CLOSTRIDIUM DIFFICILE – PATHOGEN OR NOT FOR NEWBORNS?

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Abstract

Clostridium difficile (CD), described initially as a commensal germ in infants under 1 year of age, is more and more often recognized as enteric pathogen in newborns and children, even in those without risk factors. Aim: The paper is shortly reviewing available data regarding CD colonization and infection in neonates and presents a case report. Material and method: The case of a preterm infant, positive for A and B toxins of CD, with gastro-intestinal complications during neonatal period is presented. Results: In the 9th day of life, the preterm infant develops symptoms of colitis, concomitant with maternal CD infection. Despite antibiotic treatment and intensive care, peritonitis developed during the 24th day of life. The clinical course was further complicated by an umbilical fistula, prolonging hospitalization. No other pathogen except CD has been isolated starting birth but the clinical course suggests for neonatal infection with CD. Conclusion: Given the increased incidence of CD in the latest period, we do have to reconsider the role of CD as a potential etiological agent in neonatal infections.

Keywords: Clostridium Difficile, newborn, necrotizing enterocolitis, colitis, peritonitis

Introduction

Clostridium difficile (CD) has been described initially as a commensal germ in infants under 1 year of age, is more and more often recognized as enteric pathogen in children, even in those without risk factors [1-3]. Clostridium difficile is more often encountered in infants admitted to neonatal intensive care unit (NICU) [4] and in preterm infants [5]. Most often, CD is colonizing preterm and term neonates and different factors are suggested as protectors against neonatal infections with CD [1, 4, 6]. Also, risk factors for neonatal colonization with CD were identified [1, 4], yet transmission from mother to child, irrespective the delivery mode, is debated [1, 6, 7]. Even though Clostridium Perfringens, C. Butyricum, and C. neonatale were clearly identified as etiologic agents in neonatal necrotizing enterocolitis (NEC), a causal link between CD and NEC is still controversial [8-11]. This paper briefly reviews the data in the literature regarding colonization and infections with CD during neonatal period and presents the case of a neonatal infection with CD.

Clostridium Epidemiology

Clostridium difficile is a gram positive, sporulated bacillus with toxic and non-toxic forms. A and B toxins, produced by CD, are mediating the pathogenesis of CD infections. Based on toxin production, there are three types of CD: toxin A producing CD (A+B-), toxin B producing CD (A-B+), and non-toxic CD (without virulence genes), non-pathogenic [12]. Transmission occurs by direct contact between individuals or with contaminated surfaces, and by aerosolized spores. The spores are resistant to heat, acidity, and many disinfectants [1]. After ingestion, spores are surviving in the stomach and germinate into vegetative forms inside colon. In newborns, CD transmission occurs mostly from the environment or from other children [1, 13] although identical strains of CD were also isolated per partum from mother and neonate pairs [6, 7]. Most of the studies showed no differences of the CD colonization rates according to the delivery mode [1, 6, 14], although some studies reported an increased colonization rate after cesarean section [4, 15]. Symptomatic and asymptomatic carriers of CD toxigenic strains are representing a reservoir [1].

Old epidemiological studies flagged asymptomatic CD colonization in infants [13] but during the last decade CD was recognized as potential enteric pathogen in children, even in those without risk factors [1-3]. Rates of neonatal CD colonization are variable: 25-30% of the newborns, over 33% in the NICU[4], 61-79% in preterm infants discharged from the NICU[5,13,16,17], 14-71% of infants under one year of age[1] (usually with non-toxigenic strains), decreasing to 4% between 12-18 months, rates similar to that of non-hospitalized adults[1]. Most of the toxigenic strains are found in preterm infants with intestinal conditions [17].

