B-Vitamins Reduce the Long-Term Risk of Depression After Stroke: The VITATOPS-DEP Trial

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Objective: The consumption of certain B-vitamins through diet or supplementation decreases the total plasma concentration of homocysteine (tHcy) and may enhance response to standard antidepressant treatment. It is unclear if treatment with B-vitamins can reduce the long-term prevalence of depression in people at risk, such as stroke survivors. The purpose of this research was to determine if treatment with B-vitamins reduces the hazard of poststroke depression compared with placebo.

Methods: Randomized, double-blind, placebo-controlled trial of tHcy-lowering treatment with daily folic acid (2 mg), vitamin B6 (25 mg), and vitamin B12 (0.5 mg) for 1 to 10.5 years in survivors of stroke. The primary endpoint was the onset of Diagnostic and Statistical Manual of Mental Disorders, 4th edition (DSM-IV) major depression after randomization. Secondary outcomes were the prevalence of DSM-IV major or minor depression at the end of treatment. Other measured factors included age, gender, poststroke handicap associated with stroke, recurrence of strokes, cognitive impairment, and use of antidepressants.

Results: Among 273 people who completed the final assessment after 7.1 ± 2.1 years (mean ± standard deviation) of follow up, random assignment to B-vitamins was associated with a lower hazard of major depression compared with placebo (18.4% vs 23.3%, adjusted hazard ratio [HR] = 0.48; 95% confidence interval [CI] = 0.31–0.76) and a trend toward a lower odds of major or minor depression at the end of the trial compared with placebo (19.1% vs 27.7%; adjusted odds ratio [OR] = 0.58; 95%CI = 0.31–1.09).

Interpretation: Long-term treatment of poststroke survivors with folic acid, B6, and B12 was associated with a reduction in the hazard of major depression in our patient population. If these findings can be validated externally, B-vitamin supplementation offers hope as an effective, safe, and affordable intervention to reduce the burden of poststroke depression.

Depression affects 1 in every 3 stroke survivors,1 and represents an added source of morbidity, functional impairment, and mortality in this population.2,3 Attempts to prevent the onset of depressive symptoms after stroke through the prophylactic use of antidepressants have been largely unsuccessful,4–7 which, together with the increased risk of adverse events associated with the regular use of these medications,8,9 highlights the need for the introduction of more efficacious and safer interventions.

Stroke and depression share numerous risk factors,10,11 and some, like high total plasma homocysteine (tHcy),12,13 are potentially modifiable. Treatment with B-group vitamins (folic acid, B6, and B12) reduces tHcy by about 20%14,15 and may reduce the relative risk of stroke by 18% (95% confidence interval [CI] = 0.58; 95%CI = 0.31–1.09).
0.0–32%). In addition, existing evidence suggests that the consumption of these vitamins through diet or supplementation may improve response to standard antidepressant treatment and reduce the prevalence of depression, although conclusive data from randomized trials is still lacking. The possible antidepressant effect of these vitamins has been attributed to the facilitation of methylation reactions that ultimately lead to the synthesis of neurotransmitters (such as noradrenaline, dopamine, and serotonin) or, alternatively, to a decrease in cerebrovascular burden, as postulated by the vascular hypothesis of depression. Finally, it is not known whether lowering tHcy can prevent the onset of depressive symptoms in people at risk, such as stroke survivors.

We designed the present study to determine if treatment with folic acid and vitamins B6 and B12 would reduce the incidence and prevalence of depression among stroke survivors living in Perth, Australia. We hypothesized that participants randomly allocated to treatment with vitamins would have lower risk of developing major depression during long-term follow-up than participants treated with placebo.

Patients and Methods

Participants and Setting

The present study (VITATOPS-DEP) is an add-on substudy of the VITamins TO Prevent Stroke (VITATOPS) trial, which is a large multicenter, double-blind, placebo-controlled randomized trial that was designed to test if B-vitamin therapy (folic acid 2 mg, vitamin B6 25 mg, and vitamin B12 0.5 mg) reduces the risk of major cardiovascular events in people who have had a recent stroke or transient ischemic attack (TIA). Details about the design of VITATOPS have been described elsewhere.

We included in VITATOPS-DEP patients presenting within 7 months of a stroke (ischemic or hemorrhagic) or TIA (eye or brain) to Royal Perth, Sir Charles Gairdner, and Fremantle Hospitals in Western Australia. The diagnosis of stroke and TIA followed the guidelines of the International Classification of Diseases, 10th revision (World Health Organization, Geneva, Switzerland). We excluded people who had been prescribed folic acid, vitamin B6, or methotrexate, who were pregnant or of child-bearing age, or who had a chronic terminal illness such as cancer. VITATOPS-DEP participants did not fulfill criteria for the diagnosis of a major depressive episode at the time of randomization (retrospective assessment).

