Early Use of Antibiotics Is Associated with a Lower Incidence of Necrotizing Enterocolitis in Preterm, Very Low Birth Weight Infants: The NEOMUNE-NeoNutriNet Cohort Study

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Objective To determine whether commencement of antibiotics within 3 postnatal days in preterm, very low birth weight (VLBW; ≤1500 g) infants is associated with the development of necrotizing enterocolitis (NEC).

Study design Preplanned statistical analyses were done to study the association between early antibiotic treatment and later NEC development, using the NEOMUNE-NeoNutriNet cohort of VLBW infants from 13 neonatal intensive care units (NICUs) in 5 continents (n = 2831). NEC incidence was compared between infants who received early antibiotics and those who did not, with statistical adjustments for NICU, gestational age, birth weight, sex, delivery mode, antenatal steroid use, Apgar score, and type and initiation of enteral nutrition.

Results The incidence of NEC was 9.0% in the group of infants who did not receive early antibiotics (n = 269), compared with 3.9% in those who did receive early antibiotics (n = 2562). The incidence remained lower in the early antibiotic group after stepwise statistical adjustments for NICU (OR, 0.57; 95% CI, 0.35-0.94, $P < .05$) and other potential confounders (OR, 0.25; 95% CI, 0.12-0.47; $P < .0001$).

Conclusions In this large international cohort of preterm VLBW infants, a small proportion of infants did not receive antibiotics just after birth, and these infants had a higher incidence of NEC. It is important to better understand the role of such variables as time, type, and duration of antibiotic treatment on NEC incidence, immune development, gut colonization, and antibiotic resistance in the NICU. *(J Pediatr 2020;227:128-34)*.

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Necrotizing enterocolitis (NEC), a severe intestinal inflammatory complication related to gut immaturity, occurs in 1%-13% of very low birth weight (VLBW) infants.[1-3] Although early postnatal treatment with enteral antibiotics has been shown to reduce bacterial load in the gut and protect against NEC in animal models and clinical studies, this approach is not used to prevent NEC, owing to concerns about antimicrobial resistance and negative consequences of antibiotic therapy.[4-6] Antibiotics administered intravenously can potentially decrease microbial diversity, delay commensal colonization, and increase pathogenic bacteria.[7,8] Prolonged exposure to empirical intravenous antibiotics given in the first week of life has been associated with an increased risk of NEC, late-onset sepsis, and death in preterm infants in some studies.[9-14]

Owing to the risk of early-onset sepsis, intravenous antibiotics are administered shortly after birth in the majority of VLBW infants, but >20% have a very low risk of infection with questionable treatment indications.[14,15] The effect

NEC	Necrotizing enterocolitis
NICU	Neonatal intensive care unit
PMA	Postmenstrual age
SGA	Small for gestational age
VLBW	Very low birth weight

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of such early antibiotic intervention on later NEC development compared with no early antibiotic treatment is unclear. Data collected in the NeoNutriNet cohort from 13 neonatal intensive care units (NICUs) on 5 continents reported wide differences in enteral feeding practices and antibiotic use in VLBW infants.[1] Using data from this cohort, we investigated the within-hospital correlation between early use of antibiotics and NEC in VLBW infants. We hypothesized that initiation of antibiotics within 3 days of birth would increase the incidence of NEC.

Methods

The NEOMUNE-NeoNutriNet cohort is a web-based database established on September 15, 2013, to collect data on in-hospital nutrition and clinical measures for VLBW infants. Each participating unit was requested to collect at least 100 infants, born consecutively between January 2011 and September 2014. A detailed description of the cohort has been published previously.[1] Inclusion criteria included infants with a birth weight ≤1500 g who were admitted to a participating NICU within 24 hours after birth. Exclusion criteria included major congenital abnormalities, metabolic disease, or death or transfer to another hospital within 24 hours of birth. Thirteen hospitals participated across Europe (n = 3), Oceania (n = 2), North America (n = 1), Africa (n = 1), and Asia (n = 6). A total of 2947 infants were included from 13 participating NICUs, of whom 2831 had data on both antibiotic use and NEC outcomes and were included in the current study (**Figure 1**; available at www.jpeds.com). The incidence of NEC was defined as Bell stage ≥II, as described previously.[1,16] According to the timing of antibiotic use, these 2831 infants were divided into 2 groups: the early antibiotic group, if the infant received any antibiotic treatment within 3 days of birth or the no-early antibiotic group if the infant received antibiotic treatment beyond the third postnatal day, or never received any antibiotics. Antibiotics were administered intravenously in all cases. We compared the incidence of NEC between the early antibiotic and no early antibiotic groups with adjustments for potential confounders.

