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Low cobalamin (vitamin B_{12}) levels in multiple myeloma: a retrospective study

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ABSTRACT

Background: In patients with multiple myeloma a variety of metabolic events may occur. One of these are changes in the serum cobalamin (vitamin B12) concentration. Elevated as well as decreased serum cobalamin levels have been reported. The prevalence and clinical consequences of low cobalamin levels are largely unknown.

Objective: To investigate the prevalence of low serum cobalamin levels in patients with multiple myeloma and to describe the clinical features, haematological parameters and outcome in patients with multiple myeloma with low and normal serum cobalamin levels.

Methods: A retrospective study was conducted in the Deaconess Hospital in Eindhoven. Thirty-two patients were identified who fulfilled the diagnostic criteria for multiple myeloma and had at least one serum cobalamin level tested during the diagnostic or treatment period. A number of clinical characteristics, haematological parameters and outcome were scored.

Results: Twenty-one (66%) patients had a normal serum cobalamin level, nine (28%) patients had a low one and two (6%) patients had an elevated serum cobalamin level. Between the group with a normal and a low serum cobalamin level there were no differences in patients characteristics such as sex and age, tumour characteristics such as the type of paraprotein, tumour load or tumour stage nor in haematological parameters such as haemoglobin level, mean corpuscular volume and megaloblastic changes in the bone marrow. The median survival was not statistically different between both groups.

INTRODUCTION

Multiple myeloma is characterised by a clonal expansion of malignant plasma cells which produce monoclonal heavy and/or light chain immunoglobulins. The malignant plasma cells reside primarily in the bone marrow leading to suppression of the normal haematopoiesis and increased bone turnover eventually resulting in osteolysis. The relationship between malignant plasma cells and bone marrow stromal environment is complex.[1] Plasma cells produce a variety of cytokines, such as interleukin 1 and tumour necrosis factor (TNF), which influence the growth and activity of bone marrow cells such as osteoblasts and osteoclasts. The increased activity of these cells results in increased bone turnover and production of interleukin 6, which acts as a growth-promoting factor for the plasma cells. Common metabolic sequelae of the plasma cell expansion include hypercalcaemia, hypogammaglobulinaemia, and renal failure, either due to the hypercalcaemia or due to deposition of immunoglobulins in the kidney. There are a variety of less common or less well-known metabolic events in myeloma patients. One of these is a change in the serum cobalamin (vitamin B_{12}) concentration. Elevated as well as decreased serum cobalamin levels have been reported in small series of patients.[2-7] In these reports the prevalence of low cobalamin levels ranges from 0 to 70%. To investigate the prevalence and clinical characteristics of a low cobalamin level in patients with multiple myeloma a retrospective study was performed in myeloma patients treated in the Deaconess Hospital in Eindhoven during the period from 1996 to 2001.

PATIENTS AND METHODS

Patients potentially eligible for the study were identified using the laboratory data system (LABOSYS) which found patients who had undergone immunoelectrophoresis; the records of the bone marrow examinations were screened for results reporting more than 10% plasma cells. To be included in the study the patients identified had to fulfil the diagnostic criteria for multiple myeloma according to Durie and Salmon[8] and the serum cobalamin concentration had to be determined at least once during the diagnostic period or during the course or treatment of the disease. At the time of determining the serum cobalamin concentration the following data had to be available: haemoglobulin level, total leucocyte and thrombocyte count, mean corpuscular volume of the erythrocytes, and serum paraprotein, creatinine, calcium and folic acid levels. In addition the serum concentration of IgG, IgM and IgA, the percentage of plasma cells in the bone marrow, urine analysis for the presence of light chains, and radiographic evaluation of the skeleton had to be recorded. The presence of megaloblastic changes in the erythropoiesis in the bone marrow was scored by an independent investigator (J. Bury) who was unaware of the results of the serum cobalamin concentration. A low cobalamin level was defined as a serum concentration below 120 pmol/l. The results were compared using the Student's *t*-test of unpaired observations.

