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# Bone marrow failure syndromes

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## Bone marrow failure syndromes: what they comprise

Bone marrow failure syndromes are characterised by the primary failure to produce one or more blood cell lineages. Secondary causes of marrow failure such as chemotherapy or radiotherapy, marrow infiltration or peripheral autoimmune destruction of blood cells are excluded. Marrow failure may occur at the level of the haemopoietic stem cell resulting in aplastic anaemia or at a later developmental stage of haemopoiesis affecting a single lineage. Single lineage marrow failure disorders may later progress to aplastic anaemia. Bone marrow failure syndromes are most commonly acquired but there are rare congenital forms.[1]

In aplastic anaemia, there is overlap with clonal disorders such as paroxysmal nocturnal haemoglobinuria (PNH) and myelodysplastic syndromes (MDS, pre-leukaemia). Previously, abnormal cytogenetic clones were thought to indicate MDS, but it is now clear that abnormal clones as well as PNH clones may be present in aplastic anaemia or arise during the course of the illness and then disappear.[2]

## Aplastic anaemia

### *Definition, disease severity and causes*

Aplastic anaemia is defined by:

- pancytopenia
- hypocellular bone marrow where normal haemopoietic cells are replaced by fat cells and there is no increase in reticulin or fibrosis (Fig 1), and
- absence of abnormal cells in the bone marrow.

There are three grades of severity of aplastic anaemia, as defined by peripheral blood counts and the degree of marrow hypocellularity. In non-severe aplastic anaemia the neutrophil count is $>0.5 \times 10^9$/l, in severe aplastic anaemia $<0.5 \times 10^9$/l, and in very severe aplastic anaemia $<0.2 \times 10^9$/l.[3]

Aplastic anaemia is a rare idiosyncratic disorder, with an incidence in the West of 1–2 per million population per annum. It is 2–3 times more common in the Far East. The disease is idiopathic in many cases but may be drug-induced or occur following an episode of acute viral hepatitis (Table 1).

### *Pathogenesis*

Aplastic anaemia is characterised by a defect in the haemopoietic stem cell compartment, with both deficiency and

## Key Points

**Aplastic anaemia is a potentially life-threatening disorder, but treatment outcomes with bone marrow transplantation and immunosuppressive therapy have improved considerably with time**

**In many patients there is evidence of an autoimmune basis for acquired bone marrow failure disorders**

**The genetic basis of congenital forms of bone marrow failure is now more clearly understood**

**KEY WORDS: amegakaryocytic thrombocytopenia, aplastic anaemia, cyclic neutropenia, pure red cell aplasia, severe congenital neutropenia**

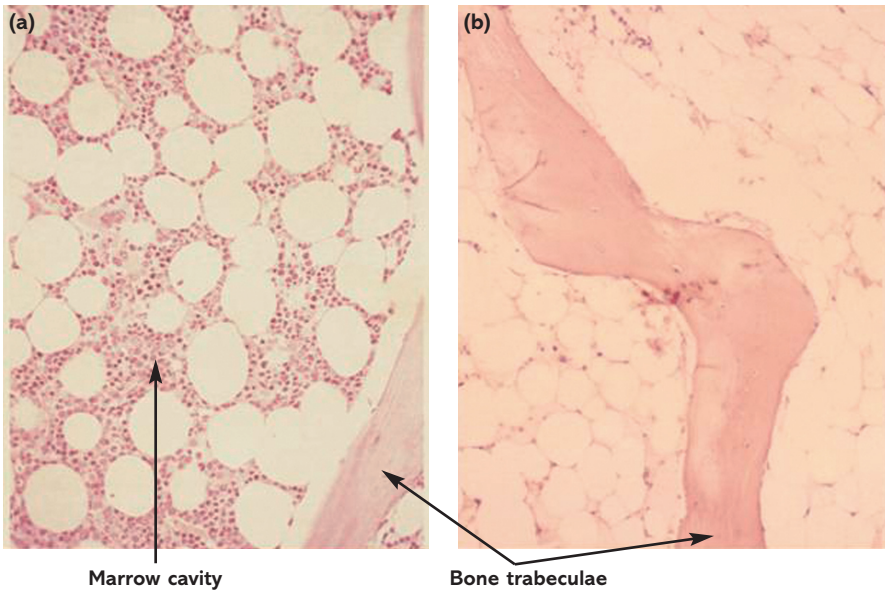


**Fig 1. Normal and severe aplastic anaemia bone marrow biopsies. Histological sections (a)** normal bone marrow with a normal proportion of haemopoietic cells interspersed with fat cells in the marrow cavity; **(b)** bone marrow from a patient with severe aplastic anaemia showing virtual absence of haemopoietic cells in the marrow cavity and replacement with fat cells.

