

Supplementary Table SI. Bell's modified criteria for necrotizing enterocolitis [13]

Stage	Systemic signs	Abdominal signs	Radiographic signs	Treatment
IA Suspected	Temperature instability, apnea, bradycardia, lethargy	Gastric retention, abdominal distention, emesis, heme-positive stool	Normal or intestinal dilation, mild ileus	NPO, antibiotics × 3 days
IB Suspected	Same as above	Grossly bloody stool	Same as above	Same as IA
IIA Definite, mildly ill	Same as above	Same as above, plus absent bowel sounds with or without abdominal tenderness	Intestinal dilation, ileus, pneumatosis intestinalis	NPO, antibiotics × 7 to 10 days
IIB Definite, moderately ill	Same as above, plus mild metabolic acidosis and thrombocytopenia	Same as above, plus absent bowel sounds, definite tenderness, with or without abdominal cellulitis or right lower quadrant mass	Same as IIA, plus ascites	NPO, antibiotics × 14 days
IIIA Advanced, severely ill, intact bowel	Same as IIB, plus hypotension, bradycardia, severe apnea, combined respiratory and metabolic acidosis, DIC, and neutropenia	Same as above, plus signs of peritonitis, marked tenderness, and abdominal distention	Same as IIA, plus ascites	NPO, antibiotics × 14 days, fluid resuscitation, inotropic support, ventilator therapy, paracentesis
IIIB Advanced, severely ill, perforated bowel	Same as IIIA	Same as IIIA	Same as above, plus pneumoperitoneum	Same as IIIA, plus surgery

DIC – disseminated intravascular coagulation, NPO – “nil per os” or nothing by mouth.

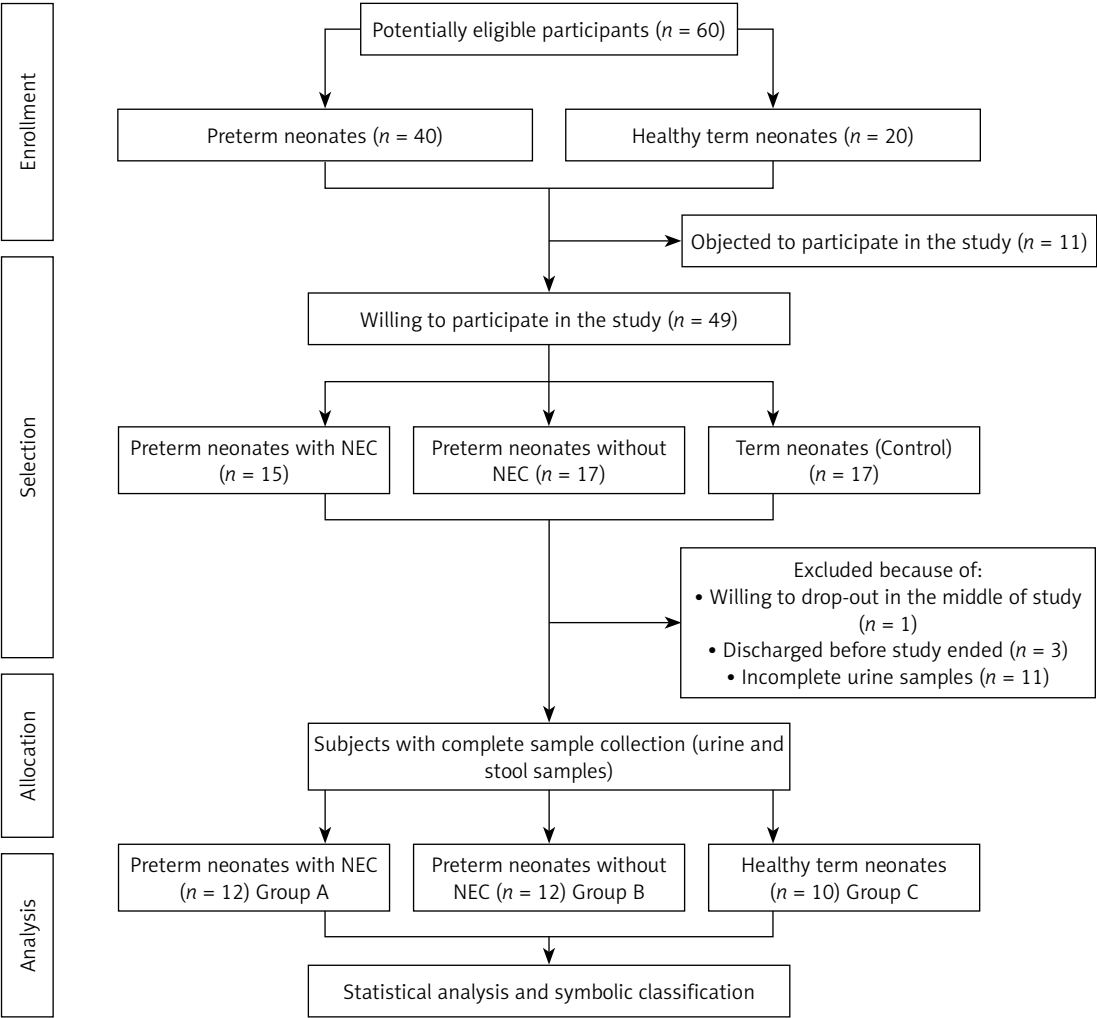
Supplementary Table SII. Primer sequences for qPCR intestinal microbiota

