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Cognitive and psychiatric effects of vitamin B_{12} replacement in dementia with low serum B_{12} levels: a nursing home study

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Abstract

Background—The aim of this study is to determine whether B_{12} replacement would ameliorate cognitive and psychiatric symptoms in elderly subjects with dementia and low serum B_{12} levels.

Methods—A test group (n=28) of nursing home residents with low serum B_{12} levels (<250 pg/mL) and a matched comparison group (n=28) with normal serum B_{12} levels (>300 pg/mL) were evaluated by blinded raters while the test group received intramuscular (IM) B_{12} replacement therapy. All subjects were assessed at baseline, 8 weeks, and 16 weeks with the Dementia Rating Scale, Brief Psychiatric Rating Scale, and Geriatric Depression Scale.

Results—Although B_{12} replacement produced significant improvement in hematologic and metabolic parameters, it yielded no significant effect on cognitive or psychiatric variables. A few subjects evidenced notable individual treatment responses; however, these were not statistically more frequent than in the normal B_{12} group.

Conclusions—These results suggest that B_{12} replacement is unlikely to benefit cognitive or psychiatric symptoms in the vast majority of elderly dementia patients with low serum B_{12} levels.

Keywords

vitamin B_{12}; cobalamin; dementia; methylmalonic acid; homocysteine

Introduction

Serum vitamin B_{12} (cobalamin) testing has long been – and continues to be – recommended as part of the routine screening of patients with dementia (American Psychiatric Association, 2007). This recommendation reflects the widespread assumption that B_{12} deficiency is a reversible cause of dementia, or at least is commonly associated with cognitive impairment that may be partially correctable. However, surprisingly little research has systematically

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Conflict of interest None.

Description of authors' roles CHvD designed the study, evaluated subject eligibility, performed data analyses, and wrote the paper. JML, RMR, and APS performed psychometric assessments and assisted with the paper.

examined the cognitive and behavioral response to treatment of low serum B_{12} levels in the setting of dementia. A full diagnostic work-up is frequently not undertaken until a dementing illness is well established; and when B_{12} deficiency is diagnosed in a demented patient, it is usually not the only (or even primary) etiological process involved. Furthermore, B_{12} deficiency is often incidentally detected during the course of a dementia of other etiology (e.g. Alzheimer's disease). In such circumstances there is little prognostic data available to guide clinicians regarding the likely response to B_{12} replacement therapy.

Further diagnostic challenges are posed by patients who present with subtle B_{12} deficiency (Carmel *et al.*, 1987) or B_{12}-related neuropsychiatric syndromes without anemia or macrocytosis (Lindenbaum *et al.*, 1988). These diagnostic uncertainties prompted the quest for biochemical markers of tissue B_{12} deficiency and the development of assays for two metabolites that accumulate if B_{12} is lacking: methylmalonic acid (MMA) and homocysteine (Hcys) (Stabler *et al.*, 1986; 1988). The measurement of serum MMA and Hcys in combination with serum B_{12} levels has enlarged the understanding of B_{12} deficiency considerably beyond the classically described megaloblastic anemia and subacute combined systems disease. For example, if defined by an elevated serum concentration of MMA, then B_{12} deficiency is a relatively common condition affecting at least 15% of the elderly (Pennypacker *et al.*, 1992; Lindenbaum *et al.*, 1994).

Apart from case reports, few studies have reported outcome data on the cognitive and behavioral response to treatment of low serum B_{12} levels in the setting of dementia. There have been particularly few placebo-controlled trials of B_{12} replacement in dementia (de la Fournière *et al.*, 1997; Hvas *et al.*, 2004), presumably because placebo controls have been deemed unethical by many investigators (Martin *et al.*, 1992; Teunisse *et al.*, 1996). Previous prospective studies have thus largely involved open-label designs (Martin *et al.*, 1992; Carmel *et al.*, 1995; Cunha *et al.*, 1995), in some cases with comparison to neuropsychological test performance by a reference group (Teunisse *et al.*, 1996) or matched controls (Eastley *et al.*, 2000) with dementia and normal B_{12} levels (for a review of these studies, see Discussion). However, an intermediate solution not previously reported is to use a virtual control group of patients with normal levels for blinding of outcome raters.

In the present project we examined whether B_{12} replacement would ameliorate cognitive and behavioral measures in one particular population: nursing home residents with low serum B_{12} levels and relatively established cases of dementia. A test group of elderly nursing home residents with dementia and low B_{12} levels and a matched comparison group with normal B_{12} levels were both rated on cognitive and psychiatric scales in a single-blind manner (rater blind) while the test group received standard intramuscular (IM) B_{12} replacement therapy. In addition, we used assays of MMA and Hcys to measure tissue B_{12} deficiency and to relate this to clinical response to B_{12} replacement. This study was conducted nearly 20 years ago but has not previously been published. Given the up-to-date methodology employed, the unique protocol design, and the current interest in B-vitamins and dementia, this study provides an important extension of contemporary knowledge.