defective function. The marrow microenvironment functions normally, as assessed by long-term marrow culture.[4] Aplastic anaemia occurs as an idiosyncratic reaction and there is increasing evidence for an autoimmune basis to the disease (Table 2; Fig 2).[5–7]

### *Clinical presentation*

Patients present with symptoms of anaemia or thrombocytopenia or bacterial infection due to neutropenia. Purpura, bruising, gum bleeding, epistaxes and menorrhagia are common.

**Table 1. Aetiology of acquired aplastic anaemia.**

| | |
|---|---|
| Idiopathic | 70–80% of cases |
| Drugs* | Antibiotics: chloramphenicol (no evidence for chloramphenicol eye drops), sulphonamides<br>Antirheumatics: gold, penicillamine<br>Anti-inflammatory drugs: phenylbutazone, indomethacin, diclofenac, naproxen, piroxicam<br>Anticonvulsants: phenytoin, carbamazepine<br>Antithyroids: carbimazole (more likely to cause neutropenia), thiouracil<br>Antidepressants: dothiepin, phenothiazines<br>Antidiabetics: chlorpropamide<br>Antimalarials: quinine |
| Chemicals | Benzene<br>Pesticides: organochlorines (eg lindane, organophosphates)<br>Cutting oils and lubricating agents<br>Recreational drugs: methylenedioxy-methamphetamine (MDMA, ecstasy) |
| Viruses | Viral hepatitis: non-A, non-B, non-C and non-G in most cases<br>EBV rarely |
| PNH | Haemolytic PNH in 5%; small PNH clone can be detected by flow cytometry in at least 20–25% of AA patients at presentation |
| Rarely | SLE, pregnancy, anorexia nervosa, thymoma |

* Drugs currently licensed in the UK reported to have a rare association with aplastic anaemia (AA).
EBV = Epstein-Barr virus; PNH = paroxysmal nocturnal haemoglobinuria; SLE = systemic lupus erythematosus.

There is a risk of spontaneous life-threatening haemorrhage such as cerebral haemorrhage. A careful drug and occupational history should be obtained. Children and young adults should be examined for features of congenital aplastic anaemia such as short stature, skeletal anomalies, hypo- or hyperpigmentation of the skin. Examples of congenital aplastic anaemia are Fanconi anaemia and dyskeratosis congenita.[1]

### *Investigations*

The following investigations should be performed:

- full blood count and blood film (pancytopenia with no abnormal cells such as blasts)
- reticulocyte count (reticulocytopenia)
- bone marrow aspirate and trephine biopsy (hypocellular bone marrow with no evidence of infiltration or blasts or increased fibrosis)
- marrow cytogenetics (to detect an abnormal clone)
- flow cytometry of glycosyl-phosphatidyl inositol-anchored proteins on blood cells (deficient expression on red cells, neutrophils and monocytes indicates a PNH clone)
- liver function tests and viral hepatitis screen (to exclude posthepatitic aplastic anaemia)
- serum vitamin B12 and folate (to exclude severe megaloblastic anaemia which may cause pancytopenia)
- autoimmune profile (systemic lupus erythematosus is a rare cause of aplastic anaemia)
- for children and young adults below 35 years old, exclude congenital aplastic anaemia – in Fanconi anaemia there are increased spontaneous and diepoxybutane-induced chromosome breakages in cultured peripheral blood lymphocytes, and many cases of dyskeratosis congenita show mutations of the telomerase gene complex.[8]

### *Treatment*

*Supportive care.* Initial treatment is to stabilise the patient clinically with blood and platelet transfusions and to administer broad-spectrum intravenous antibiotics for fevers. When the platelet count is below $10 \times 10^9$/l prophylactic platelets should be given and prophylactic antibiotics and antifungal drugs if there is severe neutropenia. HLA antibodies may complicate transfusions, necessitating HLA-matched platelet transfusions.[3]

