Masked vitamin B_{12} and folate deficiency in the elderly

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1. A high incidence of vitamin B_{12} or folate deficiency, or both, may be found in the elderly, particularly those in hospital. This report concerns fifty cases detected in an inner-city-area geriatric unit during the course of routine clinical investigation. The majority had none of the classical haematological signs of vitamin B_{12} or folate deficiency, and all the patients reported had a mean corpuscular volume (MCV) of less than 100 fl.

2. There was a significant negative correlation between the MCV and the erythrocyte folate ($P < 0.01$), supporting earlier published work using a low serum folate as an index of folate deficiency.

3. There was no correlation between the MCV and the serum vitamin B_{12}. Published work differs on this point.

4. Serum iron, total Fe-binding capacity and percentage Fe saturation results were available in forty patients in this series. There was a significant positive correlation between the serum Fe and the MCV ($P < 0.01$) and 34% of patients had haematological evidence of Fe deficiency. In the majority, however, there was no evidence that associated Fe deficiency had masked the haematological signs of vitamin B_{12} or folate deficiency.

5. More attention should be paid to the problem of 'masked' vitamin B_{12} and folate deficiency in the elderly. There is a case for routine screening of the elderly for vitamin B_{12} and folate deficiency irrespective of the MCV.

Langdon (1905) observed a number of patients 'whose chief complaints were of a distinctly mental and nervous character and who in the course of time were found to present the characteristic blood state and somatic phenomena of pernicious anaemia'. Later, Strachan & Henderson (1965) recommended screening for vitamin B_{12} deficiency in all patients diagnosed as having senile or pre-senile dementia, cerebral atheroma or depression, and pointed out that normal peripheral blood and bone marrow appearances should not be regarded as excluding vitamin B_{12} deficiency. Routine screening for folate and vitamin B_{12} deficiency in a geriatric hospital population has been advocated (Munasinghe & Pritchard, 1978) but is not widely practised.

The mean corpuscular volume (MCV) is classically raised in vitamin B_{12} or folate deficiency, but subnormal vitamin B_{12} and folate levels have been reported with an MCV less than 100 fl (Sheridan *et al.* 1974), especially in the presence of iron deficiency (Pederson *et al.* 1957; Cox *et al.* 1959). Nevertheless, some clinicians and haematologists are still reluctant to measure serum vitamin B_{12} or folate levels in the absence of overt macrocytosis, and there is no doubt that many cases of vitamin B_{12} or folate deficiency, or both, are undetected. The purpose of the present paper is to highlight the problem of masked vitamin B_{12} and folate deficiency in the elderly, to re-examine the relation between the serum Fe, vitamin B_{12}, erythrocyte folate and the MCV, and to demonstrate the need for screening for vitamin B_{12} and folate deficiency irrespective of the MCV in the elderly.

EXPERIMENTAL

Patients studied

This report concerns fifty patients with vitamin B_{12} or folate deficiency, or both, with an MCV of less than 100 fl detected during routine clinical investigation of patients admitted to an inner-city-area geriatric unit during 1 year. With three exceptions all the patients were on one clinical firm. They varied in age from 71 to 94 years, with a mean of 81 years. Twenty

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were male and thirty female. Forty-nine were investigated on admission to an acute assessment ward, and one was on a long-stay ward.

The Department of Geriatric Medicine at St Thomas's Hospital has no formal policy for the screening of patients for vitamin B_{12} or folate deficiency. In the present series, vitamin B_{12} and folate status were investigated in patients with anaemia irrespective of the MCV; in patients with macrocytes, hypersegmented polymorphs or large platelets reported on the blood film; in patients with depression, dementia or a confused state; and those with a past history of partial gastrectomy. Two patients had been vitamin B_{12} deficient in the past, but had discontinued replacement therapy. Serum Fe, total Fe-binding capacity (TIBC) and percentage Fe saturation were measured in forty of the patients in this series.

Venous blood samples were usually taken in the morning after breakfast. All vitamin B_{12}, folate and Fe results quoted are based on a single estimation of each index that corresponded most closely with the full blood count. No patient was on methotrexate, trimethoprin or therapeutic folate analogues at the time of folate assay. Vitamin B_{12} absorption tests were not undertaken routinely in patients with vitamin B_{12} deficiency, nor was the bone marrow examined routinely in the patients in this series. Diet was not formally assessed.

