Virchows Arch (2012) 460:123–127
DOI 10.1007/s00428-011-1183-x

CASE REPORT

Eosinophilic myenteric ganglionitis as a cause of chronic intestinal pseudo-obstruction

Ariadne H. A. G. Ooms · Joanne Verheij · Jessie M. Hulst · John Vlot · Cynthia van der Starre · Lissy de Ridder · Ronald R. de Krijger

Received: 13 August 2011 / Revised: 30 November 2011 / Accepted: 5 December 2011 / Published online: 16 December 2011

Abbreviations

Anti-SMA	Anti-smooth muscle actin
CIPO	Chronic intestinal pseudo-obstruction
ENS	Enteric nervous system
GINMD	Gastrointestinal neuromuscular disorders
HPF	High power field
ICC	Interstitial cells of Cajal
IL-5	Interleukin-5

Introduction

Chronic intestinal pseudo-obstruction (CIPO) represents a rare and severe condition characterized by failure of the intestinal tract to propel its contents normally [1, 2]. The condition presents with clinical features such as abdominal pain, vomiting, distended abdomen, constipation, and diarrhea [1, 2]. These symptoms resemble an intestinal mechanical obstruction in the absence of a demonstrable lesion occluding the gut. It is thought that this can result from disturbance of the intestinal motor function, supplied by the enteric nervous system (ENS). The neurons of the ENS are contained in two groups of ganglia: the myenteric (Auerbach's) and the submucosal (Meissner's) plexuses.

The ENS has the unique ability to control most gut functions, such as regulating vascular tone, secretion/absorption, and gut motility [3, 4]. Given these important functions of the ENS, it is not surprising that damage to the ENS results in digestive disorders and reduced quality of life. Human and experimental evidence indicates that inflammation can occur in the ENS, resulting in severe intestinal motor impairment. Inflammation of the ENS has indeed been observed in some cases of CIPO [3, 5, 6].

Many efforts have been made to classify CIPO. Based on histological examination, CIPO may be categorized as primary, secondary, or idiopathic in nature [1]. Primary CIPO can be classified into three major categories of gastrointestinal neuromuscular disorders (GINMD): mesenchymopathies, myopathies, and neuropathies, depending on the predominant involvement of interstitial cells, smooth muscle cells, or enteric neurons, respectively [1, 7]. The enteric neuropathies underlying CIPO can be subdivided into two major entities: (a) degenerative neuropathies, without any evident inflammatory response and (b) inflammatory neuropathies, referred to as myenteric ganglionitis [3, 8]. Inflammation within the myenteric ganglia is a recognized primary cause of CIPO, but the mechanisms through which the dysfunction occurs and the mechanisms leading to enteric neuropathies remain poorly understood [3, 4]. In this case report, we present the first male neonate with eosinophilic myenteric ganglionitis

A. H. A. G. Ooms (✉) · R. R. de Krijger
Department of Pathology, Josephine Nefkens Institute,
Erasmus MC—University Medical Centre,
P.O. 2040, 3000 CA, Rotterdam, The Netherlands
e-mail: a.ooms@erasmusmc.nl

J. Verheij
Department of Pathology, AMC,
Amsterdam, The Netherlands

J. M. Hulst · L. de Ridder
Department of Pediatric Gastroenterology, Erasmus MC—Sophia,
Rotterdam, The Netherlands

J. Vlot
Department of Pediatric Surgery, Erasmus MC—Sophia,
Rotterdam, The Netherlands

C. van der Starre
Neonatal and Pediatric ICU, Erasmus MC—Sophia,
Rotterdam, The Netherlands

underlying CIPO and report his successful recovery following steroid treatment.