For explorative analyses, postmenstrual age (PMA) at onset of NEC was compared between the early antibiotic and no early antibiotic groups. NEC incidence was also assessed for correlation with the duration of early antibiotic treatment and the receipt of prolonged antibiotic treatment (>3 days). In addition, to evaluate whether the association between NEC and early antibiotic use differed in different geographical regions, we performed similar analyses within the 5 Guangdong (South China) NICUs and the 6 Western NICUs, respectively (**Table I**).

Statistical Analyses

All statistical analyses were performed using R version 3.2.2 (R Foundation for Statistical Computing, Vienna, Austria). *P* values were 2-sided, and statistical significance was defined as $P < .05$. Data were summarized using mean and SD or number and percentage as appropriate. For baseline comparisons between the early antibiotic and no early antibiotic groups, ANOVA was used for continuous outcomes (eg, body weight, birth weight) and logistic regression was used for binary outcomes (eg, small for gestational age [SGA] status) without any adjustment.

For our primary analysis, logistic regression was used to assess the difference in NEC incidence between the early antibiotic and no early antibiotic groups. With regard to adjustment for covariates and potential confounders, a predefined stepwise approach was followed using logistic mixed-effects models.[17] Model A was adjusted using NICU as the random effect. Model B was also adjusted for gestational age, birth weight, and sex. Model C added adjustments for delivery mode, use of antenatal steroids, and 5-minute Apgar score. Model D added adjustments for time when enteral nutrition was initiated and type of enteral nutrition provided in the first week of life. To test whether SGA could influence the primary outcome, birth weight *z*-score and SGA status were used in the model instead of birth weight in explorative analyses. Birth weight *z*-score was calculated based on the Fenton 2013 growth chart, and SGA was defined as <10th percentile for weight at birth, as described previously.[1,18] Unadjusted ORs of each NICU were visualized in a forest plot, and the heterogeneity across NICUs was examined by the Cochran Q test and quantified by the I^2 index.

For further explorative analyses, a logistic mixed-effects model was used to assess associations between NEC incidence and duration of early antibiotic therapy and the difference in NEC incidence among predefined subgroups, and a linear mixed-effects model was used to assess the difference in PMA at NEC onset between the early antibiotic and no early antibiotic groups and among predefined subgroups. Similarly, the stepwise approach with 4 models was used to adjust for covariates and potential confounders, as described above. ORs and regression coefficients are reported for all models together with their 95% CIs. To address possible bias from the loss of data of infants who died or were discharged or transferred early to step-down units or home before NEC development, we performed proportional hazards regression analysis for the primary outcome to test the robustness of our findings. Tukey post hoc analysis was used to adjust for multiple comparisons when required.

Results

Across the NICUs, the majority of infants (2562 of 2831; ~90%) received antibiotic treatment within the first 3 days after birth. The prevalence of early antibiotic treatment ranged from 70% in the Department of Neonatology, Rigshospitalet, Denmark to 99% in the Shenzhen Maternity & Child Health Care Hospital, China, and the mean duration of early antibiotics treatment ranged from 3 ± 2 days in NEW to 19 ± 12 days in the Shenzhen Nanshan People's Hospital, China (**Table I**).

