van der Kaay DC, De Kleijn ED, De Rijke YB, Hop WC, De Groot R,
  461 Hazelzet JA. Procalcitonin as a prognostic marker in meningococcal
  462 disease. Intensive Care Med. 2002 Nov;28(11):1606-12. Epub 2002 Oct
  463 4
- 464

465	Legends
466	
467	Figure 1 : Study flow diagram
468	
469	Figure 2 : Box-and whisker plot showing PCT concentration according to the
470	infectious status. PCT level is expressed in ng/ml. Box represent first and third
471	quartiles, horizontal lines represent medians with the corresponding numerical
472	value, whiskers give 10/90 percentiles. Open circle represents oulier. Dotted line
473	represents the cut-off of 0,6 ng/ml. p represents the comparison of medians
474	
475	Figure 3 : A. Receiver Operating Characteristics (ROC) curves of PCT, CRP1
476	and CRP2 in the whole population. B. Areas under the ROC curve of PCT,
477	CRP1 and CRP2 in the whole population with confident interval 95%
478	
479	Figure 4 : Nomogram of Bayes representing positive and negative post test
480	probability according to pre test probability and positive and negative likelihood
481	ratios in the whole and preterms populations. In the whole population pre test
482	probability was 1.2% and post test probability was 28% in case of positive test
483	and < 0.001% in case of negative test. Among preterms, pre test probability was
484	2% and post test probability was 30% in case of positive test and $< 0.001\%$ in
485	case of negative test
486	

Characteristic	All ne	wborns	Full	-terms	Preter	ms (<37
	(n=	2151)	(n=	1339)	G	iW)
	· ·	,		,	(n=	=812)
Mean Gestational age (SD)	36.9	(3.8)	39.4	(1.2)	32.7	(2.9)
(weeks)						
<28 weeks	52	(2.4)				
28-32 weeks	248	(11.5)				
32-36 weeks	512	(23.8)				
>36 weeks	1339	(62.3)				
Male sex	1141	(53)	684	(51)	446	(55)
Mean birth weight (SD) (g)	2783	(843.8)	329	(453.8)	2394	(649.2)
<1000 g	72	(3.3)	0	(0)	68	(8.4)
1001-1500 g	151	(7)	0	(0)	149	(18.3)
1501-2500g	472	(21.9)	59	(4.4)	415	(51.1)
>2500 g	1456	(67.7)	1280	(95.6)	180	(22.2)
Prenatal corticosteroids	423	(19.7)	19	(1.4)	404	(49.8)
exposure						
Gastric fluid colonisation	312	(14.5)	208	(15.5)	102	(12.6)
Membrane rupture >24h	578	(26.9)	348	(26)	232	(28.6)
Caesarean	691	(32.1)	276	(20.6)	414	(51)
Antenatal antibiotics exposure	699	(32.5)	409	(30.5)	290	(35.7)
Chorioamnionitis	82	(3.8)	10	(0.7)	72	(8.8)
Pre-eclampsia	164	(7.6)	42	(3.1)	122	(15)
APGAR score <7 at 5 min	61	(2.8)	14	(1)	47	(5.8)
pH ,at birth –mean	7.24		7.23		7.26	
<7.09	91	(4.2)	61	(4.6)	30	(3.7)
7.09-7.14	118	(5.5)	94	(7)	24	(3)
>7.14	1945	(90.4)	1187	(88.6)	758	(93.3)
Intrauterine growth retardation	136	(6.3)	28	(2.1)	108	(13.3)
Death before discharge from the	28	(1.3)	3	(0.2)	25	(3.1)
hospital						
Multiple pregnancy	281	(13.1)	17	(1.3)	264	(32.5)
Infection	26	(1.2)	9	(0.7)	17	(2.1)
Certain	3	(0.1)	1	(0.1)	2	(0.2)
Probable	23	(1.1)	8	(0.6)	15	(1.8)