The Human Research Ethics Committees of the Royal Perth, Sir Charles Gairdner, and Fremantle Hospitals approved the study, and all participants provided written informed consent.

Baseline Characteristics

At baseline, the following data were recorded: date of birth, date of assessment, gender, ethnic background, smoking status, medical history of diabetes, hypertension, and dyslipidemia, stroke classification, handicap associated with stroke, and use of antidepressants at the time of randomization. We did not collect information regarding prior use of antidepressants and did not record the indications for antidepressant use at the time of randomization.

Randomization

Between November 1998 and December 2008, participants were randomly assigned to treatment with B-vitamins or placebo by means of a central 24-hour telephone service or interactive website (http://www.health.wa.gov.au/VITATOPS). The treatment allocation was derived from a list of computer-generated random numbers in permuted blocks stratified by hospital.

Intervention and Blinding (Exposure)

Eligible and consenting participants were randomly allocated to treatment with B-vitamins (2 mg of folic acid, 25 mg of vitamin B6, and 0.5 mg of vitamin B12) or placebo in a double-blind fashion. Placebo and active tablets had the same shape, size, weight, color, and smell. They were administered orally or, if necessary, in crushed form via a nasogastric feeding tube. Participants were advised to take 1 study tablet every morning after breakfast. Participants and investigators remained blind to treatment allocation for the duration of the study.

Procedures for the Collection of Endpoints of Interest

Follow-up in the VITATOPS trial was scheduled for every 6 months after randomization until completion of the trial on June 30, 2009. For the final VITATOPS trial follow-up visit, we invited participants to undergo an additional assessment for the purposes of this substudy.

Participants who consented to the additional assessment were enrolled in the VITATOPS-DEP substudy and assessed by means of the Mini-International Neuropsychiatric Interview (MINI) to determine the presence of current minor or major depressive episode and past major depressive episode according to Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition (DSM-IV) criteria (American Psychiatric Association, Washington, DC, 1994). The MINI (2006 version) is a structured neuropsychiatric interview that collects detailed information about the presence of current and past depressive symptoms. In the case of major depression, we asked participants to report the total number of episodes they had experienced since randomization, and the approximate date of onset and recovery of each episode. As the MINI does not record information relating to past episodes of minor depression, we were unable to determine whether minor depression had occurred between randomization and the VITATOPS-DEP assessment.

In addition, we asked participants to record the average number of days per week that they had consumed the study tablets, whether they had had a recurrence of stroke or TIA since they started taking vitamins/placebo, and whether they had used antidepressants between randomization and the VITATOPS-DEP assessment. We provided participants with a list of all antidepressants available in the Australian market, including both generic and trade names to facilitate identification of such a therapy.
Cognitive function was rated with the Mini-Mental State Examination (MMSE) and participants with total scores <24 were considered to have cognitive impairment.

We measured tHcy according to standard procedures in a subset of consenting participants at enrolment and the end of the trial (86 and 90 participants assigned placebo and vitamins, respectively).

**Outcomes**

The primary endpoint of interest for VITATOPS-DEP was the development of a major depressive episode from randomization until the end of follow-up (we did not collect information from participants about episodes of depression occurring before randomization). Secondary outcomes were prevalent depression at the end of the trial (major, minor, and both), and changes in tHcy.

**Other Measured Factors**

We collected data on severity of handicap associated with the primary stroke, recurrence of strokes during the trial, presence of cognitive impairment at the last visit, and use of antidepressants during the trial.

**Statistical Methods**

We used the statistical package Stata release 11.0 (StataCorp, College Station, TX) to manage and analyze the data. The results were summarized as mean ± standard deviation (SD) or proportions. We compared the distribution of the relevant variables according to treatment group using Student t tests and the Pearson chi-square test, as appropriate. We then calculated the unadjusted odds of depression according to treatment allocation and adjusted the analyses for known confounders and effect modifiers using logistic regression. In addition, we used Cox proportional regression to investigate the hazard of major depression during the trial according to treatment allocation. This model was adjusted for age at the time of randomization, gender, use of antidepressant medications, post-stroke handicap, recurrence of stroke, and presence of cognitive impairment. Time-varying covariates (eg, recurrence of stroke) were entered in the Cox regression models as time-independent variables that either had occurred or not. Finally, we used imputation by chained equations (ICE) to complete an intention-to-treat (ITT) analysis of the outcomes of interest. As we did not have access to the dates of loss to follow up of participants, ICE was also used to estimate time-to-event, as described by others. We created multiple imputation files based on the following variables: use of antidepressants (baseline or during the trial), age at randomization, gender, poststroke handicap, recurrence of stroke, and presence of cognitive impairment. We generated 5 imputed datasets to achieve stable estimates and 95% CIs.

