Investigation of patients with polycythaemia

TC Pearson, M Messinezy

Summary

Polycythaemia may complicate or be the presenting feature of a wide variety of different pathologies. Early diagnosis and treatment of primary polycythaemia will significantly reduce the morbidity and mortality associated with this condition. Patients with a raised packed cell volume are divided into those with a raised red cell mass (absolute polycythaemia), and those with a red cell mass within their normal range (apparent polycythaemia). A standard investigative approach of an absolute polycythaemia enables patients with primary and secondary polycythaemia to be identified, leaving a group termed idiopathic erythrocytosis. There are a number of physiological situations and pathological events associated with idiopathic erythrocytosis and apparent polycythaemia. Careful follow-up of both groups of these patients is essential to identify possible causative mechanisms.

Keywords: polycythaemia, clinical guidelines

Packed cell volume
A sustained value for males > 0.51, for females > 0.48 = some form of polycythaemia

Box 1

Division of Haematology, United Medical and Dental Schools of Guy's and St Thomas's Hospitals, St Thomas' Hospital, Lambeth Palace Road, London SE1 7EH, UK
TC Pearson
M Messinezy

Accepted 12 December 1995

Polycythaemia, defined as an increased packed cell volume (haematocrit), is known to be caused by a variety of pathological events and factors. In its primary form, up to 50% of patients present with vascular occlusive signs or symptoms¹ and untreated patients have a median survival of only 18 months,² the majority dying of vascular occlusion. However, adequately treated patients have a median survival of 10–13 years.³ Secondary polycythaemia may be the presenting feature of many, often treatable, pathological states or complicate the management of known pathology. From these observations, it is clear that the early recognition and an understanding of the causes of polycythaemia is relevant to improved patient care and to doctors in both primary care and hospital settings.

Packed cell volume

In normal adults, venous haemoglobin and packed cell volume values show a small diurnal variation, but are relatively stable for any individual. Thus, the oxygen-carrying capacity of the blood is maintained at a fairly constant level. The rate of red cell production is dominantly controlled by a feedback loop consisting of erythropoietin secretion by renal cells, at a level governed by the oxygen delivery to these cells. The mechanisms controlling plasma volume are more complex and include secretion of renin, aldosterone, catecholamines, atrial natriuretic peptide, and antidiuretic hormone. Venous packed cell volume is determined by the volume of circulating red cells, their distribution in the various parts of the circulation, and the plasma volume.

When considering a diagnosis of polycythaemia, it is better to use the packed cell volume than the haemoglobin value. The reason for this is that there is a closer correlation between red cell volume or mass and packed cell volume value than between haemoglobin concentration and red cell mass. Also, some patients with polycythaemia can have iron-deficient red cell indices. In this situation, the Hb concentration is disproportionately lower than the packed cell volume and a diagnosis of polycythaemia may be overlooked if only the haemoglobin value is evaluated. It is important to collect venous blood samples under standard conditions, particularly avoiding significant venous stasis, which causes haemoconcentration. Individuals should have a minimum of two results on blood samples collected on separate occasions before stating that their packed cell volume value is raised. While packed cell volume can be accurately determined by electronic cell counters, it must be appreciated that some, notably those manufactured by Coulter Electronics Limited, underestimate the true packed cell volume when there are iron-deficient red cell indices.⁴ For example, at a mean corpuscular haemoglobin of 20 pg, the packed cell volume is underestimated by 8–10%. In these circumstances, a correction must be applied or the packed cell volume measured by the micro-haematocrit method. The normal range of packed cell volume for men extends up to 0.51 and for women up to 0.48. These values represent two standard deviations above the mean. By definition, all patients with a raised packed cell volume have some form of polycythaemia. These patients are further investigated by measurement of their red cell mass and plasma volume.

