THE EFFECT OF TEA AND ITS CONSTITUENT L-THEANINE ON ANXIETY:
A REVIEW OF THE LITERATURE

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A Senior Project submitted
In partial fulfillment of the requirements for the degree of
Bachelor of Science in Nutrition

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California Polytechnic State University
San Luis Obispo, CA
December 2014
ABSTRACT
Mental health disorders are a universal issue and affect millions of individuals every day. Anxiety is the most common type of mental disorder, affecting social, physical, and cognitive health. Although clinicians usually suggest prescriptive medications, they carry side effects, are expensive, and only acutely alleviate symptoms. As a result, recent interest has been directed towards nonconventional dietary therapies, including tea and its constituent L-theanine, for anxiolytic effects. Tea is the most prevalent beverage consumed after water; it is historically and globally accepted. The purpose of this literature review is to explore whether tea and its constituent L-theanine carry anxiolytic effects. To identify research in human subjects, PubMed was searched for human studies from 1999 through 2014 of the relation between tea, its constituent L-theanine, and anxiety. Seven studies (2 cross-sectional, 5 clinical) were identified as relevant to measuring anxiety either subjectively or objectively. Three clinical studies showed an effect of tea and L-theanine on anxiety, however they lacked sufficient quality to give confidence of a cause-effect relationship. Stronger studies showed an inconsistent relationship between tea and its constituent, L-theanine, on anxiety. Due to anxiety’s high prevalence and health consequences, more clinical trials using systematic measurements for assessing exposures and outcomes in a large number of subjects need to be undertaken. One key research question that persists is whether L-theanine in isolation is similar to L-theanine found in tea. More valid research is needed before definite conclusions can be made about the consumption of tea and its constituent, L-theanine, for anxiolytic properties.
Introduction

Mental health disorders are a worldwide issue that affect 12% of individuals at some point during their lives (Kessler et al., 2009) and 7.3% of the global population at any given time (Baxter, Vos, Scott, Ferrari, & Whiteford, 2014). Mental disorders include anxiety, mood, impulse control, and substance abuse disorders. Disorders are specifically widespread in the United States and impact 30% of the adult population, with anxiety the most prevalent disorder at 18.1%. It is expected that anxiety will affect 28.8% of individuals in the United States for at least a 12-month span (Kessler, Chiu, Demler, Merikangas, & Walters, 2005). Some of the many detrimental consequences of anxiety include an overall lower quality of life, leading to impaired physical health, disturbed social relationships, and a decreased function at work (Olatunji, Cisler, & Tolin, 2007).

Current treatments for anxiety include cognitive, behavioral, and pharmacological approaches (Barlow, 2004). Non-traditional approaches include nutritional therapy and dietary supplements. A recently proposed dietary treatment for anxiety is tea and its constituent L-theanine (Kimura, Ozeki, Juneja, & Ohira, 2007). Recent research suggests that tea may have a positive effect on mental health and mood. Tea may also be beneficial to physical health since it contains bioactive compounds that may potentially reduce the risk of cardiovascular disease, cancer, and diabetes (Matthews, 2010). Besides water, tea is a traditional part of lifestyle and is the most frequently consumed beverage internationally (Matthews, 2010).

Historically, tea has been associated with comfort and relaxation. However, it has only recently been studied whether tea and its constituent L-theanine truly lowers
psychological stress (Einöther & Martens, 2013). The purpose of this literature review is to investigate whether tea and its constituent L-theanine carry anxiolytic effects. The majority of this paper will explore human studies of the effects of tea and L-theanine on anxiety. First, however, an anxiety overview will be presented, including its definition, diagnosis, complications, and treatments. Tea varieties, consumption data, and health effects will also be described.

Anxiety Overview

Definition & Prevalence

Anxiety is a mood state characterized by irrational anticipation and sensitivity towards futuristic threats (American Psychiatric Association [APA], 2013). Mild or brief anxiety may be considered as a normal response to stress. However, anxiety disorders are severely debilitating. They last at least 6 months and lead to a host of mental, physical, and social health issues (Craske et al., 2009). Anxiety disorders are separated into multiple subcategories including generalized anxiety, panic, post-traumatic stress, social anxiety, and obsessive-compulsive disorder (National Institute of Mental Health [NIMH], 2009).

Around 40 million, or 18%, of American adults are currently suffering with an anxiety disorder. The lifetime prevalence of having an anxiety disorder in the United States is 28.8%. Anxiety disorders currently affect nearly 8% of teenagers (NIMH, 2009). Figure 1 further specifies the percentages of various age groups that have an anxiety disorder in America. Evidently, it is most common for individuals ages 30-44 to have an anxiety disorder, and least common for those older than 60 to have an anxiety disorder.
Overall, 19 million Americans have a phobia, 15 million Americans are diagnosed with social anxiety disorder, almost eight million carry post-traumatic stress disorder, nearly seven million Americans are diagnosed with generalized anxiety disorder, six million are affected by panic disorder, and over two million have obsessive-compulsive disorder (Anxiety and Depression Association of America [ADAA], 2014a). About two thirds of these individuals do not receive any treatment and daily experience the many complications of anxiety (NIMH, 2009).

Etiology

Combinations of mental, environmental, developmental, and genetic components are suggested to cause anxiety (NIMH, 2009). Anxiety mainly arises from brain regions called the amygdala and hippocampus. The amygdala responds to a threatening situation by generating fear. The hippocampus then encodes this fearful response into emotional memories. Stored memories of this negative event can trigger feelings of threat and anxiety. Chronic and excessive cases of anxiety are termed anxiety disorders (NIMH, 2009).
Diagnosis

In order to determine whether an individual has an anxiety disorder, a skilled physician must carefully analyze and evaluate symptoms that an individual is experiencing (NIMH, 2009). Most physicians rely on the Diagnostic and Statistical Manual of Mental Disorders (DSM-5) to diagnose an anxiety disorder (Craske et al., 2009). Due to the separate diagnostic criteria for individual anxiety subtypes, the diagnosis of anxiety with the DSM criteria is complicated to summarize. However, example criteria for generalized anxiety disorder will be given. Criteria include difficulty in managing worry; an extreme apprehension about normal circumstances; worry that disturbs everyday functioning; anxiety that is not due to another medical or physiological occurrence; an issue is not more properly defined by another psychological disorder; and a state that exhibits at least three of the subsequent manifestations: restlessness, lethargy, problems with focusing, annoyance, muscular tension, and sleep issues (APA, 2013). Criteria common to all disorders are based on self-report and include panic, self-restraint, perspiration, nausea, cardiac palpitations, muscle tightness, and tremor (Craske et al., 2009). Although anxiety shares many comparable symptoms with other mental disorders like depression, physiological hyperarousal is a distinguishing characteristic that separates an anxiety disorder apart from other mental disorders (Craske et al., 2009).

Due to the complex features of anxiety disorders, psychological questionnaires can aid a physician in diagnosing a patient with an anxiety disorder (Craske et al., 2009). Disorder-specific scales are used in research settings and accurately indicate the severity of psychological distress (APA, 2013). Disorder-specific scales can also address any
changes in the severity of an anxiety disorder. Multiple scales have been created for different sets of focus, including the measurement of cognitive, behavioral, and physical symptoms (APA, 2013). Specific anxiety tests that are commonly used for research purposes will now be discussed.

**Questionnaire-based anxiety tests.** The State-Trait Anxiety Inventory Questionnaire (STAI) is a universal instrument that relies on two separate anxiety levels, state and trait anxiety, to measure the level of psychological distress. The STAI has two sections with twenty questions each, with answer options using a 4-point Likert scale. Scores range from 20 to 80 for each section (American Psychological Association, 2014). Subscores above 40 are suggested to indicate symptoms of anxiety, with higher numbers indicating more extreme anxiety disorder cases. Since this test is considered to be one of the most consistent, reliable, and valid, it is one of the most widely used instruments in research settings (Julian, 2011).