Methods

Human subjects

The study sample consisted of 28 elderly (age ≥65 years) subjects with dementia and low serum B_{12} levels (<250 pg/mL) and 28 subjects with dementia and normal B_{12} levels (>300 pg/mL), who resided in a nursing facility with both skilled and intermediate care units (the Jewish Home for the Elderly, Fairfield CT, U.S.A.). Subjects were enrolled between October 1988 and August 1989 (and completed the study by December 1989). B_{12} levels were obtained as part of routine annual medical examination, which included medical

history, physical and neurological examinations, serum chemistries (including lactate dehydrogenase, LDH), thyroid function studies, complete blood count (CBC) (including hemoglobin, hematocrit, red blood cell (RBC), erythrocyte mean corpuscular volume (MCV), white blood cell (WBC) and differential, and platelets), B_{12}, folate (RBC or serum), urinalysis, electrocardiogram, and chest X-ray. All residents with B_{12} levels <250 pg/mL were invited to participate as members of the test group. From residents with normal B_{12} levels (>300 pg/mL), comparison subjects were selected to participate in parallel with low B_{12} subjects. Normal B_{12} subjects were group-matched with low B_{12} subjects for age, gender, and severity of dementia – as measured by the Mini-mental State Examination (MMSE; Folstein *et al.*, 1975), which was also performed as part of routine annual assessment. Cutoffs of 250 pg/mL and 300 pg/mL were adopted because of previous reports that some patients with levels up to 300 pg/mL may show B_{12}-responsive neuropsychiatric disorders (Lindenbaum *et al.*, 1988).

Eligibility criteria for both subject groups included DSM-IIIR criteria (American Psychiatric Association, 1987) for dementia and a score of ≤ 24 on MMSE (Folstein *et al.*, 1975). Subjects were excluded who were currently receiving B_{12} replacement therapy; had severe sensory impairment (e.g. blindness) which precluded use of standard cognitive and psychiatric rating scales; or had a neurological disease likely to produce marked cognitive or behavioral change during the study (e.g. brain tumor). However, eligibility criteria were otherwise very liberal in order to study effects of B_{12} replacement across a broad spectrum of clinical dementia in the nursing home setting. Thus subjects were included across the full severity range of dementia (MMSE=0–24), with stable neurological diseases, or receiving central nervous system (CNS) active medications. Psychotropic medication use was common in both the low B_{12} (antipsychotics 7, benzodiazepines 7, antidepressants 3) and normal B_{12} (antipsychotics 11, benzodiazepines 5, antidepressants 4) groups. All subjects who met eligibility criteria for low or normal B_{12} groups agreed to participate and were enrolled during a 10-month period. All subjects (or their responsible next of kin) provided informed consent, and were studied under a protocol approved by the Yale Human Investigation Committee.

Subjects were initially evaluated by a geriatric psychiatry fellow (CHvD) to establish the diagnosis, classification, and characteristics of dementia (American Psychiatric Association, 1987). This evaluation included a review of symptoms, neurological examination, interview of family members, and detailed chart review. In addition to annual medical examinations (see above), a VDRL (test for syphilis) had been performed on all subjects at the time of nursing home admission. Twenty-seven of the 56 subjects had had a previous documented head CT (computed tomography scan) or MRI (magnetic resonance imaging) performed since onset of dementia. Subjects were classified according to the probable etiology of dementia, assuming that B_{12} deficiency was not exclusively causal. Classification of dementia in the low B_{12} group included: primary degenerative dementia of the Alzheimer type (DAT) (American Psychiatric Association, 1987) (18), multi-infarct dementia (MID) (American Psychiatric Association, 1987) (3), DAT plus MID (patients who met some criteria for both etiologies, e.g. a progressive dementia with a recent stroke) (3), DAT plus Parkinson's disease (PD) (3), and DAT plus Herpes encephalitis (1). Classification of dementia in the normal B_{12} group included: DAT (22), MID (2), DAT plus MID (2), DAT plus PD (1), and DAT plus dementia due to head trauma (1).

Procedure

All low B_{12} subjects were tested for tissue deficiency with serum MMA (Stabler *et al.*, 1986), and serum Hcys (Stabler *et al.*, 1988) and administered baseline cognitive and psychiatric ratings (see below). They then received replacement B_{12} therapy in a

standardized manner: 1,000 μg IM daily for 1 week, then 1,000 μg IM weekly for the duration of the study (16 weeks total).

