Post-splenectomy and hyposplenic states

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The spleen is crucial in regulating immune homoeostasis through its ability to link innate and adaptive immunity and in protecting against infections. The impairment of splenic function is defined as hyposplenism, an acquired disorder caused by several haematological and immunological diseases. The term asplenia refers to the absence of the spleen, a condition that is rarely congenital and mostly post-surgical. Although hyposplenism and asplenia might predispose individuals to thromboembolic events, in this Review we focus on infectious complications, which are the most widely recognised consequences of these states. Because of the high mortality, the fulminant course, and the refractoriness to common treatment of overwhelming infections caused by encapsulated bacteria, prevention through vaccination and antibiotic prophylaxis is the basis of the management of patients who have had splenectomy or have hyposplenism. In this Review, we critically assess clinical and diagnostic aspects of splenic dysfunction and highlight new perspectives in the prevention of overwhelming post-splenectomy infections.

Introduction

The initial experimental evidence of the protective role of the spleen against infections was provided in the early 1900s by Morris and Bullock,[1] who indicated that splenectomised rats had a significantly higher post-surgical mortality than did rats who had sham operations, and this high mortality was attributed to sepsis by the bacillus that causes rat plague. Many years afterwards, the crucial function of the spleen in immune defence emerged as a result of two brief studies. King and Schumacker[2] reported a series of cases of overwhelming post-splenectomy infections (OPSI) caused by encapsulated bacteria in a cohort of children who had had splenectomy. Dameshek[3] coined the term hyposplenism to describe a patient with coeliac disease in whom Howell-Jolly bodies were detected on peripheral blood smear and an atrophic spleen was confirmed at post-mortem examination. Hyposplenism is now regarded as an acquired disorder, potentially associated with several diseases and sometimes accompanied by a reduction in spleen size. Asplenia refers to the absence of the spleen, a disorder that is rarely congenital and is more frequently a result of surgery.

Although basic research has provided detailed information on the role of the spleen in the immune response,[4,5] and data from many studies have confirmed the association of asplenia or impaired splenic function with increased morbidity and mortality from infectious complications,[6] this information has not been sufficiently translated into appropriate clinical practice. Most physicians are unaware of the diseases for which a systematic search for splenic dysfunction should be done, what the clinical predictors of this dysfunction and the means for assessing it are, what the real risks of sepsis are, and what would be the best ways to prevent sepsis. Treatment of post-splenectomy thrombotic complications has been recently reviewed;[7] in this Review, we clarify the risk factors for spleen dysfunction, clinical signs, diagnostic techniques, and prophylaxis options against infection.

Search strategy and selection criteria

We searched Medline by using the medical subject heading terms "asplenia", "hyposplenism", "pneumococcal vaccination", "splenectomy", "splenic atrophy", and "splenosis" for articles published between January, 1998, and June, 2010, but we did not exclude commonly referenced and highly regarded older publications. We also searched the reference lists of review articles on splenectomy and hyposplenism for additional papers we judged to be relevant to this Review.

Immunological function of the spleen

The spleen consists of three functional inter-related compartments—the red pulp, white pulp, and marginal zone (figure 1).[4] The red pulp is a sponge-like structure filled with blood flowing through sinuses and cords. The white pulp is distributed along the central arteriole branching from the splenic artery. T cells form an envelope (the periarteriolar lymphoid sheath) around the central arteriole, and also surround the B-cell follicle in a thin layer. This thin layer is formed by an outer dark zone—the mantle zone, containing dominantly proliferating small B lymphocytes—and a light central zone—the germinal centre, the area of B-cell selection. The marginal zone, containing memory B cells, is the extreme periphery of the white pulp in direct contact with the perifollicular area, where macrophages and fibroblasts positive for mucosal addressin cell adhesion molecule 1 are located.

The spleen functions as a phagocytic filter, removing senescent and damaged cells (culling), solid particles from the cytoplasm of erythrocytes (pitting), and blood-borne microorganisms, and also produces antibodies. When blood enters the splenic cords of the red pulp and passes through the fenestrated epithelium going into the venous sinus, the flow slows down, which helps the removal of defective erythrocytes and bacteria by splenic macrophages. Some bacteria are recognised directly by macrophages, but many first need to be opsonised, during which the bacterial surface is coated by complement or other spleen-derived opsonising molecules (properdin, tuftsin), which in turn interact with receptors on phagocytes.[8] Opsonised bacteria are removed efficiently by macrophages in the spleen and liver. However, poorly opsonised bacteria, such as encapsulated bacteria—in