Table I. Overview of antibiotic treatment at each participating NICU

Hospital	FOS	SWC	SNP	BWC	PWC	AUC	CHI	COP	AMS	NEW	PER	TAI	IBA
Total included, n	403	455	93	167	241	156	177	281	174	134	152	249	149
Use of early antibiotics, n (%)	394 (98)	451 (99)	91 (98)	140 (84)	228 (95)	144 (92)	172 (97)	196 (70)	131 (75)	112 (84)	138 (91)	228 (92)	137 (92)
Duration of early antibiotics, d, mean (SD)	14 (9)	10 (8)	19 (13)	10 (7)	11 (6)	6 (5)	6 (4)	6 (4)	5 (2)	3 (2)	4 (2)	5 (4)	12 (7)
Type (class) of early antibiotics, n							UNK						
Penicillin	370	449	5	112	224	144		129	131	111	137	227	129
Aminoglycoside	4	9	1	0	1	140		192	131	111	137	200	135
Cephalosporin	316	435	89	123	155	19		108	0	46	7	46	97
Carbapenem	125	62	29	63	46	4		127	33	12	12	42	31
Macrolide	77	102	7	57	0	0		3	0	1	0	2	3
Lincosamide	1	2	0	1	0	0		3	0	0	1	0	0
Glycopeptide	30	21	7	36	16	10		69	47	45	46	35	11
Other	8	10	0	5	4	19		49	13	30	5	58	42

AMS, Amsterdam University Medical Center, Vrije Universiteit, Emma Children's Hospital, The Netherlands; *AUC*, National Women's Health, Auckland City Hospital, New Zealand; *BWC*, Shenzhen Bao'an Maternal and Child Health Hospital, China; *CHI*, Rush University Children's Hospital, Chicago, Illinois; *COP*, Department of Neonatology, Rigshospitalet, Denmark; *FOS*, Foshan Woman and Children's Hospital, China; *IBA*, University College Hospital Ibadan, Nigeria; *NEW*, Newcastle Hospitals National Health Service Foundation Trust, United Kingdom; *NICU*, neonatal intensive care unit; *PER*, King Edward Memorial Hospital, Perth, Australia; *PWC*, Guangdong Women and Children Hospital, China; *SNP*, Shenzhen Nanshan People's Hospital, China; *SWC*, Shenzhen Maternity & Child Health Care Hospital, China; *TAI*, Children's Hospital of China Medical University, Taichung, Taiwan; *UNK*, unknown.
Guangdong NICUs: FOS, SWC, SNP, BWC, and PWC; Western NICUs: AUC, CHI, COP, AMS, NEW, and PER.

The various agents used in early antibiotic therapy are listed in Table I. The predominant type of antibiotics used in the Guangdong units was a combination of penicillin and cephalosporin, except for the Shenzhen Nanshan People's Hospital, China, which used primarily cephalosporin. In the remaining units (non-Guangdong), antibiotics was mostly a combination of penicillin and aminoglycoside. Compared with the early antibiotic group, the no early antibiotic group had higher birth weight and gestational age, lower birth *z*-score, a higher prevalence of SGA infants, fewer males, more births by cesarean delivery, and earlier receipt of enteral nutrition (all unadjusted $P < .05$; NICU-adjusted $P < .001$) (Table II). Other demographic and nutritional characteristics did not differ between the 2 groups after adjustment for NICU (Table II). Mortality adjusted by NICU tended to be higher in the early antibiotic group ($P = .07$).

The incidence of NEC was 9.0% in the no early antibiotic group, compared with 3.9% in the early antibiotic group. As the primary outcome, unadjusted NEC incidence was lower in the early antibiotic group ($P < .001$; Table II); the respective ORs per NICU are shown in a forest plot in Figure 2. There was little statistical heterogeneity among NICUs ($I^2 = 20\%$; $t^2 = 0.23$; $P = .24$). When adjusted for NICUs, the incidence of NEC remained lower in the early antibiotic group (OR, 0.57; $P < .05$) (model A, Figure 2). The significance level increased when other potential confounders were added to the models (ORs of 0.29, 0.27, and 0.25 for models B, C, and D, respectively; $P < .0001$ for all) (Figure 2). The difference between the 2 groups remained robust when changing the covariates in the analysis from birth weight to *z*-score or SGA as a binary variable (data not shown). Analysis of the primary

Table II. Early use of antibiotics, demographic, nutritional, NEC, and mortality data