All subjects (low and normal B_{12} groups) were rated by a second examiner (JML, RMR, APS; geriatric psychiatrist or psychiatry resident), blind to B_{12} status, at 0 weeks (prior to initiation of B_{12} treatment for the low B_{12} group), 8 weeks, and 16 weeks with the following scales: (1) Dementia Rating Scale (DRS, scoring range 0–144) (Mattis, 1976) which has been found to be sensitive to change in patients with moderate to severe dementia (Salmon *et al.*, 1990); (2) Brief Psychiatric Rating Scale (BPRS, 16-item, scoring range 16–112) (Overall and Gorham, 1962); (3) Geriatric Depression Scale–abbreviated version (GDS, scoring range 0–15) (Brink *et al.*, 1982; Sheikh and Yesavage, 1986). Some subjects with severe dementia were considered unratable using the BPRS and/or GDS and their data were not included in the analysis of those variables.

After 8 weeks, CBC, LDH, MMA and Hcys were repeated for low B_{12} subjects to measure hematologic and metabolic response to treatment.

Laboratory studies

All laboratory studies were performed by Metpath-Columbia Laboratory (Bridgeport, CT and Teterboro, NJ). Serum B_{12} and RBC folate assays utilized competitive protein binding. MMA and Hcys assays employed capillary gas chromatography-mass spectrometry. Initially, they were performed (under contractual agreement with Metpath) at the laboratories of Robert H. Allen and Sally P. Stabler (University of Colorado) where they were developed before this technology was transferred to Metpath's Teterboro, NJ laboratory. Peripheral blood smears were read by a blinded technician in the laboratory of John E. Lindenbaum at the College of Physicians and Surgeons of Columbia University.

Statistical analysis

Baseline differences between low B_{12} and normal B_{12} groups were analyzed by two sample Student t-tests. Effects of B_{12} treatment on hematologic and metabolic parameters in the low B_{12} group were analyzed by paired t-tests at 0 and 8 weeks of treatment. However, the data for MMA were not normally distributed (Lilliefors test) and were analyzed instead by Wilcoxon matched-pairs test.

The primary efficacy variable in this study was change from baseline to week 16 in DRS score, and the secondary efficacy variables were change from baseline to week 16 in BPRS and GDS scores. All three variables were analyzed using the last observation carried forward (LOCF) approach. For each of these variables, a comparison between the two groups was performed using an analysis of covariance model (ANCOVA) with group as factor and baseline score as covariate. Change in DRS scores was not normally distributed (Lilliefors test) and were re-analyzed post-hoc by Mann-Whitney U test. The number of subjects who showed a significant individual cognitive improvement (defined as ≥12 points on DRS – an improvement equivalent to the average deterioration in score over one year in DAT patients (Salmon *et al.*, 1990)) was compared between groups by χ^2. Correlations between variables were analyzed using Pearson's product moment correlation (r) or Spearman's rank correlation coefficient (ρ) depending on the normality of data.

All statistical analyses employed two-tailed tests of significance, using a significance level of α 0.05. All statistical analyses were performed using=SPSS (SPSS Inc., Chicago, IL) or SYSTAT (SYSTAT Inc., Evanston, IL) software.

Results

Subject characteristics

Demographic, clinical, and baseline neuropsychological characteristics of low B_{12} and normal B_{12} dementia subjects are displayed in Table 1. The two groups did not differ significantly in age, gender, education, duration of dementia, or Hachinski ischemia score (Hachinski *et al.*, 1975). They also did not differ in any neuropsychological measure at baseline (MMSE, DRS, GDS, or BPRS), although there was a nonsignificant trend for low B_{12} subjects to have higher BPRS scores than normal B_{12} subjects (t=1.80, df=46, p=0.079). Twenty-five of 28 low B_{12} and 23 of 28 normal B_{12} subjects were considered ratable on the BPRS. BPRS scores for these subjects were similar to those reported in other dementia samples (Gottlieb *et al.*, 1988). Twenty-two low B_{12} and 21 normal B_{12} subjects were considered ratable on the GDS. GDS scores consistent with probable depression (>5) were common in both low (10/22) and normal (6/21) B_{12} subjects.

Baseline hematologic findings

Baseline hematological characteristics of low B_{12} and normal B_{12} subjects are also displayed in Table 1. RBC folate levels were available for 24 low B_{12} and 24 normal B_{12} subjects (remaining subjects instead had serum folate levels). Low B_{12} subjects also had significantly lower RBC folate levels (364±157 ng/mL) than normal B_{12} subjects (524±161 ng/mL; t=3.48, df=46, p=0.001), although no folate level in either group was abnormal (RBC folate <200 μg/L or serum folate <3 μg/L). Low and normal B_{12} groups showed no significant differences in LDH, WBC, RBC, hemoglobin, hematocrit, MCV, or platelets. Seventeen low B_{12} subjects were anemic (hemoglobin <12.0 g/dL for females or <14.0 g/dL for males), compared to 13 normal B_{12} subjects. Only two low B_{12} subjects had a MCV greater than 100 fL (the same number of normal B_{12} subjects with MCV greater than 100 fL), consistent with previous reports (Thompson *et al.*, 1987; Lindenbaum *et al.*, 1988) that MCV is frequently normal in clinically important B_{12} deficiency.

