Austin Journal of Clinical Pathology

Open Access

Austin Publishing Group

Review Article

Hirschsprung Disease; Insights into Developmental Etiology, Pathophysiology and Postoperative Long-Term Outcomes

Abdelrahman ST[1], Ezzat TA[1], Saleh MI[1], Sakr SM[1], Abdeldayem MS[1], Alalfy AA[1], Ayad MA[1], Abdelshafy AM[1]* and Domouky AM[2]

[1]Third Year Medical Student, Faculty of Medicine, Zagazig University, Egypt

[2]Department of Human Anatomy & Embryology, Faculty of Medicine, Zagazig University, Egypt

***Corresponding author:** Aliaa M Abdelshafy, Third Year Medical Student, Faculty of Medicine, Zagazig University, Egypt

Received: December 02, 2021; **Accepted:** January 06, 2022; **Published:** January 13, 2022

Abstract

Hirschsprung disease, or aganglionic megacolon, is a devastating neurocristopathy characterized by the absence of the enteric nervous system in the distal bowel, resulting from a failure of migration of the neural crest cells during embryological development. The enteric nervous system's vital role is demonstrated in children with Hirschsprung disease, in which absence of the enteric nervous system in even a few centimeters of the intestine can be lethal. Functional obstruction results from the absence of neural mediated propulsive motility and persistence of bowel contraction. The disease occurs in about 1 in 5000 children. Hirschsprung disease is a multifactorial disorder influenced by both genetic and environmental factors and is associated with several hereditary disorders. Children with Hirschsprung disease have symptoms of bowel obstruction or perforation, abdominal distention, bilious vomiting, growth issues, fatigue, severe constipation, along with an increased risk of Hirschsprung-associated enterocolitis that presents the leading cause of morbidity. Current management necessitates surgery; nevertheless, many individuals experience long-term complications. This article aims to review the association between the basic science principles of Hirschsprung disease and its predisposing factors, disease mechanisms, clinical manifestations, together with its ongoing functional problems after surgical correction. Insights into the genetic basis and mechanisms of the disease might pave the way for new diagnostic and treatment measures hereafter.

Keywords: Hirschsprung disease; Enteric nervous system; Hindgut; Neural crest cells; Aganglionic megacolon; RET; EDNRB; HAEC

Introduction

Hirschsprung disease (HSCR), a life-threatening birth defect, is a multifactorial genetic disorder and neuro-congenital condition of aganglionosis along the distal colon ascribable to a defect in migration of neural crest cells towards the hindgut, leading to functional intestinal obstruction [1-3]. The developmental anomaly that describes Hirschsprung disease has various clinical manifestations and a complex etiology, making diagnosis and management mysterious. Disturbances in intestinal function, including Hirschsprung-associated enterocolitis (HAEC), can be deadly even after reconstructive surgery [4,5].

Most published studies have focused on surgical management of the disease; however, little interest has been given to the disorder's anatomic background and multigenic nature [6,7]. It is essential to shed light on the malformation's anatomic and embryological features, incidence, multi-genetic nature, pathophysiology, clinical picture, and long-term complications after corrective surgery aiming to clarify how current studies provide insight into embryological defects and pathophysiology of Hirschsprung disease which might lead to novel regenerative approaches to reduce disease morbidity.

Developmental Anatomy and Embryology

The gastrointestinal (GI) tract is endodermal in origin, beginning at the mouth and terminating at the anus. The fetal GI tract is divided into three segments: foregut, midgut, and hindgut (Figure 1) [8,9]. Distal part of the hindgut consists of the cloaca, which gets separated into a dorsal anorectal compartment and a ventral urogenital compartment by a septum [10]. The hindgut differentiates to give rise to the distal one-third of the transverse colon, descending colon, sigmoid colon, rectum, superior anus (down to the dentate line in the anorectum), and urinary epithelium (Figure 1) [8,11,12]. Its arterial supply comes mainly from the inferior mesenteric artery (IMA) which arises from the anterior or left anterolateral aspect of the abdominal aorta. The venous and lymphatic drainage correspond to its arterial supply [11]. The IMA classically terminates into three branches. From proximal to distal, these branches are the left colic artery supplying the descending colon, the sigmoid artery supplying the sigmoid colon, and the superior rectal artery supplying the rectum and upper part of the anal canal [13-15]. The nerve supply of the hindgut is represented by intrinsic and extrinsic innervation. The enteric nervous system (ENS) represents the intrinsic innervation, while the extrinsic innervation of the hindgut is mediated through sympathetic and parasympathetic nerves. Sympathetic fibers originate from T5 to L2 levels, and parasympathetic fibers originate from the dorsal vagus complex and pelvic nerves S2-S4 [11].

The GI tract responds to the state of the lumen and gut wall by

Citation: Abdelrahman ST, Ezzat TA, Saleh MI, Sakr SM, Abdeldayem MS, Alalfy AA, et al. Hirschsprung Disease; Insights into Developmental Etiology, Pathophysiology and Postoperative Long-Term Outcomes. Austin J Clin Pathol. 2022; 9(1): 1077.