Characteristics	Early antibiotics	No early antibiotics	*P* value*	*P* value†
Number of patients	2562	269		
Birth weight, g, mean (SD)	1163 (240)	1200 (246)	<.05	<.0001
Gestational age, wk, mean (SD)	29.5 (2.4)	31.0 (2.6)	<.0001	<.0001
Birth weight *z*-score, mean (SD)	−0.38 (0.98)	−0.99 (1.08)	<.0001	<.0001
SGA, n (%)	448 (18)	99 (37)	<.0001	<.0001
Male sex, n (%)	1366 (53)	111 (41)	<.001	<.001
Cesarean delivery, n (%)	1538 (60)	217 (81)	<.0001	<.0001
Antenatal steroid use, n (%)	1500 (68)	180 (74)	<.05	.25
Apgar score at 5 min <8, n (%)	1504 (63)	145 (58)	.21	.81
Enteral nutrition start, n (%)‡			<.0001	<.0001
Slow	479 (19)	10 (4)		
Medium	763 (31)	28 (11)		
Quick	1233 (50)	226 (86)		
Enteral nutrition type, n (%)§			<.0001	.890
IF	1013 (45)	54 (22)		
HM	1258 (55)	193 (78)		
Probiotics use, n (%)	675 (28)	94 (38)	<.01	<.01
Initiation in week 1, n (%)	362 (15)	71 (29)	<.0001	.53
Mortality, n (%)	152 (5.9)	15 (5.6)	.83	.07
NEC, n (%)	99 (3.9)	24 (9.0)	<.001	<.05

HM, human milk; *IF*, infant formula.
*Unadjusted *P* values.
†Adjusted for NICU as a random effect.
‡Time of initiation of enteral nutrition was initiated: slow, never started or started later than postnatal day 4; medium, started on postnatal day 3 or 4; quick, started by postnatal day 2.
§Type of enteral nutrition used in postnatal week 1: IF, exclusive IF or mixed feeding; HM, exclusive HM feeding (mothers milk and/or donor milk).

