

NECROTISING ENTEROCOLITIS – ETIOPATOGENIC FACTORS IN PREMATURE VERSUS TERM NEONATES

Elena Hanganu¹, S.G. Aprodu¹, Simona Gavrilescu¹,
Andreea Avasiloaiei², Mihaela Moscalu³, Maria Stamatin²

Abstract

Objectives. The aim of this paper is to analyze the elements that define the etiopathological profile of NEC cases diagnosed in term and preterm neonates admitted in the Regional Center of Neonatal Intensive Care Unit “Cuza Voda” and “Sf. Maria” Emergency Children Hospital Iasi.

Material and methods. An analytic retrospective study was performed and all cases of necrotizing enterocolitis diagnosed in the Regional Center of Neonatal Intensive Care Unit “Cuza Voda” and “Sf. Maria” Emergency Children Hospital Iasi were included. A datasheet was developed for study group (premature) and control (term neonates) group and the statistical analysis of the 205 cases was performed in correlation with the following variables: gender, gestational age (GA), birth weight (BW), APGAR score, maternal and neonatal risk factors, the presence of associated anomalies, type of feeding, age, symptoms at onset and stage at diagnosis according to Bell criteria. The statistical analysis of all data was performed in SPSS Statistics 20 program.

Results. The incidence of NEC was 3.32%, with a total number of 205 cases identified: 155 cases in premature group and 50 cases in control group. In study group 65.81% were born naturally versus 46% in control group respectively 34.19% were born through cesarean section versus 54% in control group. Birth weight was < 1000g in 12.9% of cases and < 1500g in 43.9% in the study group with only 14% below 2500g in control group. 98 cases were diagnosed in stage I Bell (63.23%), 46 cases in stage II (29.68%) and only 11 cases were diagnosed in stage III (7.1%). In study group 21.9% presented persistence of ductus arteriosus (PDA) whereas in the control group PDA was identified in 12%. Perinatal asphyxia was present in 17.42% of cases in the study group, mild respiratory distress in 32.9%, moderate distress in 27.1% and severe respiratory distress in 40%. Bacterial colonization was identified with a frequency of 45.2% whereas in the control group frequency was 20%. Mechanical ventilation in the study group was identified as a risk factor in 67.1% of cases and in the control group in 12% of cases. The type of feeding in the

study group was: 39.35% of cases were on total parenteral nutrition (TPN), 30.97% parenteral nutrition and milk formula, 18.71% parenteral nutrition and maternal milk, 8.39% formula milk and only 2.58% were exclusively feed with maternal milk. The mean age at the time of diagnosis was of 10.9 days for the stage I NEC, 11.5 days for stage II and 14.3 days for stage III.

Conclusions. The only cert pathogenic factors in NEC are represented by prematurity and low birth weight. The significant number of cases in term neonates should bring the attention on the necessity of modified diagnostic criteria because under the generic term of NEC we can treat other NEC-like entities with a particular evolution pattern.

Key words: necrotizing enterocolitis, newborn, premature

Abbreviations

NEC: necrotizing enterocolitis, LBW: low birth weight, VLBW: very low birth weight, ELBW: extremely low birth weight, BW: birth weight, GA: gestational age, TPN: total parenteral nutrition, SIP: spontaneous intestinal perforation, PDA: persistence of ductus arteriosus.

Introduction

The first descriptive data suggestive for necrotizing enterocolitis (NEC) are dated in 1888 in pathological reports of neonates with intestinal perforation as a possible cause of death. The first medical report of ileal isolated perforation successfully treated belongs to Argenty in 1943.

Later in 1952, we found the first information regarding necrotizing enterocolitis in the medical literature in 2 articles published in “Z Kinderheikd” journal referring to the pathological (1), and clinical (2) characteristics of a specific form of enteritis which the authors Schmid and Quaiser are calling „Enterocolitis ulcerosa necroticans”. After 1 year, the same authors have published a new article (3) in which the disease is called necrotizing enterocolitis (NEC) and the terminology is used until today. In 1964 in United States at Children Hospital from New York, Berdon published the first description of the clinical and radiological characteristic of the disease in premature newborn (4).