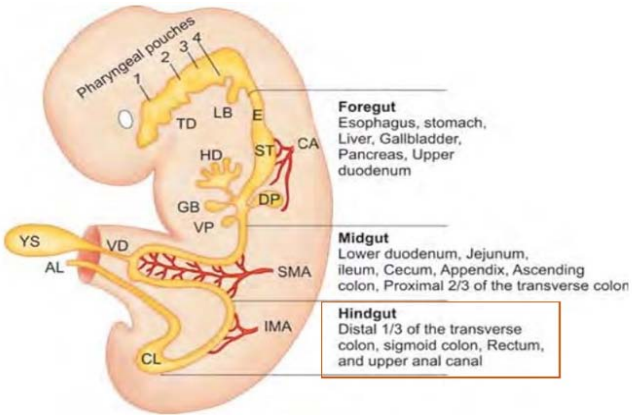

Figure 1: The fetal Gastrointstinal tract divided into foregut, midgut, and hindgut [8].

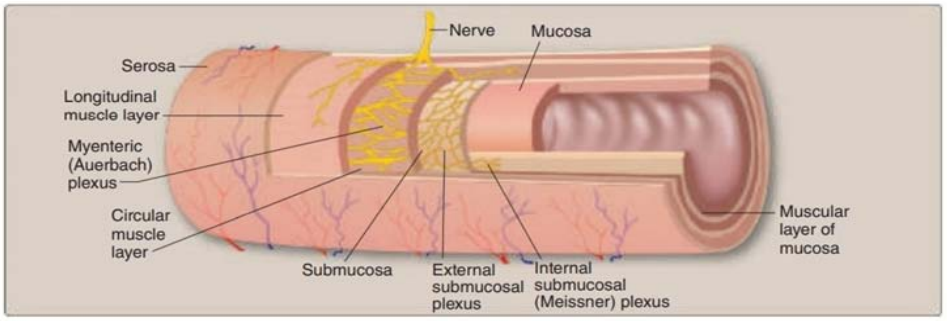

Figure 2: Wall of the Gastrointestinal tract showing the intrinsic nerve supply, the enteric nervous system, consistig of the myenteric (Auerbach's) as well as the submucosal (Meissner's) plexus [17].

secretions, activating peristalsis, and controlling blood flow and thus maintain suitable physiological balance. This potential depends on the presence of a well-functioning enteric nervous system [16]. The formation of a well-functioning ENS entails coordination of the survival, migration, proliferation, and differentiation of its progenitor cells within the GI tract. The enteric nervous system is derived from a multipotent, migratory cell population called the neural crest cells (NCCs); besides, it comprises the part of the autonomic nervous system which is responsible for direct control of the GI tract [9]. The ENS is formed of an extensive network of neurons and glia within the wall of the intestine [17]. Within the small and large intestines, enteric neurons are arranged into two main plexuses, the myenteric (Auerbach's) as well as the submucosal (Meissner's) plexus (Figure 2) [18]. The myenteric plexus clusters into ganglia between the circular and longitudinal muscle layers, while the submucosal plexus is present within the connective tissue of the submucosa. Some neurons might also be found within the mucosa [19].

Embryogenesis of the Enteric Nervous System

Enteric nervous system neurons and glia are derivatives of the vagal segment of the neural crest. Vagal segment of neural crest cells migrates along the path of the vagus nerves, penetrate the foregut mesenchyme, and spread cranio-caudally across the gastrointestinal tract. This takes 7 weeks in humans to be completed. By the 5th, 7th, and 8th weeks, neural crest derivatives have infiltrated the foregut, distal ileum, and mid-colon, respectively, infiltrating the myenteric plexus before the submucosal plexus. By the 12th week of gestation, ENS derivatives have colonized the entire colon [1]. Following vagal neural crest-derived cells' colonization, sacral level neural crest cells also contribute to the colonic ENS. However, even in the most distal regions of the bowel, most enteric neurons are vagally derived [20]. Enteric neural crest- derived cells (ENCCs) simultaneously proliferate and migrate towards and within the gut [21,22]. Furthermore, a subpopulation of ENCCs starts to undergo neuronal differentiation. ENCCs' colonization of the intestine necessitates coordinated proliferation, migration, and neuronal differentiation [23]. Any defect in such development gives rise to disorders termed neurocristopathies. One such disorder is Hirschsprung disease which is characterized by absence of the ganglion cells in the myenteric (Auerbach's) and submucosal (Meissner's) plexuses of the distal intestine [3,9,24]. The essential role for enteric neurons in peristalsis is demonstrated by the bowel obstruction that occurs in the aganglionic region of GIT [23]. In children with Hirschsprung disease, there are two main hypotheses on how this mechanism is disrupted. The ganglion cells either never enter the distal intestine because they mature or differentiate into ganglion cells earlier than they should, or they reach the distal intestine but do not survive or proliferate [25].