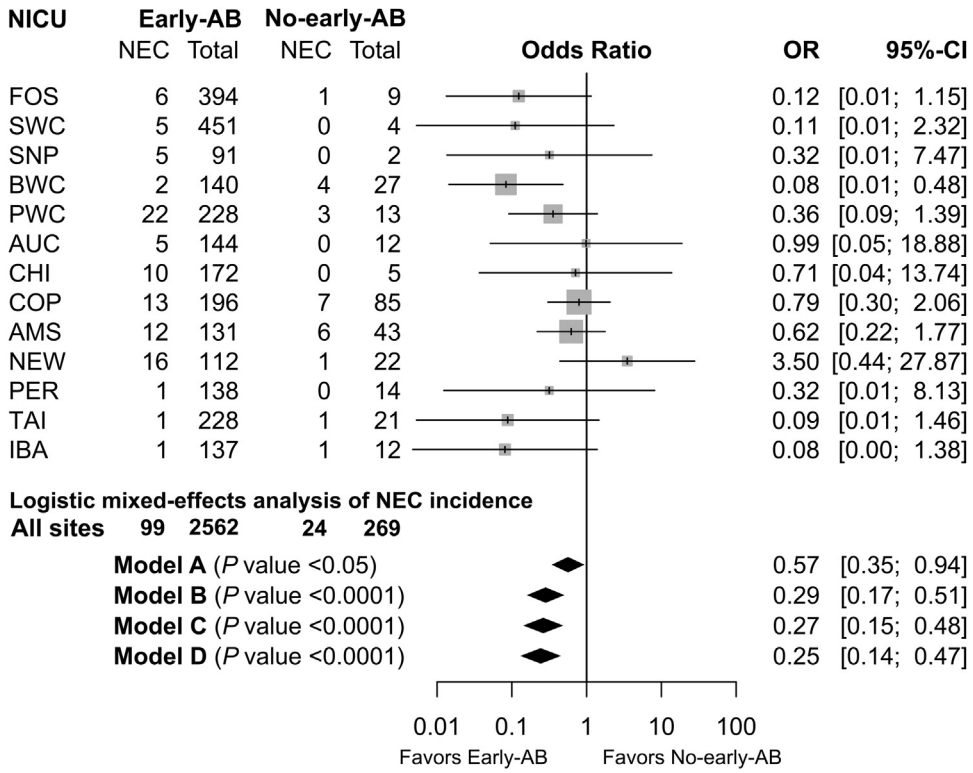

Figure 2. Forest plot of unadjusted OR of NEC incidence between the early antibiotics and no early antibiotics groups for each NICU. Model A: NICU site as the random effect and no adjustment for other fixed effects; model B: model A with adjustment for gestational age, birth weight, and sex as fixed effects; model C: model B with additional adjustment for delivery mode, use of antenatal steroids, and Apgar score at 5 minutes; model D: model C with additional adjustment for time of initiation of enteral nutrition and type of enteral nutrition in the first week of life.

outcome in a subgroup of infants of <28 weeks' gestational age revealed even more pronounced differences between the early antibiotics (n = 634) and no early antibiotics (n = 39) groups, with an NEC incidence of 8.0% vs 20.5% (OR, 0.39-0.34; $P < .05$ for all models). In the 123 NEC cases, the demographic, nutritional, and mortality characteristics differed in the same way as in all infants when comparing the 2 groups, except for birth weight (**Table III**). Compared with the early antibiotic group, the no early antibiotic group had higher birth weights in all infants (**Table II**), but the 2 groups had similar birth weights among infants with NEC. When considering all-cause mortality and early discharge, the proportion of infants diagnosed with NEC was still lower in the early antibiotic group compared with the no early antibiotic group after adjustment for potential confounders (hazard ratio, 0.23; 95% CI, 0.15-0.36; $P < .0001$).

Among the infants with NEC, there was no significant difference in postnatal age at NEC onset between the 2 groups (**Table III**), whereas PMA at NEC onset was significantly greater in the no early antibiotic group both before and after adjustment for NICU (OR, 0.96 for model A; both $P < .05$) (**Figure 2**). When adjusted for other potential confounders, including gestational age, the between-group difference in PMA became nonsignificant in models B, C, and D (OR, 1.00-1.02; $P = .20$-$.93$).

The differences in NEC outcomes were further investigated in additional subgroup analyses. Categorizing the infants by different geographic regions showed that the OR for NEC in the early antibiotic group compared with the no early antibiotic groups was lower with stronger statistical significance in the 5 Guangdong units than in the 8 non-Guangdong units (**Table IV**; available at www.jpeds.com). When analyzing data only from the 5 Western NICUs, the ORs for NEC were attenuated but remained significant after adjustment for potential confounders, such as birth weight and gestational age ($P < .05$, for models B, C, and D) (**Table IV**). The duration of early antibiotic treatment did not correlate with NEC incidence (OR, 0.99 for all models) for each 1-day increase in early antibiotic duration. When the infants were grouped into duration of early antibiotic treatment duration of either 1-3 days (n = 559), or >3 days (n = 1991), the incidence of NEC was 4.3% and 3.7%, respectively, and did not differ between the 2 groups.

Discussion

Contrary to our hypothesis, NEC incidence was lower in infants who received early antibiotic treatment. This association was robust across NICUs and was strengthened

Table III. Demographic, nutritional, and mortality characteristics in infants with NEC

Characteristics	Early antibiotics	No early antibiotics	*P* value*	*P* value†
Number	99	24		
Birth weight, g, mean (SD)	1012 (255)	1026 (331)	.82	.32
Gestational age, wk, mean (SD)	27.8 (2.5)	29.5 (2.4)	<.01	<.001
Birth weight *z*-score, mean (SD)	−0.10 (0.89)	−1.00 (0.89)	<.0001	<.0001
SGA, n (%)	13 (13)	9 (34)	<.01	<.05
Male sex, n (%)	61 (62)	15 (63)	.94	.73
Cesarean delivery, n (%)	52 (53)	21 (88)	<.01	<.01
Antenatal steroid use, n (%)	68 (72)	17 (77)	.64	.89
Apgar score at 5 min <8, n (%)	52 (62)	12 (52)	.40	.39
Enteral nutrition start, n (%)‡			<.001	<.01
Slow	27 (29)	0 (0)		
Medium	23 (25)	2 (8)		
Quick	43 (46)	22 (92)		
Enteral nutrition type, n (%)§			.76	.54
IF	20 (26)	7 (29)		
HM	57 (74)	17 (71)		
Mortality, n (%)	18 (18)	7 (29)	.23	.97
Probiotics, n (%)	22 (26)	10 (43)	.10	.53
Probiotics prior to NEC onset, n (%)	17 (20)	8 (35)	.13	.67
Age at NEC onset, postnatal d, mean (SD)	18 (15)	14 (7)	.78	.62
PMA at NEC onset, wk, mean (SD)	30.2 (2.5)	31.4 (2.4)	<.05	<.05
Bell stage, n (%)			1.00	.79
II	58 (67)	16 (67)		
III	29 (33)	8 (33)		

