Examining the enhancement drink NeuroBliss®: Lack of effect on mood and memory in late adolescents

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NeuroDrinks® are beverages designed to enhance certain aspects of consumers' lives, such as sleep, attention, or mood. The NeuroBliss® beverage is marketed as a product that helps reduce stress, enhance mood, provide focus and concentration, and promote a positive outlook (drinkneuro.com/bliss). NeuroBliss® contains some anxiolytic and memory boosting ingredients, such as L-theanine, chamomile, Alpha GPC, and Vitamin D. We tested the impact of NeuroBliss® consumption on measures of mood, memory, and anxiety in undergraduate students. After a mood pre-assessment test, undergraduate students consumed either one bottle of the original NeuroBliss® blend (n=14) or a placebo (n=16). Participants then attended a college level biology class and took a mood post-assessment test and a memory test approximately three hours later. Statistical analysis using both paired and unpaired t-tests indicated that NeuroBliss® had no significant effects on anxiety levels, mood, or memory. The results indicate that NeuroBliss® did not provide the advertised effects in an undergraduate population. Future studies could determine the effects of other NeuroDrink® beverages.

Abbreviations: FDA-Food and Drug Administration; VDD-Vitamin D deficient; GABA-gamma-aminobutyric acid; VAS - Visual Analogue Scale; HADS - Hospital Anxiety and Depression Scale.

Keywords: L-theanine, NeuroBliss®, phosphatidylserine, supplements, mood, anxiety, depression, stress.

Introduction

Widely available, supplement-containing drinks are marketed to consumers as providing myriad benefits, including increased energy, more focus, increased muscle mass, body health, or mental well-being. Supplements are defined as substances that cannot treat, diagnose, prevent, or cure a disease, whereas a drug is a substance that can implement these effects (U.S. FDA, 2014). The U.S. Food and Drug Administration (FDA) monitors food safety and regulates the categories of supplements and drugs. The FDA does not, however, evaluate supplements or investigate efficacy claims made by supplement products, leaving the consumer to determine the validity of each product's claim for his or herself.

The original NeuroBliss® drink is claimed to "help reduce stress, enhance mood, provide focused concentration, and promote a positive outlook" (Neuro®, 2014). The NeuroDrink® products are said to provide a dose of essential vitamins and nutrients from naturally derived ingredients found around the

world (Neuro®, 2014). The claims for NeuroBliss® are believed to be made from existing evidence on the effects of the ingredients contained in the "proprietary blend." This blend is composed of six ingredients: L-theanine, chamomile, alpha GPC and phosphatidylserine, Vitamin D, and blended B Vitamins (B1, B3, B6, and B12) for a total of 185 mg of proprietary blend ingredients per bottle. NeuroBliss® (original) also contains filtered water, crystalline fructose, citric acid, natural flavors, magnesium citrate, potassium sorbate, sodium benzoate, sucralose, acesulfame potassium, and beta carotene.

Existing Science behind the Ingredients

L-theanine is one of the most studied compounds found in NeuroBliss®. L-theanine is an amino acid that is found in tea, and has been shown to reduce responses to stress and anxiety (Kimura et al., 2007; Kobayashi et al., 1998; Lu et al., 2004).

Chamomile is linked to having mild anxiolytic properties, although these results have been somewhat contested between studies (Amsterdam et al., 2009; Awad et al., 2007; Srivastava et al., 2010). It has also been shown to decrease sleep latency (the time it takes to fall asleep (Shinomiya et al., 2005). It has also been linked to anxiety relief (Amsterdam et al., 2009).

Alpha GPC and phosphatidylserine are precursors to the neurotransmitter acetylcholine, which is implicated in memory function and formation. These two ingredients have been shown to boost memory function (Kato-Kataoka et al., 2010).