Red cell mass and plasma volume

There are standard isotope dilution methods for measuring red cell mass and plasma volume. Although strictly only red cell mass is required to evaluate patients further, the additional measurement of plasma volume takes very little extra time and effort and helps in the interpretation of the results and subclassification of patients. Traditionally, normal values and patients' results for red cell mass and plasma volume were expressed in terms of their total body weight (ml/kg). It has been recognised for a long time that this leads to erroneous interpretation in obese patients. Fat tissue is relatively avascular

Red cell mass

- >25% above mean normal predicted value
= absolute polycythaemia
- <25% above mean normal predicted value
= apparent polycythaemia

Box 2

Box 3

compared with lean tissue and normal values expressed in ml/kg are lower in obese than lean subjects. Various formulae using both height and weight or lean body mass have been proposed to obviate the problem of obesity. The Radionuclide Panel of the International Council for Standardization in Haematology have recently published their recommendations based on an analysis of published data on red cell mass and plasma volume values in normal individuals⁵ and have proposed a formula for the derivation of mean normal values based on surface area, derived from height and weight. Using this formula, the normal ranges for male and female red cell mass are 25% above and below the mean normal value. This includes 98% and 99% of the normal male and female populations, respectively. Thus, by comparing a patient's red cell mass with the normal range predicted for their surface area, patients can be divided into those with an absolute polycythaemia and those with apparent polycythaemia (box 2). The next part of the investigation is largely determined by which of these two groups the patient's red cell mass falls into.

Subclassification of absolute polycythaemia

The aim of further investigation is to place the patient in one of three diagnostic groups: primary polycythaemia (also termed primary proliferative polycythaemia, polycythaemia rubra vera or polycythaemia vera), secondary polycythaemia, or idiopathic erythrocytosis (box 3). Primary polycythaemia is a chronic clonal myeloproliferative disorder arising from a 'malignant' pluripotent stem cell. Secondary polycythaemia simply denotes that the absolute polycythaemia arises from some demonstrable pathology and the marrow response is a secondary phenomenon. Idiopathic erythrocytosis is used for patients who, at the time of initial investigation, cannot be defined as either primary or secondary polycythaemia.

Investigation of absolute polycythaemia

While rather complicated algorithms have been proposed to separate primary from secondary polycythaemia, a standard approach to investigation of all patients makes certain that key investigations are performed. The starting point is a knowledge of the causes of secondary polycythaemia (box 4) and of the diagnostic criteria of primary polycythaemia (box 5).

Causes of secondary polycythaemia

- hypoxaemia (arterial oxygen saturation <92%), eg, chronic obstructive airways disease, cyanotic congenital heart disease, high altitude
- renal ischaemia or pathology, eg, polycystic kidneys, hydronephrosis, renal artery stenosis, post-renal transplantation
- erythropoietin-secreting tumours, eg, hypernephroma, hepatoma, fibroids, cerebellar haemangioblastoma, lung cancer
- familial polycythaemia, high oxygen affinity haemoglobin, autonomous high erythropoietin secretion, truncated erythropoietin receptor

Box 4

Diagnostic criteria of primary polycythaemia

A1 Absolute polycythaemia

A2 Absence of cause of secondary polycythaemia

A3 Palpable splenomegaly

A4 Clonality marker, eg, abnormal marrow karyotype

B1 Raised platelet count (>400 × 10⁹/l)

B2 Raised neutrophil count (>10 × 10⁹/l)

B3 Splenomegaly demonstrated by ultrasound or isotope scan (when spleen is palpable)

B4 Reduced serum erythropoietin or characteristic BFU-E culture results

A1+A2+A3 or A4 = primary polycythaemia

A1+A2+any two of B criteria = primary polycythaemia

Box 5

Figure 1 Microvascular occlusion due to an elevated platelet count in primary polycythaemia presenting with areas of purplish discolouration of the toes. The peripheral pulses were present

Figure 2 Embolic lesions in toes of patient with primary polycythaemia and extensive peripheral vascular disease of the lower extremities. The peripheral pulses were absent

Investigations in patients with absolute polycythaemia	
Stage 1	Stage 2
full blood count	bone marrow
ferritin	marrow
vitamin B ₁₂	karyotype
folate	serum erythro-
urea, creatinine	poietin
liver function	BFU-E culture
tests	oxygen disso-
urate	ciation curve
arterial oxygen	sleep study
saturation	lung function
COHb	tests
abdominal	chest X-ray
ultrasound	echocardiogram

Box 6

HISTORY AND CLINICAL EXAMINATION

Apart from taking a careful systematic history, attention should be focused on possible causes of secondary polycythaemia, features of primary polycythaemia and risk factors in the development of vascular occlusion. In particular, a history of renal disease and symptoms of lung pathology should be sought. A family history of polycythaemia should be sought, particularly in young patients. Some symptoms are more often associated with primary than secondary polycythaemia, although these should only be used as a general guide and do not replace detailed investigation. Often the patient is elderly and the possibility of dual pathology should always be remembered. Vascular occlusive lesions and transient cerebral ischaemia are more common in primary than secondary polycythaemia. Microvascular occlusion, typically involving the toes (figures 1 and 2), aquagenic pruritis and haemorrhagic manifestations particularly occur in primary polycythaemia. Gout is more common in primary than secondary polycythaemia.