*Bone marrow transplantation.* Early bone marrow transplantation (BMT) from an HLA identical sibling is indicated as first-line therapy if the patient has severe or very severe disease and is younger than 40 years of age.[9] An immunosuppressive, non-myeloablative preparative regimen is used to help prevent graft rejection (cyclophosphamide 200 mg/kg with antithymocyte globulin (ATG)). Ciclosporin and methotrexate are used to prevent graft versus host disease. Irradiation is not necessary, so children grow and develop normally post-BMT and fertility is well preserved. The success rate is good, with a 70–90% chance of long-term cure for those patients younger than 40 years of age (Fig 3).[10]

BMT from an HLA matched volunteer donor (MUD BMT) may be considered for those patients with severe or very severe disease who do not have an HLA compatible sibling and who have failed to respond to immunosuppressive therapy (see below).[3,11]

*Immunosuppressive therapy.* Immunosuppression, using the combination of ATG and ciclosporin, is indicated for all patients over 40 years old, all those with non-severe disease and for younger patients with severe or very severe disease who lack an HLA compatible sibling donor. Although 60–80% will respond and achieve normal or near-normal blood counts, recovery is unstable. Relapse may occur in 10–30%, necessitating further immunosuppression, approximately 10% later develop haemolytic PNH, and a further 10% MDS or acute myeloid leukaemia. Nevertheless, a good quality of life is possible for most patients.[12]

**Table 2. Pathogenesis of aplastic anaemia.**

| Evidence | |
|---|---|
| **Haemopoietic stem cell defect** | **Autoimmune basis to the disease** |
| Bone marrow colony forming cells of all cell lineages reduced or absent | HLA restriction: over-representation of HLA-DR15 |
| Long-term marrow cultures show reduced marrow repopulating ability | Syngeneic BMT unsuccessful in at least 50% of patients unless prior immunosuppression given |
| Reduced or absent long-term culture initiating cells (LTC-IC) in bone marrow | 60–80% patients respond to immunosuppressive therapy using ATG and ciclosporin |
| Reduced % bone marrow haemopoietic progenitor (CD34+) cells compared with normal bone marrow | Increased levels of cytokines that inhibit haemopoiesis (IFN-γ, TNF-α) and upregulate Fas-antigen expression in blood and marrow |
| Bone marrow CD34+ cells are more apoptotic than normal CD34+ cells | Increased Fas-antigen expression on bone marrow CD34+ cells |
| Bone marrow cells have shortened telomere lengths compared with normal bone marrow cells | Activated cytotoxic CD8 T cells present in blood and bone marrow |
| | T cell repertoire analysis shows oligoclonal expansion of CD8 T-cells |
| | Upregulation of apoptosis and immune response genes |

ATG = antithymocyte globulin; BMT = bone marrow transplantation; IFN = interferon; LTC-IC = long-term culture initiating cells; TNF = tumour necrosis factor.