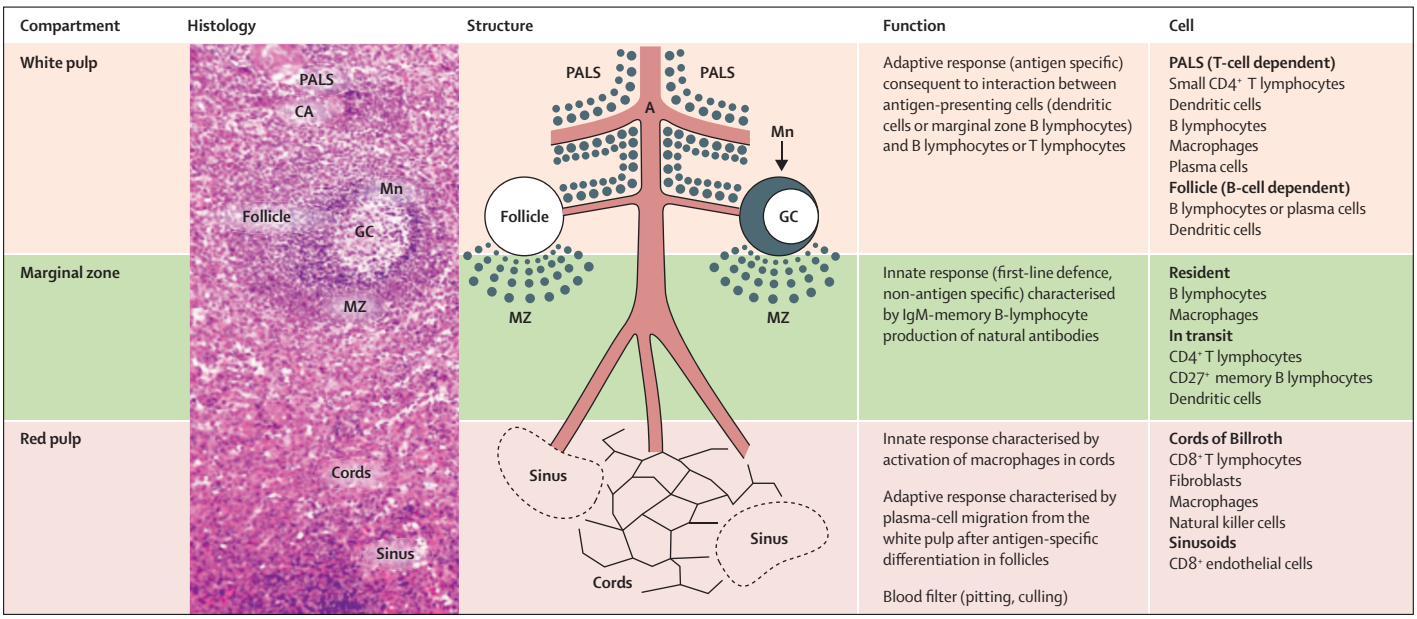

Figure 1: **Structure, function, and cell populations of the three functional compartments of the spleen**
The histology panel shows a haematoxylin and eosin-stained section of normal spleen. CA=central arteriola. GC=germinal centre. Mn=mantle zone. MZ=marginal zone. PALS=periarteriolar lymphoid sheath. The structure panel provides a schematic representation of the spleen. The function panel lists a concise description of the roles of each compartment of the spleen. The last panel provides a description of the different cells within each compartment.

particular *Streptococcus pneumoniae* whose polysaccharide capsule impedes binding of complement or prevents complement assembled on the capsule from interacting with macrophage receptors—are only cleared by the spleen. For removal of these bacteria in the course of initial infection, natural antibodies are needed, which are pentameric IgM able to facilitate phagocytosis either directly or via complement deposition on the capsule and are produced by IgM memory B cells—a unique B-cell population in the marginal zone of the spleen. The key role of the spleen in initiating an immune response against encapsulated bacteria is indicated by the large reduction of IgM memory B cells after removal of the spleen.[5,9]

Nearly half of the total B cells in the blood express the memory marker CD27 and carry somatic mutations, and are therefore thought to be memory B cells. Two populations of memory B cells have been identified in human beings—switched memory B cells and IgM memory B cells. Switched memory B cells, which are the final product of germinal centre reactions, produce high-affinity antibodies and have a protective function against reinfection.[10] IgM memory B cells, which need the spleen for their survival and generation, have a unique ability to produce natural antibodies, including those directed against *S pneumoniae*, *Neisseria meningitidis*, and *Haemophilus influenzae* type b, and can initiate T-cell-independent immune responses on infection or vaccination with capsular polysaccharide antigens. A decreased number of IgM memory B cells has been reported in children younger than 2 years because of marginal zone immaturity, and in patients with common variable immunodeficiency, splenectomised patients, individuals with congenital asplenia or hyposplenism, and elderly people. All these patients have an increased susceptibility to infections by encapsulated bacteria because of their inability to initiate a protective antibody response to polysaccharide vaccines.[5,11,12] However, the role of IgM memory B cells is debatable after results in *SCID/SCID* mice transplanted with human B-lymphocyte subsets and immunised with pneumococcal capsular polysaccharides[13]—the data from this study seem to contradict the belief that the IgM memory B-cell pool participates in only T-cell-independent immune response.[13] These results, together with the report that splenectomy alone might not be the only reason for loss of IgM memory B cells and reduced IgM antipneumococcal antibodies,[14] suggest that additional work to understand the role of IgM-positive CD27+ memory B cells is needed. In mice, the spleen is a reservoir for a specific population of monocytes that deploy to distant sites to favour wound healing.[15]

Diagnosis of spleen dysfunction

Diagnosis of spleen dysfunction is generally based on assessment of the spleen's filtering function by radioisotopic methods or quantitation of erythrocyte morphological abnormalities (table 1). Radioisotopic methods enable a morphofunctional assessment of the spleen by injection, uptake, and clearance of particulate substances or radiolabelled tracers.[16,17] However, their use in clinical practice is limited by their high costs and some technical difficulties. Detection methods of

	Description	Comments
Technetium-99m-labelled sulphur colloidal scintiscan[16]	Quantitation of splenic uptake of colloidal sulphur particles enables a fairly accurate static assessment of spleen function	Hypertrophy of the left hepatic lobe might be a limiting factor (this technique does not clearly show whether the mass originated in the liver or the spleen in the presence of an overlapping hypertropic left hepatic lobe)
Technetium-99m-labelled or rubidium-81-labelled heat-damaged autologous erythrocyte clearance[17]	Measurement of clearance time allows a dynamic evaluation of spleen function	Pre-existing erythrocyte defects, difficult erythrocyte incorporation of the radioisotope, false positive or negative results in relation to excessive or insufficient heat damage make the test not suitable for clinical practice
Detection of Howell-Jolly bodies[18,19] by staining	Erythrocytes with nuclear remnants	No need for special equipment; inaccurate in the quantitation of splenic hypofunction
Detection of pitted erythrocytes[20] by phase-interference microscopy	Erythrocytes with membrane indentations (4% upper limit of the normal range)	Need for phase-interference microscopy; counts enable a wide range of measurements and correlate with radioisotopic methods