*Unadjusted *P* values.
†Adjusted for NICU as a random effect.
‡Time when enteral nutrition was initiated was grouped into: slow, never started or started later than postnatal day 4; medium, started on postnatal day 3 or 4; quick, started within postnatal day 2.
§Type of enteral nutrition used in postnatal week 1.

after adjusting for potential confounders, including enteral nutrition type and initiation time. The association was also unaffected by the use of probiotics, which varied widely across the different sites, although information on type of probiotics was not available. The association persisted when taking early death or discharge into account and was robust to changing the covariates from birth weight to *z*-score or SGA as a binary variable, as well as to subgroup analyses with only infants <28 weeks of gestational age. Furthermore, broad-spectrum antibiotic combinations are used as empiric treatment, and the negative association between NEC rate and early antibiotic treatment did not appear to be restricted to specific types of antibiotics. In addition, the PMA at NEC onset was not affected by early antibiotic treatment when adjusted for gestational age, and no association was found between the duration of early antibiotic treatment and NEC incidence.

The major strengths of this study include the specified single a priori hypothesis before secondary analysis of the data from a large cohort of VLBW infants collected to study the relationship between feeding and NEC, along with the primary analysis addressing the within-hospital association. Although the incidence of NEC as well as mortality varied widely among the study NICUs, the ranges are comparable to those in previous reports of NEC incidence in VLBW infants.[19] However, the diagnosis of NEC is known to be variable, and this study did not permit diagnosis of NEC after transfer to a step-down unit or to home before meeting discharge criteria for socioeconomic reasons.[1,20] Nevertheless, the robust association across NICUs from several continents indicates potential general relevance despite the widely differing guidelines for antibiotic treatment.

Although the total cohort size for this study was large, an important limitation is the small number of subjects in the no early antibiotic group, and this group was characterized by significantly more mature infants (by approximately 2 gestational weeks) who were disproportionately SGA owing to the ≤1500 g birth weight limit of the cohort. A critical question is whether the adjustment for SGA in the statistical analysis was effective. We used 3 different sets of variables as covariates: gestational age and birth weight, gestational age and *z*-score, and gestational age and SGA status. These analyses showed very similar results. Furthermore, in the no early antibiotic group, the proportion of SGA in infants diagnosed with NEC (34%) was similar to the overall proportion of SGA (37%), indicating that SGA was not a main reason for increased NEC in the no early antibiotic group. In addition, when only including those of gestational age <28 weeks, this finding was still significant and even more pronounced. The proportion of cesarean deliveries was also higher in the no-early-antibiotic group. This likely reflected a higher incidence of physician-induced preterm birth, where the absence of risk factors for infection makes it relevant to omit postnatal antibiotic treatment. Poor fetal growth associated with fetal Doppler ultrasound abnormalities may increase the risk of NEC, however, and thus the true effects of the prenatal causes of NEC might not be fully represented by SGA.[21,22] Cesarean delivery can potentially lead to abnormal colonization in newborns, although several studies have found no increased

risk of NEC after cesarean delivery.[23-27] Mode of delivery does not appear to be a confounding factor on the effect of early antibiotic use, as this would have been identified through our statistical analysis (model D).