Methods

All blood counts and assays were undertaken as part of the routine work of the Department of Haematology, St Thomas's Hospital. Serum vitamin B_{12} was measured by a competitive protein-binding radioassay using pooled human serum as a binder giving a normal range of 200–1000 ng/l. The assay had been validated by comparison with *Euglena gracilis* and *Lactobacillus leichmannii* bioassays.

Serum folate and erythrocyte folate (RBC folate) were measured by standard radioassay techniques. During the first 6 months of the study a single isotope method using the Amersham folate radioassay kit (method a) was used. The normal range for the laboratory with this method was 4–10 μg/l for serum folate and 150–750 μg/l for RBC folate. When this kit was withdrawn the Becton Dickinson folate radioassay kit (^{125}I) (method b) was used. This was more sensitive, giving a lower limit of 135 μg/l for RBC folate.

Serum Fe was measured by a standard AutoAnalyzer method using ferozine (Technicon Instrument Co. Ltd, 1976).

Standard haematological indices such as the MCV (normal 84 (SD7) fl), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were obtained by analysis of samples on a Coulter model S or S + (phase 1) counter. Bone-marrow samples were examined by routine methods.

RESULTS

The mean results, the range of results and normal values for each index measured are shown in Table 1. The majority of patients had a normal haemoglobin, but 32% were anaemic as judged by a haemoglobin less than 120 g/l. The mean MCH and MCHC results were within the normal range, but 34% of patients had an MCHC below 320 g/l. The MCV was below 100 fl in all cases by virtue of the cut-off point chosen for inclusion in this series. Classical vitamin B_{12} or folate deficiency causes an increase in erythrocyte size, which is reflected in a high MCV. Although all the patients in this series had vitamin B_{12} or folate deficiency, the peripheral blood film showed macrocytes in only two cases, hypersegmented polymorphs in three, and some large platelets in three cases. Most blood films gave no hint of vitamin B_{12} or folate deficiency. Anisocytosis was reported in twenty-eight cases (56%), anisocytosis and poikilocytosis in twenty (40%), elliptocytes in six and hypochromic erythrocytes in five cases.

Table 1. *Mean values for blood indices in elderly patients with folate or*
vitamin B_{12} deficiency

Measurement	n	Mean	Range	Normal range Mean	SD
Haemoglobin (g/l)	50	125	75–163	♂160	20
				♀140	20
MCV (fl)	50	88	65–99	84	7
MCH (pg)	48	28·8	16·8–33·1	29	3
MCHC (g/l)	50	323	279–353	340	20
Serum vitamin B_{12} (ng/l)	50	267	79–922	200–1000	
RBC folate (μg/l)	47	164	70–519	(150–750 method a)*	
				(135–750 method b)*	
Serum folate (μg/l)	30	1·7	0·3–4·5	4–10	
Serum iron (μmol/l)	40	10·1	2·7–21	13–32	
TIBC (μmol/l)	40	53·4	29·7–80·1	45–70	
Percentage Fe saturation	40	19	3–39	18–71	

MCV, mean corpuscular volume; MCH, mean corpuscular haemoglobin; MCHC, mean corpuscular haemo-globin concentration; RBC, erythrocytes; TIBC, total iron-binding capacity.
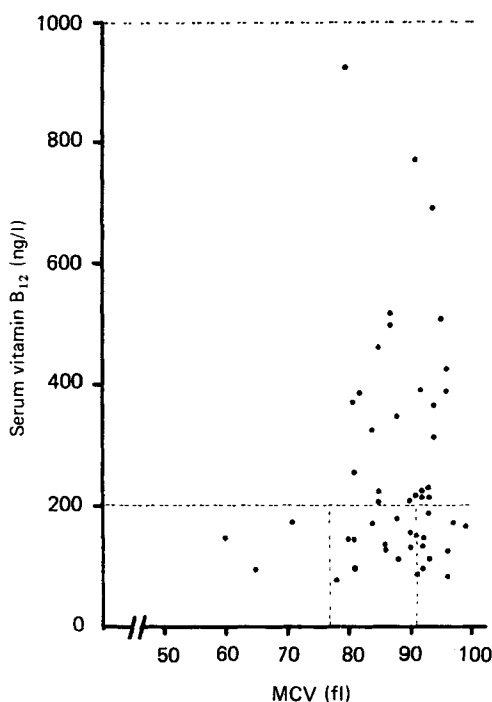
* For details, see p. 614.