The Beck Anxiety Inventory (BAI) emphasizes the cognitive, subjective, and objective indications of anxiety (Beck & Steer, 1990). Using a 4-point Likert scale, the 21 multiple-choice questions ask about symptoms specific to anxiety and help distinguish anxiety from depression. Each question ranges from 0-3 points and contributes to the maximum score of 63. Summative scores of 21 or less illustrate minimal anxiety, scores ranging from 22-35 suggest moderate anxiety, and scores above 36 indicate that someone has severe anxiety. Like the STAI, the BAI is demonstrated to have sound consistency, validity, and reliability (Beck & Steer, 1990).

**Scale-based anxiety tests.** Another common means of measuring anxiety levels is the Visual Analogue Mood Scale (VAMS) by Robert Stern (Stern, Arruda, Hooper,
Wolfner & Morey, 1997). In the VAMS, individuals describe how they feel by marking lines across various emotional statements or neutral symbolic faces. This scale has a greater variety of response options when compared to a multiple-choice questionnaire. Consequently, health experts may be more likely to recognize subtle changes in mood behavior (Stern et al., 1997). Eight specific mood states are analyzed and scored from 0-100 by measuring the distance with a ruler on the marked line. Scores are adjusted into gender- and age-specific T-scores. If the measure of the fear scale is above 60T-70T, an individual is suggested to have an anxiety disorder (McCallum, 2003). This method is rapid, easy to administer, and simple to fill out (Stern et al., 1997).

A test similar to the VAMS is the Bond-Lader scale. The Bond-Lader scale is a self-reported questionnaire that measures subjective feelings with a series of 16 visual analogue scales (VAS) (Bond & Lader, 1974). This scale measures current anxious feelings and physiological functioning through measures of alertness, calmness, and contentedness. Subjects mark along a 100 mm scale that is juxtaposed with mood-related adjectives. Scales are measured with a ruler and compared with the mean score in the given manual (Bond & Lader, 1974). Scoring below the average for calmness and contentedness and above the average for alertness may indicate that an anxiety disorder is present. When compared to conventional scales, the Bond-Lader scale offers a wider variety of categorical responses and is therefore suggested to carry greater accuracy (Bond & Lader, 1974).

The Kessler Psychological Distress Scale (KPDS) is a quick, easy, and validated questionnaire measuring level of psychological distress (Kessler et al., 2003). It is based on contemporary psychometric theory and can include a various amount of questions
In the 6-item scale test (K-6), an individual answers six questions on whether they feel a certain way “all of the time” (4 points) to “none of the time” (0 points). Out of the total 24 points, individuals with 13 points or higher are suggested to have psychological distress (Hozawa et al., 2009). In the 10-question scale test (K-10), the patient answers 10 questions scored on a scale from 1-5 and selects if they feel a certain way none, a little, some, most, or all of the time. If the patient scores 20 or above, he or she is suggested to carry a mild mental disorder, with higher scores associated with severity of psychological distress (Kessler et al., 2003).

**Risk Factors**

Certain populations are at a higher risk for developing an anxiety disorder (NIMH, 2009). Non-modifiable risk factors, such as genetics, race, and gender, play an important role in determining the development of an anxiety disorder. Hispanics have a 30% lower risk and African Americans carry a 20% lower risk of developing the disorder compared with Caucasians (NIMH, 2009). Compared to males, females have a 60% increased likelihood of developing any kind of anxiety disorder (NIMH, 2009) and are twice as likely to develop phobias, generalized anxiety disorder, and panic disorder (ADAA, 2014a).

Additionally, individuals who have another psychiatric disorder, physical illness, or suffer with substance abuse are more likely to carry an anxiety disorder (NIMH, 2009). Specifically, depression is commonly associated with anxiety disorder: almost half of individuals with depression also have an anxiety disorder (ADAA, 2014a). Individuals who experience highly traumatic events, such as child sexual abuse, are also more likely to develop an anxiety disorder. For instance, 65% of men and almost 50% of
women who are sexually assaulted develop post-traumatic stress disorder (ADAA, 2014a). Additionally, individuals who are divorced or have been diagnosed with severe medical conditions are at a higher risk of developing an anxiety disorder (National Institutes of Health, 2014).

Consequences and Complications of the Disorder

At nearly one third of America’s mental health bill, anxiety disorders annually cost the country $42 billion (ADAA, 2014a). Anxiety is a comorbid condition and individuals diagnosed with an anxiety disorder have a 3-5 times higher likelihood of seeking medical care for various health complications. Physiological complications include lightheadedness, fainting, and gastrointestinal distress (ADAA, 2014a). Chronic pain disorders, like arthritis and migraines, are also very common in individuals with anxiety disorder (ADAA, 2014b). Clinical illnesses including respiratory disease, cardiovascular disease, and gastrointestinal disorders are attributed to anxiety disorders (Harvard Medical School, 2008). In particular, anxiety disorders have been shown to be highly associated with cardiovascular disease. Women with the highest anxiety have been shown to be 59% more likely to experience and 31% more likely to die from a myocardial infarction than women with the lowest anxiety (Albert, Chae, Rexrode, Manson, & Kawachi, 2005).

Additional complications can result from anxiety triggering other psychological disorders. One example of a mental disorder is somatoform disorder, which is when an individual has unexplainable physical symptoms that are suggested to solely arise from mental illness. Common somatoform disorders include hypochondriasis, conversion disorder, and body dysmorphia (Harvard Medical School, 2008). Body dysmorphia can
lead to disturbances with eating, where almost half of individuals have history of an anxiety disorder prior to the onset of their eating disorder (ADAA, 2014b). Other mental disorders that can arise from anxiety include bipolar disorder, sleep disorder, attention deficit hyperactive disorder, and depression (ADAA, 2014a).

Treatments for Anxiety

Anxiety disorders are most commonly treated with psychotherapy, prescription drugs, or a combination of the both. For most individuals, combining psychotherapy with medication is considered to be the best treatment (NIMH, 2009). Psychotherapy, commonly known as counseling, is the practice of therapeutic interaction with a mental health professional. Psychotherapy usually involves cognitive-behavioral therapy, an approach that helps individuals change the way they think and react in fearful situations (NIMH, 2009). Other common therapies include acceptance and commitment therapy, exposure therapy, and dialectical behavioral therapy (ADAA, 2014c).

Prescriptions drugs are also used to treat, but not cure, anxiety disorders (NIMH, 2009). The medications alleviate major symptoms of psychological and physiological distress. Benzodiazepines are the most common psychoactive drugs for anxiety disorder (NIMH, 2009). Benzodiazepines include the medications alonazepam, iorazepam, and alprazolam. Clinicians also prescribe antidepressants and beta-blockers for the treatment of anxiety disorder (NIMH, 2009). However, these treatments do not alleviate symptoms with all individuals. The medications may produce adverse side effects that include fatigue, sexual malfunction, gastrointestinal issues, hypotension, and fluctuations in weight. Further issues involve tolerance, dependence, and a high expense for doctors visits and prescriptions (NIMH, 2009).
Due to the high expense of prescription medication and its unwanted side effects, nonconventional therapies are receiving increasing universal interest (Lakhan & Vieira, 2010). Acupuncture, aromatherapy, and mindfulness-based medication are more familiar types of complementary and alternative medicine for treating anxiety (Van der Watt, Laugharne, & Janca, 2008). Nontraditional approaches also involve dietary practices and supplements (Lakhan & Vieira, 2010). Common supplements include kava, passionflower, L-lysine, and L-arginine. Although some scientific evidence supports the use of these supplements for the reduction of anxiety disorder symptoms, these treatments are not a proven therapy (Lakhan & Vieira, 2010). Additionally, treatments like kava may cause harmful side effects, such as severe liver damage (National Center for Complementary and Alternative Medicine, 2012). Amidst these findings, recent attention has been drawn to the use of tea and its amino acid L-theanine for the enhancement of relaxation (Vuong, Bowyer, & Roach, 2011). The rest of this review will examine tea composition and consumption, as well as human studies of its effect on anxiety.