The commensal colonization of the neonatal gastro-intestinal tract, mainly with non-toxic strains, begins during birth and with the first feedings [1]. Factors increasing the risk for neonatal colonization and infection with CD are: antibiotic therapy (disrupting endogenous flora)(mainly due to Clindamycin, cephalosporin, penicillin)[1,4,18], increased hospitalization duration[1,4,6], significant chronic conditions[1], increased contamination of the neonatal environment[1,19], feeding method and mode, and use of acid suppressor agents[1].

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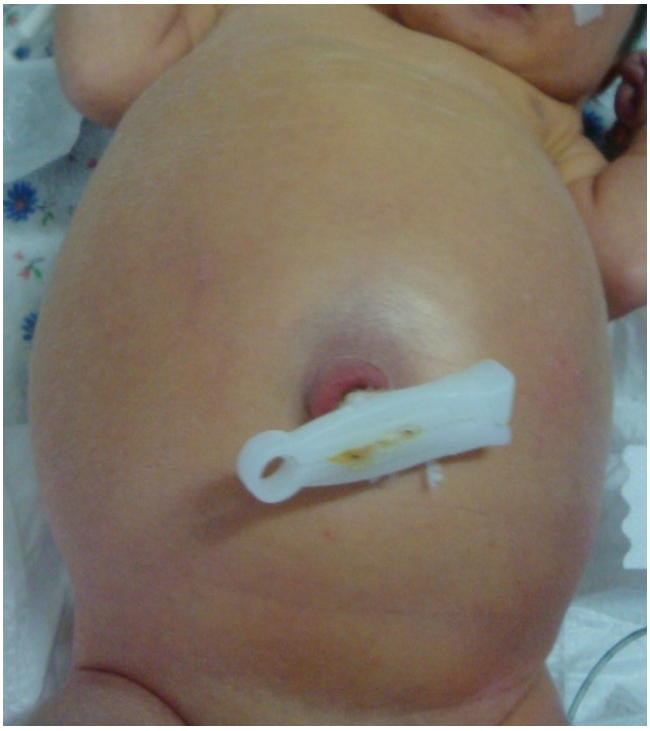

