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Selected topics in pediatric pathology

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Perinatal gastrointestinal pathology

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While much perinatal gastrointestinal pathology is concerned with confirmation of diagnoses, such as necrotizing enterocolitis, meconium ileus and intestinal atresia in surgically resected specimens, there are two clinical situations in the neonatal period in which the primary diagnosis rests with the histopathologist. The first is constipation (which usually presents as delayed passage of meconium) in which a rectal suction biopsy is required to confirm or exclude the diagnosis of Hirschsprung's disease. The second is congenital diarrhea, in which a small bowel biopsy is performed to confirm or rule out a number of specific disease entities, chief among which are microvillus inclusion disease, tufting enteropathy and autoimmune enteropathy.

Hirschsprung's disease

Hirschsprung's disease (HD) is characterized by the absence of ganglion cells in the distal portion of the intestine and by their presence in the proximal portion. It can be categorized according to the length of the aganglionic segment. The incidence is 1 in 5,000 live births. Approximately 80% of patients are male but this ratio decreases as the length of the aganglionic segment increases, such that the sex incidence is equal in total colonic aganglionosis. The genetic basis is complex with an increased incidence of Down's syndrome in HD. Mutations on chromosome 10q11.2 involving the receptor tyrosine kinase gene RET are present in approximately 20% of patients.

Rectal mucosal biopsy obtained by suction biopsy machine is the established method for diagnosing HD. Two biopsies are preferred, at 2 and 5 cm above the pectinate line. The biopsies should be kept moist on saline-soaked filter paper and transported to the laboratory within 15 min. One biopsy is formalin-fixed and paraffin-embedded. Sixty serial sections in a plane horizontal to the mucosal surface are stained with hematoxylin and eosin, and examined for the presence of ganglion cells. In HO there is an absence of ganglion cells and an increase in large nerve trunks.

The second biopsy is stained for acetylcholinesterase (AChE) activity. There is a well-attested body of literature, both in journals and standard textbooks, supporting the value of AChE in confirming and excluding the diagnosis of HD. The biopsy is orientated on a block of animal liver and frozen for cryostat sections in a plane perpendicular to the mucosal surface. Sections are taken at 5 mm for H&E and at 10 mm for AChE staining. In HD, there is a marked increase in coarse AChE-positive fibers in the muscularis mucosae and lamina propria. Numerous large nerve trunks are present in the submucosa.

The following *provisos* apply: i) An adequate biopsy must include enough submucosa to provide a reasonable chance of finding ganglion cells. ii) The biopsy must be taken from above the pectinate line. It is important that the pathologist be able to recognize a low biopsy, which is normally anatomically hypoganglionic. iii) In the neonate, ganglion cells are small and immature and look like neuroblasts. The AChE pattern is less dramatic than in the older child with only the muscularis mucosae showing increased nerve fibers. iv) In total colonic aganglionosis, there may be no increase in prominent submucosal nerves and the AChE pattern may be normal. v) With regard to interpretation of AChE staining, we routinely stain all rectal suction biopsies for AChE, even those that are clearly ganglionic. The reason for this is to keep the eye in for the range of normality as well as for the abnormal. Uniformity of staining from case to case in setting a "departmental standard" of normal and abnormal pattems is of importance and we have found that the preparation of batches of incubation media and storing in preprepared aliquots at —20 °C is useful.

At Great Ormond Street, some 250 rectal suction biopsies are currently performed annually, of which approximately 30 will prove to be HD. Two-to-three cases per year produce an "equivocal" pattern. These would all be aganglionic samples but showing only a slight abnormality in the pattern of AChE not amounting to that seen in classical HD. In these circumstances, we suggest either rebiopsy or defunctioning colostomy with intraoperative biopsy, as clinically indicated.