Clinical history

A boy was born prematurely at 25 weeks through a primary Caesarean section, weighing 940 grams (>−2.5 SD). Two weeks later he suffered from necrotizing enterocolitis for which he underwent a right hemicolectomy as well as resection of 10 cm of jejunum. However, in the months following surgery, the infant continued to have poor feeding tolerance with a distended abdomen and severe constipation. At 6 months of age, the boy weighed 4.1 kg (−2.5 SD), despite total parental feeding. At 4 months of age, he developed signs of obstruction (distended abdomen, vomiting, and gastric retentions) and a laparotomy was performed. During surgery, there was no stricture at the ileocolonic anastomosis. Although there was no stricture, a milk curd was found proximal to the anastomosis and the anastomosis was resected. Rectal full thickness biopsies were taken to rule out Hirschsprung's disease. The laboratory results showed a variable number of eosinophils (0.1×10^9/l to 1.0×10^9/l) and during a short period of time peripheral eosinophilia was present until treatment started. Laboratory results further showed a normal amount of serum IgE and tests for antinuclear and smooth muscle autoantibodies were negative. Because of the feeding difficulties, the patient was treated with an amino acid-based infant formula for 4 months. Major improvement of the infant's condition only occurred after starting anti-inflammatory treatment, using intravenous prednisone and subsequently oral beclomethasone 200 mg three times daily. Within 1–2 weeks after starting the beclomethasone, enteral nutrition could be increased to complete enteral feeding. After 6 months of hospitalization, the patient was discharged in good condition. Follow-up for 7 months after discharge showed that the patient still needs corticosteroids and steroid-containing enemas have been added to the therapy during flares of the gastrointestinal problems, which has a positive effect on his feeding tolerance. After 18 months of follow-up after hospitalization, the child eats normally and does not need any medication.

Materials and methods

Patient

Informed consent for publication was obtained from the parents.

Histological methods

A hemicolectomy specimen as well as 10 cm of jejunum was received. Later, full thickness rectal biopsies were taken during surgery for a pseudo-obstruction as well as the anastomosis created during the previous surgery. Histological sections were prepared and examined by routine methods. Enzyme histochemistry with acetylcholinesterase was performed on frozen sections of the biopsies to exclude Hirschsprung's disease. Immunohistochemistry using CD117 was applied on formalin-fixed paraffin-embedded sections form the resection specimen to demonstrate the presence of interstitial cells of Cajal (ICCs). The number of eosinophils per high power field (HPF, i.e., 400×) was counted in the resection specimens.

On the same slides, we performed immunohistochemical analysis with PGP9.5 to demonstrate the presence of enteric neurons. In addition, we applied anti-smooth muscle actin (anti-SMA) to demonstrate the presence of a normal layer of smooth muscle.

Results

Histological findings

In the biopsies, there was no evidence of hypo- or aganglionosis. Histological examination of the resected bowel segment showed a striking presence of eosinophils, especially in the myenteric plexus with affinity for the neurons and ganglion cells (range 8–43/HPF; mean 16/HPF) (Fig. 1). This was located in the ileum as well as in the colon. There was no significant increase of lymphocytes surrounding the ganglion cells as in lymphocytic ganglionitis. Retrospectively, histological examination of slides of the previously removed necrotic bowel also showed the presence of eosinophils with affinity for the ganglion cells.

Immunohistochemical findings

Immunohistochemistry with CD117 showed an apparently normal c-Kit labeling pattern, since we found CD117 positive cells in the colon as well as in the ileum. These were located in the myenteric plexus, in the submucosa, and interspersed in the muscle layer. This indicated a normal number of interstitial cells of Cajal (Fig. 2a, b). Immunohistochemistry with acetylcholinesterase showed a normal amount of acetylcholinesterase-positive neuronal fibers in the muscularis mucosae and