Fig. 1. Abdominal distension, mild edema of the abdominal wall

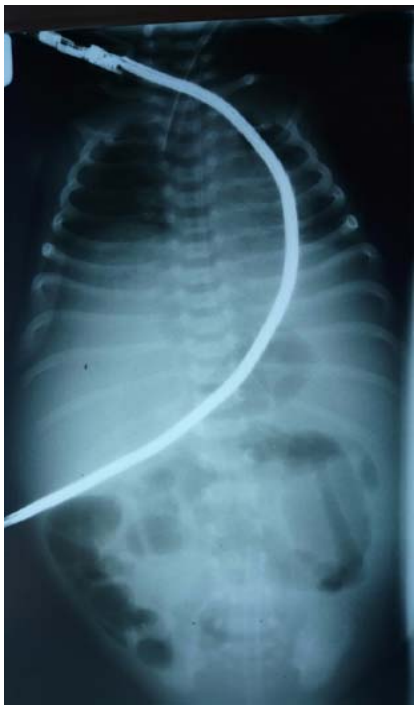

Fig. 2. Abdominal radiography – 9th day of life

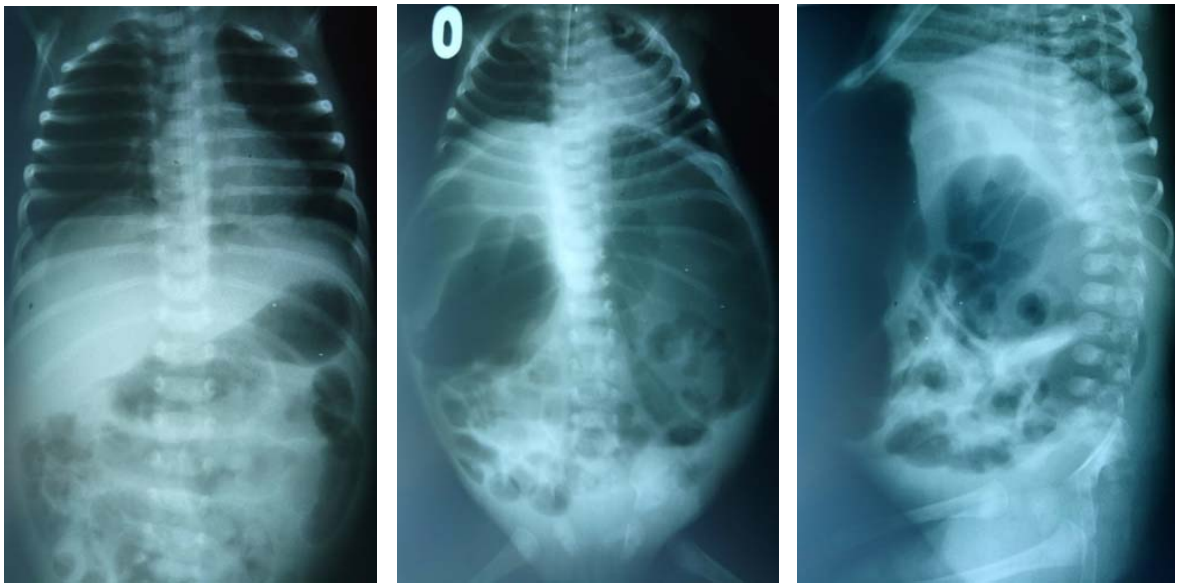

Fig. 3. Abdominal radiography survey after the 24th day of life

Clinical aspects. Laboratory diagnostic

In neonates, symptoms are varying from asymptomatic colonization to diarrhea or severe pseudo-membranous colitis. Bloody stools are the most frequent symptom seen in newborns [22, 23]. In infants, diarrhea with CD is usually mild, aqueous, accompanied by fever, anorexia, and abdominal cramps [1].

Infections with CD may be classified according to the severity – presence of complications, lab markers, and clinical signs – [1]. Pseudo-membranous colitis, toxic mega colon, gastro-intestinal perforation, intestinal pneumatics are all possible complications of neonatal CD infection, responsible both for NICU admission and surgical interventions. Laboratory tests are often showing leukocytosis or leukopenia, decreased albumin levels and increased creatinine levels [1].

Molecular testing is more and more used to identify CD. Due to the high rates of neonatal CD colonization, routine testing of the newborns is not recommended [1]. Enzyme immunoassay is a rapid, easy to perform, and very specific test for A and B toxins of CD, used currently by most of the laboratories [1]. Detection of glutamate dehydrogenase (an antigen present on toxigenic and non-toxigenic CD) has an increased negative predictive value and can be used in the diagnostic algorithm. Polymerase chain reaction (PCR) is more expensive but has increased sensibility and specificity for A and B toxins [1].

Prevention and treatment

Limiting unjustified antibiotic use, isolation of the contacts, decontamination (with agents capable to kill CD spores) of the surfaces, and hand washing are critical for prevention and limiting transmission of CD colonization and infections in the NICU [1]. Alcohol-based hand disinfectants are less efficient as compared to hand washing with warm water and soap since CD spores are resistant to alcohol [24]. A vaccine is in study [1].

Basic principles of the treatment are symptomatic care (intravenous fluids), cessation of useless antibiotic therapy (sometimes mild symptoms are vanishing [1]), and initiation of efficient antibiotics. Metronidazole (30 mg/kg/day in 4 doses, a maximum of 2 g/day) is recommended in moderate CD infections [1, 25]. Metronidazole has reduced oral absorption, rapid intestinal transit and diffusion through inflamed colonic mucosa [1]. Prolonged use is associated with increased risk for peripheral neuropathy [1, 6]. For children with severe CD infection an association of oral vancomycin (40 mg/kg/day in 4 doses, a maximum of 2 g/day) and intravenous metronidazole is recommended [25]. Vancomycin has also a reduced absorption rate therefore intestinal vancomycin levels are increased [1]. Enema with vancomycin is an alternative to oral administration [25]. The minimum duration of the treatment is 10 days [3]. A second course of treatment is recommended for the first recurrence (as in adults) [1]. Fidaxomycin is a new macrocyclic antibiotic approved in 2011 for adult treatment, in study for children [1]. Nitazoxanide and rifaxamin are other possible efficient agents, under study [6]. Restoration of the microbioma can be achieved using probiotics and is recommended mainly in children with recurrent CD infections [1].