Microvillus inclusion disease

Microvillus inclusion disease (MID) is a rare autosomal recessive enteropathy in which infants present in the first few days of life with severe watery diarrhea. Jejunal biopsy shows severe partial villous atrophy without crypt hyperplasia and without inflammation of the lamina propria. Surface enterocytes lack a well-defined brush border. Periodic acid Schiff (PAS) staining and staining for alkaline phosphatase show distinctive inclusions in the apical cytoplasm in surface and midcrypt enterocytes. Electron microscopy (EM) of surface enterocytes shows intracellular microvillus inclusions with a lack of, or shortened, irregular microvilli on the surface. Jejunal biopsy is optimal for the histological diagnosis but peroral biopsy is only obtainable in infants of 6 weeks or older. Endoscopic small bowel biopsy can be procured in younger infants. A rectal biopsy that can be performed in the perinatal period does not exhibit PAS and alkaline phosphatase positivity but scattered intracellular inclusions may be seen by EM in occasional enterocytes in MID. An absence of inclusions in a rectal biopsy, however, does not exclude the diagnosis. An acquired form of MID is recognized and we have seen the disease in a neonate in whom sequential jejunal biopsies over a 3-week period revealed a progression from an almost normal brush border to fully developed MID. The prognosis for patients with MID is invariably fatal. Long-term parenteral nutrition used to be the only treatment but bowel transplantation is now considered the treatment of choice.

Tufting enteropathy

First described in 1994, tufting enteropathy is a clinicopathological entity in which infants present with watery diarrhea in the first month of life. The pattern of inheritance appears to be autosomal recessive. Jejunal biopsy shows partial or subtotal villous atrophy with normal or hyperplastic crypts. The lamina propria is not inflamed and intraepithelial lymphocytes are not increased. The most striking finding is of epithelial cell tufts, that is, closely packed enterocytes with rounded apical plasma membranes. While this appearance may be seen focally in other enteropathies, in tufting enteropathy between 80% and 90% of the surface exhibits the change. Treatment at present is with total parenteral nutrition.

Patients with autoimmune enteropathy present with diarrhea in the first year of life and may present in the neonatal period. A family history of autoimmune disease is often present. In some patients, the disorder may occur in the context of a primary immunodeficiency syndrome. Jejunal biopsies show mild, partial to subtotal villous atrophy with crypt hyperplasia. Surface enterocytes show numerous intraepithelial lymphocytes and numerous vacuolated and individually necrotic surface enterocytes as a marker of cytopathic damage. The lamina propria shows dense lymphocytic and plasma cell infiltration (although in agammaglobulinemia and hypogammaglobulinemia plasma cells are absent) and there may be crypt abscesses. Antigut antiepithelial or antigoblet cell antibodies are detected in the blood. Treatment is directed to the underlying immune defect in immunodeficient patients and immunosuppressive therapy in those who do not have a demonstrable immunodeficiency.

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Gastric pathology in pediatric Crohn's disease

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Systematic endoscopic evaluation of the upper gastrointestinal (GI) tract of patients with Crohn's disease (CD) has revealed frequent endoscopical and histological abnormalities (1, 2). Recent studies have shown that there is gastric involvement in over three-fourths of patients with CD. However, in these studies, only a minority of these gastric lesions were specific cases of granulomatous gastritis. The most striking lesion was a focal acute inflammation of the stomach (3, 4). Other studies have shown that the majority of the gastritis that occurs in CD is not associated with *Helicobacter pylori* infection (5). The greatest challenge for understanding upper gastrointestinal involvement in CD is to define the value of this focal inflammation more precisely.

Focal inflammation may be defined as the presence of inflammatory cells surrounding at least a foveolar pit or a small group of foveolae or glands. The inflammatory cells are mononuclear cells (histiocytes and lymphocytes) and polymorphonuclear neutrophils. The neutrophils infiltrate the epithelium that shows alterations, such as discontinuities or cuboidal-shaped cells. The inflammation is often deep within the mucosa and is rarely associated with granulomas. This histological definition of focal inflammation rules out activity when the majority of crypts or glands are infiltrated by inflammatory cells or when adjacent normal mucosa cannot be found in a given biopsy specimen. Focal inflammation is characteristic of *H.* pyloni-negative CD and is uncommon in non-CD cases. The value of focal enhanced gastritis is so great that some authors claim that this type of gastritis should encourage further investigations of patients not yet known to have CD, or that it could help to classify indeterminate colitis.