Table 1: **Diagnostic techniques for and features of spleen dysfunction**

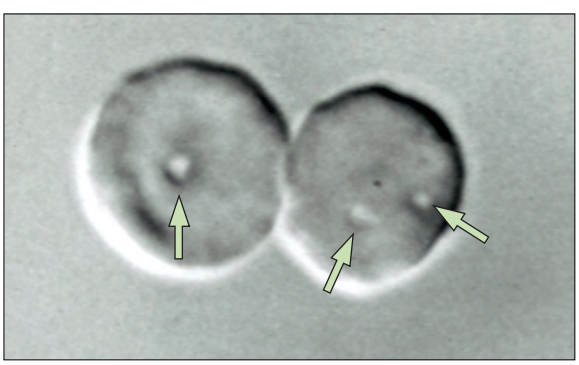

Figure 2: **Characteristic pitted erythrocytes**
A pitted erythrocyte is recognisable on phase-interference microscopy by the characteristic "pit" on the cell membrane (arrows).

morphological alterations of erythrocytes are more suitable for clinical use because these tests are easy to do, are inexpensive, and are less invasive than radiolabelling. The detection of Howell-Jolly bodies (ie, erythrocytes with nuclear remnants) is a useful method of screening for asplenia,[18] although the specificity[19] and sensitivity[18,21] of this test has been disputed, particularly in the quantitation of mild forms of hyposplenism. In one uncontrolled study,[22] quantitation of Howell-Jolly bodies by flow cytometric analysis was proposed.

Erythrocytes with characteristic membrane pits, visible with phase-interference microscopy (figure 2), were originally reported in premature neonates and splenectomised adults.[23] Because one of the functions of the spleen is the removal of pits from the circulating erythrocytes (pitting), asplenia and hyposplenism cause an increased number of circulating pitted erythrocytes. Because pitted erythrocyte counting is a simple, repeatable, and quantitative test, which accurately correlates with spleen volume,[20,24] this test is regarded as the gold standard for the assessment of spleen dysfunction in most studies. However, pitted erythrocyte counting needs specific equipment (Nomarski optics), which explains its limited use in clinical practice. The inverse correlation seen between pitted erythrocytes and IgM memory B cells in splenectomised and hyposplenic patients indicates that the impairment of the filtering function, as defined by pitted erythrocyte counting, might mirror a parallel impairment of the immunological compartment.[25,26]

The incidental finding of a small spleen during an abdominal imaging procedure should always prompt clinicians to undertake one of the appropriate tests for quantitation of splenic dysfunction.

Splenectomy

In tertiary referral centres, the frequency of splenectomy for haematological, immunological, or oncological reasons (54%) is substantially higher than that for trauma surgery (16%),[27] although this ratio might be different in non-academic settings. The increasing awareness of the risk of OPSI[28] has led to a more conservative approach, and a notable reduction in the incidence of splenectomy for trauma was recorded in children between 1970 and 2000.[29] In 2005, the Society for Surgery of the Alimentary Tract provided guidelines according to which post-traumatic splenectomy is only indicated if the patient is haemodynamically unstable, if the patient has lost more than 1000 mL of blood, if transfusion of more than two units of erythrocytes is needed, or if there is evidence of bleeding in progress. Therefore, current approaches use haemostatic repair agents on tissue lacerations, ligation and partial angioembolisation of the splenic artery, simple splenorrhaphy, and partial splenectomy. Partial splenectomy leads to only a transitory depression of humoral immunity and enables an adequate phagocytic response.[30] In patients with thalassaemia who have had partial splenectomy, the risk of OPSI was lower than in those undergoing total removal of the spleen,[31] and results from long-term follow-up studies did not indicate a significantly increased frequency of infections.[32] However, no definite information is available as to whether antibiotic treatment or pneumococcal vaccination is necessary in patients who undergo conservative surgery. Thus, the same measures recommended for total splenectomy should also be adopted for patients undergoing partial removal of the spleen.

When the extent of the traumatic lesions makes removal of the whole spleen necessary, deliberate heterotopic autotransplantation of splenic tissue in omental pockets might lead to sufficient maintenance of splenic function.[33] The spontaneous occurrence of this phenomenon, after rupture of the splenic capsule, was first reported in 1939[34] and is known as splenosis. Splenotic nodules, which consist of vital and functioning splenic tissue, are usually numerous, measure from a few mm to a few cm in diameter, and stain the omentum, the peritoneum, and the mesentery (figure 3). These nodules can be distinguished from accessory spleens by their high number, the absence of hilum, capsule, and trabeculae, the vascularisation provided by small omental vessels, and their localisation that is often separate from the splenopancreatic and gastrosplenic ligaments.[33]

A high frequency of splenosis, confirmed by the reduction of circulating pitted erythrocytes, was found in children splenectomised for trauma (59% of 22 patients).[35] Additionally, normal pitted erythrocyte counts were reported in patients after partial splenectomy or splenorrhaphy, whereas patients with deliberate autotransplantation or spontaneous splenosis had pitted erythrocyte counts intermediate between normal counts and counts seen in splenectomised patients without splenosis.[36]

In splenectomised patients with splenosis, the proportion of pitted erythrocytes is indicative of the volume of regenerated splenic tissue, and it has been calculated that 30 mL is the minimum volume of reimplanted splenic tissue needed to ensure a return of splenic function sufficient to guarantee some protection.[37]

Although there is evidence that reimplantation of splenic tissue ensures a good antibody response after pneumococcal vaccination, splenosis does not seem to guarantee full protection against OPSI. Possible factors that restrict the defensive efficacy of the splenotic nodules include, in addition to their small size, poor vascularisation, which reduces the contact between particulate antigens and splenic phagocytes.