Clostridium Difficile and necrotizing enterocolitis

Necrotizing enterocolitis (NEC) is the most frequent gastro-intestinal emergency in premature infants, occurring in 7% of infants weighting under 1500 g at birth [8]. Prematurity, abnormal intestinal colonization, and formula feeding are cited as main risk factors for NEC [8, 26]. Recently, abnormal intestinal microbioma has been shown as a key factor in NEC pathogenesis, bacteria having an important role in the progression and severity of the intestinal lesions [26]. Clinical signs of NEC are very unspecific, varying form signs limited to gastro-intestinal tract – feeding intolerance, increased gastric residuals, abdominal distension, ileus, and bloody stools – to signs of severe disease with multiple organ failure – lethargy, apnea, metabolic acidosis, shock, intravascular disseminated coagulation, and even death [8]. Different types of Clostridium – C. Perfringens, C. Butyricum, C. neonatale – were cited in association with NEC both in animal studies and human newborns [8, 26, 27]. However, implication of CD in NEC pathogenesis is controversial: a causal connection could not be identified, toxigenic strains of CD are rarely described in NEC but outbreaks of CD infections associated with NEC were also reported [4, 8, 10, 11]. For example, CD was isolated in 12 in 13 infants with NEC versus 2 in 17 controls in an outbreak finished after oral vancomycin was administered in sick infants, contacts were isolated and strict control measures were implemented [10].

Case report

The female newborn was delivered vaginally, in cranial presentation, after 6 hours after amniotic membranes rupture, at 36 weeks gestation with a birth weight of 2140 g, height 46 cm, cranial circumference 29 cm. The mother, 23 years old, primigravida, primipara had an uneventful pregnancy except non-specific vaginitis and one episode of urinary tract infection one month before delivery, incompletely treated with antibiotics. Two hours after delivery, the premature infant developed signs of respiratory distress (tachypnea, intercostal retractions, low oxygen saturations), was admitted in the NICU and placed on Bubble CPAP on nasal cannula with 30% oxygen. An umbilical line was placed for partial parenteral nutrition and antibiotics (Penicillin and Amikacin, 7 days). Leukocytosis (37.420/mm3) and a shift to the left on the differential (1% myelocytes, 1% metamyelocytes, 4% immature granulocytes, 71% mature granulocytes) were seen on the blood count and C reactive protein was elevated – 13,6 mg/L (N<5 mg/L). Oxygen and pressure need decreased continuously, so that the CPAP support was removed after 3 days, also the umbilical line, CRP decreased to 5,13 mg/L, the infant had a good enteral tolerance and was transferred to the Premature Infants Department. Blood culture performed in the first hours of life came back negative, while yeasts grew in the gastric aspirate collected at birth and in the pharyngeal exudate in the 4th day of life. The infant was fed by gavage with maternal milk, freshly

expressed by the mother every 3 hours, supplemented by formula without lactose in the first 3 days and special formula for preterm infant afterwards.

The clinical course was uneventful until the 9th day of life when the preterm infant suddenly presented with altered general status, large gastric residuals, abdominal distension with mild edema of the abdominal wall (Figure 1), tachycardia, mild hypotonia, and fever. Enteral feedings were interrupted and total parenteral nutrition was started. Antibiotics (gentamycin and meronem) were started intravenously. Laboratory tests revealed elevated C reactive protein (250 mg/L), leukopenia (3470/mm3, 53.3% mature granulocytes), immature/total neutrophil ratio was 0.32, and abdominal radiography showed a tendency to hydroaeric levels in the abdominal flanks (Figure 2). Blood culture, repeated at this time, showed no bacterial growth. At the same time, the mother presented diarrhea and altered general status, being diagnosed with CD infection.