487 Tab 1 : Baseline characteristics of patients suspect of infection.

488 Results are numbers (percentages) unless stated otherwise489

Whole		Whole population		Preterms		Fullterms	
population							
Sensitivity	0.9	(0.75 - 0.98)	1	(0.82 - 1)	0.78	(0.45 -	
Specificity	0.9 7	(0.96 - 0.98)	0.9	(0.93 -	0.98	(0.94) (0.97 - 0.99)	
Negative predictive value	0.9 9	(0.99 - 0.99)	1	(0.99 - 1)	0.99	(0.99 - 1)	
Positive predictive value	0.2 8	(0.2 - 0.36)	0.3	(0.3 - 0.31)	0.23	(0.18 - 0.28)	
Positive likelihood ratio	32	(24 - 41)	20	(14 - 27)	45	(26 - 77)	
Negative likelihood ratio	$\begin{array}{c} 0.0 \\ 8 \end{array}$	(0.02 - 0.3)	0	(0 - 0.45)	0.23	(0.07 - 0.8)	

# 491 Table 2. Diagnosis values of PCT according to the groups of term.

492 Values are percentages (95% CI)

Parameters	Crude OR (95%	р	Adjusted OR	р
	CI)		(95% CI)	
Chorioamnionitis	25.5 (10.6 -	< 0.001	8.3 (2.3 - 29.8)	0.001
	61.3)			
Intrapartum fever	1.8 (0.8 - 2.3)	0.07	1.12 (0.5 - 3.1)	0.2
Spontaneous delivery < 37	2.6 (0.6 - 10.9)	0.2	1 (0.9 - 1.1)	0.9
GW				
Length of membrane rupture >	2.4 (1 - 5.5)	0.04	1.16 (0.4 - 3.7)	0.8
12h				
Maternal colonization with	0.84 (0.2 - 2.94)	0.5	0.8 (0.2 - 3.9)	0.8
streptococcus B				
Fetal asphyxia	13.4 (4.9 - 35.3)	< 0.001	2.6 (0.7 - 10.2)	0.2
PCT > 0.6 ng/ml	399.3 (88.6 -	< 0.001	257.6 (57.2 -	< 0.001
-	2501.9)		1159.1)	

Table 3 : Analysis of factors related with infection diagnosis by univariate
 analysis and multivariate analysis by logistic regression in the whole population.

496 Values are odds ratios with 95% confident interval.

497

499 Table 4 : Analysis of factors increasing PCT value in cord blood by univariate 500 analysis and multivariate analysis by logistic regression in the whole population

analysis and multivariate	e analysis by logisti	c regress	ion in the whole pop	ulation.
Parameters	Crude OR (95%	р	Adjusted OR (95%	р
	CI)		CI)	
Gestational age <28 weeks	11.7 (5.5 - 25)	< 0.001	6.2 (0.8 - 48.9)	0.06
Gestational age 28-32 weeks	4.9 (2.8 - 8.5)	< 0.001	4.4 (1.2 - 16.2)	0.02
Gestational age 32-36 weeks	1.9 (1.1 - 3.3)	0.03	1.9 (0.8 - 4.6)	0.11
Birth weight<1000 g-	5.7 (2.7 - 11.9)	< 0.001	0.7 (0.1 - 5.6)	0.69
Birth weight 1001-1500	3.9 (2.1 - 7.3)	< 0.001	0.6 (0.2 - 2.5)	0.57
g-				
Birth weight 1501-	1.7 (1 - 2.9)	0.4	0.9(0.3 - 2.3)	0.81
2500g-				
Intra uterine growth retardation	0.3 (0.1 - 1.4)	0.13	0.2(0.05 - 1.2)	0.08
Preeclampsia	0.4 (0.1 - 1.35)	0.15	0.3 (0.1 - 1.2)	0.07
pH < 7.10	2.4 (1.1 - 5.1)	0.02	2.9 (1.1 - 7.4)	0.04
pH 7.10–7.14	0.6 (0.2 - 2.1)	0.4	0.6 (0.1 - 2.7)	0.59
Apgar <7 at 5 min	8.1 (4.3 - 15.3)	< 0.001	2.1 (0.8 - 5.6)	0.07
Multiple pregnancy	1.2 (0.6 - 2.2)	0.6	0.6 (0.3 - 1.3)	0.16
Infection	393.5 (91 -	< 0.001	304.5 (63.7 -	< 0.001
	1701.5)		1456.3)	
Prenatal corticosteroids exposure	2.9 (1.9-4.6)	< 0.001	1.8 (0.8-3.9)	0.12