¹Department of Paediatric Surgery, “Sf. Maria” Emergency Children Hospital, “Gr. T. Popa” University of Medicine and Pharmacy, Iasi

²Department of Neonatology, Regional Center of Neonatal Intensive Care Unit “Cuza Voda”, “Gr. T. Popa” University of Medicine and Pharmacy, Iasi

³Department of Medical Informatics and Biostatistics, “Gr. T. Popa” University of Medicine and Pharmacy, Iasi

E-mail: dr.elenahanganu@gmail.com, sgaprodu@yahoo.com, drgavrilescu@yahoo.com, andreea_avasiloaiei@ymail.com, mmoscalu@yahoo.com, maria.stamatin@yahoo.com

In 1975 Santulli et al have published the first article reporting their experience regarding some forms of necrotizing enterocolitis in low birth weight neonates which needed aggressive surgical treatment. It is the time when it was first launched the etiopathogenic hypothesis of the disease based on the 3 components: lesions of the intestinal mucosa, alteration of the bacterian colonization and the presence of a metabolic substrate (5). An important moment in the disease description is represented by the publication of some staging criteria by Bell et al based on the clinical and radiological characteristics. This is known in the medical literature as Bell staging criteria (6). The importance of this classification is that it offers data of clinical and practical utility for an adequate therapeutic decision. In 1979 the International Register of Classification of Disease settles a code for death by necrotizing enterocolitis and in this way a more correct centralization of the epidemiological data has become possible. From that moment many scientific articles based on data review or on research activity have been published with the intention to complete the information regarding the incidence, clinical characteristics and possibilities of treatment in necrotizing enterocolitis.

Necrotizing enterocolitis affects 1 to 5% from the total neonate number in every Neonatal Intensive Care Unit and the incidence in very low birth weight (VLBW) is between 7-14% (7, 8). Published data regarding incidence of necrotizing enterocolitis vary by geographical area and the time period in which the report is done and on the other hand vary by the degree of prematurity and the birth weight (BW) of the neonate. One of the most important sources of variability of the incidence of NEC is represented by the criteria of diagnosing the disease.

NEC is a pathological entity which belongs to the premature neonate by definition but may also be encountered in term neonate. It is estimated that from the total number of NEC between 7 to 13% of cases may be encountered in term neonate with an onset of the symptoms at approximately 5 days of age and especially in the presence of some associated risk factors that can affect the mesenteric blood flow: perinatal hypoxia, congenital heart malformation, multiple births, history of umbilical artery catheterization, sepsis, polycythemia, gastroschisis (9, 10). A particular form of disease can appear in the first 2 weeks of life in extremely low birth weight (ELBW) neonate before the initiation of enteral feeding and is represented by the isolated spontaneous intestinal perforation (SIP). This entity has distinct anatomo-pathological characteristics and the prognosis of the neonates appears to be better than in the classical forms of NEC (11).

For the premature neonate the peak of incidence is situated between 2 to 3 weeks postnatal, after the immediate recovery period when the neonate is enteral feed. The sooner the enteral feed is begin the earlier the onset of NEC is. It was calculated a rate of NEC onset in weeks of life in correlation with birth weight and it was concluded that the risk period for disease onset is lower with the progression of

the BW. Also the calculated risk of NEC suddenly drops when the gestational age of 35-36 weeks is reached. These observations have promoted the pathogenic theory of gastrointestinal tract maturation as a risk factor for NEC.

NEC is a multifactorial disease characteristic for premature newborns with an incidence that is in constant progression both in Romania and worldwide. In the medical literature there are still multiple controversies regarding the etiopathogeny of the disease, the role of risk factors and the clinical distinction between different forms of enterocolitis (the term of NEC covers a spectrum of disease that includes spontaneous isolated perforation and other NEC-like disease) (12,13,14).

The aim of this paper is to analyze the elements that define the epidemiological and etiopathogenic profile of NEC cases diagnosed in the Regional Center of Neonatal Intensive Care Unit “Cuza Voda” Iasi and “Sf. Maria” Emergency Children Hospital Iasi and to identify the particular aspects of NEC in premature neonate cases.