**Results**

A total of 563 patients were randomly allocated to treatment with placebo (n = 279) or B-vitamins (n = 284) between November 1998 and March 2008. Figure 1 shows the flow of participants during the trial.

Between randomization and the final visit, 157 people (27.9%) died and a further 133 (23.6%) were lost to follow up. The remaining 273 (48.5%) participants completed the last assessment, when the endpoints for VITATOPS-DEP were collected. The demographic and clinical characteristics at the time of randomization of participants included and lost to the study are summarized in Table 1, whereas Table 2 shows the characteristics of 273 VITATOPS-DEP participants according to their treatment allocation. The groups were well balanced for age, gender, ethnic background, smoking status, clinical history of diabetes, hypertension and high cholesterol, use of antidepressants, type and severity of stroke, biochemical parameters, treatment allocation, and duration of follow-up (mean 6.9 years placebo vs 7.2 years B-vitamins, p = 0.30).

**Other Measured Factors**

Participation in the trial ranged from 1 to 10.5 years and compliance was about 80% for both treatment groups (see Table 2). There were no differences between the groups regarding stroke recurrence, poststroke handicap, and antidepressant use.

**Primary Outcome**

A major depressive episode occurred during the trial in 25 (18.4%) patients assigned B-vitamins and 32 (23.3%) patients assigned placebo; adjusted hazard ratio (HR) = 0.48, 95% CI = 0.27–0.86). We found no evidence of interaction between antidepressant and B-vitamin use on the occurrence of depression during the trial (HR = 0.82; 95% CI = 0.25–2.68). Because we observed a
nonsignificant excess of stroke recurrences among people treated with placebo, we repeated the Cox regression analysis excluding participants who had experienced a recurrence of stroke during the trial—the results remained unchanged (HR = 0.43; 95%CI = 0.22–0.81). ITT analysis using multiple imputation for participants lost to follow-up yielded similar results (Fig 2). The annual rate ratio of depression was 34 (95%CI = 25–50) and 25 (95%CI = 17–38) cases per 1,000 person-years for participants assigned placebo and vitamins, respectively.

**Secondary Outcomes**

At the final follow-up assessment, neither major nor minor depression were significantly more prevalent among people assigned placebo than B-vitamins (Table 3), although the distribution of people with major and minor depression were consistent with the results obtained for the primary outcome (27.7% vs 19.1%, respectively; \( \chi^2 = 2.83, p = 0.093 \)). Logistic regression adjusted for other measured factors (age, gender, use of antidepressants, handicap, recurrence of strokes, and cognitive impairment) showed that treatment with B-vitamins decreases the odds of depression (odds ratio [OR] = 0.48; 95%CI = 0.25–0.91), although this effect did not reach statistical significance in the ITT analysis (OR = 0.58; 95%CI = 0.31–1.09) (see Table 3).

A total of 86 and 90 people treated with placebo and vitamins, respectively, consented to donate a blood sample for the measurement of tHcy at the end of the trial. Treatment with vitamins was associated with a 28% reduction in tHcy compared with placebo.

**Discussion**

The results of this trial suggest treatment with 25 mg of vitamin B6, 0.5 mg of vitamin B12, and 2 mg of folic acid reduces the hazard of a major depressive episode...
compared with placebo among survivors of a stroke or TIA. B-group vitamin treatment was also associated with a nonsignificant trend toward a reduction in odds of prevalent major or minor depression at the end of the trial.

The randomized, double-blind, placebo-controlled design is a strength of our study. Loss to follow-up was within acceptable limits for both treatment groups, particularly given the 7-year average study duration. In addition, as the diagnosis of major depression was based on the use of a validated structured clinical interview and standardized criteria for the diagnosis of major and minor depression, our findings should have clinical relevance. Moreover, we are not aware of any breaches of protocol during the trial.