The manufacturers of NeuroBliss® claim that Vitamin D has a role in neurotransmission. Tenenhouse et al. (1991) showed that chronic Vitamin D deficient (VDD) and hypocalcemic rats had higher levels of gamma-aminobutyric acid (GABA) in most of the regions studied, whereas VDD-only (normal calcemic) rats did not show this increase; therefore, Vitamin D was not likely affecting GABA activity. Since GABA is a common inhibitory neurotransmitter, it can be suggested that an increase in GABA activity would result in decreased responses by the brain. The researchers in this study concluded that, since VDD/hypocalcemic rats showed this increase in GABA activity and VDD/normal calcemic rats did not, calcium was likely the determining factor. Low Vitamin D levels have also been linked to alterations in learning and memory (Becker et al., 2005).

The B Vitamins, particularly Vitamin B12, are implicated in metabolism and brain functioning. Vitamin B deficiency has been shown by several studies to be linked to brain degeneration, development, and various neuropathies (Weir and Scott, 1999; van de Rest et al., 2012; Vogiatzogluo et al., 2008). Some of these studies have implicated low levels of Vitamin B12 in brain atrophy and subacute, combined degeneration (Weir and Scott, 1999; Vogiatzogluo et al., 2008); so in pathological situations resulting from low B12, it is possible that Vitamin B12 consumption might increase cognitive capacity. However, a more recent review of this study has shown that there is not enough — and sometimes contradictory — evidence for the effects of B Vitamins in the brain and that more research needs to be done to give any formal recommendation (van de Rest et al., 2012).

Current Study

Based on our initial review of NeuroBliss® ingredients, we sought to evaluate its efficacy. We chose to measure variables that reflected elements of the NeuroBliss® marketing claims (decreased stress, enhanced mood, positive outlook, and increased concentration). Therefore, we evaluated the effect of the NeuroBliss® drink on measures of memory, mood, and anxiety in undergraduate students.

Materials and Methods

Participants

The study was done at a small liberal arts college in Ashland, Virginia. Randolph-Macon College has a population of 1,300 students, approximately 47% male and 53% female. The study's participants consisted of (n= 39) 11 males and 23 females, all between the ages of 18-20.

All protocols used were approved by Randolph-Macon College's Institutional Review Board. Human participants were used in the study to effectively receive information on specific testing ways and to gain specific data about mood. The subjects were recruited from an introductory college level Biology course and incentivized with the chance of winning a food gift card. Students were not asked if they had taken any other enhancement drinks that day or if they had taken other supplements or drugs. Statistical analysis was performed using both paired and unpaired t-tests.

Materials

Strawberry-flavored sparkling water was chosen as the placebo because it most closely matched the treatment, although it contained none of the same active ingredients. NeuroBliss® is lightly carbonated with a sweet, fruity taste. The bottle calls the flavor "citrus lychee." Lychee are a strawberry-looking fruit that grow on trees and possess a sweet, floral taste. This fruit is popular in China and Asian countries. The researchers' own taste testing of NeuroBliss® matched the taste most closely to a strawberry flavor. NeuroBliss® and the placebo had only one of the same inactive ingredients: acesulfame potassium, which is an artificial sweetener (Horne et al., 2002).

Methods

Before receiving a drink, a pre-test assessment was given to each participant for a baseline comparison. Fourteen participants received an entire bottle of the original formulation of NeuroBliss® treatment, while 16 participants received the same amount of strawberry-flavored sparkling water. All potential participants read over the informed consent and a combined ingredient lists for both drinks. The ingredients were also read aloud to participants.

Once informed consent was obtained, each participant was asked to fill out a mood Visual Analog Scale (VAS) for researchers to obtain a baseline. This scale had 14 pairs of concrete adjectives. One word was separated by a 100 millimeter line leading to an opposite-meaning word. For each of the 14 questions, students were to place an "X" anywhere on the line that described their current mood. For data collection a ruler was used to measure the location of the "X" (Bond and Lader, 1974). Table 1.

Table 1: Sample of the VAS test (adapted from Bond and Lader, 1974) This scale contained 18 questions with different concrete adjectives. Students were to place an "X" on the 100 mm line where they felt their current mood was at that very moment.

1. Alert	———	Drowsy
2. Calm	———	Excited
3. Strong	———	Feeble
4. Muzzy	———	Clear- headed

Following completion of the mood VAS, participants were numbered in order of completion of the pre-tests and given a beverage in a 16 ounce red Solo® cup. Every odd-numbered student received 14.5 ounces of NeuroBliss® (the entire bottle), and every even-numbered student received the placebo of 14.5 ounces of strawberry-flavored sparkling water.