Hyposplenism

The natural history of many diseases, including congenital, haematological, immunological, gastroenterological, infectious, and iatrogenic disorders, can be complicated by splenic abnormalities ranging from a reversible mild hyposplenism to severe splenic atrophy (panel).[6] In most of these conditions, the pathogenesis of hyposplenism is mostly unknown.[8] Table 2 describes the main clinical aspects of the most frequent diseases complicated by splenic hypofunction or atrophy. Because of the relevance of hyposplenism and its consequences in sickle-cell anaemia, bone marrow transplantation, graft-versus-host disease, coeliac disease, and HIV infection, we discuss these disorders in more detail in this section.

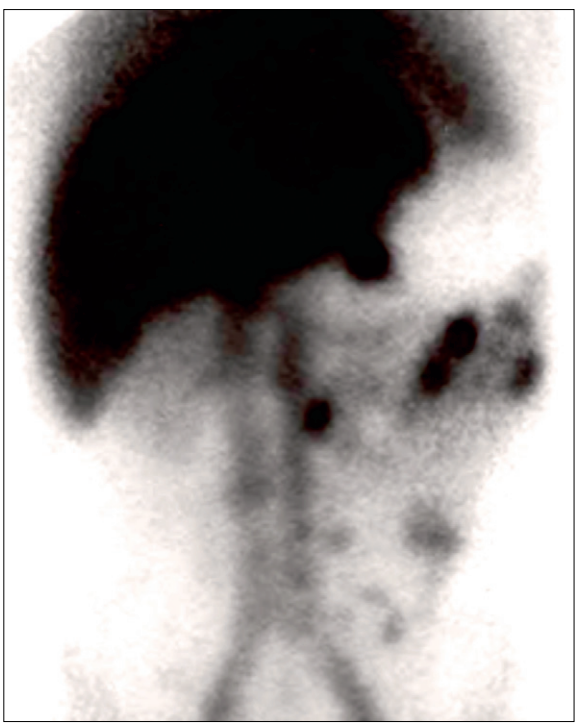

Figure 3: **Splenosis in a splenectomised patient**
Posteroanterior technetium-99m heat-damaged erythrocyte scan shows at least ten splenotic nodules in the left-upper abdomen of a patient undergoing deliberate splenic autotransplantation after splenectomy.

Sickle-cell anaemia

Sickle-cell anaemia is invariably associated with severe splenic dysfunction.[38] In the first years of life of patients with sickle-cell anaemia, the size of the spleen increases because of vaso-occlusive congestion secondary to recurrent episodes of erythrocyte sickling. These events lead to engorgement of the sinusoids, although severe functional hyposplenism is already present and documented by scintiscan and pitted erythrocyte count.[39] As a consequence, patients are highly susceptible to sepsis by encapsulated bacteria, particularly before 3 years of age, reaching a risk level of sepsis or meningitis that is about 300–600 times higher than in the general population.[66]

Sustained high concentrations of haemoglobin F (≥20%) are recognised as a protective factor against impaired splenic function and are associated with mild disease and low mortality.[40] Drugs that substantially increase production of haemoglobin F, such as hydroxycarbamide, can improve clinical outcomes in patients with sickle-cell anaemia. Both treatment with hydroxycarbamide and blood transfusions revert hyposplenism in young children.[41–43] Hypertransfusion therapy even reversed splenic involution and fibrosis in some children with sickle-cell anaemia.

Although the general outcome of children with sickle-cell anaemia has substantially improved as a result of better comprehensive care, transfusion therapy, and

Panel: **Diseases associated with hyposplenism or splenic atrophy**

Congenital forms
- Normal and premature neonates
- Isolated congenital hyposplenia
- Ivemark's syndrome
- Autoimmune polyendocrinopathy-candidiasis-ectodermal dystrophy (APECED) syndrome
- Hypoparathyroidism syndrome
- Stormorken's syndrome

Gastrointestinal disorders
- Coeliac disease
- Inflammatory bowel diseases
- Whipple's disease
- Dermatitis herpetiformis
- Intestinal lymphangiectasia
- Idiopathic chronic ulcerative enteritis

Hepatic disorders
- Active chronic hepatitis
- Primary biliary cirrhosis
- Hepatic cirrhosis and portal hypertension
- Alcoholism and alcoholic hepatopathy

Oncohaematological disorders
- Haemoglobin S diseases
- Bone marrow transplantation
- Chronic graft-versus-host disease
- Acute leukaemia
- Chronic myeloproliferative disorders

Autoimmune disorders
- Systemic lupus erythematosus
- Rheumatoid arthritis
- Glomerulonephritis
- Wegener's granulomatosis
- Goodpasture's syndrome
- Sjögren's syndrome
- Nodous polyarteritis
- Thyroiditis
- Sarcoidosis

Infectious diseases
- HIV/AIDS
- Pneumococcal meningitis
- Malaria

Iatrogenic forms
- Exposure to methyldopa
- High-dose steroids
- Total parenteral nutrition
- Splenic irradiation

Alteration in splenic circulation
- Thrombosis of splenic artery
- Thrombosis of splenic vein
- Thrombosis of coeliac artery

Miscellaneous
- Amyloidosis

treatment with hydroxycarbamide,[44] repeated splenic infarcts and the consequent fibrosis lead to spleen atrophy (autosplenectomy) and irreversible hyposplenism.[45] However, prophylactic treatment with benzylpenicillin, vigilance at times of fever, and vaccination can reduce this risk. Acute chest syndrome and multiorgan failure syndrome are now the leading causes of death in this disorder rather than bacterial sepsis.[44]