Species	Primer name	Primer sequence (5' to 3')	Annealing temp [°C]	Amplicon size [bp]	Reference (number)
<i>Klebsiella</i> spp. 16s rRNA	<i>Kleb-F</i>	AGCACAGAGAGCTTG	55	127	(Kurupati <i>et al.</i> , 2004) [37]
	<i>Kleb-R</i>	ACTTTGGTCTTGCGAC			
<i>Escherichia coli</i> Uropathogenic (UPEC) <i>hlyD</i> gene	<i>hlyD-F</i>	ACAGGCTTCAGTAATCAG	54	173	(Enderle <i>et al.</i> , 2014) [38]
	<i>hlyD-R</i>	CGACGTTAATAAAACCAATATC			
<i>Pseudomonas</i> spp. 16s rRNA	<i>Pseudo-F</i>	ACTTTAAGTTGGGAGGAAGGG	60	252	(Bergmark <i>et al.</i> , 2012) [39]
	<i>Pseudo-R</i>	ACACAGGAAATCCACCACCC			
<i>Lactobacillus</i> spp. 16s rRNA	<i>Lacto-F</i>	TGGAAACAGRTGCTAATACCG	62	233	(Byun <i>et al.</i> , 2004) [40]
	<i>Lacto-R</i>	GTCCATTGTGGAAGATTCCC			
<i>Clostridium</i> spp. 16s rRNA	<i>Clost-F</i>	TACCHRAGGAGGAAGCCAC	55	200	(Song <i>et al.</i> , 2004) [41]
	<i>Clost-R</i>	GTTCTTCCTAATCTCTACGCAT			
Genus <i>Bacteroides</i> 16s rRNA	<i>Bctd-F</i>	CGTTCATTAGGCAGTTGGT	60	110	(Converse <i>et al.</i> , 2009) [42]
	<i>Bctd-R</i>	CGTAGGAGTTTGGACCGTGT			
Genus <i>Prevotella</i> 16s rRNA	<i>Prev-F</i>	GGGATGCGTCTGATTAGCTTGTT	55	176	(Datcu <i>et al.</i> , 2013) [43]
	<i>Prev-R</i>	CTGCACGCTACTTGGCTGGTTC			
<i>Bifidobacterium</i> spp. 16s rRNA	<i>Bifido-F</i>	GGGATGCTGGTGTGGAAGAGA	60	231	(Haarman and Knol, 2005) [44]
	<i>Bifido-R</i>	TGCTCGCGTCCACTATCCAGT			

Supplementary Table SIII. Parameter definitions in the scoring system with their respective Aiken indices

Parameter	Definition	Aiken index of validity*
Abdominal cellulitis	Abdominal physical examination showed tenderness and discoloration of abdominal wall. Yes = 1, No = 0	0.83
Abdominal plain X-ray	Abdominal plain X-ray showed pneumatosis intestinalis, portal venous gas, or pneumoperitoneum. Yes = 1, No = 0	1.0
Advanced neonatal resuscitation	The need of positive-pressure ventilation, compression and tracheal intubation, or epinephrine administration at birth. Yes = 1, No = 0	0.17
Gestational age < 32 weeks or birthweight < 1500 g	Gestational age < 32 weeks or birthweight < 1500 g (very low birthweight criteria or less). Yes = 1, No = 0	0.5
Platelet count	Platelets count from venipuncture showed < 150 000/ml. Yes = 1, No = 0	0.5
Sepsis	Clinical picture of temperature instability and bradycardia, supported with positive blood culture confirming bacteremia or procalcitonin level > 2.0 ng/ml. Yes = 1, No = 0	0.5
(Oro)gastric retention	Emesis or residual retention > 20% feed volume or discolored emesis/residual. Yes = 1, No = 0	0.5
Abdominal distention	Abdominal girth (circumference) at the level of umbilicus increase > 3 cm from baseline. Yes = 1, No = 0	0.67
Metabolic acidosis	Metabolic acidosis from blood gas analysis showed decrease of pH < 7.35 and plasma bicarbonate (< 22 mmol/l). Yes = 1, No = 0	0.5

Aiken (1980) introduced a measure to assess the validity of items and determine content validity (Aiken, 1980). Aiken's validity test entails evaluating whether an item is valid from the perspective of evaluators (raters). Three experts assessed the validity of the observational guidelines using Aiken's method. According to Retnawati (2016), Aiken's agreement index can be classified into three categories: (1) low validity if the score is below 0.4, (2) moderate validity if the score is in the range from 0.4 to 0.8, and (3) high validity if the score exceeds 0.8.



Supplementary Figure S1. Participants flow diagram