Anaemia, inflammation, renal function, and the diabetic foot: What are the relationships?

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Article points

1. Anaemia and iron deficiency are common problems in patients with diabetic foot disease.
2. Anaemia and inflammation are associated with stage of diabetic foot disease.
3. Anaemia was found to be independent of renal function, which may be explained by an underlying inflammatory process.

Keywords
- Anaemia
- Diabetic foot disease
- Inflammatory state

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Diabetic foot disease has major implications for a patient's quality of life – from reduced mobility to potential loss of limb. Anaemia and inflammation have been shown to play a role in diabetic foot disease. Here, the authors report the results of a retrospective analysis of patients with diabetes treated at University College London Hospital, and the relationship between haemoglobin, C-reactive protein (CRP), creatinine levels and foot ulcer risk score. An inverse correlation was found between haemoglobin and CRP, and foot disease progression was linked to haemoglobin decline and CRP rise.

For patients with diabetes – many of whom already have an impaired quality of life – anaemia constitutes an unwelcome additional burden. Anaemia has been shown to impact significantly on quality of life leading to chronic fatigue and reduced energy levels (Thomas, 2007). Classically, anaemia in patients with diabetes is described as anaemia of chronic disease. It is the second most prevalent after anaemia caused by iron deficiency and commonly occurs in patients with acute or chronic immune activation, hence the condition has been termed 'anaemia of inflammation'. Several mechanisms have been proposed, including disturbances of iron haemostasis, impaired proliferation of erythroid progenitor cells and a blunted erythropoietin response to anaemia (Weiss and Goodnough, 2005).

Many efforts have been initiated to identify certain risk factors and their impact on development and persistence of diabetic foot disease, however, published work in this area is limited and incomplete. The authors set out to investigate anaemia, inflammation and renal function, and their elusive association with diabetic foot disease.

Aims

To undertake a retrospective study to assess the relationships between haemoglobin, C-reactive protein and creatinine in patients with varying levels of diabetic foot disease.

Methods

A multidisciplinary diabetic foot team coordinates diabetic foot care at the University College London Hospital (UCLH) NHS Foundation Trust. One-hundred-and-seventy-

five patients with diabetes who attended the clinic in 2010 were identified from the Trust database.

The cohort of 175 patients were subsequently stratified to four groups, according to NICE's (2011) *Diabetes in Adults: Quality Standards* Quality Statement 10 on foot risk*:

- D1 – normal sensation, palpable pulses (low risk)
- D2 – neuropathy, absent pulses, other risk factors (increased risk)
- D3 – neuropathy, absent pulses, deformity, skin changes, previous ulcer (high risk)
- D4 – ulcerated foot

In the present study, patients with Charcot foot, other foot deformities, gangrene, and any previous amputations were excluded. Patients with existing venous ulcers, those on oral antibiotics, and those awaiting revascularisation were also excluded.

Management of such patients involves a multidisciplinary approach by a team consisting of vascular surgeons, diabetologists, podiatrists, microbiologists, and nurse specialists. To adhere to health service targets, a large number of patients were treated at a different hospital if there was a delay in achieving timely specialist input. Therefore, only those who received all of their care at UCLH were included. Thus, patients on dialysis were not included as this patient group are managed at a different hospital.

Data collected included age, sex, presence of anaemia as defined by the World Health Organization (Blanc et al, 1968) (haemoglobin [Hb] <13 g/dL for men and <12 g/dL for women), elevated C-reactive protein (CRP) level defined as >1.0 mg/dL and renal function assessed by serum creatinine levels (µmol/L), which was 60–120 µmol/L. As this was a retrospective database, blood profiles of a number of patients were absent, as was staging of diabetic foot disease. Thus, only those patients who had documented evidence of all the above blood profiles and stage of diabetic foot disease were included in the study.

Ultimately, data on a total of 40 patients that fulfilled the criteria were analysed and found to fall evenly into each of the sub-groups (D1, D2, D3, D4). Two investigations of this cohort were conducted: (i) cross-sectional analysis of prevalence of anaemia, inflammation, and deteriorating renal function in patients with diabetic foot disease; and (ii) a longitudinal analysis of diabetic foot disease progression, D1 to D4, over a 6-month period.

Statistical analysis was performed using Prism for Windows version 4.0 (GraphPad Software, La Jolla, CA, USA).