Classification of Hirschsprung Disease

According to the distal extent of aganglionosis, Hirschsprung disease is subdivided into ultrashort (distalmost rectum), short

(rectosigmoid), and long segment (proximal to splenic flexure) subtypes [24,26,27]. Ultra- short segment Hirschsprung disease is defined by the presence of an aganglionic rectal segment less than 1-2 cm in length [28]. The finding of aganglionosis in the rectum with a transition at the rectosigmoid junction indicates short-segment Hirschsprung disease, and it can sometimes extend into the rectosigmoid region [29]. Long-segment Hirschsprung disease is further subcategorized into long-segment colonic aganglionosis, total colonic aganglionosis, or total colonic and small bowel aganglionosis [30]. Long- segment colonic aganglionosis is the extension of aganglionosis from the rectum to the level of the transverse colon [31]. Total colonic aganglionosis and small bowl aganglionosis, also known as extensive aganglionosis, are the least common and extend beyond 30-50 cm proximal to the ileocecal valve [27,30].

Incidence

Every 1 in 5000 live births is diagnosed with HSCR [32,33]; however, this does not account for multi-ethnic differences. Moreover, the incidence in populations with high consanguinity rates can be significantly higher [1]. Concerning subcategories of Hirschsprung disease, short-segment Hirschsprung disease is the most common presentation, comprising 60%-85% of cases [34]. Long segment colonic aganglionosis constitutes approximately 10% of all Hirschsprung disease. Total colonic aganglionosis incidence is approximately 5% [35]. The male-to-female ratio is variable. According to Best et al. [36] male sex increases the risk of HSCR by about 2 folds, while it is estimated to be 4 to one times more common in males than females as suggested by Thakkar and Curry [37]. The male to female ratio is also affected by the length of the aganglionic segment, being a 1:1 ratio rather than 4:1 in those with a total colonic aganglionosis.

Pathophysiology

Normal motility of the GIT is primarily controlled by the enteric nervous system, known as the second brain. Its activation mostly leads to intestinal smooth muscle relaxation, which is mediated by nitric oxide (NO) as well as other neurotransmitters. Therefore, when the enteric ganglia are absent as in Hirschsprung disease, the bowel remains tonically contracted, due to the effect of the extrinsic cholinergic fibers that are mainly excitatory, leading to functional obstruction of the affected segment [1,38].

Furthermore, it is becoming increasingly clear that the etiology of the disease extends beyond the aganglionic area. Imbalance in the expression of important neurotransmitters and changes in neuronal density in the colon proximal to the aganglionic site are thought to contribute to bowel dysfunction and dysmotility, as well as the persistence of bowel symptoms in some patients, even after corrective surgery [39].

Coyle et al reported minimal expression of NO in the aganglionic area, as opposed to higher expression in the proximal ganglionic bowel, which may contribute to the unopposed muscle spasm. Conversely, choline acetyltransferase (CHAT) expression was higher in the aganglionic bowel but lower in the proximal dilated part [40]. Additionally, neuronal density is markedly lowered in the proximal part of the colon, and the ganglia in this area are smaller and more isolated, while the nerve cell bodies are increased in size [39,41].

The pathological picture of the colon in this disease consists of 3 zones. The first is the aganglionic zone that is spastic, making the passage of stool or gas difficult. The second zone is the dilated area proximal to the diseased site, caused by the accumulation of the intestinal contents due to the functional obstruction and failure of peristalsis. The third zone is 1-2 inches between the 2 areas and is called the transitional zone [42].

Etiological Factors

HSCR is a multifactorial disorder that is influenced by both genetic and environmental factors. It is thought to be caused by mutations in several genes, which may explain its diverse characteristics [43,44]. The RET proto-oncogene, as well as other associated genes like glial cell line-derived neurotrophic factor (GDNF), are the most frequently found gene mutations in Hirschsprung disease. Endothelin-3 and endothelin-B receptor genes are different gene families that also cause Hirschsprung disease [25]. There is a well-established association between HSCR and Down Syndrome (trisomy 21), with the reported incidence being 2-15% [45]. Ophthalmological, auditory, genitourinary, cardiac, and neurological anomalies are found in up to 60% of patients with HSCR. Most cases of HSCR are sporadic in nature, while the rest are familial [37].

As discussed before, HSCR is caused by failure of migration of neural crest cells to the distal colon. Mutations in any of the genes responsible for the migration, proliferation, differentiation, or survival of ENCCs can result in failure of ENS development, leading to HSCR phenotype [44,46]. Around 20% of HSCR cases are familial, while the rest are sporadic. The pattern of inheritance is complex, and the penetrance is variable.

Heuckeroth [46] states that the pattern of inheritance is non-mendelian, while Tjaden & Trainor [9] suggest that the genes show an autosomal dominant pattern. Extensive research has recognized a number of genes that are involved in the pathogenesis of Hirschsprung disease, including RET, GDNF, GFRα1, NRTN, EDNRB, ET3, PHOX2b, SOX10, and SHH [9].

In addition, 10% of cases have a chromosomal/syndromic association, including Waandenburg-Shah syndrome (loss of pigmentation and defective hearing) [47], Congenital central hypoventilation syndrome (insufficient breathing or apnea during sleep) [48], Mowat-Wilson syndrome (intellectual disability, facial dysmorphism, delayed development, and Hirschsprung disease) [49], or other malformations, such as Meckel diverticulum, Imperforate anus, Ventricular septal defect, Neuroblastomas, or Cryptorchidism [1,9,46,50]. Down syndrome (Trisomy 21) is one of the most common associations with the disease. Children with Down syndrome are at a 100-fold higher risk of developing HSCR than the normal population, as many genes are triplicated in the Hirschsprung disease risk region of chromosome 21 [1,46].