The concept that immunological changes play a key role in the pathogenesis of CD is now established (6). The cytokine production profiles in CD strongly suggest the prevalence of a Th1 response (7). In contrast, murine models of ulcerative colitis, such as the T-cell receptor alpha mutant, indicate the critical role of the Th2 cytokine in the pathogenesis of this colitis (8). The cytokine interleukin-12 (IL-12) is composed of two subunits, p40 and p35, encoded by two separate genes (9). IL-12 is produced mainly by monocytes and macrophages and plays a pivotal role in the differentiation of Th1 T-cells, as it induces naive T-cells to produce interferon-γ (10,11). Murine models of experimental granulomatous colitis mimicking CD indicate that IL-12 plays a major role in the disorder (12). Recent studies have shown greater amounts of IL-12 in the intestinal tissues of CD patients than in the normal and inflamed tissues of controls (13).

The important part played by IL-12 in CD prompted us to study its expression in the gastritis of pediatric patients with confirmed distal CD in an attempt to identify a link between cytokine production and nongranulomatous and granulomatous gastritis occurring in CD. We demonstrated *in situ* enhanced IL-12 production in the gastric mucosa of pediatric CD patients. The patterns of IL-12-positive cells in the small bowel and gastric mucosa were the same, with small to large foci of positive cells in the lamina propria. However, no IL-12-positive cells were found in the gastric mucosa of the patients with ileal CD and normal gastric mucosa, or in the controls and only few scattered positive cells were found in 50% of the gastric mucosa specimens from patients with *H.* pylori-positive chronic gastritis. Also, the amounts of IL-12 expression in the gastric biopsy specimens exhibiting granulomatous gastritis and non granulomatous gastritis were similarly enhanced. A recent study has shown that gastritis-associated CD is characterized by a focal periglandular accumulation of CD68+ histiocytes and CD3+ lymphocytes with a high content of TiA1+ cytotoxic T lymphocytes (3). As IL-12 is mainly produced by macrophages and enhances T-cell and natural killer cell cytotoxicity, with increased productions of granzyme B and perforin messenger RNA, IL-12 may play a major role in the pattern and accumulation of immune cells in gastritis-associated CD. The demonstration of focally enhanced IL-12 expression in both nongranulomatous gastritis and granulomatous gastritis, as well as in ileal specimens from CD patients, highlights the link betweennall these lesions in CD.

Finally, IL-12 expression is elevated at the time of CD diagnosis and remains increased for months or years during the course of CD, suggesting that IL-12 is important for both initiating and maintaining inflammation in this disease. In conclusion, IL-12 expression is focally enhanced in the gastric mucosa of pediatric patients

with CD, with or without granulomatous lesions, suggesting that both IL-12 and the lesions are indeed linked to CD (14,15). These data reinforce the idea that focal inflammation of the gastrointestinal tract is a hallmark of CD.

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Childhood gastrointestinal infections and infestations as seen in the tropics

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The spectrum of infection and infestation that occurs in the tropics is generally not very different from that which is encountered in subtropical and temperate regions but the frequency of particular infections and infestations, as seen in the tropics, varies considerably and also varies from one tropical region to another.

Diarrheal disease

Acute childhood diarrhea is common and is frequently lethal among poor people throughout the tropics. There are many causative agents and the frequency with which they occur varies from area to area and with the season of the year. Viral infections due to Norwalk virus, rotavirus, enterovirus and enteric adenovirus are highly prevalent. Identified bacterial pathogens account for about 30-45% of acute childhood diarrheal disease (1). Particular strains of *Escherichia coli* have an important role and their identification and separation from commensal strains is often a major problem, especially in tropical countries with limited resources. Enterogenic *E. co/i* is a significant cause of diarrhea in children younger than 3 years and also causes traveler's diarrhea in older individuals. Infection generally occurs from contaminated food and water. Enteropathogenic *E.* co/i mainly affects children less than 3 months of age in developing countries. Enteroinvasive *E. co/i* rarely causes epidemics of diarrhea in developing countries and is acquired from contaminated food. Enterohemorrhagic *E. coli* occurs worldwide and causes a watery diarrhea that becomes bloody and is associated with neurologic manifestations. Some strains are associated with the hemolytic uremic syndrome. Enteroaggregative *E.* co/i causes acute persistent diarrhea in infants in developing countries but infection is generally self-limiting. The *Campylobacter* group of organisms is now considered to be the dominant identifiable bacterial pathogen causing severe gastroenteritis in semi-urban shack dwelling children (2) where informal butcheries and home slaughtering of chickens is common practice. During recent years there has been an accumulation of evidence to implicate *C. jejuni* in the occurrence of the acute motor axonal neuropathic form of Guillain-Barré syndrome (3). *Salmonella* and *Shigella* infections in the tropics are no different from those that occur in temperate areas except that they are more prevalent and there is a wide spectrum of virulence. In recent years there have been increased reports of hemolytic uremic syndrome complicating *Shigella* infection. Cholera is becoming more widespread throughout the whole of the tropics and manifests as an acute illness with a rapid loss of fluids and electrolytes from the intestines that leads to hypovolemic shock (4). *Yersinia enterocolitica* gastroenteritis is uncommon in the tropics.

