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REVIEW

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ABSTRACT

Anaemia is common in older individuals and, because of its association with various negative outcomes, adequate diagnosis and treatment is important. The present review focuses on prominent factors included in diagnostic and therapeutic algorithms for anaemia.

Although pernicious anaemia is associated with severe vitamin B12 deficiency, evidence of an association between subnormal vitamin B12 and anaemia in older persons in the general population is limited and inconclusive. Accumulating evidence suggests that clinicians should at least reconsider the risks of a low vitamin B12 level before starting vitamin B12 supplementation in older individuals. Although clinicians may be reluctant to measure ferritin in older individuals due to its acute phase properties, such measurements are important in older persons with anaemia, especially in those with signs of inflammation. While a severe age-related decline in renal function may lead to a blunted erythropoietin response and anaemia, elevated erythropoietin levels are associated with increased mortality. More studies are needed to identify the clinical relevance and therapeutic implications of low and high erythropoietin levels in older persons. In contrast to other age-related diseases, telomere length is not associated with anaemia in older individuals in the general population.

In conclusion, many issues regarding the aetiology of anaemia in old age remain unresolved. Because current guidelines on anaemia are based on the classic notions of the aetiology of anaemia, they may need to be revised for the highest age groups.

KEYWORDS

Anaemia, mortality, ferritin, vitamin B12, folate, erythropoietin, myelodysplasia, telomere length, aged

INTRODUCTION

Anaemia is very common in older individuals. The reported prevalence ranges from <3% in healthy persons aged ≥65 years to 61% in older patients newly admitted to geriatric wards.[1,2] This wide variance can be due to various definitions of anaemia, and to large differences in study populations with respect to gender, age, race, living situation, and health status.[1,2] In the Third National Health and Nutrition Examination Survey (NHANES III), a nationally representative study of non-institutionalised civilian adults in the USA, the overall prevalence of anaemia among adults aged ≥65 years was 11.0% in men and 10.2% in women.[3] In that study, anaemia was defined according to World Health Organisation criteria (haemoglobin concentration ≤12 g/dl in women and ≤13 g/dl in men).[4] Interestingly, the prevalence of anaemia increased significantly with age, i.e. up to 26.1% in men and 20.1% in women aged 85 years and over.[3]

In older persons, anaemia is associated with impaired survival,[5-10] decreased physical performance, disability in daily living, cognitive impairment, depression, diminished quality of life, and with an increased number of hospital admissions.[1,11-21] Considering the steep increase in the prevalence of anaemia in older individuals, and the exponential rise in the number of older individuals in our ageing society, anaemia in older individuals may have a significant impact on healthcare needs and costs in the future.[22] Adequate diagnosis and treatment of anaemia in older persons is therefore of vital importance.

In clinical practice, older patients with anaemia are carefully examined to detect and treat the underlying cause of the anaemia. Treating physicians will enquire about recent blood loss, signs and symptoms from the digestive tract, nutritional habits, weight loss, and drugs and alcohol intake.[23] In most diagnostic laboratory algorithms for anaemia, the mean corpuscular volume (MCV) plays a central role.[23-25] In patients with microcytic anaemia

(MCV <80 fl), ferritin, iron and transferrin levels are measured to determine the presence of iron deficiency anaemia. Vitamin B12 and folate are measured in patients with macrocytic anaemia (MCV >100 fl) to determine or rule out the presence of vitamin B12 or folate deficiency. Normocytic anaemias (MCV 80-100 fl) are often caused by chronic diseases, malignancies or bone marrow conditions.[23-25]

Anaemia is a unique condition in the sense that diagnostic and therapeutic guidelines are based on assumed aetiology and pathophysiology. Interestingly, most studies on anaemia have been performed in selected patient groups (e.g. patients in hospital wards and residents in institutions for older persons) and not in very old persons from the general population. Increasingly, data have become available that question the extrapolation of 'common' medical knowledge to the highest age groups. For instance, the effects of some classical determinants of disease and mortality in middle age (e.g. hypothyroidism, hypertension and hypercholesterolaemia) have been shown to disappear or even reverse in the oldest old,[26-29] indicating that physiological processes in the oldest old may be distinct from those in younger individuals.

The present review focuses on some of the most prominent factors included in diagnostic and therapeutic algorithms for anaemia to assess whether these factors also apply for older persons in the general population.