The most commonly identified gene involved in the pathogenesis of HSCR is the receptor tyrosine kinase or RET gene, which is a proto-oncogene present on chromosome 10 [1]. Inactivating mutations of this gene account for about 50% of familial cases of HSCR and about 15-20% of sporadic cases. Very rarely, activating RET mutations can cause congenital megacolon associated with MEN2. So and colleagues [51] explained that RET common variants are associated

with the common manifestations of the disease (sporadic, male, short-segment aganglionosis), while the rare coding sequences are involved in the less frequently encountered forms (familial, female, long/total colonic aganglionosis).

Glial cell line-derived neurotrophic factor (GDNF) and Neurturin (NRTN or NTN) are RET ligands [46]. Patients with heterozygous GDNF mutations have been diagnosed with HSCR, as GDNF is important for activating the RET pathway, that influences migration, proliferation, differentiation, and survival of ENCCs [52]. Rare human cases of HSCR were reported with NTN genes, and some of these occasionally had mutations of the RET gene or other HSCR genes. This suggests a modifier role for NTN in HSCR pathogenesis [53].

Transcription of RET depends on SOX10 and PHOX2B. Mutations of the genes encoding these transcription factors cause syndromic forms of HSCR [46]. SOX10 is a transcription factor that regulates key genes important for ENS, glial, and melanocyte development [9]. Mutations of its gene are associated with Waardenburg-Shah syndrome, where patients suffer from other neurologic phenotypes, such as hearing loss as well as pigmentation defects, and a more severe form involves demyelination of the CNS & PNS. SOX10 mutations are responsible for about 4% of both syndromic and non-syndromic Hirschsprung disease. It was also noted that the male sex is at higher risk of the disease, as the sex-determining region Y (SRY) protein, competes with SOX10 at RET regulatory elements [54,55].

PHOX2B is a transcription factor required for RET expression. It is expressed in migrating ENCCs, enteric neurons, and glial cells. Its mutation accounts for 0.5% of HSCR and is associated with congenital central hypoventilation syndrome (CCHS) and neuroblastoma (NB), which is often referred to as CCHS-HSCR-NB association [56]. Sonic Hedgehog (SHH) is another key regulator of the migration of ENCCs of the gut. SHH has opposing effects to GDNF, so the balance between the two is important for normal enteric plexus positioning [56]. Another family of genes involved in the etiology of Hirschsprung diseases is the EDN3 and EDNRB family. These genes encode proteins that are involved in endothelin signaling, which is also necessary for the normal migration of ENCCs. Mutations of these genes represent 5-10% of cases of HSCR and are also associated with both syndromic (Waardenburg-Shah) and non-syndromic forms of the disease [54,58].

It is obvious that HSCR is a complex disorder that results from the interaction between genetic and environmental factors. Despite the large number of genes that were found to be involved in the disease, they only account for about 50% of the cases [9].

Clinical Presentation

80% of patients have symptoms in the first few months of life [59], while 15% of cases remain undiagnosed until the age of 5 years [60]. Asymptomatic cases until adolescence are rare. The most frequent presentation of HSCR present in a term newborn infant is distal intestinal obstruction (DIO), which manifests as severe constipation, abdominal distention, bilious vomiting, feed intolerance, and failure to thrive. Delayed passage of meconium (no bowel movements for more than 48 hours after birth) is pathognomonic for the disease. However, the normal passage of meconium can be seen in around 40% of infants. Perforation of the cecum or appendix can occur in some neonates [46,61]. Children with Hirschsprung disease may present growth failure, while other children with the disease develop normally even without treatment. The combination of growth failure and abdominal distension (or other Hirschsprung disease symptoms) should raise suspicions, but similar symptoms may also be caused by other medical conditions. Growth deficiency in children with Hirschsprung disease is poorly known; nonetheless, it is most common in children with severe abdominal distension and is thought to be caused by a lack of nutrients [46].

Hirschsprung Disease-Associated Enterocolitis (HAEC)

Enterocolitis, which is defined as an inflammatory disorder of the bowel, is a dangerous clinical symptom complex that can occur both before and after surgical correction of HSCR. The etiology of HAEC is not fully understood. However, several hypotheses like impaired mucosal barrier function [4,62,63], dysbiosis of the intestinal microbiome [64], altered innate immune responses [65], and bacterial translocation have been introduced [67]. Furthermore, it is seen more commonly in patients with long-segment disease and those with Trisomy 21 [67]. Explosive diarrhea (often with blood in the stool), abdominal distension, lethargy, and fever are all manifestations of HAEC [68-70]. When a perirectal examination is performed, a gush of stool and gases are seen. The onset of enterocolitis can be sudden, with symptoms changing abruptly over several hours, or it can be chronic and associated with failure to thrive. Enterocolitis can cause fatal septicemia, that's why early identification of its symptoms is essential. Treatment involves gut rest, fluid resuscitation, broad-spectrum IV antibiotics, and bowel decompression [46].

Ongoing functional problems after surgical correction HSCR treatment are primarily surgical. The procedure is based on the excision of the aganglionic segment and then anastomosing the normally innervated bowel to the anus, just above the anal sphincter, in order to preserve the function of the sphincter for the child to remain focally continent [32].