All the above reported data refer to individuals who are homozygous for the haemoglobin S gene or who are affected by haemoglobin $S\beta^0$-thalassaemia. The splenic deficit is less severe in patients who are homozygous for the haemoglobin S gene associated with α-thalassaemia, in individuals with haemoglobin $S\beta^+$-thalassaemia, and in patients with haemoglobin SC disease.[39]

Bone marrow transplantation and graft-versus-host disease

After earlier case reports, results from two studies using pitted erythrocyte counting and Howell-Jolly bodies detection indicated that functional hyposplenism might complicate 15–40% of allogeneic bone marrow transplantation.[46,67] Additionally, a positive correlation was identified between reduction in spleen size, the duration of chronic graft-versus-host disease, and an increased risk of life-threatening pneumococcal infections.[47] The report of deficiency of IgM memory B cells in allogeneic haemopoietic stem-cell transplantation recipients[48] might provide a pathophysiological explanation for the high susceptibility of these patients to infections by encapsulated bacteria, mainly *S pneumoniae*. *S pneumoniae* causes severe and sometimes fatal invasive infections months or years after transplantation, with an incidence of up to 27% in long-term survivors.[49] In conclusion, functional impairment of the spleen should be carefully taken into account in all individuals who undergo allogeneic bone marrow transplantation, especially in those who develop chronic graft-versus-host disease; furthermore, these patients have been proposed to receive life-long antibiotic prophylaxis.[68]

Coeliac disease

Coeliac disease, an immune-mediated enteropathy induced in genetically susceptible individuals by the ingestion of gluten,[69] is the most frequent disorder associated with hyposplenism.[50] Splenic hypofunction, with or without splenic atrophy, is a frequent complication of coeliac disease, with a prevalence ranging from 33% to 76%.[51,52] When these data are categorised according to clinical severity, the prevalence

	Diagnostic method	Prevalence of hyposplenism (%)	Degree of hyposplenism	Evidence of OPSI	Additional information
Sickle-cell anaemia[38-45]	Pitted erythrocyte count	100	Severe	+ + +	Hyposplenism worsens with decreasing concentrations of haemoglobin F
Bone marrow transplantation or GVHD[46-49]	HJB detection; pitted erythrocyte count	40; 15	Moderate to severe	+ + +	Hyposplenism is more frequent in patients with extensive GVHD; need for antibiotic prophylaxis
Coeliac disease[25,50-55]	Pitted erythrocyte count	33–76	Moderate to severe	+ + +	Hyposplenism reversible after gluten-free diet; decreased concentrations of IgM memory B cells; poor prognosis in patients with splenic atrophy
HIV/AIDS[56-60]	Pitted erythrocyte count	36	Moderate to severe	+ + +	Decreased concentrations of IgM memory B cells
Alcoholic liver disease[61]	Pitted erythrocyte count	37–100	Moderate to severe	+ + +	Abstinence improves splenic function
Inflammatory bowel disease[26,62]	Pitted erythrocyte count	35–45 (UC); 9–37 (CD)	Mild to moderate (UC>CD)	+ +	Decreased concentrations of IgM memory B cells; poor prognosis in patients with splenic atrophy
Whipple's disease[63]	Pitted erythrocyte count	47	Mild	–	Thrombocytosis; thrombotic events
Primary amyloidosis[64]	HJB detection	28	Moderate	+ +	Poor prognosis in hyposplenic patients
Systemic lupus erythematosus[65]	HJB detection; pitted erythrocyte count	7; 5	Mild to moderate	+ +	Hyposplenism unrelated to disease activity

–=no evidence. +=weak evidence. ++=moderate evidence. +++=strong evidence. OPSI=overwhelming post-splenectomy infections. GVHD=graft-versus-host disease. HJB=Howell-Jolly bodies. UC=ulcerative colitis. CD=Crohn's disease.

***Table 2*: Clinical features of the most common hyposplenism-associated disorders**

of hyposplenism increases from 19% in patients with uncomplicated disease, to 59% in those with associated autoimmune diseases, and up to 80% in those with premalignant or malignant complications.[25] Hyposplenism does not complicate coeliac disease in infancy, and its occurrence in adults correlates with the duration of pre-exposure to gluten.[51] A gluten-free diet is effective in restoring splenic function except in patients who already have an irreversible loss of splenic tissue.[52,53] Splenic atrophy, which is now recognised as an indicator of poor prognosis in patients with coeliac disease and is often associated with mesenteric lymph node cavitation (figure 4), affects the size of both the marginal zone and the white pulp B-cell compartment, and causes splenic IgM memory B-cell deficiency. This B-cell deficiency might contribute to the increased susceptibility of patients with coeliac disease to severe infections. This predisposition is supported by data from two ad-hoc studies, which indicate a substantially higher relative risk of pneumococcal sepsis in adult patients with coeliac disease than in the general population.[54,55] These data are in accordance with the increased mortality reported in these patients because of major infections.[70]

HIV infection

Impaired reticuloendothelial function was first reported in patients with AIDS in 1985,[56] and data from a subsequent study that used pitted erythrocyte counting confirmed that functional hyposplenism can affect patients who are HIV positive, with a prevalence of 36%.[57] Moreover, in patients with AIDS, splenic function was correlated with the activity of the spleen-derived opsonising factor tuftsin.[58] More recently, depletion of IgM memory B cells has been reported in untreated patients with HIV who have a CD4 T-cell count of less than 300 cells per µL.[59] This depletion might be a risk factor for pneumococcal disease, which occurs in patients with HIV with an incidence that is 100-times greater than in the general population. Antiretroviral therapy seems to be effective in restoring the IgM memory B-cell pool,[59] which might partly explain the low rates of pneumococcal disease in patients with HIV receiving this therapy. In another series,[60] 70% of 84 patients with HIV had similar numbers of IgM memory B cells to those in splenectomised individuals, and this loss was associated with diminished vaccination responses to both T-cell-dependent and T-cell-independent antigens.