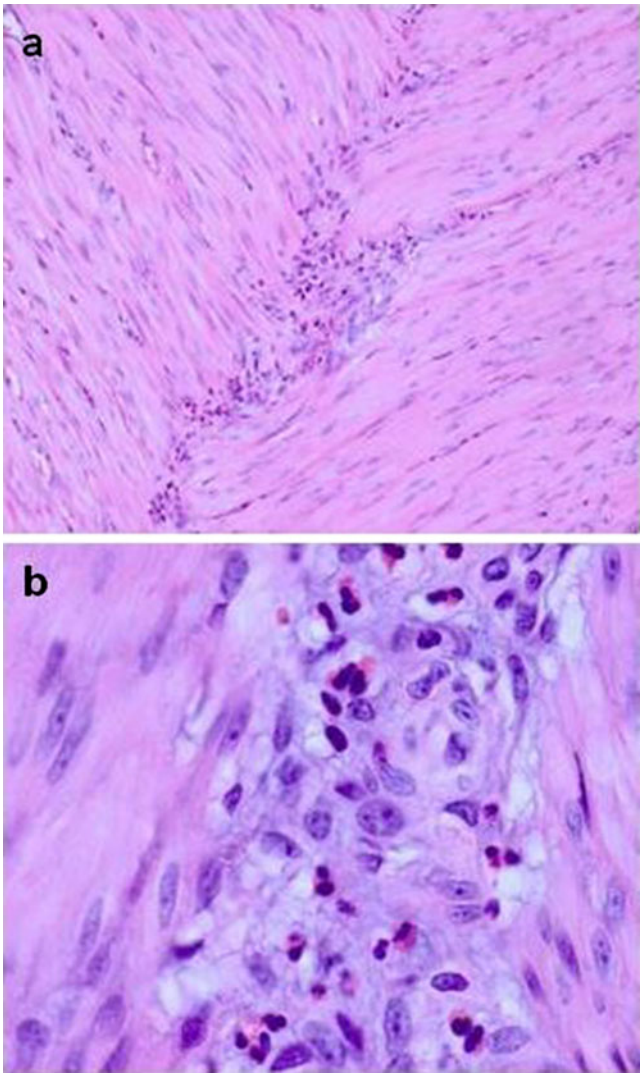

Fig. 1 Hematoxylin/eosin (H/E)-stained sections of full thickness biopsies from our patient demonstrating the presence of eosinophils in the myenteric plexus. **a** Overview of muscular layer with myenteric plexus. **b** Detail of myenteric plexus with infiltrating eosinophilic granulocytes between the ganglion cells and neurons without obvious cellular destruction

lamina propria, as well as in the submucosal neurons (data not shown).

Immunohistochemical analysis with PGP9.5 showed the presence of an apparently normal number of enteric neurons, even with the presence of eosinophils in the immediate vicinity (Fig. 2c). This indicates that the eosinophilic infiltrate did not lead to degeneration of the neurons. In addition we used immunohistochemical analysis with antibodies against smooth muscle actin (anti-SMA) to show there was a normal amount of smooth muscle cells (Fig. 2d).

Discussion

We describe the second neonatal case of eosinophilic myenteric ganglionitis, causing a rare and highly morbid functional intestinal obstruction syndrome, referred to as CIPO [2]. This represents a rare syndrome characterized by impaired gastrointestinal propulsion, which causes symptoms of gastrointestinal obstruction, in the absence of any lesion occluding the intestinal lumen [2]. The intestine depends on the ENS for its functionally important patterns of movement [9]. Current evidence indicates that alterations in the ENS, including functional impairment of neurons, are associated with uncoordinated motor activities, resulting in altered transit of intestinal contents potentially leading to CIPO [8].

Schappi et al. [2] described three cases, all female in contrast to our patient. The three cases described were aged 1 month, 11 and 15 years. The clinical presentation of the first two patients is similar to our case: vomiting, constipation, and marked abdominal distension for several days. The third case had a more acute event. The neonate also experienced a peripheral blood eosinophilia (1.15×10^9/l) as did our case before he was treated. However, in our case there was no elevation in serum IgE, as was described in the first two cases. Smooth muscle and antinuclear autoantibodies were weakly positive in the neonatal case, not described in the second, and negative in the third case as in our patient. In our patient no anorectal inflammatory manometry was performed.

In our patient the clinical picture was obscured at first by necrotizing enterocolitis in this premature infant. However, after surgery the patient continued to have feeding difficulties. The patient developed signs of a gastrointestinal obstruction, but during surgery this turned out to be a pseudo-obstruction. When examining the bowel specimen, it was noticed that there was an eosinophilic myenteric ganglionitis. The eosinophils were specifically located around the ganglion cells, with some scattered eosinophils in the submucosa and tunica muscularis. In the second and third cases described by Schappi et al. there seems to be a more extensive involvement of all layers of the bowel, but with a larger collection of eosinophils around the myenteric plexus. Unfortunately, they do not provide a count of the eosinophils. IL-5, a potent eosinophil chemoattractant, has been suggested as a humoral factor playing a role in gut dysmotility. IL-5 expression by the myenteric ganglions was demonstrated by Schappi et al. in his patients with eosinophilic myenteric ganglionitis. Unfortunately we did not have the means to demonstrate IL-5 expression in our patient.