AETIOLOGY OF ANAEMIA

Vitamin B12 deficiency

Pernicious anaemia is a form of anaemia that is undeniably associated with severe vitamin B12 deficiency. Finding the cure for pernicious anaemia in fact led to the discovery of vitamin B12.[30-36] Undoubtedly, patients with very low vitamin B12 concentrations (in case of pernicious anaemia) have to be treated. Patients with pernicious anaemia or food-vitamin B12 malabsorption show large increases in haemoglobin after vitamin B12 administration.[37-40]

The outcomes of studies in patients with pernicious anaemia are often extrapolated to patients with subnormal vitamin B12 concentrations in the general population. As a result, subnormal vitamin B12 concentrations are considered to be associated with (mild) anaemia in general, but also with other conditions such as dementia, neuropathy and subacute combined degeneration of the spinal cord.[41-44] Therefore, physicians routinely measure vitamin B12 in patients with anaemia. Individuals with low serum concentrations of vitamin B12 (and normal folate concentrations) are frequently given intramuscular vitamin B12 supplements, often for many years.[23,43,44] Also, since low serum vitamin B12 concentrations are very common in older individuals,[45] screening older people for vitamin B12 deficiency has often been recommended.[46,47] However, although the biological role of vitamin B12 in haematopoiesis is well defined,[44,48-50] current evidence suggests that the outcomes of these studies in patients with severe vitamin B12 deficiency should not be extrapolated to patients with subnormal vitamin B12 concentrations in the general population.

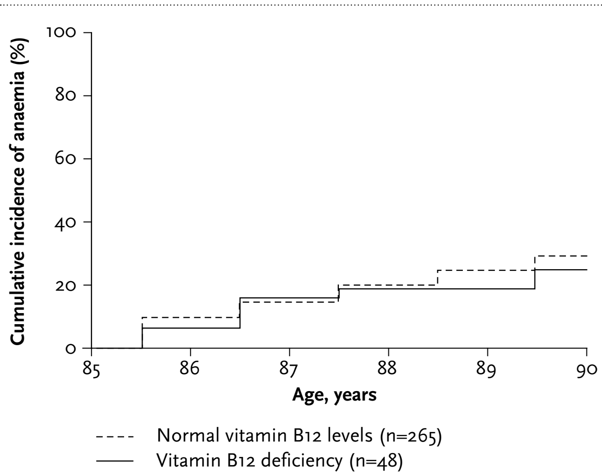
Results from the Leiden 85-plus Study

In the Leiden 85-plus Study, a population-based prospective follow-up study of 85-year-old individuals (living in Leiden, the Netherlands), we showed that low vitamin B12 concentrations (<150 pmol/l) in 85-year-old persons are not associated with the presence of anaemia at age 85 years.[51] Also, participants with low vitamin B12 concentrations did not have a higher risk to develop anaemia from age 85 onwards (*figure 1*). Adjustment for possible confounders did not change our results.[51]

Results of a systematic literature review

Interestingly, our study was not the first to cast doubt on the relationship between subnormal vitamin B12 concentrations and anaemia in older individuals. In a systematic review of the literature, we evaluated the association between subnormal vitamin B12 concentrations and anaemia in older people.[52] Twenty-two observational studies showed inconsistent results with regards to the association between subnormal vitamin B12 concentrations or vitamin B12 deficiency and anaemia in older subjects. Three randomised placebo-controlled trials (RCTs), with

Figure 1. *Effect of vitamin B12 deficiency (<150 pmol/l) on anaemia during follow-up in subjects without anaemia at age 85 years (n=313); hazard ratio 0.85; 95% confidence interval 0.43-1.65.[51] Reprinted with permission. Arch Intern Med 2008;168(20):2241. Copyright: American Medical Association. All rights reserved.*



a total of 210 participants, met the inclusion criteria for intervention studies for our review.[53-55] Due to clinical heterogeneity (differences in methods of administration, dose of vitamin B12, outcome measures and treatment follow-up time) we did not combine the results in a meta-analysis. However, the three RCTs (considered to be of methodologically high quality) showed no beneficial effect of vitamin B12 administration on haemoglobin concentrations, MCV, cognitive function and neurological symptoms.[53-55] Moreover, there was no treatment effect for participants who were anaemic.[54]