Continuous data are expressed as mean ± the standard error of the mean (SEM). Differences in continuous variables were compared using one-way analysis of variance (ANOVA). Pearson correlation was used to analyse univariate associations between continuous variables.

Results

Of the 40 patients analysed (24 men, 16 women; mean age 65 ± 8 years). There was an association between stage of diabetic foot disease and anaemia at first assessment (*Figure 1*). Mean haemoglobin for each stage is shown in *Table 1*.

The authors found no association between renal function and diabetic foot disease stage. Mean creatinine was 77.2 ± 7.8 µmol/L, 75.1 ± 7.3 µmol/L, 94.0 ± 4.9 µmol/L and 86.0 ± 5.8 µmol/L in groups D1, D2, D3, and D4, respectively (P=0.2; *Figure 2*). There was no correlation between baseline creatinine and baseline haemoglobin levels (r= –0.21, P=0.2).

There was a positive relation between rising CRP levels with worsening diabetic foot disease. Mean CRP levels of 6.1 ± 1.9 mg/dL, 22.8 ± 2.6 mg/dL, 32.4 ± 6.2 mg/dL, 39.5 ± 4.9 mg/dL were found in groups D1, D2, D3, and D4, respectively (P<0.0001; *Figure 3*). A strong inverse correlation (r= –0.56, P<0.0001) was found between baseline CRP levels and haemoglobin levels. These data suggest that as CRP levels increase,

Please note: These four categories were defined by NICE (2011), but this document was revised in July 2012. Active ulceration is no longer included as a level of risk (see http://guidance.nice.org.uk/QS6)

haemoglobin levels decrease at presentation (*Figure 4*).

A subgroup of six patients (four men and two women; mean age 69 ± 14 years) progressed from D1 to D4, over the course of 6 months. Results were consistent with the cross-sectional analysis. Mean haemoglobin was calculated as 13.2 ± 0.2 g/dL, 12.3 ± 0.4 g/dL, 11.0 ± 0.2 g/dL, and 9.3 ± 0.2 g/dL for D1, D2, D3, and D4, respectively (P<0.0001; *Figure 5*). Mean CRP levels were also compared for each stage and are shown in *Figure 6* (P<0.0001).

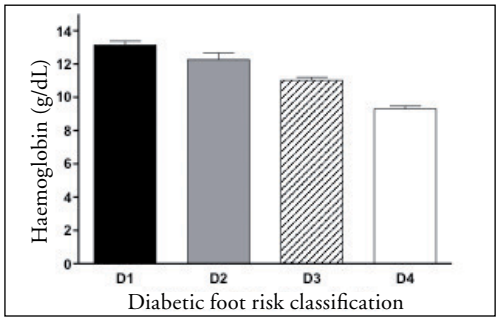

Figure 1. The relationship between haemoglobin levels and diabetic foot disease stage (one-way ANOVA, P<0.0001; n=10 in each group).

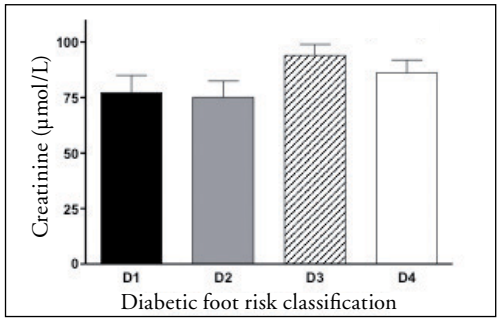

Figure 2. The relationship between creatinine and diabetic foot disease stage (one-way ANOVA, P=0.2; n=10 in each group).

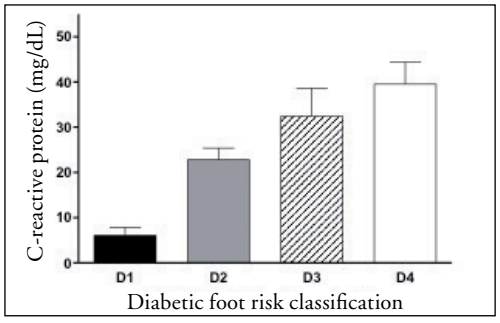

Figure 3. The relationship between C-reactive protein and diabetic foot disease stage (one-way ANOVA, P<0.0001; n=10 in each group).

Discussion

The high prevalence of anaemia is well recognised in patients with diabetes, but its association with diabetic foot disease has not been fully addressed. Data presented here from both cross-sectional and longitudinal analysis suggest an association between stage of diabetic foot disease and both haemoglobin and CRP levels. This is in keeping with a prospective cohort study by Hokkam (2009) who demonstrated a relationship was present between the probability of having diabetic foot disease and anaemia.