Some patients may continue to have some long-term complications even after corrective surgery. The most common complications are obstructive symptoms, constipation, enterocolitis, and fecal incontinence [37,71]. Postoperative obstructive symptoms can occur in 8-30% of patients. The obstruction can be mechanical, histopathological, or physiological. Mechanical obstruction can be caused by anastomotic stricture, adhesions, and scarring or bowel torsion proximal to the site of anastomosis. Histopathological obstruction can be caused by residual aganglionosis, usually occurring with long-segment disease [72-75] or acquired aganglionosis, that occurs due to degenerative loss of ganglion cells in the distal pulled through the bowel, possibly by vascular compromise or the scarring associated with the surgery [73,76]. Lastly, physiological obstruction can be caused by anal achalasia (nonrelaxing internal anal sphincter) with elevated internal anal sphincter (IAS) pressures, withdrawal behavior, poor pelvic floor function, or colonic dysmotility [4,77-79]. Patients usually present with abdominal distension, bloating, vomiting, and severe constipation [4,80,81]. Another important complication is fecal incontinence, which occurs in around 1-2% of patients [37]. It can be classified into true incontinence, caused

by iatrogenic damage to the anal sphincter and/or the dentate line, or pseudo-incontinence, due to gross constipation, for example by a chronic large stool burden, or abnormal colonic motility [4,73,77,80,81]. The most dangerous and possibly lethal complication of HSCR is Hirschsprung disease-associated enterocolitis, which can occur in 10-15% of cases [82]. It typically occurs within months following the surgery, so it is relatively uncommon after the age of 5 [4,33]. Although considerably decreased in the past decades, HSCR mortality rate ranges between 1-10%. Newborns and infants are at higher risk of developing life-threatening complications, especially those with cardiovascular anomalies [82]. Reports of the long-term outcomes of the disease are variable. Thakkar & Curry [37] state that 20-30% of adults present with significant diarrhea and incontinence, and that sexual function is sometimes also affected. On the other hand, Sood and colleagues [83] reported that in the absence of any ongoing obstructive symptoms or enterocolitis, the quality of life of teenagers and young adults affected by the disease was comparable to the healthy population. Therefore, postoperative close follow-up is recommended. Families should also be educated for quick recognition and treatment of severe life-threatening complications [82].

Conclusion

Hirschsprung disease, a congenital malformation resulting from aganglionosis along a segment of the distal bowel, leads to impaired colonic motility and complications which decrease the patients' life expectancy rates. It usually manifests with impaired ability to pass meconium, abdominal distension, and discomfort that usually necessitates surgical resection of the affected part of the bowel. This review is important as its increases awareness about this disease and how it affects quality of life in the aim to decrease the incidence of complications, morbidity, and mortality rates. Hirschsprung disease is still challenging to be diagnosed and managed; therefore, we recommend more and more research to be done to further understand the embryologic and genetic basis of the disease in hopes to help the practicing clinicians in the care of individuals with Hirschsprung disease.