RESULTS

A total of 32 patients fulfilled all the above-mentioned criteria and were included in this retrospective study. Twenty-one (66%) patients had serum cobalamin levels within the normal range (130 to 850 pmol/l), nine patients (28%) had a low cobalamin level and two (6%) had a serum cobalamin concentration of more than 1476 pmol/l. In six of the nine patients with a low level of cobalamin, this low level was present at the time of diagnosis of multiple myeloma, while in three patients the low cobalamin level developed during the treatment and the course of the disease. The characteristics of the patients with a normal and a decreased cobalamin concentration are summarised in *table 1* and show no differences between the two groups. As a measure for the tumour load the stage of the disease according to the Durie and Salmon criteria,[8] level of the serum paraprotein, level of the remaining serum IgM and percentage of plasma cells in the bone marrow were used. As depicted in *table 2* patients with low cobalamin levels had no greater tumour load than patients with a normal serum cobalamin level. The haematological parameters are shown in *table 3*. There was no difference in the serum haemoglobulin level, mean corpuscular volume of the erythrocytes, and total leucocyte and

Table 1
The clinical characteristics of patients with multiple myeloma with a normal and low serum cobalamin level

	NORMAL COBALAMIN N=21	LOW COBALAMIN N=9
Cobalamin (pmol/l)		
Mean	300.4	93.8
Range	125-845	46-114
Age (years)		
Mean	66.5	68.8
Range	44-84	55-86
Sex		
Male	6	4
Female	15	5
Paraprotein		
IgA	4	5
IgG	10	4
Light chain	6	-
Nonsecreting	1	-

Table 2
The tumour load in patients with multiple myeloma with a normal and low serum cobalamin level

	NORMAL COBALAMIN N=21	LOW COBALAMIN N=9
Stage		
I	1	2
II	4	-
III	16	7
M component (g/l)		
Mean	27.6	27.3
Range	4.3-65.3	11.9-76.3
IgM (g/l)		
Mean	0.4	0.26
Range	0.02-1.32	0.02-0.74
Bone marrow plasma cells (%)		
Mean	51.75	52.2
Range	15-100	15-100

Table 3
Haematological parameters in patients with multiple myeloma with a normal and low serum cobalamin level

	NORMAL COBALAMIN N=21	LOW COBALAMIN N=9
Haemoglobulin (mmol/l)		
Mean	6.9	7.1
Range	3.7-8.7	5.4-8.6
Mean corpuscular volume (fl)		
Mean	93.4	96
Range	80-110	89-104
Leucocytes (10^9/l)		
Mean	6.6	5.6
Range	2.9-13.8	2.7-12.3
Thrombocytes (10^9/l)		
Mean	240	181
Range	101-591	83-286
Megaloblastic features		
Present	5	1

thrombocyte counts in the two groups. In addition there was no difference in the number of patients with megaloblastic changes in the erythropoiesis in the bone marrow. The serum folic acid levels did not differ between the two groups (data not shown). The median survival in patients with a normal cobalamin was 20.5+ months (range 1 to 76+ months) and in those with a low cobalamin level this was 16+ months (range 6 to 61+ months), which was statistically not significant (p=0.715).
Two patients had an elevated serum cobalamin level at the time of diagnosis. Both had an IgA paraprotein-producing myeloma with very high concentrations (69.6 and 77 g/l). One patient was lost for follow-up. In the other patient the serum cobalamin level normalised during cytostatic treatment with reduction of the serum IgA level.