Fig. 1. Serum vitamin B_{12} (normal range 200–1000 ng/l) and mean corpuscular volume (MCV) values (normal range 84 (SD 7) fl) in elderly patients with folate or vitamin B_{12} deficiency.
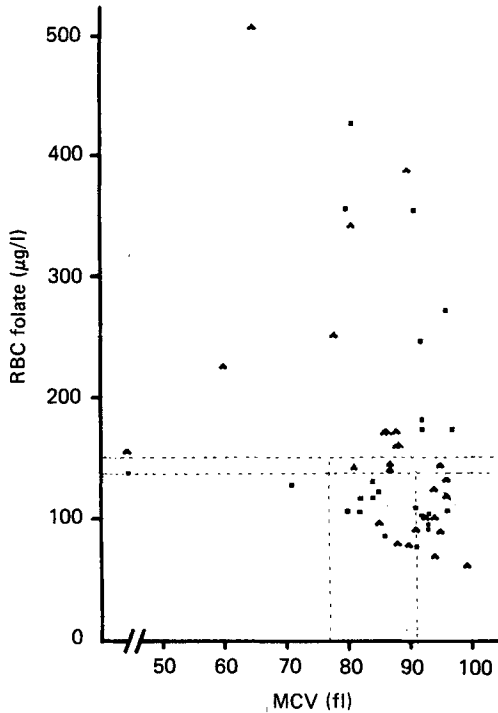
Fig. 2. Erythrocyte folate (RBC folate; μg/l) and mean corpuscular volume (MCV; fl) in elderly patients with folate or vitamin B_{12} deficiency. RBC folate results by method a (▲, normal range 150–750 μg/l); results by method b (■, normal range 135–750 μg/l). For details of methods, see p. 614.

Vitamin B_{12} and folate

Serum vitamin B_{12} values were obtained in all fifty patients, and RBC folate was measured in forty-seven. In three patients a diagnosis of folate deficiency was based on a low serum folate only. Twenty-five patients in this series had a low serum vitamin B_{12}, thirty-four had evidence of folate deficiency, and eight had both vitamin B_{12} and folate deficiency. All had an MCV below 100 fl. Fourteen patients with a low serum vitamin B_{12} had a normal or low MCV, and thirteen patients with a low erythrocyte folate level had a normal or reduced MCV. There was a significant negative correlation between the MCV and the RBC folate (R −0.43, $P < 0.01$, n 47) but there was no correlation between the MCV and the serum vitamin B_{12} (R 0.07). These data were also re-examined after log transformation. This did not alter the conclusions reached. The significant negative correlation between MCV and RBC folate persisted ($R - 0.342$, $P < 0.05$), and there was still no significant correlation between the MCV and serum vitamin B_{12}.

Comparison of the RBC folate results in patients with and without vitamin B_{12} deficiency showed a mean RBC folate of 110 μg/l (n 23) in those with a normal vitamin B_{12}, compared with a mean RBC folate of 214 μg/l (n 24) in those with a low vitamin B_{12}. The difference was highly significant ($P < 0.001$, Student's t test). This suggests that the low RBC folate results were due to tissue folate depletion rather than to failure of erythrocytes to take up folate as may occur in vitamin B_{12} deficiency (Tisman & Herbert, 1973). There was, overall, a significant negative coefficient of correlation between RBC folate and serum vitamin B_{12} results ($R - 0.527$, $P < 0.001$, using log-transformed data).
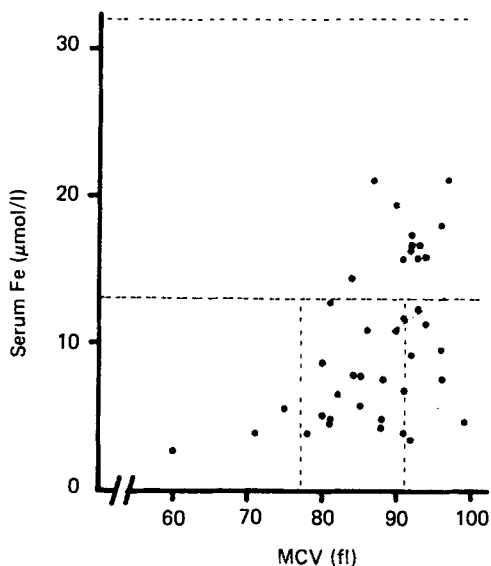
Fig. 3. Serum iron (μmol/l) *v.* mean corpuscular volume (MCV; fl) in elderly patients with folate or vitamin B_{12} deficiency.

