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Baseline Vitamin B12 and Folate Levels Do Not Predict Improvement in Depression After a Single Infusion of Ketamine

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Abstract

Introduction—Deficiencies in both vitamin B12 and folate have been associated with depression. Recently, higher baseline vitamin B12 levels were observed in individuals with bipolar depression who responded to the antidepressant ketamine at 7 days post-infusion. This study sought to replicate this result by correlating peripheral vitamin levels with ketamine's antidepressant efficacy in bipolar depression and major depressive disorder (MDD).

Methods—Baseline vitamin B12 and folate levels were obtained in 49 inpatients with treatment-resistant MDD and 34 inpatients with treatment-resistant bipolar depression currently experiencing a major depressive episode. All subjects received a single intravenous ketamine infusion. Post-hoc Pearson correlations were performed between baseline vitamin B12 and folate levels, as well as antidepressant response assessed by percent change in Hamilton Depression Rating Scale (HDRS) scores from baseline to 230 min, 1 day, and 7 days post-infusion.

Results—No significant correlation was observed between baseline vitamin B12 or folate and percent change in HDRS for any of the 3 time points in either MDD or bipolar depression.

Discussion—Ketamine's antidepressant efficacy may occur independently of baseline peripheral vitamin levels.

Keywords

vitamin B12; folate; ketamine; bipolar depression; major depressive disorder

Introduction

Ketamine is an N-methyl-D-aspartate (NMDA) receptor antagonist. Studies have found that a single administration of ketamine at subanesthetic doses has rapid-acting antidepressant and anxiolytic effects that occur within hours and persist for days (for a review, see [1]). Response rates to ketamine (defined as a 50 % reduction on a standard depression

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Conflict of Interest

All other authors report no biomedical financial interests or other potential conflicts of interest.

measure) at 72 h post-infusion range from 14–70 % for individuals with major depressive disorder (MDD) and bipolar depression [2]. In bipolar depression, antidepressant effects have been observed as soon as the end of ketamine infusion [3]. In MDD, decreased suicidal ideation has also been reported within 40 min and has persisted for up to 4 h [4]. More persistent antidepressant effects have also been observed with repeated infusions of ketamine at subanesthetic doses [5]. Vitamin B12 and folate are critical enzymatic cofactors that reduce levels of the pro-oxidant homocysteine in the S-adenosylmethionine cycle [6]. Both vitamins are necessary for neurological functioning, and deficiencies have been associated with MDD [7,8]. In outpatients, higher baseline vitamin B12 levels were found to predict antidepressant response to monoaminergic anti-depressants [9]. In the June 2013 issue of *Pharmacopsychiatry*, Permoda-Osip and colleagues [10] reported that baseline serum vitamin B12 levels positively predicted antidepressant response to ketamine at 7 days post-infusion in patients with bipolar disorder currently experiencing a depressive episode. In contrast, folate, homocysteine, and various other factors did not differ significantly between ketamine responders and non-responders. The authors postulated that exogenous vitamin B12 supplementation might be an effective augmentation and/or maintenance strategy for post-antidepressant response to ketamine.

Because the sample size in the study by Permoda-Osip and colleagues [10] was relatively small (10 responders and 10 non-responders), we sought to replicate these results in a larger independent sample to further assess whether baseline vitamin B12 and/or folate levels predicted antidepressant response to ketamine. We performed a post-hoc analysis of baseline vitamin levels and percent change in Hamilton Depression Rating Scale (HDRS) scores in 34 inpatients with treatment-resistant bipolar depression and 49 inpatients with treatment-resistant MDD; all subjects received a single ketamine infusion at subanesthetic doses. Many patients who respond to ketamine do so at one day post-infusion, and the third day post-infusion is associated with high relapse rates [2]. Thus, we selected the time points of 230 min, 1 day, and 7 days post-infusion to best contrast the maximal acute (230 min and day 1) and minimal sustained (day 7) antidepressant effects of ketamine. It should also be noted that our laboratory has previously used these time points in similar post-hoc correlational analyses [11, 12].