Materials and methods

We carried out an analytic retrospective study that included all cases of necrotizing enterocolitis diagnosed in the Regional Center of Neonatal Intensive Care Unit “Cuza Voda” and “Sf. Maria” Emergency Children Hospital Iasi. We realized a database on the evidence of disease code that was introduced in Electronic Evidence System of the two hospitals and afterwards the information from the medical records and the operatory registers were noted. Cases were divided in two groups: study group (premature newborns) and control group (term newborns). We developed an original datasheet and the statistical analysis was performed in correlation with the following variables: gender, gestational age (GA), birth weight (BW), APGAR score, maternal and neonatal risk factors, the presence of associated anomalies, type of feeding at onset, age at onset, symptoms at onset and stage at diagnosis according to Bell criteria. The statistical analysis of all data was performed in SPSS Statistics 20 program.

Results

From January 2008 to December 2013 the general incidence of NEC was 3.32%, from a total number of 6183 neonates that were admitted in the Regional Center of Neonatal Intensive Care Unit “Cuza Voda” with a total number of 205 cases identified (155 cases at premature neonates with GA below 37 weeks and 50 cases at term neonates). The cases in premature neonates represented 75.6% from the total number of NEC diagnosed. In the statistical analysis of data the term neonate group represented the control group used to identify the particular aspects of the diagnosis and evolution of the premature babies with NEC.

The mean age of gestation in the general study group was 33 weeks. The statistical parameters of the age of gestation from the study group are presented in table 1.

Tabel 1. Statistical parameters of GA.

Mean GA	Mean		Std. Dev.	Min	Max	Q25	Median	Q75
	-95%	+95%						
33.0	32.4	33.5	4.1	24	41.0	30	33	36

The analysis of the association between NEC onset and type of delivery revealed that in premature neonates from the total number of 155 cases 102 (65.81%) were born naturally and 53 (34.19%) were born through cesarean section. In the control group of 50 term neonates 23 cases were born naturally (46%) and 27 through cesarean section (54%). The statistical analysis revealed that vaginal delivery

is statically significant correlated with prematurity in NEC (65.81%) whereas in term neonates with NEC vaginal delivery was identified in 46% of cases (Pearson Chi-square $\chi^2=6.23$, $p=0.0125$, 95%CI).

Birth weight (BW) in the premature study group was as follows (table 2):

Tabel 2. Birth weight in NEC.

Birth Weight	< 1000g	1000-1499g	1500-2499g	>2500g	Total
Premature newborns	20	68	57	10	155
	12.9%	43.9%	36.8%	6.5%	
Term newborns	0	0	7	43	50
			14.0%	86.0%	
%	9.76%	33.17%	31.22%	25.85%	

In the premature group of NEC we found out that 12.9% of cases presented a birth weight lower than 1000g and 43.9% presented with birth weight below 1500g. In comparison with the term neonate control group with NEC there are statistically significant differences ($\chi^2=127.19$, $p<<0.01$), in this group only 14% of the neonates presented with birth weight between 1500 and 2499g.

The distribution according to Bell, Walsh and Kliegman in the group of 155 premature cases of NEC was

as follows: 98 cases were diagnosed in stage I Bell (63.23%), 46 cases in stage II (29.68%) and only 11 cases were diagnosed in stage III (7.1%).

The graphic representation of NEC staging at the time of diagnosis revealed that advanced stage of disease are correlated with prematurity unlike term neonates cases in which there were no case of diagnosis of NEC in stage III Bell- Figure 1.