Harwant et al (2000) similarly demonstrated that lower haemoglobin concentration and higher white cell count (WCC) were factors associated with an increased incidence of amputations compared to foot sparing/local procedure. Further studies have revealed that low haemoglobin levels and elevated WCC/leucocytosis are associated with poor prognosis in patients with diabetic foot disease (Akanji et al, 1989; Ekpebegh et al, 2009).

It is important to note that, while a reduced haemoglobin level and inflammatory state identifies patients with diabetic foot disease as being at increased risk of adverse outcomes, none of the above authors investigated the association between haemoglobin and inflammation with stage of diabetic foot disease.

In healthy individuals, reduced circulating haemoglobin levels can be compensated for by increased peripheral perfusion (Jonsson et al, 1991), increased vasoreactivity (Schmidt-Lucke et al, 2000), and elevated erythropoietin levels that stimulate endothelial cell mitosis and motility important in neovascularisation, but such compensatory responses are significantly impaired in patients with diabetes, particularly in those with microvascular complications. Correction of anaemia by selective vasodilatation (Migliori et al, 1999) may be beneficial in patients with chronic ulcers and

may facilitate angiogenesis in granulation tissue during wound healing (Haroon et al, 2003; Galeano et al, 2004).

Patients with diabetes have an increased susceptibility to infection, largely due to macrophage dysfunction and inhibition of cell-mediated immunity (Eliashiv et al, 1978). Anaemia may also have significant effects on the immune system as erythropoietin is not only a haemopoietic factor, but also an immunomodulatory cytokine (Coleman and Brines, 2004). Interestingly, in the present study, despite exclusion of patients with evident infection, an inverse correlation was found between CRP levels and baseline haemoglobin levels.

Chronic and overt inflammation associated with diabetic microvascular disease reduces the effect of erythropoietin on the proliferation of erythroid precursor cells. This can be attributed to suboptimal response to erythropoietin (Bosman et al, 2002) associated with an increased production of cytokines, such as tumour necrosis factor-alpha, interleukin-1, or interferon-gamma (Goicoechea et al, 1998), which may suppress erythropoietin production (Zanjani et al, 1982). Furthermore, iron retention within macrophages and upregulation of hepcidin hormone result in inhibition of intestinal absorption, thus reducing its availability for bone marrow erythropoeisis (Weiss and Goodnough, 2005).

Failure to increase circulating erythropoietin concentrations in response to falling haemoglobin levels is the causative factor leading to anaemia associated with diabetic nephropathy. Several plausible mechanisms by which diabetes impairs the renal erythropoietin response to reduced haemoglobin levels have been proposed; microvascular damage, chronic hypoxia, oxidative stress, systemic inflammation, autonomic neuropathy, urinary erythropoietin loss and hyperfiltration (Thomas et al, 2003). Contrastingly, the present study shows that development of anaemia is independent of renal function assessed by creatinine, which is consistent with the findings of Dikow et al (2002), Thomas et al (2003) and the National Health and Nutrition Examination Survey-II (Astor et al, 2002). The mechanism whereby anaemia occurs without evidence of renal impairment in patients with diabetes is poorly understood. Although the authors excluded patients on dialysis, inevitably some patients may have a degree of nephropathy (Ritz and Haxsen, 2005).

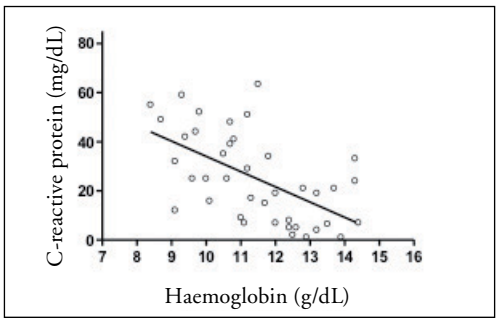

Figure 4. A scatter plot showing an inverse correlation between C-reactive protein and haemoglobin (P<0.0001).