Serum Fe

Serum Fe, TIBC and percentage Fe saturation results were obtained in forty patients in this series. The serum Fe was low in twenty-eight of forty patients (70%) (see Fig. 3). Of these twenty-eight patients, four had a raised TIBC, seventeen a normal TIBC and seven a low TIBC. On this basis four had classical Fe deficiency and twenty-four the Fe pattern of chronic disease. Mean results are given in Table 1. The MCHC, MCH and MCV are all reduced in classical Fe deficiency resulting in a microcytic hypochromic anaemia. In our patients there was a significant positive correlation between the serum Fe and the MCHC (R 0·52, $P < 0·001$) and between the serum Fe and the MCH (R 0·54, $P < 0·001$). On the basis of an MCHC below 320 g/l, 34% of patients were Fe deficient. Serum Fe results were available in twenty patients with a low vitamin B_{12} and twenty with a normal vitamin B_{12}. There was no significant difference between the serum Fe from each group (Student's *t* test). Overall, there was no significant correlation between the serum Fe and the serum vitamin B_{12} (R 0·04).

Serum Fe results were available in twenty-six patients with a low RBC folate and thirteen with a normal RBC folate. The mean serum Fe in each group was identical. There was no correlation between the serum Fe and the erythrocyte folate ($R - 0·05$).

Overall there was a significant positive correlation between the MCV and the serum Fe (R 0·46, P 0·01, n 40), but no correlation between the MCV and the MCHC (R 0·1).

Bone marrow

A bone marrow examination was deliberately not done as a routine. Results in seven patients showed that two had a frankly megaloblastic marrow, one was marginally megaloblastic, two had occasional giant metamyelocytes in a normoblastic marrow, and two were normal. Both patients with a frankly megaloblastic marrow had absent stainable marrow Fe.

DISCUSSION

The evidence suggests that patients with classical megaloblastic anaemia or subacute combined degeneration of the spinal cord represent only the tip of the iceberg. The majority of elderly people with vitamin B_{12} or folate deficiency do not have overt macrocytosis or abnormal neurological signs. In one report from a geriatric hospital, almost half the cases with a low serum folate or vitamin B_{12} deficiency had an MCV below 90 fl (Munasinghe & Pritchard, 1978). A survey carried out in Ireland showed that 11% of people over the age of 65 years living outside an institution had either vitamin B_{12} deficiency or a low serum folate, and 69% of cases had an MCV below 100 fl (Sheridan et al. 1974). The erythrocyte folate is a more accurate measure of folate status than the serum folate, thus our results confirm and extend these earlier reports. Whereas Sheridan et al. (1974) found a statistically significant correlation between the MCV and the serum vitamin B_{12} level neither we, nor Munasinghe & Pritchard (1978) could confirm this. It is likely that other factors complicate the picture in a geriatric hospital population as opposed to the apparently fit elderly in the community. It is clear that the MCV is a poor guide to vitamin B_{12} status in the elderly, and no reliance should be given to it for screening purposes.

Several reports indicate that vitamin B_{12} deficiency or folate deficiency can co-exist with otherwise classical Fe deficiency (Pederson et al. 1957; Cox et al. 1959; Roberts et al. 1977) and the problem is not confined to the elderly. The low serum vitamin B_{12} may return to normal with treatment of the Fe deficiency alone (Cox et al. 1959). Malabsorption of protein-bound vitamin B_{12} has been reported in chronic Fe deficiency (Dawson et al. 1984). There was haematological evidence of Fe deficiency in 34% of our patients. Although there was no direct correlation between the serum Fe and the serum vitamin B_{12} or RBC folate, the correlation between erythrocyte indices and serum Fe suggests that associated Fe deficiency may have masked morphological evidence of vitamin B_{12} or folate deficiency in the peripheral blood in a number of our patients. In the majority of patients, however, the reason for the lack of classical haematological evidence of deficiency was not clear.

Of the fifty patients we report, forty-seven were drawn from not more than 246 admissions under the care of one consultant. Thus our results indicate that approximately one in five admissions to the South Western Hospital Geriatric Unit have 'masked' vitamin B_{12} or folate deficiency. A low serum folate has been reported in 16–40% of admissions to a geriatric hospital (Hurdle & Picton Williams, 1966; Sneath et al. 1973): the incidence is especially high in those who are poorly motivated and withdrawn and folate deficiency in the elderly is usually due to a poor diet rather than to folate malabsorption (Hurdle & Picton Williams, 1966). Folate is required in DNA synthesis, some amino acid conversions, and possibly monoamine metabolism and synaptic events (Reynolds, 1976). The role of folate deficiency in dementia, depression and memory impairment should not be underestimated (Sneath et al. 1973; Reynolds, 1976).