Clinical implications and implications for future research

Taking these findings into account, one may conclude that strong evidence is lacking for a positive association between subnormal vitamin B12 concentrations and anaemia in older persons in the general population. The above-mentioned findings do not imply that patients with pernicious anaemia or food-vitamin B12 malabsorption (with tissue depletion of vitamin B12 and very low vitamin B12 concentrations) should be withheld from vitamin B12 administration.[38,39] However, apart from the undisputed reality of pernicious anaemia, the clinical impact of a subnormal vitamin B12 concentration in older persons in the general population remains unclear. The fact that several observational studies and RCTs also showed no effect of vitamin B12 administration on cognitive function raises even more doubt about the consequences of subnormal vitamin B12 concentrations in older persons in the general population.[56-58]

Many older persons in primary care may receive vitamin B12 injections without evidence for clinical improvement. In addition, these findings raise doubt about the value of vitamin B12 measurement in diagnostic guidelines for anaemia as this may distract attention from other possible underlying causes. If a subnormal vitamin B12 concentration is not the cause of the anaemia, supplementation with vitamin B12 will not lead to a rise in haemoglobin concentration. Additional proof of the (lack of) effectiveness of vitamin B12 treatment in older patients with anaemia and subnormal vitamin B12 concentrations should come from a randomised double-blind placebo-controlled trial. However, before such a trial is performed, this accumulating evidence suggests that clinicians should at least reconsider the risks of a low vitamin B12 concentration before starting cyanocobalamin or hydroxocobalamin supplementation in older individuals.[59]

Interestingly, in contrast to vitamin B12, folate deficiency is still associated with anaemia in older individuals.[51] Early detection of folate deficiency by screening may identify older individuals at risk of developing anaemia. The biochemical pathways suggest that folic acid supplementation is beneficial, but it remains unclear whether folic acid fortification of grain and cereal products (as employed in the USA[60,61]) has a positive effect on the incidence of anaemia in older persons and should also be employed in the Netherlands.[62] This is a topic for future studies.

Iron deficiency and inflammation

Iron deficiency is a common cause of anaemia, being found in ≥15% of older persons with anaemia.[3,24,63] Serum ferritin levels strongly correlate with body iron stores[64,65] and are considered the best noninvasive test for the diagnosis of iron deficiency.[63,66,67] Therefore, ferritin plays a central role in diagnostic and therapeutic algorithms for iron-deficiency anaemia in clinical practice.[23-25]

Results of the Leiden 85-plus Study

Ferritin is also a well-known acute phase protein and may be elevated in acute and chronic inflammatory conditions, such as (respiratory tract) infections, rheumatoid arthritis and cancer.[63,68,69] In case of acute and chronic inflammatory conditions, serum ferritin may not accurately reflect true iron status.[63,68,69] Clinicians may be reluctant to measure ferritin in older individuals, especially in those with infections or inflammation; however, findings from the Leiden 85-plus Study suggest that ferritin measurements are important in these persons.[70] Low ferritin was associated with lower haemoglobin levels and lower MCV, but this association was more pronounced in participants with elevated C-reactive protein (CRP) levels than in subjects with normal CRP levels. It is hypothesised that low ferritin is such a specific marker of iron status in individuals with inflammation due to its 'acute phase' properties, i.e. iron status must be poor when low ferritin levels are found in the presence of inflammation.[70]

Potential role for hepcidin

It has been hypothesised that upregulation of hepcidin (the main regulator of iron homeostasis) plays an important role in the anaemia of inflammation.

An inflammatory stimulus activates monocytes and T cells to produce pro-inflammatory cytokines.[71] These cytokines, particularly interleukin 6, induce the production and secretion of hepcidin by hepatocytes.[72] Hepcidin binds to the membrane protein ferroportin, an iron efflux channel on the surface of absorptive enterocytes, macrophages and hepatocytes, and induces its internalisation and degradation in lysosomes, thereby blocking the export of iron from cells.[73] Consequently, duodenal enterocytes deliver less dietary iron to extracellular fluid, macrophages fail to release iron recycled from senescent erythrocytes and hepatocytes retain stored iron, leading to a rapid drop in iron levels,[74] iron-restricted erythropoiesis, and anaemia.[71,75,76] Moreover, transgenic mice overexpressing hepcidin and mice receiving synthetic hepcidin develop mild-to-moderate microcytic, hypochromic anaemia.[77-80]