LABORATORY AND CLINICAL INVESTIGATION

A list of tests are given in box 6. They are presented in two stages. Selected tests of those listed in stage 2 are undertaken following an evaluation of the results of stage 1 investigations, which should be performed in all patients.

Stage 1

Full blood count The red cell indices should be examined for evidence of iron deficient changes (severe iron deficiency may mask polycythaemia). A raised granulocyte count (typically neutrophilia but occasionally with basophilia and/or eosinophilia) and raised platelet count would support primary polycythaemia. However, in some ‘reactive’ states, notably infection, chronic inflammation and neoplasia a granulocytosis and/or thrombocytosis may occur. In addition, smokers have been shown to have higher neutrophil counts than non-smokers. In the heaviest smokers, it would be appropriate to modify the relevant minor criterion for primary polycythaemia to neutrophils greater than $12 \times 10^9/l$.

Serum ferritin, vitamin B₁₂ and folate Low serum ferritin levels are more common in primary than secondary polycythaemia. Vitamin B₁₂ levels may be elevated in primary polycythaemia due to transcobalamin release from an increased granulocyte mass. Folate deficiency has been reported in primary polycythaemia. These tests are not diagnostic in terms of the polycythaemia but provide useful information relating to these essential components of erythropoiesis.

Urea and creatinine Mild renal impairment has been associated with secondary polycythaemia⁷ but the red cell mass is usually only marginally above the normal range. Generally, with advanced renal impairment, the red cell mass falls. Polycystic renal disease may produce hypertension, impaired renal function and a significantly raised packed cell volume and red cell mass. Renal ultrasound (see below) is an important investigation to demonstrate renal lesions that may be responsible for secondary polycythaemia.

Liver function tests Cirrhosis and inflammatory liver disease have been associated with secondary polycythaemia and increased red cell production, respectively.

Arterial oxygen saturation (SaO₂) The measurement of arterial oxygen saturation is most easily accomplished by the use of a pulse oximeter. However, the values given by these oximeters include carboxyhaemoglobin (COHb). Thus, COHb levels should be measured simultaneously and subtracted from the oximeter reading to give the true oxygen saturation. Formal arterial blood gas analysis should be reserved for those with low SaO₂ values. The critical value for SaO₂ which implicates it as a cause of secondary polycythaemia is equal to or below 92%. However, the SaO₂ is not a static figure and secondary polycythaemia may be caused by intermittent desaturation events if these are sufficiently prolonged. It is important to enquire about typical symptoms of nocturnal oxygen desaturation such as a history of snoring, nocturnal restlessness, nocturia, waking unrefreshed and daytime somnolence. This is more commonly found in obese patients. Nocturnal reduction in SaO₂ below 92% with normal daytime values have been shown to explain the cause of polycythaemia in 10–20% of patients who would otherwise have been classified as idiopathic erythrocytosis.⁸ A useful screen for this possibility is to measure the SaO₂ by pulse oximetry when the patient has been in the supine position for 5–10 minutes.

Figure 3 Renal arteriogram showing a highly vascular hypernephroma. The patient had presented with a retinal artery occlusion presumably due to his high packed cell volume. An absolute polycythaemia was demonstrated

COHb COHb interferes with oxygen carriage and delivery. Smokers normally have packed cell volume values ± 0.02 above nonsmokers with COHb levels of 3–5% (normally less than 2%). However, some heavy smokers have higher COHb values and secondary polycythaemia can result,⁹ although this usually occurs when additionally there is chronic lung disease with some reduction in SaO_2 , which provides a further stimulus to red cell production.

Abdominal ultrasound This is an essential investigation in all patients with an absolute polycythaemia except when the cause is obvious. Careful examination of the kidneys is obviously important. A renal arteriogram should be performed if there is any suggestion of pathology on ultrasound examination (figure 3). Difficulty can arise when simple renal cysts are demonstrated. These become increasingly common with advancing age.¹⁰ It is exceedingly uncommon for simple cysts to be the cause of an absolute polycythaemia. Abdominal ultrasound can also establish the presence of relevant liver pathology and splenic size. While palpable splenic enlargement is a major criterion for the diagnosis of primary polycythaemia, enlargement demonstrated only on ultrasound examination is taken as a minor criterion (box 4). The reason for this is that the range of normal spleen size for a population of individuals of varying dimensions and age has not usually been firmly established. Finally, although large fibroids have been shown to cause secondary polycythaemia, the association is very rarely found.