**Tea Varieties, Consumption, and Health Impacts**

Since 1700 BC, tea has been perceived as a necessary substance for the maintenance of health (Heiss & Heiss, 2007). Deemed the “elixir of life,” tea has universally played a role in cultural, religious, social, and medicinal situations. Today, tea is still recognized for its herbal-healing traditions and continues to thrive as one of the world’s favorite beverages (Heiss & Heiss, 2007).
Tea Varieties

All teas originate from the *Camellia sinensis* plant (Matthews, 2010). Differences in processing methods produce the independent varieties of tea (Carlson, Bauer, Vincent, Limburg, & Wilson, 2007). The four predominant types include green, white, black and oolong (Matthews, 2010). In order to make green tea, bush leaves are immediately steamed and dried after picking, which prevents oxidation and produces larger amounts of catechins. Although green tea is recognized as having a simpler flavor, thousands of varieties and flavors exist (Matthews, 2010). Similar to green tea, white tea is unfermented. It is restricted from the sun in order to maintain its white hue and pure taste (Alcázar et al., 2007).

Unlike green and white tea, black and oolong teas are fermented. In black tea bush leaves are picked, withered, and crushed to promote oxidization (Heiss & Heiss, 2007). The high amounts of theaflavins and thearubigins found in black tea produce its dark, rich color and flavor (Yang, Chen, & Wu, 2014). Oolong teas are much more difficult to create since the leaves have specific size and sunlight requirements (Carlson et al., 2007). After its leaves are prematurely plucked, they are crushed and semioxidized. Consequently, they have a chemical combination like green and black teas (Carlson et al., 2007) and produce a variety of rich and fruity aromas (Heiss & Heiss, 2007).

Consumption Data and Consumer Trends

*Worldwide.* Every year, individuals consume an average of 40 liters of tea (Alcázar et al., 2007), contributing to the annual worldwide production of almost 4.7 million tons (Statista, 2014). As one of the top consumers in tea, China is responsible for
40% of worldwide tea production (Statista, 2014). Per capita, Ireland and Britain have the greatest number of tea drinkers (Richardson, 2010). In all countries besides Asia, black tea is the most frequently consumed tea type (Bryan, 2008). In response to this popular demand, 75% of all tea produced is black tea (Carlson et al., 2007).

United States. Over half, or around 158 million Americans, drink tea daily (Tea Association of the USA, 2013). An average American will drink almost 9 gallons of tea each year (Statista, 2014), with 85% of it being iced. Retail markets annually sell about $2.25 billion dollars worth of tea, with sales increasing nearly 16% within the past 5 years (Tea Association of the USA, 2013). Although black tea is at 84% of all tea sales, green tea has been growing in popularity (Tea Association of the USA, 2013). Continuous growth of this industry from all areas is expected, primarily due to the highly publicized health benefits of tea (Bryan, 2008).

Proposed Health Effects

Due to the increasing interest of tea’s benefits, health effects have recently been widely studied. Most studies have involved green tea (Yang et al., 2014). Due to its anti-inflammatory activity, tea has been proposed to reduce cancer, high blood pressure, gum disease, diabetes (Matthews, 2010), obesity, and metabolic syndrome (Yang et al., 2014). More than half of randomized controlled trials involving tea have reported a reduction in cardiovascular disease (Kuriyama, 2008). Additionally, a strong inverse association has been observed for tea with stroke mortality (Kuriyama, 2008). Tea may also have neuroprotective effects, potentially reducing the risk of Parkinson’s, Huntington’s, and Alzheimer’s diseases (Yang et al., 2014). Although health effects may be promising, the exact mechanism behind tea’s protection remains obscure (Kuriyama,
Several bioactive compounds are considered to contribute to tea’s suggested health effects.

**Bioactive Compounds**

Tea has a variety of compounds that influence its flavor, mouthfeel, and nutritional value (Heiss & Heiss, 2007). For example, a variety of vitamins and minerals are naturally found in scant amounts within tea. Typical compounds include thiamine, riboflavin, vitamin C, and fluoride (Heiss & Heiss, 2007). Better characterized constituents include those discussed below.

**Polyphenols.** There is a variety of polyphenols found within tea that are proposed to reduce levels of oxidative stress. Common polyphenols include catechins and flavonols (Heiss & Heiss, 2007). An example of a well-known catechin is epigallocatechin-2-gallate (EGCG), an antioxidant that makes up almost half of the total polyphenols within green tea (Matthews, 2010). EGCG is suggested to be an anticarcinogenic molecule that modulates gene expression by inhibiting deleterious enzymes, angiogenesis, and intracellular peroxides (Matthews, 2010). Flavonols include tannins, which give tea its astringent characteristics (Heiss & Heiss, 2007). Tannins are also suggested to reduce gastrointestinal discomfort (Matthews, 2010).

**Caffeine.** Contributing to tea’s bitter taste, caffeine is a natural component found in green, black, oolong, and white tea varieties (Heiss & Heiss, 2007). Multiple factors, such as brewing time and water temperature, affect a tea’s caffeine level. On average, an 8-ounce serving of tea on average has 40 mg of caffeine (Bryan, 2008), or about one third the amount regularly found in coffee (Matthews, 2010). Caffeine amounts in specific tea types will later be presented in this literature review and can be found in
Table 1. Caffeine crosses the blood-brain barrier within 30 minutes and may strengthen alertness, concentration, physical and cognitive ability. Highest levels of caffeine are found within 30-120 minutes after ingestion (Bryan, 2008).

*L-theanine.* The tea constituent L-theanine may interact with some of caffeine’s stimulatory effects. It is proposed to induce tranquility and psychological well-being while maintaining high levels of alertness (Bryan, 2008). L-theanine is suggested to counteract caffeine’s arousing effects by lowering the body’s central nervous system response (Bryan, 2008). For example, caffeine artificially raises serotonin and gamma-aminobutyric acid levels, which are neurotransmitters that may contribute to anxiety. L-theanine is suggested to modulate the amount of these chemicals and reduce the adverse, anxious consequences of cognitive overstimulation (Bryan, 2008). At the same time, L-theanine appears to increase an individual’s acute and chronic attention span by enhancing speed of information processing and improving mental accuracy with complex activities. It may also decrease mental inactivity and exhaustion (Bryan, 2008).

L-theanine’s structure is comparable to glutamic acid, an excitatory neurotransmitter. Figure 2 displays the chemical structure of L-theanine. Along with regulating serotonin and gamma-aminobutyric levels, L-theanine may also contribute to

![L-Theanine](image)

*Figure 2. Chemical structure of L-theanine. From Yang et al., 2014.*
relaxation by down-regulating glutamic acid through antagonistic pathways (Bryan, 2008).

L-theanine is a non-protein amino acid restrictedly found in tea leaves (Heiss & Heiss, 2007) and contributes to the sweet umami flavor found in tea (Alcázar et al., 2007). L-theanine constitutes over half the total amount of amino acids in tea (Alcázar et al., 2007). Generally, L-theanine constitutes around 1-2% of tea by weight, resulting in about 25-60 mg of L-theanine per 200 mL tea serving (Bryan, 2008). L-theanine’s amount varies according to tea manufacturing and cultivation processes (Yang et al., 2014). For instance, higher amounts of L-theanine are found in a tea plant with lower fermentation (Alcázar et al., 2007) and reduced sun exposure (Heiss & Heiss, 2007).

Table 1 depicts the different types of tea in relation to their L-theanine and caffeine content. When analyzing L-theanine levels, 0.25 g dried tea leaves were combined with 10 mL of water at 80°C for 25 minutes (Alcázar et al., 2007). When analyzing caffeine amounts, 0.1 g of dried tea leaves were combined with 20 mL of boiling water for 10 minutes (Sereshti & Samadi, 2014).
L-theanine can cross the blood-brain barrier within 30 minutes and may contribute to the production of alpha brain waves (Tea Association of the USA, 2013). Alpha brain waves are associated with relaxation and calmness (Nobre, Rao, & Owen, 2008). The remainder of this paper will examine human studies of the effects of tea and its constituent L-theanine on anxiety.