Intestinal tuberculosis

In rural Africa milk is generally not pasteurized and children are increasingly becoming infected with bovine mycobacteria. Such infection usually results in a primary complex occurring in the ileum and mesenteric lymph nodes. However, most cases of intestinal tuberculosis occur as a secondary phenomenon when infected sputum from primary pulmonary tuberculosis from human strains is swallowed. The condition manifests as circumferential ulcers in the lower ileum, caseous mesenteric adenitis and peritonitis. Fibrosis many cause intestinal obstruction and in older children. Rectal infection may cause an schiorectal abscess or fistula.

Tropical sprue

This malabsorption syndrome responds to prolonged antibiotic therapy. It mainly occurs in the Middle East, India and the Caribbean. Patients have watery diarrhea, abdominal discomfort or pain with distention, anorexia, weight loss, glossitis, stomatitis, alteration of skin pigmentation and edema. Persistence of symptoms leads to megaloblastic anemia. Microscopy of intestinal mucosa shows atrophy with an inflammatory infiltrate that includes many eosinophils (5).

Protozoal infections

Giardiasis due to *Giardia duodenalis* is a worldwide disease causing upper abdominal pain and diarrhea or steatorrhea in children. The organisms occur in abundance in the duodenum, common bile duct and upper jejunum. The intestines undergo villous atrophy with excessive loss of surface enterocytes and an accumulation of interepithelial lymphocytes (6). In malnourished or hypogammaglobulinemic children there is reduced production of secretory IgA and nodular lymphoid hyperplasia of intestinal mucosa may occur. The biological behavior of the disease is not significantly influenced by concurrent HIV infection (7). Amebiasis occurs worldwide but is prevalent in the tropics and accounts for many deaths. The controversy as to whether the large and small ameba found in the gut and associated with amebiasis represent different species or distinct pathogenic and commensal forms of the same species *(Entamoeba histolytica* and *E. hartmanni* or *E. dispar)* continues (7) but the balance of evidence points to a single species that changes when enzymes from ingested bacteria and/or host erythrocytes are acquired (8). *E. histolytica* occurs in the lower intestine and colon and release quadrinucleate cysts into the feces. It is estimated that in about 10% of cases with luminal amebiasis the trophozoites become invasive, phagocytose red blood cells and penetrate the colonic wall to cause flask-shaped ulcers and spread through the blood stream to the liver and beyond. In invasive intestinal amebiasis the ameba may also invade retrogradely along the walls of arteries and progressively along the tributaries of the portal vein to cause edema of vessel walls and thrombosis with intestinal infarction and perforation (9,10). In addition, amebiasis may manifest as polypoid structures with many organisms in the cecum of infected children. Rectal amebiasis can involve the anus and perianal skin to produce pseudoepitheliomatous hyperplasia and may rarely become a sexually transmitted disease (11). Amebic liver abscesses that occur as a consequence of hematogenous spread through the portal vein has been seen during the first mouth of life at the Red Cross Children's Hospital. In children there are often multiple small abscesses which rarely perforate. Balantidiasis due to the large protozoan *Balantidium coil* occasionally causes intermittent diarrhea in rural areas after contact with pigs, rats or baboons.

Colonic ulcers are similar to those of amebiasis but may be complicated by hemorhage, perforation or hematogenous spread to liver and genitourinary system. Isosporiasis due to *Isospora bern* is an intestinal opportunistic infection mainly occurring in children in developing countries who are either malnourished or have HIV infection. In common with *Sarcocystis, Cryptosporidium* and microsporidia they cause villous atrophy with an inflammatory cell infiltration into the lamina propria that includes eosinophils. Clinically there is diarrhea, malabsorption and steatorrhea. For diagnosis the parasites may be seen in epithelial cells or oocysts may be identified in Giemsa stained stools. *Cryptosporidium* is a prevalent and underdiagnosed cause of diarrhea in malnourished and immunosuppressed children. The very small organisms occur in intraepithelial vacuoles but are also recognized in stools stained with Ziehl-Nielsen or auramine (12).