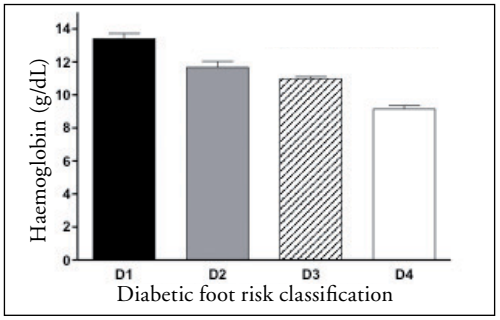

Figure 5. The relationship between haemoglobin and diabetic foot ulcer stage on patients who progressed from D1 to D4 (P<0.0001).

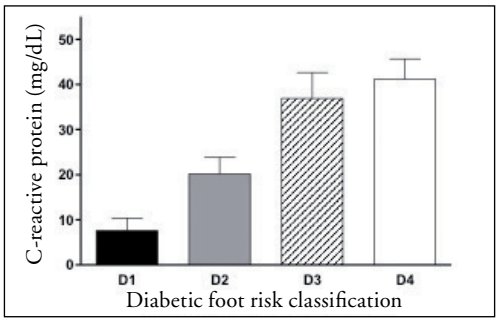

Figure 6. The relationship between C-reactive protein and diabetic foot ulcer stage on six patients who progressed from D1 to D4 (one-way ANOVA, P<0.0001).

Table 1. Values (mean ± standard error of the mean)of haemoglobin, C-reactive protein and creatinine in patients stratified to diabetic foot ulcer group according to NICE (2011) Quality Statement 10.

	D1 (*n*=10)	D2 (*n*=10)	D3 (*n*=10)	D4 (*n*=10)
Haemoglobin (g/dL)	13.2 ± 0.2	12.3 ± 0.4	11.0 ± 0.2	9.3 ± 0.2
C-reactive protein (mg/dL)	6.1 ± 1.9	22.8 ± 2.6	32.4 ± 6.2	39.5 ± 4.9
Creatinine (µmol/L)	77.2 ± 7.8	75.1 ± 7.3	94.0 ± 4.9	86.0 ± 5.8

D1 = normal sensation, palpable pulses (low risk); D2 = neuropathy, absent pulses, other risk factors (increased risk); D3 = neuropathy, absent pulses, deformity, skin changes, previous ulcer (high risk); D4 = ulcerated foot.

The overall prevalence of all-cause anaemia reported in a US study in people 65 years of age and older was 10.6%, with a prevalence of 11.0% for men and 10.2% for women (Guralnik et al, 2004). The authors found that anaemia was mild in the majority of cases (Hb >10 g/dL). Less severe degrees of anaemia in older people have typically not received much clinical attention.

Several studies have demonstrated poorer 5-year survival rates in older persons with mild anaemia, than in non-anaemic persons of the same age (Kikuchi et al, 2001). Whether worsening anaemia causes progression of diabetic foot disease, or vice versa, is worth debating as their pathogenesis are remarkably intricate and often exist in parallel. As the authors' analyses progressed, it came to light that patients with diabetic foot disease had thorough investigations, bar one; namely haemoglobin.

Screening routinely for anaemia in diabetic foot/podiatry follow-up clinics may be a cost-effective way of identifying and managing those with the condition and deteriorating diabetic foot disease. However, pending further research, a question exists with regard to the benefits of treatment of mild anaemia in similar patients.

Conclusion

In the present study, the authors showed that diabetic foot disease stage is associated with anaemia and the development of anaemia is independent of renal function, which may be explained by an underlying inflammatory process. Using retrospective records a large proportion of relevant data entry was absent. Lack of relevant documentation resulted in a small sample size, which was the main limitation of the study. Due to insufficient information regarding the duration of diabetes and HbA_{1c}, degree of neuropathy, and other significant comorbidities, causality cannot be substantiated.

Despite the small sample size and seemingly organised sub-group stratification, the authors' results demonstrated statistical significance indicating an association of anaemia and inflammation in patients at each stage of diabetic foot disease. For that reason, the authors are in the process of developing a prospective database to test this association in a larger cohort of patients with diabetes and foot disease that will have fewer of the biases and inconsistencies that have been reported here. Preliminary results from this prospective study have already revealed a higher incidence of anaemia and iron deficiency in patients with D4 diabetic foot disease classification.

Currently, there is no conclusive evidence that anaemia correction improves clinical outcomes in diabetic foot disease. Consequently, rather than treating diabetes complications, the indication for correcting anaemia is based on a prediction that correction will relieve symptoms, and improve patients' quality of life (Thomas, 2007). Further studies are needed to elucidate the link between anaemia and inflammation and their mechanistic role in diabetic foot disease. ■

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