Methods

All patients (n = 83) were admitted to our inpatient Mood and Anxiety Disorders Research Unit at the National Institute of Mental Health (Bethesda, MD, USA). Patients were recruited through advertisements in local newspapers and public transportation, the Internet, and referrals from local and nationwide physicians and psychiatric inpatient units. All subjects provided informed consent prior to entering the study, and all protocols were approved by the Combined Neuroscience Institutional Review Board of the National Institutes of Health for the duration of the study.

All patients with MDD or bipolar depression were 18–65 years of age, currently experiencing a major depressive episode without psychotic features that had lasted at least 4 weeks prior to hospitalization. Subjects had been hospitalized on average for 6 weeks prior to the first ketamine infusion, received a standard hospital diet, and took no vitamin

supplements during the study. All Axis I diagnoses were confirmed by Structured Clinical Interview for DSM-IV Disorders (SCID): Patient Edition [13]. Subjects were deemed to be in good physical health as determined by hospital records, physical examination, blood and urine laboratory tests, chest x-rays, and electrocardiogram. Subjects were excluded if they had comorbid substance abuse or dependence (excluding caffeine or nicotine) within 3 months of enrollment in the study, had a positive urine toxicology screen, a serious unstable medical disorder or condition, or if they had a current or past diagnosis of schizophrenia or any other psychotic disorder according to the SCID and/or clinical history. Female subjects could not be nursing or pregnant. All patients were free of psychotropic medications in the 2 weeks prior to ketamine infusion (5 weeks for fluoxetine) aside from therapeutic dose lithium or valproate for those with bipolar depression. 3 groups of depressed patients were studied. The first group, comprising 34 patients with bipolar I ($n = 19$) or II ($n = 15$) depression, was collated from our double-blind, placebo-controlled, cross-over, add-on studies (13 males, 21 females; 23–62 (mean $\pm$ SD = 46 ± 11) years old) [3, 14]. These patients were experiencing a current major depressive episode of at least moderate severity (Montgomery Asberg Depression Rating Scale (MADRS) total score ≥ 20) lasting 18 ± 20 months; mean duration of illness was 29 ± 11 years. 22 patients were maintained on therapeutic doses of lithium, and 12 received a therapeutic dose of valproate; the patients received no other psychotropic medications throughout the study. Treatment-resistance was confirmed by history of non-response to at least one adequate dose/duration antidepressant trial and at least one adequate mood stabilizer (lithium or valproate) trial during the current depressive episode, as assessed via the Antidepressant Treatment History Form (ATHF) [15].

The second and third groups comprised 49 unmedicated patients with active, treatment-resistant MDD (26 males, 23 females; 19–65 (mean $\pm$ SD = 48 ± 12) years old). 20 of these subjects participated in a double-blind, placebo-controlled, cross-over study and were required to have a score ≥ 18 on the 21-item HDRS at screening and at the start of each infusion. The remaining 29 participated in an open-label ketamine study and were required to have a score ≥ 22 on the MADRS at screening and on the day of ketamine infusion, with no greater than a 25 % decrease in total MADRS score between screening and infusion. All MDD patients were experiencing a current major depressive episode lasting on average 73 ± 116 months; mean duration of illness was 25 ± 14 years. Treatment-resistance was confirmed by history of non-response to at least 2 adequate antidepressant trials, as assessed via the ATHF [15].

Baseline vitamin B12 and folate levels were assessed during the screening phase. The baseline HDRS was administered 1 h before patients received a single subanesthetic dose (0.5 mg/kg) ketamine infusion over 40 min. All subjects were monitored for the next 7 days with a battery of neuropsychiatric ratings as previously described [3, 14]. To mirror the analysis used by Permoda-Osip and colleagues [10], we classified responders as those with a ≥ 50 % decrease from baseline HDRS at 7 days post-infusion and nonresponders as those with < 50 % decrease at 7 days post-infusion. Vitamin B12 and folate levels in our patient sample were not normally distributed; therefore, the data were natural log-transformed. We performed an independent samples t -test to compare the means of log-transformed baseline

vitamin B12 levels between responders and non-responders. To further investigate whether baseline levels were associated with antidepressant efficacy, we performed Pearson correlations between log-transformed baseline vitamin B12 and folate levels and percent change from baseline HDRS at 230 min, 1 day, and 7 days post-ketamine infusion.