Stage 2

The following tests may be useful where the diagnosis is still uncertain after stage 1 investigations. Some are not widely available or are still in the process of development and evaluation.

Bone marrow examination and karyotype Typically, the marrow is hypercellular with erythroid, granulocytic and megakaryocytic hyperplasia in primary polycythaemia. However, in difficult diagnostic situations marrow morphology is often rather unhelpful. Examination of the marrow chromosomes can occasionally be useful (box 5). At presentation, approximately 10–15% of primary polycythaemia patients have an abnormal karyotype,¹¹ although no specific diagnostic change has been demonstrated. The absence of an abnormal karyotype does not therefore exclude the condition.

Serum erythropoietin The results of serum erythropoietin estimation are not totally discriminatory for either primary polycythaemia or the various causes of secondary polycythaemia. However, the measurement can be helpful in the evaluation of an uncertain diagnosis. About two-thirds of patients with primary polycythaemia have a reduced serum erythropoietin value, and results for the remaining patients fall in the low normal range.¹² In secondary polycythaemia, erythropoietin values are in the normal range or raised. High values are typically found in arterial hypoxaemia. In secondary polycythaemia of renal origin, however, values often fall in the normal range. A few families have been described where some members have permanently raised serum erythropoietin levels without any underlying cause being found (autonomous high erythropoietin; box 3).

Burst-forming units–erythroid (BFU-E) culture Culture of peripheral blood mononuclear cells given the right culture conditions yields BFU-E in all individuals. BFU-E growth in samples from patients with primary polycythaemia shows an increased sensitivity to a variety of haemopoietic growth factors. Growth of BFU-E in the absence of erythropoietin in serum-containing media,¹³ or characteristic response of BFU-E to haemopoietic growth modifiers¹⁴ have been used to distinguish between primary and secondary polycythaemia (box 5).

Oxygen dissociation curve Measurement of the oxygen dissociation curve of haemoglobin and determination of the P_{50} value is essential in all patients with unexplained polycythaemia. Although only rarely encountered, over 30 haemoglobins, typically β -globin chain variants, with high oxygen affinity and 'left-shifted' dissociation curves have been described. Despite normal arterial oxygen saturations in these patients, the delivery of oxygen to the renal sensor is reduced. This causes an increased erythropoietin secretion and red cell production.

Sleep study, lung function tests, chest X-ray, echocardiogram These investigations should be performed in patients with a history suggesting nocturnal oxygen desaturation or relevant lung or cardiac pathology.

X-linked chromosome probes These investigations are by no means routine but are currently under investigation in research laboratories. They offer promise as a definitive method of recognising the clonal proliferation of primary polycythaemia (box 5). For this reason an outline of these studies is given below.

Women have only one active X-chromosome in each cell, randomly either the maternal or paternal X-chromosome. The classical observation was made that women with primary polycythaemia, who were heterozygotes for two recognisably different glucose-6-phosphate dehydrogenase enzymes, showed the presence of only one of the two enzymes in all lineages of their peripheral blood cells.¹⁵ This is the basis for defining primary polycythaemia as a clonal proliferation (arising from one ‘malignant’ pluripotent cell). In the last few years, a number of X-linked probes have been used in an attempt to determine whether a clonal proliferation is present or not. Obviously the technique can only be used in women. The basis of the technique is to use restriction enzymes in two stages. The first stage leads to the recognition of maternal and paternal strands of DNA from the two X-chromosomes by Southern blotting. Only a proportion of women, depending on the probe used, show the so-called ‘heterozygous pattern’, which is essential for further useful analysis. In the second stage, a restriction enzyme is used which cleaves further only the inactive X-chromosome strand. In normal women, generally half of each of the X-chromosomes (either maternal or paternal) is active and both of the two X-chromosome sequences are further digested. In women with primary polycythaemia, only one of the X-chromosomes is active and only one of the two bands, seen after the first stage, is further digested. Apart from any technical considerations, there are a number of problems with this approach in establishing clonality. Approximately 12% of normal women show extreme lyonisation with one or other X-chromosome dominantly active and different tissues in normal women show different patterns of X-inactivation.¹⁶ Thus, it has been proposed that a pure population of granulocytes should be examined taking the patient’s T lymphocytes as a control, on the assumption that T lymphocytes do not form part of the abnormal clone in primary polycythaemia.¹⁷ Only when the granulocytes show a ‘monoclonal’ pattern and T lymphocytes show a ‘polyclonal’ pattern can one conclude that there is a clonal proliferation of granulocytes and hence primary polycythaemia. Additionally, in the early stages of the disease, it is likely that circulating granulocytes are derived partly from normal stem cells as well as from the clonal proliferation. Thus, a polyclonal pattern of granulocytes is not necessarily against a diagnosis of primary polycythaemia.