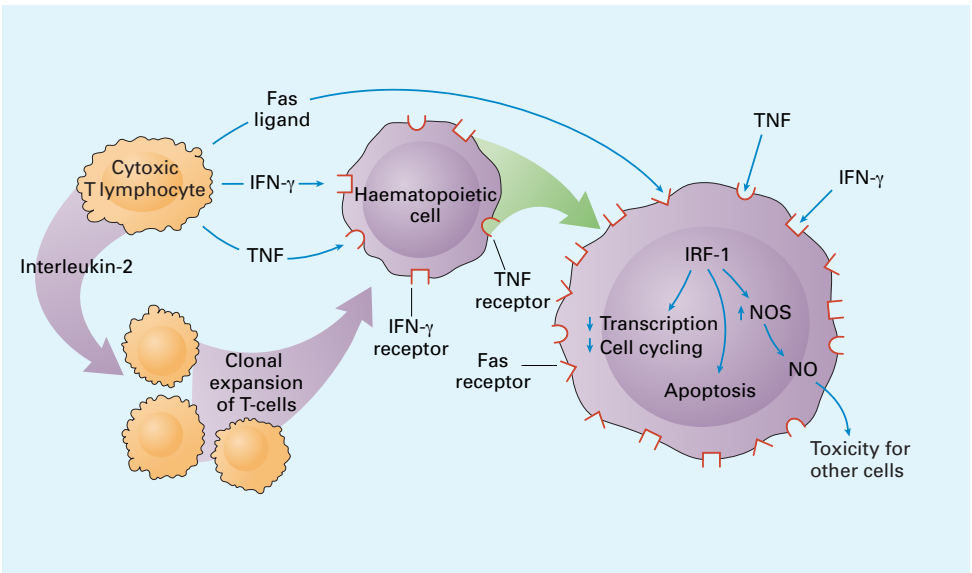


**Fig 2. Immune-mediated aplastic anaemia (AA).** In AA, activated cytotoxic T cells secrete cytokines such as tumour necrosis factor (TNF)-α and interferon (IFN)-γ which inhibit haemopoietic progenitor cells (HPC). TNF-α and IFN-γ upregulate the expression of the Fas receptor on HPCs, triggering apoptosis. Increased production of interleukin-2 results in expansion of T cells. TNF-α and IFN-γ also increase nitric oxide synthase (NOS) and nitric oxide (NO) production by marrow cells, which may contribute to immune-mediated cytotoxicity and elimination of HPCs[5] (IRF-1 = IFN regulatory factor-1) (reproduced, with permission, from Ref 7).

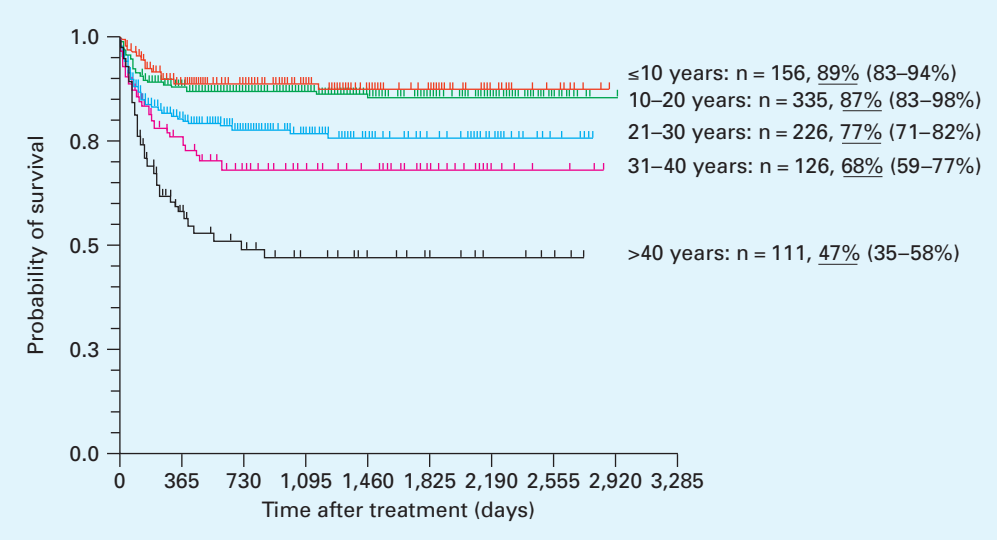


**Fig 3. Actuarial survival for patients undergoing HLA identical sibling bone marrow transplantation for severe aplastic anaemia according to age.**[10]

## Pure red cell aplasia

### *Definition, mechanisms and aetiology*

There is defective maturation of red cell precursors in pure red cell aplasia (PRCA), with failure to produce reticulocytes and hence mature red cells. The block in maturation may occur at the level of early (burst-forming unit (BFU-E)) or late (colony-forming unit (CFU-E)) erythroid progenitors, resulting in an absence of erythroid precursors in the bone marrow, or at the early normoblast stage producing a maturation arrest in the bone marrow with absence of late normoblasts.