Results

Due to the non-normal distribution of both vitamin B12 and folate in the MDD and bipolar depression patient groups, we first calculated the natural log-transformed means. We then calculated the geometric means in order to revert the values back to the original, more standard, scale. Log-transformed mean baseline vitamin B12 levels were 6.3 ± 0.44 for the bipolar group and 6.5 ± 0.40 for the MDD group; log-transformed mean baseline folate levels were 2.6 ± 0.39 for the bipolar group and 2.8 ± 0.46 for the MDD group. Geometric mean baseline vitamin B12 levels were 551 in the bipolar group and 648 in the MDD group; mean folate levels were 14.1 in the bipolar group and 17.0 in the MDD group. While these values are higher than those reported by Permoda-Osip and colleagues [10], the values in both studies are in the normal range (193–982 pg/mL for vitamin B12 and 3.0– 17.0 ng/mL for folate).

Unlike the patient sample in the study by Permoda-Osip and colleagues [10], which maintained a large antidepressant effect at one week post-infusion, in the present study only 7 % of the bipolar depression group and 24 % of the MDD group remained responders at day 7. The small number of responders vs. nonresponders at day 7 did not permit reliable statistics for dichotomous analysis. Response rates for the bipolar group were 38 % at 230 min and 33 % at day 1. Response rates for the MDD group were 40 % at 230 min and 39 % at day 1. While response rates were higher at the 2 earlier time points, we found no significant differences in either diagnostic category for vitamin B12 or folate. P-values from independent samples *t*-tests with B12 levels for responders vs. non-responders were as follows for bipolar depression and MDD, respectively: $p = 0.59$ at 230 min and $p = 0.73$ at day 1; $p = 0.51$ at 230 min and $p = 0.70$ at day 1. In a similar responders vs. non-responders analysis for folate, p-values for independent samples *t*-tests were as follows for bipolar depression and MDD, respectively: $p = 0.24$ at 230 min and $p = 0.59$ at day 1; $p = 0.54$ at 230 min and $p = 0.30$ at day 1.

To take advantage of the entire dataset, we performed Pearson correlations between vitamin B12 levels and HDRS percent change at 230 min, 1 day, and 7 days post-ketamine infusion. In patients with bipolar depression, baseline vitamin B12 levels did not correlate with HDRS change from baseline at 230 min, day 1, or day 7 ($\triangleright$ **Table 1**). Similarly, these correlations were also not significant in patients with MDD at 230 min, day 1, or day 7. We further examined the relationship between baseline folate levels and antidepressant response to ketamine. In patients with bipolar depression, there was again no relationship between baseline folate levels and percent change in HDRS score at 230 min, day 1, or day 7. In patients with MDD, baseline folate levels and change in HDRS score were similarly not significant at 230 min or day 1. A significant correlation was noted at day 7 ($r = 0.35$, $p = 0.02$), but this did not survive Hochberg's correction for multiple comparisons [16]. To display the individual data points and assess potential outliers, scatterplots of the log-

transformed baseline B12 levels at the 3 post-infusion time points are presented in ► **Fig. 1**. All B12 and folate analyses were also performed on the non-normally distributed raw data, but this did not affect the results.

Finally, to ensure that the difference in MDD study design (double-blind, placebo-controlled, cross-over [17] vs. open-label [18]) did not affect our results, we correlated HDRS percent change and vitamin levels for all 3 time points across the 2 study designs. We then performed Fisher r-to-z-transformations [19] with the correlation coefficients and found that none of the correlation coefficients from the open-label study significantly differed from those of the randomized controlled trial at any time point for either vitamin B12 or folate, confirming that the differences in MDD study design did not alter our major outcomes.