Peripheral blood smears were available from 25 low B_{12} and 23 normal B_{12} subjects at screening. Hypersegmentation of neutrophils (1 six-lobed or >5 five-lobed granulocytes per 100 polymorphonuclear neutrophils) was noted on blinded review of smears from only 3/25 low B_{12} and 5/23 normal B_{12} subjects. Macroovalocytes were present in only 3/25 low B_{12} and 3/23 normal B_{12} subjects. Intrinsic factor antibodies were obtained in 17 low B_{12} subjects and were positive in only one case. Schilling tests were not performed in any subject. Twenty-two low B_{12} subjects had abnormally elevated MMA levels (>270 nmol/L), and 13 had abnormally elevated Hcys levels (>16.0 μmol/L) (Table 2).

Responses to B_{12} therapy

LABORATORY EFFECTS—The effect of IM B_{12} replacement (after 8 weeks of treatment) on pertinent laboratory values in low B_{12} subjects is displayed in Table 2. B_{12} replacement resulted in small but statistically significant increases in RBC number (t=2.26, df=27, p=0.032, paired t-test) and hemoglobin (t=2.13, p=0.043) as well as a decrease in MCV (t=2.36, p=0.025). Predicted sharp declines with treatment were seen in serum MMA (from 592±626 to 179±66 nmol/L; Z=−4.62, p <0.001, Wilcoxon matched-pairs test) and total Hcys (from 17.8±8.2 to 12.3±5.6 μmol/L; t=6.11, p<0.001). Whereas 22 low B_{12} subjects had abnormally elevated MMA levels (>270 nmol/L), and 13 had abnormally elevated Hcys levels (>16.0 μmol/L) pre-treatment, only 3 MMA levels and 7 Hcys levels remained elevated after 8 weeks of B_{12} replacement. Baseline MMA 0.30, (r=−0.26, n=28, p=0.18) and Hcys (r=0.07, p=0.71) levels and the degree of MMA (r=−0.30, p=0.12) and Hcys (r=−0.13, p=0.52) response with treatment were all uncorrelated with B_{12} levels.

Residual elevations in MMA and Hcys post-treatment appeared to be related in part to variations in renal function, as described in previous reports (Rasmussen *et al.*, 1990; Allen *et al.*, 1993; Savage *et al.*, 1994). Post-treatment Hcys levels were significantly correlated with blood urea nitrogen (BUN) (r=0.69, n=28, P <0.001) and serum creatinine (r=0.50, p=0.006). Post-treatment MMA levels were also significantly correlated with BUN (r=0.52, p=0.005) but not serum creatinine (r=0.31, p=0.11). Mean values of renal function tests for low B_{12} subjects were BUN=24±11 (range 12–57) mg/dL and serum creatinine=1.2±0.6 (range 0.7–3.8) mg/dL.

Cognitive and psychiatric effects—Outcome on cognitive and psychiatric rating scales through 16 weeks is displayed in Figures 1, 2 and 3. Two subjects (both normal B_{12}) did not complete 16-week ratings, and their data were included in the analysis using 8-week ratings, per the LOCF analysis plan. One of these subjects died suddenly of unknown cause, and the other suffered a major stroke. Although subjects were not blinded with regard to treatment, none of the low B_{12} subjects was aware of receiving B_{12} replacement at the end of 16-weeks, and only one subject recalled that she had received IM injections.

On the DRS (Figure 1) low B_{12} subjects improved by 4.4±17.6 points from baseline to week 16, and normal B_{12} subjects declined by −2.0±9.9 points. This difference trended in favor of treated low B_{12} subjects according to the planned ANCOVA (F=3.24; df=1,53; p=0.077, ANCOVA). However, change in DRS scores was not normally distributed (Lilliefors Test Statistic=0.16, df=56, p=0.002) was therefore re-analyzed post-hoc and by Mann-Whitney U test and found not to be significant (Z=−0.97, p=0.33, Mann-Whitney U test). On the BPRS (Figure 2) low B_{12} subjects (n=25) improved (declined) by −1.4±6.6 points, and normal B_{12} subjects (n=23) worsened (increased) by 0.5±5.6 points (F=0.12; df=1,45; p=0.73, ANCOVA). On the GDS (Figure 3) low B_{12} subjects (n=22) improved by 0.1±2.8 points, and normal B_{12} (n=21) subjects improved by points 0.3±2.5 (F=0.00; df=1,40; p=0.97, ANCOVA). Although baseline score was the only covariate specified in the statistical plan, none of these results was altered by the addition of age, sex, and RBC folate level to the ANCOVA models.