DISCUSSION

In this study a low level of cobalamin was found in more than one fourth (28%) of the patients with multiple myeloma. The presence of low cobalamin levels was not related to patient characteristics such as sex and age or tumour characteristics such as the type of paraprotein or tumour load. As reported by others,[4] the presence of a low cobalamin level could not be predicted by haematological parameters. In contrast to selected reported cases[3,9] no association with megaloblastic haematopoiesis in the bone marrow was found in this study and previous reports.[2,6] In addition, the presence of a low cobalamin level seems no poor prognostic factor since the median survival was not significantly impaired.
Cobalamin is an essential vitamin for DNA synthesis and consists of a number of analogues of which methylcobalamin and deoxyadenosylcobalamin are metabolically active in humans.[10] Cobalamin is primarily present in a protein-bound form in foods of animal origin. After digestion cobalamin is released from the protein by gastric acid and peptic enzymes and bound to cobalamin-binding proteins such as the gastric intrinsic factor (IF). After resorption of the intrinsic factor-cobalamin complex in the terminal ileum through endocytosis by the mucosal cells, cobalamin is intracellularly bound to transcobalamin II (TC II) and the intrinsic factor shed into the lumen of the bowel. Cobalamin is transported by binding onto TC II which is produced by hepatocytes, endothelial cells and probably by enterocytes. The TC II molecule binds methylcobalamin as well as deoxyadenosylcobalamin. However, only 5 to 20% of cobalamin is bound to TC II. Only the TC II-bound cobalamin can enter the cell to be metabolically active. Methylcobalamin acts as a co-factor for methionin synthetase which regulates the formation of methionin from homocysteine whereas deoxyadenosyl-cobalamin acts as a co-factor of methylmalonic-coenzyme A mutase, which regulates the formation of succinyl CoA from methylmalonic CoA.
The remaining 80 to 95% of cobalamin is bound to haptocorrines (HC), which are mainly produced by myeloid cells, and consist of a sialic poor and a sialic rich analogue. The physiological role of HC is not fully established.
A number of clinical, especially haematological, conditions are associated with changes in the serum cobalamin concentration. Strongly elevated serum cobalamin levels are mainly found in myeloproliferative disorders such as chronic myeloid leukaemia, polycythaemia vera and the hypereosinophilic syndrome.[10] The elevation of the serum cobalamin level in chronic myeloid leukaemia is the result of an increased production of HC by the proliferating myeloid cells.[11] Also in lymphoproliferative diseases such as plasma cell disorders as multiple myeloma, changes in the serum cobalamin, TC II and HC are found.[12,13] As indicated in this study too, myeloma patients may have strongly elevated serum cobalamin levels. These increased serum cobalamin levels are thought to be the result of elevated TC II levels.[12,13]
In the past, several hypotheses have been postulated on the cause of the low levels of cobalamin in myeloma patients. Initially it was thought that the low cobalamin level was the result of gastric malabsorption due to pernicious anaemia, which is occasionally found,[14-16] or to hypochlorhydria or achlorhydria as frequently seen in old age.[4] There was scattered evidence of a pseudodeficiency of cobalamin as the result of an increased plasma volume in some myeloma patients[6] and it has also been hypothesised that the decreased serum cobalamin levels result from binding of cobalamin to the paraprotein.[17] Some investigators have found a decreased HC serum level in association with a decreased serum cobalamin level.[4] From the reported studies and our data it is not clear whether the low level of serum cobalamin found reflects an actual cobalamin deficiency or is merely the result of changes in the cobalamin binding proteins.[4,6,17,18]
An *in vitro* study provided some evidence for the assumption that the myeloma cells themselves may consume cobalamin.[5] In comparison with healthy controls the concentration of cobalamin in bone marrow suspension of myeloma patients was significantly higher with a positive correlation between the cobalamin concentration and the amount of bone marrow plasma cells. In addition, the investigators found an increased transcobalamin II-mediated cobalamin uptake in bone marrow cells of myeloma patients in comparison with healthy controls.
The consequences of low cobalamin levels found in myeloma patients are unclear. This study shows that this condition does not seem to influence the prognosis. Theoretically, low cobalamin levels may have an effect on the bone turnover contributing to the myeloma-associated osteoporosis. Older studies in nonmyeloma patients with cobalamin deficiency provided scattered evidence for a

diminished osteoblastic activity leading to osteoporosis, which is reversible after suppletion with cobalamin.[19-22] Several *in vitro* and clinical studies indicate that cobalamin may indeed have a direct or indirect effect on the osteoblast and osteoclast activity. Firstly, the activity and proliferation of human bone marrow stromal osteoprogenitor and osteoblastic cells have been shown to be dependent on the cobalamin concentration.[23] Furthermore, clinical studies have shown that cobalamin deficiency results in a significantly increased homocysteine as well as TNF-α level.[24] The elevated homocysteine levels may be caused by an insufficient activity of methionine synthetase which may be the result of a decrease in methylcobalamin, a cobalamin analogue that is disproportionately decreased in myeloma patients.[18] It has been demonstrated that high serum homocysteine levels may be associated with impaired cross-linking of collagen.[25] As mentioned previously, experimental studies indicate that TNF-α has a regulatory effect on the osteoclast activity.[1] Taken together, these data may suggest that, if the low cobalamin level were to reflect an actual cobalamin deficiency, the low cobalamin level in myeloma patients may have a contributory effect on the increased and pathological bone turnover observed in myeloma patients.

In conclusion, changes in serum cobalamin levels is a frequent finding in patients with multiple myeloma. Especially a low cobalamin level is a common metabolic feature in this disorder. The reason for this low level of cobalamin is not fully understood and the question whether cobalamin substitution is warranted in myeloma patients with an established low cobalamin level cannot be answered. A well-defined study is needed to establish whether a low cobalamin level reflects an actual cobalamin deficiency, to evaluate the metabolic consequences of the observed low cobalamin level and to explore the potentially additional role of a low cobalamin level in the bone turnover in patients with multiple myeloma.

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