References

1. Chhabra S, Harwood R & Kenny SE. Hirschsprung's disease. Surgery (Oxford). 2019; 37: 640-645.
2. Klein M & Varga I. Hirschsprung's disease-recent understanding of embryonic aspects, etiopathogenesis and future treatment avenues. Medicina (Lithuania). 2020; 56: 1-13.
3. Davidson JR, Kyrklund K, Eaton S, Pakarinen MP, Thompson DS, Cross K, et al. Long-term surgical and patient-reported outcomes of Hirschsprung's Disease. Journal of Pediatric Surgery. 2021.
4. Gosain A, Frykman PK, Cowles RA, Horton J, Levitt M, Rothstein DH, et al. Guidelines for the diagnosis and management of Hirschsprung- associated enterocolitis. Pediatr Surg Int. 2017; 33: 517-521.
5. Lewit RA, Veras LV, Cowles RA, Fowler K, King S, Lapidus-Krol E, et al. Reducing Under diagnosis of Hirschsprung- Associated Enterocolitis: A Novel Scoring System. Journal of Surgical Research. 2021; 261: 253-260.
6. Neuvonen MI, Kyrklund K, Lindahl HG, Koivusalo AI, Rintala RJ, Pakarinen MP. A population-based, complete follow-up of 146 consecutive patients after transanal mucosectomy for Hirschsprung disease. J Pediatr Surg. 2015; 50: 1653-1658.
7. Meinds RJ, Van Der Steeg AFW, Sloots CEJ, Witvliet MJ, De Blaauw I, Van Gemert WG, et al. Long-term functional outcomes and quality of life in patients with Hirschsprung's disease. Br J Surg. 2019; 106: 499-507.
8. Kaushal RK. Self Assessment and Review of Anatomy, Jaybee Brothers Medical Publishers. 2018.
9. Butler Tjaden NE & Trainor PA. The developmental etiology and pathogenesis of Hirschsprung disease. Transl Res. 2013; 162: 1-15.
10. Kluth D, Fiegel HC & Metzger R. Embryology of the hindgut. Seminars in Pediatric Surgery. 2011; 20: 152-160.
11. Kostouros A, Koliarakis I, Natsis K, Spandidos DA, Tsatsakis A & Tsiaoussis J. Large intestine embryogenesis: Molecular pathways and related disorders (Review). Int J Mol Med. 2020; 46: 27-57.
12. Kahai P, Mandiga P, Wehrle CJ & Lobo S. Anatomy, Abdomen and Pelvis, Large Intestine. StatPearls. Treasure Island (FL): StatPearls Publishing Copyright © 2021, StatPearls Publishing LLC. 2021.
13. Firetto MC, Lemos AA, Marini A, Avesani EC & Biondetti PR. Acute bowel ischemia: analysis of diagnostic error by overlooked findings at MDCT angiography. Emerg Radiol. 2013; 20: 139-147.
14. Shaikh H, Wehrle CJ & Khorasani-Zadeh A. Anatomy, Abdomen and Pelvis, Superior Mesenteric Artery. StatPearls. Treasure Island (FL): StatPearls Publishing Copyright © 2021, StatPearls Publishing LLC. 2021.
15. Heanue TA & Pachnis V. Enteric nervous system development and Hirschsprung's disease: advances in genetic and stem cell studies. Nature Reviews Neuroscience. 2007; 8: 466-479.
16. Gershon MD. Developmental determinants of the independence and complexity of the enteric nervous system. Trends Neurosci. 2010; 33: 446-456.
17. Leeper-Woodford SK & Adkison LR. Lippincott Illustrated Reviews: Integrated Systems Wolters Kluwer. 2016.
18. Metzger M, Caldwell C, Barlow AJ, Burns AJ & Thapar N. Enteric nervous system stem cells derived from human gut mucosa for the treatment of aganglionic gut disorders. Gastroenterology. 2009; 136: 2214-2225.e1-3.
19. Wang X, Chan AK, Sham MH, Burns AJ & Chan WY. Analysis of the sacral neural crest cell contribution to the hindgut enteric nervous system in the mouse embryo. Gastroenterology. 2011; 141: 992-1002.e1-6.
20. Barlow AJ, Wallace AS, Thapar N & Burns AJ. Critical numbers of neural crest cells are required in the pathways from the neural tube to the foregut to ensure complete enteric nervous system formation. Development. 2008; 135: 1681-1691.
21. Simpson MJ, Zhang DC, Mariani M, Landman KA & Newgreen DF. Cell proliferation drives neural crest cell invasion of the intestine. Dev Biol. 2007; 302: 553-568.
22. Mckeown SJ, Stamp L, Hao MM & Young HM. Hirschsprung disease: a developmental disorder of the enteric nervous system. Wiley Interdiscip Rev Dev Biol. 2013; 2: 113-129.
23. Gunadi Ivana G, Mursalin DA, Pitaka RT, Zain MW, Puspitarani DA, Afandy D, et al. Functional outcomes of patients with short-segment Hirschsprung disease after transanal endorectal pull-through. BMC Gastroenterology. 2021; 21.
24. Langer JC. Hirschsprung disease. Curr Opin Pediatr. 2013; 25: 368-374.
25. Widyasari A, Pravitasari WA, Dwihantoro A & Gunadi. Functional outcomes in Hirschsprung disease patients after transabdominal Soave and Duhamel procedures. BMC Gastroenterol. 2018; 18: 56.
26. Wang XJ & Camilleri M. Hirschsprung disease: Insights on genes, penetrance, and prenatal diagnosis. Neurogastroenterol Motil. 2019; 31: e13732.
27. Meier-Ruge WA, Bruder E, Holschneider AM, Lochbühler H, Piket G, Posselt HG, et al. Diagnosis and therapy of ultrashort Hirschsprung's disease. Eur J Pediatr Surg. 2004; 14: 392-397.
28. Kenny SE, Tam PKH & Garcia-Barcelo M. Hirschsprung's disease. Seminars in Pediatric Surgery. 2010; 19: 194-200.
29. Moore SW. Total colonic aganglionosis and Hirschsprung's disease: a review. Pediatric Surgery International. 2015; 31: 1-9.

30. Szylberg L & Marszałek A. Diagnosis of Hirschsprung's disease with particular emphasis on histopathology. A systematic review of current literature. Prz Gastroenterol. 2014; 9: 264-269.

31. Langer JC & Levitt MA. Hirschsprung Disease. Current Treatment Options in Pediatrics. 2020; 6: 128-139.

32. Bernstein CN, Kuenzig ME, Coward S, Nugent Z, Nasr A, El-Matary W, et al. Increased Incidence of Inflammatory Bowel Disease After Hirschsprung Disease: A Population-based Cohort Study. The Journal of Pediatrics. 2021.

33. Alnajar H, Murro D, Alsadi A & Jakate S. Spectrum of Clinicopathological Deviations in Long-Segment Hirschsprung Disease Compared With Short-Segment Hirschsprung Disease: A Single-Institution Study. Int J Surg Pathol. 2017; 25: 216-221.

34. Burkardt DD, Graham JM Jr, Short SS & Frykman PK. Advances in Hirschsprung disease genetics and treatment strategies: an update for the primary care pediatrician. Clin Pediatr (Phila). 2014; 53: 71-81.

35. Best KE, Glinianaia SV, Bythell M & Rankin J. Hirschsprung's disease in the North of England: prevalence, associated anomalies, and survival. Birth Defects Res A Clin Mol Teratol. 2002; 94: 477-80.