Discussion

In our sample of 83 treatment-resistant, currently depressed patients with either bipolar depression ($n = 34$) or MDD ($n = 49$), baseline vitamin B12 and folate levels did not significantly predict antidepressant response to a single subanesthetic dose of ketamine.

Several possible explanations exist for the discrepancy with prior results. First, our response rate was significantly lower than that of Permoda-Osip and colleagues [10], who observed a 50% response rate at 7 days post-ketamine infusion. While previous ketamine studies (including some that overlap with the present sample) have shown larger response rates at day 7 post-infusion – 35 % in MDD [17], 26 % in bipolar depression [3], and 43 % in MDD [20] – some studies used the MADRS to assess response [3, 20] while here we used the HDRS. This could lead to potential measure-specific response rate differences. In addition, a recently published MDD randomized controlled trial of ketamine by Murrough and colleagues [20] used an active placebo (midazolam) rather than the normal saline solution used in our studies. While there was a 43 % response rate at day 7 post-ketamine in their study, this did not significantly differ from the midazolam group, suggesting a large placebo response. Next, in the study by Permoda-Osip and colleagues [10], patients with bipolar depression were receiving multiple first- or second-generation mood stabilizers (including atypical antipsychotics); in contrast, our patients with bipolar depression received only lithium or valproate. These multiple adjunctive psychotropic medications could have potentially augmented ketamine's antidepressant effects. Second, as mentioned above, patients with bipolar depression in the study by Permoda-Osip and colleagues [10] experienced a response rate of 50% at 7 days post-infusion; this is significantly greater than the day 7 response rate observed in our previous randomized controlled studies of ketamine in bipolar depression [3, 14], and suggests that our sample may be more severely ill and/or treatment-resistant. Another interesting difference was a disproportionate sex distribution; their sample was 90% female and ours was 62% female. One study found that sex did not significantly predict antidepressant response to ketamine in a total sample of 108 treatment-resistant depressed inpatients [11]. However, other studies found that antidepressant response to both serotonergic antidepressants [21 – 23] and the muscarinic antagonist scopolamine [24] was greater in females. Therefore, the preponderance of females in the study by Permoda-Osip and colleagues [10] may have contributed to the disproportionate response rate seen at 7 days post-infusion and, hence, to the baseline vitamin B12 predictor

difference. Finally, it should be noted that while baseline peripheral vitamin levels have been reported to predict antidepressant response to tricyclic antidepressants (TCAs) as well as selective serotonin reuptake inhibitors (SSRIs) [9], they may not predict response to ketamine, which is hypothesized to primarily affect glutamatergic neurotransmission [25].

In conclusion, the conflicting results between our analysis and that of Permoda-Osip and colleagues [10] suggest that larger sample sizes will be necessary to better assess these pharmacoepidemiological relationships in active depression.

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Fig. 1.

Baseline B12 levels after a natural log transformation and percent change in Hamilton no period. Depression Rating Scale (HDRS) scores from baseline to 3 time points after a subanesthetic infusion of ketamine in patients with bipolar depression and major depressive disorder.

Table 1

Pearson correlations between baseline serum vitamin levels and percent Hamilton Depression Rating Scale (HDRS) change from baseline after a single subanesthetic ketamine infusion at 3 time points in patients with active bipolar depression and major depressive disorder.

		230 Min		Day 1		Day 7	
		r	p	r	p	r	p
Bipolar Depression	Vitamin B12	0.05	0.79	−0.002	0.99	0.19	0.33
	Folate	−0.20	0.27	−0.13	0.49	−0.28	0.14
Major Depressive Disorder	Vitamin B12	0.14	0.36	0.14	0.36	−0.01	0.94
	Folate	0.23	0.12	0.18	0.21	0.35	0.02

Vitamin B12 and folate underwent natural log transformations to account for non-normal distributions