Post-hoc analyses were performed to relate evidence for tissue deficiency (MMA or Hcys) to cognitive response to B_{12} replacement. First, since normal MMA and Hcys levels may exclude clinical deficiency in subjects with low B_{12} levels, a secondary analysis was done, considering only those low B_{12} subjects with an elevation (2SD) in either MMA or Hcys (n=24). Using this more stringent criterion of deficiency, the magnitude of improvement on the DRS in low B_{12} subjects improved slightly (to 4.8±18.9 points), although the comparison to normal B_{12} subjects was still nonsignificant (F=2.45; df=1,49; p=0.12, ANCOVA) (Z=−0.94, p=0.35, Mann-Whitney U test). Second, when the relationship between change in cognition and metabolites was examined, the magnitude of improvement on the DRS paradoxically showed an inverse correlation with the magnitude of reduction in both MMA (ρ =−0.42, n=28, p=0.025) and Hcys (ρ =−0.51, p=0.006).

The number of subjects who showed a significant cognitive improvement (≥12 points on the DRS) during the study was numerically, but not statistically, greater for treated low B_{12} subjects (n=6) than comparison subjects (n=2) (χ^2 =1.31, df=1, p=0.25). Six treated low B_{12} subjects who met this criterion gained 71, 38, 21, 16, 15, and 13 points on the DRS, compared to two normal B_{12} subjects who gained 13 and 12 points on the DRS. The subject who gained 71 points was described at baseline as emotionally withdrawn with psychomotor retardation, although she was considered ratable on DRS. Her DRS improvement was accompanied by a drop (improvement) in BPRS score of 17 points. Similarly, the subject who gained 38 points on the DRS had a reduction (improvement) in GDS score of 6 points.

Therefore these largest DRS increases may have been related to improvement in noncognitive behavioral symptoms, whether or not these were B_{12} related.

Discussion

This study examined whether B_{12} replacement would ameliorate cognitive and psychiatric variables in elderly nursing home residents with dementia and low serum B_{12} levels. Low B_{12} subjects demonstrated tissue deficiency, as B_{12} replacement was associated with robust reductions in serum metabolites (MMA and Hcys) as well as small but statistically significant changes in hematologic parameters. However, B_{12} treatment failed to effect statistically significant improvements in cognitive (DRS) or psychiatric (BPRS, GDS) scores. Although a few subjects evidenced notable individual treatment responses, these were not statistically more frequent than in normal B_{12} subjects.

This study possessed significant limitations associated with the use of a comparison group of untreated subjects with normal B_{12} levels, rather than a randomized, placebo-controlled design. This virtual control group was successful in maintaining blinded ratings of outcome measures. And although dementia subjects themselves were not technically blinded with regard to treatment status (and no placebo injections were administered to normal B_{12} subjects), this likely had negligible effect, as none of the low B_{12} subjects recalled receiving B_{12} replacement at end of study. The major limitation of this design likely involved the inherent differences between subjects as a function of B_{12} status, despite careful demographic matching and post-hoc examination of other subject variables, which may have been associated with different outcome at 16 weeks.

The ethics of placebo controls remains a significant problem for the study of cognitive and psychiatric effects of B_{12} replacement in subjects with low serum B_{12} levels. Although placebo-controlled trials of B_{12} and other B-vitamins are widely accepted in dementia patients without evidence of deficiency, no consensus exists regarding the ethics of placebo controls in patients with low levels. As detailed in the following section, other investigators have handled this problem by conducting open-label studies (Martin *et al.*, 1992; Carmel *et al.*, 1995; Cunha *et al.*, 1995), sometimes with comparison to historical controls (Teunisse *et al.*, 1996; Eastley *et al.*, 2000) without low B_{12} levels, and only rarely with full placebo-controlled designs (de la Fournière *et al.*, 1997; Hvas *et al.*, 2004). In elderly samples with no dementia, placebo-controlled trials of B_{12} have sometimes handled the ethical dilemma by excluding subjects with any evidence of anemia or neurological symptoms (Hughes *et al.*, 1970; Seal *et al.*, 2002). However, this strategy is difficult to implement in practice, since minor hematological and neurological symptoms are prevalent in the elderly, and not necessarily related to low B_{12} levels. Furthermore, this practice may exclude the very subjects most likely to demonstrate behavioral improvement with treatment.