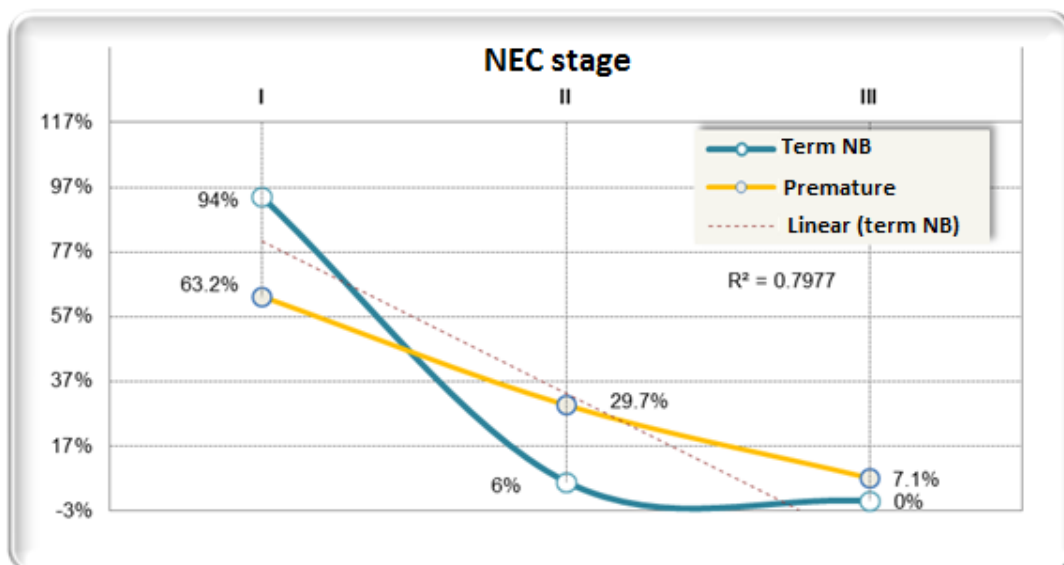


Fig. 1 Stage of NEC in premature versus term neonates.

The risk factors involved in the pathogenesis of NEC that were analyzed were the following: persistence of ductus arteriosus, perinatal asphyxia, respiratory distress, bacterial colonization, mechanical ventilation and the type of feeding at the onset of symptoms.

Persistence of ductus arteriosus (PDA) was encountered in 34 cases of premature newborns and was associated statistically significant with NEC. In the study group of premature newborns 21.9% presented PDA whereas in the control group of term neonates PDA was identified in 12% of cases, value that is statistically significant lower ($\chi^2=6.37$, $p=0.012$, 95%CI).

Perinatal asphyxia as a risk factor for the neonates with NEC was present in 27 cases (17.42%), value that is statistically significant lower than the value in the control group of 2% ($\chi^2=7.62$, $p=0.005$, 95%CI).

Mild respiratory distress was identified in 51 cases (32.9%) and moderate distress in 42 cases (27.1%). Severe respiratory distress was encountered in 40% of the premature neonates with NEC and in 8% of the term cases, these data proving that there is a statistically significant correlation between NEC in premature and the severity of respiratory distress ($\chi^2=46.18$, $p<<0.01$, 95%CI).

Bacterial colonization in cases of premature NEC was identified with a frequency of 45.2% whereas in the control group presented a lower frequency of 20% ($\chi^2=10.05$, $p=0.001$, 95%CI). The analysis of the colonization type in

premature NEC revealed the implication of Gram negative bacteria mostly Enterobacter and Klebsiella.

Mechanical ventilation in the study group was identified as a risk factor in 67.1% of cases and in the control group only in 12% of cases. The statistical analysis revealed that there is a significant correlation ($\chi^2=46.15$, $p<<0.01$, 95%CI) between mechanical ventilation and the onset of NEC in premature newborns.

The type of feeding at the onset of symptoms in the study group was as follows: 39.35% of cases were on total parenteral nutrition (TPN), 30.97% parenteral nutrition and milk formula, 18.71% parenteral nutrition and maternal milk, 8.39% formula milk and only 2.58% were exclusively feed with maternal milk. Regarding to parenteral nutrition we did not found statistically significant differences between the study group and the control group but this differences were identified in formula feed cases with a frequency of 8.4% in premature and 34% in term neonates with NEC ($\chi^2=30.82$, $p<<0.01$, 95%CI).

The mean age of the neonates at the time of diagnosis was of 10.9 days for the stage I NEC, 11.5 days for stage II and 14.3 days for stage III- figure 2.