Helminthic infestations

Nematode (round worm) infestations are prevalent in tropical and subtropical areas among poor people living in conditions of substandard housing, water and sewage provision. Cestodes (flat worms) and Trematodes (flukes) occur worldwide but are more prevalent among poor people in tropical or subtropical climates. The hookworms, *Ancylostoma duodenale* and *Necator americanus* are indigenous to the moist tropics and particularly occur amongst subsistence farmers. Adult worms live in the lumen of the duodenum below the level of the ampulla of Vater and upper jejunum. They attach to a mucosal villus and suck blood and intestinal fluid. Depletion of iron and protein causes symptoms (13).

Hookworms are a recognized cause of protein losing enteropathy amongst anemic children in the tropics. *Strongyloides stercolaris* has the capacity to undergo successive generations of reproduction within a host so that symptoms may occur years after initial infection when the host could have left an endemic area. Fatal massive autoinfection particularly occurs in immunocompromised hosts (14). Urticarial wheals occur where the infective larvae enter the body through the skin and in heavy infections larvae may cause asthma, watery diarrhea with dehydration and renal failure. Stool larvae may seem to be scanty because of the dilution effect of diarrhea. Secondary Gram-negative septicemia and disseminated intravascular coagulopathy are the most frequent complications and causes of death.

Ascariasis is caused by *Ascaris lumbricoides,* the common large round worm. It is extremely prevalent in moist areas of the tropics and subtropics where up to 100% of children may carry the worm. Heavy infestation is considered to cause nutritional problems, fever, malaise, urticaria, nausea, vomiting, intestinal colic and diarrhea. Complications that occur include: volvulus due to a worm bolus (15); obstructive jaundice and pancreatitis from duct obstruction by one or more worms; cholangitis, liver abscess and hepatic granulomas from embryonating ova may occur subsequently (15,16); irritation and obstructive symptoms occur from worms getting into respiratory passages and eustachian tubes; pneumonitis and bronchospasm attributed to larval migration through the lungs are extremely rare amongst indigenous inhabitants of endemic areas; and mesenteric adenitis has been attributed to infestation and a fatal worm embolus to the lungs has been reported as well as involvement of the kidney and renal pelvis (17,18).

Intestinal capillariasis due to *Capillaria philippinensis* occurs in the Philippines and Thailand with infestation occurring from eating fresh water fish followed by autoinfection. The worms cause abdominal pain, diarrhea, anorexia, nausea and vomiting. Prolonged diarrhea leads to cachexia, muscle wasting and death. Whipworm infestation due to *Trichuris trichiura* is particularly prevalent in Southeast Asia and most infections are light and asymptomatic. In heavy infestation there is colitis with blood and mucus in stools, which can lead to anemia and edema. A tendency to rectal prolapse has been reported and in rare instances acute appendicitis has been attributed to the worm.

Intestinal angiostrongyliasis due to *Angiostrongylus costaricensis* occurs in Central America and *A. cantonensis* in the Far East when man is accidentally infected by ingesting vegetable leaves smeared with the mucus of slugs containing infective larvae. Infected children develop a high pyrexia, anorexia, vomiting, right lower abdominal pain with a blood eosinophilia. The worms lodge in the ileocecal legion producing colonic and intestinal edema with granulomas and lymph adenopathy. The liver, omentum, testes and arteries may become involved in an inflammatory process with prominent eosinophilia around ova or larvae (19).

Schistosomiasis is particularly prevalent in Africa and the Far East. *Schistosoma haematobium* and *S. japonica* mainly affect the genitourinary system while *S. mansoni* tends to involve bowel as well as other sites such as the central nervous system and geni-

tourinary system. The cercariae of S. *mansoni,* which emerge from snails, penetrate the skin to cause an itchy eruption in previously exposed individuals. About 8 weeks after infection Katayama fever with chills, pyrexia, sweating, headache, cough, hepatosplenomegaly and lymphadenopathy occurs. The worms may cause pseudotuberculous granulomas to develop in the liver but ova are mainly deposited in the colon and rectum. Complications from S. *mansoni* infection include: hepatic fibrosis leading to portal hypertension, and portal, mesenteric and vertebral vascular inflammation and fibrosis with subsequent involvement of the pulmonary vascular bed leading to cor pulmonale or neurological deficit and membrane-proliferative glomerulonephritis from circulating immune complexes (19).