Previous treatment studies of low B_{12} in dementia

Apart from case reports, remarkably few studies have reported outcome data for treatment of dementia associated with low serum B_{12} levels. Martin *et al.* (1992) studied cognitive function before and after at least 6 months of open-label B_{12} treatment in 18 older subjects with low B_{12} levels (<150 pmol/L) and cognitive dysfunction. Eleven of 18 subjects improved on DRS scores, two subjects by 31 and 28 points. The only clinical characteristic found to predict improvement was short duration (<12 months) of cognitive dysfunction. Carmel *et al.* (1995) studied metabolic, neurologic, and electrophysiologic response to open-label B_{12} replacement in 16 patients with low B_{12} levels (<190 pg/mL), including 13 with dementia. They found that B_{12} improved several abnormalities but not cognitive function in the 13 patients with dementia. Cunha *et al.* (1995) investigated 19 patients with dementia and B_{12} deficiency (<200 pg/mL) and observed that 16 of 19 patients showed persistent

decline with B_{12} treatment at follow-up (3–24 months). All patients who demonstrated improvement on MMSE score had mild dementia with a history of less than two years.

Two previous studies have examined effects of open-label B_{12} treatment on neuropsychological function in dementia patients – with comparison to a reference group (Teunisse *et al.*, 1996) or matched controls (Eastley *et al.*, 2000) with dementia and normal B_{12} levels. Teunisse *et al.* (1996) gave open-label IM B_{12} replacement to 26 dementia patients with low B_{12} levels (<200 pg/mL). After 6 months of treatment they found no cases of dementia reversal and no difference in cognitive change scores in comparison to a reference group of 69 patients with Alzheimer's disease. Eastley *et al.* studied 66 patients who presented with dementia and 22 with cognitive impairment who were seen for a second assessment after treatment. Changes in neuropsychological test scores were compared with those of patients with normal serum B_{12}, matched by age and diagnosis. B_{12}-treated dementia patients showed no significant improvement and no less neuropsychological deterioration than their matched group. However, cognitively impaired patients improved significantly compared to matched patients on a verbal fluency test. In these two studies, psychometricians were not blinded to the B_{12} treatment status of patients.

Placebo-controlled trials of B_{12} replacement in dementia associated with low serum B_{12} levels are exceedingly few. In their Cochrane Review of all randomized, double-blind, placebo-controlled trials of B_{12} on cognitive function, Malouf and Areosa Sastre (2003) included only three trials (de la Fournière *et al.*, 1997; Seal *et al.*, 2002; Hvas *et al.*, 2004). de la Fournière *et al.* (1997) studied 11 subjects with a diagnosis of Alzheimer's disease with low serum B_{12} levels (<240 pg/ml). Subjects were randomly assigned to receive injections of B_{12} 1,000 μg or placebo daily for 5 days and then one injection monthly for 5 months. Treatment and placebo groups did not differ significantly in cognitive outcome as measured by ADAS-Cog scores. Seal *et al.* (2002) aimed to determine the minimum dose of oral B_{12} required to maintain normal serum levels. They studied 31 geriatric inpatients (one-third of whom had dementia) with low serum B_{12} levels (100–150 pmol/L) and no history of anemia or neurological disorder other than stroke. They compared treatment groups randomly assigned to receive 50 μg or 10 μg of oral B_{12} or placebo daily for one month and found that only the 50 μg dose produced a significant increase in serum B_{12}. No significant changes were observed in MMSE scores. Hvas *et al.* (2004) examined effects of B_{12} treatment (1,000 μg IM weekly for 4 weeks) on cognitive function and depression in a randomized placebo-controlled study of 140 subjects with elevated serum MMA (0.40–2.00 mmol/l). At baseline 40 (29%) subjects had cognitive impairment (MMSE <25), and 18 (13%) had symptoms of depression. At 3-month follow-up, no improvement was found in cognitive or depression scores. The available randomized clinical trials of B_{12} in dementia samples are thus remarkably limited, with only one small dedicated study (de la Fournière *et al.*, 1997) and another study with a large subsample with dementia (Hvas *et al.*, 2004). We would concur with the conclusions of Malouf and Areosa Sastre (2003) that these trials do not provide statistically significant evidence of treatment effect for B_{12} supplementation compared with placebo, on cognitive function.

Limitations of present study

In addition to the absence of a placebo-controlled design, this study had a number of additional limitations that warrant comment. First, the cutoff selected for B_{12} deficiency (<250 pg/mL) was somewhat higher than in most other studies. A lower cutoff may have yielded a greater degree of tissue deficiency and more significant effects of B_{12} replacement. Nonetheless, when a more restrictive definition of tissue deficiency was used – i.e. when only subjects with an elevation (2SD) in either MMA or Hcys were included – the results were still nonsignificant. Second, the follow-up period (16 weeks) may have been too brief to observe full effects of B_{12} replacement on cognitive and psychiatric variables. The limited