**Human Studies of Tea and Anxiety**

**Cross-Sectional Studies**

Few large-scale cross-sectional studies have examined the relationship between tea drinking and anxiety disorder. However, Hozawa et al. (2009) completed one extensive study in 2006. This study examined whether average green tea intake was associated with lower levels of psychological distress in Ohsaki City, Japan. At 24.4%, Japan has one of the lowest lifetime risks of mental health disorders. In comparison, the United States has one of the highest risks, at over 55%. This information may suggest that a part of the Japanese culture may be responsible for this drastic difference.

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Table 1. Typical caffeine and L-theanine contents in different tea types

<table>
<thead>
<tr>
<th>Tea Type</th>
<th>L-theanine (mg/g)&lt;sup&gt;a&lt;/sup&gt;</th>
<th>Caffeine (µg/mL)&lt;sup&gt;b&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Green</td>
<td>1.62-3.37</td>
<td>120.5</td>
</tr>
<tr>
<td>Black</td>
<td>0.49-4.12</td>
<td>149.5</td>
</tr>
<tr>
<td>Oolong</td>
<td>0.49-4.12</td>
<td>145.2</td>
</tr>
<tr>
<td>White</td>
<td>5.3-33.37</td>
<td>165.8</td>
</tr>
</tbody>
</table>

<sup>a</sup> L-theanine in milligrams per gram of dried tea. From Alcázar et al., 2007.

<sup>b</sup> Caffeine in micrograms per milliliter of liquid tea. From Sereshti & Samadi, 2014.
tea drinking is a more prevalent custom in Japan than the United States, it may be one factor that is responsible for this observation (Hozawa et al., 2009).

Three different questionnaires were mailed to all 77,235 households living in Ohsaki City (Hozawa et al., 2009). Exposures were measured through a food-frequency questionnaire (FFQ). Although the main exposure assessed was green tea, the FFQ also measured intake of a variety of foods, coffee, black and oolong tea. The FFQ tested for validity with 3-day food records given on 4 different days. A 6-item KPDS measured mental health distress, the main outcome. The questions focused on mental stability over the past month. Additionally, a baseline survey gathered characteristics of participants, including age, gender, lifestyle factors, history of diseases, and social activities. Four different multivariate logistic regression analyses controlled for differences in baseline characteristics. Since all pertinent data were completed by 42,093 individuals, they were the only participants included in the analyses (Hozawa et al., 2009).

Results from the KPDS displayed that 6.6% of subjects exhibited psychological distress (Hozawa et al., 2009). Without adjusting for any confounders, the highest prevalence of psychological distress (8.4%) was found among those consuming the least amount of green tea, or less than 1 cup. On the contrary, the lowest prevalence of psychological distress (5.1%) was found among those consuming the most green tea, or 5 or more cups daily. Odds ratio data was presented among four models that adjusted for different confounding variables. Even in the simplest and most complex adjusted model, the protective relationship between tea and levels of psychological distress remained significant, in which the confidence intervals did not contain the null. In relation to drinking less than 1 cup of green tea a day for the most fully adjusted model, the odds
ratio of psychological distress for drinking 5 or more cups of green tea a day compared to less than 1 cup was 0.80 (0.70, 0.91). However, with further addition of confounders to each of the four models, relationships were mediated and attribution due to tea was less. Nonetheless, all models suggested a dose response relationship, with p values for trend < 0.001 (Hozawa et al., 2009). This supports that a higher green intake was associated with a lower prevalence of psychological distress.

Hozawa et al. (2009) hypothesized that green tea consumption would be associated with a lower risk of psychiatric stress. Their findings support their hypothesis, in that all odds ratios revealed that higher amounts of green tea intake were significantly associated with lower likelihood of psychological distress. Hozawa et al. (2009) limited variability and erroneous results by using a large sample population and a significant amount of individuals in each level of tea intake.

Although the Hozawa et al. (2009) study had statistically significant findings, results are still not applicable to the general population. This study only included individuals that were 40 years or older living in Japan. Cultural, habitual, and economic factors are some of the few differences that separate Japan with other individuals worldwide. Additionally, this study was mainly restricted to green tea. The few results given for black tea were insignificant, with confidence intervals containing the null. Although this study tested the validity of the food frequency questionnaire with a 3-day food record, only 113 subjects completed the food records. The food records also revealed that the reliability of the FFQ reliability was low (0.63-0.71). This suggests that self-reporting is prone to human error and are not completely reliable.
Compared to Hozawa et al. (2009), the Bryan et al. (2012) study focused on individuals in a somewhat stressful, mentally-challenging occupational environment (Bryan et al., 2012). Although Bryan et al. (2012) assessed many exposures and outcomes, this literature review will focus on the intake of tea and its effect on mood assessed with the Bond-Lader VAS relaxation subscale. One hundred and forty-two volunteers were interested in this experiment, however individuals were excluded if they did not complete all necessary information, disclosed burnout, or reported work abuse within the past 6 months. Thus, 95 professional academic and administrative staff members from three different South Australian colleges were included in the study (Bryan et al., 2012). Besides a few descriptive characteristics, other baseline information regarding the subjects was not given.

Exposures were measured through beverage diaries that assessed habitual intake of tea (Bryan et al., 2012). The diaries also took into consideration the preparation style and accompaniments of milk and sugar, along with other beverages including coffee, other caffeinated drinks, and non-caffeinated drinks. Non-caffeinated beverages included various hot beverages, juices, sodas, alcohol, and herbal teas. Although other outcomes were recorded, including various work performance factors, tiredness, self-awareness, commitment to work, work-related stress and rehabilitation, this literature review will mainly focus on the outcome of self-reported mood with the VAS relaxation subscale. The Bond-Lader VAS was earlier described in this literature review. Bryan et al. (2012) used a subscale that specifically measures calmness, with lower scores associated with improved relaxation. Beverage diaries and self-reported mood were
taken 4 times a day for 10 working days: prior to work, during the middle of work, after work, and prior to sleeping (Bryan et al., 2012).

Since subjects provided data on different days, analyses were completed through multilevel modeling. Bryan et al. (2012) assessed the direct association of beverage exposure on calmness, as well as whether there was an association or interaction of adding milk and sugar to tea. Results were displayed as separate data points between and across hierarchical structures. The three levels of analysis included the time-frame, day, and subject. The data were adjusted to eliminate daily variations and incidental trends among participants. Although missing data points were infrequent, the data points were recovered by spontaneously inputting numerical values, substituting average numbers, or classifying the data as missing data points (Bryan et al., 2012). The study does not mention why it would choose one option over the other, which could lead to inconsistent recovery of data.

Within the 10 days reported, 64 subjects drank tea, with daily intakes ranging from 0-7.5 cups, with the median being 1 cup (Bryan et al., 2012). Tea was associated with a higher perception of work accomplishment, lower fatigue, alleviation of adverse consequences of nighttime rehabilitation, and enhancement of morning mindfulness (p < 0.1). However, tea consumption was not associated with enhanced relaxation. Even when adjusting for the addition of milk or sugar, effects of tea on relaxation were still insignificant. The only significant relationship found with relaxation was a positive relation among non-caffeinated beverages (p < 0.01), but not tea specifically (Bryan et al., 2012). Although herbal teas were a subcategory of non-caffeinated beverages, this
study did not provide results specific to herbal teas. Results could have been more comprehensive if they individually measured herbal tea and its effect on relaxation.

In this observational experiment, tea and its association with relaxation were uniquely studied within a common workplace environment. Therefore, Bryan et al.’s (2012) results extended outside of a typical laboratory setting. By adjusting data, Bryan et al. (2012) limited daily variations and supported a high internal validity. Unlike Hozawa et al. (2009), Bryan et al. (2012) considered additives to tea, reflecting different ways tea could be consumed. However, the multiple comparisons of many exposures and outcomes gave a high opportunity for spurious relationships, making it difficult to determine if a significant finding is real or a statistical anomaly.