Participants drank their assigned beverage under experimenters' supervision within 15 minutes.

Once finished, all students attended several sections of the same entry-level science course. The class lasted approximately three hours with a lecture and lab component. Class is

usually run with a one-hour lecture and two-hour lab. Upon finishing class, students were administered post assessments, including the same mood VAS and a Hospital Anxiety and Depression Scale (HADS) (Table 2; Zigmond and Snaith, 1983).

Table 2: Two example questions from the Hospital Anxiety and Depression Scale (HADS). This 14-question scale measures anxiety and depression Students were to choose which description fit them at that very moment.

I feel tense or wound up
most of the time
3. most of the time
2. a lot of the time
1. from time to time, occasionally
0. not at all

I still enjoy the things I used to enjoy
0. definitely as much
1. not quite so much
2. only a little
3. hardly at all

Students were also given an immediate word-recall test that consisted of one investigator reading a list of 15 unrelated, concrete words at a rate of 1 second apart. Once the investigator finished reading the words, the students were given 3 minutes to write down as many of the words as they could remember. The memory recall was administered to help researchers test the concentration claim NeuroBliss® advertises, as well as the stress reduction. Because we were only testing short-term memory, there was no memory pre-test given.

Statistics

After the single blind test, the data was analyzed using Microsoft Excel. All assumptions were met for each of the tests, using a histogram, Shapiro-Wilk normality test, and a normal probability plot. The Mood VAS data was collected by researchers by using a ruler to measure where participants placed an "X" on the line and then assigning a number 0-10. Lower numbers indicated a more positive mood. The Mood VAS can be divided into two factors: one based on alertness and the other on tranquility. Paired t-tests were run before and after Mood VAS, while unpaired t-tests were run for HADS and memory recall.

Results

Statistical analysis using both paired and unpaired t-tests indicated that NeuroBliss® had no significant effects on anxiety levels, mood, or memory. The results suggest that NeuroBliss® did not provide the advertised effects.

Mood Visual Analogue Scales

Consumption of NeuroBliss® did not improve overall Mood VAS scores after the 3 hour experiment ($n = 14$, before mean = 3.55 ± 0.19 SE and after mean = 3.76 ± 0.23 SE). No differences were seen in the mood of the placebo group before and after drink consumption either ($n = 16$, before mean = 3.65 ± 0.19 SE and after mean = 3.82 ± 0.16 SE see Figure 1A). Using a paired t-test, the difference in the means before and after consumption was insignificant for the NeuroBliss® ($n = 14$, $p > 0.05$) and the placebo ($n = 16$, $p > 0.05$). A pooled variance t-test failed to find a significant difference between the differences when comparing the control and NeuroBliss® ($n = 34$, $p > 0.05$), indicating no placebo effect.

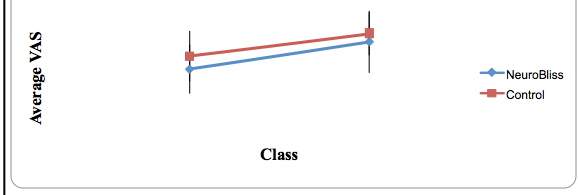

Figure 1A: Average Mood VAS values before and after class. The mood of participants was measured before and after a 3 hour undergraduate Biology class. Results are based on a 0-10 scale with lower values indicating a more positive mood. NeuroBliss® consumption did not significantly alter mood scores before and after consumption as well as for the placebo drink. A pooled variance t-test failed to find a significant difference between the treatment levels ($n = 34$, $p > 0.05$). It compared the differences of the before and after data for each treatment level to each other.

The average Factor 1 (a measure of alertness) Mood VAS value was similar for the

NeuroBliss® before consumption (n = 14, mean = 3.55 ± 0.12 SE) and after consumption (mean = 3.71 ± 0.17 SE). It was also similar for the control before consumption (n = 16, mean = 3.66 ± 0.18 SE) and after consumption (mean = 3.97 ± 0.16 SE). A pooled variance t-test found no significant difference between the differences of the two treatment levels (Figure 1B; $p > 0.05$).