501 Values are odds ratios with 95% confident interval.

502

503

505	Legends
506	
507	Figure 1 : Study flow diagram
508	
509	Figure 2 : Box-and whisker plot showing PCT concentration according to the
510	infectious status. PCT level is expressed in ng/ml. Box represent first and third
511	quartiles, horizontal lines represent medians with the corresponding numerical
512	value, whiskers give 10/90 percentiles. Open circle represents oulier. Dotted line
513	represents the cut-off of 0,6 ng/ml. p represents the comparison of medians
514	
515	Figure 3 : A. Receiver Operating Characteristics (ROC) curves of PCT, CRP1
516	and CRP2 in the whole population. B. Areas under the ROC curve of PCT,
517	CRP1 and CRP2 in the whole population with confident interval 95%
518	
519	Figure 4 : Nomogram of Bayes representing positive and negative post test
520	probability according to pre test probability and positive and negative likelihood
521	ratios in the whole and preterms populations. In the whole population pre test
522	probability was 1.2% and post test probability was 28% in case of positive test
523	and $< 0.001\%$ in case of negative test. Among preterms, pre test probability was
524	2% and post test probability was 30% in case of positive test and $< 0.001\%$ in
525	case of negative test
526	

Characteristic	All ne	wborns	Full	-terms	Preter	ms (<37
	(n=	2151)	(n=	1339)	G	W)
					(n=	=812)
Mean Gestational age (SD)	36.9	(3.8)	39.4	(1.2)	32.7	(2.9)
(weeks)						
<28 weeks	52	(2.4)				
28-32 weeks	248	(11.5)				
32-36 weeks	512	(23.8)				
>36 weeks	1339	(62.3)				
Male sex	1141	(53)	684	(51)	446	(55)
Mean birth weight (SD) (g)	2783	(843.8)	329	(453.8)	2394	(649.2)
<1000 g	72	(3.3)	0	(0)	68	(8.4)
1001-1500 g	151	(7)	0	(0)	149	(18.3)
1501-2500g	472	(21.9)	59	(4.4)	415	(51.1)
>2500 g	1456	(67.7)	1280	(95.6)	180	(22.2)
Prenatal corticosteroids	423	(19.7)	19	(1.4)	404	(49.8)
exposure						
Gastric fluid colonisation	312	(14.5)	208	(15.5)	102	(12.6)
Membrane rupture >24h	578	(26.9)	348	(26)	232	(28.6)
Caesarean	691	(32.1)	276	(20.6)	414	(51)
Antenatal antibiotics exposure	699	(32.5)	409	(30.5)	290	(35.7)
Chorioamnionitis	82	(3.8)	10	(0.7)	72	(8.8)
Pre-eclampsia	164	(7.6)	42	(3.1)	122	(15)
APGAR score <7 at 5 min	61	(2.8)	14	(1)	47	(5.8)
pH ,at birth –mean	7.24		7.23		7.26	
<7.09	91	(4.2)	61	(4.6)	30	(3.7)
7.09-7.14	118	(5.5)	94	(7)	24	(3)
>7.14	1945	(90.4)	1187	(88.6)	758	(93.3)
Intrauterine growth retardation	136	(6.3)	28	(2.1)	108	(13.3)
Death before discharge from the	28	(1.3)	3	(0.2)	25	(3.1)
hospital						
Multiple pregnancy	281	(13.1)	17	(1.3)	264	(32.5)
Infection	26	(1.2)	9	(0.7)	17	(2.1)
Certain	3	(0.1)	1	(0.1)	2	(0.2)
Probable	23	(1.1)	8	(0.6)	15	(1.8)