The Bryan et al. (2012) study has several limitations. Bryan et al. (2012) pulled information out of context in order to support their hypothesis. They hypothesized that tea, when compared to other beverages, would have beneficial effects on work related factors, including relaxation. This study did not fully discuss that tea did not contribute to relaxation. Only significant relationships were reported; insignificant data were ignored. Additionally, findings supported that the absence of caffeine encouraged daytime relaxation. Since tea often contains caffeine, this finding may provide counter-evidence for tea enhancing relaxation.

Since the sample population was very distinct and small, Bryan et al.’s (2012) findings cannot be extended to the general population. Only a few subject characteristics were given: most participants rated themselves with good health (4.08 on a scale of 1-5), were female (78.9%), and were from a specific work environment and region (academic and administrative staff in South Australia). Unlike Hozawa et al. (2009), Bryan et al.
(2012) did not adjust for any differences in characteristics. Confounding factors could have influenced any of the correlations that were given.

Table 2 compares the two recently discussed cross-sectional studies. Hozawa et al. (2009) found that subjects who green drank tea were less likely to have psychological distress, however Bryan et al. (2012) did not find a significant relationship between intake of tea and relaxation. Although there are a limited number of studies in this comparison, this table shows that there is an inconsistent relationship between tea and its proposed anxiolytic effects.

Table 2. A summary of two cross-sectional studies examining the relationship between consumption of tea and anxiety

<table>
<thead>
<tr>
<th>Study</th>
<th>Subjects</th>
<th>Treatment</th>
<th>Outcome Measure</th>
<th>Findings</th>
</tr>
</thead>
</table>
| Hozawa et al., 2009 | 42,093 volunteers 40+ years living in Ohsaki City, Japan | Average cups of green, black, and oolong tea with one FFQ                   | Prevalence of psychological distress using K-6 at same time of treatment | • Lowest prevalence of psychological distress among those consuming the most amount of green tea  
  • Odds ratios revealed that higher intake of green tea was associated with lower prevalence of psychological distress with a dose response relationship  
  • No significant findings with black tea |
| Bryan et al., 2012  | 95 South Australian academic employees with an average age of 37 | Average habitual intake of tea including milk and sugar additives 4x day for 10 days | Level of relaxation with VAS³ calmness subscale at same time of treatment | • Tea consumption not associated with enhanced relaxation  
  • Non-caffeinated beverages positively associated with relaxation |

³K-6: Six question Likert scale test from the Kessler Psychological Distress Scale that measures levels of psychological distress.
³VAS: Visual analogue scales that measure subjective anxiety through a series of 100 mm scales.
The inherent design of cross-sectional studies limit their findings. It cannot be interpreted whether the exposure (tea) led to the outcome (lower psychological distress), or vice versa. In order to make any definite conclusions, it is necessary to examine clinical studies involving tea and anxiety. Only one has been published to date.

**Clinical Study**

An experiment by Steptoe et al. (2007) explored whether 6 weeks of black tea intake would influence subjective responses toward acute stress. Although Steptoe et al. (2007) considered objective measurements including cardiovascular, platelet, and cortisol levels, this evaluation will mainly focus on the subjective stress and relaxation ratings as the primary outcomes. In this double-blind experiment, 105 volunteers were recruited by means of email and print. Subjects had to be healthy, nonsmoking, tea-drinking men that were between 18-55 years old. Participants were excluded if they had a previous psychiatric or medical history, were taking prescription medications, or followed any diet. After a preliminary lab evaluation, subjects underwent a 4-week washout period where all participants were restricted from caffeinated beverages, tea, coffee, painkillers, nutritional supplements, and flavonoid rich foods. During this time, all subjects were instructed to drink a placebo in the form of a fruit-flavored tea. The washout period allowed for greater reliability of dietary adherence. Participants were tested for adherence with self-report and saliva samples at the end of the washout period; 13 individuals did not follow the dietary instructions and were dropped from the study (Steptoe et al., 2007).

After omitting 17 other participants due to inadequate data, 75 subjects were left eligible for the study (Steptoe et al., 2007). Baseline characteristics were taken, which
included habitual tea intake, race, age, educational degree, marital status, previous smoker, and physical activity levels. Participants were then randomized into the treatment or control group. Primarily as a result of randomization, subjects did not differ in any measured baseline characteristics. Thirty-seven subjects were assigned to the active treatment: a fruit-flavored, powdered black tea added to hot water 4 times daily. Since the treatment group was unaware they were drinking tea, confounding variables associated with drinking tea were likely eliminated. Thirty-eight subjects drank the placebo: a fruit-flavored, tea-colored powder dissolved in hot water 4 times daily. The experimental and control beverages had identical organoleptic qualities. Both treatments also had equal amounts of caffeine, so the two conditions only differed in other tea constituents (Steptoe et al., 2007).

Both groups followed their designated treatment for 6 weeks (Steptoe et al., 2007). Compliance was tested for with self-report and caffeine saliva samples. Subjects were instructed to consume one beverage before testing for outcomes. Prior to and after the 6-week dietary intervention, subjects’ stress and relaxation levels were tested with 7-point Likert scales. The scales measured perception of stress from low (1) to high stress (7). Subjective measures were completed before (baseline), during, and 30 minutes after assigning two behaviorally difficult activities. The activities included a stressful speech challenge and a laborious mirror-tracing task. Although not described in detail in this review, objective measurements were obtained prior to the subjective tests (Steptoe et al., 2007).

During the span of recovery from baseline to post-stress recovery, the experimental tea group had a significantly greater sense of relaxation (+6.36%), whereas
the control group had a reduced sense of relaxation (-3.19%) \( (p = 0.032) \) (Steptoe et al., 2007). Combined, this difference in relaxation is nearly 10%. However, when analyzing average subjective stress ratings after adjusting for pretreatment responses, differences between the two groups were insignificant (Steptoe et al., 2007). This finding is depicted in figure 3, where the solid line depicts the experimental tea group and the dashed line represents the control group, with groups nearly indistinguishable.

![Figure 3. Average subjective stress ratings (1=low stress, 7=high stress) adjusted for pretreatment responses after 6 weeks of treatment. Solid line is tea, dashed line placebo. Responses include baseline, speech task, mirror tracing (MT), and recovery trial. From Steptoe et al., 2007.](image)

The Steptoe et al. (2007) experiment has several strengths. This study was double-blinded, limiting bias. Steptoe and his colleagues openly admitted that their first hypothesis was not supported—that tea would lower cardiovascular and platelet stress responses. Another strength is that this study provided a 6-week intervention period. Many clinical trials obtain results after just one trial, however Steptoe et al. (2007) had a long-run experiment that allowed for the development of substance response patterns. Additionally, most of the results for task responses were logical and according to
expectations: subjects correspondingly had greater acute stress ratings as a result of the behavioral challenging tasks. Accordingly, all ratings of stress were lowered during post-stress recovery. There was also no disparity in perception of task difficulty between groups. Subjective post-relaxation results were supported and consistent with objective outcomes. For example, lower subjective relaxation ratings were directly correlated with lower levels of cortisol, a hormone associated with stress response (Steptoe et al., 2007).

However, not all of Steptoe et al.’s (2007) conclusions are fully substantiated or sensible. Although Steptoe et al.’s study (2007) supported that tea improved relaxation, they did not support that tea had protected against subjective stress. Since relaxation and stress are opposing concepts, it would be assumed that findings between the two would be in agreement. The Steptoe et al. (2007) experiment has some additional shortcomings. Subjects were healthy, tea-drinking, middle-aged men. Since this is a narrow population subgroup, results cannot be compared to the general society. Additionally, results cannot be extended to other living conditions, such as a stress-free environment. Findings only pertain to a condition involving a stressful situation within a laboratory facility. Also, only one subjective test measured stress and relaxation post-treatment. Validity would have been further improved if additional tests during the intervention measured outcomes. Besides caffeine, adherence to dietary constraints was only assessed through self-report during the two tests. Results could have also been more valid if objective quantifiers verified beverage compliance throughout the intervention.