We noted the fact that in cases of premature newborns the age at onset of symptoms was significantly higher comparing to the age at onset in term neonates ($F=8.21$, $p=0.000004$, 95%CI).

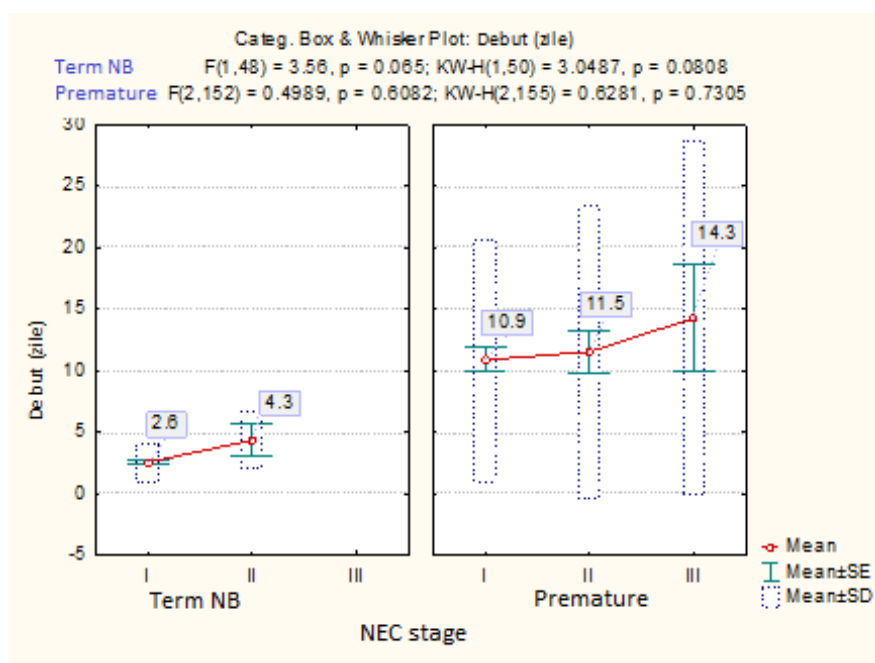


Figure 2. Statistical parameters of age at onset versus stage of NEC.

Discussion

Necrotizing enterocolitis is the number one digestive emergency encountered in premature neonate. It is universally accepted that NEC is a disease characteristic for prematurity; up to 90% from the total number of cases are

diagnosed at newborns with gestational age below 36-37 weeks (15). In the study group the general incidence of NEC was 3.32% between January 2008 and December 2013. From the total number of cases 75.6% were diagnosed in premature newborns value that is below the reported number

in the medical literature of 80-90%. The high frequency of term neonates with NEC (24.4%) could be explained by a high incidence of congenital megacolon with neonatal clinical manifestation or by some possible NEC-like cases that were introduced in the study group. The risk of NEC onset is in direct relationship with the decrease of gestational age and with the very low and extremely low birth weight (12). In the study group we identified a high frequency of NEC between 28 to 30 weeks GA with 22.44% of cases diagnosed at premature less than 22 weeks of gestation. It was demonstrated a reverse correlation between the GA of the premature and the age of onset of symptoms with cases reported in the literature with onset at more than 4 weeks after birth at extremely premature. In premature with very low birth weight (VLBW < 1500g) the reported incidence of NEC is 10 to 15% (11, 12, 13, 14).

The pathogenesis of NEC remains controversial but it is universally accepted that it is a multifactorial disease. The most frequently used pathogenic theory explains the fact that NEC can occur in the event of association between the immaturity of the premature gut with the decrease of the barrier function of the mucosa, the abnormal bacterial colonization, intestinal ischemia-hypoxemia mechanism and the enteral feeding.