527 Tab 1 : Baseline characteristics of patients suspect of infection.

528 Results are numbers (percentages) unless stated otherwise

<sup>529</sup> 

Whole	Wh	Whole population		Preterms		Fullterms	
population							
Sensitivity	0.9	(0.75 - 0.98)	1	(0.82 - 1)	0.78	(0.45 - 0.94)	
Specificity	0.9 7	(0.96 - 0.98)	0.9 5	(0.93 - 0.96)	0.98	(0.97 - 0.99)	
Negative predictive value	, 0.9 9	(0.99 - 0.99)	1	(0.99 - 1)	0.99	(0.99 - 1)	
Positive predictive value	0.2 8	(0.2 - 0.36)	0.3	(0.3 - 0.31)	0.23	(0.18 - 0.28)	
Positive likelihood ratio	32	(24 - 41)	20	(14 - 27)	45	(26 - 77)	
Negative likelihood ratio	$\begin{array}{c} 0.0 \\ 8 \end{array}$	(0.02 - 0.3)	0	(0 - 0.45)	0.23	(0.07 - 0.8)	

531 Table 2. Diagnosis values of PCT according to the groups of term.

532 Values are percentages (95% CI)

analysis and multivariate analysis by logistic regression in the whole population.								
Parameters	Crude OR (95%	р	Adjusted OR	р				
	CI)		(95% CI)					
Chorioamnionitis	25.5 (10.6 -	< 0.001	8.3 (2.3 - 29.8)	0.001				
	61.3)							
Intrapartum fever	1.8 (0.8 - 2.3)	0.07	1.12 (0.5 - 3.1)	0.2				
Spontaneous delivery < 37	2.6 (0.6 - 10.9)	0.2	1 (0.9 - 1.1)	0.9				
GW								
Length of membrane rupture >	2.4 (1 - 5.5)	0.04	1.16 (0.4 - 3.7)	0.8				
12h								
Maternal colonization with	0.84 (0.2 - 2.94)	0.5	0.8 (0.2 - 3.9)	0.8				
streptococcus B								
Fetal asphyxia	13.4 (4.9 - 35.3)	< 0.001	2.6 (0.7 - 10.2)	0.2				
PCT > 0.6  ng/ml	399.3 (88.6 -	< 0.001	257.6 (57.2 -	< 0.001				
	2501.9)		1159.1)					

Table 3 : Analysis of factors related with infection diagnosis by univariate analysis and multivariate analysis by logistic regression in the whole population.

536 Values are odds ratios with 95% confident interval.

537

Table 4 : Analysis of factors increasing PCT value in cord blood by univariate analysis and multivariate analysis by logistic regression in the whole population

analysis and multivariate	analysis by logisti	c regress	ion in the whole pop	ulation.
Parameters	Crude OR (95%	р	Adjusted OR (95%	р
	CI)		CI)	
Gestational age <28 weeks	11.7 (5.5 - 25)	< 0.001	6.2 (0.8 - 48.9)	0.06
Gestational age 28-32 weeks	4.9 (2.8 - 8.5)	< 0.001	4.4 (1.2 - 16.2)	0.02
Gestational age 32-36 weeks	1.9 (1.1 - 3.3)	0.03	1.9 (0.8 - 4.6)	0.11
Birth weight<1000 g-	5.7 (2.7 - 11.9)	< 0.001	0.7 (0.1 - 5.6)	0.69
Birth weight 1001-1500	3.9 (2.1 - 7.3)	< 0.001	0.6 (0.2 - 2.5)	0.57
g-				
Birth weight 1501-	1.7 (1 - 2.9)	0.4	0.9(0.3 - 2.3)	0.81
2500g-				
Intra uterine growth retardation	0.3 (0.1 - 1.4)	0.13	0.2(0.05 - 1.2)	0.08
Preeclampsia	0.4 (0.1 - 1.35)	0.15	0.3 (0.1 - 1.2)	0.07
pH < 7.10	2.4 (1.1 - 5.1)	0.02	2.9 (1.1 - 7.4)	0.04
рН 7.10–7.14	0.6 (0.2 - 2.1)	0.4	0.6 (0.1 - 2.7)	0.59
Apgar <7 at 5 min	8.1 (4.3 - 15.3)	< 0.001	2.1 (0.8 - 5.6)	0.07
Multiple pregnancy	1.2 (0.6 - 2.2)	0.6	0.6 (0.3 - 1.3)	0.16
Infection	393.5 (91 -	< 0.001	304.5 (63.7 -	< 0.001
	1701.5)		1456.3)	
Prenatal corticosteroids exposure	2.9 (1.9-4.6)	< 0.001	1.8 (0.8-3.9)	0.12