In the neonatal case and the second case of Schappi et al. Hirschsprung's disease was considered, as was done in our case. Full thickness rectal biopsies in those children and in

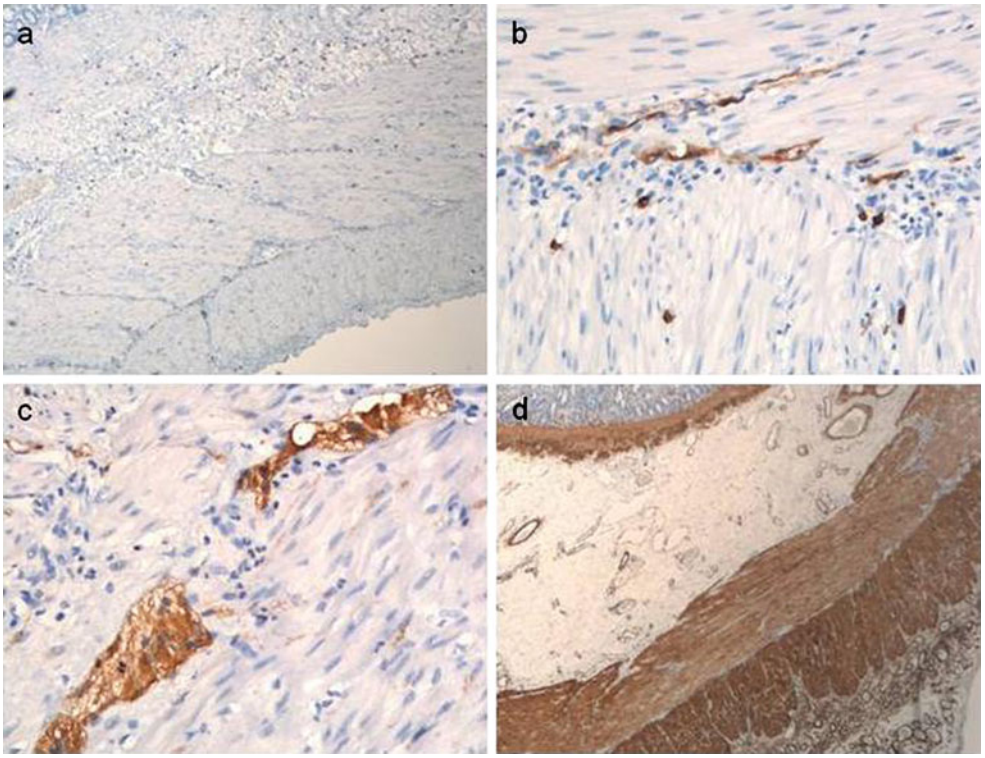

Fig. 2 Immunohistochemistry on sections of the resection specimen from our patient. **a**, **b** Overview (**a**; 40×) and detail (**b**; 200×) of a slide form the bowel resection stained for CD117, highlighting the ICCs, with an apparently normal distribution and number. **c** Detail (200×) of the myenteric plexus stained for PGP9.5, highlighting the neurons. An apparently normal number can be appreciated. **d** Overview (40×) of a slide stained for anti-SMA, in which a normal pattern of smooth muscle cells can be seen

our case also showed the presence of normal ganglia and neurons within the submucosal and myenteric plexus, as well as a normal acetylcholinesterase staining pattern. In all cases described as well as in our case, there was no significant lymphocytic infiltrate.

We applied immunohistochemistry with CD117 antibodies to analyze the number of interstitial cells of Cajal, which are thought to be required for normal gut motility and are also implicated in some forms of GINMD. ICCs are found widespread throughout the gastrointestinal tract, distributed along a variety of locations. In the colon, distinct networks can be found in the myenteric plexus and more loosely in the submucosa. In the small bowel, the same networks can be found in the myenteric plexus and in addition more loosely in the deep muscular plexus. Single ICCs can also be found interspersed between muscle cells in the muscle layer [10, 11]. CD117 also stains mast cells, which were excluded by comparing with the H&E and by looking at the contour of the cell, since mast cells do not have processes whereas ICCs do [10, 11]. Until now, there are no normal reference values for the range of ICC numbers when applying immunohistochemistry. Most reports use immunofluorescence on frozen tissue samples instead of immunohistochemistry on paraffin-embedded material [10, 11]. It is believed that immunohistochemistry does not detect all ICCs [10]. Therefore, we examined our slides for the presence of the ICCs in the previously named locations, especially in those with the presence of eosinophils. In addition we used PGP-9.5 and anti-SMA to demonstrate the presence of a normal amount of enteric neurons and smooth muscle cells, respectively. These immunohistochemical stainings also showed a normal amount of neurons and smooth muscle cells.