The factors that are influencing the bacterial colonization of the premature gut are: type of delivery (vaginal or cesarean), using of the wide spectrum antibiotics, type of feeding (maternal versus formula milk) and other factors related to the Neonatal Intensive Care Unit (Neu et al., 2008). Data from the medical literature do not show any correlations between the incidence of NEC and the demographic factors (gender, race, place of birth, age of the mother, socioeconomic status and type of delivery) but there are significant differences between the incidence of prematurity in different centers for premature based on the level of technical expertise. In the study group we did not identified significant differences in NEC cases in correlation with gender but the vaginal delivery was associated statistically significant with NEC in premature (65.81%). This finding is in contradiction with data from the medical literature where cesarean section is associated more frequent with NEC possible based on the perturbation of the normal colonization process that comes with the vaginal delivery (16).

Ischemia-hypoxemia mechanism was initially considered as the primary factor in NEC onset (Neu et al. 2008). Recent studies have shown a greater incidence of NEC in premature newborns that needed maneuvers of resuscitation at birth, mechanical ventilation in the first days after birth, in newborns with intrauterine growth restriction caused by insufficiency of the placenta or by history of abruptio placentae (Chokshi et al., 2008). In our study the severe respiratory distress was identified with a frequency of 40% in premature newborns with NEC and of 8% in term newborns with NEC, fact that proves the statistical significant correlation between the severity of distress and the EUN onset in premature ($\chi^2=46.18$, $p<<0.01$, 95%CI). Regarding the mechanical ventilation we found out in our study that there is a statistical significant correlation

($\chi^2=46.15$, $p<<0.01$, 95%CI) between mechanical ventilation and the premature cases with NEC, 67.1% of them being on mechanical ventilation.

The implication of Gram negative bacteria in the disease pathogenesis is confirmed in the study group, the most frequently identified species being Klebsiella, Enterobacter and E Coli. In the medical literature a so called colonization "microbioma" of premature gut that was identified and is extremely vast but is impossible to isolate by conventional methods (16,17). The type, the volume and the moment of feeding initiation are key factors that are influencing the pathogenic mechanisms of NEC onset. Human milk contains lactobacillus and bifidobacterium species that facilitates the normal colonization process of the gut. More than 90% of the premature that will develop NEC have been fed. Regarding the type of feeding a higher incidence of NEC was found in formula milk versus maternal milk fed premature (Chokshi et al., 2008; Grave et al., 2007; Neu et al., 2008). In a randomized prospective study premature newborns that were feed with maternal milk developed NEC 6 to 10 times less than those feed exclusively with formula milk and 3 times less than those feed with formula and maternal milk (Lucas & Cole, 1990). In our study 39.5% of the premature with NEC were on TPN feeding at the time of symptoms onset, 8.39% were on formula milk and only 2.58% were exclusively on maternal milk. Statistic significant differences were noticed in cases with formula milk feeding which had a frequency of 8.4% for the premature newborns and of 34% for the term newborns ($\chi^2=30.82$, $p<<0.01$, 95%CI) fact that could explain the high frequency of NEC for the term neonates group. The diagnosis of disease was established early in 92.91% of cases (stage I Bell 63.23% and stage II 29.68%), results that are consistent with the published data from the literature (Neu et al., 2008) with only 7.10% of cases that were diagnosed in advanced stage III Bell. The future finding of some clinical parameters and biochemical markers will lead to left deviation of the diagnosis curve and could create the premises of NEC diagnosis exclusively in the medical stage of disease improving the general prognosis of the affected neonates.

Conclusions

Despite the evident improvement regarding the survival of the premature neonates, NEC remains a challenge both for the neonatologist and for the pediatric surgeon involved in diagnosing and treating this disease. In the time period 2008-2013 the incidence of NEC remained almost constant (3.32%) although the total number of the neonates admitted in Regional Center of Neonatal Intensive Care Unit "Cuza Voda" is in continuous progresion. The only cert pathogenic factors are represented by prematurity and low birth weight and this statement is confirmed by the analysis in the study group.

The presence of a significant number of cases in term neonates should bring the attention on the necessity of modified diagnostic criteria because it is possible that under the generic term of NEC to find a larger spectrum of

pathologic entities NEC-like with a particular evolution pattern.

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Correspondence to:

Simona Gavrilescu
 ”Gr. T. Popa“ University of Medicine and Pharmacy,
 16 Universitatii Street
 Iași, România
 E-mail: drgavrilesco@yahoo.com