541 Values are odds ratios with 95% confident interval.

542

543

### 545 Legends

546 547 Figure 1 : Study flow diagram

548549 Figure 2 : Box-and whisker plot showing PCT concentration according to the infectious

550 status. PCT level is expressed in ng/ml. Box represent first and third quartiles, horizontal lines

- represent medians with the corresponding numerical value, whiskers give 10/90 percentiles.
- 552 Open circle represents oulier. Dotted line represents the cut-off of 0,6 ng/ml. p represents the
- 553 comparison of medians
- 554 555 Figure 3 : A. Receiver Operating Characteristics (ROC) curves of PCT, CRP1 and CRP2 in

the whole population. B. Areas under the ROC curve of PCT, CRP1 and CRP2 in the whole

- 557 population with confident interval 95%
- 558

559 Figure 4 : Nomogram of Bayes representing positive and negative post test probability

- 560 according to pre test probability and positive and negative likelihood ratios in the whole and
- 561 preterms populations. In the whole population pre test probability was 1.2% and post test
- 562 probability was 28% in case of positive test and < 0.001% in case of negative test. Among
- 563 preterms, pre test probability was 2% and post test probability was 30% in case of positive
- test and < 0.001% in case of negative test
- 565

566	Tab 1 :	Baseline	characteris	tics of	patients	suspect	of infection.
-----	---------	----------	-------------	---------	----------	---------	---------------

Characteristic	All newborns		Full-terms		Preterms (<37 GW)	
	(n=	2151)	(n=1339)		(n=812)	
Mean Gestational age (SD) (weeks)	36.9	(3.8)	39.4	(1.2)	32.7	(2.9)
<28 weeks	52	(2.4)				
28-32 weeks	248	(11.5)				
32-36 weeks	512	(23.8)				
>36 weeks	1339	(62.3)				
Male sex	1141	(53)	684	(51)	446	(55)
Mean birth weight (SD) (g)	2783	(843.8)	329	(453.8)	2394	(649.2)
<1000 g	72	(3.3)	0	(0)	68	(8.4)
1001-1500 g	151	(7)	0	(0)	149	(18.3)
1501-2500g	472	(21.9)	59	(4.4)	415	(51.1)
>2500 g	1456	(67.7)	1280	(95.6)	180	(22.2)
Prenatal corticosteroids exposure	423	(19.7)	19	(1.4)	404	(49.8)
Gastric fluid colonisation	312	(14.5)	208	(15.5)	102	(12.6)
Membrane rupture >24h	578	(26.9)	348	(26)	232	(28.6)
Caesarean	691	(32.1)	276	(20.6)	414	(51)
Antenatal antibiotics exposure	699	(32.5)	409	(30.5)	290	(35.7)
Chorioamnionitis	82	(3.8)	10	(0.7)	72	(8.8)
Pre-eclampsia	164	(7.6)	42	(3.1)	122	(15)
APGAR score <7 at 5 min	61	(2.8)	14	(1)	47	(5.8)
pH ,at birth –mean	7.24		7.23		7.26	
<7.09	91	(4.2)	61	(4.6)	30	(3.7)
7.09-7.14	118	(5.5)	94	(7)	24	(3)
>7.14	1945	(90.4)	1187	(88.6)	758	(93.3)
Intrauterine growth retardation	136	(6.3)	28	(2.1)	108	(13.3)
Death before discharge from the hospital	28	(1.3)	3	(0.2)	25	(3.1)
Multiple pregnancy	281	(13.1)	17	(1.3)	264	(32.5)
Infection	26	(1.2)	9	(0.7)	17	(2.1)
Certain	3	(0.1)	1	(0.1)	2	(0.2)
Probable	23	(1.1)	8	(0.6)	15	(1.8)