Cestode infestations are less common in children than in young adults. The commonly occurring tapeworms are Taenia *soilum* (pork tapeworm) and the Echinococcus species. Cysticercosis due to *T sollum* affects diverse tissues but especially the brain, heart and muscles when autointection occurs from ova being passed in the feces. In *Echinococcus* infestation (hydatid disease) the embryos from ingested eggs very quickly pass through the gastrointestinal tract to the portal circulation to be filtered out in the liver, lungs and other sites where classical hydatid cysts containing larvae appear.

Infestation by the dwarf tapeworm *Hymenolepis nana* is commonly seen by microbiologists examining wet preparations of stool but the worms are rarely recognized by anatomical pathologists because of their small size. Heavy infestation may cause diarrhea and abdominal pain and constitutional symptoms associated with moderate eosinophilia. The cestode requires a single host but also infects mice, fleas and beetles (19).

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Examination of the placenta in late intrauterine death: What can we tell about cause and recurrence risk?

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During the past decades, there has been considerable advancement in the care of a fetus and its mother and, later on, of the newborn. With the great reduction of overall perinatal mortality in highly developed countries we have come to a stage where 50% of all perinatal losses are constituted by intrauterine death affecting a viable fetus older than 26 weeks of gestation. Severe fetal malformations are already excluded from this number, as are the few deaths that are caused by obvious bacterial or viral infections. Thus, the majority of fetal demise in the third trimenon is due to some kind of placental insufficiency that occurs in two quite different clinical settings and which can be described as "chronic" or "acute" placental insufficiency.

"Chronic" placental insufficiency

Chronic insufficiency comprises all conditions in which reduced placental function is reflected by fetal growth retardation. Obviously there are cases where the growth of a child for genetic reasons is below the 5th percentile and the placenta is small simply because the corresponding child is small. Growth retardation proper is characterized by a declining curve of fetal growth caused by *in* utero starving: it is thus necessarily "chronic". Starvation itself can be compatible with fetal survival for periods up to 6 and even 8 weeks. In general, compromise of the maternal circulation is better tolerated, especially when it occurs in "succession" as opposed to the compromise of fetal circulation, which is also much more ominous in regard to the underlying disease condition. *Post mortem* findings in the fetus will reveal severe reduction in the weight of internal organs, especially the liver, thymus and spleen with some growth parameters remaining around the 50th percentile for a long time, especially brain weight and foot length. The placenta of a growth-retarded child is usually characterized by areas that have ceased to function. These usually constitute gross morphological findings of a focal nature, the most typical being infarcts of different age, size and location. It is important to estimate the volume of placental tissue lost and to denote whether all infarcts have the same appearance (thus having occurred eventually all at the same time) or whether they exhibit different color and Consistency (giving the impression that they have occurred in succession). The most typical accompanying microscopic finding is an increased vascularization of the chorionic villi which have remained viable and which thus try to compensate for the losses. At the time when chronic placental insufficiency leads to fetal malnutrition, many of the mothers are also unhealthy; they have symptoms of preeclampsia, are

found to smoke or to be addicted to other drugs or are found to suffer from acquired or hereditary thrombophilic conditions.

"Acute" placental insufficiency

Acute insufficiency is the term used when fetal death is the first noted symptom of a placental disturbance. Thus, neither the intrauterine growth curve nor the *post mortem* examination of the fetus indicate *in utero* starvation. Fetuses that have succumbed to acute placental insufficiency are mostly around the 50th percentile when their age-related weights and measurements are taken. Many of them will show petechial bleedings at the surface of the lungs, heart and thymus as well as congestion of the meninges. Although the pathophysiology of these findings is not quite understood, they are interpreted as signs and symptoms of hypoxia. The corresponding placenta is mostly perfect in size and shape and does not exhibit focal changes. However, the color of the cut surface is paler (resembling in this regard a placenta of 20 weeks or less) and does not exhibit the dark red appearance that is so characteristic of a placenta during the last 4-6 weeks of pregnancy. Microscopic examination will reveal the cause of this reduction in color to be a largely reduced vascularization of the chorionic villi. Mothers are usually completely healthy with the exception of a few who suffer from diabetes mellitus type I.