Summary of investigations of an absolute polycythaemia

Following detailed investigation as given, approximately 80% of patients can be defined as primary polycythaemia or secondary polycythaemia of known cause. However, there remain a significant proportion of patients who cannot be assigned to either of these groups. These patients constitute the idiopathic erythrocytosis group.

Idiopathic erythrocytosis

Undoubtedly, this is a heterogenous group of patients.¹⁸ The possible causes are listed in box 7. Based on the selection of the normal red cell mass range, an occasional normal individual physiological variant will have a value more than 25% above the normal range. Transformation into primary polycythaemia, with other demonstrable features of the myeloproliferative disease, occurs in about 10% over many years.¹⁹ The recognition of some causes of secondary polycythaemia can be difficult, particularly intermittent hypoxaemia and small renal tumours. In addition, new causes of secondary polycythaemia will be established in future. For example, a large Finnish family containing many individuals with polycythaemia, has recently been described.²⁰ The cause has been established as a defective gene for the erythropoietin receptor leading to a shortened cytoplasmic portion of the receptor (box 4) which is responsible for switching off the receptor’s signal. As a result in these individuals, there is enhanced erythropoietin activity, despite reduced serum erythropoietin levels, and their BFU-E are hypersensitive to erythropoietin, features previously only associated with primary polycythaemia (box 5). Finally, it is possible that a clonal proliferative process showing just enhanced erythropoietic activity might exist.

Follow-up and judicious re-investigation of patients with idiopathic erythrocytosis is essential in case features of either primary polycythaemia or a possible cause of secondary polycythaemia emerge with time.

**Idiopathic erythrocytosis:
possible nature**

- physiological variant
- early primary polycythaemia
- underlying cause of secondary polycythaemia unrecognised
- clonal proliferation predominantly showing erythropoietic expansion

Box 7

Apparent polycythaemia:
alternative terms

- relative polycythaemia
- stress polycythaemia
- spurious polycythaemia
- Geisbock's syndrome
- pseudo-polycythaemia

Box 8

Apparent polycythaemia:
possible causes

- physiological variant
- early stage of development of absolute polycythaemia
- obesity
- fluid loss and diuretics
- smoking
- hypertension
- alcohol
- arterial oxygen desaturation
- renal disease
- pheochromocytoma

Box 9

Nature, pathogenesis and investigation of apparent polycythaemia

By definition, these patients have raised packed cell volume values but their red cell mass is within the normal range. A variety of terms has been used for this group (box 8). About one-third have a reduced plasma volume below the normal range but the remainder generally show an increase in red cell mass and reduction in plasma volume within their normal ranges. The possibilities in these patients include a normal physiological variation, an early absolute polycythaemia (that is, the red cell mass has increased in the individual patient but not outside the wide normal range) and various other possible underlying causes (box 9).^{21,22} Many of these factors increase the red cell mass and reduce the plasma volume independently and it is the combination of these two changes that cause apparent polycythaemia. Perhaps surprisingly arterial hypoxaemia appears in this list. In a small percentage of these patients, the hypoxaemia produces a reduction in plasma volume and any increase in red cell mass is within the normal range.

As far as investigation is concerned, it is important to exclude renal disease and arterial hypoxaemia. This necessitates a renal ultrasound, SaO₂ and a sleep study, when there are any symptoms suggesting nocturnal arterial oxygen desaturation. All patients should be followed-up to exclude the development of primary polycythaemia or a cause of secondary polycythaemia.