hematological response (in hemoglobin and MCV) in the treated low B_{12} subjects may be due in part to the still shorter laboratory retest period (8 weeks, as compared to average RBC life span of 120 days). However, we were concerned that a longer follow-up period would result in more study non-completers and more confounding intercurrent illnesses in this fragile population. Third, the long duration of dementia in this nursing home population may have mitigated against observing cognitive and behavioral effects of B_{12} replacement. Some previous investigators have reported a time dependency of cognitive recovery with B_{12} replacement (Martin *et al.*, 1992; Cunha *et al.*, 1995). Martin *et al.* observed that only patients symptomatic for less than a year showed improvement with therapy (Martin *et al.*, 1992), whereas Cunha *et al.* (Cunha *et al.*, 1995) reported improvement only in patients with mild dementia of less than two years duration. Our low B_{12} sample contained only nine subjects with mild dementia (MMSE ≥20) and only six with duration ≤2years. The six treatment responders had a mean MMSE score of 10.5 and mean duration of 3.3 years and thus did not differ markedly from the overall sample. Nonetheless, the preponderance of studies in this field would suggest that future research of cognitive and psychiatric effects of B_{12} should focus on patients with mild symptoms of short duration.

Conclusion

These results suggest that B_{12} replacement is unlikely to benefit cognitive or psychiatric symptoms in the vast majority of elderly dementia patients with low serum B_{12} levels. However, future research is warranted for patients with mild symptoms of short duration.

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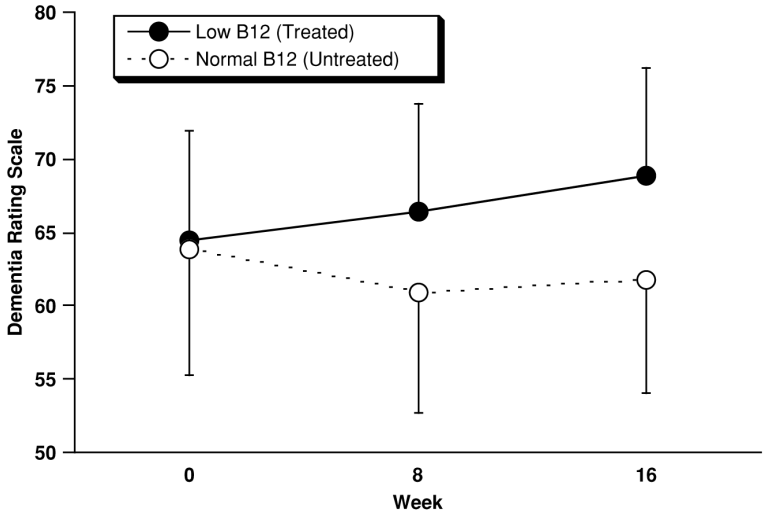

Figure 1.

Outcome on the Dementia Rating Scale (DRS) for low B_{12} (n=28) and normal B_{12} (n=28) subjects. Displayed values represent mean±SEM (SEM used for legibility). From 0 to 16 weeks low B_{12} subjects improved by 4.4±17.6 (SD) points, and normal B_{12} subjects declined by −2.0±9.9 points. However, this difference was not significant (F=3.24; df=1,53; p=0.077, ANCOVA) (Z=−0.97, p=0.33, Mann-Whitney U test).

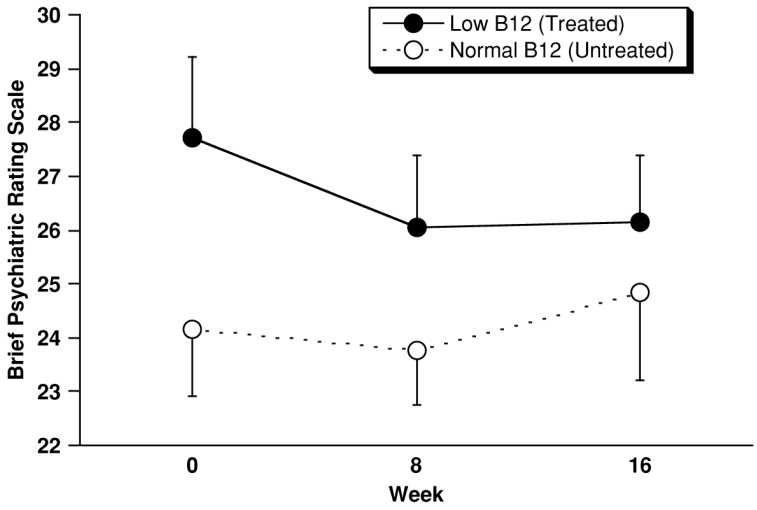

Figure 2.

Outcome on the Brief Psychiatric Rating Scale (BPRS) for low B_{12} (n=25) and normal B_{12} (n=23) subjects. Displayed values represent mean±SEM (SEM used for legibility). From 0 to 16 weeks low B_{12} subjects improved (declined) by −1.4±6.6 (SD) points, and normal B_{12} subjects worsened (increased) by 0.5±5.6 points (F=0.12; df=1,45; p=0.73, ANCOVA).