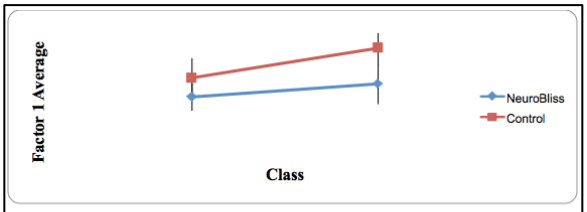

Figure 1B: Average Factor 1 values before and after class. The NeuroBliss® group had a higher score than the control group after consumption. Results are based on a 0-10 scale with 0 indicating a positive mood. Factor 1 focused on the alertness questions and a pooled variance t-test found no significant difference between the differences of the treatment levels ($p > 0.05$). It compared the differences of the before and after data for each treatment level to each other.

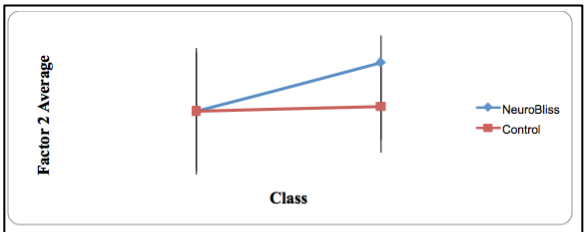

Figure 1C: Average Factor 2 values before and after class. The control group had a higher Factor 2 score than the NeuroBliss® group when averaged. Results are based on a 0-10 scale with lower scores indicating a more positive mood. Factor 2 examines the tranquility questions of the groups. A pooled variance t-test found no significant difference between the differences of the two treatment levels ($p > 0.05$). It compared the differences of the before and after data for each treatment level to each other.

The average Factor 2 (a measure of tranquility) Mood VAS were equivalent for the NeuroBliss® before intake (n = 14, mean = 3.54 ± 0.47 SE) and after intake (mean = 3.90 ± 0.57 SE). It was also equivalent for the control before drinking (n = 16, mean = 3.54 ± 0.44 SE) and after drinking (mean = 3.58 ± 0.34 SE). A pooled variance t-test found no significant difference between the differences of the two treatment levels (Figure 1C ; $p > 0.05$).

Word Recall Test

The average number of words remembered for both NeuroBliss® (n = 14, mean = 7.06 ± 0.45 SE words) and the placebo (n = 16, mean = 7.83 ± 0.56 SE words) was not significantly different by t-test (Figure 2; $p > 0.05$).

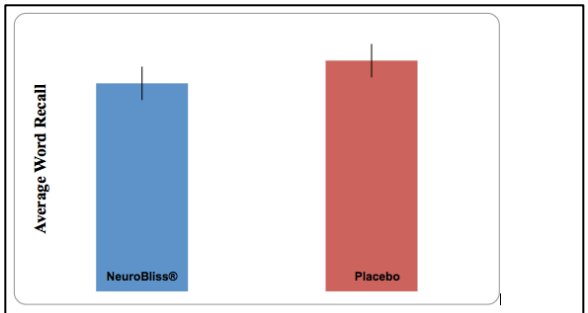

Figure 2: Average number of words recalled for the treatment levels. Participants were read fifteen words one second apart and then immediately asked to write down as many as they could recall. Only correct answers were recorded. A single tailed pooled variance t-test found no significant difference ($p > 0.05$). Error bars indicate standard error of the mean.

Hospital Anxiety and Depression Scale

The average of the Hospital Anxiety and Depression Scale (HADS) scores for the NeuroBliss® (n = 16, mean = 11.37 ± 1.29 SE) and the control (n = 18, mean ± SE = 13.50 ± 1.35 SE) were not significantly different by an equal variance t-test ($p > 0.05$).