It must be appreciated that, increasingly, patients are investigated at an earlier stage of the development of pathology. It is therefore essential that neither idiopathic erythrocytosis nor apparent polycythaemia is considered to be the final diagnosis but only a descriptive title for two groups of patients, where the mechanism of the raised red cell mass in idiopathic erythrocytosis and of the raised packed cell volume in apparent polycythaemia has not been established.

1 Barabas AP, Offen DN, Meinhard EA. The arterial complications of polycythaemia vera. *Br J Surg* 1973; **60**: 183–7.

2 Chievitz E, Thiede T. Complications and causes of death in polycythaemia. *Acta Med Scand* 1962; **172**: 513–23.

3 Berk PD, Goldberg JD, Donovan PB, Fruchtman SM, Berlin NI, Wasserman LR. Therapeutic recommendations in polycythemia vera based on polycythemia vera study group protocols. *Semin Hematol* 1986; **23**: 132–43.

4 Guthrie DL, Pearson TC. PCV measurement in the management of polycythaemic patients. *Clin Lab Haematol* 1982; **4**: 257–65.

5 Pearson TC, Guthrie DL, Simpson J, et al. Red cell mass and plasma volume interpretation in normal adults. *Br J Haematol* 1995; **89**: 748–56.

6 Du Bois D, Du Bois EF. Clinical calorimetry. A formula to estimate the approximate surface area if height and weight be known. *Arch Intern Med* 1916; **17**: 863.

7 Hoppin EC, Depner T, Yamuchi H, Hopper J. Erythrocytosis associated with diffuse parenchymal lesions of the kidney. *Br J Haematol* 1976; **32**: 557–63.

8 Moore-Gillon JC, Treacher DF, Gaminara EJ, Pearson TC, Cameron IC. Intermittent hypoxia in patients with unexplained polycythaemia. *BMJ* 1986; **293**: 588–90.

9 Smith JR, Landaw SA. Smokers' polycythemia. *N Engl J Med* 1978; **298**: 6–10.

10 Laucks SP, McLachlan MSF. Aging and simple cysts of the kidney. *Br J Radiol* 1981; **54**: 12–4.

11 Diez Martin JL, Graham DL, Pettitt RM, Dewald GW. Chromosome studies in 104 patients with polycythemia vera. *Mayo Clin Proc* 1991; **66**: 287–99.

12 Messinezy M, Westwood NB, Woodcock SP, Pearson TC. Low serum erythropoietin – a strong diagnostic criterion of primary polycythaemia even at normal haemoglobin levels. *Clin Lab Haematol* 1995; **17**: 217–20.

13 Reid CDL. The significance of endogenous erythroid colonies (EEC) in haematological disorders. *Blood Rev* 1987; **1**: 133–40.

14 Dudley JM, Westwood N, Leonard S, Eridani S, Pearson TC. Primary polycythaemia: positive diagnosis using the differential response of primitive and mature erythroid progenitors to erythropoietin, interleukin 3 and alpha-interferon. *Br J Haematol* 1990; **75**: 188–94.

15 Adamson JW, Fialkow PJ, Murphy S, Prchal JF, Steinmann L. Polycythemia vera: stem-cell and probable clonal origin of the disease. *N Engl J Med* 1976; **295**: 913–6.

16 Gale RE, Wheadon H, Boulous P, Linch DC. Tissue specificity of X-chromosome inactivation patterns. *Blood* 1994; **83**: 2899–905.

17 Gale RE, Wheadon H, Boulous P, Linch DC. Tissue specificity of X-chromosome inactivation patterns. *Br J Haematol* 1993; **84** (suppl 1): 59.

18 Messinezy M, Sawyer B, Westwood NB, Pearson TC. Idiopathic erythrocytosis – additional new study techniques suggest a heterogenous group. *Eur J Haematol* 1994; **53**: 163–7.

19 Pearson TC, Wetherley-Mein G. The course and complications of idiopathic erythrocytosis. *Clin Lab Haematol* 1979; **1**: 189–96.

20 de la Chapelle A, Träskelin A-L, Juvonen E. Truncated erythropoietin receptor causes dominantly inherited benign human erythrocytosis. *Proc Natl Acad Sci US* 1993; **90**: 95–9.

21 Messinezy M, Pearson TC. A retrospective study of apparent and relative polycythaemia: associated factors and early outcome. *Clin Lab Haematol* 1990; **12**: 121–9.

22 Pearson TC. Apparent polycythaemia. *Blood Rev* 1991; **5**: 205–13.