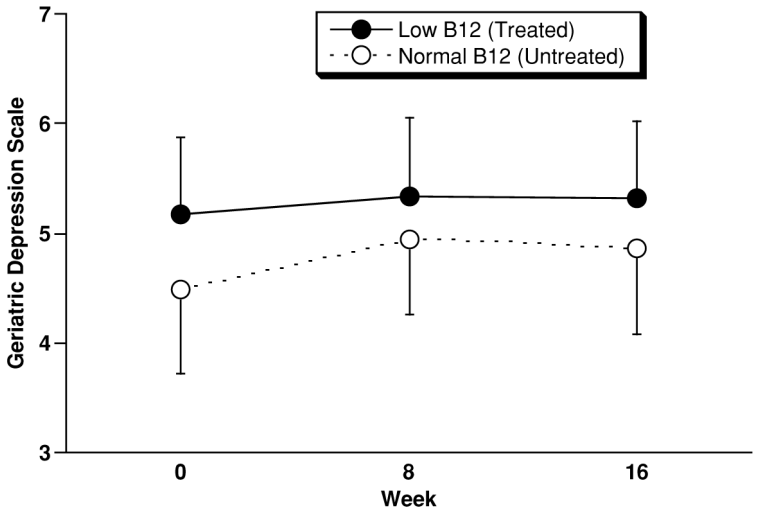

Figure 3.
Outcome on the Geriatric Depression Scale – abbreviated version (GDS) for low B_{12} (n=22) and normal B_{12} (n=21) subjects. Displayed values represent mean±SEM (SEM used for legibility). From 0 to 16 weeks low B_{12} subjects improved by 0.1±2.8 (SD) points, and normal B_{12} subjects improved by 0.3±2.5 points (F=0.00; df=1,40; p=0.97, ANCOVA).

Table 1

Subject characteristics

	LOW B_{12} (N = 28)			NORMAL B_{12} (N = 28)		
VARIABLE	**MEAN**		**SD**	**MEAN**		**SD**
Demographics						
Age	86.8	±	6.9	87.0	±	5.8
Gender		25F, 3M			25F, 3M	
Education (years)	10.7	±	3.1	10.7	±	2.8
Dementia Characteristics						
Duration (years)	4.8	±	3.3	5.5	±	4.2
Hachinski Score	3.9	±	3.0	3.9	±	2.9
Neuropsychological						
MMSE	11.9	±	8.9	11.6	±	8.0
DRS	64.4	±	39.6	63.8	±	45.4
GDS	5.2	±	3.3	4.5	±	3.5
BPRS	27.7	±	7.6	24.4	±	5.9
Hematological						
Serum B_{12} (pg/mL)	186	±	43*	521	±	226
RBC Folate (ng/mL)	364	±	157†	524	±	161
Serum LDH (U/L)	148	±	38	165	±	39
WBC (×1000/mm^3)	6.8	±	1.8	7.4	±	2.1
RBC (×1000/mm^3)	4.0	±	0.4	4.3	±	1.2
Hemoglobin (g/dL)	12.1	±	1.4	12.4	±	1.5
Hematocrit (%)	36.3	±	4.4	37.2	±	4.7
MCV (fL)	91.9	±	5.5	91.9	±	6.3
Platelets (×1000/mm^3)	255	±	56	258		90

Note: MMSE, Mini-Mental State Examination; DRS, Dementia Rating Scale; GDS, Geriatric Depression Scale; BPRS, Brief Psychiatric Rating Scale; LDH, lactate dehydrogenase; MCV; erythrocyte mean corpuscular volume.

*Differs from Normal B_{12} group, P<0.001, t-test;

†Differs from Normal B_{12} group, p=0.001, t-test. RBC folate levels were available for 24 subjects in each group.

Table 2

Effect of B_{12} Replacement on Laboratory Variables in Low B_{12} Dementia Subjects

	BASELINE			8 WEEKS			
	MEAN		SD	MEAN		SD	P
Methylmalonic Acid (nmol/L)	592	±	626	179	±	66	<0.001
Homocysteine (μmol/L)	17.8	±	8.2	12.3	±	5.6	<0.001
Serum LDH(U/L)	148	±	38	145	±	46	0.514
WBC (×1000/mm^3)	6.82	±	1.79	6.98	±	1.75	0.625
RBC (×1000/mm^3)	3.95	±	0.45	4.09	±	0.51	0.032
Hemoglobin (g/dL)	12.1	±	1.4	12.5	±	1.7	0.043
Hematocrit (%)	36.3	±	4.4	37.1	±	5.0	0.148
MCV (fL)	91.9	±	5.5	90.6	±	4.7	0.035
Platelets(×1000/mm^3)	255	±	56	261	±	58	0.569

Note: LDH, lactate dehydrogenase; WBC, white blood cell count; RBC, red blood cell count; MCV, erythrocyte mean corpuscular volume.

P-values are from paired t-tests for all variables except Methylmalonic Acid, which is from Wilcoxon matched-pairs test.