Steptoe et al. (2007) claim that tea may strengthen psychophysiological response post-stress. They do not provide a proven mechanism behind their reasoning, but do
allude to certain constituents within tea, including L-theanine, which may be responsible for stress reduction. Clinical trials involving L-theanine and its effect on anxiety will now be discussed.

**Clinical Studies of L-theanine and Anxiety**

Human trials of L-theanine and anxiety have been conducted using different outcomes measures, including self-report anxiety questionnaires and changes in alpha brain waves. This next section will discuss experiments that have measured anxiety either subjectively or objectively.

*Measurements using anxiety questionnaires.* A double-blind, randomized study by Haskell and his colleagues compared the acute effects of L-theanine and caffeine together and individually on psychological measures of mood (Haskell, Kennedy, Milne, Wesnes, & Scholey, 2008). Although this study measured other factors including reaction time, memory accuracy, and word recognition, this literature review will focus on the outcome of relaxation measured by the Bond-Lader VAS. Nine males and 15 females from ages 18-34 years volunteered for this study. Participants were undergraduate students in good health and free from drugs, medications, and smoking. Subjects were required to restrict themselves from caffeinated and alcoholic beverages during and for at least 12 hours before testing (Haskell et al., 2008).

This balanced crossover study used 250 mg L-theanine, 150 mg caffeine, 250 mg L-theanine and 150 mg caffeine combined, and a placebo containing neither component as exposures in a 250 mL Lipton Iced Tea drink (Haskell et al., 2008). The drink tasted similar between the placebo and treatment groups. The Bond-Lader’s calm, content, and alert subscales measured changes in subject anxiety. Details about this scale were earlier
described in this paper. Outcomes were measured 5 times 7 days apart at baseline, 30 minutes post-dose, and 90 minutes post-dose (Haskell et al., 2008). The multiple days of nonconsecutive testing improved reliability and decreased results due to chance.

Statistical analyses were completed through ANOVA (Haskell et al., 2008). Paired t-tests compared the three conditions to the placebo. There was a significant positive effect of L-theanine plus caffeine versus the placebo on the Bond-Lader alert subscale (p < 0.01), as shown in Figure 4. There were no other significant effects on anxiety with L-theanine in isolation and L-theanine taken with caffeine (Haskell et al., 2008).

![Figure 4. Average baseline change (mm) in the Bond-Lader’s subscale of alert with placebo, 150 mg caffeine, 250 mg L-theanine, and a combination of 150 mg caffeine and 250 mg L-theanine. More positive values indicate higher positive ratings (p < 0.01). From Haskell et al., 2008.]

Like Steptoe et al. (2007), Haskell et al. (2008) considered the confounding variable caffeine within their analysis. Since caffeine is commonly found within tea, it is important to consider the effects of both exposures taken together. Subjects were tested for their adherence to caffeine intake with saliva samples. Higher levels of caffeine were
Accordingly found in the caffeine and in the combined caffeine and L-theanine treatments (p < 0.0001) (Haskell et al., 2008).

Although Haskell et al. (2008) found a significant effect of L-theanine and caffeine on alertness, caffeine in isolation also improved factors of cognitive ability. This may suggest that caffeine, not L-theanine, could be the primary contributor to the effect on alertness. It is also important to note the potential side effects when supplementing with L-theanine. Compared to the other treatment conditions, subjects who took the isolated form of L-theanine had impairment with an attention span activity and also had elevated reports of headaches (p < 0.05) (Haskell et al., 2008). It is also critical to consider that the given levels of L-theanine (250 mg) and caffeine (150 mg) in this experiment are much greater than what are found in a typical tea bag. Haskell et al. (2008) would need to examine the effects of L-theanine and caffeine at more realistic dietary levels (50 mg, 40 mg) in order to extend any findings to tea.

In another clinical trial with crossover design, Lu et al. (2004) compared L-theanine’s acute effects on anxiety to a placebo and positive control, alprazolam (Lu et al., 2004). Outcomes of anxiety were measured before and after treatment administration with the BAI, VAMS, and STAI. In this study, 12 males and four females ages 18-34 were found through University announcements. Subjects were screened and interviewed for the following requirements: subjects had to be non-smokers, healthy, could not have any individual or ancestral history of psychological disorders, and could not be taking any medication. Treatments included 200 mg of L-theanine as the supplemental form Suntheanine, 1 mg of alprazolam as the commercial brand Xanax, and a placebo pill. All participants were given each treatment 3 times on separate days, with at least 7 days in
between testing. In an effort to account for metabolism disparities, participants were told to consume a similar breakfast the day of treatment, to not drink caffeine or alcohol 24 hours before testing, and were given a standard meal 3.5 hours after their designated treatment. In an attempt to control for variations in mood due to a woman’s menstrual cycle, females were only tested during their follicular phase (Lu et al., 2004). These procedures improved consistency among subjects.

After giving subjects their treatments, this double-blind, repeated measure experiment assessed behavioral anxiety in two task conditions: a state of relaxation and a state of experimentally induced anticipatory anxiety (Lu et al., 2004). Anticipatory anxiety is a common response to stressful, critical situations and can be elicited by electrical shocks. The experimental condition asked subjects to focus on their feelings while staring at a computer screen. When a red border framed the screen, participants in the experimentally induced anxiety group were informed they would randomly receive electrical stimulation to their right hand. Contrary, the participants in the relaxed condition were correctly told they would not receive the electrical shocks and had a blue border frame their computer screen. The colored stimulus appeared for a total of 180 seconds within each 5-hour experiment (Lu et al., 2004).

Anxiety was measured with the BAI, VAMS, and STAI 1 hour before treatment, 2.5 and 5 hours post-treatment (Lu et al., 2004). The BAI, VAMS, and STAI are all self-reported subjective questionnaires that were previously discussed in this literature review. All of the questionnaires were all-inclusive besides the VAMS. The VAMS only focused on three of its eight different mood subscales: calm-excited, relaxed-tense, and tranquil-troubled. Average treatment scores with respect to time were calculated and
compared to baseline values. Scores were analyzed with multivariate analysis of variance and task conditions were compared with a paired-sample t-test. Subjects were also monitored for any changes in physiological symptoms (Lu et al., 2004).

Scores for the tranquil-troubled subscale of the VAMS showed that L-theanine had slight calming effects when compared to alprazolam or a placebo during subjects’ relaxed states ($p = 0.03$) (Lu et al., 2004). However, L-theanine had insignificant effects for all of the remaining questionnaires in both task conditions. With only one subscale having a significant relationship, effects of L-theanine on anxiety may be considered weak. However, Lu et al. (2004) still argued that their experiment provided evidence that L-theanine has calming effects when an individual is in a state of relaxation.

Unlike Steptoe et al. (2007) who mostly reported logical subjective and objective outcomes in their study involving black tea intake on acute levels of stress, Lu et al. (2004) had multiple controversial issues when analyzing their results. As a prescription drug designed for the relief of anxiety, alprazolam was the experiment’s positive control—it was expected to reduce subjective anxiety scores. Contrary to expectations, alprazolam didn’t have any calming effects for any of the questionnaires in the relaxed or anxiety-induced state when compared to the placebo. In fact, alprazolam actually heightened STAI anxiety scores when compared to the placebo in the relaxed condition. These paradoxical results may lessen the validity of the experiment’s findings with L-theanine. If the subjective measures were too insensitive to detect alprazolam’s effects on anxiety, it may also be too insensitive to detect L-theanine’s effect on anxiety.

Further questions emerge when analyzing the findings. The anxiety-induced condition was designed to elevate anxiety. However, when compared to the relaxed...
condition, average anxiety scores were only heightened in the BAI and the tranquil-
troubled subscale of the VAMS (p < .05) (Lu et al., 2004). The other subjective tests did
not show any differences between the relaxed and anxiety-induced condition. Outcomes
with their conditions contrast with Steptoe et al. (2007), where subjects appropriately
had greater average stress ratings from the behavioral challenging tasks. Lu et al. (2004)
tried to rationalize the insignificant effects seen with L-theanine and alprazolam,
suggesting that results were insignificant because the experimentally induced anxiety
level was too strong. However, if this were true, then the analyses should have shown
that anxiety was heightened during the anticipatory anxiety state. As previously
mentioned, the majority of their results did not confirm this.