It is important to distinguish acquired PRCA from the rarer congenital form, Diamond Blackfan anaemia (DBA), in young patients. DBA is often associated with short stature and skeletal anomalies, typically the red cell adenine deaminase level is elevated and mutations in the ribosomal protein S19 (RPS19) gene are found in 25% of affected individuals. A second DBA gene has been located on chromosome 8.[1]

Many cases of acquired PRCA are idiopathic and immune in origin (Table 3). PRCA often occurs in association with other systemic autoimmune disorders. It is associated with a thymoma in 10–15% of cases, 50% of whom will respond to thymectomy. Some cases are drug-induced.[13] Severe PRCA due to recombinant human erythropoietin (EPO) has recently been reported, in most cases with epoetin-alpha (Eprex®) when given subcutaneously in chronic kidney disease. This is associated with anti-EPO antibodies and appears to have been due to changes in the formulation of the drug.[14]

**Table 3. Conditions associated with acquired pure red cell aplasia (PRCA).**

| Aetiology | Mechanisms |
|---|---|
| Idiopathic | Immune-mediated |
| SLE, RA, Sjögren's disease | Immune-mediated |
| Thymoma (sometimes with myasthenia gravis and/or hypo-γ-globulinaemia) | Immune-mediated |
| Drugs: phenytoin, isoniazid, azathioprine | Marrow toxicity, drug-dependent antibodies |
| EPO in chronic kidney disease | Anti-EPO antibodies |
| B cell lymphoproliferative disorders (eg CLL, lymphoma) | Immune-mediated |
| Parvovirus B19 infection<br>CMV, EBV rarely | Lysis of late erythroid progenitors (CFU-E) with parvovirus infection |
| Myelodysplastic syndrome | Mechanism of PRCA uncertain:<br>? extreme apoptosis of red cell progenitors |

CFU = colony forming unit; CLL = chronic lymphocytic leukaemia; CMV = cytomegalovirus; EBV = Epstein-Barr virus; EPO = erythropoietin; RA = rheumatoid arthritis; SLE = systemic lupus erythematosus.

### *Investigations*

The following investigations should be performed:

- full blood count and blood film (normocytic, normochromic anaemia)
- reticulocyte count (severe reticulocytopenia)
- bone marrow aspirate and trephine biopsy (absent red cell precursors or maturation arrest (see above); examine for a secondary cause of PRCA such as lymphoma, chronic lymphocytic leukaemia, MDS; immunophenotyping and gene rearrangement studies for heavy chain and T cell receptor monoclonal expansion)
- chest X-ray and computed tomography scan of thorax (to exclude thymoma)
- autoimmune profile
- parvovirus B19, cytomegalovirus and Epstein-Barr virus serology.

### *Treatment*

Blood transfusions are required until recovery occurs. Thymectomy should be considered in patients with a thymoma and intravenous immunoglobulin is indicated for parvovirus-induced PRCA. For the remaining patients, immunosuppressive therapy is given, initially prednisolone starting at 1 mg/kg/day for four weeks, to which 50% of patients will respond. For non-responders, ciclosporin or azathioprine are often used. Monoclonal antibody therapy with anti-CD20 (rituximab) and anti-CD52 (alemtuzumab) is being evaluated for refractory cases.[15]

## Amegakaryocytic thrombocytopenia

Acquired amegakaryocytic thrombocytopenia is a rare cause of severe thrombocytopenia and bleeding. Megakaryocytes are absent from the bone marrow, in contrast to immune thrombocytopenia where megakaryocyte numbers are normal or increased. Patients may later develop aplastic anaemia or MDS. Regular platelet transfusions are required; some patients respond to ATG and ciclosporin. The underlying defect in most cases of congenital amegakaryocytic thrombocytopenia is due to a mutation in the thrombopoietin receptor (c-mpl).[1] Thrombocytopenia with absent radii is another example of congenital thrombocytopenia associated with absent or reduced megakaryocytes, but the gene defect in unknown.[16]

## Severe congenital neutropenia and cyclic neutropenia

Severe congenital neutropenia (SCN) is a rare inherited or sporadic form of marrow failure, with maturation arrest at the promyelocyte-myelocyte stage in the bone marrow. It presents in early infancy with severe neutropenia and recurrent bacterial infections. The autosomal recessive form is known as Kostmann's syndrome. Mutation in the neutrophil elastase gene (ELA2) occurs in many affected children and in all patients with cyclic neutropenia – a stem cell disorder resulting in periodic neutropenia, often severe, usually occurring every 21 days. Severe infections may occur during the neutrophil nadir. Most patients with cyclic neutropenia and SCN respond to granulocyte-colony stimulating factor.[17]

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