OPSI

The term OPSI defines fulminating sepsis, meningitis, or pneumonia triggered mainly by *S pneumoniae*, *N meningitidis*, and *H influenzae* type b in splenectomised and hyposplenic individuals.[29] Although data from several studies[6] have confirmed that asplenia and hyposplenism are major risk factors for sepsis, prevention of these infections is often overlooked. Nevertheless, extrapolation

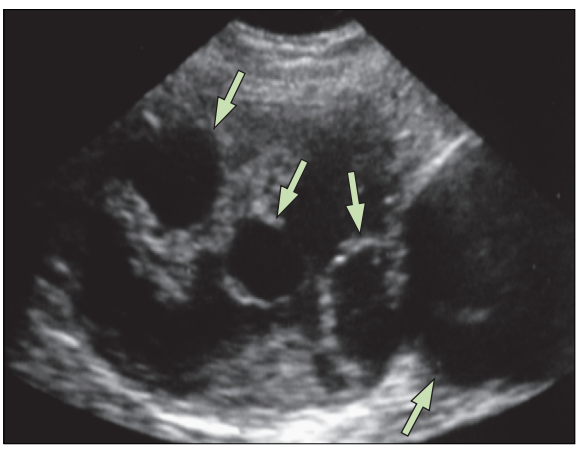

Figure 4: **Mesenteric lymph node cavitation in a patient with refractory coeliac disease complicated by ulcerative jejunoileitis and splenic atrophy**
Multiple cavitations of mesenteric lymph nodes appear on abdominal bowel sonography as anechoic cystic masses with posterior acoustic enhancement (arrows).

of definite figures on the epidemiology of OPSI from a series of retrospective studies is difficult because of the tendency to over-report the events, the variability in the selection and inclusion criteria, and the differences in the duration of the follow-up and in the stratification of patients by age, underlying disease, and type of splenectomy. Similarly, the distinction in terms of risk of OPSI between splenectomised patients and patients with acquired hyposplenism is not possible because of the absence of comparable data.

An analysis of 78 studies done between 1966 and 1996 enabled investigation of 19 680 splenectomised patients for a mean period of 6·9 years.[28] The prevalence of infections was 3·2% with a mortality rate of 1·4%. A more detailed analysis was only possible in a few patients, in whom there was an unexpected similar prevalence of infections between children (3·3%) and adults (3·2%), with a higher mortality rate in children (1·7% *vs* 1·3%). The risk of sepsis and death was strongly associated with the reason why splenectomy was done. The indications for splenectomy most frequently associated with a risk of infection and death were thalassaemia major (8·2% and 5·1%, respectively), sickle-cell anaemia (7·3% and 4·8%), Hodgkin's lymphoma (4·1% and 1·9%), spherocytosis (3·1% and 1·3%), and idiopathic thrombocytopenic purpura (2·1% and 1·2%). The prevalence of OPSI and the mortality rate in splenectomy for trauma were 2·3% and 1·1%, respectively. The results of this analysis do not noticeably differ from those of the individual studies analysed, but might underestimate the risk of OPSI because of the short duration of the follow-up in many of the studies. The opinion that OPSI occur in the first few years after surgery[71] is not universally shared.[72] The risk of sepsis in the absence of the spleen is a permanent condition; cases of OPSI have been described 20–40 years after removal of the spleen,[73] and the cumulative prevalence of infections needing admission to hospital rose progressively from 17% at 1 year to 33% at 10 years.[74] The underlying disease and concomitant factors that depress the immune system (malnutrition, chemotherapy, alcoholism, diabetes) have an important role. The rare Wiskott-Aldrich syndrome, which was not included in Bisharat and colleagues' survey,[28] was reported as the disorder with the highest risk of post-splenectomy infection (30·7%) and death (12·8%).[75] Finally, in patients who had splenectomy because of idiopathic thrombocytopenic purpura, the not-infrequent coexistence of common variable immunodeficiency substantially increased the risk of sepsis.

The risk of OPSI in splenectomised patients is more than 50-times higher than in the general population,[29] and *S pneumoniae* is the most common causal organism (50–90% of cases), followed by *H influenzae* type b and *N meningitidis*.[71] A predominant pneumococcal serotype has not been documented, and serotype distribution did not differ between OPSI and other forms of pneumococcal infections. Two surveys on invasive pneumococcal disease reported different results: one concluded that host factors are more important than isolate serotype in establishing the severity and outcome of infections,[76] whereas the other reported that serotypes 18C, 9N, 6B, 16F, 23A, 19F, and 3 are those most commonly associated with increased case-fatality rates.[77] The prominence of *Salmonella* sp in patients with sickle-cell disease, which mostly causes osteomyelitis, seems to be associated with further determinants in addition to impaired splenic function—eg, abnormal humoral immunity, bone ischaemia, or infarction.[78] Causative organisms that are less frequently implicated are *Escherichia coli*, *Pseudomonas aeruginosa*, *Capnocytophaga canimorsus* (transmitted by dog bites), and, more rarely, *Enterococcus* sp, *Bacteroides* sp, and *Bartonella* sp.