Details of "acute insufficiency" morphology and pathophysiology

A fetus exhibiting signs of hypoxia dies from suffocation, which is almost always an "acute" condition. However, initiating events leading to reduced vascularity of chorionic villi must date back quite some time. The morphological hallmark of reduced vascularity is found in the terminal villi which during the last 6 weeks of pregnancy are characterized by an increasing number of dilated capillaries seeking close contact with the maternal blood. This sinusoidal transformation leads to thin cytoplasmic membranes, which comprise the only remaining barrier between fetal and maternal blood. It has been postulated, but not proven, that these membranes are the site of oxygen exchange. Assuming that this is the case, placentae with underdeveloped "syncytio-capillary membranes" have not fully developed their respiratory capacity. Nutrition of the fetus is not disturbed as the transport of many nutrients requires active transport by carriers and does not occur at the syncytio-capillary membranes. The disturbance of vascular development presents with two different morphological patterns. One is a concomitant increase of villous stroma and enlarged placental weight, often considered the typical finding in cases of maternal diabetes mellitus, The other features a pure lack of terminal villi. The examination of placentae from normal pregnancies reveals on average three well-developed sinusoids within terminal villi that have formed syncytiocapillary membranes covering about 35% of the circumference of a terminal villus. In contrast, terminal villi of fetuses that have succumbed to *in utero* hypoxia, on average present with less than one transformed capillary and the syncytiocapillary membranes covers at the most 10% of the villous circumference. These differences are highly significant, however, when placentae from children who had shown severe alteration of their cardiotocogram before and during birth were examined again: only one transformed capillary with membrane was found on average at the terminal villi. This indicates that the reduction in vascular maturation is a prerequisite of hypoxia but that it does not necessarily lead to fetal demise. Currently, most fetuses survive this developmental abnormality of the placenta because they are rescued by birth.

Epidemiology and cause of vascular maturation defect

It has been known for a long time that maternal diabetes mellitus can lead to fetal and placental overgrowth endangered by sudden intrauterine death and increased fetal mortality. Invariably, in nearly all cases the placentae exhibit strongly increased diameters of stroma-rich terminal villi that do not exhibit sinusoids and membranes. We retrospectively studied the obstetric history of 54 women who had experienced an *in utero* death. In 32, placental insufficiency had been "acute" due to maturation defect of the chorionic villi. Only three of these mothers were diabetic during pregnancy (<10%). Although the event dated back to between 5-20 years, none of the other 29 women with placental morphology "typical of diabetes mellitus" developed diabetes later on. Furthermore, it transpired that women with this type of placental abnormality did not experience another in *utero* death, in contrast to mothers who lost their child due to a compromise of maternal or fetal circulation (Table I).

Perspectives

Chronic placental insufficiency, sometimes termed "nutritional insufficiency", can be detected by careful monitoring of the fetal growth curve, offering the chance to rescue a baby from the adverse uterine environment by preterm delivery. In many cases, mothers have underlying disease and there is a risk of recurrence in the eventual case of another pregnancy. Acute insufficiency, sometimes termed "respiratory insufficiency", is very rarely detected when CTG examination is performed during a hypoxic period and which is sometimes provoked by so-called stress tests. With the exception of maternal diabetes mellitus there seems to be no predisposing maternal disease and the condition is thus termed "idiopathic". An increased risk of recurrence regarding *in utero* death has not been found. However, the morphological finding of striking immaturity of chorionic villi can occur in consecutive pregnancies. Overall, it seems that most children do not succumb to this developmental abnormality as they are rescued by birth. Nevertheless, the lack of a method to detect the latent stage of a defect that can manifest acutely with fetal hypoxia and death is disappointing.

Table 1. Obstetric history of women experiencing intrauterine death of a fetus grouped by the pathology-pattern circulatory compromise ("chronic") *versus* maturation defect ("acute").

	Number of women included	Mothers with children	Liveborn after	Fetal death in a previous pregnancy	Women remaining without children	Average number of live children
Chronic insufficiency	22/54	5	16	3 (13.6%)	3 (13.6%)	1.8
Acute insufficiency	32/54	16	22	0	4(18.2%)	1.9

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