By excluding the above-mentioned cells as a cause of the pseudo-obstruction, it appears more likely that there is a functional rather than anatomically detectable cause for the pseudo-obstruction in this patient. However, we cannot entirely exclude this, as we have not performed quantitative analysis of the enteric neurons. Similar to the patients demonstrated by Schappi et al. there was no clinical improvement in the gastrointestinal dysmotility syndrome with an amino acid-based formula as the sole source of nutrition for at least 3 months in our patient as compared to 2 months in the neonatal case described by Schappi et al. However, all patients, including our patient, did respond rapidly to steroids [2]. We used the same steroid (beclomethason) in the same dosage as in the previously published neonatal case. The other cases were treated with (methyl)prednisolone and azathioprine. Our patient could tolerate complete enteral feeding within 1–2 weeks. In contrast to the previously published patients, our patient is completely without medication after 18 months of follow-up, not requiring a maintenance dose of steroids.

In conclusion, this case illustrates that a GINMD, such as myenteric ganglionitis, should be considered in patients with CIPO, since it may be treated with an elemental diet and steroids.

Conflict of interest statement We declare that we have no conflict of interest.

References

1. Antonucci A, Fronzoni L, Cogliandro L, Cogliandro RF, Caputo C, De Giorgio R, Pallotti F, Barbara G, Corinaldesi R, Stanghellini V (2008) Chronic intestinal pseudo-obstruction. World J Gastroenterol 14(19):2953–2961
2. Schappi MG, Smith VV, Milla PJ, Lindley KJ (2003) Eosinophilic myenteric ganglionitis is associated with functional intestinal obstruction. Gut 52(5):752–755
3. De Giorgio R, Camilleri M (2004) Human enteric neuropathies: morphology and molecular pathology. Neurogastroenterol Motil 16(5):515–531. doi:10.1111/j.1365-2982.2004.00538.x
4. Lacy BE, Weiser K (2006) Gastrointestinal motility disorders: an update. Dig Dis 24(3–4):228–242. doi:10.1159/000092876
5. De Giorgio R, Barbara G, Stanghellini V, De Ponti F, Salvioli B, Tonini M, Velio P, Bassotti G, Corinaldesi R (2002) Clinical and morphofunctional features of idiopathic myenteric ganglionitis underlying severe intestinal motor dysfunction: a study of three cases. Am J Gastroenterol 97(9):2454–2459. doi:10.1111/j.1572-0241.2002.06002.x
6. Khan WI, Collins SM (2006) Gut motor function: immunological control in enteric infection and inflammation. Clin Exp Immunol 143(3):389–397. doi:10.1111/j.1365-2249.2005.02979.x
7. Knowles CH, De Giorgio R, Kapur RP, Bruder E, Farrugia G, Geboes K, Lindberg G, Martin JE, Meier-Ruge WA, Milla PJ, Smith VV, Vandervinden JM, Veress B, Wedel T (2010) The London Classification of gastrointestinal neuromuscular pathology: report on behalf of the Gastro 2009 International Working Group. Gut 59(7):882–887
8. Di Nardo G, Blandizzi C, Volta U, Colucci R, Stanghellini V, Barbara G, Del Tacca M, Tonini M, Corinaldesi R, De Giorgio R (2008) Review article: molecular, pathological and therapeutic features of human enteric neuropathies. Aliment Pharmacol Ther 28(1):25–42. doi:10.1111/j.1365-2036.2008.03707.x
9. Furness JB (2008) The enteric nervous system: normal functions and enteric neuropathies. Neurogastroenterol Motil 20(Suppl 1):32–38. doi:10.1111/j.1365-2982.2008.01094.x
10. Farrugia G (2008) Interstitial cells of Cajal in health and disease. Neurogastroenterol Motil 20(Suppl 1):54–63
11. Gomez-Pinilla PJ, Gibbons SJ, Sarr MG, Kendrick ML, Shen KR, Cima RR, Dozois EJ, Larson DW, Ordog T, Pozo MJ, Farrugia G (2011) Changes in interstitial cells of cajal with age in the human stomach and colon. Neurogastroenterol Motil 23(1):36–44. doi:10.1111/j.1365-2982.2010.01590.x, Epub 02010 Aug 01519