Although Lu et al. (2004) used a positive control and three reputable tests to
measure outcomes, most of their results were conflicting. Lu et al. (2004) only focused
on the positive findings to support their hypothesis: that alprazolam and L-theanine
would reduce subjective anxiety. The Lu et al. (2004) study also had a low sample of 16
subjects. Participants were educated, young adults in a laboratory setting, and findings
cannot be generalized to a natural environment. Study results also cannot be compared
to the amount of L-theanine found in tea. Like Haskell et al. (2008) who used 250 mg of
L-theanine as the treatment, Lu et al. (2004) also used a dose of L-theanine (200 mg)
that is much greater than the average cup of tea (50 mg). Additionally, although Lu et al.
(2004) mentioned that outcome measurements taken at 2.5 and 5 hours post-treatment
coincided with peak pharmacokinetic effects of L-theanine, L-theanine is suggested in
the literature to have effects within half an hour (Tea Association of the USA, 2013).
The analysis may have been more thorough if it included the immediate effects of L-theanine on anxiety.

*Measurement of alpha-brain waves.* A few studies have relied on using electroencephalography (EEG) to examine L-theanine’s effect on alpha brain waves. As previously mentioned, a higher proportion of alpha brain waves are associated with a state of relaxation. If L-theanine enhances brain wave activity, it may be identified as the constituent responsible for calmness.

Juneja and his colleagues were some of the first individuals who studied the effects of L-theanine on alpha brain waves. Outcomes of alpha wave production were measured on the parietal and occipital brain areas (Juneja, Chu, Okubo, Nagato, & Yokogoshi, 1999). Although this study is highly referenced in subsequent papers involving L-theanine and anxiety, it carries limited information because it is an unpublished experiment within a review paper (Juneja et al., 1999). This author will describe all pertinent information that Juneja et al. (1999) provided.

Eight subjects, ages 18-22 years, were selected from 50 female volunteers (Juneja et al., 1999). The study does not mention how or why the subjects were chosen. Participants were equally split into high- and low-anxiety groups based on results from the Manifest Anxiety Scale (MAS). Specific information regarding treatments was limited, however the study mentions that the treatment group switched off receiving 50 or 200 mg of Suntheanine, a commercial product for L-theanine. Suntheanine was prepared by dissolving the prescribed dose in 100 mL of water, whereas the control group only received water. Measurements were completed in 10-minute intervals from
time of intake until 60 minutes with EEG topography. Measurements were recorded twice a week for 2 months (Juneja et al., 1999).

Juneja et al. (1999) proposed that L-theanine, in doses of either 50 mg or 200 mg, would promote the generation of alpha waves (Juneja et al., 1999). After half an hour, subjects who ingested the L-theanine solution had a significant, dose-dependent enhancement of alpha brain waves compared to the control group. This finding is supported by the proposition that L-theanine crosses the blood-brain barrier within half an hour. The subjects produced the greatest amounts of alpha brain waves after 40 minutes of intake. Juneja et al. (1999) did not provide p-values or the specific quantitative changes in brain wave intensities between the control and placebo groups. Additionally, Juneja et al. (1999) did not describe the differences in brain waves between the 50 mg or 200 mg doses of L-theanine. Results between the high- and low-anxiety subjects also were not compared.

Although Juneja et al. (1999) depicted images of both alpha-1 and alpha-2 brain waves, alpha-1 brain waves are the only alpha subtype associated with a state of relaxation (Niedermeyer, 1997). Thus, only alpha-1 waves will be examined here. Figure 5 depicts the results of alpha-1 wave topographies from an EEG comparing the treatment and placebo groups (Juneja et al., 1999). This study did not describe if this image depicts the average scan of all individuals. This study also does not note if the subjects were relaxed or mentally aroused while receiving the brain scans. In this figure, a darker color (black) is associated with stronger brain waves. After 30 minutes, alpha waves appear to be greater in the L-theanine group compared to the placebo, which is consistent with the study’s proposed results. Alpha brain waves appear to be stronger for
water at 20 minutes than at other time periods; Juneja et al. (1999) do not provide reasoning for why this may occur.

![Diagram of alpha wave production](image)

Figure 5. Alpha wave production shown in 10-minute intervals after intake of either water or L-theanine in volunteers. Topographies are from EEG data. From Juneja et al., 1999.

Although results showed a positive effect of L-theanine on alpha brain waves, multiple limitations of this study need to be considered. Four authors in the Juneja et al. (1999) review are in a company that makes Suntheanine. Since the authors repeatedly promote the sale of Suntheanine, authors have a recognizable conflict of interest. Their biased position leads them to jump to the conclusion that Suntheanine effectively improves relaxation. As previously mentioned, this study comes from a review paper and its data were not individually published or peer reviewed. This can contribute to biased results and also limits available information that is necessary for analyzing the credibility of the study. Since this study does not mention that it was double-blinded or randomized, it could have had a biased sample selection and distribution. Without proper randomization, results could have been attributed to confounders. Baseline characteristics were not considered or adjusted for in the analysis. Additionally, the small sample size increases likelihood of spurious results.
Compared to Juneja et al. (1999) who used 50 and 200 mg of L-theanine as their exposure, Nobre et al. (2008) solely gave 50 mg doses of L-theanine to the treatment group. Nobre et al. (2008) examined alpha brain wave measurements in relation to time as the primary outcome. Sixteen young, healthy volunteers were given 50 mg of powdered L-theanine mixed in a tea beverage. Nineteen participants in the control group were given 100 mL of chilled drinking water (Nobre et al., 2008). Baseline characteristics about the subject were not studied.

During the outcome measurement, participants were watching television and under a relaxed, alert mentality (Nobre et al., 2008). Nobre et al. (2008) used EEG’s to examine the changes in alpha brain waves at 45, 60, 75, 90, and 105 minutes after the tea or water treatment. In the experimental group, alpha brain waves significantly increased in linear correlation with elapsed time ($p < 0.05$) (Nobre et al., 2008). Figure 6 displays the average alpha brain wave activity over time. In this figure, brain waves are measured in microVolts$^2$ and greater amount of alpha brain waves are associated with a red-orange hue. Clearly, the L-theanine group appears to have a greater average production of alpha waves.
At all time intervals, alpha intensity appears to be significantly greater in the group consuming L-theanine compared to the control. These results support that L-theanine may affect the central nervous system and enhance relaxation (Nobre et al., 2008). When compared to the Juneja et al. (1999) study, the Nobre et al. (2008) study was much more descriptive in experiment specifications. For instance, they noted specifics involving the selection of subjects and included the quantitative measurements of alpha bands. This study also had a larger sample size (35 subjects) than the Juneja et al. (1999) experiment (8 subjects), improving the applicability of findings. Compared to Juneja et al. (1999) who used 50 and 200 mg of L-theanine, Nobre et al. (2008) also used more consistent, realistic dietary amounts of L-theanine (50 mg).

The Nobre et al. (2008) study did have distinct limitations, similar to the Juneja et al. (1999) study. The authors are affiliated with the sale of L-theanine products. This

Figure 6. Average power of alpha brain waves (microVolts²) over time (minutes) after consumption of water or 50 mg L-theanine beverage. From Nobre et al., 2008.
may lead to publication bias, limiting findings. Also, these experiments were not randomized or double-blinded, and baseline characteristics were not assessed or adjusted for. Nobre et al. (2008) did not assess anxiety levels in patients, yet differences in anxiety levels among subjects could be a major confounding variable. Whereas Juneja et al. (1999) measured outcomes twice a week for a period of 2 months, Nobre et al. (2008) only measured outcomes once.