As a result, hepcidin is considered to be the main mediator of anaemia of inflammation,[75,81,82] also known as anaemia of chronic disease, which is commonly found in patients with chronic infections or with inflammatory disorders, such as rheumatoid arthritis, inflammatory bowel disease, cancer and chronic kidney disease.[71,83,84] Although a preliminary analysis in the InChianti study (a population-based study of older persons in Tuscany, Italy) could not demonstrate higher urinary hepcidin levels in older individuals with anaemia of inflammation,[85] this hypothesis should still be tested in other population-based prospective follow-up studies, preferably using serum hepcidin assays which have recently become available.[86,87] Depending on the outcomes of these additional studies, future diagnostic algorithms for anaemia may incorporate markers of inflammation such as CRP or even hepcidin to discriminate between classic iron-deficiency anaemia (low hepcidin levels) and iron-deficiency anaemia in the context of anaemia of inflammation or chronic disease (elevated hepcidin levels).[87] The results of these studies may also lead to innovative clinical trials, for instance by treating older patients with anaemia of inflammation with anti-inflammatory agents or hepcidin antagonists such as agents that inhibit hepcidin production (e.g. anti-interleukin 6 receptor antibodies), hepcidin neutralising antibodies, targets against hepcidin binding site of ferroportin or agents that inhibit ferroportin internalisation.[83,88]

Erythropoietin

Renal function, erythropoietin and anaemia

Decreased oxygen availability in the kidney triggers the production of erythropoietin (the principal regulator of red blood cell mass) by the peritubular capillary lining cells within the kidney.[89] Impaired oxygen delivery to the kidney can result from various pathophysiological mechanisms, such as anaemia, hypoperfusion due to renal arteriosclerosis, lowered renal blood flow or heart failure, or decreased oxygen saturation due to diseases such as chronic obstructive pulmonary disease.[89-93] In the InChianti study, participants with a creatinine clearance of 30 ml/min or lower had significantly lower age and haemoglobin-adjusted endogenous erythropoietin levels than their counterparts with normal renal function.[94] Thus, severe age-related decline in renal function may lead to a blunted erythropoietin response and anaemia.[94] It is also known that erythropoietin substitution therapy is effective in raising haemoglobin levels and improving the quality of life in (pre) dialysis, cancer patients, and also in community-dwelling older persons with unexplained chronic anaemia.[95-99]

Erythropoietin and mortality

Interestingly, studies in chronic heart failure patients indicated that high erythropoietin is a predictor of impaired survival.[100-105] In addition, in the Leiden 85-plus Study, we also observed a dose-dependent positive association between increasing erythropoietin levels and mortality, independent of gender, creatinine clearance, haemoglobin level, comorbidity, smoking and C-reactive protein level.[106] It is not exactly clear why elevated erythropoietin levels mark excess mortality. Elevated erythropoietin levels could be a physiological response to a chronically increased hypoxic stimulus due to yet undiagnosed subclinical disease.[107,108] Elevated erythropoietin may also be compensating for removal of erythrocytes from the blood, either because of erythrocyte fragility, subclinical chronic haemolysis, or blood loss.[107,108] Further studies are needed to shed light on the mechanisms involved and to identify the clinical and therapeutic implications of a high erythropoietin level in old age, especially since a number of unexpected nonhaematopoietic functions of erythropoietin have recently been identified.[109] Our findings do not necessarily implicate that older individuals with renal failure, cancer or unexplained anaemia should not be treated with recombinant erythropoietin. However, recent meta-analyses of randomised trials showed that treatment with erythropoiesis-stimulating agents in patients with chronic kidney disease or cancer had a negative influence on survival,[110-112] which clearly emphasises the need for further studies on the aetiology and effects of high erythropoietin levels in older individuals.

Unexplained anaemia

In approximately one third of older patients with anaemia, the cause of the anaemia is unknown; their anaemia is 'unexplained'.[3] Since older subjects with unexplained anaemia often present with low leucocyte counts,[113,114] myelodysplastic syndromes or other types of bone marrow failure may be the underlying diagnosis for unexplained anaemia.[3,114-116]