The main limitation of this study resides in the fact that VITATOPS was designed primarily to test the hypothesis that the prolonged administration of B-group vitamin supplements would reduce the combined incidence of nonfatal recurrent strokes and myocardial infarction, and death due to vascular causes. VITATOPS-DEP was an add-on study performed at the final VITATOPS assessment, and the collection of depression endpoints was retrospective and cross-sectional. Because these subjects were consenting survivors, some degree of bias may have occurred (recall bias for past events and survivor bias due to differential loss to follow-up). Nonetheless, the proportion of participants lost to follow-up was similar in the 2 treatment groups, and ITT analyses

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<tr>
<th>TABLE 2: Demographic and Clinical Characteristics of VITATOPS-DEP Participants According to Their Treatment Allocation</th>
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<tr>
<td>Characteristics at randomization</td>
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<tr>
<td>Age at entry, yr, mean (SD)</td>
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<tr>
<td>Male gender, n (%)</td>
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<tr>
<td>Caucasian ethnicity, n (%)</td>
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<tr>
<td>Current smoker, n (%)</td>
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<tr>
<td>Diabetes, n (%)</td>
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<td>Hypertension, n (%)</td>
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<td>High cholesterol, n (%)</td>
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<tr>
<td>Using antidepressant, n (%)</td>
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<tr>
<td>Stroke classification, n(%)&lt;sup&gt;b&lt;/sup&gt;</td>
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<tr>
<td>TACS</td>
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<td>PACS</td>
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<tr>
<td>LACS</td>
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<tr>
<td>POCS</td>
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<tr>
<td>Monocular blindness</td>
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<td>tHcy, geometric mean (SD)</td>
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| Clinical characteristics at final assessment | Placebo (n = 137) | Vitamins (n = 136) | Statistic<sup>a</sup> | p   |
| Time in trial, yr, mean (SD) | 6.9 (2.1) | 7.2 (2.1) | 1.03 | 0.303 |
| Vitamin compliance, n (%) | 102 (79.7) | 108 (85.0) | 1.26 | 0.262 |
| Stroke recurrence, n (%) | 26 (19.1) | 15 (11.2) | 3.29 | 0.070 |
| Moderate/severe handicap, n (%) | 32 (23.5) | 42 (31.3) | 2.07 | 0.150 |
| Cognitive impairment, n (%)<sup>b</sup> | 11 (8.0) | 15 (11.0) | 0.40 | 0.713 |
| Antidepressant use during trial, n (%) | 23 (16.8) | 35 (25.7) | 3.27 | 0.071 |

<sup>a</sup>Student t tests were used to compare mean differences and Pearson’s chi-square statistic to compare frequency distributions according to group allocation.

<sup>b</sup>Data missing for 23 participants in each treatment group.

LACS = lacunar stroke; PACS = partial anterior circulation stroke; POCS = posterior circulation; TACS = total anterior circulation stroke.
produced effect estimates of treatment that remained virtually unchanged for incident major depression. Of note, we included some factors in these multivariate models that were collected during the study and conservatively assumed that they could have modified the effect of the intervention throughout the trial (e.g., cognitive impairment). In addition, our trial is likely to have included an unknown number of people with past history of depression, so that the major depressive episodes which occurred between randomization and the end of the trial represent a mixture of recurrent and incident episodes. As all participants had a similar chance of being randomly assigned placebo or vitamins, the presence of prevalent cases of recurrent depression in the sample should not have biased the results. Moreover, remission of prevalent cases and prevention of recurrence are as important as the prevention of new episodes of depression after stroke. We also acknowledge that clinically significant depressive symptoms that lead to functional impairment are not limited to major depression,²⁹,³⁰ and this is the reason why we assessed participants for the presence of minor depression at the end of the trial (the MINI did not provide sufficient information to enable us to diagnose past episodes of minor depression).

Because participants were randomized an average 7 months poststroke, we cannot infer potential benefits of B-group vitamin supplementation up to this time. This is an important question that will need to be addressed by future studies, as a large proportion of people develop depression within 6 months of a stroke.¹,³¹ We also acknowledge that the power of the VITATOPS-DEP trial was limited by its modest sample size, and this may have compromised our ability to conclusively demonstrate a treatment effect for prevalent depression. Finally, our data shows that there was a nonsignificant excess of antidepressant use during the trial among patients assigned vitamins. As we did not have access to information regarding the underlying reasons for the prescription of antidepressants, we were unable to determine how this could have influenced the results of our study. We

![FIGURE 2: Survival curves showing the proportion of participants meeting criteria for a major depressive episode during the trial according to treatment group. The HR of depression associated with vitamin treatment was 0.48 (95% CI = 0.27–0.86; intention-to-treat HR = 0.48; 95% CI = 0.31–0.76; adjusted for age at the time of randomization, gender, antidepressant use between randomization and the VITATOPS-DEP assessment, handicap, cognitive impairment, and recurrence of stroke). [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.](image-url)](image-url)