Another limitation is that we did not include other potential confounding factors, such as maternal status, use of surfactant, respiratory and circulatory challenges, and patent ductus arteriosus. Furthermore, potential confounding events that occurred between early antibiotics and NEC onset were not recorded in the database. Related to this aspect, it is relevant to note that the 2 groups are defined by the very initial antibiotic treatment, whereas many subsequent diagnoses, complications, and treatments before NEC onset are unclear. This includes antibiotics initiated after the first 3 postnatal days, which can comprise several regimen variations for different reasons. In a subgroup of infants who had initiation of antibiotics beyond the third postnatal day, the NEC incidence was still lower in the early antibiotic group (n = 949) compared with the no early antibiotic group (n = 137) (7.9% vs 16.8%; $P < .01$ for all models).

Two previous studies in preterm infants support the association between early antibiotics and lower NEC incidence. A case-control study with 56 NEC cases and 280 controls found a lower incidence of NEC in infants who received antibiotic treatment within 24 hours of birth.[21] A large cohort study in VLBW infants (n = 14 207) showed a small but significant reduction in the risk of NEC in infants treated with a short duration of early antibiotics (≤3 days) compared with infants not receiving early antibiotic treatment.[14] In contrast, others have found an increased NEC incidence with early antibiotic treatment, which was defined as initiation within the first week of life.[8] Early antibiotic treatment may delay rapid colonization after birth in the immature preterm intestine.[28-30] A brief delay may provide time for intestinal postnatal adaptation of immune defense mechanisms, such as mucosal barrier function, which undergoes significant maturation within the first few days after birth in preterm infants, as also supported by studies in preterm pigs.[6,31,32]

A prolonged duration of antibiotic treatment, starting in the early neonatal period, has been linked to higher incidence of NEC as well as disruption of the intestinal microbial composition.[9-12,14] However, other studies and the present study have not found this association.[12,14,21] Nonetheless, compared with shorter duration, prolonged antibiotic therapy promotes the development of antimicrobial resistance, which is a growing challenge and concern.[33-35] This is a significant problem in NICUs, and infants who develop NEC often need long treatment courses of antibiotics.[35-37] Furthermore, the use of antibiotics early in life may have adverse consequences after the neonatal period.[38] Studies have found an increased risk of obesity after antibiotic exposure in early life in both infant and mouse studies.[35,39] Although antibiotics are a mainstay in the treatment and prophylaxis of infections, excessive use of antibiotics should be avoided.[35,38,40]

In conclusion, in this large international cohort of VLBW infants, the 10% of infants who did not receive antibiotics within 3 days of birth showed a higher NEC incidence. We do not yet suggest empirical antibiotic administration for every VLBW infant without antenatal risk factors or signs of neonatal infection given the other antibiotic-related neonatal morbidities and potential long-term side effects, as well as the risks of antibiotic resistance.[14,35] Nonetheless, the no early antibiotic group in this study may represent an underrecognized group of infants with an elevated risk of NEC, despite their apparent lack of indication for antibiotic treatment. A clinical trial using early and short-duration antibiotic treatment to prevent NEC, coupled with clinical signs of gut dysfunction (eg, feeding intolerance, bacterial dysbiosis, fecal and blood biomarkers of immune dysfunction) could be considered. Furthermore, mechanistic animal studies on the optimal timing, dose, regimen, and type of antibiotic, incorporating detailed microbiological, immunologic, and nutritional data, may help clarify if, when, and how antibiotics should be used for VLBW infants. A better understanding of antibiotics-related gut mechanisms could pave the way for more and better alternatives to the widespread antibiotic treatment of VLBW infants. ■