In the next 24-48 hours, the infant continued to show and altered general status, pallor, abdominal distension, bilious gastric residuals, intermittent fever, stools with mucus and blood. Stool tested positive for A and B CD toxins and negative for Shigella and Salmonella. Colistin, vancomycin and nystatin were added in the antibiotic therapy (replacing gentamycin) and total parenteral nutrition was continued.

Irritability and respiratory distress syndrome (mainly due to intense abdominal distension), sub-occlusion, and intermittent elevations of the temperature occurred in the next days so that in the 14th day of life the infant was intubated and mechanically ventilated. After infectious disease consult, review of the clinical, laboratory, and imagistic data, the preterm infant was diagnosed with toxic megacolon and septic shock, and the antibiotic treatment was changed again – meronem, teicoplanin and vancomycin -, intravenous immunoglobulin and pentoxifylline were added to therapeutic plan, total parenteral nutrition and mechanical ventilation were continued. C reactive protein remained at high values the next days while all other laboratory tests were in normal limits (creatinine, blood urea nitrogen, liver function tests, blood gas analysis). In the 24th day of life abdominal radiological survey showed gigantic colic distension and pending hydroaeric levels in the lower abdomen consistent with increased abdominal distension (with increased need of oxygen and pressure on the ventilator) (Figure 3), occurrence of collateral abdominal circulation, persistent sub occlusion.

With a diagnosis of pseudomembraneous colitis, possible secondary to CD infection, and suspected peritonitis, the infant was submitted to pediatric surgery. Laparotomy performed the same day revealed thickened peritoneum, loss of its shiny luster, air and fecaloid fluid in the peritoneal cavity, lavage and drainage were performed and repeated regularly. Complex antibiotic and antifungal therapy, mechanical ventilation and total parenteral nutrition were continued the next month. The infant was extubated after 2 weeks, the drainage tube was removed after one month, enteral nutrition was initiated and gradually increased after 26 days. Blood cultures, peripheral cultures, cultures from stools, and repeated cultures from the drainage fluid showed no bacterial growth.

The infant was submitted to Premature Infant Department after 44 days with 2350 g for nutritional recovery. The next 3 weeks were complicated by an umbilical fistula and an associated inflammatory syndrome but finally the fistula spontaneously closed, the infant grew up, and has been discharged on special preterm formula.

A final diagnosis of necrotizing enterocolitis with secondary peritonitis was formulated at discharge.

Discussion

The presented case is illustrative for difficulties encountered in neonatal gastro-intestinal tract conditions, as NEC. Difficulties had started with correct diagnosis and timely decision as regards the need for surgical intervention. Obviously, another major difficulty was to find the correct antibiotic therapy. Decisions regarding antibiotic therapy had been hampered at least by two things: failure to isolate an etiologic agent and concurrent infection of mother and infant with CD. As most of the literature shows that colonization with CD is frequent in the neonatal intensive care unit and in preterm infants and only very few cases of infection had been cited, it was the initial clinical course that suggested that CD may be the etiological agent in the presented case therefore vancomycin had been one of the antibiotic used. But evidence was not convincing for the neonatologist-pediatric infection disease specialist team, so other antibiotics were added and antibiotic strategy was changed many times resulting in antibiotic overuse..

Conclusions

The final correct diagnosis is still unclear since NEC rarely presents only with peritonitis. Fortunately, the infant survived and has a good clinical evolution up to one year. Given the increased incidence of CD in the latest period, we do have to reconsider the role of CD as a potential etiological agent in neonatal infections.

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