<table>
<thead>
<tr>
<th>Outcome Events</th>
<th>Placebo (n = 137)</th>
<th>Vitamins (n = 136)</th>
<th>Crude OR (95%CI)</th>
<th>Adjusted OR (95%CI)²</th>
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<tbody>
<tr>
<td>Major depression at the end of the trial, n (%)</td>
<td>24 (17.5)</td>
<td>18 (13.2)</td>
<td>0.72 (0.37–1.39)</td>
<td>0.52 (0.24–1.13)</td>
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<td>Minor depression at the end of the trial, n (%)</td>
<td>14 (10.2)</td>
<td>8 (5.9)</td>
<td>0.55 (0.22–1.35)</td>
<td>0.48 (0.19–1.23)</td>
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<tr>
<td>Major or minor depression at the end of the trial, n (%)</td>
<td>38 (27.7)</td>
<td>26 (19.1)</td>
<td>0.62 (0.35–1.09)</td>
<td>0.48 (0.25–0.91)³</td>
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<tr>
<th>tHcy, µmol/l, mean (SD)</th>
<th>Placebo (n = 86)</th>
<th>Vitamins (n = 90)</th>
<th>Mean Difference (95%CI)³</th>
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<tbody>
<tr>
<td>tHcy, µmol/l, mean (SD)</td>
<td>13.3 (0.5)</td>
<td>9.9 (0.4)</td>
<td>3.33 (2.00–4.66)³</td>
</tr>
</tbody>
</table>

²ORs adjusted for age, gender, use of antidepressants between randomization and the VITATOPS-DEP assessment, handicap, stroke recurrence, and cognitive impairment.
³ITT following multiple imputation: OR = 0.58, 95% CI = 0.31–1.09.
⁴Percentage change of tHcy from baseline associated with active treatment: 28% (data missing for 51 and 46 participants in the placebo and vitamin groups, respectively).
attempted to circumvent this limitation by systematically adjusting our analyses of the outcomes of interest for the use of antidepressants between randomization and the VITATOPS-DEP assessment, but acknowledge that this type of data should be collected prospectively.

The results of our analyses further indicate that the effect of the intervention only becomes apparent after several years of treatment (about 6 years, according to our data). The reasons for such a delayed clinical action are unclear, but our findings suggest that the physiological changes associated with the use of B-vitamins that reduce the prevalence of depression after stroke cannot be a direct consequence of the acute lowering of tHcy, which can be achieved within a few weeks or months.14,15 Other possible explanations are that B-vitamin supplementation prevents or retards the progression of vascular disease,13 random error, or another as yet unknown mechanism.

Our findings are in accord with those of observational studies showing that the odds of depression are higher among people with high tHcy.12,14,32–38 A recent meta-analysis showed that the odds of depression among older adults with the MTHFR C677T TT genotype (which increases basal tHcy by 20%39) are about 20% greater than in people with the CC genotype.12 Our tHcy data are also consistent with interim evidence from VITATOPS38 and other studies14,37 showing that treatment with 2 mg of folic acid, 25 mg of B6, and 0.5 mg of B12 is effective at reducing tHcy by more than 20%.

Five small trials have used folic acid alone or in combination with B12 to alleviate symptoms of depression,17,40–43 although none of those trials produced conclusive results, possibly because of lack of power and limited duration of treatment. Despite the existence of this supportive evidence, no previous study had been able to demonstrate that the use of folic acid, vitamin B6, and vitamin B12 reduces the onset of a major depressive episode in people at risk. Our results are, therefore, novel and important, although we cannot be sure they can be extended to adults who have not had a stroke. Moreover, these promising results should be tempered by concerns that treatment with high dosages of B-vitamins in people with preexisting cardiovascular disease or diabetes may increase the risk of fatal and nonfatal cardiovascular events.44,45

In summary, the results of the present trial indicate that participants treated with vitamin supplements (folic acid, B6, and B12) for 7 years are about 50% less likely than adults treated with placebo to experience a major depressive episode after a stroke. Independent prospective data would be valuable to confirm if the use of B-vitamin supplements should become an integral part of the routine management of stroke patients to reduce their long-term risk of depression.

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The sponsors had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; or preparation, review, or approval of the manuscript.

VITATOPS Trial registration: NCT00097669, Australian and New Zealand Clinical Trials Registry.

Potential Conflict of Interest
Nothing to report.

References