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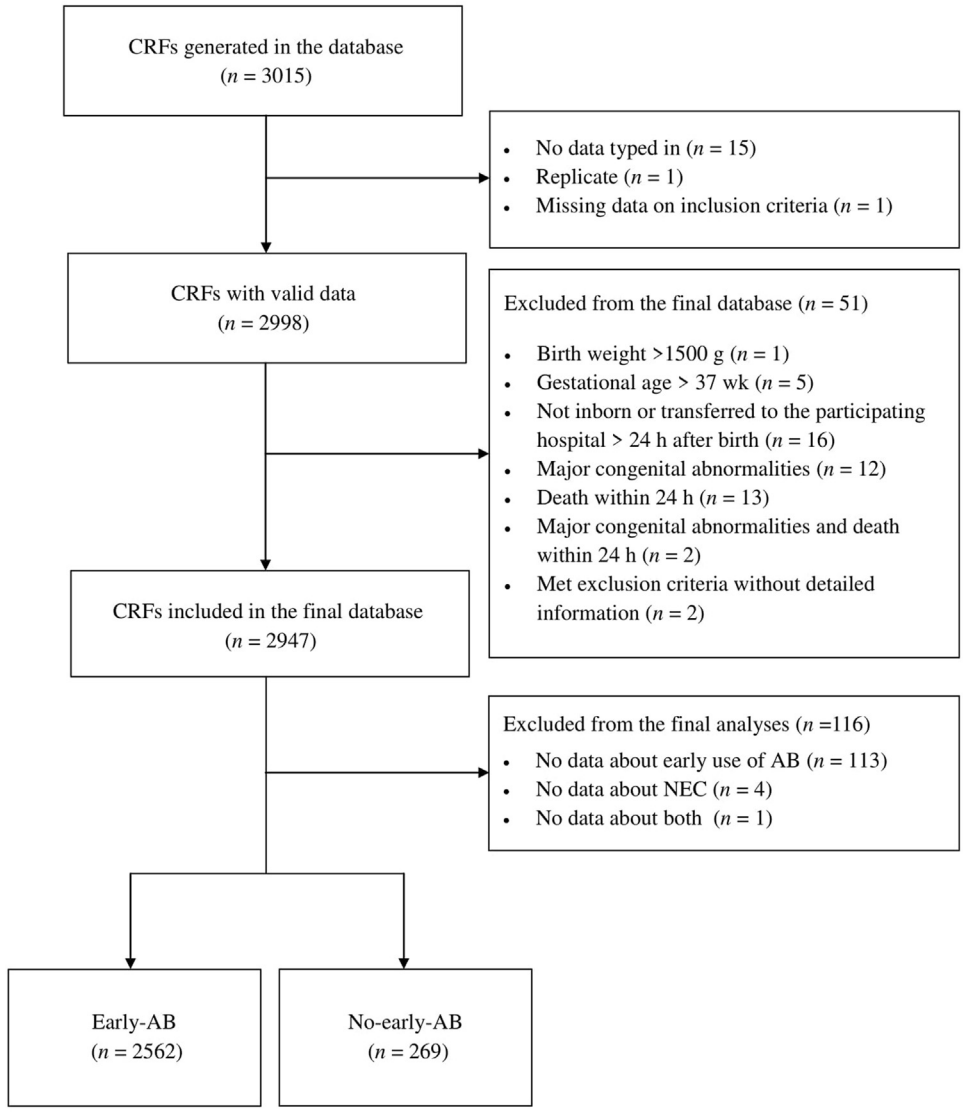

Figure 1. Flowchart for inclusion of study participants. *AB*, antibiotics; *CRF*, case report form.

Table IV. NEC incidence in the early antibiotic and no early antibiotic groups by geographic region

	5 Guangdong NICUs		8 non-Guangdong NICUs		6 Western NICUs	
Models*	OR (95% CI)	*P* value	OR (95% CI)	*P* value	OR (95% CI)	*P* value
A	0.22 (0.09-0.51)	<.001	0.82 (0.45-1.49)	.49	0.96 (0.52-1.80)	.91
B	0.14 (0.05-0.35)	<.000	0.34 (0.17-0.69)	<.01	0.39 (0.19-0.81)	<.05
C	0.13 (0.05-0.34)	<.000	0.32 (0.15-0.70)	<.01	0.38 (0.17-0.87)	<.05
D	0.11 (0.04-0.32)	<.000	0.33 (0.15-0.72)	<.01	0.39 (0.17-0.89)	<.05

*Adjustment of models: model A, NICU as the random effect and no adjustment for other fixed effects; model B, model A with adjustments for gestational age, birth weight, and sex as fixed effects; model C, model B with additional adjustments for delivery model, use of antenatal steroids, and Apgar score at 5 min; model D, model C with additional adjustments for time of initiation of enteral nutrition and type of enteral nutrition in the first week of life.