36. Thakkar H & Curry J. Hirschsprung's disease. Paediatrics and Child Health. 2020; 30: 341-344.

37. De Lorijn F, Boeckxstaens GE & Benninga MA. Symptomatology, pathophysiology, diagnostic work-up, and treatment of Hirschsprung disease in infancy and childhood. Current Gastroenterology Reports. 2007; 9: 245-253.

38. Zaitoun I, Erickson CS, Barlow AJ, Klein TR, Heneghan AF, Pierre JF, et al. Altered neuronal density and neurotransmitter expression in the ganglionated region of Ednrb null mice: implications for Hirschsprung's disease. Neurogastroenterol Motil. 2013; 25: e233-244.

39. Coyle D, O'donnell AM, Gillick J & Puri P. Altered neurotransmitter expression profile in the ganglionic bowel in Hirschsprung's disease. J Pediatr Surg. 2016; 51: 762-769.

40. Bhave S, Arciero E, Baker C, Ho WL, Guyer RA, Hotta R, et al. Pan-enteric neuropathy and dysmotility are present in a mouse model of short-segment Hirschsprung disease and may contribute to post-pullthrough morbidity. J Pediatr Surg. 2021; 56: 250-256.

41. Kapur RP. Practical pathology and genetics of Hirschsprung's disease. Semin Pediatr Surg. 2009; 18: 212-213.

42. Tam PK & Garcia-Barceló M. Genetic basis of Hirschsprung's disease. Pediatr Surg Int. 2009; 25: 543-558.

43. Tilghman JM, Ling AY, Turner TN, Sosa MX, Krumm N, Chatterjee S, et al. Molecular Genetic Anatomy and Risk Profile of Hirschsprung's Disease. New England Journal of Medicine. 2019; 380: 1421-1432.

44. Moore SW. Advances in understanding the association between Down syndrome and Hirschsprung disease (DS–HSCR). Pediatric Surgery International. 2018; 34: 1127-1137.

45. Heuckeroth RO. Hirschsprung disease - Integrating basic science and clinical medicine to improve outcomes. Nature Reviews Gastroenterology and Hepatology. 2018; 15: 152-167.

46. Saleem MD. Biology of human melanocyte development, Piebaldism, and Waardenburg syndrome. Pediatr Dermatol. 2019; 36: 72-84.

47. Zaidi S, Gandhi J, Vatsia S, Smith NL & Khan SA. Congenital central hypoventilation syndrome: An overview of etiopathogenesis, associated pathologies, clinical presentation, and management. Auton Neurosci. 2018; 210: 1-9.

48. Conces MR, Hughes A & Pierson CR. Neuropathology of Mowat-Wilson Syndrome. Pediatr Dev Pathol. 2020; 23: 322-325.

49. Amiel J, Sproat-Emison E, Garcia-Barcelo M, Lantieri F, Burzynski G, Borrego S, et al. Hirschsprung disease, associated syndromes and genetics: a review. Journal of Medical Genetics. 2018; 45: 1-14.

50. So MT, Leon TY, Cheng G, Tang CS, Miao XP, Cornes BK, et al. RET mutational spectrum in Hirschsprung disease: evaluation of 601 Chinese patients. PLoS One. 2011; 6: e28986.

51. Eketjäll S & Ibáñez CF. Functional characterization of mutations in the GDNF gene of patients with Hirschsprung disease. Hum Mol Genet. 2002; 11: 325-329.

52. Gianino S, Grider JR, Cresswell J, Enomoto H & Heuckeroth RO. GDNF availability determines enteric neuron number by controlling precursor proliferation. Development. 2003; 130: 2187-2198.

53. Bondurand N, Natarajan D, Barlow A, Thapar N & Pachnis V. Maintenance of mammalian enteric nervous system progenitors by SOX10 and endothelin 3 signalling. Development. 2006.

54. Lang D, Chen F, Milewski R, Li J, Lu MM & Epstein JA. Pax3 is required for enteric ganglia formation and functions with Sox10 to modulate expression of c-ret. J Clin Invest. 2000; 106: 963-971.

55. Garcia-Barceló M, Sham MH, Lui VC, Chen BL, Ott J & Tam PK. Association study of PHOX2B as a candidate gene for Hirschsprung's disease. Gut. 2003; 52: 563-567.

56. Fu M, Lui VC, Sham MH, Pachnis V & Tam PK. Sonic hedgehog regulates the proliferation, differentiation, and migration of enteric neural crest cells in gut. J Cell Biol. 2004; 166: 673-684.

57. Barlow A, De Graaff E & Pachnis V. Enteric nervous system progenitors are coordinately controlled by the G protein-coupled receptor EDNRB and the receptor tyrosine kinase RET. Neuron. 2003; 40: 905-916.

58. Amiel J & Lyonnet S. Hirschsprung disease, associated syndromes, and genetics: a review. J Med Genet. 2001; 38: 729-739.

59. Feldman M, Friedman L & Brand TL. Hirschsprung's disease: congenital megacolon In: Feldman, M, Friedman, L. & Brandt, L. (eds.) Sleisenger & Fodtran's Gastrointestinal and Liver Disease: Pathophysiology, Diagnosis, Management 7th ed. Philadelphia, PA Saunders. 2002.