Results are numbers (percentages) unless stated otherwise

Whole	Wh	ole population		Preterms		Fullterms	
population							
Sensitivity	0.92	(0.75 - 0.98)	1	(0.82 - 1)	0.78	(0.45 - 0.94	
Specificity	0.97	(0.96 - 0.98)	0.95	(0.93 - 0.96)	0.98	(0.97 - 0.99)	
Negative predictive value	0.99	(0.99 - 0.99)	1	(0.99 - 1)	0.99	(0.99 - 1)	
Positive predictive value	0.28	(0.2 - 0.36)	0.3	(0.3 - 0.31)	0.23	(0.18 - 0.28)	
Positive likelihood ratio	32	(24 - 41)	20	(14 - 27)	45	(26 - 77)	
Negative likelihood ratio	0.08	(0.02 - 0.3)	0	(0 - 0.45)	0.23	(0.07 - 0.8)	

#### Table 2 Diagnosis values of PCT according to the groups of term 570

571 572 Values are percentages (95% CI)

Table 3 : Analysis of factors related with infection diagnosis by univariate analysis and
 multivariate analysis by logistic regression in the whole population.

+ multivariae analysis by logistic regression in the whole population.								
Parameters	Crude OR (95% CI)	р	Adjusted OR (95% CI)	р				
Chorioamnionitis	25.5 (10.6 - 61.3)	< 0.001	8.3 (2.3 - 29.8)	0.001				
Intrapartum fever	1.8 (0.8 - 2.3)	0.07	1.12 (0.5 - 3.1)	0.2				
Spontaneous delivery < 37 GW	2.6 (0.6 - 10.9)	0.2	1 (0.9 - 1.1)	0.9				
Length of membrane rupture > 12h	2.4 (1 - 5.5)	0.04	1.16 (0.4 - 3.7)	0.8				
Maternal colonization with streptococcus B	0.84 (0.2 - 2.94)	0.5	0.8 (0.2 - 3.9)	0.8				
Fetal asphyxia	13.4 (4.9 - 35.3)	< 0.001	2.6 (0.7 - 10.2)	0.2				
PCT > 0.6 ng/ml	399.3 (88.6 - 2501.9)	< 0.001	257.6 (57.2 - 1159.1)	< 0.001				

575 Values are odds ratios with 95% confident interval.

576

Table 4 : Analysis of factors increasing PCT value in cord blood by univariate analysis and
 multivariate analysis by logistic regression in the whole population.

Parameters	Crude OR (95% CI)	р	Adjusted OR (95% CI)	р
Gestational age <28 weeks	11.7 (5.5 - 25)	< 0.001	6.2 (0.8 - 48.9)	0.06
Gestational age 28-32 weeks	4.9 (2.8 - 8.5)	< 0.001	4.4 (1.2 - 16.2)	0.02
Gestational age 32-36 weeks	1.9 (1.1 - 3.3)	0.03	1.9 (0.8 - 4.6)	0.11
Birth weight<1000 g-	5.7 (2.7 - 11.9)	< 0.001	0.7 (0.1 - 5.6)	0.69
Birth weight 1001-1500 g-	3.9 (2.1 - 7.3)	< 0.001	0.6 (0.2 - 2.5)	0.57
Birth weight 1501-2500g-	1.7 (1 - 2.9)	0.4	0.9(0.3 - 2.3)	0.81
Intra uterine growth retardation	0.3 (0.1 - 1.4)	0.13	0.2(0.05 - 1.2)	0.08
Preeclampsia	0.4 (0.1 - 1.35)	0.15	0.3 (0.1 - 1.2)	0.07
pH < 7.10	2.4 (1.1 - 5.1)	0.02	2.9 (1.1 - 7.4)	0.04
pH 7.10–7.14	0.6 (0.2 - 2.1)	0.4	0.6 (0.1 - 2.7)	0.59
Apgar <7 at 5 min	8.1 (4.3 - 15.3)	< 0.001	2.1 (0.8 - 5.6)	0.07
Multiple pregnancy	1.2 (0.6 - 2.2)	0.6	0.6 (0.3 - 1.3)	0.16
Infection	393.5 (91 - 1701.5)	< 0.001	304.5 (63.7 - 1456.3)	< 0.001
Prenatal corticosteroids exposure	2.9 (1.9-4.6)	< 0.001	1.8 (0.8-3.9)	0.12

580 Values are odds ratios with 95% confident interval.