The five clinical studies that were analyzed in this literature review are summarized in Table 3. Although the findings support that tea and L-theanine have significant anxiolytic effects, it is important to consider each study’s limitations. Juneja et al. (1999) and Nobre et al. (2008) were parallel-arm trials that lacked randomization and were not double-blinded. Other limitations include publication bias and the lack of adjustment for confounders. As a result, they may be considered as studies of insufficient quality to confirm a meaningful relationship between L-theanine and anxiety. Stronger studies included Steptoe et al. (2007), Haskell et al. (2008), and Lu et al. (2004). All studies in this group were double-blinded, and Steptoe et al. (2007) and Haskell et al. (2008) were randomized studies. The Steptoe et al. (2007) study was parallel-arm, whereas Haskell et al. (2008) and Lu et al. (2004) were experiments with a crossover design. Although these studies had their individual limitations, including small sample sizes and acute doses of L-theanine that are too high to be relatable to a single serving of tea, they are higher quality studies.
<table>
<thead>
<tr>
<th>Study and design</th>
<th>Subjects</th>
<th>Treatment</th>
<th>Outcome Measure</th>
<th>Findings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Steptoe et al., 2007; parallel-arm</td>
<td>75 healthy, tea-drinking men ages 18-55</td>
<td>Fruit-flavored black tea vs fruit-flavored beverage taken 4x/day for 6 weeks</td>
<td>Subjective measurement of stress and relaxation during behavioral challenging tasks with 7-point Likert scales taken pre- and post-treatment</td>
<td>• Tea group had 9.54% greater sense of relaxation vs. control amongst span of recovery • Similar ratings of average subjective stress levels when adjusting for pretreatment responses</td>
</tr>
<tr>
<td>Haskell et al., 2008; crossover</td>
<td>24 healthy undergraduate students ages 18-34</td>
<td>250 mg L-theanine, 250 mg L-theanine &amp; 150 mg caffeine vs. neither components in 250 mL iced tea taken 5x 7 days apart</td>
<td>Subjective measurement of anxiety during cognitive tasks with Bond-Lader VAS subscales alert, content and calm. Measurements taken prior, 30 &amp; 90 minutes post-treatment</td>
<td>• Caffeine &amp; L-theanine together improved alertness 30 &amp; 90 minutes post-dose • No significant relationships found between just L-theanine and outcome</td>
</tr>
<tr>
<td>Lu et al., 2004; crossover</td>
<td>16 healthy students ages 18-34</td>
<td>200 mg L-theanine as Suntheanine, 1 mg alprazolam vs. placebo pill taken 1x/week for 3 weeks</td>
<td>Subjective measurement of behavioral anxiety in state of relaxation and induced anticipatory anxiety with VAMS, BAI &amp; STAI</td>
<td>• Only tranquil-troubled subscale of VAMS showed slight calming effects</td>
</tr>
<tr>
<td>Juneja et al., 1999; parallel-arm</td>
<td>8 female subjects ages of unknown ages</td>
<td>50 mg, 200 mg L-theanine as Suntheanine in 100 mL water vs. water. Unknown length &amp; interval of intake</td>
<td>Objective measurement of alpha waves on unknown state with EEG 2x/week for 2 months</td>
<td>• L-theanine had dose-dependent enhancement of alpha brain waves vs. control after 30 minutes • L-theanine had greatest amounts of alpha waves after 40 minutes of intake</td>
</tr>
<tr>
<td>Nobre et al., 2008; parallel-arm</td>
<td>35 young, healthy volunteers of unknown ages</td>
<td>50 mg powdered L-theanine mixed in tea beverage vs. 100 mL water taken once</td>
<td>Objective measurement of alpha waves with EEG during relaxed state at 45, 60, 75, 90, 105 minutes post-treatment</td>
<td>• Alpha intensity greater in L-theanine group vs water at all measured time intervals</td>
</tr>
</tbody>
</table>

*a Outcome measure abbreviations:
VAS: Visual analogue scales. Measures subjective anxiety through a series of 100 mm scales.
VAMS: Visual analogue mood scales. Measures mood behavior through the marking of various statements or symbolic faces.
BAI: Beck Anxiety Inventory. Measures cognitive, subjective, and objective indicators of anxiety using a 4-point Likert scale.
STAI: State-Trait Anxiety Inventory. Measures state and trait anxiety through 4-point Likert scales.
EEG: Electroencephalography. Records electrical brain wave activity.
Summary & Future Research Needs

This literature review assessed two cross-sectional and five clinical trials, all of the available literature relevant to this topic. The two observational studies and first clinical trial focused on the consumption of tea as the primary exposure and used subjective measurements of stress as the primary outcome. The four remaining clinical trials assessed tea’s constituent, L-theanine, as a potential relaxation treatment by using subjective and objective outcome measurements. Despite the suggestion that tea and its constituent L-theanine may provide anxiolytic effects, the seven studies evaluated in this review provide contradictory evidence to support this claim. Although the studies focused on similar subject matter, the studies are not truly comparable. The studies had different types and lengths of exposures, methods of measurement, and outcomes. Overall, however, the majority of studies had limitations that outweighed their strengths.

Most of the studies in this literature review had a small sample size involving healthy, young adults as sample subjects. In order to extend findings to the general population, larger sample sizes, different ages, ethnicities, educational levels, and regions need to be considered. Testing in different environments also needs to be performed. Experiments should include a non-stress control condition along with an experimentally induced stress condition to examine the effects of social circumstances. Additional intervening variables, including caffeine, need to be adequately recognized and controlled for. Therefore, results can more likely be attributed to the designated exposure.
This author suggests that additional research is needed before forming any definite conclusions involving tea and its constituent L-theanine on anxiety. Specifically, more double-blinded, randomized controlled clinical trials that are crossover design need to be performed. In order to form any significant conclusions, future studies need to be undertaken using consistent exposures. This way, results from separate studies can be more accurately compared. If a study uses L-theanine as an exposure, doses at realistic dietary levels need to be prescribed. If a study uses tea as an exposure, studies should involve all common tea types, so that the exposures can be relevant to the standard tea-drinking population. It is also recommended that future studies give exposures for longer periods of time. A chronic treatment may give a more accurate understanding of how habitual intake of tea or L-theanine affects anxiety.

Future studies also need to include systematic measurements for assessing exposures and outcomes. Tools need to be practical and validated. In order to obtain the most accurate and comprehensive data from subjects, it would be beneficial for studies to use both subjective and objective measurements during their assessments. Subjective measurements, including self-reported questionnaires, elicit issues such as bias and human error. Objective measurements, such as the measurement of alpha brain waves, can be considered to be an indirect, crude measurement. When used together, corresponding subjective and objective outcomes may help verify data.

One key research question that still remains unanswered is whether the effects of L-theanine in isolation are truly comparable to L-theanine found in tea. Although L-theanine can be provided as a pharmacological compound in higher amounts than what is normally found in tea, this is irrelevant to the main focus of this literature review. The
majority of studies used acute amounts of L-theanine much higher, or around 4-6 times
greater, than what is usually found in a single serving of tea. The mechanism and
bioavailability behind L-theanine is unknown. Thus, isolated L-theanine may have
different outcomes than when it is found in its holistic entity, tea. To make appropriate
comparisons, future studies need to focus on amounts of L-theanine that can be easily
achieved in an individual’s typical diet. If studies had significant protective findings
with L-theanine, the protective relationship could be more relatable to tea.

Due to anxiety’s high prevalence and co-morbidity, it is important to continue
research on this topic. The United States has a high prevalence of anxiety, yet all of the
studies in this review were either investigated or published from authors around the
globe. As seen, our scientific community has not given sufficient time and resources to
investigate this topic. Yet, current treatments for anxiety are expensive, addicting, carry
adverse side effects, and do not mitigate symptoms for everyone. Alternative methods,
including tea, may be promising. Tea is a low-cost beverage that is universally accepted,
does not have any serious side effects, and may have a variety of health benefits. With
the growing number of tea-drinkers and diagnoses of anxiety disorders, it is worthwhile
to determine whether there is a significant relationship between tea, its constituent L-
theanine, and anxiety.


Julian, L. J. (2011). Measures of anxiety: State-Trait Anxiety Inventory (STAI), Beck Anxiety Inventory (BAI), and Hospital Anxiety and Depression Scale-Anxiety