The significance of a low serum vitamin B_{12} is the subject of debate. Doubt has been cast on some vitamin B_{12} radioassays (Brynskow et al. 1983; Guthrie & Priest, 1983) and some people disregard a low serum vitamin B_{12} in the absence of haematological evidence of deficiency (Pathy & Newcombe, 1980). We do not advocate this approach, since neurological damage can occur in the absence of anaemia. The demonstration of malabsorption of protein-bound vitamin B_{12} in subjects with a low serum vitamin B_{12} level indicates that low vitamin B_{12} results should not be too readily discounted (Dawson et al. 1984). Indeed, it has been argued that the lower limit of normal for serum vitamin B_{12} may be too low, since factors such as smoking can increase requirements (Smith, 1960). It is prudent to consider a moderately reduced serum vitamin B_{12} level to be a sign of severe B_{12} deficiency (Mollin et al. 1976).

It is not known whether vitamin B$_{12}$ levels decrease with age in the absence of pathology, nor whether low vitamin B$_{12}$ levels in the elderly are due to abnormal transcobalamin levels. There is, however, a trend towards reduced absorption of protein-bound vitamin B$_{12}$ with increasing age (Dawson *et al.* 1984). An association between a low serum vitamin B$_{12}$ and Alzheimer-type dementia has been reported (Cole & Prchal, 1984). Since neurological and psychiatric syndromes due to vitamin B$_{12}$ deficiency are treatable unless cerebral demyelination has occured (Smith, 1960), it is important to detect and treat all patients with vitamin B$_{12}$ or folate deficiency before irreversible damage has occurred.

Our results show that a high index of clinical suspicion, irrespective of the MCV, will uncover a great many cases of vitamin B$_{12}$ and folate deficiency in the elderly. There is a strong case for routine screening for vitamin B$_{12}$ and folate deficiency in the elderly.

REFERENCES

Brynskow, J., Anderson, K., Gimsing, P. & Hippe, E. (1983). *Lancet* i, 1104–1105.

Cole, M. G. & Prchal, J. F. (1984). *Age and Ageing* 13, 101–105.

Cox, E. V., Meynell, M. J., Gaddie, R. & Cooke, W. T. (1959). *Lancet* ii, 998–1001.

Dawson, D. W., Sawers, A. H. & Sharma, R. K. (1984). *British Medical Journal* 288, 675–678.

Guthrie, D. L. & Priest, C. J. (1983). *Medical and Laboratory Science* 40, 95.

Hurdle, A. D. F. & Picton Williams, T. C. (1966). *British Medical Journal* ii, 202–205.

Langdon, F.W. (1905). *Journal of the American Medical Association* 45, 1635–1638.

Mollin, P. L., Anderson, B. B. & Burman, J. F. (1976). *Clinics in Haematology* 5, 524–529.

Munasinghe, D. R. & Pritchard, J. G. (1978). *British Journal of Clinical Practice* 32, 16–18.

Pathy, M. S. & Newcombe, R. G. (1980). *Gerontology* 26, 34–42.

Pederson, J., Lund, J., Ohlsen, A. S. & Kristenson, M. P. O. (1957). *Lancet* i, 448–453.

Reynolds, E. H. (1976). *Clinics in Haematology* 5, 661–696.

Roberts, P. D., St. John, D. J. B., Sinha, R., Stewart, J. S., Baird, I. M., Coghill, N. F. & Morgan, J. V. (1977). *British Journal of Haematology* 20, 165–176.

Sheridan, D. J., Temperley, I. J. & Gatenby, P. B. B. (1974). *Journal of the Irish Colleges of Physicians and Surgeons* 4, 39–45.

Smith, A. D. M. (1960). *British Medical Journal* ii, 1840–1845.

Sneath, P., Chanarin, I., Hodkinson, H. M., McPherson, C. K. & Reynolds, E. H. (1973). *Age and Ageing* 2, 177–182.

Strachan, R. W. & Henderson, J. G. (1965). *Quarterly Journal of Medicine* 34, 303–317.

Technicon Instrument Co. Ltd (1976). *Technicon Methodology Sheet* SF4–0025 F14. Basingstoke: Technicon Instrument Co. Ltd.

Tisman, G. & Herbert V. (1973). *Blood* 41, 465–469.