OPSI is a medical emergency for which only prompt diagnosis and immediate treatment can reduce mortality.[79] The clinical course of OPSI is measured in hours rather than days. The mortality rate is 50–70%, and most deaths occur within the first 24 h.[71] Bacteraemia commonly has an unknown origin; after a brief prodrome characterised by fever, shivering, myalgia, vomiting, diarrhoea, and headache, septic shock develops in just a few hours, with anuria, hypotension, hypoglycaemia, and, commonly, disseminated intravascular coagulation and massive adrenal gland haemorrhage (Waterhouse-Friderichsen syndrome), to multiorgan failure and death.[79]

In patients at risk and with indicative symptoms of OPSI, particularly fever, treatment with empirical antibiotics is essential, and involves infusion with third-generation cephalosporins (cefotaxime 2 g every 8 h or ceftriaxone 2 g every 12 h), combined with gentamicin (5–7 mg/kg every 24 h) or ciprofloxacin (400 mg every 12 h) if a possible urinary or intestinal focus is suspected, or vancomycin (1–1·5 g every 12 h) in case of resistance to benzylpenicillin.[79] While waiting for the results of

blood-culture tests, bacteria can be visualised by Gram or Wright staining in the buffy-coat, or even in the peripheral blood smear. This empirical approach can be changed to a more specific treatment once the nature of the pathogen is known. A real-time PCR test, which enables the simultaneous identification of the three main encapsulated bacteria causative for OPSI, is available.

Prevention of OPSI

Education

Up to 84% of splenectomised individuals are thought to be unaware of their increased susceptibility to severe sepsis,[80] and provision of proper information reduces infectious complications.[81] In particular, patients and relatives should be instructed to notify their physicians of any acute febrile illness, especially if associated with rigor and systemic symptoms,[81] and of visits to tropical countries because of the high risk of parasitic infections, such as malaria or babesiasis. In particular, travellers should take preventive measures such as antimalarial prophylaxis, mosquito repellents, and other barrier precautions. Other measures include immediate treatment in the event of dog bites or bites of other animals, although there is no consensus on the need for prophylactic antibiotics in case of dental procedures. Disappointingly, these recommendations are ignored by doctors themselves,[72] whereas constant monitoring is essential because the risk of OPSI is long term, compliance with prevention tends to decrease over time, and vaccination itself might gradually lose efficacy. Careful monitoring of these patients would be greatly helped by specific registers, and this approach has a favourable cost-benefit ratio, in terms of prevention and reduction of mortality.[82]

Antibiotic prophylaxis

The use of antibiotics for the prevention of OPSI is not evidence based. The actual effectiveness of antibiotics is unknown and there is no agreement on how long these drugs should be taken for or which subgroups to treat, especially when factors such as poor compliance and possibility of favouring the development of resistant bacterial strains in the long term are taken into account.[83] Moreover, antibiotics might reduce but not abolish the risk of OPSI. Apart from data from two controlled trials published in the 1980s that indicated the efficacy of prophylactic benzylpenicillin against pneumococcal infections in children with sickle-cell anaemia,[84,85] we are not aware of any study that has assessed antibiotic prophylaxis in other subgroups of asplenic or hyposplenic individuals. Furthermore, we identified only two retrospective studies done on large cohorts of splenectomised children, which reported a substantial reduction of the occurrence of OPSI during treatment with benzylpenicillin.[86,87] Most of the patients, however, had concomitant pneumococcal vaccination, which makes estimation of the contribution of antibiotics difficult.

Compliance with long-term antibiotic treatment is often inadequate and remains suboptimal in children even after ad-hoc education of parents.[81] Guidelines recommend antibiotic prophylaxis in children younger than 5 years, but there is no agreement between Canadian, British, and American guidelines on when to discontinue prophylactic benzylpenicillin. In adults, guidelines recommend prophylaxis with 250–500 mg per day of amoxicillin or 500 mg per day phenoxymethylpenicillin. In children allergic to these antibiotics, possible alternatives are co-trimoxazole or erythromycin, although these options are becoming less effective because of the increasing development of resistant pneumococcal strains.[83] Although there is no consensus on the duration of treatment, the British guidelines propose life-long treatment with regards to the persistent risk of sepsis. Long-term treatment is recommended in patients with concomitant haematological diseases or an impaired immune system. Episodic short-term antibiotics at the time of febrile illnesses might be a useful strategy in asplenic patients, although robust data are needed.

Vaccine prophylaxis

Vaccines used in patients at risk of OPSI are the 23-valent pneumococcal polysaccharide vaccine (PPV-23; serotypes include 1–5, 6B, 7F, 8, 9N, 9V, 10A, 11A, 12F, 14, 15B, 17F, 18C, 19A, 19F, 20, 22F, 23F, and 33F), the 7-valent diphtheria cross-reactive material 197 (CRM197) protein-conjugate pneumococcal vaccine (PCV-7; serotypes include 4, 6B, 9V, 14, 18C, 19F, and 23F), the *H influenzae* type b conjugate vaccine, and the meningococcal vaccine.

Although PPV-23 reduces (but does not abolish) the occurrence of OPSI in splenectomised patients,[81] this vaccine is not commonly used. In 2005 in England and Wales, only 366 (13·2%) of 2770 splenectomised patients had been vaccinated in the previous 12 months and 1479 (53·4%) had been vaccinated in the previous 5 years.[88] For patients with sickle-cell anaemia, 473 (13·2%) had been vaccinated in the previous 12 months and 1914 (53·4%) had been vaccinated in the previous 5 years. Of the 3584 patients with either sickle-cell disease or coeliac disease, 154 (4·3%) had been vaccinated in the previous 12 months, and 570 (15·9%) had been vaccinated in the previous 5 years.