Telomere length and anaemia

Telomeres are DNA-protein complexes at the ends of chromosomes. Telomeres are critical for chromosome stability and function, since they protect chromosome ends against fusion, degradation and recombination. In somatic and haematopoietic cells, telomeres shorten with every cell division as a result of the end-replication problem (i.e. the inability of the DNA replication machinery to replicate the lagging DNA strand after removal of the RNA primer) and oxidative damage.[117] Telomerase can preserve telomere length by adding *de novo* tandem repeats at chromosome ends, but its activity in somatic cells and haematopoietic progenitor cells is very low. Consequently, mean somatic cell and peripheral blood mononuclear cell telomere length shortens with age.[117] When telomere length falls below a critical level, replicative senescence (permanent growth arrest) is induced.[118,119]

Telomere length is considered a marker of biological and cellular ageing and has been correlated with a number of major age-related diseases such as dementia,[120-123] myocardial infarction,[124] heart failure,[125] atherosclerosis,[126] and solid tissue tumours.[127]

Myelodysplastic syndromes or other types of bone marrow failure are thought to explain the increased frequency of (unexplained) anaemia in older individuals.[3,115,116] Adult haematopoietic stem cells show a severe loss of telomeric DNA compared with cells from foetal liver or umbilical cord blood,[128] and aged mice have a decreased capacity to replace blood cells during haematopoietic stress compared with younger mice,[115,129] indicating a loss of replicative potential for bone marrow stem cells with age[128,130] and a possible incapacity to react to the physiological demand for blood cell replenishment with age.[115,116,129] Since earlier studies indicate that patients with myelodysplastic syndromes or other types of bone marrow failure syndromes have shortened telomeres,[131-133] shorter telomere length has been associated with an increased risk of anaemia in chronic heart failure patients[134] and was an independent predictor of lower red blood cell counts in a study of middle-aged subjects (aged 35-55 years),[135] telomere length may be a marker of haematopoietic ageing and bone marrow failure and, as a result, may be associated with anaemia in older individuals in the general population. Therefore, we investigated the relation between telomere length and the presence of anaemia (and unexplained anaemia in particular) in two population-based studies of individuals aged 85 years and over: the Newcastle 85-plus Study, and the Leiden 85-plus Study. In both cohorts, no difference was observed in telomere length between participants with anaemia and without anaemia, nor did telomere length correlate with any other haematological parameter.[136] Thus, in contrast to other age-related diseases, telomere length is not associated with anaemia or any other haematological parameter in older individuals in the general population, despite the plausible biological mechanism underlying this association. Our findings are supported by another study in which no correlation was found between telomere length and blood counts in a population-based sample of 717 women aged 38 to 100 (median 72) years.[137] To further investigate this intriguing matter, studies incorporating bone marrow biopsies are needed.

CONCLUDING REMARKS

Although researchers and clinicians have paid much attention to the clinical implications and pathophysiology of anaemia in older individuals, the consequences and underlying pathophysiological mechanisms of anaemia in the oldest old in the general population are still relatively unknown. However, it has become clear that, while folate deficiency at age 85 years is still associated with the development of anaemia during follow-up, this does not seem to be the case for vitamin B12 deficiency. Nowadays, many older subjects with subnormal vitamin B12 concentrations receive hydroxocobalamin treatment. Further trials are needed to verify whether older individuals with anaemia and subnormal vitamin B12 levels should be treated with hydroxocobalamin. Furthermore, in old age, low ferritin is associated with the presence of anaemia, particularly in older persons with elevated CRP levels, indicating that ferritin measurements are still important, especially in older persons with signs of inflammation. Serum hepcidin measurements may elucidate the complicated interrelation between iron deficiency, inflammation and anaemia. Additionally, severe age-related decline in renal function may lead to a blunted erythropoietin response and anaemia. Elevated erythropoietin levels are associated with increased mortality, independent of haemoglobin and other comorbidities. Additional studies are needed to identify the clinical relevance and therapeutic implications of a low and a high erythropoietin level in older people in the general population. Moreover, in contrast to other age-related diseases, telomere length is not associated with anaemia in older individuals in the general population, despite the plausible biological mechanism underlying this association.

Finally, future studies should focus on improving the diagnostic algorithms for anaemia in older individuals by examining the additional diagnostic value of erythropoietin, homocysteine, methylmalonic acid, CRP or hepcidin in these algorithms. Since the prevalence of anaemia is highest in the highest age groups, more studies are needed to elucidate the specific causes of anaemia in these age groups. As current diagnostic and therapeutic guidelines are based on the classic notions of the aetiology of anaemia, the guidelines on anaemia may have to be revisited for the highest age groups in the coming years.

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