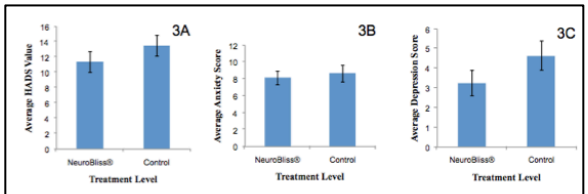

Figure 3: Results of the Hospital Anxiety and Depression Scale (HADS) (Zigmond and Snaith, 1983). There was no statistically significant difference in average depression score to support the claims that NeuroBliss® "promotes positive outlook," "reduces stress," and "enhances mood" (NeuroBliss® n = 16, control n = 18). All data was analyzed with an equal variance t-test, $p > 0.05$. This analysis was administered at the post-assessment 3 hours after consumption. Error bars indicate standard error of the mean. The three graphs show the overall HADS results (3A), the anxiety questions results (3B), and the depression questions results (3C).

It was found that there was no significant difference between the anxiety questions for each treatment group and the depression questions for each treatment group (Figure 3; $p > 0.05$).

Discussion

The results show that consumption of NeuroBliss® did not significantly alter mood or memory in undergraduates, as measured by VAS, memory test, and HADS data. We conclude that drinking a full bottle of NeuroBliss® does not provide decreased stress or promote a more positive outlook in our population of undergraduates. Of note, subjects in both treatment groups found it hard to consume the entire 14.5 ounces in 15 minutes. More studies should be done with a larger number of participants to evaluate these claims in other populations. We also did not test a completely untreated group, and it would be worth testing whether or not drinking anything at all has a significant effect on mood and memory.

Although over half the participants were female, sex was not treated as a variable during data analysis. Depending on where each female was in her menstrual phase, this could have played a role in memory tests or affected depression and anxiety levels. But in the end this was not accounted for in the experiment (Konishi et al., 2009; Golub, 1976).

There is scientific support in the literature for the use of their "proprietary blend" of ingredients, including L-theanine and alpha GPC/phosphatidylserine, but each NeuroBliss® bottle contains just 185 mg of this "proprietary blend." Some of these ingredients have been well studied and can reduce responses to stress (Kimura et al., 2007; Kobayashi et al., 1998, and Lu et al. 2004), but only if used at effective concentrations. The effective dose appears to be higher than the dose contained in NeuroBliss®. For example, researchers found that 200 mg of L-theanine induced calming effects in patients (Kimura et al., 2007, and Kobayashi et al., 1998). By comparing the effective dose with total blend amount, we can infer that there was simply not enough present.

Kato-Kataoka et al. (2010) used both 100 mg and 300 mg per day of phosphatidylserine in their experiments to measure effectiveness; it was noted that the higher dosage had more of an effect, and this dosage is not included in the "proprietary blend," as the entire NeuroBliss® blend is only 285 mg. Since Vitamin D helps with the absorption of calcium, the inclusion of calcium in NeuroBliss® has the ability to have long-term benefits, such as prevention of osteoporosis, but will not have any desired effects on the brain immediately (Tenenhouse et al., 1991). Therefore, the addition of Vitamin D to NeuroBliss® may have no noticeable effects. Since there is no consensus on the exact effects of B Vitamins on the brain, we cannot determine if the addition of these vitamins to NeuroBliss® will have any immediately noticeable effects (Weir and Scott, 1999; van de Rest et al., 2012).

It is possible that alterations in mood or concentration could have been reached at an earlier time-point than our three-hour measurement. Unno et al. (1999) found maximum plasma concentration of l-theanine between 30-60 minutes, but a recent study by Scheid et al. (2012) found absorption at 10-24 minutes, although this is not maximum absorption. van der Pijl and Chen (2010) found that absorption of L-theanine can start quickly (10 minutes) and reaches peak absorption around 50 minutes after consumption.

NeuroBliss® does not meet the claims to reduce stress, increase mood, and promote a more positive outlook in a group of undergraduates when assessed at 3 hours after consumption. The immediate word recall test seemed to frustrate some students, as they wanted to do well, which may have been a

confounding variable. It should be tested whether NeuroBliss® would be more effective in a clinically depressed or anxious population, or perhaps at an earlier time point in healthy individuals. If the manufacturers added a higher dosage of L-theanine, alpha GPC and phosphatidylserine, and perhaps chamomile, then the product should be re-evaluated for improved efficacy.

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