The protective action of PPV-23 is based on the production of opsonising anticapsular antibodies by means of a T-cell-independent mechanism.[89] PPV-23 is recommended for adults or children older than 5 years who are scheduled for elective splenectomy. This vaccine should be given at least 2 weeks before surgery or, in the event of emergency splenectomy, 2 weeks after surgery.[90] PPV-23 provides protection against many capsulated antigens, albeit only temporarily. Therefore, revaccination after 5 years is advisable, although the serological response declines more rapidly and frequent reimmunisation in specific subgroups is needed (eg, sickle-cell anaemia, myeloproliferative diseases,

and lymphoproliferative diseases). However, these recommendations are made despite only few data on antibody persistence after vaccination and on antibody responses to revaccination. Monitoring of antibody titres at intervals with ELISA might be helpful for the assessment of whether revaccination is needed; measurement of functional antibody titres by opsonophagocytic assay seems to be preferable to ELISA to predict the extent of protection or to establish an ad-hoc retreatment schedule,[91] although the availability of this assay is limited to a few research laboratories at present because of the increased technical difficulty.

In addition to the limited persistence of circulating antibodies post-vaccination, other factors that counteract effectiveness of PPV-23 are acquired immune deficiency, increasing age, and a genetically determined inability to respond to most or all polysaccharides contained in this vaccine. Ineffectiveness of this vaccine might also occur when bacteraemia is caused by a serotype not included in PPV-23.[92]

The more recent and costly conjugate vaccines protect against fewer serotypes than does PPV-23 but are more immunogenic.[93] In PCV-7, conjugation of polysaccharides to the CRM197 protein changes the antipolysaccharide response from T-lymphocyte-independent responses to T-lymphocyte-dependent ones. This effect makes this vaccine particularly suitable for infants, especially those younger than 2 years who still have an immature B-cell-dependent splenic compartment, and for patients who have already undergone splenectomy or have hyposplenism. In these groups, the pre-existing deficit of IgM memory B cells might be an additional cause of ineffectiveness of PPV-23,[5] which has been repeatedly reported in hyposplenic and splenectomised patients.[72,94] By contrast, there was an adequate immune response after PCV-7 in both experimental and human asplenia,[95,96] and vaccination of splenectomised patients who had invasive pneumococcal disease despite previous PPV-23 immunisation was protective.[97] Finally, combined use of PCV-7 followed by PPV-23 was effective in hyposplenic patients with sickle-cell anaemia,[98] HIV infection,[99] and individuals splenectomised for β-thalassaemia.[100] The explanation for this effectiveness is the less prominent role of the spleen in the anamnestic response in comparison to the generation of the primary response to polysaccharides.[95]

The novel protein-conjugate vaccine PCV-13 (serotypes include 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F)[101] offers coverage against several additional pneumococcal strains in comparison to PCV-7 or PCV-10 (serotypes include 1, 4, 5, 6B, 7F, 9V, 14, 18C, 19F, and 23F),[102] and is expected to replace these other pneumococcal vaccines.[103] PCV-13 includes all seven serotypes most frequently involved in antibiotic resistance (6A, 6B, 9V, 14, 19A, 19F, 23F).[104] Finally, conjugating capsular polysaccharides to pneumococcal surface peptides might offer new therapeutic options in the future.

Current guidelines recommend immunisation against *H influenzae* type b and *N meningitidis*, for adults and children. The *H influenzae* type b conjugate vaccine was immunogenic in splenectomised patients, although the antibody response was lower and faded more rapidly in these patients than in control individuals.[105] There is no agreement on the need for reimmunisation in children, and a single vaccination in adults seems sufficient. For meningococcal immunisation, the conventional tetravalent vaccine protects against strains A, C, Y, and W135 capsular polysaccharides. Independent of T cells, this vaccine has little efficacy in infants younger than 2–3 years,[106] whereas a conjugate vaccine against meningococcal serogroup C is highly immunogenic in the first few months of life and in adults. A single dose of this vaccine is thought to be sufficient,[107] but because revaccination increases the effective antibody response from 80 to 93%, revaccination has been proposed if protective titres are not achieved. The availability of a new tetravalent conjugate meningococcal vaccine[108] might be useful, especially for hyposplenic or splenectomised patients who travel to countries in which *N meningitidis* is hyperendemic or epidemic.

Conclusions

Although post-splenectomy and hyposplenic states might predispose individuals to thromboembolic complications, the main adverse events are immunological and infectious. Spleen-preserving surgical techniques have become increasingly common both for emergency and elective splenectomy; however, the morbidity and mortality associated with the absence and the dysfunction of the spleen are still unacceptably high. In disorders associated with hyposplenism, spleen function should be assessed by means of an appropriate technique (scintiscan, pitted erythrocyte counting, or Howell-Jolly body detection), and, if splenic function is reduced—particularly if associated with atrophy—then all preventive measures against OPSI should be adopted. For children scheduled for splenectomy, postponement of the operation is advisable when possible until the age of 6–12 years. During splenectomy for trauma, surgeons should try to save as much tissue as possible, resorting to deliberate splenosis if necessary. In view of the high mortality rate of OPSI, prophylaxis against encapsulated bacteria is an unavoidable option, and the choice of the most appropriate vaccine is secondary to awareness that vaccination is necessary. Clear clinical and experimental evidence suggests that conjugate vaccines are preferable to the traditional PPV-23 in young children, although there are as yet insufficient data to recommend the use of these vaccines in splenectomised patients and in those with major hyposplenism. Patients, family members, and general practitioners need to be aware of the long-term risk of OPSI and of the advisability of antibiotic treatment not only in children soon after surgery and in patients who

are immunologically impaired, but also in particular situations such as animal bites and visits to at-risk countries. In asplenic or hyposplenic states, any fever must be promptly and carefully assessed and treated.

Contributors

ADS and GRC wrote the clinical part of the Review. ADS and RC wrote the immunological part of the Review. RC participated in writing the section on pneumococcal vaccines.

Conflicts of interest

We declare that we have no conflicts of interest.

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