60. Hackam DJ, Reblock KK, Redlinger RE & Barksdale EM. Diagnosis and outcome of Hirschsprung's disease: does age really matter? Pediatric Surgery International. 2004; 20: 319-322.

61. Thiagarajah JR, Yildiz H, Carlson T, Thomas AR, Steiger C, Pieretti A, et al. Altered goblet cell differentiation and surface mucus properties in Hirschsprung disease. PLoS One. 2014; 9: e99944.

62. Yildiz HM, Carlson TL, Goldstein AM & Carrier RL. Mucus Barriers to Microparticles and Microbes are Altered in Hirschsprung's Disease. Macromol Biosci. 2015; 15: 712-718.

63. Ward NL, Pieretti A, Dowd SE, Cox SB & Goldstein AM. Intestinal aganglionosis is associated with early and sustained disruption of the colonic microbiome. Neurogastroenterol Motil. 2012; 24: 874-e400.

64. Frykman PK, Cheng Z, Wang X & Dhall D. Enterocolitis causes profound lymphoid depletion in endothelin receptor B- and endothelin 3-null mouse models of Hirschsprung- associated enterocolitis. Eur J Immunol. 2015; 45: 807-817.

65. Pierre JF, Barlow-Anacker AJ, Erickson CS, Heneghan AF, Leverson GE, Dowd SE, et al. Intestinal dysbiosis and bacterial enteroinvasion in a murine model of Hirschsprung's disease. Journal of Pediatric Surgery. 2014; 49: 1242-1251.

66. Jiao C-L, Chen X-Y & Feng J-X. Novel Insights into the Pathogenesis of Hirschsprung's- associated Enterocolitis. Chinese medical journal. 2016; 129: 1491-1497.

67. Pastor AC, Osman F, Teitelbaum DH, Caty MG & Langer JC. Development of a standardized definition for Hirschsprung's-associated enterocolitis: a Delphi analysis. J Pediatr Surg. 2009; 44: 251-256.

68. Frykman PK & Short SS. Hirschsprung-associated enterocolitis: prevention and therapy. Semin Pediatr Surg. 2012; 21: 328-335.

69. Gosain A & Brinkman AS. Hirschsprung's associated enterocolitis. Curr Opin Pediatr. 2015; 27: 364-369.

70. Wester T & Granström AL. Hirschsprung disease-Bowel function beyond

childhood. Seminars in Pediatric Surgery. 2017; 26: 322-327.

71. Gobran TA, Ezzat A, Hassan ME & O'Neill J. Redo transanal endorectal pull-through: a preliminary study. Pediatric Surgery International. 2007; 23: 189-193.

72. Langer JC, Rollins MD, Levitt M, Gosain A, Torre L, Kapur RP, et al. Guidelines for the management of postoperative obstructive symptoms in children with Hirschsprung disease. Pediatr Surg Int. 2017; 33: 523-526.

73. Kapur RP, Smith C & Ambartsumyan L. Postoperative Pullthrough Obstruction in Hirschsprung Disease: Etiologies and Diagnosis. Pediatr Dev Pathol. 2020; 23: 40-59.

74. Smith C, Ambartsumyan L & Kapur RP. Surgery, Surgical Pathology, and Postoperative Management of Patients With Hirschsprung Disease. Pediatr Dev Pathol. 2020; 23: 23-39.

75. Ralls MW, Coran AG & Teitelbaum DH. Redo pullthrough for Hirschsprung disease. Pediatric Surgery International. 2017; 33: 455-460.

76. Hyman PE. Defecation Disorders after Surgery for Hirschsprung's Disease. Journal of Pediatric Gastroenterology and Nutrition. 2005: 41.

77. Zhang SC, Bai YZ, Wang W & Wang WL. Stooling patterns and colonic motility after transanal one-stage pull-through operation for Hirschsprung's disease in children. J Pediatr Surg. 2005; 40: 1766-1772.

78. Chumpitazi BP & Nurko S. Defecation disorders in children after surgery for Hirschsprung disease. J Pediatr Gastroenterol Nutr. 2011; 53: 75-79.

79. Levitt MA, Dickie B & Peña A. Evaluation and treatment of the patient with Hirschsprung disease who is not doing well after a pull-through procedure. Seminars in Pediatric Surgery. 2010; 19: 146-153.

80. Calkins CM. Hirschsprung Disease beyond Infancy. Clin Colon Rectal Surg. 2018; 31: 51-60.

81. Demehri FR, Halaweish IF, Coran AG & Teitelbaum DH. Hirschsprung-associated enterocolitis: pathogenesis, treatment and prevention. Pediatr Surg Int. 2013; 29: 873-881.

82. Pini Prato A, Rossi V, Avanzini S, Mattioli G, Disma N & Jasonni V. Hirschsprung's disease: what about mortality? Pediatr Surg Int. 2011; 27: 473-478.

83. Sood S, Lim R, Collins L, Trajanovska M, Hutson JM, Teague WJ, et al. The long-term quality of life outcomes in adolescents with Hirschsprung disease. J Pediatr Surg. 2018; 53: 2430-2434.